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INVITED SPEAKER PRESENTATIONS

I1

Hepatitis E: state of the art

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Retrovirology 2010, **7(Suppl 1):**I1

Hepatitis E (HEV) has traditionally been thought to be a disease of developing countries. In this setting, HEV causes hepatitis in young adults. It is generally a self-limiting illness, with a good prognosis, except in pregnant women and patients with pre-existing chronic liver disease, in whom the mortality is 20% and 70% respectively.
Traditionally HEV was considered rare in developed countries, and confined to travellers returning from endemic areas. However, recent data has shown that locally acquired HEV infection is common in developed countries. Acute HEV infection in this setting is caused by HEV genotype 3 and appears to have a predilection for middle aged/elderly males. It has a significant morbidity (15%) and carries an adverse prognosis in chronic liver disease. HEV infection is commonly misdiagnosed as drug-induced liver injury. The source and route of infection are currently uncertain, but evidence suggests that HEV genotype 3 is a porcine zoonosis, which can be transmitted via the human food chain.
Until very recently chronic HEV infection was thought not to occur. However studies from Europe have shown that HEV genotype 3 can cause chronic infection with rapidly progressive cirrhosis in patients with immunocompromise. This includes patients taking immunosuppressive therapy following solid organ transplantation and patients with HIV-1 infection (NEJM 2009; 361(10): 1025-7).
HEV IgG seroprevalence in developed countries is variable, with rates of 16% reported from the UK and SW France. These data indicate that HEV infection is either commonly subclinical, or commonly overlooked as a diagnostic possibility. However, the burden of HEV disease in both the developing and developed world is currently uncertain. The worldwide burden of chronic HEV infection may prove to be more considerable than previously thought likely, as countries where HEV is endemic also have a high seroprevalence of HIV.

I2

Lessons learned from Chikungunya

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Retrovirology 2010, **7(Suppl 1):**I2

Chikungunya virus (CHIKV) is an Arbovirus belonging to the Alphavirus genus, which is transmitted by *Aedes* spp mosquitoes. Since late 2004, CHIKV caused several outbreaks in coastal Kenya, in the Indian Ocean (especially on the island of La Reunion), and on the Indian subcontinent, where it caused more than 1.5 million cases. Although *Aedes aegypti* is the most commonly involved vector, other *Aedes* spp mosquitoes, such as *Aedes* albopictus, appear to have a good vectorial capacity for CHIKV. The vectorial capacity of *Aedes albopictus* is of special concern, since this mosquito is widespread in several countries in Southern Europe and in other areas of the world, outside tropical areas.
In the summer of 2007, an unexpected outbreak of CHIKV fever, caused more than 200 human cases in the Emilia-Romagna Region of Italy; most of the cases were recorded in two villages in the Province of Ravenna, but smaller clusters were also detected in other towns in the same Region (i.e., the towns of Cervia, Cesena, Ravenna, Rimini, and Bologna). The tropical virus was introduced in Italy by a man from Kerala (an Indian district affected by a large outbreak) and sustained by local mosquitoes (*Aedes albopictus*, "the tiger mosquito"), which transmitted the infection to other persons. The epidemic can be said to have been the result of the globalisation of vectors and humans, which occurred through a two-step process: i) the introduction and adaptation of *Aedes albopictus* to a new environment (i.e., an area with a temperate climate); and ii) the introduction of Chikungunya virus (CHIKV) in a previously infection-free country, as a result of population movement. The CHIKV strains introduced in Italy contained a mutation in the E1 glycoprotein which was responsible for a single amino-acid substitution (A226V) able to increase the infectivity of the virus for *Aedes albopictus*. CHIKV outbreaks did not reoccur in the areas that had been affected by the 2007 outbreak. This was probably the consequence of three combined factors: i) the lack of human cases during the winter season, due to extremely reduced *Ae. albopictus* activity; ii) the mosquito control activities performed during and after the outbreak; and iii) the extremely low rate of transovarial transmission of the infection (i.e., transmission from the adult mosquito to larvae). By contrast, on the island of La Reunion, although the number of human cases decreased during the dry season, they did not completely disappear, and a second epidemic wave ravaged the local population determining significant increase in mortality.
Investigations conducted during the recent outbreaks provided a unique opportunity to improve our knowledge on epidemiological, clinical and virological aspects of CHIKV infection (i.e., estimate of CHIKV infection R_0, definition of the clinical spectrum of the disease, duration of viremia, antibody dynamics), and on evolutionary changes leading to virus adaptation to different vectors.
In conclusion, the re-emergence of CHIKV is paradigmatic of the infectious threat in the era of globalisation and emphasises the need for preparedness and response to vector-borne infections and other emerging infectious threats.

BioMed Central

I3

The influenza challenge

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Retrovirology 2010, **7(Suppl 1):**I3

The presentation aims at introducing the various challenges faced at global level during the first pandemic of the 21st century.
On 25 April 2009, Mexico, under the International Health Regulations (2005) (IHR (2005)), notified the World Health Organization (WHO) about an outbreak of influenza-like illness. The responsible virus was identified a few days later by a WHO Collaborating Centre for Reference and Research on Influenza and a reference laboratory in Canada. This was the first reported event related to Influenza A (H1N1). In less than nine weeks, the pandemic virus spread in the six WHO regions. On 11 June 2009, WHO announced a pandemic phase 6.
Because of the threat posed by the re emergence of A(H5N1)virus in 2003 in Asia, most of the influenza experts and modellers were predicting that the next pandemic would start in Asia with similar characteristic as the H5N1 disease currently observed with a high case fatality ratio. One of the lessons learned is that predictions are very difficult when related to influenza viruses due their unpredictable evolution and the complexity of factors leading to pandemic.
The pandemic (H1N1) 09 is challenging in many aspects: firstly, the uncertainty about the evolution of the virus makes mid term preparedness and planning difficult.
Secondly, the weakness of some surveillance system does not allow an easy monitoring of the disease and its spread. The oseltamivir resistance monitoring has also been difficult in some parts of the world.
Thirdly, the modern communications means enable rumours to spread faster than ever. Public health authorities have to include risk communication in their strategy to respond to the outbreak.
The nature of this pandemic with moderate severity and very rapid spread imposed to revise national pandemic preparedness plans during the course of the event. The public and the media often misunderstood the rational for the modification.
The international community has responded quite well to the pandemic threat. In particular, countries have been especially collaborative regarding the exchange of information and the global access to supplies such as vaccine and antiviral drugs.

I4

Ageing with HIV: next challenges

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Retrovirology 2010, **7(Suppl 1):**I4

With the advent of effective HAART and subsequent improved survival and also with ongoing HIV transmission including amongst older persons, the HIV positive population accessing care is ageing and a significant proportion of patients (up to 25% in some cohorts) are now aged over 50.
Older age has an effect on both HIV natural history and response to HAART. Older persons in the pre-HAART era progressed more rapidly to AIDS or death after seroconversion and AIDS diagnoses occurred at higher CD4 counts. Virological response to HAART is typically greater in older patients, likely mediated by greater levels of adherence, although immunological responses appear blunted. Some HAART toxicities occur at a higher frequency on older patients, although tolerance of adverse effects may be greater.
Additionally, it is increasingly suggested that HIV may result in accelerated ageing. This is thought to result from a combination of lifestyle factors, drug toxicities, and chronic inflammation/immune activation which persists even in the setting of virological suppression. Morbidities that were not historically considered to be HIV-related (e.g. cardiovascular disease, osteopaenia, "non-AIDS" malignancies) are now considered to be related to both ongoing HIV replication and chronic immune activation, and all are associated with increasing age.
It is therefore likely that HIV treatment recommendations (such as when to start HAART and what to start with) may need to be adapted for the older patient. Furthermore, older patients with HIV are likely to increasingly experience multiple co-morbidites which will require complex management and significant challenges to healthcare delivery.

I5

Cardiovascular disease and HIV

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Retrovirology 2010, **7(Suppl 1):**I5

Different population studies have consistently shown that HIV-infected patients have approximately 2-fold higher incidence of coronary artery disease or myocardial infarction than non-HIV-infected persons.
However, all these studies have shown a higher prevalence of traditional cardiovascular risk factors (smoking, hypertension, diabetes mellitus, and dyslipidemia) in HIV+ relative to HIV-. These factors are the most common determinants of the cardiovascular risk in a given HIV-infected patient. They should be screenned for in any patient and agressively treated if estimated cardiovascular risk is moderate/high.
HIV infection itself additionally contributes to a higher risk of cardiovascular disease through inflammation, immune depression (low CD4 cell counts), and inmmune activation. There is a need to control HIV infection with antiretroviral therapy to decrease the cardiovascular risk associated with HIV infection. The overall effects of any effective antiretroviral therapy on cardiovascular disease are definitely more positive than not giving therapy at all. Antiretroviral therapy contributes to decrease the effect of HIV infection, although it may be unable to lower it to a level similar to that of uninfected persons.
Antiretroviral therapy may contribute to cardiovascular risk in a more modest way than that of traditional risk factors and uncontrolled HIV infection, through the induction of metabolic abnormalities (dyslipidemia and insulin resistance). This effect has been proven for protease inhibitors, and it remains controversial for thymidine nucleoside reverse transcriptase inhibitors. It should be beard in mind that not all protease inhibitors and not all patients receiving a protease inhibitor necessarily develop metabolic abnormalities. It is unclear whether antiretroviral therapy may have pathogenetic mechanisms other than metabolic abnormalities contributing for a higher cardiovascular risk.
Abacavir has been identified as a marker of cardiovascular disease, but its potential role as a causative agent is confounded by multiple factors that are impossible to adjust for completely in cohort studies. A plausible underlying mechanism is not known either. The results of a BICOMBO sub-study sugested that abacavir does not not cause inflammation, endothelial dysfunction, hypercoagulability, or insulin resistance in virologically suppressed HIV-infected patients. As of January 2010, health authorities in Europe and in the United States (EMEA and FDA, respectively) have concluded that there is no definitive information proving that abacavir may induce cardiovascular disease.

I6

Ageing, metabolism and HIV

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Retrovirology 2010, **7(Suppl 1):**I6

HIV infection is now considered as a chronic disease, most patients being well-controlled and experiencing long-term survival. However, these patients encounter an increasing number of complications and in particular of age-related comorbidities occurring earlier than in the general population: cardiovascular disease, dyslipidemia, diabetes, osteoporosis, liver and kidney failure, neurocognitive impairment, non-AIDS defining cancers. Therefore, it is now considered than some patients

present a phenotype of premature aging, the origin of which remains unknown.
In the general population, most age-related comorbidities including cancer have been linked to long-term chronic inflammation (inflammaging). This inflammation could partly result from adipose tissue redistribution and hypertrophy leading to insulin resistance, dyslipidemia and altered glucose tolerance. Inflammation is also involved in the occurrence of atherosclerosis and increased cardio-vascular risk, osteoporosis, liver dysfunction and neurocognitive disorders. Two main contributors to cellular senescence and inflammation are activation of the monocyte/macrophage system and increased oxidative stress.
In HIV-infected patients, recent works indicate that even well-controlled patients present low-grade inflammation as shown by increased level of CRP. Links between CRP, increased intima-media thickness and the occurrence of myocardial infarction have been shown. Similarly, it is hypothesized than brain inflammation could play a role in neurocognitive impairment presented by some patients. The origin of increased inflammation is probably complex and multifactorial. A role for persistent viral infection is postulated and infected immune cells such as macrophages can produce deleterious viral proteins, induce an oxidative stress and release pro-inflammatory cytokines. Long-term immune activation could result in immunosenescence and increased proinflammatory cytokines level. Some antiretroviral drugs induce an oxidative stress. Finally, patients' linked parameters are important to consider: age, smoking, metabolic disorders, hypertension, vitamin D deficiency and life-style environment (lipid-rich diet, sedentarity).
It is important to control these alterations: treat early, avoid drugs with specific tissue toxicity in patients with risk factors, take in charge the metabolic alterations (hypertension, dyslipidemia, diabetes), compensate vitamin D deficiency if present. A safe life-style (stopping smoking, exercise, diet) is strongly recommended for these patients.

I7

Neurodegeneration in the HAART era

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Retrovirology 2010, **7(Suppl 1):**I7

Highly active antiretroviral therapy (HAART) has led to dramatic changes in HIV disease. Patients are now living for a significantly longer time; with increasing numbers over the age of 60. Most have been living with HIV disease for many years, some in excess of 20 years. Additionally, the number of patients over 50 years old who have recently acquired HIV infection is increasing. Despite the benefits of HAART, cognitive impairment remains. Whilst the incidence of HIV associated dementia (HAD) has significantly fallen with HAART, its prevalence is increasing partly because patients are living longer with fixed deficits. Furthermore, the prevalence of milder cognitive impairment, now termed minor neurocognitive disorder and included under the broader term HIV-associated neurocognitive disorder (HAND), has not changed despite the introduction of HAART. While there are many potential reasons for this, there is concern that the longer duration of HIV disease, as a consequence of HAART, together with the increasing age of infected persons may have a compounding detrimental effect on cognitive function. Additionally, these two factors may facilitate and perhaps enhance the expression of a variety of neurodegenerative diseases as HIV-infected patients approach the age where such disorders become increasingly common. This presentation will review the evidence for the potential compounding effect of age on cognition in HIV disease by examining the evidence for persistent and developing neurodegeneration in HAART treated patients, particularly in those maximal suppression of HIV viral load in the blood and cerebrospinal fluid (CSF). Analysis of three different cohorts has shown an approximate rate of HAND of 35% in patients with advanced HIV disease who are virally suppressed. Evidence for the facilitation of neurodegenerative diseases by HIV and age, will also be detailed. This primarily centres around evidence for the development of an Alzheimer like illness and Parkinsonism. This is followed by a delineation of the potential mechanisms firstly by a review of the general aspects of the pathogenesis of neurodegenerative diseases. Then the effects of normal ageing and how they intersect with HAND will be discussed, followed by a review of the overlapping features and mechanisms in HIV and neurodegenerative diseases. Lastly two potential therapeutic interventions, the rationale for optimising HAART to ensure adequate brain penetration of antiretroviral drugs, and the clinical value of risk factor reduction for neurodegenerative diseases will be reviewed.

I8

Diffusion of new antiretroviral drugs in CSF

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Retrovirology 2010, **7(Suppl 1):**I8

An aim of the presentation will be to review the relative importance of ARV penetration into the central nervous system (CNS) for optimizing virological response and minimizing cognitive deficits. A brief summary of clinical studies addressing the importance of ARV penetration will be included. A focus of the discussion will be on pharmacological factors driving CNS penetration (e.g. protein binding, lipophilicity and interaction with membrane transporters). The talk will summarize current knowledge for the most widely prescribed ARVs. Methodological approaches will be discussed in that studies addressing CNS penetration often rely on sparse PK measurements within the cerebrospinal fluid (CSF) coupled with population PK modelling. Few studies include intensive pharmacokinetic (PK) measurements in both CSF and plasma for estimating the CSF/plasma exposure ratio. Pharmacodynamic studies assessing the importance of CSF PK exposure for virological response will be reviewed with an emphasis on the newer ARVs including raltegravir. Raltegravir achieves CSF concentrations providing adequate HIV inhibition in the majority of patients. Results for additional new ARVs will also be presented.

I9

Inhibitory quotient in HIV pharmacology

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Retrovirology 2010, **7(Suppl 1):**I9

Aim: This presentation will review the concept of the inhibitory quotient and its use in HIV pharmacology.
Materials and methods: Recent publications including research articles, abstracts and review articles were retrieved to provide an overview of recent advances in the field.
Results: The inhibitory quotient is the ratio of drug exposure to viral susceptibility. There are a number of different ways to calculate the inhibitory quotient. The trough concentration is the most frequently used pharmacokinetic parameter to represent drug exposure in inhibitory quotient calculations. To represent resistance, both phenotypic and genotypic data can be used to calculate the inhibitory quotient. Using population averages for drug exposure and resistance it is possible to compare the inhibitory quotient of different drugs. More frequently the inhibitory quotient is used in the context of therapeutic drug monitoring. The inhibitory quotient has mainly been studied for protease inhibitors. For non-nucleoside reverse transcriptase inhibitors a single mutation can cause high level resistance, whereas for PIs, mutations have a smaller but cumulative effect. For this reason it is unlikely that the inhibitory quotient will be helpful in the therapeutic drug monitoring of non-nucleoside reverse transcriptase inhibitors. For the newer drugs in the classes of CCR5 and integrase inhibitors it is not yet clear what the role for inhibitory quotient could be. In terms of therapeutic drug monitoring cutoff values have been proposed for the genotypic inhibitory quotient as well as for the phenotypic inhibitory quotient.
Discussion: The inhibitory quotient has been topic of discussion for the past decade. We have moved from the initial discussions on how to compare inhibitory quotients for different drugs into the use of inhibitory quotient as a useful parameter for therapeutic drug monitoring. Further data are needed to confirm the respective roles of different forms of inhibitory quotient in daily practice.

I10

Drug-drug interactions: it is not only CYP450's which matter

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Retrovirology 2010, **7(Suppl 1):**I10

The objectives of this review are:

• To highlight that although CYP450's are a major target for drug-drug interactions other mechanisms need to be considered.
• To appreciate that important interactions also occur due to a change in pH (gut) and inhibition of non P450 enzymes (eg UGTs).
• To discuss the increasing importance of understanding transporter based interactions in the gut, liver, kidney and blood brain barrier.
• To outline new data on clinically important drug-drug interactions between antiretrovirals and between antiretrovirals and other drugs.
• To discuss how to predict, manage and avoid drug-drug interactions including an outline of on-line resources available for this purpose.

Pharmacokinetic drug interaction studies performed during the drug development process, or post-licensing provide the substantive data base from which recommendations regarding the use of certain drug combinations are made. However given the sheer number of potential interactions extrapolation on the basis of potential mechanism of interaction is also important. Thus a foundational knowledge of drug disposition (enzymes, transporters involved etc) is essential so that *in vitro* data (is the drug a substrate; is the drug an inhibitor of a particular enzyme or transporter) can be used to underpin a clinical study or be the basis for an informed decision re the potential for an interaction. While the major focus in the HIV field has been on CYP450 enzymes (for the obvious reason that many of the drugs are extensively metabolised and/or are inducers/inhibitors) there is a growing awareness of the key role for other proteins – in particular UDP-glucuronyltransferases (UGTs) and transporters (ABC transporters such as P-pg, MRP1,2,7; SLCO transporters such as OATP1B1, OATP1B3, OATP1A2, OCT1,2, OAT1,2). This is a rapidly emerging field and one which is going to impact not only on our understanding of mechanisms of drug-drug interactions but also on seeing the bigger picture in relation to the role of pharmacogenetics in inter-individual variability.
Unexpected interactions will continue to emerge and will need to be managed.
Ultimately the key to management of patients on multiple drugs is clinical vigilance, access to adequate resources to help inform (eg web based resources), utility of therapeutic drug monitoring where available and close follow up of patients.

Recommended reading:

Dickinson L, Khoo S, **Back D**. Pharmacokinetics and drug-drug interactions of antiretrovirals: An update. Antiviral Res. 2009 Aug 7 (Epub ahead of print).

I11

Evidence-based TDM for antiretroviral drugs

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Retrovirology 2010, **7(Suppl 1):**I11

Aim: Evidence-based practice is essential to improving patient safety and the effectiveness of health care practices. The impact of therapeutic drug monitoring (TDM) on patients outcomes must be evaluated accordingly. Members of the 'TDM group' of the French Society of Pharmacology and Therapeutics worked together to address the role of TDM for various drugs. HIV protease inhibitors (PI) and non nucleoside reverse transcriptase inhibitors (NNRTI) have been evaluated in this context.
Methods: A systematic review of the literature was done. Published studies were analyzed and classified according to the methodology used: randomized or not, number of patients, exposition-efficacy and/or toxicity relationship, statistic method, pharmacokinetic parameters evaluated.
Results: The level of evidence for the TDM of atazanavir, amprenavir, lopinavir, indinavir, saquinavir, efavirenz and nevirapine was evaluated. Several controlled and non-controlled studies have been performed for these drugs, reporting pharmacokinetic-pharmacodynamic correlations on treatment efficacy and for some adverse effects, such as atazanavir and bilirubin elevations or efavirenz and neurological disorders.
Discussion: The level of evidence of the interest of first generation PI and NNRTI TDM is recommended, which is in accordance with the French and European recommendations for the management of HIV-infected patients.

I12

Is there a role for cytokine-immune based therapy in HIV disease therapy?

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Retrovirology 2010, **7(Suppl 1):**I12

Background: The partial capacity of HAART of reverting the immunological dysfunction associated with HIV-1 infection has stimulated the clinical investigation of cytokines as molecules endowed with strong immunological reconstitution potential.
Methods: Among other cytokines, cytokines utilizing a common γ-receptor chain for signal transduction (such as interleukin-2, IL-2, and IL-7) have been particularly studies for their high potential of boosting the adaptive immune response and of stimulating the proliferation of mature CD4+ and CD8+ T lymphocytes or of their precursors. In addition, GM-CSF, sharing some signaling pathways and clinical effects with IL-2 (i.e. boosting of $CD4^+$ T cell counts), has also been studied. The recent failure of phase III trials on the use of intermittent IL-2 administration in spite of its proven effect of increasing CD4+ T cell counts poses the question of whether these approach maintain interest and rationale to be pursued in the context of successful HAART regimens.
Results: Clinical experimentations with IL-7 and GM-CSF are still ongoing and maintain promising potential. We have recently reported that intermittent IL-2 therapy counteracts the *in vivo* evolution of HIV-1 from CCR5 monotropic use to dualtropic (CCR5 and CXCR4) use (*S. Ghezzi et al., HIV Medicine, 2009 Dec 8*). IL-7 is also considered a powerful tool to purge viral reservoir embedded in resting memory CD4+ T cells. In this regard, most HIV + individuals show the constitutive activation of a cleaved form of signal transducer and activator of transcription-5 (STAT5), commonly activated by IL-2, IL-7 and GM-CSF, therefore generating a transdominant negative inhibitor of STAT-5 dependent gene expression *(A. Crotti et al., Blood, 2007)*.
Conclusions: Cytokines maintain their potential to complement HAART at least in those patients discordantly responding to antiretroviral agents. Furthermore, certain cytokines may be useful in the purge or attrition of viral reservoirs in the perspective of eradicating HIV infection.

I13

Immunotherapy in HIV infection; current and future challenges

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Retrovirology 2010, **7(Suppl 1):**I13

Administration of HAART has resulted in significant improvements in the survival of HIV-infected patients. However, despite now reaching a point where we can achieve durable, maximal suppression of plasma viral load in most of our HAART-treated patients, non-AIDS-related morbidity and mortality among these patients remain a concern. Conditions typical of aging, such as cardiovascular disease and cancer, are seen at a higher rate in HIV-infected patients compared to the general population, potentially because the ability of HAART to restore immunocompetence appears incomplete–even in patients who have long-term undetectable HIV-1 RNA. New insights into the pathogenesis of HIV-1 infection highlight several new and promising areas of investigation for immune-based therapies, including strategies that target T-cell homeostasis and immune activation, as well those targeted at restoring immune responses directed against HIV. The rationale behind the investigation of a of cytokines such as IL-2 and IL-7 as adjunctive therapies to antiretroviral treatment is to improve the restoration of the immune system and improve HIV-directed immune responses. Among cytokines, IL-2, was extensively studied in several phase II and two large phase III studies. Results from these studies showed that IL-2 increases signficantly CD4 counts in the long term.

However, this biological effect did not translate into clinical benefit. These results raise several questions about the functionality of IL-2 expanded CD4 T cells that will be discussed.
The potential interest of IL-7 is based on its crucial role on T cell homeostasis both in thymic output and peripheral T proliferation and survival. This new promising cytokine is currently under evaluation is several I/II clinical trials in chronically HIV-infected patients with low level of immune restoration despite controlled viral load. Results from these studies will be presented and discussed.

I14

NK Cells and immune activation in HIV-1 infection

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Retrovirology 2010, **7(Suppl 1):**I14

Natural Killer (NK) cells have the potential to eliminate HIV-1 infected target cells and to influence the rate of HIV-1 disease progression. NK cells are, however, depleted during HIV-1 chronic infection and their functions remain diminished in individuals receiving antiretroviral therapy. Chronic immune activation may contribute to loss of NK cell functional potency in HIV-1 infection.
Aims: Here we investigate the contribution of HIV-1 viraemia and exposure to human cytomegalovirus (HCMV) to chronic NK cell activation and differentiation. We also assess the impact of HIV-1 on NK cells in the gastrointestinal (GI) tract and the association between microbial products translocated from the (GI) tract and chronic activation of NK cells.
Methods: Blood and colonic lamina propria NK cell activation and differentiation were studied in aviraemic and viraemic HIV-1 infected and HIV-1 seronegative control individuals without or with inflammatory bowel diseases (IBD). NK cell activation and receptor expression were assessed by flow cytometry, CMV exposure by anti-HCMV IgG Elisa and plasma lipopolysaccharide (LPS) by limulus amoebocyte assay.
Results: HIV-1 plasma viraemia has a dominant impact on NK cell losses and chronic NK cell activation whereas differentiation of blood NK cell receptor expression is associated with exposure to HCMV in HIV-1+ individuals. HIV-1 plasma viraemia is also associated with depletion of NK cells in the colonic lamina propria whilst atypical gut NK cell differentiation occurs in both HIV-1+ individuals and in HIV-1- IBD patients. Plasma LPS is elevated both in viraemic and aviraemic HIV-1+ individuals and in IBD patients. Chronic NK cell activation is, however, reduced in aviraemic HIV-1+ individuals and is absent in IBD patients.
Discussion: Depletion and chronic activation of NK cells is associated with HIV-1 viraemia whilst concomitant infections and ongoing inflammatory processes influence functional differentiation of NK cells in the blood and GI tract. These data have implications for innate immune competence in HIV-1 infected individuals.

I15

Molecular mechanisms involved in HIV latency and implications for HIV treatment and eradication

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Retrovirology 2010, **7(Suppl 1):**I15

Aim: The aim of this presentation is to review the molecular mechanisms necessary for the establishment of HIV-1 latency, their relationship with different cellular and anatomic reservoirs, as well as the current treatment strategies targeting viral persistence in latent reservoirs, their main limitations and future perspectives.
Methods: For years, the molecular mechanisms leading to HIV-1 reactivation have been characterised in detail but the study of latency has remained elusive due to the technical limitations that arise when a negative phenomenon, like the absence of replication, is studied. Development of new techniques for studying HIV-1 latency, the identification of factors that restrict retroviral infections, the characterisation of chromatin structure in the setting of viral integration, and the discovery of new systems regulating gene expression.
Results: Resting lymphocytes represent an extremely restrictive environment for HIV-1 replication. In contrast, immune activation of CD_4^+ T lymphocytes provides an optimal context for robust HIV-1 replication. Most mechanisms to maintain HIV-1 latency operate at transcriptional level such as the chromosome environment at the site of integration or the availability of viral and host transcription factors. In addition, HIV-1 integration and expression can be restrained or enhanced by different host cell factors such as IkBa, COMMD1, APOBEC3G, LEDGF and Emerin. Finally, both cellular and viral miRNAs could be involved in maintaining HIV-1 latency or in controlling low-ongoing viral replication. Identification of new cellular elements restricting the viral cycle provides a new paradigm on HIV-1 latency.
Discussion: As a lentivirus, HIV-1 is able to infect resting, non-dividing cells where the viral genome can be permanently integrated into the host cell chromosomes. Latent HIV-1 reservoirs are established early during primary infection in lymphocytes and macrophages and constitute a major barrier to eradication even in the presence of highly active antiretroviral therapy (HAART). HIV-1 latency should no longer be considered a merely passive event due to the lack of positive factors but as an active process that is maintained by cellular elements that regulate the gene expression program in the infected cell.

I16

Update on HIV reservoirs

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Retrovirology 2010, **7(Suppl 1):**I16

Aim: During combination antiretroviral therapy, reduction of HIV-1 RNA levels to less than 50 copies/ml is frequently achieved; however, residual low-level viremia has been detected using ultrasensitive assays. The source of persistent viremia is currently unknown: it could arise from ongoing cycles of replication in a sanctuary site, long-lived productively infected cells, and/or activation of viral expression from latently infected cell reservoirs. This presentation will discuss the characteristics and probable sources of persistent viremia.
Materials and methods: Insights into the source and dynamics of residual virus are determined using ultrasensitive assays for: 1) quantifying persistent viremia and 2) sequencing persistent HIV populations from plasma and cells.
Results: Using sensitive assays it has been shown that HIV persists for at least 7 years of therapy. The decay of persistent viremia in plasma of patients on suppressive therapy appears to be biphasic with short-lived cells contributing to the viremia initially followed by a second more constant release of virus from long-lived HIV-infected cells. An indicator that current treatment regimens halt ongoing viral replication is that intensifying a patient's treatment regimen does not result in a decrease in persistent viremia. Moreover, persistent viremia in patients under long-term treatment is genetically homogeneous and the viral population which rebounds during treatment interruption is also genetically homogeneous.
Discussion: The genetic homogeneity of the viral populations in patients on suppressive thereapy indicates that persistent viremia arises from long-lived latently infected cells, such as resting memory CD4+ T-cells, tissue CD4+ T-cells, tissue macrophages or stems cells, and reflects an evolutionary bottleneck during prolonged therapy. However, it is possible viral production may occur in such anatomical compartments as the gut or central nervous system where drug penetration is suboptimal.

I17

Treatments for persistent HIV infection: the road ahead

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Retrovirology 2010, **7(Suppl 1):**I17

Effective antiretroviral therapy (ART) suppresses viremia and allows immunological recovery, but the intrinsic features of retroviral biology allow HIV infection to persist despite ART. Persistent infection is primarily characterized by the twin phenomenon of latent infection of long-lived cells of the immune system, and continued virus release from undefined cellular sources.

Of late has there been a reawakening of interest in strategies to purge these latent reservoirs of HIV with the goals of a drug-free remission of viremia and, ultimately, virus eradication. To achieve this, therapeutics that target host restrictions to proviral expression that exemplify latent infection, such as epigenetic modifications of chromatin about the HIV promoter, or deficiencies of key host transcription factors within resting CD4 T cells, have been explored in laboratory models of latency, and emerging humanized mouse and non-human primate model systems.
Most clinical studies of ART intensification have thus far shown little effect on persistent infection or persistent viremia, but plans for further human studies are underway. However, as multiple molecular mechanisms appear to underlie the establishment and maintenance of persistent, latent HIV infection, combined approaches may ultimately be necessary to effectively purge residual HIV genomes.
Beyond these near-term efforts, renewed translational efforts seek to re-examine the possibility of transplanting HIV-infected patients with cells resistant to virus infection, alongside expanding laboratory studies to directly excise, damage, or silence proviral genomes. It is clear that to control the HIV pandemic, the scientific, medical, and pharmaceutical communities must marshal new technological and logistical approaches to sustain innovative efforts to prevent and treat HIV, and to work towards a cure for HIV infection.

I18

Translational research at acute HIV Infection

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Retrovirology 2010, **7(Suppl 1):**I18

The initial interaction between HIV and its host is crucial in determining disease progression and uniquely provides the ability to explore the determinants of HIV acquisition. Upon this first encounter, the virus will induce an innate and adaptive immune response that will determine the severity of symptoms and the level of T cell activation. To rationally design the optimal treatment strategy, whether it immunotherapies or ART, this brief time period is of the utmost importance. The challenge here lies in the conflict between the drive for scientific knowledge and supporting the patient though this emotional stage. However, only a delicate balance between these necessities will produce successful translational research. In addition, integrating virological, immunological and behavioural factors has become the sole valid approach of properly addressing both HIV pathogenesis and patient needs.
After two decades of HIV research, mucosa immunity has emerged as a cornerstone of future research. Several studies in both SIV and HIV infection have emphasized the importance of depletion of mucosal CD4 T cells and high levels of T cell activation immediately following infection. The contribution of mucosal immunity has shed light on the importance $\alpha 4\beta 7$ mucosal homing receptors, massive loss of memory CD4 T cells, microbial translocation, and T cell activation. Recently, dendritic cells, monocytes, NK cells as well as the interleukin-7 and interleukin-21 signalling pathways have been shown to contribute to the early immune dysfunction observed after infection. The timing of ART initiation plays a major role on the quality of immune recovery. In addition, the establishment of viral reservoir occurs within days after infection and its size is largely determined by the CD4 memory/effector ratio and CD4 cell nadir.
New frontiers in acute HIV infection research need to include blood sampling by leukapheresis to collect large quantities of immune cells, mucosa biopsies, and neurocognitive evaluation to pave the route toward HIV eradication and rational design of vaccines and immunotherapies.

I19

Use of new technologies to detect and understand HIV drug resistance

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New sensitive genotyping technologies can detect low level drug resistant variants that are often missed by standard methods. New technologies have the potential to help the field investigate important viral population characteristics which may impact antiretroviral treatment responses. Specific viral factors such as the number of viral variants with drug resistance mutations in the circulating quasispecies (resistance mutational load), genetic linkage (mutations within the same viral genome), and specific mutation patterns may all impact treatment response. The detection level for particular resistance mutations and the relative mutational loads required to predict virologic failure for different regimens requires better definition. The ability to identify mutation linkage may improve the prediction of virologic failure. It should be emphasized that not all low level resistant variants lead to virologic failure as other active agents in a regimen (with different genetic barriers) may suppress the resistant variant. New sensitive genotyping methods have limitations due to both the inability to extract a fully representative sample of all viral variants present in blood and by the intrinsic error rates of the enzymatic and detection processes employed. These limitations can lead to discordant results between different sensitive technologies. The impact of low level resistant variants on treatment response is likely a multifactorial process with mutation load, linkage, and the genetic barrier of the regimen all contributing. The development of new genotypic resistance technology will allow the field to examine how these factors interact and impact therapy. This paper will describe some of the new sensitive technologies to detect genotypic drug resistance and the important questions that need to be addressed to help improve their clinical utility.

I20

Resistance to new antiretrovirals (ETV, anti-integrases, entry inhibitors)

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Retrovirology 2010, **7(Suppl 1):**I20

HIV is extremely variable and newly arisen variants may be rapidly selected upon drug pressure. New antivirals are available, new therapeutic options are at hand for all patients, either naïve or multiexperienced. Individualization is the hallmark of new approaches to antiretroviral therapies. Knowledge of drug resistance is important to guide clinicians in devising treatment regimens and to optimally preserve later treatment options for patients.
This presentation will focus on the new technologies, methodologies, models and research tools to enhance detection and interpretation of HIV-1 drug-resistance. Particular attention will be dedicated to the resistance to the new antivirals.
The development of the new second-generation NNRTI etravirine, which has a higher genetic barrier than the first-generation drugs, has led to new understanding of the complexity of NNRTI resistance. To date, 17 mutations have been identified as being associated with etravirine resistance and reduced virological response.
Integrase inhibitors are very potent drugs providing a great opportunity for clinicians, to construct new regimens for individuals with HIV infection resistant to all three traditional classes of antivirals. However, these drugs have a relatively low genetic barrier to resistance and thus require both the selection of an optimised backbone to ensure durable viral suppression and a clear understanding of their residual activity in a failing integrase regimen. Resistance tests able to sequence and detect mutations in integrase are crucial for their proper use in clinical practice.
Viral escape of entry inhibitors may occur through 'classic' viral resistance to the compounds, but also by usage of alternative cell co-receptors. To date the main focus of research on co-receptor antagonists has been on determining and quantifying the CCR5, CXCR4, dual/mixed coreceptor tropism. Recent data begun to emerge on how resistance develops with the accumulation of mutations in gp120 to maraviroc, the first-in-class R5 co-receptor antagonist.

I21

Use of new resistance markers to predict virologic response to antiretrovirals

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Aim: There can now be little doubt that certain drug-resistance mutations are more likely to emerge in viruses of certain subtypes than others. Two of

the best and most practical examples of this are the emergence of the V106M NNRTI mutation and the K65R NRTI mutation in viruses of subtype C.
Methods: It is relevant that the use of newer more sensitive methods have been key in establishing the role that certain drug-resistance mutations may play in both transmitted and acquired drug resistance. In particular, the use of allele-specific PCR assays (AS-PCR) and pyro-sequencing have been very useful in providing new insights.
Results: Among the reasons that more ultrasensitive assays may sometimes be needed for more accurate assessments of drug resistance is the differential effect of certain mutations on viral replicative capacity. As an example, the K65R mutation is known to adversely affect HIV replication, and this may be one of the reasons that it is found relatively infrequently among individuals who fail antiretroviral therapy. In contrast, the use of AS-PCR for K65R in subtype C viruses has shown that this method was able to detect the presence of this mutation in an additional 4 of 30 samples who had tested negative by bulk sequencing methods. Now, it also appears as though the transmission of the K65R mutation, while rare, can also be detected in higher numbers by AS-PCR than bulk sequencing, and that this is also more common among subtype C than subtype B viruses. The likely reason is that subtype C viruses are more prone to develop K65R as a consequence of treatment failure and are therefore more likely than subtype B viruses to contain this mutation at the time that transmission takes place. In the case of the M184V mutation, it has also been observed that AS-PCR methods can detect this substitution more efficiently than bulk sequencing among newly-infected individuals.
Discussion: Thus, the reason that some resistance-associated mutations are not commonly observed in newly-infected subjects is not because they impact on the ability of HIV to be transmitted but rather because they may quickly revert to wild-type in the absence of drug pressure and then be rapidly overgrown by wild-type variants.

I22

HIV/HPV coinfection: state-of-the-art

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Retrovirology 2010, **7(Suppl 1):**I22

Aim: The risk of HPV-related cancers is higher among persons with HIV/AIDS, even under HAART treatment. This data prompted us to analyse HPV distribution, persistence and changes in HPV multiplicity of infections before and during antiretroviral treatment.
Materials and methods: Prevalence and persistence of mucosal HPV genotypes and HPV16 variants were analysed in a prospective cohort of HIV-positive and HIV-negative Italian women.
Results: HIV-positive women were more likely than HIV-negative women to be infected by HPV at the initial examination (39.3 vs 13.9%, $P < 0.001$) and to have a higher period-prevalence of HPV infection over a 3-year follow-up (43.8% vs 17.4%, $P < 0.001$), regardless of CD4+ cell counts and anti-retroviral therapy. 'High-risk' and 'probable high-risk' HPVs (types 16, 18, 31, 33, 35, 45, 52, 58 and 66), among the 20 different viral genotypes identified, were predominant in HIV-positive (33.9%) compared with HIV-negative (13.9%) women. Among HIV-infected women, with normal cytology as well as with SIL of any grade, the most common genotypes were HPV16 followed by HPV81, -58, -72, -33 and -62. HPV16 isolates from 18 HIV-positive and eight HIV-negative women were classified into variant lineages based on sequencing analysis of E6 and E7 genes and the long control region. Whilst the HPV16 G350 European variant was prevalent in both HIV-positive (10.7%) and -negative women (3.5%), HPV16 African 2 variant was only detected in HIV-positive women (3.6%), suggesting different sexual mixing behaviours.
Discussion: The high prevalence of HPV-related lesions in our cohort study, of HIV-positive patients under HAART-treatment, is consistent with the reported high standardized incidence rates (SIRs) of HPV-related in situ cervical (SIR 8.9, 95% CI = 8.0 to 9.9) and anal cancers (SIR 68.6, 95% CI = 59.7 to 78.4) as well as for invasive oropharyngeal (SIR 1.6, 95% CI = 1.2 to 2.1) and anal cancers (SIR 34.6, 95% CI = 30.8 to 38.8). The high prevalence of uncommon viral genotypes and HPV16 variants in HIV-positive women underscores the need to target a wide range of HPV types in cervical screening of HIV-positive women.

I23

Hepatitis B: treatment strategies and resistance

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More than 350 million people worldwide are chronically infected with HBV. The prevalence is particularly high in South East Asia, Sub-Saharan Africa and South America. In developed regions, the rate of chronic hepatitis B is declining mainly as result of universal HBV vaccine programs, although immigration from HBV endemic areas represents a continuous source of incident cases.
Viral load and HBV genotype influences the natural history of chronic hepatitis B and progression of liver fibrosis. Treatment must be considered for individuals with serum HBV-DNA above 2000 IU/ml, given their increased risk of cirrhosis and liver cancer. Non-invasive tools for assessing liver fibrosis, such as elastometry (FibroScan) are rapidly replacing liver biopsy to guide treatment decisions.
Besides peginterferon, five nucleos(t)ide analogues (lamivudine, adefovir, telbivudine, entecavir and tenofovir) have been approved for the treatment of chronic hepatitis B; it is unlikely that any more will be available in the near future. Cross-resistance is common between all these drugs, although in different extent. Therefore, an adequate design of effective therapeutic strategies is crucial for long-term benefit using these drugs. Patients that experience virologic breakthrough or partial response to their primary therapy can often be successfully treated with a second nucleos(t)ide analogue, preferentially when the rescue intervention is done at early stages of failure. However, best strategies for preventing drug resistance include first-line use of the most potent antivirals with a high barrier to resistance (namely tenofovir or entecavir) and/or combination therapy. At this time, the use of lamivudine or adefovir as single agents in first-line therapy is no longer recommended, given their low-genetic barrier to resistance and weak activity, respectively.
The genes that encode the polymerase and envelope proteins of HBV overlap, so resistance mutations in polymerase usually affect the hepatitis B surface antigen; these alterations affect infectivity, vaccine efficacy, pathogenesis of liver disease, and transmission throughout the population. Primary lamivudine resistance-associated mutations in HBV are now being seen in 10% of new HBV infections in Western Europe and North America.
A particular considerations merit the coinfection with HIV or the superinfection by delta virus. Specific guidelines for managing HBV in HIV+ individuals have been released (AIDS 2008, 22:1399–1410). The fixed combination of tenofovir plus emtricitabine (Truvada) is the preferred option in most HIV-HBV coinfected patients. The role of entecavir or telbivudine in this setting is hampered by the recognition of low anti-HIV activity and possibility of selecting resistance mutations in HIV. With respect to hepatitis delta, preliminary data suggest that potent nucleotide analogues as tenofovir may exert some benefit in a subset of individuals with chronic hepatitis delta, although data are too preliminary.

I24

New anti HCV drugs in the pipeline

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Retrovirology 2010, **7(Suppl 1):**I24

Several novel therapeutic approaches are currently in the pipeline for HCV treatment. They include new target host immune system as novel interferon, toll-like receptor agonists, therapeutic vaccines or interleukins, new target cell replication drug as ribavirin analogues, cyclophilin inhibitors, alpha glucosidase inhibitors, dephosphorylationinhibitors of eukaryotic initiation factors 2alpha, antisense molecule, entry inhibitors

and specifically targeted antiviral therapy (STAT-C) such as viral enzyme inhibitor (protease and polymerase). Several STAT-C molecules are currently in development and will soon be at hand and will offer new treatment opportunities to patients infected with hepatitis C. Promising results leading to more than 70% of sustained virological response in genotype 1 naïve patients have been reported with two proteases inhibitors (telaprevir and boceprevir) in combination with pegylated interferon and ribavirin that are currently in phase III. These studies also demonstrated the potential to shorten treatment duration in those with a rapid viral response and the realistic hope for retreatment success (40 and 75%) in previous non responders or relapsers to interferon based therapies. In addition there is early indication that STAT-C drugs may help to overcome negative host factors that have historically been associated with poor response rates (such as ethnicity, insulin resistance, steatosis and cirrhosis). However these trials also emphasize the limitations of protease inhibitors and viral resistance data have provided important new lessons for small molecule drug development. In the near feature, it is likely that IFN-based therapy plus ribavirin will remain the backbone of the treatment of chronic hepatitis C. PEG-IFN and ribavirin are needed in order to prevent HCV resistance to STAT-C drugs and subsequently increase SVR. Genotypic and phenotypic resistance tests will also enter the therapeutic arena. Once several STAT-C agents without cross resistance become available, treatment strategies will include a combination of several drugs with different mechanisms of action (protease inhibitors and polymerase inhibitors) that could hopefully results in IFN and/or ribavirin sparing regimen. The first dual – combination clinical trial with oral antiviral is still ongoing. Patients receiving this combination for 14 days had undetectable HCV RNA in 63% of naïve genotype1 patients. This "early success" has moved IFN-free regimen a step closer to reality for patients. In the future, there might be combinations of antivirals having additive potency, lacking cross resistance and with a good safety profile.

I25

Liver transplantation in HIV-1-infected patients

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With the advent of highly active antiretroviral therapy in 1996, patients infected with HIV are now living longer and dying from illnesses other than acquired immunodeficiency syndrome (AIDS). Liver disease due to chronic hepatitis B and C is now a leading cause of mortality among HIV-infected patients in the developed world. For these patients, liver transplantation (OLT) is the only therapeutic option and HIV infection alone is not a contraindication. The current HIV selection criteria for HIV-infected OLT candidates are as follows: 1) ideally no history of opportunistic infections or HIV-related cancer, although some treatable and preventable opportunistic infections are not exclusion criteria; 2) CD4 cell count >100 cells/mm^3; and, 3) plasma HIV RNA viral load that is undetectable or can be suppressed with antiretroviral treatment. Drug users must abstain from heroin and cocaine, although patients can be in a methadone programme. Accumulated experience in North America and Europe in the last few years indicates that five-year survival in liver recipients coinfected with HIV and HCV is lower than that of HCV-monoinfected recipients, being the five-year survival of around 50%. Conversely, 3-5–year survival of non-HCV-HIV-coinfected liver recipients is very good and it was similar to that of HIV-negative patients. Pharmacokinetic (PK) and pharmacodynamic interactions between NNRTI- or protease-inhibitor based regimens and immunosuppressors have been one of the most important problems in the post-trasplant period, although with the new NNRTI- and protease inhibitor-sparing raltegravir-based regimens we can avoid them. Other problems in the post-transplant period are the high rates of acute rejection, and the HCV re-infection in HIV-infected liver recipients, that is the main cause of mortality. Better anti-HCV management and therapy could improve the long-term outcome of OLT recipients coinfected with HIV and HCV.

I26

HIV controllers: state of the art

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HIV controllers are rare chronically HIV-1-infected patients in whom viral replication is undetectable in the absence of antiretroviral treatment. Most such patients are nonetheless infected by replication-competent viruses. An effective multifunctional HIV-specific CD8+ T cell response and functional CD4 T cells are thought to be central to viral control in these individuals. The mechanisms underlying this spontaneous control of HIV infection are the focus of intensive investigations, as they should help to unravel the pathogenesis of AIDS and to provide new clues for the design of effective vaccine strategies. In this review we examine recent findings from these studies.

I27

The role of cytotoxic T cells

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Introduction: The development of anti-HIV T cell-based vaccines is a current major objective in the strategy to halt AIDS pandemic. For this purpose the understanding of the mechanisms underlying effective HIV-specific CD8+ T cell responses is of great importance. One of the most appealing models for such efficient responses is found today in HIV controllers (HICs), rare individuals able to control HIV infection to undetectable levels for more than ten years in the absence of therapy.
Results and discussion: Despite very low levels of antigen in blood, most HICs have high frequencies of HIV-specific CD8+ T cells that preferentially target the viral Gag protein. Studies of CD8+ T cell responses in HICs have revealed important characteristics of functional HIV-specific CD8+ T cells in HIV infection. Contrary to cells from viremic individuals, HIV-specific CD8+ T cells from HICs can, upon stimulation with their cognate antigen, proliferate and generate a multifunctional response. This could be related to a peculiar (HLA-DR+CD38-) activation phenotype of these cells and to constitutive telomerase activity that protects them against senescence. Our lab has recently shown that CD8+ T cells from most HICs are endowed with a striking capacity to suppress HIV infection ex vivo through a cytotoxic mechanism, a property that is likely to be relevant in vivo. This is likely related to a higher capacity of HIV-specific CD8+ T cells from HICs to upregulate perforin and granzyme. Extending this observation, we have found that HIV-suppressive capacity of CD8+ T-cells is strongly correlated to the frequency of HIV-specific CD8+ T-cells in HIV controllers (but not in viremic individuals), and in particular to the frequency of Gag-specific CD8+ T-cells. Actually, the depletion of Gag-specific CD8+ T-cells but not other specificities abrogates HIV suppression, suggesting that not all the cells in HICs have the same anti-HIV potential.

I28

Potential inflammatory consequences in HIV controllers

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Retrovirology 2010, **7(Suppl 1):**I28

Aim: Most HIV controllers have strong HIV-specific T cell responses that likely contribute to viral control. However, there may be negative

inflammatory consequences to the immunologic control of viral replication in these individuals.

Materials and methods: We compared the frequency of activated (CD38+ HLA-DR+) T cells and carotid intima-media thickness (a measure of atherosclerosis) between untreated HIV controllers maintaining plasma HIV RNA levels <75 copies/ml and HIV-infected ART-suppressed, untreated HIV-infected "non-controllers," and HIV-uninfected controls. We also assessed the relationships between the frequencies of Gag-specific and activated T cells, and cell-associated HIV RNA and DNA levels in HIV controllers.

Results: The 52 HIV controllers had higher frequencies of activated CD4+ and CD8+ T cells than HIV-uninfected controls ($P < 0.001$ for both) and higher CD8+ T cell activation than the ART-suppressed ($P = 0.017$). HIV controllers also had higher carotid intima-media thickness than HIV-uninfected individuals even after adjustment for traditional cardiac risk factors ($P = 0.003$). In HIV controllers, higher CD4+ and CD8+ T cell activation was associated with lower CD4 counts in HIV controllers ($P < 0.001$ for both). While HIV controllers had the highest frequencies of Gag-specific CD4+ T cells of any group, suggesting a role in the control of viral replication, higher Gag-specific CD4+ (but not CD8+) T cells were associated with both higher CD8+ T cell activation ($P < 0.001$) and higher cell-associated HIV DNA levels ($P = 0.019$).

Discussion: Strong HIV-specific CD4+ T cell responses in HIV controllers may assist in the control of viral replication, but may also contribute to viral persistence and generalized immune activation, which may drive both CD4+ T cell depletion and subclinical cardiovascular disease even in the absence of clinically detectable viremia.

I29

Progeria, a model for accelerated aging exhibited by HIV patients?

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Aim: To confirm, among HIV1-infected patients, data from *in vitro* studies showing that antiretroviral therapies (ART) induce an accelerated aging through the same mechanism than genetic laminopathies (progeria) and « physiological » aging, *i.e.* through the synthesis and persistence of farnesylated prelamin A. The perspective is to minimize ART side effects using the same drug combination yet given to treat progeria children in Marseille.

Materials and methods: A multicentric (Marseille, Nice and Montpellier Hospitals) 3 year-long study will analyse 50 HIV1-infected patients without any ART (A group), 100 infected patients receiving ART for at least 12 months (B group) and 50 age- and sex-matched seronegative control subjects. Infected patients will be submitted to 4 successive investigations (M0, M12, M24 and M36).

Biological tests are performed in *Timone Hospital labs* (Marseille): i/ viral load, PBMC isolation, DNA extraction, proviral DNA measurement [*Virology*]; ii/ CD4, CD8, glycemia, insulinemia, HOMA, total-, LDL- and HDL-cholesterol, triglycerides [*Biochemistry labs from the 3 Hospitals*]; iii/ ART assay [*Pharmacokinetics Lab*]; iv/ detection (western blot, immunocytochemistry) of PBMC nuclear, cytosolic and mitochondrial ART targets: A and B lamins, NF-kB and I-kB (proteasome activity), CD36 (glycosylation), mitochondrial Hsp70, ROS production, inner membrane potential, cytochrome C oxidase subunits 2 and 4 [*Cell Biology*]; v/ genotyping the ART targets: prelamin A and B processing proteases, Golgi SREBP-releasing proteases, mitochondrial deoxynucleoside transporters and proteases involved in nuclear-encoded protein import; telomere length [*Molecular Genetics*]. *CIC-UPCET* collaborated to the protocol design, recruits control subjects and is in charge of data statistical treatment.

Results and discussion: The M0 collection just finished. Mitochondrial data will be presented.

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I30

Accelerated immune senescence in HIV infection

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Aim: The prospect that immune activation or inflammation may be directly related to the increased incidence in HIV infected donors of manifestations that are reminiscent of the human aging process (i.e. cardiovascular disease, malignancies, osteoporosis, cognitive impairment, depression and frailty) is raising increasing concerns. On the immunological side, in addition to promoting viral replication as well as $CD4^+$ T cell apoptosis, HIV associated immune activation may also lead to an accelerated decline of immune competence resembling the phenomenon of immunosenescence. Our aim is to explore further the potential relationship between immune activation, HIV disease progression and immunosenescence.

Materials and methods: We performed a comparative analysis of immunologic markers in different groups of HIV infected donors and healthy controls. The initial investigation of blood lymphocyte populations led us rapidly to turn our attention onto the lymphocyte primary immune resources, i.e. the $CD34^+$ hematopoietic progenitor cells (studied directly from the blood of patients).

Results: Human aging and HIV-1 infection exhibit a number of parallels with regards to immunological attributes, that are evocative of premature immunosenescence in HIV-1 infected patients and reflect a reduced production of lymphocytes. Analyses of $CD34^+$ hematopoietic progenitor cell number, phenotype and clonogenic potential underline a manifest impairment of primary immune resources with age or HIV-1 infection. Systemic immune activation emerges as a major correlate of altered lymphopoiesis, which can be partially reversed with prolonged antiretroviral therapy. Poor $CD4^+$ T cell count recovery despite successful virological response on antiretroviral treatment is associated with persistent damage to the lymphopoietic system.

Discussion: Our findings provide new insights into the consequences of persistent immune activation in HIV-1 infection, and demonstrate the importance of primary hematopoietic resources in HIV pathogenesis and the response to antiretroviral treatments, but also more generally in the development of immunosenescence.

I31

Bone ageing and HIV

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Retrovirology 2010, **7(Suppl 1):**I31

As HIV-infected patients live longer they are experiencing diseases normally associated with ageing, sometimes at higher frequency and at younger ages than expected. Low bone mineral density (BMD) and osteoporosis is one example. Rates of low BMD are high in HIV-infected patients and fracture rates occur more commonly and at a younger age than the general population. Factors thought to contribute to low BMD in HIV include HIV infection itself, exposure to antiretroviral therapy (ART) and over-representation of traditional risk factors (such as smoking, hypogonadism and low body mass index) for low BMD in HIV-infected populations. Current evidence suggests that patients lose BMD after ART initiation and, to some extent, with switch in ART. Rates of BMD loss appear higher when protease inhibitors or some NRTI are used as part of the initial ART regimen. This presentation will outline current data on low BMD in HIV, discuss ways of monitoring for low BMD and current approaches to management of this common condition.

I32

Towards a vaccine against AIDS

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Aim: The CD4 depletion in the chronic phase of HIV infection is mostly due to the loss of uninfected cells. We previously showed that this lost was related to the expression of NKp44L, a cellular ligand of the natural cytotoxicity receptor NKp44, which renders CD4 cells sensitive to NK killing. NKp44L is specially induced by the highly conserved 3S motif of the HIV-1 gp41 envelope protein. We also have shown that NKp44L is present on bystander non-infected CD4 cells, but absent from HIV-infected cells, through a Nef-dependent mechanism. In this study we sought to determine whether the loss of uninfected bystander CD4 cells could be prevented by a 3S therapeutic immunization in a macaque model chronically infected with SHIV162P3.
Methods: Ten cynomolgus macaques were chronically infected with SHIV162P3. Seven months after infection, the animals were primed/boosted in IFA with 3S-KLH or free KLH, as control. Lymphocyte samples and sera were periodically tested, while secondary lymphoid organs after euthanasia, at 7 months post-immunization.
Results: We discovered that immunization with 3S-KLH, significantly decreases of NKp44L expression on CD4 cells, and NK cells cytotoxicity against autologous CD4 cells, when compared to infected group with free KLH-immunization. Interestingly, the frequency of CD4 central memory T cells from immunized animals remains stable, while decreasing in the control group. Finally, in lymphoid organs, including spleen, lymph node and gut, a significant decrease of the cell-activation and the caspase-3 dependent apoptosis was observed in macaques immunized by 3S-KLH, as compared to control.
Discussion: These results emphasize the deleterious role of NK cells on CD4 depletion and demonstrate for the first time, its prevention by a therapeutic vaccine, which should also inhibits and/or delay disease evolution to AIDS.

KEYNOTE PRESENTATION

K1

25 years after discovering HIV as the cause of aids: prospects for a vaccine

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Live attenuated HIV vaccines cannot be tested in humans because of their danger. Inactivated virions are still dangerous and also poorly immunogenic. For that reason subunit vaccines in one form or another have been the approach of almost all investigators since 1984 when adequate HIV culture systems became available.
With the exception of using small animals for testing immune responses all in vivo experimental vaccine testing research utilize subhuman primates, especially macaques, challenged with SIV or the chimeric SHIV (using an HIV envelope with SIV). Thus, we begin with two handicaps – a limitation to subunit vaccines and a limitation in our animal model. However, the limits of the animal model in my view are not due to any inadequacy of the model but rather the limitation of monkeys. This has been a problem from the beginning, but is one that could have been overcome long ago with adequate investments from funders. Instead, we witness most quality investigators limited in their experiments to 4 to 6 animals per group, so as to require multiple repeated experiments and years of delay, while a few groups can afford 20 to 30 animals per group. Obviously, this needs to change. A third handicap is HIV variation. Generally, investigators have challenged macaques with homologous SIV (or SHIV), but this is inadequate. However, in recent years the need for heterologous viral challenges has finally been recognized. It would be ideal to challenge with several different heterologous challenges, but there are serious limitations in available primate viral stocks, particularly SHIV. The best protocol for testing a candidate vaccine in macaques is controversial. Many have favored a single relatively high dose challenge, while recently several groups argue for multiple low dose challenges. The fact is arguments can be made for either, and no one knows which one will be more predictable for a HIV vaccine in man.
By far the greatest hurdle to an effective vaccine is the retrovirus characteristics of HIV, namely its capacity to rapidly integrate its genes into our DNA. This establishes life-long infection, rapid diminished function of immune responses, and the emergence of variants. This feature has major implications all too often forgotten in the 1984 to 2008 period of HIV vaccine research, namely an effective HIV vaccine: (1) must protect against infection not just reduce HIV after infection; (2) must be long lasting; and (3) must be broad.
Early in HIV vaccine history (roughly 1984-1990) the candidate immunogen was focused on the gp120 envelope delivered as protein, DNA, or vector with an HIV env gene insert. Mostly these led to type specific immune responses so vaccines of this type failed in the monkey model when challenges were made with heterologous strains of SIV or SHIV, especially with protein alone. Nonetheless, a company (VaxGen) still went forward with a gp120 protein clinical trial, and all too predictably, completely failed, and caused much negativity for HIV vaccine research.
Many investigators then turned to CMI based vaccines. Though CMI is important and likely helpful to any vaccine candidate, CMI alone is a predictable failure, and indeed such vaccines have failed (e.g., recent NIAID – Merck trials in Africa).
We and some other groups think the answers will be in finding, "fixing," and properly presenting *conserved* sequences of gp120 which are *functionally* required for HIV infection. Finding conserved sequences is an obvious need and a readily achievable one. "Fixing" means making a *constrained* envelope because the mobile envelope is a difficult and ever changing target. We have chosen the CCR5 binding domains of gp120 as the conserved *and functionally necessary* region. This region of gp120 contains novel epitopes called CD4i epitodes and Abs to them are known as CD4i Abs. We approach this by linking gp120 to domains of CD4 that bind gp120. We have developed a candidate vaccine based on inducing CD4i Abs. These Abs are often broadly reactive, and although we have not as yet reached sterilizing immunity, the vaccine ultimately protects against a high dose heterologous challenge and often has ADCC activity [1].
From our primate experiments and our studies in human elite viral suppressors [2] *we predict that a successful HIV vaccine* may consist of an immunogen that induces CD4i Abs and ADCC type responses.

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ORAL PRESENTATIONS

O1

Evolution of tuberculosis/HIV co-infection in California during the HAART Era, 1996-2007

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Background: California reports the highest annual number of tuberculosis (TB) cases and over 12% of all persons living with HIV in the US. To assess changes in the intersection of these diseases, we analyzed state-wide data from both registries.

Methods: Incident cases reported to the California TB registry from 1996 to 2007, inclusive, were cross-matched with the state HIV/AIDS registry. Incidence rates of TB with and without HIV co-infection were analyzed for changes in trend. Sociodemographic, clinical characteristics, and treatment outcomes were examined.
Results: 2,392 (6%) of 39,718 TB cases during the study period were matched in the state AIDS registry. From 1996 to 2006, annual TB incidence among persons with HIV declined from 368/100,000 to 73/100,000, and without HIV declined from 12.7/100,000 to 7.3/100,000. Comparing the period 1996-2000 with 2001-2007, the proportion of TB/HIV cases increased among Hispanics (48% to 57%; $p < .001$), foreign born (46% to 63%; $p < .001$), and women (14% to 18%; $p = .06$), and decreased among Blacks (28% to 21%; $p < .001$). Male-to-male sexual contact (44% to 41%; $p < .01$) and injection drug use (21% to 15%; $p < .01$) decreased, and high-risk heterosexual contact (9% to 16%; $p < .01$) increased as HIV transmission routes. 77% of all foreign born patients with TB/HIV co-infection originated from Mexico or Central America; the median time from immigration to TB diagnosis was 10.7 (IQR 4-19) years. CD4 count at TB diagnosis (median 78 (IQR 30-167)) was available for 54% of patients. Patients with HIV co-infection were more likely to be sputum smear-positive (OR 1.17 (95% CI 1.08-1.28). Both isoniazid resistance (5% to 7%; $p = .07$) and pyrazinamide monoresistance (6% to 8%; $p = .01$) increased over time in TB/HIV cases but not in cases without HIV. In multivariate analysis, Hispanic ethnicity, older age, and injection drug use were inversely associated with treatment success among TB/HIV cases.
Discussion: In California, the epidemiology of TB/HIV co-infection in the post-HAART era has shifted towards persons of foreign birth, women, Hispanics, and those who acquired HIV through heterosexual sex. In addition, drug resistance has increased in TB/AIDS cases. These changes should be considered in focusing TB and HIV disease prevention and treatment efforts.

O2

Abstract withdrawn

Retrovirology 2010, **7(Suppl 1):**O2

O3

Screening of differentially expressed gene in HIV/HCV co-infected patients

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Background: Most research has shown that HIV/HCV coinfected people tend to experience faster liver disease progression and develop more AIDS-defining illnesses than mono infected patients. The objective of this study was to screen the differentially expressed genes in HIV/HCV co-infected patients, and further investigate its possible mechanism.
Methods: Eligibility Criteria including 20-40 year-old male, similar social-economic background, treatment naive, same transmission route methods (Sex or Injection drug), and CD4 count >200 cells/mm^3. All research objects signed on informed consent. The project proceeded after approval of the local REC.
Human blood samples were collected from 13 HIV/HCV coinfected patients, 8 mono infected patients, and 8 normal control. At first, CD4 and CD8 positive T cells were separated with magnetic activated cell sorting, then followed by RNA extraction. After reverse transcripted to cDNA with anchor primer and amplified with random primers, the products were run on PAGE electrophoresis followed by silver stain. The bands with different expression level were extracted from gel and cloned for sequencing. The expression changes were confirmed by SYBR Green real-time quantitative PCR.
Results: Totally 307 differentially expressed bands were extracted. 25 fragments were sequenced and found 3 for ring-finger protein 141(RNF141), 5 for cyclic AMP phosphoprotein (ARPP-19), 5 for mitochondrial cytochrome (cytochrome c and b-245). Most of the other fragments were function unkown sequences located on X chromosome and mitochondrial. The change of expression of cytochrome c and ARPP-19 were confirmed with a significant down-regulation in HIV/HCV coinfected patients and a slight down-regulation in mono-infected group compared to negative population.
Discussion: 1. The expression of RNF-141, ARPP-19, cytochrome c and b-245 mRNA are probably associated with HIV/HCV co-infection.
2. The technique of mRNA differential display together with SYBR Green quantitative real-time PCR is useful for screening for differentially expressed genes in HIV/HCV co-infection.
3. Since mitochondrial cytochrome has been reported to associated with prognosis of AIDS. Is it possible to act as a useful marker for prognosis of HIV/HCV coinfection? More clinical data are needed.

O4

NK-pDC cross-talk leads to the generation of mTRAIL+IFN-α+cytotoxic pDCs following HIV-1 infection of pDCs. Consequences on the control of viral replication

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Background: Plasmacytoïd dendritic cells (pDCs) play a central role in orchestrating innate and adaptive immunity, especially by secreting large amounts of IFN-α in response to viral stimuli. IFN-α production by pDCs depends upon a cross-talk with NK cells that triggers the activation of NK cells. Considering that NK cells are essential for the elimination of virus-infected cells, and pDC required for their antiviral activity, we addressed the impact of HIV on NK-pDC cross-talk, and the consequences on viral control.
Methods: pDCs and NK cells were sorted with magnetic beads from the blood of healthy donors. NK cells were kept either resting or activated with PMA/ionomycine for 2 hrs. pDC were stimulated with either IL-3 or CpG, or were infected with R5-HIV-1 at various concentrations. pDCs were cocultured with NK cells at various ratios for 24 hrs. The influence of NK-pDC interaction on both cell types was analyzed by multiparametric flow cytometry, combining maturation and cell death/survival markers with cytokine detection, and released cytokines in cultures supernatants were identified and quantified by the MAP luminex technique.
Results: Our results show that high amounts of HIV-1 induced the maturation of pDCs, characterized by the expression of HLA-DR, CD40, CD80, CD86, CCR7 and CD83. This phenotypic maturation was coupled to a functional maturation since HIV-1-infected pDCs were able to activate the production of IFN-γ and TNF-α by NK cells after 24 hrs of co-culture. Consequently NK cells induced, in synergy with the virus, the emergence of killer mTRAIL+IFN-α+ pDCs. In contrast, NK-pDC cross-talk did not induce pDC maturation at low concentration of HIV, and it had a poor effect on the activation of NK cells. Finally, the release of β-chemokines and IFN-α was found dependent both on NK-pDC cross-talk and HIV concentration.
Discussion: We report for the first time that NK-pDC cross-talk induces the maturation and differentiation pDCs into mTRAIL+ IFN-α+ cytotoxic pDCs once infected with HIV-1. Furthermore, depending on virus concentration, NK-pDC was found involved in the viral control, either by triggering or by suppressing the release of anti-HIV molecules, such as β-chemokines and IFN-α.

O5

Th1/Th17 gammadelta T cells are expanded in HIV-1 infected patients and respond to *Candida albicans*

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Background: Circulating Vdelta2 T lymphocytes are involved in the response to mycobacteria and certain viruses, while Vdelta1 T cells,

resident in the mucosal tissues, participate in the immunity against intracellular microrganisms. Vdelta2 T cells recognize non-peptidic phosphorylated antigens expressed by mycobacteria, whereas Vdelta1 T cells interact with MHC-related molecules (MICA, MICB) and with receptors for the UL-16 protein produced by CMV-infected cells. Vdelta1 T cells release IFNgamma upon challenge with $MICA^+$ cells, while Vdelta2 T cells secrete this cytokine upon stimulation with phosphate antigens. We reported that in early HIV-1 infection Vdelta1 T lymphocytes, producing IFNgamma, are increased in the peripheral blood. We addressed the question of whether this T cell subset can also be involved in the response to fungal infections.

Methods: Thirty untreated HIV-1-infected patients were studied, compared to ten healthy subjects. Patients were staged according to the Center for Disease Control criteria. Serum HIV-1 RNA was quantitated and CD4 count performed. Cytokine production was determined by intracytoplasmic immunofluorescence and ELISA. Proliferation of Vdelta1 and Vdelta2 T cells to C. albicans, PPD, CMV and P. Carinii was determined by flow cytometry after CFSE staining. Gene expression was evaluated by Q-RT-PCR.

Results: We show that: 1) a population of circulating Vdelta1 T lymphocyte producing both IFN-gamma and IL-17 is expanded in HIV-1 infected patients; 2) this population is capable of proliferating and enhancing cytokine production in response to Candida albicans, while Vdelta2 T cells respond to mycobacterial antigens; 3) IFN-gamma/IL-17 double producers express the RORC and the TXB21 transcription factors, the CCR7 homing receptor, the CD161 molecule involved in transendothelial migration, and the CCR4 and CCR6 chemokine receptors.

Discussion: In HIV-1 patients, gammadelta T cells not only produce Th1/Th17 cytokines, but express a number of homing and chemokine receptors, thus being equipped for recirculation through lymph nodes and peripheral tissues. This circulating memory gammadelta T cell subset might play an important role in the control of HIV-1 spreading and in the defence against opportunistic infections, possibly contributing to compensate the impairment of $CD4^+$ T cells.

O6

NK-dependent survival of HIV-1 infected DCs. Pivotal role of HMGB1

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Background: Dendritic cells (DCs) are professional antigen-presenting cells that form cellular networks surveying for pathogens and providing the first immunological barrier to the external environment. The fate of DCs is dependent on a cross-talk with NK cells that may lead to DC's killing (editing) which is believed to keep in check their quality and quantity. Considering that HIV-1-infected DCs may become persistent viral reservoirs, we addressed the question of NK's role in infected DC's elimination as well as the mechanisms involved.

Methods: Immature DCs (iDCs) were derived from CD14+ monocytes cultured for 6 days in the presence of IL-4 and GM-CSF. iDCs were infected with R5-HIV-1BAL (DC_{HIV}). 24 h cocultures with autologous purified activated NK cells (aNK) were performed and DC's apoptosis was analyzed by multiparametric flow cytometry, combining 7-AAD staining with the detection of death/survival molecules. Gene array analyses were performed to detect variations in gene expression between different coculture conditions. siRNA magnetofection was performed to silence c-FLIP and c-IAP2 anti-apoptotic genes' expression in DCs. Live video microscopy was used to dissect apoptotic events during aNK-iDC contact.

Results: We show that, while iDCs were susceptible to NK editing, involving TRAIL/DR4 and not the perforin-pathway, DCHIV were resistant to NK-dependent cytotoxicity. We report that NK cells induce in DC_{HIV} a dramatic increase in the expression of two anti-apoptotic molecules, c-FLIP and c-IAP2, responsible for the resistance of DCHIV to TRAIL-induced apoptosis. Moreover, we found that HMGB1, a key mediator of NK-DC crosstalk, is responsible for the upregulation of these two inhibitors. The consequence of the escape of DC_{HIV} from NK cytotoxicity is an HMGB1-dependent increase in HIV replication in DCs, which is mediated by HMGB1.

Discussion: These observations show that under physiological conditions, the editing process of iDCs by NK cells occurs through rapid induction of TRAIL apoptosis in iDCs. Following HIV infection of DCs, NK cells increase DCs' survival through an HMGB1-dependant mechanism inducing c-IAP2 and c-FLIP upregulation. This study provides new insights into how HIV hijacks DCs and uses the NK-DC crosstalk to maintain viability of long-term reservoirs, and it identifies potential therapeutic targets to eliminate infected DCs.

O7

γδT cells are ADCC effectors in elite HIV controllers

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Background: Elite controllers have immune responses capable of modulating HIV replication. Our goal is to understand the role for gamma/delta T cells in controlling HIV. These rare individuals maintain undetectable plasma virus loads with few or no signs of disease progression despite not using antiretroviral therapy. High frequencies of "protective" MHC haplotypes implicated Class I-restricted cytotoxic T lymphocytes as one mechanism for virus control but this cannot explain the majority of individuals who do not carry these MHC alleles. Neutralizing antibodies are generally absent among elite controllers, but serum antibodies are active in antibody-dependent cellular cytotoxicity (ADCC) against target cells decorated with HIV Env glycoprotein. ADCC requires Fc receptor-expressing, cytotoxic effector cells to mediate destruction of HIV-infected targets; the nature of ADCC effectors has not been described for elite HIV controllers.

Methods: Cellular immunology studies with PBMC from elite controllers (designated by us as natural virus suppressors) and matched uninfected controls.

Results: We noted previously (Riedel, et al., AIDS 23:1955, 2009) that elite controllers have levels of gamma/delta T cells higher than matched controls, in sharp contrast to extensive depletion of this subset expected for individuals with HIV infection and disease. The Vgamma2Vdelta2+ subset is highly activated in elite controllers; these cells express CD56, a marker for cytotoxicity, and CD16, the Fc gamma receptor IIIa that is used for ADCC. In vitro studies demonstrated potent ADCC against Env-decorated cell targets using human monoclonal antibodies with expanded gamma/delta effector cells from elite controllers.

Discussion: A distinguishing feature among elite controllers is the preservation of activated Vgamma2Vdelta2+ T cells capable of mediating ADCC. These studies encourage the development of immunotherapies to activate gamma/delta T cells, enhance the effector component of ADCC, promote virus clearance and slow disease progression. The combination of ADCC antibodies and potent effector cells corresponds to durable control of viremia and disease. Early depletion of gamma/delta T cells in HIV disease may eliminate an important subset needed for ADCC and, despite strong antibody responses, allow HIV to persist with progressing disease.

O8

CpG methylation controls reactivation of HIV from latency

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Background: DNA methylation of retroviral promoter and enhancer localized in the provirus 5′ long terminal repeat (LTR) is considered to be a mechanism of transcriptional suppression that allows retroviruses to evade host immune responses and antiretroviral drugs. However, the role of DNA methylation in the control of HIV-1 latency has never been unambiguously demonstrated, in contrast to the apparent importance of transcriptional interference and chromatin structure, and has never been studied in HIV-1-infected patients.
Methods: We analyzed the relation of latent and reactivated HIV-1 promoters in a model of Jurkat cell lines and in memory $CD4^+$ T cells of long-term aviremic patients by means of bisulfit sequencing and chromatin immunoprecipitation in cell-sorted populations. To assess the resistance of latent HIV-1 to reactivation we exposed the cells to TNF-α, protein kinase C agonists, inhibitors of HDAC, and inhibitors of DNA methyltransferases.
Results: We show in an in vitro model of reactivable latency and in a latent reservoir of HIV-1-infected patients that CpG methylation of the HIV-1 5′ LTR is an additional epigenetic restriction mechanism, which controls resistance of latent HIV-1 to reactivation signals and thus determines the stability of the HIV-1 latency. CpG methylation acts as a late event during establishment of HIV-1 latency and is not required for the initial provirus silencing. Indeed, the latent reservoir of some aviremic patients contained high proportions of the non-methylated 5′ LTR. In the latent reservoir of HIV-1-infected individuals without detectable plasma viremia, we found HIV-1 promoters and enhancers to be hypermethylated and resistant to reactivation, as opposed to the hypomethylated 5′ LTR in viremic patients. However, even dense methylation of the HIV-1 5′ LTR did not confer complete resistance to reactivation of latent HIV-1 with some histone deacetylase inhibitors, protein kinase C agonists, TNF-α, and their combinations with 5-aza-2deoxycytidine: The densely methylated HIV-1 promoter was most efficiently reactivated in virtual absence of T cell activation by suberoylanilide hydroxamic acid.
Discussion: The latency controlled solely by transcriptional interference and by chromatin-dependent mechanisms in the absence of significant promoter DNA methylation tends to be leaky and easily reactivable. Tight but incomplete control of HIV-1 latency by CpG methylation might have important implications for strategies aimed at eradicating HIV-1 infection.

O9

Morbidity associated with hepatitis E virus infection in endemic setting

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Background: To determine the groups most affected by hepatitis E virus (HEV) during documented acute viral hepatitis (AVH) epidemics, trends in AVH-associated mortality rate (MR) per 100,000 over a 35-year period were examined.
Methods: Reported AVH incidence data from 1971 to 2005 and AVH-associated mortality data from 1981 to 1995 were examined. Serologic markers for infection with hepatitis viruses A, B, D, and E were determined from a sample of hospitalized patients with AVH from an epidemic period (1987) and from a sample of pregnant women with AVH from a non-epidemic period (1992).
Results: Two multi-year AVH outbreaks were identified: one during 1975-1976, and one during 1985-1987. During 1985-1987, AVH-associated MRs were 12.3-17.8 for the general population. Highest AVH-associated MRs occurred among children in the first 3 years of life (40-190 per 100,000) and among women aged 20-29 (15-21 per 100,000). During 1988-1995 when reported AVH morbidity was much lower in the general population, AVH-associated MRs were markedly lower among these same age groups. In 1988, AVH-associated MRs were higher in rural (21 per 100,000) than in urban (8 per 100,000) populations (RR 2.6; 95% CI 1.16-5.93; $p < 0.05$). Serologic evidence of acute HEV infection was found in 280 of 396 (71%) patients with AVH in 1987 and 12 of 99 (12%) pregnant patients with AVH in 1992.
Discussion: In the absence of the availability of confirmatory testing, inferences regarding probable hepatitis epidemic etiologies can sometimes be made using surveillance data, comparing AVH incidence with AVH-associated mortality with an eye to population-based viral hepatitis control measures. Data presented here implicate HEV as the probable etiology of high mortality observed in pregnant women and in children <3 years. High mortality among pregnant women but not among children <3 years has been observed in previous descriptions of epidemic hepatitis E. The high mortality among younger children observed in an outbreak of acute viral hepatitis associated with hepatitis E merits corroboration in future outbreaks.

O10

Hepatic cytolysis and Hepatitis E Virus infection in HIV-positive patients

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Background: Hepatitis E is an emerging infection in developed countries and progression to chronic hepatitis has been recently reported in some organ transplant recipients. The prevalence and evolution of hepatitis E in HIV-infected patients are unknown. The aim of the study was to assess hepatitis E virus (HEV) infection in HIV-infected patients attending a French Parisian hospital.
Methods: Out of 1250 HIV-infected patients attending the clinics, 108, with elevated transaminase episodes during the last four years, were included in the study. Two hundred and twelve episodes were recorded and 191 plasma samples (1 to 8 per patient), collected simultaneously to the episodes, were retrospectively tested for the presence of anti-HEV IgM and IgG antibodies and HEV RNA.
Results: An acute infection, documented by positive tests for anti-HEV IgM, low anti-HEV IgG avidity index (10%) and plasma HEV RNA (genotype 3e), was diagnosed in an homosexual patient with a moderate immunodepression (CD4+ lymphocyte count above 200/mm^3). This infection was likely locally acquired. It was benign and resolved within two weeks. No persistent carriage of HEV occurred. In addition, three past infections were evidenced, all of them in patients originating from countries with low socio-economic status. No persistent infection was diagnosed in our cohort.
Discussion: HEV should be tested in HIV-infected patients with elevated transaminase levels. HEV RNA detection should be used to diagnose the infection and monitor recovery.

O11

Co-receptor usage prediction at quasispecies level using ultra-deep pyrosequencing on both circulating and proviral HIV in patients candidates to CCR5 antagonist treatment

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Background: Ultra-deep Pyrosequencing (UDPS) offers the opportunity to analyze the co-receptor usage of each variant present in a viral quasispecies. Aim of the study was to assess co-receptor usage by UDPS, in comparison with a reference phenotypic test, in patients candidates to CCR5 antagonists treatment, in both circulating and proviral HIV-1.
Methods: Seventeen patients were enrolled. Trofile phenotypic test was performed by standard in 9 and by enhanced procedures in 8 patients. UDPS was carried out on both plasma virus RNA and on PBMC proviral DNA, amplifying *env* V3 loop region. Genotypic prediction of co-receptor usage was obtained by Position Specific Score Matrix analysis. Comparison between phenotypic and genotypic assays was performed considering a threshold of 5% and 0.3% for X4 detection (standard and enhanced Trofile sensitivity).

Results: After editing, a total of 130,999 V3 sequences were considered, with a mean coverage per site of 5,955. Concordance between phenotypic and UDPS results was 0.60 ± 0.24 (Cohen K index ± SE). Discordant results were observed in one case with standard and in 2 cases with enhanced trofile test. Only one patient, eligible to CCR5 antagonist treatment based on Trofile results, showed X4 variants in the circulation by UDPS at a frequency higher than that of Trofile sensitivity. Quasispecies archived in PBMC tended to be more heterogeneous than that found in circulating virus. All patients, with only one exception, harboured in proviral DNA X4 variants at variable frequency (from 0.07% to 53%).
Discussion: UDPS allows a detailed characterization of HIV V3 quasispecies in both circulating and archived sequences. Almost concordant results were obtained by UDPS and Trofile for prediction of co-receptor usage of plasma virions. In proviral DNA, X4 variants were commonly observed at variable frequencies. The importance of archived X4 variants in influencing clinical response to CCR5 antagonists is a crucial point to be addressed.

O12

Semen may harbor HIV despite effective HAART: another piece in the puzzle

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Background: The aim of this study was to assess the prevalence of male patients with undetectable HIV levels in blood that had detectable levels of HIV in semen.
Methods: Three hundred and thirty-two HIV-1 infected men attending in a In vitro fertilization laboratory in Marseille (FRANCE) were included in the analysis with respect of the French law (absence of ongoing disease, CD4 count >200 cells/mm^3, and stable HIV-1 RNA level). Overall, 394 paired blood and semen samples were provided between October 2001 and March 2009. The Cobas Taqman HIV-1 assay was used to quantify HIV-1 RNA in blood and in seminal plasma as previously described with a limit of quantification of 40 copies/ml in blood and in seminal plasma.
Results: Overall, 272 (69%) paired samples were concordant between blood plasma and seminal plasma for HIV-1 detection (inter-rater agreement k = 0.12); 253 (64%) samples were HIV-1 negative both in blood plasma and seminal plasma, and 19 (5%) samples were HIV-1 positive both in blood plasma and seminal plasma. Overall, 122 (31%) paired samples were discordant between blood plasma and seminal plasma for HIV-1 detection. Among these, 10 (3%) seminal plasma samples had detectable HIV-1 RNA although blood viral load was undetectable for at least 6 months under antiretroviral treatment.
Discussion: In conclusion, between 3% of patients with undetectable HIV levels in blood had detectable levels of HIV in semen. These data suggest that undetectable plasma HIV RNA means a lower risk of viral transmission through seminal fluid on a population level, but not necessarily at the level of the individual.

O13

Tenofovir (TDF) containing first-line HAART is associated with changes in plasma parameters suggestive of increased bone resorption

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Retrovirology 2010, **7(Suppl 1):**O13

Background: Prolonged treatment with tenofovir (TDF) containing HAART is associated with reductions in kidney tubular resorption of phosphorus (P). Added to this, TDF may directly affect bone remodeling. Current study analyses the potential repercussion of Calcium (Ca) and P wasting on bone-mineral metabolism.
Methods: All consecutive asymptomatic HIV-infected patients starting first-line HAART were recruited. Patients receiving HARRT with vs without TDF were considered separately. Changes with respect to baseline values in Ca and P plasma levels, alkaline phosphatase (AP) concentration and parathyroid hormone (PTH) levels were analyzed in patients followed for longer than 12 months. Adherence >95% as assessed every 4 months was requested to remain in the study.
Results: A total of 101 patients were included, 64 (63%) received TDF and 37 (37%) non-TDF containing HAART (mean age 38 years-old, 92% males, 90% Caucasians, mean CD4 count 278 cells/uL, mean HIV-RNA 4.3 log copies/mL, without differences between groups). After a mean follow up of 18 months, mean variation in bone-mineral metabolism plasma parameters with respect to baseline values, in patients under TDF vs non-TDF, was respectively as follows: P levels -0.31 mgr/dL [p < 0.001] vs -0.05 mg/dL [p = 0.7]; Ca levels +0.24 mg/dL [p = 0.005] vs +0.23 mg/dL [p = 0.36]; AP concentration +60 IU/L [p = 0.005] vs -36 IU/L [p = 0.09]; and PTH levels +20.3 pgr/mL [p = 0.02] vs +12.6 pgr/mL [p = 0.25].
Discussion: First-line HAART containing TDF, as compared with other nucleoside analogs, is associated with significant reductions in P levels, and increase in AP and PTH concentrations. These alterations are recognized markers of enhanced bone resorption, and may herald osteopenia in the long-term.

O14

SIDE effects associated with use of nevirapine in HIV treatment naïve patients with respect to baseline CD4 count

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Retrovirology 2010, **7(Suppl 1):**O14

Background: This study aims to detect Nevirapine (NVP) side effects among patients started with lead in dose at initiation of Anti Retroviral Therapy (ART) with CD4 count >250 cells/mm^3 in female and CD4 count >400 cells/mm^3 in male.
Methods: Close monitoring was conducted for the detection of NVP based side effects among ART - naïve patients initiated on CD4 count >250 cells/mm^3 among women and CD4 count > 400 cells/mm^3 are the

Table 1 (abstract O14)

Parameter		Outcome
NVP lead in dose initiated patients		3647 (n)
	Male	2408(66.02%)
	Female	1239(33.97%)
Male with CD4 >400 cells/mm^3 and initiated NVP lead in dose (n = 2408)		47(1.95%)
Female with CD4 >250 cells/mm^3 and initiated NVP lead in dose (n = 1239)		112(9.03%)
NVP induced Rash	Male (n = 47)	0
	Female (n = 112)	5(4.46%)
NVP induced Hepatitis	Male (n = 47)	1(2.12%)
	Female (n = 112)	0
Male with CD4 <400 cells/mm^3 and initiated NVP lead in dose (n = 2408)		1629(67.64%)
Female with CD4 <250 cells/mm^3 and initiated NVP lead in dose (n = 1239)		838(67.63%)
NVP induced Rash	Male (n = 1629)	19(1.16%)
	Female (n = 838)	15(1.78%)
NVP induced Hepatitis	Male (n = 1629)	5(0.30%)
	Female (n = 838)	2(0.23%)

study target at Centre of Excellence (CoE), ART Centre, B. J. Medical College, Civil Hospital, Ahmedabad, Gujarat, India.
Results: Total 5060 patients were initiated ART during the period of may 2005 to may 2009 at the institute. Among this 3647 (72%) were initiated with NVP lead in dose as per the Indian National ART Guidelines, Table 1.
Discussion: Skin Rash was recovered on substituting with another NNRTI-Efavirenz (EFV) and the treatment was well tolerated. Hepatitis was managed with substitution to EFV and close follow-up on ALT and AST. Though western literature has a black box warning for use of NVP, this data shows if closely monitored it could be given at resources limited settings with CD4 counts >250 cells/mm^3 in females and >400 cells/mm^3 males. Nevirapine is cost effective molecule compared to Efavirenz and when given in such conditions of higher CD4 need close follow up.

O15

The MONET trial: correlation between Hepatitis C coinfection and HIV RNA responses during darunavir/ritonavir monotherapy, for patients with HIV RNA <50 copies/mL at baseline

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Background: Co-infection with Hepatitis C has been associated with higher rates of treatment failure in cohort studies.
Methods: 256 patients with HIV RNA <50 on current HAART for over 24 weeks (NNRTI based (43%), or PI based (57%), switched to DRV/r 800/100 mg once daily, either as monotherapy (n = 127) or with 2NRTI (triple therapy arm, n = 129). This sub-analysis investigated the effect of Hepatitis C co-infection on HIV RNA levels during the trial.
Results: At baseline, more patients were HCV antibody positive by serology in the DRV/r arm (17%) than in the control arm (9%). In the primary efficacy analysis at Week 48, 86.2% of patients in the monotherapy arm and 87.8% in the triple therapy arm had HIV RNA <50 copies/mL. Only four of the confirmed elevations in HIV RNA were above 400 copies/mL (two in each arm). In multivariate analysis (Per Protocol), hepatitis C co-infection was a significant predictor of confirmed HIV RNA elevations ($p < 0.01$). For patients infected only with HIV (HCV antibody negative at baseline), the percent HIV RNA <50 was 88.1% in the monotherapy arm versus 87.3% in the triple therapy arm. For patients HCV antibody positive at baseline, the percent HIV RNA <50 was 61.9% for the monotherapy arm versus 58.3% for the triple therapy arm. Three patients had acute HCV infection during the trial (all in the DRV/r arm): all three had HIV RNA elevations at the time of acute HCV infection.
Discussion: In this study for patients with HIV RNA <50 copies/mL at screening, switching to DRV/r monotherapy showed non-inferior efficacy versus 2NRTI + DRV/r. Hepatitis C co-infection was more prevalent in the DRV/r monotherapy arm, and was a significant, independent predictor of transient, low-level HIV RNA viraemia. Hepatitis C co-infection might be a marker of poor adherence, or might be directly correlated with HIV RNA viraemia.

O16

Inhibitors of human immunodeficiency virus-1 replication targeting the human DEAD-box polypeptide 3 (DDX3) RNA helicase

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Background: Compounds currently used for the treatment of HIV-1 infections are targeted to viral proteins. However, the high intrinsic mutation and replication rates of HIV-1 led to the emergence of drug resistant strains with a consequent therapeutic failure. On this basis, cellular cofactors represent attractive new targets for HIV-1 chemotherapy, since targeting a cellular factor that is required for viral replication should help to overcome the problem of viral resistance. We aimed to develop through rational design a series of non-nucleosidic inhibitors of the HIV-1 cellular cofactor DDX3 and show that they can be used to block viral proliferation.
Methods: The X-ray crystallographic structure of human helicase DDX3 in complex with AMP has been used to generate a structure-based pharmacophoric model to be inserted in a computational protocol for the identification of small inhibitors of the ATPase activity of DDX3. Next, the pharmacophore was used as the three-dimensional query of a virtual screening approach to filter databases of commercially available compounds in order to identify chemical scaffolds with putative affinity toward the DDX3 ATP binding site. Positive hits were tested in antienzymatic and antiviral assays. An iterative process of synthesis, testing and optimization was used to derive lead compounds.
Results: One positive hit was identified in the first virtual screening with an IC50 against the enzymatic activity of DDX3 of 5 μM. The mechanism of action was found uncompetitive with respect to ATP and dependent on the multimeric state of the enzyme and several derivatives have been synthesized. Among those, some compounds showed nanomolar potencies against the enzymatic activity of DDX3 and micromolar potencies against HIV-1 replication in PBMCs, with no detectable toxicities.
Discussion: We report the identification of the first non-nucleosidic compounds suppressing HIV-1 replication by targeting a cellular enzyme. Our results provide a proof-of-principle for the feasibility of blocking HIV-1 infection by rendering the host cell environment less favourable for the virus. This approach may potentially overcome the problem of drug resistance related to drugs targeting viral proteins

POSTER PRESENTATIONS

P1

HIV/AIDS infection in Ukraine: a review of epidemiological data

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Retrovirology 2010, **7(Suppl 1):**P1

Background: To better understand the HIV dynamic in Ukraine in order to adapt HIV control efforts accordingly. The number of newly diagnosed HIV infections in Ukraine ranks second in the WHO European Region after the Russian Federation according to the end-year report 2006 of the European Centre for the Epidemiological Monitoring of AIDS (EuroHIV). According to the Ukrainian Ministry of Health the cumulative number of reported HIV infections by the end of 2008 amounts to more than 140.000 cases. However, because of many unreported cases the actual figures are supposed the official reports considerably.
Methods: Reported HIV/AIDS cases from the official epidemiological register of the Ukrainian Centre for AIDS Prevention between 1987 and 2008 were analysed. Joint United Nations Programme on HIV/AIDS country fact sheets were reviewed and analysed and this information was supplemented with published HIV prevalence and sexually transmitted disease case reporting information.
Results: Between 1987 and 2000, 36,600 Ukrainian citizens were registered with HIV. The number of officially registered HIV-infections increased from 7,000 in 2001 to 18,963 in 2008. 7,009 new infections were due to IDU, 7,880 to heterosexual contact and 3,635 to vertical transmissions from HIV infected mothers to their children, Table 1.
Discussion: The Ukraine is one of the European countries with the most rapidly increasing number of newly diagnosed HIV cases, mainly transmitted through IDU, but also increasingly through heterosexual contact in the general population and mother-to-child-transmission. In order to protect people from HIV infection, it is important to find ways to empower them by implementing policies and specific prevention measures that increase their access to knowledge about the HIV/AIDS epidemic. Free HIV tests, programs for the prevention the mother's child

Table 1 (abstract O14). Newly diagnosed HIV infections, AIDS cases and AIDS deaths in Ukraine, 1987-2008

	*1987-2000**	*2001*	*2002*	*2003*	*2004*	*2005*	*2006*	*2007*	*2008*
New HIV infections									
New HIV infections in total	36600	7000	8756	10009	12491	13770	16078	17669	18963
AIDS									
AIDS cases	2040	867	1353	1915	2743	4217	4723	4573	4380
Deaths among AIDS cases	1000	473	834	1285	1775	2188	2416	2507	2710

*Cumulative HIV new infections since 1987.

transference, information and access to condoms, clean hypodermic needles, drugs sentence programs and a medical treatment of gender illness would be important measures for the prevention and health care [1].

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P2

Human papillomavirus (HPV) genotypes among HIV-infected and HIV-uninfected women in Mozambique

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Background: The objective of this study was to determine the prevalence of HPV infection and related genotypes in a group of HIV-infected women and in a control group of HIV-uninfected women. To our knowledge, it is the first study such study conducted in Mozambique.
Methods: The study was conducted in a public health center in Maputo. It was performed in the context of the Drug Resource Enhancement against AIDS and Malnutrition (DREAM) program, managed by the Community of Sant'Egidio within the national health system in collaboration with Ministry of Health. This is a prospective, two-arm, observational study. The first arm includes HIV- infected women while the second arm involves a control group of HIV-uninfected women. The enrollment period lasted 6 months (August 2007- January 2008). The observation period was 6 months. HPV detection genotyping was performed using CLINICAL ARRAYS® (GENOMICA SAU). HIV was detected with b-DNA assay (HIVç1 RNA 3.0, Bayer Health Care).
Results: The study involved 191 participants: 141 HIV-infected and 50 HIV-uninfected women. HPV was found in 126/141 (89.4%) of those HIV infected and in 33/50 (66%) of HIV-uninfected subjects ($p < 0.001$). In HIV-infected women there were 94/126 (74.6%) HPV-multiple-infections while in HIV-uninfected women there were 22/33 (66.6%) ($p = 0.375$). Twenty-nine distinct HPV types were identified among the 141 HIV-infected women, of which 16 viral types were classified as cancer high-risk or probable high-risk viruses. The most common types identified were HPV types 58 (12.1%, $p < 0.001$), 16 (10.7%, $p = 0.06$), 61 (8.4%, $p < 0.05$), 53 (7.9%, $p = 0.55$) and 6 (7.6%, $p = 0.41$). Nineteen distinct HPV genotypes were identified among the 50 HIV-uninfected women. Ten types were of high-risk or probable high-risk. The most common types identified in HIV-uninfected subjects were HPV types 53 (10.4%), 6 (9.1%), 16 (9.1%), 18 (9.1%) and 66 (7.8%).
Discussion: HPV infection was *mainly* associated with HIV positive status. HPV multiple infections were high in the population studied, independently of HIV status. HPV Genotypes are different in the two groups. The HPV types identified are partially different from those more commonly identified in Western countries.

P3

Factors affecting Human Immunodeficiency Virus/Acquired Immune Deficiency Syndrome preventive behavior among pregnant women in ANC clinic in maternity hospital, Nepal

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Retrovirology 2010, **7(Suppl 1):**P3

Background: To determine the factors affecting HIV preventive behaviors among pregnant women attending antenatal care clinic in Maternity hospital Kathmandu, Nepal.
Methods: A cross sectional study using questionnaire, among 150 pregnant women, ranging from 17 to 35 years of age, attending ANC clinic at Maternity hospital, Kathmandu, Nepal in 2005.
Results: No significant association between age and overall HIV preventive behavior. (p = 0.866); and sexual HIV prevention (p = 0.535) or asexual HIV preventive behavior. (p = 0.180). Significant association between occupational status and overall HIV preventive behavior (p = 0.000); and sexual HIV preventive behavior (p = 0.000). No significant association between occupational status and asexual HIV preventive behavior (p = 0.058), but employed women showed higher tendency to have good asexual HIV preventive behavior than unemployed (14% versus 4.3%). Significant association between financial status and overall HIV preventive behavior (p = 0.001); and sexual HIV preventive behavior (p = 0.000). There is no significant association between levels of knowledge; perception of the respondents; having experience of STI and overall HIV preventive behavior.
Discussion: Study reveals that women have high perceptions of HIV susceptibility but have low efficacy. The pattern of sexual behavior cannot be separated from wider social and cultural influences. With majority of women with only primary level education, unemployment and at low income group, the socio-cultural influence is felt heavily in their sexual behavior such that despite having good knowledge and high HIV perception, women in this study still did not possess good HIV preventive practices. Hence, innovative social marketing HIV/STD prevention is needed for women to perceive benefits of prevention behaviors greater than the barriers to the behavior. Scale-up of HIV prevention programme requires inclusion of sex education and training on negotiation skills in a sexual relationship to women at school, community, workplaces, health post and hospital counseling services.

P4

Treatment of highly pathogenic filovirus infections using advanced antisense technology

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Background: Filoviruses, comprised of the Ebola viruses (EBOV) and Marburg virus (MARV), are highly pathogenic agents with human case fatality rates up to 90%. Periodic filovirus outbreaks occur in endemic regions of sub-Saharan Africa, and a new pathogenic EBOV species was

discovered as recently as 2008, following an outbreak in Uganda. Additionally, these viruses are considered Category A biodefense pathogens. There exists an urgent need to develop effective antiviral therapeutics and vaccines to protect both civilian and military populations at risk of exposure. Our aim was to identify and develop effective antisense-based therapeutics for treatment of filovirus infections. We have previously shown that synthetic antisense phosphorodiamidate morpholino oligomers (PMOs), which target EBOV-specific transcripts, confer partial protection to infected rodents and rhesus macaques. In this report, we describe the efficacy of a new class of positively charged PMOs (PMO+).

Methods: Using established rodent models of EBOV and MARV infections, we screened virus-specific PMO+ agents to identify combinations capable of protecting against lethal challenge. Efficacious PMO+ combinations were further evaluated in non-human primate models of filovirus infection.

Results: In EBOV models of infection, PMO+ molecules specific to EBOV VP35 and VP24, the combination of which is designated as AVI-6002, protected EBOV-infected mice and guinea pigs. Delivered by a combination of subcutaneous (s.c) and intraperitoneal (i.p.) routes, AVI-6002 protected 5 out of 8 (63%) of EBOV Zaire-infected rhesus macaques when delivered using a therapeutic regimen (i.e. beginning post infection). Similar survival (60%; 3 out of 5 macaques) was observed in a separate study when AVI-6002 was delivered intravenously (i.v.). In MARV infections models, a combination treatment containing a mixture of NP- and VP24-specific PMO+ agents, designated as AVI-6003, conferred a high degree of protection to mice and guinea pigs challenged with a lethal dose of MARV. Delivered using a therapeutic regimen, AVI-6003 completely protected MARV-infected cynomolgus macaques when delivered via s.c, i.v., or a combination of s.c. and i.p. routes.

Discussion: AVI-6002 and AVI-6003 represent highly promising therapeutic candidates for treatment of filovirus infections in humans.

P5

The HIV-1 viral protein Tat modulates glutamate and GABA exocytosis from human and mouse neocortical nerve endings by acting at different binding sites

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Background: Central Nervous System (CNS) disorders often accompany the acquired immunodeficiency syndrome (AIDS) and are typified by neuropsychiatric symptoms, such as cognitive and motor impairments, sometimes paralleled by neuropathological hallmarks. Collectively, these events are referred to as HIV-1 associated dementia (HAD). Before the advent of the Highly Active Antiretroviral Therapy (HAART), about 20% of adult patients, but as many as 40% of children/adolescent infected subjects developed HAD. Nowadays, in the era of HAART, the prevalence of HAD has decreased, but a more subtle form of disorder, referred to as Minor Cognitive Motor Disorder (MCMD), has emerged in about 20% of symptomatic HIV-1 seropositive patients, including those receiving HAART. This work was aimed at investigating the impact of the HIV-1 viral protein Tat on central neurotransmission since this protein has been proposed as one of the viral component involved in the onset of central neuropsychiatric symptoms.

Methods: The approach used was the up-down superfusion of purified synaptosomes isolated from human neocortical specimens removed during neurosurgery from consenting patients suffering of brain tumours. Experiments were also carried out by using mouse purified synaptosomes or slices in an attempt to propose an animal model suitable to investigate Tat-induced modification to central nervous system. Functional studies were paralleled by biochemical investigation on the existence of receptor protein potential involved in the effects observed and changes to second messenger production.

Results: Human immunodeficiency virus-1 (HIV-1)-encoded transactivator of transcription (Tat) potentiated the depolarization-evoked exocytosis of [3H]D-aspartate ([3H]D-ASP) from human neocortical terminals. The metabotropic glutamate (mGlu) 1 receptor antagonist CPCCOEt prevented this effect, whereas the mGlu5 receptor antagonist MPEP was ineffective. Western blot analysis showed that human neocortex synaptosomes possess mGlu1 and mGlu5 receptors. Tat potentiated the K+-evoked release of [3H]D-ASP or of endogenous glutamate from mouse neocortical synaptosomes in a CPCCOEt-sensitive, MPEP-insensitive manner. Deletion of mGlu1 receptors (crv4/crv4 mice) or mGlu5 receptors (mGlu5-/-mouse) silenced Tat effects, suggesting that mGlu1 and mGlu5 receptors are co-express on glutamatergic terminals. Tat enhanced inositol 1,4,5-trisphosphate production in human and mouse neocortical synaptosomes, consistent with the involvement of group I mGlu receptors. Tat inhibited the K+-evoked release of [3H]GABA from human synaptosomes and that of endogenous GABA or [3H]GABA from mouse nerve terminals; the inhibition was insensitive to CPCCOEt or MPEP. In mouse neocortical slices, Tat facilitated the K+- and the veratridine-induced release of [3H]D-ASP in a CPCCOEt-sensitive manner and was ineffective in crv4/crv4 mouse slices.

Discussion: These observations are relevant to the comprehension of the pathophysiological effects of Tat in central nervous system and may suggest new potential therapeutic approaches to the cure of HIV-1 associated dementia.

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P6

A role for the tetraspanin CD81 on the late steps of HIV-1 replication in T-lymphoblastic cells

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Background: HIV-1 uses cellular co-factors for virion formation and release, and is able to incorporate host cellular proteins in the viral particles, such as tetraspanins which serve as gateways for HIV-1 egress. Here, we investigated the implication of several tetraspanins on HIV-1 formation and release in chronically infected T-lymphoblastic cells, a model that permits the study of the late steps of HIV-1 replication in persistent infected cells.

Methods: HIV-1 infected MOLT cells were analyzed for HIV-1 production by RT assays and Western blot analysis. Gag-Tetrapanins associations were analyzed by immunoprecipitations in the purified virions and in the infected cells and by immunofluorescence confocal microscopy analysis. Down-regulation of CD81 expression in HIV-1 chronically infected MOLT cells was performed by shRNA lentiviral vectors and infectivity was monitored on SupT1 cells.

Results: Our data revealed that HIV-1 Gag and Env structural proteins colocalized with specific tetraspanins in the form of clusters at the cell surface. Co-immunoprecipitation experiments showed that viral Gag proteins interact, directly or indirectly, with the CD81 tetraspanin, and less with CD82, but not with Lamp2 or CD45, in tetraspanin-enriched microdomains composed of CD81/CD82/CD63.

When HIV-1 producing cells were treated with anti-CD81 antibodies or upon CD81 silencing by RNA interference, HIV-1 release was significantly impaired and its infectivity on SupT1 lymphocytes was modulated. We observe that CD81 downregulation in HIV-1 infected T-lymphoblastic cells resulted in Gag redistribution at the cell surface and an increase in infectivity.

Discussion: Our results highlight a critical role for CD81 on HIV assembly in T lymphoblastic cells [1], which was also reported in HIV-1 infected monocytes derived macrophages [2], and on HIV transmission in CD4+ T cells [3] and dendritic cells [4]. In addition, our findings extend the notion that even if HIV-1 assembly can occur on tetraspanin-enriched microdomains containing CD81, the incorporation of CD81 in the viral particles restrict HIV-1 infectivity. This notion can be extend to other cell membrane proteins, such as Hdlg, a cell-cell junction protein, that can also modulate HIV-1 infectivity [5].

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P7

Highjacking of PI3K/AKT signaling pathway by Hepatitis C virus in TLR9-activated human plasmacytoid dendritic cells

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Background: Plasmacytoid dendritic cells (pDCs) are responsible for the production of type I IFN during viral infection. Viral elimination by IFN-alpha-based therapy in more than 50% of patients chronically infected with hepatitis C virus (HCV) suggests a possible impairment of production of endogenous IFN-alpha by pDCs in infected individuals. Recent studies in the HCVcc-exposed pDCs purified from healthy donors show that HCV is a weak inducer of IFN-alpha *in vitro* and that HCVcc blocks the TLR9-mediated IFN-alpha production. It has been also reported that PI3K/AKT is critical for type I IFN production by pDCs in response to TLR agonists. The specific aim of the present study is to investigate the effect of HCV on PI3K/AKT signaling.
Methods: To this end we exposed pDCs from healthy donors to insect cell-derived HCV-like particles (HCV-LP) or an insect cell control preparation in the presence or absence of TLR7 and TLR9 agonists and determined dynamics of PI3K/AKT phosphorylation by flow cytometry. By this approach we compared the early (AKT phosphorylation) and late (IFN-alpha production) steps of TLR7/TLR9-MyD88 signaling. The levels of cell-free supernatant-secreted IFN-alpha were determined by ELISA.
Results: Expression of TLR9 gene was analysed by quantitative RT-PCR. Whereas phosphorylation of AKT increased 4 times during 12-h culture of pDCs in the presence of IL-3 and it was increased further by 50% after stimulation with CpG-C, it dropped-down to the basal level, when pDCs were preincubated with HCV-LP. Expression of TLR9 during 12-h culture of pDCs in the presence of IL-3 was reduced 10^4 times, whereas it was reduced 10^6 times, when pDCs were stimulated with CpG-C. HCV-LP did not show any silencing effect on TLR9 expression.
Discussion: We conclude that HCV-LP block the TLR9-mediated IFN-alpha production upstream of PI3K/AKT pathway and that HCV-LP do not block transcription of TLR9 gene. These findings suggest that HCV impairs signalization *via* TLR9 upstream of PI3K/AKT pathway in pDCs. Furthermore, our model system will allow elucidating the mechanism of the blockade of TLR9 signaling by HCV in pDCs. (ANRS grant 2007/306).

P8

Multicolor flow cytometry analysis of innate responses following in vitro interaction of PBMC with Hepatitis C virus

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Retrovirology 2010, **7(Suppl 1):**P8

Background: Alterations in innate immunity responses might be implicated in the establishment of a chronical infection with hepatitis C virus (HCV) in more than 80% of infected patients. This hypothesis is supported by the relative success of IFN-alpha-based therapy. Our aim has been to evaluate the consequences of HCV interaction with PBMC on global innate immune functions and to compare it to interaction with other RNA viruses, influenza and HIV-1.
Methods: The complex setting and diversity of interactions among cellular sub-populations involved in the innate response was approached by a short-time virus stimulation of total PBMC population. Multicolor cytometry is a unique tool for these multi-parametric investigations. By the mean of a 17-color LSRII, we have identified simultaneously plasmacytoid dendritic cells (pDCs), myeloid dendritic cells (mDCs), NK, monocytes and CD8+ T lymphocytes and analyzed their functional response to virus stimulation by measurment of expression levels of activation markers (CD69, CD83, CD86) and intra-cellular cytokines (IFN-alpha, IFN-gamma, TNF-alpha, IL-12).
Results: The global overview of the functional markers expression in each cell sub-population shows differences between the RNA viruses tested. Influenza induces pDC and NK activations but not the activation of mDC and monocytes. Interistingly, responses to HCV and HIV clusterize together and are characterized by a sustained IL-12 production in mDC and monocytes associated to a low pDC and NK activation. HCV-infected patients cells show a lower response to TLR7/8 agonist or HCV re-stimulation as compared to uninfected donors.
Discussion: In conclusion, with the help of multicolor cytometry technology, we were able to take a time-dependant picture of innate immune responses to RNA viruses stimulations from a complex cell system. Our results show importance of mDCs for a deeper understanding of HCV interactions with the innate immunity. (ANRS grant 2007/306).

P9

Comparison of Cytotoxic activity and Interferon-g secretion by Natural Killer Cells in HIV-1 and HIV-2 infected individuals

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Background: The role of NK cells in slowing disease progression in HIV-2 infected individuals compare to HIV-1 infected individuals.
Methods: In this study peripheral blood mononuclear cells were obtained from 30 HIV-1 and 30 HIV-2 infected subjects from each of 3 categories of CD4 T-cell counts (>500, 200-500 and <200 cells/ul) together with 50 HIV uninfected control subjects. Lytic activity and IFN-g secretion by NK cells from HIV-1 and HIV-2 infected subjects were measured by chromium-release and ELISPOT assays respectively following incubation of PBMC with the NK-sensitive K562 cells. Viral load was also measured from the plasma samples of the subjects.
Results: The cytotoxic response by NK cells was significantly higher in HIV-2 than in HIV-1 infection in subjects with CD4-T cell count >500 cell/ul ($p < 0.05$) and was similar to that of the healthy controls. There was a significant correlation between the magnitude of the NK population and cytolytic activity in HIV-2 individuals ($r = 0.27$, $p = 0.01$). There was also an inverse relationship between the cytolytic activity and plasma viral load in HIV-2 infected subjects ($r = -0.27$, $p = 0.009$). Interferon-g secretion by NK cells in ELISPOT assays was similar in HIV-1 and HIV-2 infections at all categories of $CD4^+$ T cell counts.
Discussion: The data suggest an efficient cytolytic function from NK cells in early HIV-2 infection, which is associated with high CD4 T cell counts. This may imply that a strategic immune-based therapy to control HIV disease through the enhancement of NK cell activity is worthy of consideration.

P10

Late HIV infection modulates the expression and activity of Cathepsin B, and its inhibitors in macrophages: implications in neuropathogenesis

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Background: To determine the mechanism by which HIV infection alters the expression and activity of CATB and its inhibitor, cystatin B (CSTB).

Methods: Peripheral blood derived macrophages (MDM) were infected with HIV_{ADA} at a MOI of 0.1 and cultured for up to 12 days. Intracellular and extracellular expression of CATB, CST B and CSTC in uninfected and HIV-infected cells were analyzed by Western blots and ELISA at 6, 9 and 12 days post infection (days p.i.). Activity of CAT B after HIV infection was determined by fluorescence and confocal microscopy.
Results: Expression of CATB protein and its intracellular inhibitor, CSTB, was increased in HIV infected cells after 12 days p.i. compared to uninfected controls ($p =< 0.05$). However CSTC increase was not significant in HIV infected cells. CATB was secreted to similar (>400 ng/ml) levels in both HIV-infected and uninfected cells at higher levels than those proved by others to promote cell damage (100 ng/ml or more). Importantly, secreted CATB from HIV-infected MDMs was significantly ($p = 0.008$) more active than that secreted from control cells throughout the extent of the infection.
Discussion: HIV infection increases the levels of active CATB in supernatants 4 times higher than those previously reported by other groups to be toxic to neuronal cells. Although CSTB increased in HIV-infected cultures, no effective inhibition of CATB was seen at 12 days pot-infection. Our results suggest that HIV infection is capable of altering the interactions between CATB and its inhibitors promoting, an increase in active CATB secretion, which may contribute to neuronal damage.
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P11

Impact of short term HAART initiated during the acute or chronic stage on SIV infection of the male genital tract

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Background: We previously evidenced the infection of human and macaque semen-producing organs by HIV-1 and SIV (Le Tortorec et al, Plos One, 2008; Le Tortorec et al, Retrovirology, 2008; Roulet et al, Am J Pathol, 2006). The male genital tract (MGT) is suspected to constitute a viral reservoir since persistent HIV shedding is found in the semen of a subset of HIV infected individuals under effective HAART (undetectable viremia). Using a macaque model, we investigated the impact of short term HAART initiated during the acute or chronic stages, on SIV infection of the MGT.
Methods: Adult male Cynomolgus macaques were treated with AZT/3TC/IDV for 2-4 weeks at 4 h and 21 weeks post-intravenous inoculation (p.i.) of SIV_{MAC251}. The presence of SIV in the testis, epididymis, prostate and seminal vesicles was analyzed at the end of the treatment period by nested PCR for SIV gag DNA and *in situ* hybridization for SIV gag RNA.
Results: HAART initiated 4 h post-infection prevents the peak of plasma viral load (PVL) and leads to a significant decrease of the virus dissemination in MGT tissues. In macaques treated during the chronic stage, the frequency of viral DNA detection in MGT tissues is on average similar to that of placebo animals, with the exception of 2 animals with an undetectable PVL, in whom a decrease of viral DNA detection is observed in all MGT organs, but to a lesser extent in the testis. In all animals including those with undetectable viremia, SIV RNA+ cells are still detected in the MGT organs following HAART.
Discussion: Short term HAART initiated post exposure dramatically reduces SIV dissemination in the MGT. Although efficient short term HAART initiated during the chronic stage decreases the level of infection of the MGT, SIV RNA+ cells can still be detected within the tissues. Whether prolonged HAART can eradicate SIV from the MGT will next be investigated.

P12

Enhanced induction of HIV-specific CTL by dendritic cell-targeted delivery of SOCS-1 siRNA

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Background: Dendritic cells (DC) are potent antigen-presenting cells that play a critical role in the activation of T cells. Antigen-loaded dendritic cell-based vaccines have been used for immunotherapy of human cancers and chronic infections, but only with limited success. RNAi-mediated silencing of negative immunoregulatory molecules expressed by DCs may provide a strategy to enhance the potency of DC-based vaccines and immunotherapy.
Methods: We have used a novel human HLA-A2 transgenic NOD/SCID-iL2rg chain -/- mice reconstituted with CD34+ HSC from A2 donors as a preclinical model to induce a robust CD8+ T cell-mediated protective immune response to HIV infection.
Results: SOCS-1 knockdown in human DCs a) enhanced their cytokine responses to LPS, and stimulated a strong mixed lymphocyte reaction in vitro, b) elicited a strong primary in vitro response to HLA-A2-restricted Melan-A/MART-1 and HIV Gag epitopes in naïve CD8+ T cells from healthy donors and c) increased the HIV gag-specific proliferation and polyfunctional cytokine response in CD8 T cells from seropositive subjects. More importantly, injection of gag peptide-pulsed, SOCS-1 silenced, but not just peptide pulsed HLA-A2 DCs, in the novel HLA-A2 humanized mice, gave rise to a robust multi-epitope-HIV specific CD8 T cells that could dramatically reduce the replication of a HIV-Gag-vaccinia recombinant challenge virus infection.
Discussion: These results demonstrate the feasibility of using manipulated DC as a prophylactic vaccine strategy for HIV infection in a humanized mouse model.

P13

Effect of Antiretroviral Therapy (ART) on HIV-1 & 2 (Subtype C) infection and its relationships to cognitive function and quality of life

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Background: To study the effect of Antiretroviral Therapy on neuropsychological functioning and quality of life of Seropositive HIV-1 Clade C infected subjects from South India.
Methods: A sample of 128 HIV-1 positive adults was recruited during 2003-2007. Of the 128 subjects, 8 were HIV-2 positive, 6 deaths and 2 dropouts during the 4 follow-up and were excluded from analyses. The present study conducted after the 4th follow-up and the final sample consisted of 112 seropositive HIV-1 Clade C infected subjects, 83 of them are not on ART (mean age 30 ± 4.35) and 29 of them are on ART (29.7 ± 5.75 years). Neuropsychological tests were administered in the domains of attention, fluency, motor speed, verbal and visual working memory, response inhibition, planning and verbal and visual learning & memory. WHOQOL-HIV BREF was also used to analyze the following domains of quality of life viz., physical health, psychological, level of independence, social relations, environment and spiritual beliefs. Mean and SD were calculated. Analysis of Variance (ANOVA) and Correlation were used to analyse the data. All statistical analysis were done using SPSS version 15.
Results: Analysis between ART and Non-ART groups differ on design fluency score ($p < 0.05$) and figural scanning ($p < 0.05$). Similarly, QOL domain significantly differs only on social relations ($p < 0.01$). Analysis between neuropsychological measures and quality of life (QOL) scores revealed significant correlation between QOL Total and motor speed ($p < 0.01$) and verbal fluency ($p < 0.01$). Social relations significantly correlate

on verbal working memory ($p < 0.05$), auditory verbal learning ($p < 0.05$), verbal memory ($p < 0.05$), verbal fluency ($p < 0.05$) and color trails ($p < 0.05$). Spiritual beliefs significantly correlate only of color trails ($p < 0.05$).
Discussion: Neuropsychological functioning and quality of life and with ART treatment revealed that ART success was significantly related to motor speed and fluency. Better neuropsychological performance may lead to more available social contacts and increase spiritual beliefs. In addition, none of the subjects developed any functional impairment.

P14

Abstract withdrawn
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P15

Effect of range of spread of CD4 count and RNA level on the efficacy of HIV treatments
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Background: We used 32,297 HIV/AIDS patients' records obtained from different Midwest clinics (and published in our web portal http://hivdatamining.com) to study the effect of CD4 count and RNA level fluctuation on patients' response to HIV regimens. We had two aims: 1. to investigate if controlling the range of spread of RNA level is more important than controlling the rate of spread of CD4 count. 2. To investigate if there is enough evidence that there exist RNA and CD4 correlation distribution that lead to better therapeutic responses.
Methods: Our HIV data analysis web portal stores patients' records obtained from different Midwest patients. We considered patients records that contains at least five laboratory tests during different patient's treatment times. Among other things, each patient laboratory test (from now on referred to a patient's record) provides CD4 count and RNA level readings.
To ensure the selection of unique datasets, we obtained ten randomly selected datasets each with 1,300 patients' records, without replacement, from the 32,297 records. To make sure that the selected records are independent, we performed a Chi Square test which yielded a p value of 0.0032. We then calculated the CD4 and RNA range of spread by calculating the standard deviation (S.D.) for each patient's CD4 and RNA level. We also calculated the correlation value between the CD4 and RNA reading for each patient. At the end of this process, we obtained 3,900 datasets. In order to summarize the data, we used the resulting 3,900 datasets to construct twenty eight groups of patients' records where each group has no more than 5% difference between their average CD4, RNA, and correlation values.
Results: We calculated that the correlation value between our proposed stddiff variable (stddiff = CD4 S.D. - RNA S.D.) and the correlation values of the correspond CD4, RNA values to be 0.78 with p value of 0.0083. We also found that the range of values with better (highly negative) correlation between CD4 and RNA values occurs when the calculated stddiff is not normally distributed.
Discussion: Contrary to many arguments that the change of RNA level is more significant than the change in CD4 count, we conclude that there is strong evidence, based on the above mentioned 0.78 correlation value, that both changes have significant impact on the HIV treatments' regimens. We also conclude, using the distribution of the stddiff parameter, that treatments effectiveness (represented by CD4 and RNA correlation) is strongly affected by the type of distribution of our proposed stddiff variable.

P16

HIV infection in elderly (patients aged more than 65 years)
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Background: After 1996 with HAART, not only HIV infected people live longer but also new HIV infection occurs in older people.
Methods: Retrospective observational study in HIV infected patients older than 65 years old in order to find special epidemiologic and clinical aspects of HIV infection in elderly.
Results: Among 1680 registered HIV infected patients, there are 61 HIV infected patients aged 65 or older (range = 65-84) including 13 women (21%) and 48 men (79%). There are 29 homosexual-bisexual (47%), 24 heterosexual (40%), only one patient blood transfusion and 7 patients denied any risk factors (11%). 23 patients (38%) with AIDS defining condition (C), 13 patients (21%) were symptomatic (B) and 25 patients (41%) were asymptomatic (A). Mean CD4 Count before antiretroviral treatment was 231, ranged 9 to 493. There were 24 patients (40%) with CD4 count less than 200 and 7 patients (11%) with CD4 count less than 100 cells/ml. Recent CD4 count ranged 10 to 1080 and (mean 478). CD4 increase = 247 after antiretroviral treatment. Mean viral load before treatment was 230184. There are 5 untreated patients (8%). After treatment 8 patients have detectable viral load more than 500 copies/ml including 2 untreated patients. It means 6 patients (10%) suffered from confirmed virologic failure and 7 patients (11%) present detectable viral load but less than 500 copies.
Discussion: 80% of HIV infected people older than 65 are men. Homosexual contact is the major risk factor in this group. In spite of 40% asymptomatic patients there are near 40% stage C. 40% of patients were coming with low CD4 count (less than 200). Mean CD4 count before treatment was 231 which is less than younger HIV infected patients, may be because of low CD4 count in older patients and/or late diagnosis of HIV infection in elderly. Virologic response is as well as younger patients even better.

P17

PRO 2000, a broadly active anti-HIV sulfonated compound, inhibits viral entry by multiple mechanisms
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Background: PRO 2000 is a polyanionic compound under development as a topical antimicrobial gel for the prevention of HIV-1 transmission. PRO 2000 is a synthetic naphthalene polymer which is documented to bind to HIV-1 gp120 and interferes with virus binding to $CD4^+$ T cells. Peculiar is that PRO 2000 appears to be more active (~10-fold) in inhibiting X4 viruses (IC_{50}: 1.9 μg/ml) compared to R5 viruses (IC_{50}: 20.8 μg/ml). Therefore, we investigated interactions of PRO 2000 with cellular HIV (co)-receptors.
Methods: Peripheral blood mononuclear cells (PBMCs) were incubated with PRO 2000 and flow cytometric assays performed using various fluorescent mAbs. Chemokine-induced signaling experiments were performed by FLIPR. Chemotaxis experiments were performed in 5-μm pore transwell filter membranes. $CXCL12^{AF647}$ binding was monitored by flow cytometry. The Bio-Plex human cytokine 27-plex assay system was used for the detection of cytokines/chemokines in PBMCs.
Results: PRO 2000 dose-dependently interfered with the binding of several anti-CD4 mAbs (clone OKT4, Leu3a, SK3) and anti-CXCR4 (clone 2B11 and 12G5) in PBMCs, whereas minor or no effects were observed on DC-SIGN and CCR5. The compound inhibited the CXCL12-induced signal transduction (IC_{50}: 19.9 μg/ml), CXCR4 internalization (IC_{50}: 9.5 μg/ml) and chemotaxis in PBMCs (IC_{50}: 6.7 μg/ml). It inhibited $CXCL12^{AF647}$ binding to T cells with an IC_{50} of 2.2 μg/ml. PRO 2000 did not induce signaling by itself. These CXCR4 antagonistic properties of PRO 2000 are potential additional mechanism of action that could explain the observation that PRO 2000 is more active against X4 viruses. In addition, we also examined the cellular activation potential and cytokines profile of PRO 2000 in PBMCs. PRO 2000 had minor effects on the induction of the activation markers CD25, CD69 and HLA-DR on T cells, but it did enhance the production of a small number of cytokines/chemokines, and most dramatically the production (~30-fold) of the specific CCR5 ligand MIP-1β.
Discussion: Thus, PRO 2000 is not just a pure gp120 binding inhibitor to $CD4^+$ T cells, but also interacts with CD4 and CXCR4 and can induce selective chemokines with potential anti-viral activity.

P18

Potential role of HIV-1 Nef and human M6B in HIV-associated neurological disorders

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Background: "Highly active antiretroviral therapy" (HAART) has dramatically increased the life expectancies of HIV positive humans. Due to this progress, other HIV-infection associated consequences like the HIV-associated neurological disorders (HIV-ND) are becoming more and more significant. The HIV Nef protein seems to play an important role in progression of HIV-NDs. We set out to identify brain tissue specific ligands of membrane associated Nef.
Methods: We applied a membrane associated yeast two-hybrid "split-ubiquitin" based system to identify human brain tissue specific proteins as direct ligands of HIV-1 Nef. Positive hits were confirmed by co-immunoprecipitation assays (CoIP), pull-down analysis, confocal microscopy and fluorescence titration assays.
Results: From a cDNA library of human brain tissue we identified the neuronal membrane glycoprotein M6B as a novel binding partner of Nef. Relevance of the Nef-M6B interaction was confirmed by CoIP assays in yeast and pull-down analysis using rat brain extracts. Association of Nef with M6B was supported by confocal microscopic studies in Neuro-2A cells. Co-localisation of transiently expressed Nef-DsRed with endogenous M6B or transiently expressed GFP-M6B was found. Direct interaction between Nef and M6B could be demonstrated by fluorescence titration studies using recombinant Nef protein and M6B derived peptides. We found that the Nef binding determinant of M6B is contained in its cytoplasmic loop that is conserved among proteins of the PLP family.
Discussion: Nef binding to M6B and other members of the PLP family might interfere with function and/or localisation of the respective protein leading to severe consequences for the function of HIV infected cells. Our results are discussed with the known benefits of SRI (serotonin re-uptake inhibitor) treatments or the synergistic proinflammatory and neurotoxic effects of exogenous opiate drugs during HIV infection.

P19

Abstract withdrawn

Retrovirology 2010, **7(Suppl 1):**P19

P20

HIV-1 evolution and drug resistance among patients receiving antiretroviral therapy in San Mateo county, California, 1997-2006

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Background: In community public health antiretroviral therapy (ART) programs, HIV-1 viremia in patients has been reduced over the last decade through improved drug regimens, adherence, and genotypic testing to manage ART. However, a large portion (up to 25%) of ART patients remain viremic with risk of disease progression and community-level transmission of drug resistance mutations. We hypothesized that HIV-1 viral evolutionary rate (vEvol) during incomplete suppression of HIV-1 RNA is associated with ART use, selection of drug resistance, and viral load.
Methods: HIV-1 reverse transcriptase (RT) and protease (Pr) sequences were obtained from 306 patients receiving ART at San Mateo Medical Center (1997-2006). From 150 paired sequences in 75 viremic patients, vEvol was determined using the TVM+I+G nucleotide substitution model in PAUP. Resistance mutations and genotypic susceptibilities to 19 ARV drugs were determined using the HIVseq algorithm (HIVDB.stanford.edu) and drug resistance mutations were correlated using hierarchical agglomerative clustering analysis. Statistical analysis was done in SAS v9.1.
Results: 75 paired genotypes were a median of 10.9 months apart with median CD4 cells 260/cu mm and geometric mean RNA VL 3.94 log copies/mL. ART was RTI-based (reverse-transcriptase inhibitor) in 38 (51%) vs. PI-based (protease inhibitor) in 37 individuals (49%), with similar rates of drug resistance mutations (89%) in either drug class. Median vEvol was higher in those with PI- (but not RTI) associated drug resistance mutations ($p < 0.05$, Wilcoxon rank-sum [WRS]). Higher vEvol was associated with reduced genotypic susceptibility scores to nRTI, nnRTI, and PI drugs ($p < 0.0001$, Spearman rank correlation). Hierarchical cluster analyses of all 306 sequences revealed many highly correlated drug resistance mutation pairs including T215Y/M41L in RT, V82/I54 in Pr, and M184V/L90M in RT/Pr, each being associated with higher vEvol ($p < 0.05$, WRS).
Discussion: Repeated HIV-1 RNA genotyping in viremic patients showed significant drug resistance mutations in 89% and evolutionary changes in RT/Pr in approximately 75%. Viral evolution was significantly greater among those with higher HIV-1 VL, exposure to PI drugs and specific patterns of resistance mutations. The accumulation of drug resistance mutations in explicit patterns during ART treatment is an evolutionary response to drug selective pressure driven by virus replication.

P21

Length variation in HIV-1 gp120 as the product of DNA misalignment mechanism

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Background: To determine whether misalignment structures such as duplications, repeats, and palindromes are associated to insertions/deletions (indels) in gp120, indicating that indels are indeed frameshift mutations generated by DNA misalignment mechanism.
Methods: Cloning and sequencing of a fragment of HIV-1 gp120 spanning C2-C4 derived from plasma RNA in 12 patients with early chronic disease and naïve to antiretroviral therapy.
Results: Indels in V4 involved always insertion and deletion of duplicated nucleotide segments, and AAT repeats, and were associated to the presence of palindromic sequences. No duplications were detected in V3 and C3. Palindromic sequences occurred with similar frequencies in V3, C3 and V4; the frequency of palindromes in individual genes was found to be significantly higher in structural (gp120, $p \leq 3.00E-7$) and significantly lower in regulatory (Tat, $p \leq 9.00E-7$) genes, as compared to the average frequency calculated over the full genome.
Discussion: Indels in V4 are associated to misalignment structures (i.e. duplications repeat and palindromes) indicating DNA misalignment as the mechanism underlying length variation in V4. The finding that indels in V4 are caused by DNA misalignment has some very important implications: 1) indels in V4 are likely to occur in proviral DNA (and not in RNA), after integration of HIV into the host genome; 2) they are likely to occur as progressive modifications of the early founder virus during chronic infection, as more and more cells get infected; 3) frameshift mutations involving any number of base pairs are likely to occur evenly across gp120; however, only those mutants carrying a functional gp120 (indels as multiples of three base pairs) will be able to perpetuate the virus cycle and to keep spreading through the population.

P22

Nucleolin relocalization associated with pre-lethal alterations of T cell morphology: redefining cell death in HIV infection

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Background: To redefine the causal link between cell cycle dysregulation of T lymphocytes and HIV induced cell death.
Methods: Intacellualar concentration of cell cycle regulatory proteins has been measured by western blot; nucleolin (C23) concentration and localization has been established by confocal microscopy; expression of surface proteins and ultrastructural membrane damage have been

analyzed by flow cytometry and transmission electron microscopy, respectively.
Results: Here we demonstrate that circulating T lymphocytes, both CD4+ and CD8+, leave lymphoid tissues with diffused regressive lesions such as vacuolization, blebbing, nuclear evanescence and organelle swelling. Equally diffused are biochemical anomalies that accompany the overall disarrangement of cell structure, namely (i) fragmentation and diffusion into the cytoplasm of C23/nucleolin, the principal structural protein of the nucleus (ii) an accumulation of short - lived regulatory proteins (p16, p21 and p53), likely due to the progressive extinction of the ATP - ub - proteasome system and (iii) a decreased expression of membrane proteins.
Discussion: The HIV - induced demise of CD4 -T cells is thought to be a result of the execution of genetically programmed cell death that occurs in lymphoid tissue, where many resident T cells are chronically hyperactivated and primed for apoptosis. The pre-lethal lesions described here recapitulate a series of regressive events that occur in immune cells, when they grow at high mitotic activity in conditions of scarce ATP production.

P23

The immunoglobulin CH1 constant region modulates antigen binding affinity and functional activities of the broadly neutralizing 2F5 HIV specific antibody

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Background: The ability of the heavy chain constant region (CH) to affect antibody affinity and specificity could be at the origin of a stronger or weaker memory response, depending on the isotype. Using as a model the broadly neutralizing human mAb 2F5, directed against the membrane proximal region (MPER) of the HIV-1 envelope transmembrane subunit gp41, we investigated the interplay between 2F5 isotype and functional activity.
Methods: A 2F5 IgA isotype was constructed from the 2F5 IgG1. Functional monomeric 2F5 IgA and IgG1 were expressed in CHO cells and their immunochemical characteristics and anti-HIV-1 in-vitro activity were evaluated.
Results: As compared to 2F5 IgG1, 2F5 IgA sharing identical VH and VL domains but in a different CH context: (i) binds with higher affinities gp41 and MPER peptides; (ii) has an increased capacity at inhibiting endocytosis of HIV-1 by dendritic cells; (iii) has an increased HIV-1 neutralizing activity in lymphocytic CD4+ T cells; (iv) blocks more efficiently HIV-1 transcytosis across epithelial monolayers in-vitro and normal human rectal mucosa, but (v) has lower ADCC activity. Epitope mapping with a 7 mer epitope library shows that 2F5 IgA recognizes essentially the same hexapeptide epitope as its IgG counterpart.
Discussion: These results show that the CH region can fine-tune the specificity of an antibody, by modulating its binding affinity to the antigen and the neutralizing activity of variable-region of otherwise identical antibodies. The determinant role of CH region on affinity and specificity changes our understanding of vaccine responses. In the context of HIV-1, which is mainly transmitted sexually, these results strongly suggest that raising a mucosal humoral IgA based response will be superior to an IgG one in blocking HIV-1 transmission.

P24

Definition of the interacting interfaces of Apobec3G and HIV-1 Vif using MAPPIT mutagenesis analysis

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Background: The host restriction factor Apobec3G is a cytidine deaminase that incorporates into HIV-1 virions and interferes with viral replication. The HIV-1 accessory protein Vif subverts Apobec3G by targeting it for proteasomal degradation. We studied the Apobec3G homomerisation and the interaction of Apobec3G with Vif in detail.
Methods: We used the MAPPIT two-hybrid technique to analyse the Apobec3G-Apobec3G and the Apobec3G-Vif interactions in intact human cells. MAPPIT is based on the functional complementation of a cytokine receptor signalling pathway.
Results: We propose a model in which Apobec3G N-terminal domains symmetrically interact via a head-to-head interface containing residues 122 RLYYFW 127. Mutations in the head-to-head interface abrogate the Apobec3G-Apobec3G interaction. All mutations that inhibit Apobec3G-Apobec3G binding also inhibit the Apobec3G-Vif interaction, indicating that the head-to head interface plays an important role in the interaction with Vif. Only the D128K, P129A and T32Q mutations specifically affect the Apobec3G-Vif association. In our model, D128, P129 and T32 cluster at the edge of the head-to-head interface, possibly forming a Vif binding site composed of two Apobec3G molecules.
Discussion: We propose that Vif either binds directly at the Apobec3G head-to-head interface or associates with an RNA-stabilized Apobec3G oligomer.

P25

Expansion of vdelta1 T lymphocytes reactive to c. albicans IN HIV-1 infected patients: effect of influenza virus vaccine

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Background: It is known that the circulating Vdelta2 T cell subset respond to mycobacteria and certain viruses, while the Vdelta1 subset is resident in the mucosal-asssociated lymphoid tissue and participate in the immunity against intracellular microrganisms. We reported that in HIV-1 infected patients circulating Vdelta1 T lymphocytes are increased; in vitro, these cells can proliferate in response to Candida albicans. We analysed the effects of influenza virus vaccination on the function of this T cell subset in HIV-1 infected patients and healthy donors.
Methods: We analysed the effects of influenza virus vaccination on the function of Vdelta1 and Vdelta2 T cell subsets in HIV-1 infected patients and healthy donors. Cells were isolated from blood samples obtained before and after 30 or 90 days after vaccination. Proliferation to C. albicans and to hemoagglutimnin (HA) was assessed by thymidine uptake after 7 days of stimulation.
Results: First, we confirmed that the Vdelta1 T cell subset is expanded in HIV-1 infected patients (absolute number of cells/microliter range 28-30 in HIV-1 patients vs. 8-12 in healthy donors). On day 90 after vaccination the number of Vdelta1 T cells significantly increased in HIV-1 patients (59 in the group A, 48 in the group B). Interestingly, upon influenza vaccination an increase in proliferation of Vdelta1 T cells to C. albicans was observed in HIV-1 patients, at variance with healthy donors, on day 30 and day 90. A specific cellular response to HA was detectable in HIV-1 patients only on day 90 post-vaccination without MF59 adjuvant, but it was observed on day 30, as in seronegative subjects, when MF59-vaccine was used.
Discussion: We suggest that in HIV-1 infected patients, a population of Vdelta1 T lymphocytes reactive to C. albicans is present in vivo; upon challenge with influenza virus vaccine this population receives an activation signal possibly mediated by cytokines triggered by the HA antigen itself.

P26

HIV-patients discrimination according to phenotype and functional assay of T-cells subsets

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Background: To distinguish HIV-1 patients with clinical diversity by using a simplified model of T-cell interactions.
Methods: During 28 months, 1074 blood samples from 200 HIV-1 patients and 418 blood samples from healthy blood donors were prospectively collected. T lymphocyte subsets and activation markers expression (CD4, CD69, CD25, CD8, CD28, HLA-DR) were determined initially and after PHA stimulation in whole blood cultures.
Results: Two step Cluster Analysis followed by a discriminant function analysis of the lymphocyte activation assay from the first blood sample, allowed the separation of HIV-1 patients in two groups: Cluster1 (67%) and Cluster2 (33%). Clusters definition relied on the level of three T-cells subsets: a) stimulated $CD4^+CD69^+CD25^{high}$, b) unstimulated $CD4^+CD69^+CD25^+$ and c) unstimulated $CD4^+CD25^{high}$. PHA stimulated $CD4^+CD69^+CD25^{high}$ subset level alone allowed to classify correctly patients with 92% sensitivity and 87% specificity. Cluster-2 expressed more CD69 and HLA-DR activation markers on CD4 and CD8 lymphocytes, less $CD8^+CD28^+$ and responded less to mitogen even if viral load undetectable. Cluster-2 presented poorly clinical profile in terms of previous AIDS events, current $CD4^+$ count, viral load, length of treatment. Over the time most patients (64%) were keeping their cluster category.
Discussion: We propose an algorithm to identify a subset of HIV patients with an over-determined immunodeficiency status characterized by a lower ability to reverse inappropriate activation of CD4 and CD8 T-lymphocytes leading probably to earlier exhaustion of their immunological resources. This subgroup of patients could display a worst clinical evolution, lower control capacity of viral load, even under antiretroviral therapy-mediated viral suppression.

P27

Evaluation of adult immunological outcomes from kimironko health center art program, 2007 to 2008

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Background: ARTservices are expanding in Sub Saharan African countries. In Rwanda the demographic and health survey of 2005(DHS) showed a prevalence rate of 3%at national level and by the end of June 2009, 70234 persons had initiated ARV at 217 sites in Rwanda (TRACnet report June 2009). Kimironko health center has provided care and treatment to patients since 2003, at this point there are 1776 patients followed in the HIV clinic at this health center. It is important to evaluate immunologic responses and efficacy of antiretroviral therapy in resource poor settings.
Methods: We conducted a retrospective cohort study to assess immunological outcomes among adult patients who initiated ART from January 1, 2007 to December 31, 2008. 426 patients were eligible for the study and data were analyzed using Epi info 3.2.2 and SPSS 17.
Results: The median age of our patients at ART initiation was 34 years and 66,9% were female. At ART initiation the median baseline CD4+ cell count was 192 cells per micro liter, the median CD4+ cell count increased by 90 cells per micro liter at 6 months(n = 393) [interquartile range (IQR) 191-372] and 112 cells per micro liter [IQR 230-408] at 12 months(n = 266). Women had higher median baseline CD4 cell counts than men (197 vs. 176/μL). First-line ART regimens contained Lamivudine (100%), Stavudine (68,3%), Zidovudine (31%), and either Efavirenz (14%) or Nevirapine (85,7%).
Discussion: These data demonstrate robust and sustained CD4 response to ART among patients continuing on therapy. Despite not having viral load testing, using CD4 criteria alone indicates efficacy of first line therapies. Public health and programmatic interventions leading to earlier HIV diagnosis and initiation of ART could substantially improve patient outcomes in resource-limited settings.

P28

Implications of hcv natural genetic diversity on HCV NS5B inhibitor NM283

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Background: The HCV NS5B RNA polymerase is a new target for drug development for HCV disease. Valopicitabine (NM283), the prodrug of 2'-C-Methylcytidine (NM107), has been the most clinically advanced NS5B nucleoside inhibitor. Nucleoside inhibitors exhibit similar activity among genotype 1 strains, but their efficacy among other genotypes is largely unknown. In this study NS5B amino acid polymorphisms in positions affecting activity and drug efficacy were investigated in sequences of all HCV genotypes.
Methods: NS5B amino acid positions significant for catalytic activity, drug binding and resistance were recovered from bibliography and molecular modelling. NS5B sequences were located and downloaded from the HCV sequence database, and added to experimentally derived NS5B sequences from drug-naïve patients in order to analyse significant amino acid positions for natural polymorphisms. The most frequent polymorphisms in resistance-conferring position 282 were further investigated by docking analysis.
Results: The results revealed a highly conserved active site. Natural polymorphisms at position 282 were found at low frequencies, in particular the drug resistant S282T, and S282R, whose effect is unknown. No genotype-specificity of polymorphisms could be confirmed.
Discussion: The selection of S282T as a drug-resistant variant when S282R also exists naturally at the same frequencies implies that the latter may not confer resistance to NM283. Molecular modelling suggests that loss of NM107 activity in the presence of the S282T mutation may be a result from improper alignment of the drug at the active site. Overall, the results imply the need for resistance testing when 2'-C-methyl nucleotide inhibitors are widely available.

P29

Viral variability study in follow-up sera from HIV-HBV-HCV coninfected patients

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Background: The genetic variability of hepatitis B virus in sera from HBV-DNA positive patients, HBsAg negative and antiHBc positive, coinfected with both HIV and HCV, was studied, to describe the natural history of HBV occult infections.
Methods: The follow-up sera, encompassing a six to nine years period, from eight patients with triple coinfection (HIV, HBV, HCV) were tested by a real time PCR HBV-DNA assay. Four patients that were HBsAg negative and HBV-DNA positive were classified as affected by occult infection. The remaining four that were HBsAg positive were used as controls. HBV-DNA was amplified by PCR and the sequence of the whole HBV genome was characterized by phylogenetic analysis (Neighbor Joining method, implemented by MEGA 3.1 software) and for the presence of specific mutations.
Results: Three out of four HBV patients with occult infection, showed reactivation phases of HBV viremia. Different mutations were observed,

with differences between pre- and post-reactivation sera. HBV-DNA remained at low levels during the entire study period also in absence of specific anti-HBV therapy. The phylogenetic analysis showed that, for each patient with HBV reactivation, all the isolates were originated from a unique parental virus. Specific mutations of PreS/S, Core and X regions were observed.
Discussion: Mutations in the "a" determinant of the S protein could be responsible for the absence of HBsAg detection. The presence of stop codon in the pre-core region and of mutations in the X region could, in part, explain the reactivation of HBV viremia.

P30

Pilot trial of oral therapeutic HIV vaccine, V-1 Immunitor, on HIV and HIV/HCV patients in Russia

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Background: V-1 Immunitor (V1) is a therapeutic AIDS vaccine formulated as an oral pill comprising heat-inactivated HIV antigens derived from pooled blood of HIV-positive donors. Several studies carried out in Thailand have reported beneficial effects of V1 in AIDS patients including the increase in CD4 counts; decrease in viral load; body weight gain; improved clinical symptoms; and extended survival. The goal of our study was to substantiate these effects independently in another country.
Methods: We have tested V1 in seven randomly chosen HIV-positive patients at our regional AIDS Center. Five patients were therapy naïve patients but two have been receiving HAART during study period. Patients were administered one V1 pill per day for three months.
Results: No adverse effects due to V1 administration were observed at any time. The average increase in CD4-positive lymphocytes was 50 cells (22% or 228 vs 278; p = 0.055); absolute CD8 cell counts increased by 29 (8% or 356 vs 385; p = 0.03); plasma viral load as measured by PCR decreased in all patients (p = 0.018 by Wilcoxon signed-rank test); the average weight gain was 4.6 kg (7.8% or 58.8 vs 64.4; p = 0.034). Clinical symptoms as observed by physicians improved in all patients, including three patients co-infected with hepatitis C virus. Patient-reported outcomes, i.e., appetite, energy, mood, and sense of well-being were also ameliorated.
Discussion: Despite small sample size, results of this study are statistically significant and support unequivocally the results of earlier trials in Thailand. Increase in CD4 counts and decrease in viral load can serve as immune correlates of vaccine efficacy in other AIDS vaccine clinical trials. Surprisingly V1 benefited HCV co-infection as well.

P31

Evaluation of immune response profiles of individuals with chronic Hepatitis C treated with interferon alpha and ribavirin, in the foundation of tropical medicine of Amazonas

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Background: The HCV is considered the main etiological agent involved in the hepatitis parenteral transmission. The most frequent genotypes in Brazil are 1, 2 and 3, and genotype 1b is the most frequent in blood donors. Data from the serological screening of the Foundation of Hematology and Hemotherapy of Amazonas (FHEMOAM) show that 0.32% of donors are seropositivity for anti-HCV. Some studies showed that 15 to 25% has good prognostic but 80% develops chronic hepatitis. The purpose study was to describe the clinical course and immunological profile of chronic infection by HCV in patients treated with interferon-alpha and ribavirin.
Methods: Clinical and laboratory evaluation, including viral genotype, viral load, and cellular and humoral immune response, during the first 24 weeks of therapy.
Results: Partial results showed that genotype 1 (51.72%) is more prevalent in the Amazon, followed by 3 (31.03%) and 2 (17.24%). Significant changes of AST and ALT concentrations showed an increase in the 4 weeks of treatment. We observed a trend to increase cell populations in time 0 (pretreatment) to lymphocyte (63.3 ± 88.7), monocytes (10.6 ± 21.5), neutrophils (86.7 ± 126, 1), had not statistically significant difference. The analysis by flow cytometry showed an increase in total T cells and CD4 + in 4 weeks, returning to baseline at 12 and 24 weeks after treatment. Furthermore, there was a decrease of LTCD8 + in 12 and 24 weeks after treatment.
Discussion: Partial results showed that HCV infection changes the profile of immune response in treated of patients with Interferon-alpha and ribavirin.
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P32

Abstract withdrawn

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P33

HIV-related morbidity rate, thirteen years after the introduction of highly active antiretroviral therapy (1996-2009)

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Background: After the availability of combined antiretroviral therapy (cART), we quantified the consequences on the general morbidity rates, and HIV-related hospitalization rates, in the period 1992-2008.
Methods: HIV-associated hospitalizations were assessed according to three different periods of time: before cART introduction (1992-1995), immediately after first cART availability (1996-1998), and the last one, referred to the fully established cART era (1999-2008).
Results: During the three examined periods, an undetectable viremia was never detected in any patient in the pre-cART era, in 21% of cases in the first years of cART, and in 41% of patients in the last years of cART ($p < .0001$). In parallel, the mean CD4+ T-lymphocyte count in the three study groups tested 27.2 ± 11.3 cells/μL, 39.3 ± 14.6 cells/μL, and 89.6 ± 38.2 cells/μL, respectively ($p < .001$). During time, an increased frequency of hospitalization of heterosexual and female patients occurred, while the frequenct of IVDA had a significant drop (from 69% in the pre-cART period, to 57% during initial cART era, to 39% at the time of consolidated cART era; $p < .0001$). The patients with a prior diagnosis of full-blown AIDS represented 86%, 57%, and 33%, respectively ($p < .0001$), while hospitalized inpatients who experienced a diagnosis of AIDS concurrently with the first detection of HIV infection (the so-called "AIDS presenters"), showed an evident temporal increase (11%, 21%, and 39%, respectively; $p < .0001$). Among concurrent illnesses, a huge rise of chronic liver diseases was registered from the pre-cART time (18%), to the first years of cART availability (29%), to the current time of advanced cART (48%) ($p < .001$), while an increased mortality due to hematological and solid malignancies also occurred, although at a lesser extent (8.2%, 11.7%, and 17.8% respectively; $p < .001$).
Discussion: The introduction of cART profoundly acted on the general morbidity for HIV infection and AIDS, although the epidemiological-clinical-laboratory scenario significantly changed over time. These modifications need a careful monitoring, in order to ensure a timely diagnostic and clinical disease recognition by all involved health caregivers who face HIV-infected patients, and to plan an adequate allocation of available resources, funding, structures, and dedicated personnel.

P34

The "AIDS Presenters" phenomenon, thirteen years after the availability of potent, combination antiretroviral therapy

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Background: Notwithstanding the availability of potent, combined antiretroviral therapy (cART), AIDS notifications continue to occur, with increasing prevalence for patients (p) who missed or neglected their condition, or refused or took with insufficient compliance the recommended antiretroviral medications.
Methods: All cases of AIDS notified since the year 2001 were compared with those found in the decade preceding cART availability (1986-1995).
Results: Compared with the pre-cART era, a significant drop of frequency of overall AIDS cases occurred: from a mean 58.3 ± 11.2 patients-year observed in the decade 1986-1995, to 13.7 ± 6.0 patients-year during years 2001-2008 ($p < .001$), together with an increased mean age ($p < .002$), female gender ($p < .01$), sexual *vs* i.v. transmission ($p < .001$), and proportion of immigrant *versus* native p ($p < .02$). In the cART era, the most evident drop of frequency interested opportunistic diseases linked to a CD4+ lymphocyte count below 50-100 cells/μL, while a proportional rise of tuberculosis, pneumonia, lymphomas, and other neoplasms was observed. The frequency of both *Candida* esophagitis and *Pneumocystis carinii* pneumonia remained stable, as the first two most frequent AIDS-related conditions. After cART availability, the following diagnoses in crude frequency were represented by neurotoxoplasmosis, wasting syndrome, AIDS-dementia complex, and non-Hodgkin's lymphomas. P with multiple AIDS-defining diseases, and also AIDS diagnoses made only at or after death, even showed a paradoxically increased frequency and absolute number during the cART era *versus* the prior decade ($p < .001$ and $p < .03$), while no difference was found as to the grade of HIV-associated immunodeficiency. Surprisingly, an underlying anti-HIV therapy was a more common event until 1995, *versus* p observed in the cART era ($p < .001$), since during recent years AIDS notification tends to be increasingly associated with the first diagnosis of HIV infection.
Discussion: When facing p with some form of opportunism, clinicians should maintain an elevated suspect for an advanced (but missed-untreated) HIV disease. A continued level of attention will help a more rapid recognition and an appropriate management of p who could not take benefit from cART, since they remained unaware of their disease, or refused controls and treatment during the previous years.

P35

The prevalence and presentation of heart disease in HIV positive adults attending a clinic at Mulago hospital, Kampala, Uganda

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Background: In the pre-HAART era, HIV related heart disease was described as an often underlooked consequence of the infection. Heart disease of non-infectious etiology unmasked by the clinical management of HIV is more often described. The signs and symptoms of heart disease when present portend a poor prognosis. This study sought to document the magnitude of cardiac involvement in Ugandan HIV patients. The objective was to determine prevalence, describe the clinical, echocardiographic and electrocardiographic presentations of heart disease amongst HIV positive adults attending a clinic (IDC) during the study period between September and December 2003.
Methods: This was a cross-sectional descriptive study. Five consenting HIV sero-positive adults attending IDC were selected three times weekly by systemic random sampling achieving a sample size of 128. Measurables including history and examination, electrocardiography and echocardiography were recorded in a pre-tested data collecting tool. Electrocardiography and echocardiography tests were conducted at the Cardiac Institute at the same hospital.
Results: Only 15.6% of patients were accessing antiretroviral therapy. Most cardiovascular abnormalities were sub clinical and only detected at echocardiography. 11/128 patients were in clinical heart failure; majority in WHO clinical stage IV disease. At least one cardiovascular abnormality was detected in 69% of the patients studied. Cardiovascular abnormalities were more prevalent in the more immunosuppressed patients in WHO Clinical Stage III, IV (34%) and least prevalent in WHO class II (9%) p - 0.028. 2 dimensional echocardiography detected cardiovascular abnormalities in 41.5% of patients. Electrocardiography detected abnormality in 40.6%, Doppler detected in 14.3% and clinical evaluation 10.2%. The main echocardiography abnormality was Left Ventricular Dysfunction (58.7%). The only clinical signs and symptoms alluding to cardiovascular disease was third heart sound (p - 0.014), tender hepatomegaly (p - 0.011), and oedema (p - 0.05).
Discussion: Cardiovascular abnormalities are common in adult HIV patients. The presentation is mainly sub clinical and more prevalent in severe immunosuppression. Signs and symptoms alluding to cardiovascular disease are easily masked by extra cardiac disease in these patients. Patients with advanced HIV disease should be carefully evaluated for cardiovascular disease be use of potentially cardio-toxic drugs.

P36

A 16-year prospective survey of Mycobacterium xenopi, *Mycobacterium kansasii* and *Mycobacterium fortuitum* infection in patients with HIV disease

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Background: A prompt and effective diagnosis and a timely treatment of atypical mycobacteriosis, and especially *Mycobacterium kansasii*, *Mycobacterium xenopi*, and *Mycobacterium fortuitum* disease, remains a serious challenge for clinicians engaged in the management of the immunocompromised host, including HIV disease.
Methods: Eighteen, twelve, and three HIV-infected patients with a microbiologically-confirmed *M. kansasii*, *M. xenopi*, and *M. fortuitum* respiratory infection respectively, have been observed in a 16-year period, out of over 4,700 hospitalizations performed because of HIV-associated disorders at our inpatient centre. These episodes were carefully evaluated from an epidemiological, bacteriological, clinical, and therapeutic point of view.
Results: In 15 out of the 33 overall episodes (45.5%), a concurrent bacteremia was also retrieved, as a sign of disseminated infection. A rapid and significant reductin of the crude frequency of atypical mycobacteriosis as a major HIV-related complication, occurred shortly after the introduction of potent antiretroviral combinations (cART) in the year 1996. In fact, until early nineties, the lack of potent antiretroviral regimens made frequent the association of this opportunism with full-blown AIDS, a mean CD4+ lymphocyte count of around 20-50 cells/μL, and extremely variable chest X-ray features and systemic presentations. The recent detection of 9 further episodes of atypical mycobacteriosis in the year 2009 was due to a late recognition of a far advanced HIV disease (the so-called "AIDS presenters"), which were already complicated by multiple opportunistic disorders.
Discussion: *M. kansasii*, *M. xenopi*, and *M. fortuitum* respiratory and/or disseminated infection continues to occur, and pose relevant diagnostic problems, including late or missed identification due to slow culture and frequently concurrent opportunistic disease. Serious therapeutic difficulties, due to the unpredictable *in vitro* antimicrobial susceptibility profile of these organisms, and the need to start as soon as possible an effective combination therapy which should not interfere with other medications (especially cART), are also of concern.

P37

Vitamin D and HIV: implications for chronic disease management

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Background: To investigate implications for management of Vitamin D deficiency in HIV disease. Vitamin D, the prohormone, is important in bone metabolism, renal disease, immune function and recently has been studied for its relationship to cardiovascular disease and cancer. Various reports have noted the prevalence of Vitamin D deficiency among HIV infected individuals.
Methods: Case studies of 17 HIV/AIDS patients in two community-based private practices in San Francisco caring for nearly 1000 HIV patients were initiated in 2009. Charts were reviewed for serum 25-(OH) Vitamin D, ionized calcium (Ca), parathyroid hormone (PTH), CD4 cell count, viral load, and lipid levels. Age, race, gender, current antiretroviral treatment, significant concomitant diseases and results of bone mineral density by DEXA scans were noted. We defined Vitamin D insufficiency or suboptimal levels as <30 ng/dl, deficiency as <20 ng/dl and severe deficiency as <10 ng/dl. Levels of 25-(OH) Vitamin D after at least 3 months of replacement with 800 IU of Vitamin D were noted when available.
Results: 65% were older than 50, 29% were between 40 and 50, and 6% were younger than 40. 76% were White, 12% were Hispanics, 6% were African-American and 6% were Pacific Islander. All were male except for one male to female transgender patient. All patients except one (Elite Controller on no meds) were on antiretroviral therapy and 100% were virologically suppressed at <75. CD4 counts ranged from 174 to 1058 with 69% > 350. All were on well balanced diets. Overall, 76% had 25-(OH) Vitamin D levels of less than 30 with 12% severely deficient, and with the lowest level of 8.9 in a 37 year old patient. 2 patients, both over 60 years of age had normal levels.
Discussion: A significant number of these patients (76%) had insufficient or suboptimal levels of Vitamin D. It is clear that Vitamin D is obtained from sun exposure, the diet or from supplements. What is unclear is what are the causes of the deficiency and what is the association of this deficiency with HIV itself, antiretroviral treatment or perhaps, premature aging in a chronically infected population.

P38

Abstract assessment of knowledge, attitude and risk practice for HIV infection among male prisoners: the case Maweni prison, Tanga region Tanzania

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Background: HIV prevalence in prisons has been reported to be higher compared to general population. However, prisoners have been overlooked and little has been done regarding HIV in this high risk group. Attitude and practice studies are crucial to acquire information that would be required to plan for the appropriate intervention programs that would be used to reduce risk practice among this high group. This study aimed to assess knowledge attitude and risk practice related to HIV infection among inmate prisoners.
Methods: This was a cross sectional analytical study design was conducted from April to May 2009. Structured questionnaires were used to collect relevant information from the participants through face to face interview. A systematic random sampling was used to select 203 male prisoners to participate in this study. Ethical approval was obtained before commencing this study from the Muhimbili college research ethics committee, permission to conduct the study was sought from prisons authority and informed consent was obtained from the study participants. Statistical Package for Social Sciences (SPSS) program version 14.0 was used to analyze data. Median range and proportions for different variables were estimated.
Results: About 100 (50.3%) of the study participants have high knowledge on HIV infection, 107 (53.8%) of the participants have negative attitude toward HIV prevention and 106 (53.2%) of the participants have HIV risk practice related to HIV infection. However risk practice have statistical significant association with knowledge on HIV infection (P = 0.001) and attitude towards HIV prevention (p = 0.02).
Discussion: Participants have high HIV risk practice which could have been contributed by low knowledge on HIV infection and negative attitude toward HIV prevention. The most important area to think about when plan for intervention should base on increase level of knowledge on HIV infection to alter negative attitude towards HIV prevention and to reduce risk practice related to HIV infection.
Recommendation: HIV health education in prison must become a public health priority to educate prisoners through.

P39

Clinical and immunological presentation of new HIV-infected patients in an outpatient clinic

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Background: Current guidelines of antiretroviral therapy (ART) recommend starting ART before an advanced immunological deterioration. The benefit of early ART is lost when diagnosis is established at an advanced stage of the disease. The aim of this study is to evaluate the clinical and immunological data at the first observation of HIV-infected patients in an Infectious Diseases Service.
Methods: Demographic and epidemiological data of all HIV-infected patients diagnosed between 01/January/2006 and 31/December/2008 were evaluated. Clinical and immunological status, AIDS defining entities and mortality rate were also analysed.
Results: 311 new patients were observed (305 HIV-1 infected and 6 HIV-2 infected), with a mean age of 42 ± 13,7 years. 218 (70%) patients were men. Risk factor for HIV-infection was sexual in 271 (87%) patients (18% were homo-bisexual). At the time of diagnosis the mean CD4+ cell count was 333 ± 305 cells/mm^3; 132 (42,4%) patients had CD4 cell count <200/mm^3. In 89 (28,6%) patients an AIDS-defining illness was diagnosed simultaneously with HIV-infection diagnosis; 43% of these were tuberculosis. After a mean follow-up of 15 ± 9,7 months 27 (8,6%) patients died, 24/27 (88,8%) with AIDS and the other 3 patients one each with eosophageal cancer, hepatic failure and MSSA sepsis,. In 22/27 (81,4%) of the deceased patients the CD4 cell count was <200 cells/mm^3 at the time of the HIV diagnosis.
Discussion: In the last 3 years, 42% of the new diagnosed patients had a serious immunological deficit. In 28% of them HIV-infection was diagnosed simultaneously with a defining AIDS opportunistic infection. The fact that 81,4% of the deceased patients had a CD4 cell count <200/mm^3 at diagnosis shows that HIV-infection diagnosis was done too late for ART to be of benefit.

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An HIV-infected patient with associated, lethal rhinopharyngeal actinomycosis and a rapidly progressing local adenocarcinoma

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Background: Solid malignancies, including those with atypical presentations, are increasing 12 years after the introduction of combined antiretroviral therapy (cART), and the differential diagnostic problems may be increased by the eventual concurrence of superinfections.
Methods: An extremely infrequent episode of associated nasopharyngeal squamous adenocarcinoma plus an underlying actinomycosis occurred in a HIV-infected male patient (p) with a previous diagnosis of AIDS, treated with cART taken with insufficient adherence, so that a satisfactory immune system recovery (as expressed by a CD4+ count persistingly

>400 cells/μL), was in contrast with a low-level persistance of detectable HIV viremia, and extensive genotypic drug resistance mutations.
Results: Interestingly, a number of local and specific risk factors for both neoplastic and infectious disorders were recognized by caregivers (tobacco smoke, long-term inhalatory substance abuse, in particular cocaine, and a half-professional mushroom-truffle search and evaluation also by systematic smelling). Although an appropriate and timely diagnostic workup carried out with repeated, combined computerized tomography, magnetic resonance imaging, and fiberoptic rhinoscopy with multiple biopsy and histopathologic studies, the final diagnosis of a combined, dual neoplastic-infectious pathology occurred only after a demolitive surgical intervention and subsequent pathology studies. Despite a correct antimicrobial therapy, and an associated radiotherapy and cytotoxic chemotherapy schedule, a rapid dissemination of multiple secondary lesions to the brain rapidly led our p to death.
Discussion: The particular epidemiological issues, and the imaging and histopathological diagnostic workup of dual illnesses of our HIV-infected p, and its therapeutic and outcome features, are presented and discussed on the ground of the available literature evidences. To the best of our knowledge, no cases of associated actinomycosis plus a local, underlying squamous cell adenocarcinoma of the same ear, nose, and throat district occurred until now in both HIV-infected and also non-HIV-infected p, so that health care professionals should take into careful consideration even a dual etiology, when facing p with rhinopharyngeal mass lesions, with multiple risk factors for different diseases.

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More than ten years without any detectable HIV viremia: exceptionally long-term non-progressive HIV infection

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Background: It is still unclear whether the virological-immunological steady-state observed among the small number of HIV-infected patients defined "long-term non-progressors" may have a prolonged-undefined duration, or it simply represents the extremity of a gaussian curve, and the progression of HIV disease is therefore an unavoidable event. These subjects are actively investigated, to understand this prolonged infection latency, so that many studies focused on the viremia levels predictive of disease progression.
Methods: A 42-year-old asymptomatic patient is followed since the age of 23 (year 1990), for her HIV infection.
Results: During periodic monitoring, carried out at least quarterly, HIV viremia always remained under the detectability threshold (<200 copies/mL until 2003, <40 copies/mL since 2004), in absence of any viremia "blip", while the CD4+ T-lymphocyte count ranged from 597 and 794 cells/μL (with a percentage of 28% to 36% of overall CD4+ T-lymphocytes), concurrently with an always contained expansion of CD8+ T-lymphocyte subset (52% to 56% during the entire follow-up). A genotypic resistance testing was never feasible, due to the persistingly undetectable viremia.
Discussion: In our patient followed since the age of 23 (year 1990), an HIV infection repeatedly confirmed by Western Blot assays was never associated with a detectable viremia, or a quantitative drop of CD4+ cell count, while the CD4+ percentage, although proportionally elevated (28-36%), always remained below that of CD8+ lymphocytes, which had only a moderate expansion (52-56%). Rodés *et al. [AIDS 2004;18:1109]* assessed five "long-term non-progressor" patients with a persistingly negative viremia during 6 years (1997-2003), also demonstrating a reduction of replication capacities related to the retrieval of R5 HIV strains, or the R77Q mutation of viral gene "vpr", while an homozygosis for the delta-32 variant of the CCRC co-receptor was not found. From the immunological point of view, the Author underlined a reduced expansion of CD8+ lymphocyte subset in these five subjects *[AIDS 2004;18:1109]*.Case reports like ours, although very infrequent and therefore not representative of the entire population of HIV-infected individuals, should deserve in-depth virological and immunological assessment, on the ground of the present, enlarged investigation perspectives, to collect further informations on the network which sustains and allows a so prolonged clinical-immunological HIV infection latency.

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Antiretroviral treatment and expression of the mRNA levels for Pgp, MRP1, MRP4 and MRP5 in HIV antiretroviral naïve patients. Follow-up at 48 weeks

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Background: The ATP-binding cassette genes represent the largest family of transmembrane proteins [including multidrug resistant proteins (MRPs) and P-glycoprotein (Pgp)] able to drive the transport of various molecules across cell membranes. Several studies have demonstrated that most of the above transporter are also able to transport antiretrovirals.
The aim of this study was to evaluate whether the antiretroviral treatment might affect the mRNA expression of Pgp and some MRPs.
Methods: Blood samples were collected from 13 HIV-positive patients treatment naïve. After the beginning of the treatment, samples were collected at 12, 24, 36 and 48 weeks. Eight patients were treated with Kaletra and Truvada (group I) and five patients with Efavirenz and NRTIs (truvada or combivir) (Gropu II). Expression of mRNA of the Pgp, MRP1, MRP4, and MRP5 was evaluated by real-time-PCR using the TaqMan technology (ABI Prism 7000; Applied Biosystems).
Results: MDR1 and MRP4 expression was not affected by treatment with PI and NRTI. In fact, at all time analyzed the mRNA levels of these transporters did not significantly differed from the mRNA levels detected before the beginning of treatment. As far as MRP1 and MRP5 are concerned, a modest, but not significant, reduction in the mRNA expression levels was observed after beginning of treatment. In patients belonging to the group II basically the same results were obtained. Looking at the individual trend of the mRNA expression of the above transporters in each patient it can be seen that the expression levels of these transporters seems to change during follow up but it is independent of type and time of treatment. The expression of the mRNA levels of these transporters appears to increase in some patients and decrease in other individuals suggesting that a high interindividual variability in the modulation of these mRNA does exist.
Discussion: Antiretroviral treatment does not significantly affect the expression levels of mRNA of transported analyzed. However an interindividual variability in the expression of these mRNA has been documented during the follow up and further studies are needed to evaluate whether the over-expression of these mRNA may affect the success of therapy.

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Clinical and immulogical outcomes among adult patients receiving Antiretroviral Therapy (ART) at an HIV/AIDS program in Uganda

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Background: To evaluate clinical and immunological outcomes and the associated factors among adult patients receiving ART at MJAP, Uganda.
Methods: A retrospective cohort analysis of patient data on ART between August 2005 and June 2009 at Mulago Hospital AIDS (ISS) clinic.
Changes in Body Mass Index (BMI) and CD4 cell count, incidence of Opportunistic Infections (OIs) and mortality were compared with the patients' socio-demographics, WHO stage and CD4 count at initiation of ART, ART regimen and adherence levels. Odds ratios, 95% confidence intervals, chi square tests, logistic regression and Cox proportional hazard model were used for analysis.
Results: Of the 4,824 patients on ART, 65% (3,120) were female, median age; 33 years (IQR: 28-40). Total follow up time was 5824.9 Person Years

(PY). Median CD4+ count and BMI at ART initiation was 138 cells/mm^3 (IQR: 60-200) and 21.2 kg/m^2 (IQR 19.2-23.7) respectively. 10.6% developed OIs, 239 (5%) patients died. Incidence of death was 4.12/100 PY (95% CI 3.63-4.68). Gain in CD4 count and BMI was observed in 68% and 63% of the patients respectively. Median increase in CD4 count was 174 cells/mm^3 (IQR 96-278) and mean increase in BMI was 1.5 kg/m^2 (S.D 2.82).
Increase in CD4 count and BMI was associated with increased income, adherence ≥ 95%, WHO stages III & IV and CD4 count < 100 (P < 0.05). The risk of developing an OI was associated with CD4 count < 100, WHO stages III & IV (P < 0.001) and decreasing levels of education (c^2 for trend = 7.7 (df) = 1, P = 0.005). Mortality was higher in patients in WHO stage III & IV (H.R 2.57, P < 0.001) and lower in patients with CD4 count ≥100, (H.R 0.32, P < 0.001) and adherence ≥ 95% (HR 0.55, P < 0.001).
Discussion: Early initiation of ART, good adherence, improved income and education status are associated with increased survival and positive clinical and immunological outcomes among patients on ART.

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Recombinant soluble CCR5 AND CXCR4 chemokine receptors as anti-HIV drug targets

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Background: The aim of current work was to produce recombinant soluble CCR5 and CXCR4 chemokine receptors that could be used in screening of potential HIV-1 inhibitors.
Methods: Recombinant DNA constructs were produced by using polymerase chain reaction (PCR) technique and cloning. Proteins were expressed in bacteria and purified using immobilized metal ion affinity chromatography. To validate functionality of the recombinant proteins immunoprecipitation, immunblot and ELISA assays were performed.
Results: We designed recombinant soluble CCR5 and CXCR4 proteins where functionally important regions of native receptors were connected with artificial linkers. These proteins were expressed in *E.coli* Origami 2(DE3) cells, refolded and purified. We have shown that recombinant proteins are functionally similar to native receptors since they bind to specific anti-CCR5 and anti-CXCR4 antibodies in immunoblot, immunoprecipitation and ELISA. Using the recombinant proteins for immunization we have obtained specific rabbit polyclonal serum. Finally, we established a competitive ELISA assay to search for the inhibitors of antigen and antibody binding. By applying this assay we perfomed screening of combinative drug library and found potential inhibitors.
Discussion: It is well known that CCR5 and CXCR4 chemokine receptors play a central role in the mechanism by which HIV binds to and enters white blood cells, and therefore represent key targets in the search for effective novel treatments for HIV infection and AIDS. The current results indicate that recombinant soluble chemokine receptors are functionally active and can be used in screening of potential HIV-1 inhibitors.

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Two recent, fixed associations of antiretroviral nucleos(t)ide analogues. A prospective assessment of their therapeutic use in HIV disease management: a field study

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Background: The introduction of novel, fixed NRTI combinations (emtricitabine-tenofovir, E-T, and lamivudine-abacavir, L-A), expanded the available spectrum of antiretroviral formulations, and indirectly increased patient's adherence, since both these combinations are taken as a one pill-once daily regimen.
Methods: A prospective survey of the use of these two fixed NRTI combinations was performed in our cohort of over 1,700 HIV-infected patients (p).
Results: During 12 consecutive months, 334 p received for the first time E-T (262 cases), or L-A (72 p). Among the 88 p naïve to all antiretrovirals, E-T was given to 66 p (75.0%), mostly associated with efavirenz (51 p), or different PI combinations (15 p), whereas L-A was administered to 22 p only (in 18 of them in association with PI). In the remaining 246 p, E-T or L-A therapy replaced a prior regimen, predominantly associated with PI (141 cases p), *versus* efavirenz (48 p), or oher combinations (57 p). Among the 246 pre-treated p, E-T (194 p), still prevailed over L-A (50 p), and the therapeutic change was due to failure and resistance (89 p), and in the majority of cases to toxicity or poor tolerability (146 p). Both fixed NRTI combinations were well tolerated, with only three cases of L-A suspension due to abacavir hypersensitivity, and two cases of E-T interruption due to kidney abnormalities.
Discussion: From our preliminary experience, a major role seems played by E-T in first-line treatments (preferably among "compact" regimens based on efavirenz), while the apparently increased L-A prescription to pre-treated p is attributable to the different genetic barrier of abacavir (which is often introduced in association with PI). The present availability to two more fixed NRTI combinations advantaged by once-daily administration strongly encourages further "head to head" studies in both first-line and experienced p, in order to better exploit and target their therapeutic potential and their convenience features.

P46

Factors associated with non-adherence to HAART in HIV-positive pregnant women during pregnancy, peri- and postpartum in Lima, Peru

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Background: To determine factors associated with non-adherence to HAART in HIV-positive pregnant women.
Methods: We used adherence data collected by the LILAC study in Peru based on the AACTG questionnaire, in HIV-positive women during pregnancy, peripartum and 2 months postpartum.
Results: We studied 44 women. The mean age was 27,9+5,9 years. At enrollment 86,4% were on HAART, although at delivery 100% had received HAART; of whom 27,3% for PMTCT. Majority (61,4%) were in their first HAART regimen, and the most commonly used was 2NRTI+2PI (56,8%) followed by 2NRTI+1NNRTI (34,1%), with a median of 2 tablets per day, and for an average of 31,1+41,5 months. During pregnancy, non-adherence was 7,3%, and increased in the peri- and postpartum (21,2% and 15%, respectively). More women (34,1%) reported missing a dose during pregnancy, 27,3% in peripartum and 21,9% at 2 months postpartum. Among the most common reasons cited for missing doses were "being away from home" (76,7%) and "a change in routine" (70%). Among those who had not missed a dose, women reported having trouble taking the medications during pregnancy (11,1%), at birth (3%) and 2 months postpartum (13,3%). The factors associated with non-adherence in the peri- and postpartum were taking more than 2 pills a day (p = 0,027) and a longer duration of HIV infection (p = 0.002) respectively. Factors associated with ever having missed a dose in the peripartum were taking more than 2 pills a day (p = 0.005) and taking a regimen that included PI (p = 0.021); and in the postpartum working outside the house (p = 0,047).
Discussion: Adherence to HAART decreases markedly in the peri-partum, by the change in routine that involves the cesarean section, and in the postpartum possible because women have to work outside the house. Further studies that consider others factors such as psychological are needed. The small sample size was a limitation of the present study.

P47

Treatment of HIV-2 infection with ritonavir/lopinavir: results at 60 months. A single-center study of 9 patients

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Background: In vitro and in vivo data have suggested that ritonavir/lopinavir is one of the most potent protease inhibitor against HIV-2 infection. Nevertheless, only few clinical reports have been published. So, it seemed to us important to report our clinical experience in 9 patients.
Methods: We searched in our database patients with HIV-2 who have been treated with ritonavir/lopinavir. 9 patients (4 women and 5 men) were identified. Clinical and biological evolution was analyzed.
Results: At the time of initiation of lopinavir, median age was 54 (37-66). Median duration of HIV infection was 41,6 months (6-101). Previous median duration of treatment was 29 months (4-51). No patient was naïve of treatment. The median previous number of regimens received was 2 (1-2). 5/9 patients were naïve of protease inhibitors. Initial median CD4 cells were 150 (68-478). Viral load (VL) was under the limit of detection in 6 cases. Reasons for switching to lopinavir were toxicity or intolerance (n = 2), absence of efficacy on VL (n = 3) or inadequacy of previous treatment (n = 4). In all cases, lopinavir was given in association of 2 NRTIs.
After a median follow-up of 60 months (18-84), no severe side effect was observed. VL under the limits of detection was obtained for all patients except for one patient with a very poor compliance. In this later case, after switching lopinavir to raltegravir, VL remained undetectable. CD4 increased in all cases. The median gain of CD4 from baseline was + 234 (+50 - +472).
Discussion: Because of the very low frequency of the disease in western countries, data on the treatment of HIV-2 infection are uncommon. For these reasons, treatment of HIV-2 patients remains difficult. So, reports on therapeutic options are very important. Even with a modest number of patients, we confirmed the excellent long-term efficiency of lopinavir for the treatment of HIV-2 patients.

P48

Efficacy and Tolerability of RAL, MVC and ETV used in combination in the treatment of highly treatment experienced HIV infected patients

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Background: Although the durability of antiretroviral (ARV) efficacy has improved, mainly due to better tolerability, ease of administration (adherence) and potency, some patients still encounter virological and immunological treatment failure. These patients have been on multiple regimens containing nucleoside reverse transcriptase inhibitors (NRTIs), non-nucleoside reverse transcriptase inhibitor (NNRTI) and Protease Inhibitor (PI) and have become resistant to one or more of these classes. Hence, the need for salvage therapies using newly approved antiretrovirals that are NRTI, NNRTI and PI sparing is a clinical reality. Raltegravir, maraviroc and etravirine are 3 new agents with distinguished characteristics which are now available however guidelines for their use continue to evolve and none exist yet on using them all in combination. In this study we reviewed patients who were four ARV class experienced and on salvage regimens consisting of a combination of raltegravir, etravirine and maraviroc.
Methods: We retrospectively evaluated pts who had documented failure to 3 or 4 different classes of ARVs and were treated with combination of RAL, MVC and ETV. Salvage therapy was prescribed according to: viral tropism, screening genotype and previous resistance tests. Our inclusion criteria was 1) 3 or 4 ARV class experienced, 2) CXCR5 on troFile assay 3) on a regimen containing maraviroc, raltegravir and etravirine together. Data was reviewed and analyzed at baseline and at 24 weeks.
Results: 9 out of 5500 charts fit this inclusion criterion. The mean age was 50 yeras-old (34-62) with majority being male (7/9, 78%). All pts were integrase-inhibitor (RAL) naive and harboring CCR5 tropic virus. The median ETV mutation score was 2.5 (range 0-2.5). The median CD4-cell count and HIV-RNA levels were respectively 200 cells/mm^3 (range: 19-734) and 4.4 $\log_{10}$ copies/mL (2.3-5.6) at baseline. At week-24 the CD4-cell count was 389 cells/mm^3 . The percentage of pts achieving HIV-RNA levels <400 $\log_{10}$ copies/mL and <50 $\log_{10}$ copies/mL at week 24 were respectively 78% (7/9) and 67% (6/9). In the 2 patients that did not achieve <400 $\log_{10}$ copies/mL HIV-RNA levels poor ARV adherence was documented. All pts tolerated this regimen well with no discontinuation due to adverse events up to 24 weeks.
Discussion: The combination of RAL, MVC and ETV is safe and highly efficacious in the management of HIV-treatment experienced patients with excellent CD4-cell count recovery and complete viral load suppression with good ARV adherence.

P49

Outcome of first line antiretroviral therapy (art) with respect to treatment Failure at ART CENTRE, B. J. Medical College, Civil Hospital, Ahmedabad

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Background: To Study outcome of 1st Line Anti Retroviral Therapy (ART) at with respect to treatment failure at ART Centre, B. J. Medical College, Civil Hospital, Ahmedabad, Gujarat, India.
Methods: All the ever started 1st Line ART patients were evaluated for treatment failure as per Indian National ART Guidelines at our institute.
Results: ART Centre, B. J. Medical College, Civil Hospital was started on 25th April, 2005. Till October 2009 ever registered patients are 11827 among this total 5847 patients were initiated ART as per Indian National ART Guidelines, Table 1.
Discussion: The clinical goals of HIV treatment are optimally accomplished through consistent high-level adherence to Highly Active Anti Retroviral Therapy (HAART) and sustained virological suppression through cost effective regimen (2NRTIs+1NNRTI). At the end of more than 4 years total suspected IF patients are only 73 (2.13%) of which only 49 (1.70%) has actual virological failure; shows the efficacy of the 1st Line ART. Failure to access care and discontinuation of or non-adherence to ART are the most important factors associated with the progression of HIV disease.

Table 1 (abstract P49)

Parameter	Outcome
Total Patients initiated ART	5847
Total Patients alive on ART at the end of October 2009	2880 (49.25%)
Immunological Failure (IF) patients (n = 3414; Excluding Deaths and Transfer Out)	73(2.13%)
Referred for Plasma Viral Load (PVL)	68 (94.4%)
PVL < 400 copies/ml (A)	19 (28%)
400 - 10,000 copies/ml (B)	06 (8.8%)
>10,000 copies/ml (C)	43 (63.2%)
Total patients failing as per Virological Failure (B+C) (n = 2880)	49 (1.70%)

P50

Analysis of protease treatment-associated mutations in a group of HIV-1 subtype F infected individuals with two sequences obtained in different time points

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Background: Several studies have been performed exploring HIV-1 subtype B antiretroviral resistance mutation profiles. Nonetheless, data for other HIV-1 subtypes is scarce. The aim of the present work was to analyze the subtype F protease antiretroviral related mutations.
Methods: Data from 38 HIV-1 subtype F individuals with sequences obtained in two different time points were analyzed. All patients presented virological failure to a protease-inhibitor containing regimen. The average of time between the first and the second sequence was 1.5 year. Most patients had changed therapy at least once between the first and the second genomic sequence. Protease subtyping was confirmed by phylogenetic analysis. T-pared test was performed to verify possible differences in the total number of mutations and in the number of primary mutations when comparing the group containing all first sequences and the group containing all second sequences. We searched for all primary mutations present in the group of first sequences. Once a primary mutation was present in the first sequence, we verified its presence after changing therapeutic regimen.
Results: T-pared test showed that number of total mutations increased after one or more therapy changing. The number of primary mutations stayed stable, but once a primary mutation was present in the first sequence, it was commonly found also in the second sequence. That was the case of D30N, N88D, L90M and V82A.
Discussion: Our results suggest that increasing the number of total mutations after changing therapeutic schema is a trend, as well as the persistence of some primary mutations even after at least one year under a new Protease Inhibitor related selective pressure.

P51

Efficacy and safety of TDF+FTC+EFV in naïve patients initiating HAART; an observational study comparing Atripla Vs Truvada/Sustiva exposure

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Background: Co formulation enhance adherence in HIV infected patients who initiate HAART. Aim of this study was to compare the efficacy and safety of Atripla vs Truvada plus Sustiva in naive HIV-infected patients.
Methods: This prospective and observational study was conducted at a referral outclinic in Madrid. All consecutive patients who initiated TDF+FTC+EFV coformulated (Atripla) as a first regim were compared to patients who initiated TDF+FTC (Truvada) plus EFV (Sustiva). Primary outcomes were time to viral suppression and immune recovery, assessed quarterly during regular follow up. Secondary outcomes were classified as drug-related adverse events and AIDS related events. Statistical analyses were realised using the Chi-square and the T-student test for categorical and continuous variables, respectively.
Results: The study population was composed by 33 patients included in the cohort of patients treated by Atripla and 72 patients included in the cohort of patients treated by Truvada plus Sustiva. In the Atripla cohort, mean age was 37,4 years, 97% were men, 46% were native Spaniards, 54% were MSM and 6% had a HCV coinfection. In this cohort, mean nadir CD4 count was 291 cells/μL and mean viral load at baseline was 4,3 log. All variables were similarly distributed in both cohorts except for liver stiffness: 18% had a F4 Metavir score in the Atripla group and any case at this stage was reported in the Truvada+Sustiva group. RT sequencing was available at baseline for 67% in patients initiating Atripla and for 75% in the other group. No major mutations were found and distribution was similar for minor mutations. All patients in both groups achieved viral suppression but time was inferior in the Atripla group (2,7 months vs 4,5, $p < 0,005$) and CD4 recovery was superior; 163 vs 148 ($p < 0,005$). In both groups, 15% of patients presented CNS-related adverse events. It is noteworthy that one patient treated by Tuvada plus Sustiva presented overdose of EFV.
Discussion: In current clinical practice, TDF+FTC+EFV coformulated was as effective and safe as TDF+FTC and EFV.

P52

Fusion inhibitors and their evolving role during salvage antiretroviral therapy. Seven years of experience with enfuvirtide

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Background: The need of rescue antiretroviral regimens is progressively increasing, due to the unavoidable long-term emerging of multiresistant HIV strains.
Methods: An open-label study featuring the administration of the fusion inhibitor enfuvirtide (T-20) as a part of salvage anti-HIV regimens in a cohort of hardly pre-treated and multiresistant patients (p) with advanced HIV disease, followed until at least three consecutive years, is presented.
Results: The efficacy and safety parameters of enfuvirtide adjunct to an optimized background were assessed monthly in 21 severely compromised p, with a baseline viremia ranging from 64,000 to over 500,000 HIV-RNA copies/mL, and a CD4+ lymphocyte count ranging from 11 to 213 cells/μL. At the time of enfuvirtide introduction, the background antiretroviral therapy was modified according to both genotypic-virtual phenotypic resistance assays, but only 15 of 21 p could rely on at least one *in vitro* effective drug (during the first years of our experience, raltegravir, maraviroc, etravirine, riplpivirine, and also tipranavir and darunavir were not still available). Anyway, a rapid and significant drop of viremia (at least one Log_{10} HIV-RNA copies/mL), associated with a 30-280% increase of CD4+ cell count *versus* baseline values was observed in all p, although 13 our of 15 p who could rely on an optimized background had a sustained response (12-36 months). In 11 p, a surprising dissociation between a favorable virological response and a progressive loss of CD4+ cells was observed. Although frequent, local injection site adverse effects never represented the major cause of enfuvirtide interruption.
Discussion: Expectations and concerns on the use of enfuvirtide as a novel anti-HIV compound in daily practice are still debated, since no specific recommendations have been produced (especially with regard with the novel, available compounds), when excluding the assumption that enfuvirtide appears significantly more effective when administered concurrently with at least 1-2 other active antiretrovirals, although the reported experiences often included p on very advanced salvage therapies. The management of the frequent site injection reactions represents an adjunctive concern for these multi-problematic HIV-infected p.

P53

Our experience in second line Anti Retroviral Therapy (ART) At State Aids Clinical Expert Panel (SACEP) Clinic, Centre of Excellence (CoE), Art Centre, B. J. Medical College, Civil Hospital, Ahmedabad

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Background: To share our experience in second line Anti Retroviral Therapy (ART) at State AIDS Clinical Expert Panel (SACEP) Clinic, Centre of Excellence (CoE), ART Centre, B. J. Medical College, Civil Hospital, Ahmedabad, Gujarat, India.

Table 1 (abstract P53)

Parameter			Outcome
Total Patients referred to SACEP			127
Patients Eligible and referred for Plasma Viral Load (PVL) (n = 127)			122(96.06%)
Patients recommended 2nd Line ART after PVL (n = 122)			75(61.47%)
Patients initiated Free 2nd Line ART (n = 75)			69(92.0%)
Patients under follow up TB Treatment/Counseling to initiate 2nd Line ART			6
Adherence Counseling for 2nd Line ART Patients (n = 69)			69(100%)
Undetectable PVL (<400 copies/ml) after 6 months of 2nd Line ART (n = 28)			24
Therapeutic Success according to Undetectable PVL (24/28)			85.74%
No. of Death after initiation of 2nd Line ART (n = 69)			8(11.59%)
Death Analysis	Cause of Death	WHO Stage/PVL/CD4	n = 8
	Accidental	T3/560459/17	1
	Renal Failure	T3/208048/230	1
	Malignancy	T4/384469/38	1
	Opportunistic Infections (OIs)	T3/46453/67	4
		T4/507495/26	
		T4/1166325/32	
		T4/289375/21	
	Unknown Reason	T3/4095718/29	1

Methods: First line treatment failure Patients (as per Indian National ART Guidelines) were referred to SACEP Clinic for further evaluation and if eligible as per Indian National 2nd Line ART Guidelines; enrolled and followed up for initiation of 2nd Line ART at our institute.
Results: Provision of Free 2nd Line ART to People Living with HIV/AIDS (PLHA) Program by National AIDS Control Organization (NACO), India was started in November, 2008 under SACEP Clinic for Gujarat and Rajasthan PLHAs. Till October 2009 total 127 1st Line ART Failure patients were referred to SACEP Clinic, Table 1.
Discussion: Switching to 2nd Line ART based on Immunological Failure, Clinical Failure is not recommended; as out of 122 PLHAs analyzed only 75 PLHAs have Virological Failure. Adherence Counseling for 2nd Line ART helps quite in reduction of PVL. Deaths analysis shows that late switch to 2nd Line ART at CD4 < 100 cells/mm^3 may not result in desired therapeutic goals.

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Treatment outcomes in patients receiving combination antiretroviral therapy

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Background: This study investigated mortality rate, early CD4 responses, pattern of ARVs substitutions and medication adherence of HIV-infected patients on first-line triple combination ART.
Methods: Assessment of 196 HIV-infected patients on combination ART regimens was performed after 18 months of therapy. Medication adherence assessment of 69 follow-up target groups was based on a study-specific questionnaire. Paired sample *t-test* and simple linear correlation were used to test the association of the CD4-cell Counts at different time intervals. Kaplan-Meier model used to assess survival functions and the log-rank test was used to assess statistical difference at 95% CI.
Results: Mean age of participants was 33.6 years (95%CI, 32.05-35.15); 67.9% were females. At ART initiation, 27.0% were at WHO stage II, 47.0% at stage III. Mortality rate (N = 196) was 20.32 deaths per 100 patient-months; 31.6% occurred in <30 days while 52.6% occurred post 120 days of treatment. The meanCD4-cell count (cells/mm^3) at ART initiation was 179.2 which increased to 328.5 at 3 months, 325.6 at 6 months, 357.4 at 12 months, and 366.7 at 18 months, ($p < 0.01$). Patients started on Stavudine-based or Efavirenz-based regimens were considerably more likely to have that drug substituted, compared to patients started on Zidovudine-based or Nevirapine-based regimens. 73.8% level of adherence was reported after 18 months on ART.
Discussion: In this setting, patients receiving ART showed significant improvements in CD4-cell status but adherence level was relatively poor. A large proportion of the patients presented very late for treatment with very poor baseline parameters. This finding supports the need for a rapid scale-up of counselling and testing for early detection of asymptomatic cases in developing countries. Patients were more stable on Zidovudine-based or Nevirapine-based regimens compared to Stavudine-based or Efavirenz-based regimens. Early mortality rate was high probably due to poor baseline parameters or late presentation for treatment, indicating need for early interventions. The follow-up CD4-cells measurements were not done every 6 months for all patients eligible for repeat CD4-cell Count in accordance to the Nigerian national ART guidelines, hence the need for improvement in the adherence to the national treatment guideline.

P55

Lopinavir/r + Efavirenz combination as a potent NRTI sparing antiretroviral therapy

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Background: Classical recommended HAART are NRTI+PI or NRTI+NNRTI combinations. However, some of our HIV patients have already long durations of NRTI exposure with confirmed or beginning NRTI toxicities. NNRTI+PI combinations could be proposed to avoid further exposure to NRTI.
Methods: We review the data of 19 patients heavely preexposed to NRTI and submitted to a Lopinavir/r + Efavirenz (Lpv/r 2 × 3 tablets 200 mg/50 mg/d + EFZ 1 × 1 tablet 600 mg/d) combination. Results are expressed as means ± sem.

Results: Nineteen patients (7 F/12 M; 8 Africans, 1 Asiatic, 11 Caucasians) were submitted to Lpv/r+EFZ during a total period of 618,5 patient-months (33 ± 5 months). These patients were 47 ± 2 years old and are regularly followed for their HIV seropositivity since 123 ± 9 months. Duration of exposure to any ART before inclusion in this study was 92 ± 10 m. Three patients discontinued EFZ rapidly for allergic reactions (2) or psychiatric deterioration (1), and one discontinued after 16 m due to persistently sleep disturbances. Seven patients discontinued for hyperlipemia after 30 ± 5 m. Viral loads remained undetectable (PCR<5 copies RNA/ml) and % CD4 continued to increase to reach a mean value of 25 ± 2%.
Discussion: The long term follow-up of this small cohort heavely pre-exposed to NRTI suggests that Lpv/r+EFZ is a potent antiviral therapy and that further NRTI combinations therapy can be avoided in these patients. The tolerance to each drug seems not modified by their use when combined. However, hyperlipemia is a regular preoccupation.

P56

Experience with art adherence couselling at muhimbili national hospital, Dar es Salaam, Tanzania

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Background: In July 2004, the Tanzania of Health initiated Pilot care and treatment program at the Muhimbili National Hospital. (MNH). The pilot fg program was designed to inform the National Scale – up antiretroviral therapy (ART). The goal of the pilot program was to initiate 3000 patients on ART over a period of three months. We report our initial experience in offering adherence counseling to these clients.
Methods: Clients were seen at the MNH HIV/AIDS clinic from July 2004 to October 2004 all eligible patients were offered ART adherence counseling prior to therapy initiation and thereafter at every re-fill appointment. ART was initiated only after both the counselor and clients having been satisfied with the readiness of the clients to start the therapy.
Assessment of degree of adherence was by the self-report as well as pharmacy re-fill performed quantitavely, while patients attitudes to the exercise and problems associated with offering adherence counseling were ascertained using qualitative methods.
Results: By 30th of September 2004, 1,286 patients were enrolled in care and 881 patients were put in ART having undergone adherence counseling. More than 65%of the enrollment occurred in the first eight weeks of the pilot program, indicating strong demand for HIV care and treatment services among people living with HIV/AIDS (PLWHA) in Dar es Salaam. By the end of October 2004, a total of 1655 patients were in care and 1,172 (70.82%) on ART. Patients on ART included 59% women 31% men and 10% children. The loss to follow-up rate was 11%.
Overall, clients reported good satisfication with the quality of care offered at the clinic, and 85% of clients demonstrated good understanding on issue pertaining adherence to ART, at follow-up, it was noted clients achieved 95% adherence with ART. However, long clients waiting – times merged as a significant problem when the clinic caseload exceeded 200 visits per day.
Conclusion: The achievements and experience of the MNH ART clinic showed that good adherence is possible in a resource poor setting with extreme staff shortages and should be started early before. Initiation of ARV therapy.
However, as such programe scale-up, they should be prepared to face up huge practical challenges.

P57

Interventions to promote adherence to Antiretroviral Therapy (HAART) among adult patients at an HIV/AIDS program in Uganda

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Background: To document successful interventions for promoting Adherence to ART among adult patients at Mulago hospital AIDS (ISS) clinic.
Methods: A retrospective cohort analysis was performed on data of 2,521 adult patients on ART for ≥ 1 at Mulago hospital AIDS (ISS) clinic between August 2005 and June 2009. The average adherence score for each patient was used for analysis. Adherence to ART was defined as good adherence for patients taking ≥ 95% of the prescribed ART doses and poor for patients taking < 95% of the prescribed ART doses.
Results: Of the 2,521 patients active on ART, 65% (1,638) were female. Median age was 34 years (IQR: 29-40). Median CD4+ cell count at ART initiation and Body Mass Index (BMI) were 134 cells/mm^3 (IQR: 56-198) and 21.2 kg/m^2 (IQR 19.3-23.5) respectively.
Good adherence of ≥ 95% was observed in 2,406 (95.4%) of the patients
Discussion: High rates of adherence to ART can be achieved in a setting with multiple interventions for adherence promotion.

P58

Lowering HIV fitness and replication rate by administration of lamivudine alone, in extensively resistant HIV-infected patients, as a "bridging" strategy towards optimized salvage regimens

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Background: HIV-infected patients harbouring a lamivudine-resistant virus, seem to take benefit from a continued lamivudine monotherapy, versus combined antiretroviral treatment (cART) interruption, since a reduced HIV replication is selected by the maintenance of lamivudine-related M184 mutation. The mid-term outcome of isolated lamivudine therapy in multi-drug-resistant patients with very restricted therapeutic options, waiting for novel drug classes, is reported.
Methods: Six patients aged 23-49 years (4 males and 2 females, one of them with perinatal infection), with HIV disease treated since 13.8 ± 6.2 years with 10.3 ± 4.7 therapeutic lines, experienced repeated virological-immunological failures due to an extensive HIV genotype resistance, which finally led to a complete 3-class resistance, and no residual therapeutic options, when excluding the use of a fusion/integrase/co-receptor inhibitors, without the possibility to optimize the therapeutic background. A concurrent toxicity was also present:combined lipodistrophy syndrome, dyslipidemia, and insulin resistance (3,2, and one patients, respectively).
Results: At the time of lamivudine monotherapy initiation, the median viremia was 36,000 HIV-RNA copies/mL, while the median CD4+ count was 344 cells/μL. Despite a previous diagnosis of AIDS in 4/6 patients, at the time of therapeutic switch the clinical situation was stable. During the monthly follow-up with lamivudine monotherapy, ranging from 8 to 24 months (mean 9.9 ± 5.2) months, no HIV-associated signs-symptoms occurred, previous cART-associated laboratory toxicity significantly ameliorated, and no significant differences were found as to virological-immunological markers of HIV disease. A fluctuating viremia was noticed in all cases, with a median value at the end of follow-up of 44,000 HIV-RNA copies/mL, while no significant loss of CD4+ count occurred (median final levels:322 cells/μL). Two-four nucleos(t)ide mutations, and 2-5 protease mutations were deselected during the follow-up, but the M184 mutation remained. All these patients were allowed to re-introduce a cART with novel drug classes, according to the availability of an optimized therapeutic background in the subsequent months.
Discussion: When extensive HIV resistance does not leave therapeutic options, lamivudine monotherapy performed with a strict monitoring in clinically stable patients with no compromised virological-immunological figures, is a potentially safe choice. Waiting for the novel cART associations, the exploitment of lamivudine resistance on HIV replication-fitness represents an ultimate therapeutic approach to these difficult-to-manage subjects.

P59

Impact of maraviroc on immune restoration in an advanced stage HIV-infected patient

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Background: Maraviroc is a CCR5 antagonist with clearly demonstrated virological efficacy in patients refractory to prior treatment and infected with an R5-tropic virus.
This antiretroviral drug would appear to have a special immunomodulatory property, given the significantly higher increase in CD4 count in patients treated with maraviroc in clinical trials.
We report the case of a severely immunocompromised patient in which the introduction of maraviroc reduced viral load to below the threshold of 40 copies/ml, with an increase in CD4 of over 100 in the space of 17 months.
Methods: The patient was a 53 year old man diagnosed with HIV in 1987, and classed stage C according to CDC classification for cerebral toxoplasmosis in June 1995. His history includes cerebral histoplasmosis in April 1997 (relapse in April 2006) and stage IV Hodgkin's lymphoma with visceral involvement in October 2006 (CD4 = 1/mm^3). The first antiretroviral treatment was given in October 1995 (CD4 <50/mm^3), and the first maraviroc-based combination therapy was his 28th line treatment (16 stoppages for treatment failure). In January 2008, the tropism test (trofile test) gave an R5 profile and the genotypic resistance test (ANRS algorithm July 2009) showed, for the RT gene, resistance to AZT, 3TC, D4T, ddI, ABC, EFV, NVR and possibly to TDF and ETV; for the protease gene, resistance to IDVr, SQVr, NFV, FPVr, ATV, DRVr, LPVr and possibly to TPVr.
Results: Table 1 shows viro-immunological changes with treatment:
Discussion: This case report shows the possibility of being confronted with an R5-tropic virus after 21 years' progression, and 15 years of antiretroviral treatment, with severe immunosuppression. The three successive maraviroc-based combination therapies lead to an increase in CD4 count independent of the virological response, with a delta of +120 CD4 after 17 months. A parallel increase in the CD8 count was also observed, as has been reported in clinical trials.

P60

"Self-managed", inadequate "adherence" to antiretroviral therapy, limited to one half of standard dosages, followed by an unexpected, sustained virological and immunological success

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Background: Antiretroviral adherence issues are essential for successful and sustained efficacy, and viral resistance prevention.
Methods: One exceptional case regards an ex-IVDA patient (p) with HIV infection known since 1985. Due to a severe HIV-related immunosuppression (CD4: 37 cells/μL), in 1997 3TC, d4T, and indinavir was effectively started, achieving after 3 months undetectable viremia and a CD4 count of 315 cells/μL, but recurring urolithiasis recommended a therapeutic shift. Since April 1997,3TC, d4T, and ritonavir were suggested for 5 years, followed by 3TC, d4T, and lopinavir-ritonavir (10 months), and 3TC, AZT, and lopinavir-ritonavir (6 years). However, all proposed regimens were voluntarily taken by our p (whose body weight was 75-80 Kg) at half-dose, as a single daily dosage, against any recommendation, although our p always maintained his "adherence" to his self-made regimen, as assessed by monthly visits, direct drug distribution-accountabilty, and adherence questionnaires. Surprisingly, viremia remained for 12 years at non-detectable values (save one single detection of 1,260 HIV-RNA copies/μL), so that a genotypic resistance testing was never feasible, while CD4 count ranged from a nadir of 382 cells/μL (year 2001), to 525-794 cells/μL since 2003.
Results: A second male p with a body weight of 69-73 Kg, since 2002 took all combined antiretroviral therapy at half dosage (3TC 150 mg/day, AZT 300 mg/day, and lopinavir-ritonavir 2 cp/day for 7 years, as a single daily dose), without showing detectable viremias, and CD4 counts >500 cells/μL. A third 48-y-old male, after two changes of antiretroviral regimens due to dysmetabolism, started the fixed dose AZT-3TC-abacavir combination at half dosage (one pill/day), and since 2003 had a persistingly negative viremia, and a CD4 count always >650 cells/μL. In both these last 2 p, genotypic resistance testing was not feasible (undetectable viremia).
Discussion: Although recognizing the limitation of anecdotal observations, and our impossibility to resort to resistance testing and therapeutic drug monitoring, however the long-term maintenance of an excellent virologic-immunological situation in 3 p with an adherence voluntarily limited to 50% of recommended dosages depite all counselling, deserves discussion. A 50% compliance is considered absolutely inadequate in HIV disease treatment. Anyway, all our 3 p are somewhat "adherent" to their 50% dosage regimens, and are re-enforced in their wrong consideration by checking every 3 months their excellent clinical-laboratory situation, and by their long-term, unchanged therapy response. Health care professionals are embarrassed in discussing this inappropriate mode of antiretroviral seld-administration, but lack of supporting elements to opposite to the strongloy radicated p's thoughts.

Table 1 (abstract P59)

	D0	M3	M6	M12	M15	M17
VLlog/ml	3.83	2.63	5.68	1.6	1.6	1.6
CD4/mm^3	82	138	132	147	207	200
CD4%	11.4	13.5	13.1	12.2	15.1	15.1
CD8/mm^3	346	445	524	596	661	575
CD8%	48.3	43.7	51.7	49.5	48.3	43.5
MVC add to	FTC/RAL/ETV 1 month	TPVr/ETV 5 months	TVD/DRVr/ETV 12 months			

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With out a quality management tool, all HIV Managers shall not realise any steps forward for improvement: a Taso Uganda experience

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Background: To provide direction of what needs to be done and how they will be accomplished. To set a framework for holding the HIV programme and service providers accountable for the quality of patient care.
It's a basis for self–evaluation for next cycle of improvement.
Methods: Start with your baseline assessment from where a quality statement is chosen.
The materials include Quality statement, quality program, performance measurement system, determining quality improvement goals, stakeholders & patient participation, an implementation plan and evaluation.
Determine an active case load where a sample is got through a computer automated system or manual lists.
Using performance indicators, patient's files sampled are reviewed by the quality team.
Results are shared and discussed with the quality team, staff and then another quality statement can be made.
Though many performance gaps can be identified the rule of one is employed in selecting the quality statement.
Results: The quality statements focused on "Improving CD4 monitoring" TB assessment and continuity in care and below are the results from the follow up.
Baseline 30% in sample of 100 (active Caseload = 5392) and from the follow up CD4 improved to 33% in a sample of 107 (active Caseload = 5,660).

Continuity baseline 90% follow up = 98%.
TB assessment (baseline 92% follow up = 99%.
Discussion: There was improvement in all the areas of focus due to interventions with 3% in CD4 monitoring, 8% improvement continuity in care and 6% improvement in TB assessment. Any improvement has to be appreciated by the team. For continued and sustained quality HIV programme service delivery, its very essential period monitoring and evaluations through small scale internal surveys. All HIV service delivery units should have a quality management tool.

P62

Cystatin C and cardiovascular risk in HIV infected patients

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Background: HIV infection, combined antiretroviral therapy (cART) and metabolic syndrome are associated to an increased cardiovascular risk (CVR). Cystatin C, a low molecular weight cysteine protease inhibitor involved in vascular extracellular matrix remodelling, is considered a novel marker of kidney function and CVR in general population. To value the role of cystatin C in HIV infected patients without chronic kidney disease treated with cART subdivided for CVR.
Methods: 56 Caucasian HIV+ cART treated patients without chronic kidney disease were subdivided by means of Framingham score into two groups: A) 15 patients with "high" CVR, and B) 41 patients with "low" CVR. Viro-immunological parameters, triglycerides (TGs), total cholesterol (TCh), HDL (HDL-C) and LDL (LDL-C) cholesterol, blood pressure, microalbuminuria, GFR-MDRD, fasting glucose, insulinemia, HOMA-IR, CRP, cystatin C, IL-18, IL-6, body mass index (BMI) and waist-to-hip ratio (WHR) were measured.
Results: The two groups were homogeneous for years of cART, CD4- and CD8- T cell count, viral load. Group A showed statistically higher levels of BMI ($p < 0.009$), WHR ($p < 0.001$), TCh ($p < 0.001$), LDL-C ($p < 0.001$), TGs ($p < 0.02$), systolic blood pressure ($p < 0.001$), fasting glucose ($p < 0.002$), insulinemia ($p < 0.008$), HOMA-IR ($p < 0.001$), cystatin C ($p < 0.001$), microalbuminuria ($p < 0.001$), IL-6 ($p < 0.03$) and IL-18 ($p < 0.01$). The GFR-MDRD and the inflammatory index CRP did not show significant differences between the two groups. Finally, data showed a positive correlation between CVR and cystatin C ($r = 0.39$ and $p = 0.003$).
Discussion: In the present study it is remarkable that patients in "high" CVR group showed higher values of cystatin C and microalbuminuria even though kidney function was normal. Also microalbuminuria, besides its relation with progressive renal damage, has been found to be an independent factor of CVR and mortality in the general population. Furthermore, the positive correlation between CVR and cystatin C found in this study for the first time in HIV positive patients, is in accord with the evidence existing in literature of a strong association between cystatin C and CVR in the general population. These findings may indicate that cystatin C and microalbuminuria could serve as early markers of enhanced CVR, besides of renal complications, in the HIV-infected population.

P63

Effects on insulin sensitivity and hepatic safety of Atazanavir in HCV/HIV coinfected patients versus HIV monoinfected: A prostective 48-week study

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Background: Protease inhibitors based antiretroviral therapy has been associated with elevated plasma insulin levels and insulin resistance. HCV infections are an independent risk factor for diabetes development. *In vitro* Atazanavir (ATV) has shown minimal inhibitory effect on the insulin-regulated glucose transporter GLUT4. Studies in healthy HIV-negative demonstrated a favorable metabolic profile. Clinical studies in HIV and HCV/HIV coinfected pretreated patients with underlying real conditions are needed.
Methods: 51 pretreated HIV-1 infected patients who started ATV/r were included prospectively. Insulin resistance was assessed by HOMA (Homeostasis Model Assesment). Hepatotoxicity was defined according to AIDS Clinical Trials Group criteria to ALT values. Clinical data and laboratory parameters were analyzed at baseline and every 12 weeks up to 48. It includes body mass index (BMI), fasting glucose, insulin, triglycerides (TG), total cholesterol (TC), low-density lipoprotein-cholesterol (LDL-c), CD4+ cell count and HIV-1 RNA. HIV monoinfected and HIV/HCV co-infected (pos HCV-RNA) patients were compared.
Results: 23 HIV-mono and 28 HIV-HCV coinfected patients were analyzed. Mean age was 40 ± 8 years, male 74%; median CD4 count (IQR) 288 (224-548) cells/μL. 53% plasma HIV-RNA<50 copies/mL. Only 1 case of severe hepatotoxicity (Grade 4) was seen (coinfected patient). Metabolic profile is shown in Table 1. ATV was discontinuated in 4 cases (2 poor adherence and 2 gastrointestinal intolerance). Non virological or immunological failures were documented.
Discussion: ATV/r can be safely used in patients with chronic viral hepatitis. ATV/r regimens do not induce insulin resistence and has a good lipid profile in coinfected as in monoinfected patients. These results would be better with unboosted ATV regimens.

P64

Hypertension and microalbuminuria in HIV infected patients: beneficial effects of the treatment with telmisartan

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Background: In HIV infected patients there is increasing evidence of hypertension and microalbuminuria, two important risk factors for renal and cardiovascular disease (CVD). Anti-hypertensive drugs inhibiting the renin-angiotensin system exert an antiproteinuric effect. Telmisartan, an angiotensin II receptor blocker partial agonist of the PPAR-γ approved for the treatment of hypertension, seems to exert a nephro-protective effect independent of blood pressure reduction in the general population. Aim of the study was to evaluate kidney-protective effects of telmisartan in hypertensive HIV+patients with microalbuminuria.
Methods: 8 Caucasian male HIV+ patients cART treated without therapeutic changes for over 12 months and recent diagnosis of mild hypertension, were treated with telmisartan 80 mg daily. They had suppressed viremia and CD4 cell count > 300 cell/ml up on 6 month, and microalbuminuria >5 mg/l. Systolic (SBP) and diastolic (DBP) blood pressure, triglycerides (TGs), total cholesterol (TCh), HDL (HDL-C) and LDL (LDL-C) cholesterol, CRP, ESR, microalbuminuria, MDRD-GFR, cystatin-C, IL-18, VEGF and endothelin-1 were measured at baseline (T0), one (T1), three (T3) and six months (T6). All the statistical analysis was performed with the SPSS Advanced Statistical 7.5 Software.
Results: Treatment with telmisartan improved SBP and DBP values at T1 yet ($p = 0{,}001$). Microalbuminuria were statistically decreased at T1 ($p = 0{,}006$) and further on T6 ($p = 0{,}0001$), whereas MDRD-GFR was statistically augmented ($p = 0{,}03$). Cystatin-C, endothelin-1 and VEGF were statistically reduced at T3 ($p = 0{,}0001$; $p = 0{,}01$ and $p = 0{,}0045$ respectively) and at T6. TG, TCh, LDL-C levels decreased with statistical significance at T6 ($p = 0{,}003$; $p = 0{,}03$ and $p = 0{,}02$ respectively), while HDL-C increased at T6 ($p = 0{,}04$). ESR, PCR and IL-18 decreased at T6 ($p = 0{,}04$; $p = 0{,}006$ and $p = 0{,}02$ respectively).
Discussion: Telmisartan was well tolerated and effective to improve hypertension and lipid metabolism. Decreased microalbuminuria and cystatin-C with increased MDRD-GFR are indicative of nephro-protective effects of telmisartan. Mechanisms causing microalbuminuria in HIV+ patients could be related to infection, chronic inflammation and endothelial dysfunction. Decreased endothelin-1 and VEGF in this study may be related to an endothelial protective effect of telmisartan. These data confirm renal and endothelial protective effects of telmisartan also in HIV+ patients.

Table 1 (abstract P63)

	HCV	BASAL	48-WEEK	Δ_{w48-w0}	$P(\Delta_{w48-w0})$
BMI (Kg/m^2)	-	24.5 (22-27)	26 (23-29)	0.34 (-1.0-+0.9)	>.05
	+	22 (20-26)	20 (18-23)	-0.35 (-0.8-+1.2)	>.05
Fasting glucose (mg/dl)	-	87 (83-97)	88 (85-95)	-0.5 (-5.7-+8)	>.05
	+	94 (83-103)	90 (80-105)	+2 (-24-+12)	>.05
HOMA	-	1 (0.8-3.6)	1.4 (0.9-3.2)	-0.06 (-1.5-+0.8)	>.05
	+	3.5 (1.8-6.3)	4.5 (3-7.5)	+1.8 (-4.6-+3.1)	>.05
TC (mg/dl)	-	197 (165-215)	179 (161-200)	-18 (-40- -3)	.02
	+	157 (132-174)	144 (127-165)	-5 (-32-+15)	>.05
TG (mg/dl)	-	149 (122-241)	126 (101-208)	-10 (-92-+17)	>.05
	+	139 (99-186)	114 (89-197)	0 (-19-+59)	>.05

As Median (Interquartile range). In all comparisons between HCV- vs HCV+ p > .05.

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NRTIs (ZDV and d4T) side effects in PLHAs attending the antiretroviral treatment centre of B. J. Medical College Aad Civil Hospital At Ahmedabad, Gujarat, India

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Retrovirology 2010, **7(Suppl 1):**P65

Background: This study reviews common side effects experienced by PLHAs who were initiated with Zidovudine or Stavudine based FDC ART regimen as per Indian National ART Guidelines attending ART Centre of B. J. Medical College and Civil Hospital, Ahmedabad, Gujarat, India.
Methods: All the ICTC confirmed HIV Positive Patients were registered and initiated with FDC ART at ART Center of the Institute as per Indian National ART Guidelines; recommends the use of 2 NRTIs (ZDV/d4T + 3TC) + 1 NNRTI (EFV/NVP). Baseline parameters like CD4 count, necessary laboratory investigations, TB Workup and adherence counseling were carried out and on follow up. Depending on the level of hemoglobin NRTIs were chosen.
Results: Total 4379 patients were studied during the period of May 2005 to November 2008 initiated with either ZDV or d4T based ART regimen. Out of them total 3355 (76.62%) were on either ZDV or d4T based ART regimen and regular visitor of the ART Centre are the study target, while the remaining 1024 (23.38%) were either lost to follow up (LFU), expired and transferred out were excluded, Table 1.
Few patients 15 (0.83%) had multiple side effects, while in remaining 71 (3.91%) the exact reason for regimen change was not available.
Discussion: The most observed side effects after initiating ART; Anemia (ZDV based) and Peripheral Neuropathy (d4T based) requires regular monitoring for early diagnosis. Substitution within same group must be done. While ART is becoming increasingly effective; side effects are low compared to studies in India and Cambodia by *Sharma et. al., 2008* (Anemia 20.0%, Peripheral Neuropathy 22.2%) and *Isaakidis et. al., 2008* (Anemia 21.92%). To optimize adherence and hence efficacy, clinicians must focus on preventing side effects whenever possible to yield the overall successfulness to the National Program.

Table 1 (abstract P65)

ART Regimen	Side Effect	No. of Patients	%
ZDV n = 1538 45.85%	Anemia	212	13.78%
d4T n = 1817 54.16%	Peripheral Neuropathy	220	12.11%
	Dyslipidaemias	143	7.87%
	Lipoatrophy	108	5.94%
	Lactic acidosis	6	0.33%
	Pancreatitis	5	0.27%

P66

Depression and self-esteem of patients positive for HIV/AIDS in an inland city of Brazil

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Background: Psychiatric disorders are common in the course of HIV infection, and depression is one of the most prevalent. Another consequence is the decrease in self-esteem of the individual, marked by the accumulation and fat loss in areas of the body, one of the side effects of drugs. This study aims to characterize aspects demographics, economics, clinical and epidemiological and identify symptoms of depression and assess self-esteem in individuals with HIV/AIDS at different stages of infection.
Methods: Descriptive and quantitative study, conducted in an inland city of Sao Paulo (Brazil). Data were collected through interviews with individuals with HIV/AIDS, using as instruments Beck Depression Inventory, Scale of self-esteem by Rosenberg and a questionnaire with data economic, demographic, clinical and epidemiological data.
Results: We interviewed 75 individuals, and 50.7% were male. The predominant age group was between 29 and 39 years (42.7%), 69.3% reported having been infected through sex; sexual orientation, 92% is held as heterosexual. Identified that 22 (29.4%) had depressive symptoms and as such, 4 (18.1%) with symptoms of mild to moderate, 6 (27.2%) with moderate to severe and 12 (54.5%) with severe symptoms. The self-esteem was identified scores of 14 to 23 of which 34.7% had a score of 16 points, which indicates low self-esteem.
Discussion: The analysis of data showed that the individuals interviewed are young, most were infected through sexual intercourse and almost 30% had depressive symptoms and low self-esteem. The depression associated with anxiety reflects a detrimental effect on quality of life of the individual with HIV/AIDS, with it's extremely important to invest in media and treatments to relieve pain and suffering of the individual. Regarding self-esteem, their confidence levels influence the personal care and take the individual to not seeking treatment. The correct diagnosis is essential for the proper treatment of these patients to increase adherence and improve the quality of life.

P67

A young patient with perinatal HIV infection treated for 17 consecutive years with antiretroviral therapy: extremely severe lipo-accumulation picture

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Background: Like adults, also pediatric HIV-infected patients (p) are prone to suffer from a lipodystrophy syndrome, usually accompanied by dysmetabolic disorders. The psychological consequences of the morphologic changes are expected to be particularly devastating in development ages.
Methods: Like adults, also pediatric HIV-infected patients (p) are prone to suffer from a lipodystrophy syndrome, usually accompanied by dysmetabolic disorders. The psychological consequences of the morphologic changes are expected to be particularly devastating in development ages.
Results: Notwithstanding a cumbersome anti-HIV management, the immune-virological situation remained under control (the last laboratory assays showed a viremia of 1,300 HIV-RNA copies/mL, and a CD4+ count of 612 cells/μL), and our p never suffered from AIDS-related complications. Unfortunately, starting 7 years ago, a bilateral symmetric lipomastia appeared, associated in the past two years with a bilateral, dorsal lipid accumulation (both confirmed by ultrasonography), which were responsible for severe psychological disturbances. Hormonal-metabolic workup failed in disclosing relevant abnormalities, as to serum FSH, LH, testosteron, estradiol, estrone, progesteron, dehydroepiandrosterone, and prolactin levels, thyroideal profile (TSH, fT3, fT4, and anti-thyroid antibodies), as well as metabolic pattern (fasting glucose, total cholesterolemia, HDL- and LDL- fractions, apolipoproteins, C-peptide, insulin, fruttosamin, glycosilated hemoglobin, lactate, and bicarbonate), when excluding a moderate hypertriglyceridemia.
Discussion: While in adult p the prolonged course of HIV infection and combination antiretroviral therapy (cART) usually leads to an associated dysmetabolic sindrome and lipodystrophy, characterized by co-existing peripheral lipoatrophy and visceral adiposity, usually accompanied by a mixed dyslipidemia and insulin resistance, our young p surprisingly developed an isolated mammary and dorsal lipid hyperaccumulation syndrome with lipomastia prevailing over gynecomastia. Also the recent advice to further modify cART (by including NNRTIs), is not expected to act significantly in short-mid term on the particular, focal lipoaccumulation features of our young p. A surgical option (liposuction), is the most reliable clinical option.

P68

Emerging HIV-associated concerns: osteopenia and osteoporosis. Easy prevention and management guidelines

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Background: Osteopenia and osteoporosis (O) are emerging complications of HIV infection, especially when treated with combined antiretroviral therapy (cART). The pathogenesis is multifactorial, potentially involving all classes of anti-HIV drugs, although protease inhibitor (PI) use, overall HIV and cART duration, and the male sex, seem related to a significantly greater risk.
Methods: In a preliminary DEXA screening assessing lumbar spine and femoral head of around 100 out of around 1,000 single-centre patients (p), the frequency of osteopenia-osteoporosis (based on lumbar T-score) was assessed around 48%. An increased risk was found in p treated with protease inhibitors *versus* p receiving non-nucleoside reverse transcriptase inhibitor, or triple nucleos(t)ide reverse transcriptase inhibitors.
Results: Prospective studies of extensive p samples are needed, to elucidate the epidemiology, pathogenesis, clinical issues, and evolution of HIV-associated bone metabolism abnormalities. When planning strategies for their early diagnosis, prevention, and management, also cost-effectiveness issues should be taken into consideration, since no pharmacoeconomic data still exist in this setting. Although severe consequences (pathological fractures, prosthetic implants), are expected to be proportionally infrequent events, their consequences in terms of length-intensity of hospitalization, related costs, and especially consequences on the patient's quality of life, are expected to play a remarkable role. Anyway, the most reliable diagnostic procedure of O (i.e. DEXA scan), has affordable costs (around Eur 43.40 for a scan which also offers a body composition assessment), as well as the first-line drugs for osteopenia, e.g. supplementation with calcium (Eur 6/month), and vitamin D (Eur 7/month). These costs cannot be compared with the standards costs of an asymptomatic cART-treated p (Eur 471 to 874/month), and the immunologic, virologic, laboratory, and clinical controls made at least quarterly in the same p.
Discussion: Like post-menopausal O, also HIV disease should be investigated from multiple cost-effectiveness points of view, to establish which p are the early candidates for a DEXA screening, when this examination is more useful during HIV disease course-therapy, when the DEXA scan should be repeated, and when-how to intervene pharmacologically, in order to prevent serious and potentially invalidating O-related complications.

P69

Interactive theatre for HIV/AIDS side effects on youth sexuality reproductive health and rights in pakistan to learn and practice

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Background: Pakistan has the largest demographic dividend of youth, i.e. 63% population below the age of 29 (Out of 170 million), but unfortunately majority of the youth are unaware about sexual and Reproductive health and rights and lacks in access to sexuality education and services due to Socio-cultural norms which leads to harmful sex practices, increase rate of STI/RTI including HIV/AIDS and creating myths and misconceptions in their minds.
Methods: The above stated situation urged Chanan Development Association (CDA); a youth lead NGO Working to empower young people in Pakistan, to launch an innovative program on self-help basis with the title "Interactive Theatre for HIV/AIDS Side effects on Youth Sexuality Reproductive Health and Rights in Pakistan" to Learn and Practice. The main objective of the project was Equipping young activist with the skill of Interactive Theatre (Theatre of the Oppressed) so that they can utilize this skill effectively to educate and sensitize their communities in general and young people in particular regarding sexuality and reproductive health issues of youth people and can reduce the stigma related to sexuality of youth" As part of project strategy, 22 Youth Groups were formulated all over Pakistan comprising of 10-12 members with gender, religious and ethnic equality. Then CDA Conducted 10 days training workshop on Interactive Theatre for HIV/AIDS Side effects on Youth Sexuality Reproductive Health and Rights in Pakistan for Learning and Practice for each group followed by 10 theatre performances by each group in their respective districts/communities on the said theme to raise awareness about SRHR, reduce stigma about youth sexuality and to promote informed choices among marginalized segment of the society, i.e. youth, women, Madrisah students (Religious Seminaries) and the MSM.
Results: As a result of the pilot project 1 - 250 young activists involved in Sexuality and Health Education from 18 districts of Pakistan 2 - 200 theatre performances staged 3 - 40,000 people from marginalized segment of the society reached and sensitized on Sexuality, health particularly SRH issues and informed choices for better sexual and reproductive health.
Discussion: Young people posses a great ability to challenge societal norms and bringing change in their own and other peers attitude, knowledge and practices. Through building capacity on innovative and interactive skills, youth activists can play an active role in combating the stigma related to sexuality and can promote health and safer sex practices in marginalized communities. Arts based interventions can be usefully implemented to engage marginalized communities and generate

dialogue even on every sensitive issue like sexuality and reproductive health including SRH.
Involving young activists and employing arts based approaches should have to be ensured in every awareness raising program for reaching marginalized groups and initiating dialogues in very rigid and backwards communities.

P70

Interplay between HIV and microRNAs in AIDS-related lymphomas

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Background: Human immunodeficiency virus (HIV)-induced immune activation of B cells is thought to be a contributing factor to the increased frequency of B-cell malignancies observed in HIV-infected individuals. In some cases, as in Burkitt lymphoma, tumors arise before profound immunosuppression occurs, when the $CD4^+$ cell count is still high. Therefore, immunodeficiency *per se* may not be necessary for lymphomagenesis in these patients, and that HIV itself may have an oncogenic potential.
There are no clear answers to explain how HIV leads to malignant transformation, even though several events have been proposed as co-factors in HIV-related tumorigenesis. In particular, the HIV-encoded Tat protein is thought to participate in B-cell abnormalities observed *in vivo,* as it can be released from the HIV-infected cells and then lead to differential modulation of naïve, memory and germinal center B-cells.
Recent findings indicate a complex interplay between viral proteins and host transcription regulatory machineries, including histone deacetylases (HDACs), histone acetyltransferases (HATs), histone metyltransferases (HMTs) and DNA metyltransferases (DNMTs). Tat can bind to histone acetyltransferases (HATs) p300/CBP, p300/CBP-associated factor, and hGN5. Chromatin remodelling may therefore represent a mechanism of control of gene expression, whose deregulation may eventually lead to malignant transformation.
Methods: GEP and microRNA profiles were obtained in HIV+ primary tumors.
Results: We investigated the expression of several chromatin remodelers, both in human cell lines, either transfected or not with Tat, and HIV-positive vs. HIV-negative primary tumors. Gene expression analysis revealed a differential expression of genes involved in chromatin remodelling between HIV-positive and HIV-negative patients. In particular, we observed a modulation of specific acetyltransferases and DNA metyltransferases, which seem to be regulated at the post-transcriptional level.
We then analyzed the expression of microRNAs (miRNAs) predicted to have specific chromatin remodelers as targets, and we observed that these miRNAs resulted significantly down-regulated in HIV-positive subjects.
Discussion: These findings lead to the hypothesis that HIV may interfere with physiological regulation of cell functions maintained by miRNAs, may be through viral-encoded miRNAs. We are currently analyzing which genetic/epigenetic mechanism underlies HIV-mediated miRNA silencing in host cells.
Collectively, our results support the possible oncogenic role of HIV and open new scenarios in the identification of novel therapeutic targets in HIV-related malignancies.

P71

Non-AIDS related malignancies 13 years after the availability of combined antiretroviral therapy

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Background: The introduction of combined antiretroviral therapy (cART) since the year 1996, contributed to a rapid, significant drop of frequency of all AIDS-defining opportunistic infections and some selected AIDS-related tumors (like Kaposi'sarcoma), with a consequent, remarkable reduction of both morbidity and mortality rates associated with these disease complications.
Methods: Our cohort of over 1,700 HIV-infected patients followed in two connected outpatient centres by the same physician staff were prospectively followed since the year 2000 (9 years), with special interest focused on the diagnosis, treatment and outcome of non-AIDS related malignancies.
Results: Among hematological malignancies other than non-Hodgkin's lymphoma and primary central nervous system lymphoma, we observed three cases of acute myelogenic leukaemia and 4 episodes of Hodgkin's lymphoma. A greater number of solid tumors involved different organs and sites: laryngeal cancer (8 cases, with 6 episodes of papillomatous laryngeal cancer), rhinopharyngeal squamous carcinoma (4 cases), adenocarcinoma of the lung (6 cases), gastric adenocarcinoma (3 episodes), esophageal carcinoma (2 patients), prostate cancer (4 cases), bladder adnocarcinoma (3 episodes) pancreatic adenocarcinoma (2 cases), and squamos anal carcinoma (2 episodes). Some of these malignancies have been reported with extremely rare frequency until now (usually as single-case anecdotal reports), in particular before the cART era. The patient's age ranged from 34 to 67 years, the mortality rate of these episodes was very elevated (over 80%), and occurred 3-41 months after diagnosis, despite appropriate surgical and/or cytotoxic chemotherapy and/or radiotherapy.
Discussion: The significantly increased life expectancy of HIV infected patients in the cART era was characterized by a proportionally increase of non-AIDS-defining tumors, which may depend on the advanced mean patients' age, their prolonged exposure to risk factor, the persisting functional immune system imbalance, and probably some direct oncogenic property of HIV itself, even when a "quantitative" recovery of CD4+ lymphocyte count has been achieved thanks to cART. The differential diagnosis of non-AIDS-associated tumors may be delayed by the low clinical suspicion, and their frequency to mimick and/or overlap infectious complications. Further epidemiological and clinical investigation is strongly warranted, to increase the awareness of this emerging phenomenon.

P72

Prolonged elevation of viral loads in HIV-1-infected children in a region of intense malaria transmission in Northern Uganda

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Background: Introduction: Malaria and HIV-1 infection cause significant morbidity and mortality in children in sub-Saharan Africa. Recurrent malaria infection increases HIV-1 viral load in adults and increases the rate of progression of HIV-1 infection to AIDS. The effect of malaria on viral loads in Children living with AIDS (CLWA) is not clearly known.
Objective: To assess the effect of malaria on HIV-1 viral loads in CLWA.
Methods: One hundred thirty five afebrile HIV-1 positive children having negative blood slides for malaria were recruited at Apac Hospital and followed up for one year. They were monitored for development of P. falciparum malaria, which was treated with CQ+SP and the children followed up for 28 days. HIV-1 viral loads were measured over three time-points: at enrolment (no malaria), during an episode of malaria, and at a visit about 8 weeks (range 6-19 weeks) after the malaria visit when the child had neither parasites nor any intervening malaria episodes (post-malaria). Primary analyses were restricted to children who on follow up had HIV-1 viral loads measured at the three relevant time-points.
Results: Baseline characteristics, Table 1.
Malaria increased HIV-1 viral load significantly in CLWA. Low parasitaemia <= 5/HPF transiently increased viral load by 0.42 log (95% CI 0.29-0.78, $p = 0.0002$), higher than that reported in adults. These patients' viral loads returned to levels similar to those at baseline after treatment. In 13 patients with high parasitaemia (10-20/HPF), the mean increase in viral load was 0.53 log (0.14 to 0.51), $p < 0.0001$, remaining significantly higher

Table 1 (abstract P72)

SEX	MALE/FEMALE	41.1%/58.9%
AGE (yrs)	(1.5-5)/(6-12)	44.7%/55.3%
WHO CLASSIFICATION 1, 2, 3, 4	n = 30, 55, 48, 02	percentage = 22.2%, 40.7%, 35.6%, 1.48%
HAEMOGLOBIN median, IQR	10.5	9.8 - 13.8

than at baseline after treatment i.e. mean difference (signed-rank test) in viral load "before" and "after" malaria was significant.
Discussion: *P. falciparum* malaria increased HIV-1 viral loads in children, with some viral loads remaining elevated several weeks after antimalarial treatment. Prolonged post-treatment elevation has important implications for the clinical course and the potential for transmission in sexually active adults.

P73

Women HIV education, capacity building and economic empowerment initiative

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Background: The national sentinel survey, 2005 HIV/AIDS prevalence rate in Nigeria is estimated at 4.4%. This shows a reduction from the 2004 prevalence rate of 5.0%. However, the reduction may not be significant in terms of the population of people living with HIV/AIDS.
Women have been reported to be the most affected. Women vulnerability to HIV can be attributed to social, economic and cultural factors. HIV prevention initiatives aimed at halting the spread of HIV among women population must integrate an economic empowerment programme if meaningful impact is to be made. It is in this view that Health Matters Incorporated (HMI) implemented a project on Women HIV education, capacity building and economic empowerment initiative in Makoko community in Mainland lga of Lagos state.
The goal is to halt the spread of HIV/AIDS through an integrated HIV education, capacity building and economic empowerment programme.
Methods: • Women HIV education and economic empowerment initiative project is an integrated programme aimed at building the capacity of women fish smokers and small business owners including HIV/AIDS and building their economic network for self reliance.
• HMI provided capacity building training in fish smoking, hairdressing and beads stringing for women in mainland lga of Lagos state with support from Leaders Quest and Micro Small and Medium Enterprise Development (MSME) Nigeria.
• HIV/AIDS and life skills education through training and distributions of Information, Communication and Education materials.
• Monthly women health forum to discuss challenging issues around women and HIV/AIDS and foster solutions as a group.
Results: • 200 women had their capacity build in hairdressing, beads stringing and fish smoking and this provided more economic power for women.
• 50 women were provided with soft loan which lead to increased turnover
• 25 women were linked to micro finance bank for further assistance
• Women who participated in the programme were able to negotiate for safer sex practices with their partners.
Discussion: Poverty and economic dependency on men is a driving factor for women inability to negotiate for safer sex practices with their partners.
Programme designed to address the economic, capacity and reproductive health need of women in integrated approach help to increase women participation.

P74

Tuberculosis with pulmonary involvement in HIV patients

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Background: Tuberculosis (TB) is a leading cause of death worldwide and is closely associated with HIV-infection. The higher probability of spreading TB in patients with pulmonary involvement is always a great concern and demands immediate action.
The aim of this study is to analyze, retrospectively, cases of TB with confirmed pulmonary involvement in the HIV infected population observed in an Infectious Diseases Unit and diagnosed from January 2000 to December 2008.
Results: The study included 87 patients: 96.5% HIV-1; 92.0% Caucasian; 79.3% male; mean age 41 years [25-76]; 9.2% were foreigners (5,7% from Sub-Saharian Africa); HCV co-infection in 49.4% and 51.7% with history of drug addiction.
TB was the initial manifestation of HIV infection in 52.9% and AIDS-defining in 71.3%. TB predominately occurred in late stages of HIV infection (74.7% had CD4 + T cells ≤ 200/mm^3). The average time between diagnosis of HIV infection and TB was 3.9 years. Clinical features were mainly respiratory (71.8%) and fever (66.3%). Chest – ray most frequently showed multifocal involvement (50.6%). Tuberculin test was reactive in 14 cases (n = 27). Respiratory secretion culture was positive in 83.9% - of these, 36.8% had positive Ziehl-Neelson (ZN) staining. Resistance to 1 or more anti-TB drugs was found in 12.6% (n = 11). There were 2 cases of multirresistance and 1 extensively resistance (XDR). Other pulmonary diseases were associated in 13.8%. HRZE was the most frequent treatment regimen and 14.9% did not complete it due to loss in follow-up. The average time of treatment was 7 months. Mortality rate was 18.4%.
Discussion: Most TB cases occurred with severe immunossupression and were the first manifestation of HIV infection and AIDS defining in most of the patients. ZN stain of respiratory secretions was negative in a considerable number of cases (63.2%) whose culture was positive. Mortality and loss to follow-up rates were relevant.

P75

Service outcome of antiretroviral Post-Exposure Prophylaxis (PEP) for occupational HIV exposure among health care personnel

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Background: This study aims to assessment of the outcome of Post Exposure Prophylaxis (PEP) Service at Centre of Excellence (CoE), ART Centre, B. J. Medical College, Civil Hospital, Ahmedabad, Gujarat, India among Health Care Personnel (HCP).
Methods: Potentially exposed to infectious materials HCPs of the institute were studied. Exposed HCP were enrolled and followed up as per Management of Occupational Exposure including PEP for HIV by National AIDS Control Organization (NACO) India guidelines.
Results: Total of 43 exposed HCP (18 male and 25 female with male to female ratio 0.72: 1) during October 2008 to October 2009 were studied, Table 1.
Discussion: Those tested for HIV were all Non – Reactive; shows the effectiveness of PEP services. We are finding obstacles on follow up tracking like "I am much aware about the risk, stigma of HIV Positivity, negligence for follow up and confidentiality issues at the workplace etc." that prevent the HCP for PEP and increase the risk of transmission. Practice and regular sensitization with UPW has increased the awareness among HCP for reducing HIV infection risk and transmission and prevention through PEP.

Table 1 (abstract P75)

Parameter		No. Of Cases (n = 43)
Universal Precaution Workshop (UPW) for HCP		
January 2008 – December 2008		2
January 2009 – October 2009		9
Enrolment of HCP for PEP at CoE		
October 2008 – May 2009		19(44.18%)
June 2009 – October 2009		24(55.81%)
HCP Category Enrolled for PEP at CoE		
Medics		19(44.18%)
Paramedics		21(48.8%)
Servants		3(7%)
HIV Status of the HCP for PEP enrolment		
October 2008–May 2009 (n = 19)	Reactive	0
	Non – Reactive	7(36.8%)
	Unknown	12(63.15%)
June 2009–October 2009 (n = 24)	Reactive	0
	Non – Reactive	16(66.66%)
	Unknown	3(12.5%)
Type of Exposure		
Mild		21(48.8%)
Moderate		18(41.9%)
Severe		4(9.3%)
Reporting Timeline of exposed HCP to the CoE		
<2 hours		27(62.8%)
2 – 24 hours		9(20.9%)
24 – 72 hours		5(11.6%)
>72 hours		2(4.7%)
HIV Status of the source		
Reactive		22(51.2%)
Unknown		21(48.8%)
PEP Regimen Prescribed		
Basic (Zidovudine+Lamivudine)		30(69.76%)
Expanded (Zidovudine+Lamivudine+Lopinavir/Ritonavir)		12(27.90%)
Expanded (Zidovudine+Lamivudine+Indinavir)		1(2.32%)
HIV Status of exposed HCP post PEP at 6 months (n = 18)		
Reactive		0
Non – Reactive		3(16.66%)
Unknown		15(83.33%)
HCP Not Completed 6 Months (n = 43)		25(58.13%)

P76

Glomerular filtration (GF) determined by creatinine clearance (CCR) in 24 hours urine and cockcroft & gault (cg) and modification of diet in renal disease (MDRD) equations in a large cohort of HIV+ patients

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Background: The recognition of kidney tubular dysfunction (KTD) in subjects treated with tenofovir (TDF) has prompted to include tubular and glomerular function as part of current HIV monitoring in most patients. Although CCR in 24 h urine is the most reliable method to determine GF, practical issues make CG or MDRD estimates more adequate in a daily basis. The correlation between all these methods used to assess GF, and the possible interference of KTD, has not been examined in the HIV population.
Methods: All consecutive HIV+ patients attending a reference HIV outclinic were assessed for CCR (mL/min) in 24 h urine. Subjects collecting an urine volume <450 mL were excluded. Estimates of CCR by CG and MDRD were calculated for each patient. Intraclass correlation (IC) and greatest survival-agreement plots for 75% of the population (SAP-75) were applied to test CG vs MDRD differences with respect to CCR. Correlation analyses were also performed according to the presence of glomerular impairment (CCR <60 mL/min) or KTD (at least two of the following: glucosuria, hyperaminoaciduria, hyperphosphaturia, hyperuricosuria or beta2-microglobulinuria, being present at least one of the first three).
Results: A total of 417 patients were examined (22% ARV-naïve, mean age 39 years-old, 87% males, mean CD4 count 445 cells/μL), Table 1.
Discussion: In HIV+ patients, indirect methods to assess GF show high correlation with calculated CCR. In patients with normal CCR, CG may be more accurate than MDRD to estimate GF. Glomerular dysfunction or KTD do not seem to alter the correlation between CG or MDRD and CCR. Significant reductions in GF, as assessed by CCR, CG or MDRD, are more frequent in patients with than without KTD.

P77

Factors associated with development of opportunistic infections among patients on ART at a Ugandan Program-MJAP

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Background: To establish the factors associated with development of opportunistic infections among patients on ART at an HIV/AIDS Program in Uganda.
Methods: Retrospective cohort analysis on adult patients initiated on ART between November 2005 and November 2007. The socio demographics, clinical and laboratory characteristics of patients that developed OIs were studied.
Results: Data of 4,878 patients on ART was analyzed. 3,247 (67.2%) were females. Median age 33 years (SD 8.5) and Median weight 53 kg (SD 10.0). Incidence of OIs 329 (6.7%); Oral candidiasis 106 (32%), Vaginal candidiasis 80 (24%), Kaposi's sarcoma 40 (12%), Herpes simplex 28 (9%), Oesophageal candidiasis 25 (8%), Tuberculosis 15 (5%), others 38 (10%). WHO stage 3&4 (OR 2.64, CI 2.04-3.43, P < 0.001), CD4 count <100 cells/mm^3 (OR 1.92, CI 1.52-2.42, P < 0.001), not being married (OR 1.69, CI 1.33-2.14, P < 0.001) and having no formal education (OR 1.61 CI 1.09-2.36, P = .0015) were significantly associated with development of OIs among patients on ART.
Sex, age, body weight, ART regimen and Karnofsky score were not associated with development of OIs.
Discussion: The risk of OIs is higher among patients with a low CD4 count, high WHO clinical stage, no formal education and in those that are not married.
Early identification of HIV-infected individuals and linkage to HIV care and treatment is likely to minimize the incidence of Opportunistic Infections.

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Evaluation of the therapeutic educational consultations of patients infected by the Human Immunodeficiency Virus (HIV)

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Background: The objective of this survey is to evaluate the satisfaction and feeling of HIV infected patients following weekly therapeutic

Table 1 (abstract P76)

	All	CCR >60 mL/min	CCR <60 mL/min	No KTD[a]	KTD[b]	a vs b [p]
No. of patients (%)	417	393 (94)	24 (6)	337 (86)	54 (14)	
Mean CCR (mL/min)	113	117	47	116	103	0.01
Mean CG (mL/min)	100	101	89	103	90	0.003
Mean MDRD (mL/min)	91	91	76	92	81	<0.001
IC [p]	0.64 [<0.001]	0.66 [<0.001]	0.63 [0.02]	0.59 [<0.001]	0.77 [<0.001]	
SAP-75* [CG minus CCR] (mL/min)	20.3	19.9	24.6	21.1	18.8	
SAP-75** [MDRD minus CCR] (mL/min)	21.3	21.3	20.8	24.1	14.8	
Log-Rank [SAP-75* vs SAP-75**]	0.05	0.04	0.3	0.04	0.38	

educational consultations in the hematology department (Argenteuil hospital).
Methods: Consultations are managed by a pharmacist who receives patients individually (initiation of treatment, side effects, non-compliance). A survey was carried out by a junior pharmacist. It includes 15 items on which the patient gives his opinion. The questionnaire was validated and presented to the patients. All patients had at least 2 therapeutic educational consultations implemented the survey.
Results: 24 patients filled in the questionnaire over a 3 month period. 50% of them come from sub Saharan Africa.
Factors of improvement:
§ concerning the disease: Non-acceptance of the disease: 16,6%; non-understanding of the disease: 12,5%; non-understanding of the explanations on the viral reproduction: 20,9%; information does not fit to their expectations: 8,3%.
§ concerning the treatment (explanations, information, observance, motivation), evaluation and listening: no discord.
§ concerning the progress of consultations (rythm, the duration of waiting...), more than 90% of patients are satisfied.
Discussion: An implementation of corrective measures is necessary:
§ to deal with the disease: to define clear objectives with the patients. A consultation with the psychologist must be proposed to all patients (multidisciplinary approach); the difficulties to understand the French language shows how our tools are not good enough or our vocabulary not adapted. It is necessary to develop an interactive process: creation of symptoms cards/decisions with pictures.
This survey shows that patients are satisfied with the consultations and underlines the importance to evaluate the need for modifying our professional practices.

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Abstract withdrawn

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Immune response characterization in HIV/HCV co-infected patients of medicine tropical foundation

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Background: The epidemiology of co-infection of human immunodeficiency virus and hepatitis C virus (HIV/HCV) is around 30 to 60%. Approximately one third of HIV infected shows C hepatitis, with a high rate in hemophiliacs and drug users. Recent publications demonstrated that HIV positive patients co-infection with HCV have a co-factor to develop AIDS. The purpose of this study was evaluate the cellular and humoral immune response and cytokines in HIV/HCV co-infected patients in Foundation of Tropical Medicine of Amazonas.
Methods: After consent term assignature, the population of T lymphocytes $CD4^+$ and $CD8^+$ was analyzed in the whole blood by flow cytometry and a blood sample was take to measure the serum concentration of inflammatory cytokines (interleucine – IL - 6, 8 and tumoral necrosis factor alpha-TNF-α), cytokines of T_H1 (IL-12, Interferon gamma-IFN-γ) cytokines T_H2 (IL-4) and suppression cytokine (IL-10) using ELISA BD OptEIA® *kit*.
Results: As for $CD4^+$T cells 72.2% had < 500 cls/mm^3 with a median of 271 cls/mm^3, on the T $CD8^+$ 88.9% had ≥215 cls/mm^3 with a median of 794.5 cls/mm^3. The ratio $CD4^+/CD8^+$ was 0.32 cls/mm^3. When the dose cytokines IL-4, IL-6, IL-8, IL-10, IL-12 and IFN-γ in the patients found that only the IL-6 ($p =< 0.001$) showed statistical significance especially when correlated to the logarithm of the HCV viral load (0.031).
Discussion: The results found in this study, despite the low prevalence, have annual growth of co-infection due to improvement in the research of hepatitis C in patients with HIV and the IL-6 cytokine was important marked of inflammation in this studied population.
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Advocacy and piloting the first needle and syringe exchange program in Iranian prisons

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Background: 1-Pilot the PNEP in the Iranian prisons after having received the acceptance of the authorities in the Iranian Judiciary System. 2-Schedule the piloting of the PNEP in the Iranian prisons. 3-Analyse the opinions given by the policy-makers in prisons and the staff to have a better implementation of the programme. (during pre-, while and post-stages) 4-Collect and evaluate demographic crime and drug data of the IDU's who are easily reached since in this program the IDU's can quite easily mention their difficulties with drugs. 5-Distribute the sterile syringes among the IDU's without any limitations and evaluate the data based on the number of sterile received and used returned syringes respectively.
Methods: After recieving acceptance of policy-makers, program performed in 3 prisons and all the volunteers were IV drug abusers. The program had been performed in three major prison centers in Iran, including Tehran, Isfahan and Hamadan. The prisoners were given sterile needles and syringes weekly, and the used ones were also collected regularly. Data including number of syringes and shared ones used by

each person was collected at the beginning of program; and this data was also documented on a weekly basis as the program proceeded. Information regarding blood-borne diseases was also given to the enrolled prisoners continuously.
Results: Among 341 volunteer prisoners enrolled in this program, an average of 17 syringes was reported to be used weekly for IV drug injection before starting the program. Moreover, the volunteers reported to use an average of 3.7 shared syringes during a week. Documented infection with blood-borne diseases (including Human Immunodefficiency Virus, Hepatitis B or C) was also found in 44 prisoners. At the end of the program, prevalence of using shared syringes among volunteers was decline to zero.
Discussion: 1-Iran Prisons Organisations will be going to extend the programme to the rest of the Iranian prisons in case the results are satisfactory and there are minor complications.2-Voluntary entrance of the prisoners into the PNEP shows that despite the access to the other harm reduction programmes, the need and desire to inject drugs will not disappear and if the prisoners are not given sterile syringes and needles, they will share them. 3-Programming, process, problems and their solutions during the programme can be of great help to the rest of the Iranian prisons and other prisons worldwide. 4-The average number of prisoners who entered the programme in the three target prisons has been between 25 and 35 per month and the volunteer prisoners have always intended to use syringes. 5-The average age of starting the injection has been 25 which simply shows a young majority which can help spread the diseases in the society more rapidly. And almost half of the mentioned prisoners are married and will act as a bridge to spread the disease in the society in a faster pace. 6-The other demographic characteristics of this group of IDU's including education, profession, etc. should be considered in harm reduction programmes planning. 7-The high frequency of drug-related crimes shows the necessity and importance of considering these prisoners in harm reduction programmes. 8-As mentions by the IDU's, they inject drugs three times a day and this figure can help in determining the number of needed syringes during syringe distribution. 9-A high proportion of prisoners have no laboratory records of HIV, HCV and HBV while this is the most high risk group and access to consulting services and laboratory tests must be made available for them. 10-The high proportion of prisoners with multiple partners has made this group very special in harm reduction programmes and if not properly controlled, they can be infected by HIV easily and the virus is then spread among the partners and later to the society. Syringe distribution among these prisoners can help reduce the problem.

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Small intestine enteroscopy: a new diagnostic tool not only in HIV infection

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Background: Since 2007 the department of infectious diseases of the Vivantes Auguste Viktoria Klinikum Berlin, Germany has established the single balloon enteroscopy of the small intestine. To demonstrate the usefulness of this method we report a case of a 48 year old HIV positive patient.
Methods: Case report and analysis of electronic data base for results of enteroscopies since 2007.
Results: Admission with anemia and reported bloodloss, which was not seen by professional health care workers. CD4 count 70/ml, viral load < 50 copies, HAART since 6 month. Gastroscopy and coloscopy showed no evidence of current GI bleeding and revealed no bleeding source. Staging by CT scan (cerebral, cervical, thoracical and abdominal) showed lymphnodes smaller than 1 cm, no suspicion for lymphoma, especially in the small intestine. Bone marrow biopsy was without pathological findings. The patient received blood transfusion, hemoglobin levels remained constant. To complete our diagnostic approach we performed a single ballon enteroscopy via oral routine. Approximatly 210 cm a.d. we found a semicircular ulcerating lesion followed by a circular protruding and ulcerating tumor which could not be passed. Histology showed a b-cell lymphoma (diffuse large cell, CD20 positive).
Discussion: The case report demonstrates the usefulness of the new method of single balloon enteroscopy. In our experience 10% of all enteroscopies in HIV infection discovered pathological findings with therapeutic consequences.

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Chronic HCV treatment with peginterferon-ribavirin and severe tuberculosis re-activation

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Background: Tuberculosis (T) may be reactivated following a primary, silent, and unknown T infection, when immunodeficiency (often jatrogenic in origin), or other risk factors (e.g. cancer, cachexia), become apparent. Post-primary T episodes were described also decades after a primary *M. tuberculosis* infection, in patients (p) who show apparently limited radiographic signs at chest X-ray. Some grade of immunodeficiency may also depend on the administration of associated IFN-ribavirin for an underlying chronic HCV hepatitis, as expressed by the frequent emerging of leuko-neutropenia, and altered cytokine network.
Methods: In a p aged >50 years with negative history of T, an occasional chest X-ray showed fibrous-calcified infiltrates at upper right lobe. After 11 years, due to a progressive chronic HCV hepatitis, pegylated IFN-ribavirin were started for 7 months, until a sudden occurrence of cough-hemoptisis associated with a pulmonary lesion highly suggestive of T became apparent, in the same area where some reliquates of a primary T were demonstrated 11 years before.
Results: A HRCT examination pointed out 2 different excavated infiltrates. Both direct microscropy and culture of sputum-BAL proved positive for *M. tuberculosis* (susceptible to all tested compounds), while Mantoux-Quantiferon assays also tested positive. An absolute lymnphopenia (nadir 966 cells/μL), prompted a T-cell subset study, which showed an imbalance of the CD4/CD8 ratio (30/45%), and an absolute CD4 count of 290 cells/μL. Notwithstanding 7 consecutive weeks of isoniazide, ethambutol, rifampicin and pyrazinamide administration, sputum examination remained positive, thus confirming the role of immunodeficiency is prompting a difficult-to-treat T. The adjunct of levofloxacin-amikacin-linezolid attained clinical-bacteriological cure, after 12 weeks.
Discussion: Waiting for human experimental data, two animal models demontrated that an increased release of immunosuppressive cytokines (IL-10-TGF-β), may prompt T reactivation, while a maintained T-cell competence enhances T latency. Although a few cases of non-infectious lung involvement, interstitial pneumonia, and bronchiolitis obliterans were described during IFN therapy administered to transplant p, reactivated T was excepional. The expected increase of therapeutic use of IFN and potent agents for the management of chronic hepatitis or other diseases, might support the reactivation of latent T. A careful medical history, Mantoux reaction, IGRA testing, and a chest X-ray, are mandatory before starting IFN therapy. In fact, the jatrogenic immunosuppression related to IFN-ribavirin may go beyond the expected leuko-lymphopenia, and also act against the quantitative-functional role of CD4 lymphocytes. This last circumstance may play a key role in T reactivation, when T latency is of concern.

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Severe psoriasis emerged after treatment of chronic HCV co-infection with pegylated-interferon and ribavirin, supported by neutropenia rescue with repeated filgrastim administration

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Background: Associated treatment with pegylated interferon plus specific antiviral compounds significantly improved the prognosis of chronic

hepatitis C and hepatitis B even when an underlying HIV infection is of concern, although antiviral drugs (especially interferon and its derivatives), tend to be myelotoxic and also some rescue treatments, like human recombinant granulocyte colony-stimulating factors (G-CSF, filgrastim or lenograstim) (which are extensively administered in order to correct neutropenia induced by antiviral therapy), may also be involved in prompting or exacerbating cutaneous psoriasis and its systemic complications.

Methods: A representative case report of a HCV-monoinfected woman with no personal and familial history of psoriasis, who suffered from a chronic, progressive, evolutive hepatitis C, underwent long-term treatment with combined pegylated interferon plus ribavirin, and resorted to multiple cycles of G-CSF (filgrastim), in order to recover a severe, recurring granuloytopenia caused by antiviral therapy itself, and to maintain an effective dosage of both anti-HCV antivirals.

Results: Five months after treatment initiation, she suddenly developed an extensive and severe cutaneous psoriasis, which did not benefit from all attempted local therapies, and improved only after specific cyclosporin treatment. Anti-HCV therapy was first interrupted, and then continued with a lowered dosages of peg-interferon and ribavirin, so that a sustained anto-HCV therapeutic response was not obtained.

Discussion: From a pathogenetic point of view, in our case it remains extremely difficult to distinguish the role of pergylated interferon from that of the accompanying ribavirin, from that of the frequently administered granulocyte growth factor (filgrastim in our case), since all mentioned drugs were administered concurrently during many months, and according to the existing literature evidences, all of them have a potential to induce/exacerbate psoriasis as a potential untoward effect in subjects suffering from chronic hepatitis. Cyclosporin treatment obtained a stable remission of this last severe cutaneous complication, but the efforts to contain the progression the underlying evolutive hepatitis C were blunted by the difficult-to-treat genotype 1 HCV infection, and the frequent need to lower drug dosages and/or to interrupt antiviral therapy, because of initial myelotoxicity and subsequent cutaneous complications, probably driven by anti-HCV therapy itself.

P85

The experience of pain among patients living with Hepatitis C: an assessment of prevalence and needs

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Background: It is estimated that 300,000 individuals in Canada are infected with Hepatitis C (HCV). The pain experiences reported in relation to HCV appear to vary highly in both prevalence and source. Experiences of pain/pain treatment can be complicated by feelings of depression and poor sleep; whereby, pain can contribute to both depression and poorer sleep. This study will assess the prevalence and impact of pain among HCV patients from The Ottawa Hospital (TOH) including patient interest in various pain treatment options.

Methods: A questionnaire study was conducted among HCV patients seen at the Viral Hepatitis Clinic at TOH between June-December 2008. The questionnaire package contained: Socio-demographics, CES-Depression Scale, Sleep Impairment Index, and Pain Treatment Preferences.

Results: 128 HCV patients met eligibility criteria for the study; 91 (71%) completed the survey. 56% of HCV patients reported chronic pain which commonly affected their back, legs, and joints. A majority (91%) reported that they would feel comfortable telling their healthcare providers about their pain. HCV patients with pain expressed a preference for visiting their family doctor and HCV specialist for pain treatment; almost half (47%) were interested in group-based pain management. Also, HCV patients with chronic pain reported significantly poorer sleep and greater feelings of depression.

Discussion: Overall, pain is a significant treatment concern in this sample of HCV patients. These results suggest that it may be important to consider incorporating pain assessment procedures into routine clinical care for HCV patients. The timely assessment/treatment of pain among HCV patients may also be facilitated by the development of standardized clinical tools and by providing information to healthcare workers on available options for treating chronic pain including non-pharmacological psycho-educational pain management.

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Spontaneous HCV clearance in a patient with HIV infection and a concurrent, never treated, evolutive HCV hepatitis, after over twenty years of chronic co-infection

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Background: The reciprocal virological-immunological interactions between HIV and HCV, as well as the reciprocal effect of the specific antiviral therapies, are still poorly known. A rare case report of spontaneous clearance of HCV occurred in an ex-IVDA co-infected with HIV-HCV since 20 years, and never treated for HCV, is presented.

Methods: A 49-year-old ex-IVDA patient (p) tested HIV-HCV positive since 1989, and was treated for HIV disease since 1990 with limited compliance until 2 years ago. He never attained undetectable HIV viremia until the last 6 months, although CD4+ T-lymphocyte count steadily remained >300 cells/μL.

Results: Until this last semester, against medical recommendation, our p continued alcohol consumption and irregular drug addict, despite an ongoing methadone program. Serum transaminases showed fluctuating values, always >2-3.5-fold normal levels, while HCV replication was confirmed by values of 1,200-4,000 × 10^3 IU/mL. During the last 6 months, our p first abandoned its former lamivudine-zidovudine-nevirapine therapy, leading to a combination including 2 novel nucleos(t)ide analogues (tenofovir-emtricitabine), associated to the protease inhibitors (PI) lopinavir-ritonavir, and finally in the last 3 months due to gastrointestinal intolerance-hypertriglyceridemia he introduced fosamprenavir-ritonavir on behalf of lopinavir-ritonavir. Already after lopinavir-ritonavir use, our p attained undetectable plasma HIV-RNA levels (always confirmed thereafter), while CD4+ count showed the greatest values even registered by our p (513-662 cells/μL). During 2 subsequent controls, qualitative HCV viremia tested negative for the first time, concurrently with normal transaminases.

Discussion: Single cases of apparent disappearance of a chronic HCV infection in HIV-HCV co-infected p in absence of anti-HCV therapy have been described as anecdotal reports. In these episodes, a role of anti-HIV therapy (without reference to speficic drugs and associations), and that of a concurrent, significant immune recovery was often claimed. The eventual role of HIV PI, although a negligible direct anti-HCV is known, is still debated, depending on the direct-indirect role possibly played by PI in the dynemics of HIV-HCV co-infection. Fosamprenavir is a PI with contained liver toxicity, thus recommended just in p with a concomitant chronic hepatis/liver cirrhosis. A systematic revision of safety databases of fosamprenavir in HIV-infected p with chronic viral hepatitis (to detect eventual virological-immunological changes of the concomitant HCV hepatitis), and a systematic appraisal of all literature anecdotal case reports, may shed light on novel research targets in this relevant, but somewhat unexplored situation.

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Preventive behavior among doctors with respect to Hepatitis-B in bolan medical complex hospital, Quetta, Pakistan

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Background: Acquiring the Hepatitis-B (HB) infection is an occupational hazard for doctors and other Health Care Workers (HCWs). The aim of this study was to describe preventive behavior among doctors with respect to

Hepatitis-B as well as the association of socio-demographic factors, knowledge, cues to action and perception with their behavior.
Methods: A cross-sectional survey was conducted among doctors with respect to Hepatitis-B in the Bolan Medical Complex Hospital located in Quetta, Pakistan. Questionnaires were distributed to 384 doctors; 322 (84%) were returned.
Results: The majority of the respondents were male, between the ages of 31-40 years, and were medical officers working in a medical unit, from 1 to 10 years worth of experience. Almost 97% of the respondents had a high level of knowledge concerning HB infection. Respondent's perception on severity of HB was low while susceptibility perception on HB was high. The serological test for HBsAg was negative among 94.7% of respondents. Preventive behavior was dichotomized into high and low from the mean of the responses to 21 questions about specific recommended practices consistent with Universal Precautions. Chi-square analysis showed there was an association between work department (c^2 = 22.09, P-value = 0.00) as well as perception of severity (c^2 = 9.26, P-value = 0.00) and HB preventive behavior.
Respondents with low accuracy in their perception on the severity of HB were 2 times more likely to have low HB preventive behavior as compared to those with a high level of accuracy in their perception of severity: OR (95% CI) = 2.11(1.26, 3.51). Respondents working in surgery departments were 54% less likely to have low HB preventive behavior as compared to those working in medicine departments: OR (95% CI) = 0.46 (0.04,1.00).
Discussion: Although their level of knowledge was high, the accuracy of the doctor's perception on the severity of HB was low. Therefore to increase preventive behaviors, further interventions are needed to promote the perception on severity of HB.

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Seroepidemiology of infection with Herpes Simplex Virus type 2 (HSV2) among asymptomatic students attending Islamic Azad University of Kazeroun, southwest of Iran

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Background: Herpes simplex virus (HSV) infections are among the most common infectious diseases in humans. The prevalence of herpes simplex viruse type 2 (HSV-2) varies widely across the world. Herpes simplex virus type 2 (HSV-2) is the cause of most genital herpes and is almost always sexually transmitted.
Most HSV-2 infections are consequently expected to occur after the onset of sexual activity. Genital herpes is a cause of morbidity and increases the risk of HIV acquisition, due to disruption of mucosal membranes.
Data on prevalence of herpes simplex virus type 2 (HSV-2) infections are limited in Asia.
Our study focuses on seroepidemiology of HSV-2 infection in Islamic Azad University of Kazeroun asymptomatic healthy students.
Methods: In our descriptive study, the study group comprised 360 students with the average age of 22.2. At the beginning, demographic data were recorded by using a questionnaire. For serological studies 5 ml of blood sample was collected and the serum was isolated by centrifugation. Enzyme linked immunosorbent assay (ELISA) was used for determination of immunoglobulin G (IgG) antibody titer to the HSV-2. Finally the results were analyzed by statistical methods.
Results: Overall, HSV-2-IgG antibody was positive in 84 persons (23.3%) out of 360 subjects and they had a previous infection.
We can find a significant difference in prevalence between men and women but didn't find any significant relationship between students with different field of study and their residence ($p > 0.05$).
Discussion: The overall incidence of HSV-2 infection in this study was 23.3%.
Certainly information on age- and gender-specific prevalence of HSV-2 infection is crucial to guide herpes control strategies and prevention of HSV-2 infection should target individuals before they become sexually active.

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Abstract withdrawn

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Disturbance of HDL apolipoprotein AI metabolism in severe hyperlipidemic and lipodystrophic HIV patients on a protease inhibitor treatment

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Background: The aim of this study was to characterize the metabolic abnormalities resulting in low HDL apolipoprotein AI (HDL-AI) levels in lipodystrophy HIV infected patients during protease inhibitor therapy.
Methods: Seven HIV infected patients, normolipidemic with no lipodystrophy (group A) and seven hyperlipidemic with lipodystrophy (group B) were studied. Patients are on protease inhibitors since at least six months. Patients were underwent *in vivo* kinetics of HDL-AI using a 14 h primed constant infusion of [5,5,5,$^2H_3$] leucine. Kinetic data were analyzed by monocompartmental model using SAMII program to drive metabolic parameters (FCR, Fractional Catabolic Rate, and APR, Absolute Catabolic Rate).
Results: Subjects in group B showed significantly higher plasma triglycerides ($p < 0.05$). HDL cholesterol and apolipoprotein AI (apoAI) were significantly ($P < 0.05$) lower in group B compared to group A. HDL are more enriched in triglycerides in group B compared to group A ($P < 0.005$). Kinetic study showed no change of fractional catabolic rate between two groups but significantly ($P < 0.05$) lower APR in group B compared to group A.
Discussion: These results showed that the hypertriglyceridemia and low HDL level associated with lipodystrophy in HIV infected patients during treatment is related essentially to low absolute catabolic rate. As was almost reported, the HDL enriched in TG are quickly catabolised. In our patient from group B, although HDL were enriched in TG their FCR was normal suggesting a primary abnormality in apoAI synthesis and/or secretion.

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Pulmonary and disseminated tubercular disease by Bacillus of Calmette-Guérin after administration as a local adjuvant immunotherapy of relapsing bladder adenocarcinoma

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Background: Aim of our work is to present two exemplary cases of patients (p) who received a prolonged intravesical immunotherapy with instillations of Bacillus of Calmette-Guérin (BCG), to treat an urothelial carcinoma of the bladder relapsing after endoscopic-surgical treatment, who suffered from a severe pulmonary infection caused by *Mycobacterium bovis*. Their diagnostic pathway was particularly cumbersome due to the co-existing chronic pulmonary diseases (COPD), and prior respiratory disorders (including a juvenile tuberculosis). The second presented p was also interested by a genito-urinary (penile) localization of BCG infection, to attribute to local BCG dissemination.
Methods: Both p (aged 77-58 years, respectively), had radiological remnants of a prior tubercular infection, concurrently with a severe BPCO (at HRCT scan). Histopathologic studies showed granulomatous-necrotizing lesions with a diffuse macrophage, histiocyte, and giant cell infiltrate, followed by endoalveolar fbrosis in the second patient. No mycobacteria were detected at microscopy-culture, as well as after polymerase chain reaction (PCR) assays.

Results: Our first p had a mild positive Mantoux intradermal reaction while he tested frankly positive at the interferon-gamma release assay (IGRA), thus representing the first case of BCG-itis diagnosed also with the aid of these recent techniques. From a clinical point of view, the miliary-nodular-infiltrative picture of the first p, and the granulomatous-fibrosing aspects of the second p, showed a slow but progressive ameliorement during anti-tubercolar therapy, but the long-term follow-up detected a persisting worsening of respiratory function parameters in the second p. When discussing the potential pathogenetic correlates between BCG intravesical immunotherapy of bladder carcinoma and pulmonary and lung-urogenital disease localizations, after careful exclusion of other ethiologies we have no doubt in attributing these complications to BCG, even in the absence of bacterial isolation, just because the antigenic load itself caused by repeated instillations of a live, attenuated *M. bovis* strains (BCG) is a condition sufficient to trigger a pulmonary and systemic disease, and drive its subsequent evolution.
Discussion: To our knowledge, only four cases of respiratory BCG-itis have been reported to date (as simil-tubercular forms), two of them after periodical intravesical BCG instillations, in p with relapsing urothelial cancer. The second presented p represents the first case interested by a dual, concurrent granulomatous BCG infections, involving both genito-urinary and lower respiratory tract. Particular attention should be taken by Urologists, Internal Medicine, and Infectious Diseases specialists in collecting the history and in monitoring patients who undergo adjuvant BCG immunotherapy.

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Educational intervention and HIV infection: preliminary results

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Background: To describe demographical, socio-behavioral and clinical characteristics of HIV Infected Patients included in a educational intervention program (EIP).
Methods: We initiated in March 2009 an EIP in an HIV outdoor Clinical Unit witch follow about 1000 HIV infected patients with 40% HCV coinfected. This program included at last 3 educational consultations for each patient (at least 2 initial educational consultations permitted to do a personal educational diagnosis and to fix objectives with patients and at least 1 follow up educational consultation to validate experiences) and was performed by a specifically training nurse. This analysis focused on quality of life with a self administered questionnaire at inclusion in the program.
Results: Up to now, the EIP has been proposed to 68 patients HIV infected by physicians (73.6%), by nurses (22.6%) and during pluridisciplinary staff (3.8%). 53 patients (79%) have been included (45.3% females, median age: 43.9 years, median HIV follow up: 13.9 years, median cART exposition: 11.8 years, median CD4 Cell Count/mm^3: 324, and median plasma HIV viral load copies Log/ml: 1.87) in 22 cases for bad adherence, in 22 cases for initiation or changing therapy, in 7 cases for HIV positive diagnosis and in 2 cases for virological failure.
At 31/10/2009, 160 educational consultations were realized, that is to say 3,02 consultations/patient over 8 months (mean duration of session = 45 min). The dates (meetings) were respected by 79% of patients and the self-administered questionnaires were all filled.
Amoung the 53 patients, 81,1% live alone and 42,8% have precarious situation. Vulnerability/Stress factor and handicap was found in respectively 73.3% and 46.3%. A bad knowledge of HIV infection and therapy was found in respectively 32.3% and 41.9%. Near of 20% declared to have unprotected sexual behavior.
We found a correlation between the level of knowledge of HIV infection or treatment and quality of life.
Discussion: These preliminary results stress the need of such program in HIV Clinical Unit.

P93

The frequency rate of nosocomial urinary tract infections in intensive care unit patients in Shafiieh Hospital, Zanjan

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Retrovirology 2010, **7(Suppl 1):**P93

Background: Can be considered to have a nosocomial: Hospital-acquired infections encompass almost all clinically evident infections that do not originate from patient's original admitting diagnosis. Within hours after admission, a patient's flora begins to acquire characteristics of the surrounding bacterial pool. Most infections that become clinically evident after 48 hours of hospitalization are considered hospital-acquired. Infections that occur after the patient's. This descriptive study was conducted to determine the incidence rate of nosocomial urinary tract infections among intensive care unit patients, and the predominant infecting organisms in Shafeeieh hospital of zanjan's citydischarge from the hospital origin if the organisms were acquired during the hospital stay.
Methods: Of the 150 patients studied that were chosen base on purposed oriented sampling, 75.8% were males and 24.4% were females. Data were collected by: 1.a questionnaire and a Health Evaluation Score: Informathon for each patient was collected concerning demographics(age and sex), diagnosis, underlying diseases, nutritional status, length of stay, length of catheterization an clinical status on admisstion, failure together with an Acute Physiology And Chronic Health Evaluation(APACHEII) Score calculated from raw data collected in the first 24 hours of ICU admission. In order to determine severity of illness with APACHEII Score patients categorized to three groups: sever(n = 23), moderate(n = 11)and mild(n = 4). 2. Urine culture samples were taken every 48-72 hours until patients discharge from ICU. 3. the presence or absence of infection was documented according to the standard definition of the CDC. Analytic of data was conducted by SPSS software.
Results: Incidence rate of NUTI[1] was 25%(n = 38). Most frequently reported microorganisms were: E. coli(50%), Staphylococcus(17.5%), Klebsiella(7.5%), Fungi isolated (5%), Entrobacter (2.5%), and the others (17.5%). Chi – square(X2) test showed significant variation in females in regard to males. Variance Analytic showed significant variation in age greater than 75 years was associated with NUTI(in regard to length of stay)($p < 0.05$). Female patients with NUTI were 1.28 times the risk of the men. Patients with age greater than 75 years were 1.88 times the risk of age >45 years. The odds ratio of infection increased dramatically with increasing length of stay($p < 0.05$). Patients with urinary catheter were at particular risk for urinary tract infections. Those who had urinary catheter between over 8 days were 2.08 times risk of the 1-4 days patients.
Discussion: The survey has identified and confirmed the importence of certain risk factors (in particular length of stay and urinary catheterization) that increase the risk of developing ICU-acquired Infection($p < 0.05$). the incidence rate of urinary tract infections was 25%.
Reference

1. Nosocomial Urinary Tract Infections. .

P94

Hospital based palliative care approach to HIV/AIDnt; a Nigeria experience

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Background: This study evaluates the role of Hospital Based Palliative Care services in scaling up HIV/AIDS management and reduction of social and self discrimination in HIV/AIDs patients.
Methods: The hospital medical advisory approved a Multidisciplinary team consultation and the inauguration of the HIV/AIDS clinic under the chairmanship of a trained palliative care physician. The clients consult any of the hospital specialist team depending on the needs and these include

peadiatrician, community physician, pharmacist, social workers, nutritionist, surgeon, internal physician, spiritual leader, obstetrics and gynaecologists.
Results: The clients number increased from six (6) to seventy-five (75) within 3 months. More than two-thousands patients are currently enrolled with 72% on free HAART. A significant success had been recorded in the Prevention of Mother to Child Transmission, drug adherence, home based care and support group.
Discussion: The multidisciplinary team care approach facilitated the upsurge in the number of new cases and an improved confidence in the health care providers. The previous culture of discriminating against the patients by the hospital staff is now replaced by friendship and improved quality of care.

P95

Destruction HIV viral RNA by siDNA triggering RNAse H

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Background: We are developing an alternative approach to siRNA, which may be designated as siDNA, small interfering DNA, by using hairpin-loop-structured DNA oligodeoxynucleotides (ODN), targeted to viral or cellular mRNAs. ODNs activate the viral RNase H in retroviral particles and cellular RNases H inside the cell. Also Ago2 may play a role. Other inhibitory mechanisms such as translational arrest may contribute.
Methods: We selected ODNs against various viral and mRNAs of HIV, HSV, Influenza, HCV, HBV, and the terminal repeat of telomerases in malignant melanomas in mice. The ODNs were applied with or without carriers. Furthermore their effects were directly compared to those of single-stranded antisense DNAs and siRNAs to allow comparison of the various efficiencies.
Results: The ODNs were most effective in HIV. We are able to induce HIV suicide and inactivate HIV virus particles to prevent infections, inactivate cell-free HIV in the blood from infected individuals, in the vagina of mice, and increase survival time of retroviral-infected mice. Also influenza virus replication was reduced in the lungs of a mouse model. Furthermore we could reduce malignant melanoma-formation tumor formation in mice targeting the telomerase. The effects are sequence- and dose – dependent, but the optimal algorithm is not yet known. We are analyzing whether there is a preference for G tracts, which may form higher-ordered structures and enhance uptake.
Discussion: The dsODNS are often superior to single-stranded antisense DNA and resemble the effects of siRNAs but with different kinetics. The method may complement existing silencing approaches.

P96

A research report on "positive prevention" intervention in two ethnic habitats along the border with myanmar in yunnan

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Background: "Positive Prevention", which targets on people living with HIV/AIDS (PLHIV), aims to improve their life quality and living standard, protect their families and communities, and help reducing HIV infection. This report evaluates a "positive prevention" training intervention project in Ruili and Longchuan of ethnic group Counties along the border with Myanmar in Yunnan province, China.
Methods: 312 HIV-positive volunteers of ethnic groups enrolled in invention cohort in Ruili had received "Positive Prevention" training. In contract, 308 HIV-positive volunteers of similar ethnic groups were enrolled in non intervention cohort in Longchuan after baseline surveys for the two groups. 12 months after the training, 203 participants in Ruili and 274 participants in Longchuan were traced respectively. Qualitative and quantitative methods of focused group discussion, in depth interview and in site observation were used to assess their health knowledge, health belief, health behavior, and health outcome.
Results: Demographic and occupationalcompositions of the participants from the two counties are very similar. The group in Ruili resulted in significant increase of health knowledge, improvement in perceived susceptibility and severity of diseases such as STIs and hepatitis, and perceived benefits of health behaviors, and willingness to take a role in the HIV/AIDS prevention actions. Among Ruili participants, condom protected intercourse in the latest sexual behavior increased dramatically from 77.97% at baseline to 97.69% (χ^2 = 21.438, P = 0.000 < 0.05). New HCV incidence rate was 8.0% in Longchuan and 3.97% in Ruil while new HBV incidence rate in Longchuan was 1.96% and 0.98% in Ruili. The low new incidence rate in Ruili was attributed to the changes as a result of the intervention.
Discussion: This "Positive Prevention" training intervention has improved health knowledge and health belief of intervened PLHIV and reduced their high risk behaviors to some extent. It imposes an impact on prevention of new infection for PLHIV. The lessons learnt from this project can be used for options to improve "Positive Prevention" strategies in terms of selection of target population, design of intervention activities, approaches and sustainability in ethnic habitat communities.

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Results from a multimedia testing and counseling program in an urban emergency department

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Background: This study evaluates a novel approach to counseling and testing in a high-volume inner-city ED which utilizes an HIV counselor and a multimedia tool for conveying video HIV information and electronically collecting risk factor data. We evaluated this program to assess the demographic and risk factor characteristics of all patients tested, patient-reported satisfaction with the program and outcomes for positive patients.
Methods: This prospective cross-sectional evaluation was conducted for 2 years. A convenience sample of medically stable patients presenting to an inner-city municipal hospital ED were recruited by 3 to 8 full-time equivalent HIV counselors. Previously developed and validated videos for HIV pre- and post-test counseling were used. Demographic characteristics, risk factors, and satisfaction information were collected using patient self-reporting on the touch screen computer. Data downloaded automatically into a secure database. Chart reviews were conducted by the HIV-positive patients' medical provider to assess outcomes. Data were analyzed using SPSS software.
Results: During the federal grant period, 28,995 patients were tested for HIV. Demographic characteristics of the participants were: 41.9% male, mean age 36.0 ± 14.2 years, 54.7% Hispanic, and 32.2% African-American. Risk factors were: 6.4% MSM, 31.0% had multiple sex partners in the past 3 months, 49.8% reported condom use as "never," 1.5% used injection drugs. Patient satisfaction was high: 88.8% reported learning a moderate-to-large amount of new information about HIV and 78.6% preferred the format which included both videos and an HIV counselor. There were 101 newly diagnosed or confirmed HIV positive patients and 86% were linked to outpatient HIV care; mean days to first medical visit was 7. Positive patient outcomes were as follows: 85% of eligible patients began HAART, median days to HAART treatment was 35, 62% of patients on HAART had viral load less than 400 copies/mL.
Discussion: A rapid HIV program using a multimedia tool and a counselor in a busy inner-city hospital ED can effectively test a large number of patients, provide consistent prevention messages to patients who report multiple HIV risk factors and link a large percentage HIV-positive patients to existing health care systems.

P98

Recent HIV infection among newly diagnosed with HIV cases in Turin, Italy

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Background: Knowledge of the proportion of recent HIV infections (RHI) is important for epidemiologic purposes to assess HIV transmission patterns and evaluate the impact of prevention interventions. The aim of this study was to analyze the characteristics of persons with RHI and to monitor the trend of RHI over time.
Methods: In the period 2003-2004 and in 2007-2008, serum samples from individuals newly diagnosed with HIV infection were collected in the Infectious Diseases Hospital in Turin. All serum samples were tested for the HIV antibody avidity index (AI); samples with an AI ≤ 0.80 were defined as RHI (≤ 6 months from seroconversion).
Results: In the study period, 432 serum samples were collected from newly diagnosed HIV individuals. The number of persons with RHI was 113 (26.2%), and the proportion of RHI significantly increased from 22.1% in 2003-2004 to 30.8% in 2007-2008 (p-value < 0.05). The proportion of RHI was higher among MSM compared to heterosexual individuals (33.0% vs. 21.0%, p-value = 0.06). The median age of persons with RHI was similar between MSM (35 years) and heterosexual persons (34 years), as well as between 2003-2004 and 2007-2008 (35 years). A significantly higher proportion of persons who underwent a previous HIV test was observed among RHI (51.3%) compared to those with an established infection (28.5%) (p < 0.001). The proportion of RHI among MSM increased from 25.5% in 2003-2004 to 40.7% in 2007-2008, whereas remained almost stable among heterosexual persons and injecting drug users. Disaggregating by reasons of HIV testing, different proportions of RHI were found: 34.8% among those exposed to unprotected sexual intercourse, 24.5% among injecting drug users, and 11.5% among those screened during pregnancy or blood donation.
Discussion: RHI have increased over time in Turin, especially among MSM. However, no changes were observed in the age of HIV acquisition by sexual orientation and time period. The identification of RHI can be affected by the frequency of HIV testing and this bias should be considered in estimating HIV incidence.

P99

HIV/AIDS epidemic features and trends in setif city (Algeria) from 1986 to 2009

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Background: To identify the epidemiologic characteristics of HIV and AIDS in SETIF during the period between 1986, and 2009.
Methods: This report presents a descriptive analysis of HIV and AIDS surveillance data. The subjects of this study were all notified HIV and AIDS cases in SETIF diagnosed by ELISA and confirmatory Western blot, we worked with epi info 3.5.2
Results: The HIV epidemic started in SETIF in 1986 with the first diagnosed AIDS cases. The number of cases slowly but steadily increased, to reach, by 2009, 152 cases. The median age of infected persons was 39,77 years. 40.8% were WOMEN; The most frequent mode of transmission is sexual (76,9% of all cases; with mainly heterosexual transmission 77.9 percnt; with a strong correlation to travel and migration 37.20%). the proportion of persons diagnosed late (within 12 months before AIDS diagnosis) was 82,3%; 91.4% of married men (36) have sexual relationships outside marriage as compared to 0.0% of married women (27). We report also 13.60% discordant couples among married patients, Among documented HIV-positive mothers (36), their children were screend in 61.10%, and 95.5% of them were seronegatif, 55,9% of all cases were treated with ARV. HIV/AIDS deaths 36,2% decreased markedly from 2002, associated with the advent of HAART.
Discussion: Gender distribution and distribution by mode of transmission among HIV-infected revealed an equal gender distribution of infection wich contrary to sub-Saharan Africa, where women are more affected.
a higher female prevalence of HIV infection might be expected in algeria since heterosexuality is the prevailing mode of sexual transmission.
Non-marital sex is the main risk factor, as it is worldwide for several reasons:
Our data might be explained by
ü the large number of males who travel to other countries, especially to areas with a high prevalence of HIV infection. Travel and tourism enhance the probability of having casual sex, a fact that increases the risk of exposure to sexually transmitted infections
ü The ever-decreasing religious values
Adults 23- to 45-years old were found to constitute 65,8% of all cases, a fact that points to the serious social and economical impact of the disease
The trend of mode of transmission by blood transfusion shows that most of the cases infected through contaminated blood were registered before 1995. These patients most probably received blood unscreened for HIV in the 1980s before blood safety measures were undertaken. However, the incidence of this type of transmission significantly declined after implementation of blood safety policies.
32,90% of infected had died, This high percentage can be explained by the fact that:
most infections are detected among patients, who already have a bad prognosis 66,40%
68,6% of death occurs before 1996 it means before the introduction of highly active antiretroviral therapy.

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HIV-related risk factors among male sex workers in different settings in Shenzhen, China

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Background: This study aims to investigate the prevalence of HIV infection and identify HIV-related risky behaviors of MSW in different settings in Shenzhen, China, in order to tailor the venue-based prevention interventions to this unique subgroup of men who have sex with men (MSM).
Methods: A total of 394 participants were randomly selected among MSW in Shenzhen, from April to July 2008, using time-location sampling. Each participant was asked to finish a guided self-adminstered questionaire, focusing on their behaviors and knowledge. Field observations were conducted at the same time. Their serological data were collected and tested for HIV, syphilis and HCV.
Results: Among 394 subjects, the prevalence of HIV infection was 5.1%, with 6.9% among MSWs in parks (PMSW), 11.3% among MSWs in family clubs(FMSW, a kind of home-based brothels holding small group of MSWs <= 15) and 1.7% among MSWs in entertainment venues (EMSW, holding big group of MSWs >15). Syphilis prevalence was 14.2%, with 25.0% in PMSWs, 17.6% in FMSW and 10.6% in EMSWs. Both HIV and syphilis prevalence were significantly different among different type of MSWs. Independent PMSWs and small grouping FMSWs had a higher proportion of self-identifying as homosexual/gay and fewer female sex partners, elder or higher polarization in age. In addition, a higher proportion of MSWs was found coming from provinces with higher HIV prevalence, and lower coverage by HIV-related education program and relevant services.
Discussion: Different type of male sex work is associated with HIV infection. MSWs working in family clubs and parks are comparatively with higher risk of being infected, while MSWs working in big entertainment venues showed a lowest HIV prevalence within the past few years in Shenzhen. This situation indicated that current HIV prevention intervention strategy that focus on big venues is quiet effective and should be expanded to cover the MSM population frequenting small venues.

P101

The trend of HIV/AIDS prevalence among IDU's in Iranian prisoners (1376-1386)

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Background: Prisons are recognized worldwide as important sites for transmission of blood-borne viruses (BBVs). the high prevalence of HIV infection and drug dependence among prisoners, combined with the sharing of injecting drug equipment, make prisons a high-risk environment for the transmission of HIV and the lack of supply of preventive measures (such as sterile needle and syringes or condoms or methadone maintenance therapy). In most prisons of world, because of a variety of social conditions, extra opportunities for BBV transmission are created.
HIV prevalence in Iran is generally more than 8 times higher in prisons (1.75) than in general population estimation (0.2) because of the considerable over-representation of injecting drug users (IDUs) among prisoners. We want to study the trend and outcome of interventions of HIV/AIDS in IDU's prisoners of Iran during 1997-2007.
Methods: Based on the HIV test outcome in prisons due to sentinel services during a 9-year period from 1997 to 2007, we used the data from all sentinel services in all prisons all over the country during this period. Annual HIV prevalence among prison inmates in Iran was determined and also major interventions during these times investigated.
Results: In an 11-year time period from 1997 to 2007, 107 sentinel services have been established among drug user prisoners, in which as a whole 42142 people were studied. Infection results during different years are as follow:
• 1997, prevalence rate 0.15 percent, CI = (0.13,0.16) and number (3/2022)
• 1998, prevalence rate 0.30 percent, CI = (0.28,0.31) and number (7/2367)
• 1999, prevalence rate 0.48 percent, CI = (0.46,0.50) and number (8/1670)
• 2000, prevalence rate 3.17 percent, CI = (3.07,3.27) and number (2553/81)
• 2001, prevalence rate 2.17 percent, CI = (2.13,2.22) and number (99/4556)
• 2002, prevalence rate 4.01 percent, CI = (3.92,4.10) and number (236/5881)
• 2003, prevalence rate 3.39 percent, CI = (3.31,3.47) and number (153/4515)
• 2004, prevalence rate 4.11 percent, CI = (3.99,4.22) and number (157/3824)
• 2005, prevalence rate 4.86 percent, CI = (4.74,4.98) and number (239/4920)
• 2006, prevalence rate 2.99 percent, CI = (2.92,3.05) and number (157/5226)
• 2007, prevalence rate 2.34 percent, CI = (2.29,2.39) and number (107/4571)
Two important and effective interventions performed in Iran prisons in this time period include:
1. Initiation and extending substance methadone therapy in such a way that its coverage improved from 300 prisoners in 2003 to 19500 prisoners in 2007.
2. Another important intervention was the establishment of triangular clinics (voluntary counseling testing) in Iran prisons. It started with the coverage of 1 in 2001 and reached the coverage of 105 in 2007.
Discussion: The 11-year trend of HIV among Iranian prisoners showed that although the prevalence was low in the beginning years, it gradually got a rising trend and it reached its pick of 4.86 percent in 1384 and then it started to decrease again. Although the last HIV prevalence rate among Iranian addicted prisoners was extremely more than general population of the country, its falling trend could be a symbol of effectiveness of performed interventions to decrease HIV prevalence. Therefore, two main intervention i.e. initiation of voluntary counseling & testing centers and substance methadone therapy (MMT) that were used as AIDS control strategies in Iranian prisons were greatly suitable and efficient.

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HIV infection newly diagnosed in Northern Italy; evolving trends

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Background: To assess prospectively all newly diagnosed cases of HIV infection performed at our reference centre, which serves around 800,000 inhabitants of the Bologna metropolitan area.
Methods: All patients with a newly diagnosed HIV infection were initially assessed according to several demographic, epidemiological, diagnostic, clinical, and laboratory features.
Results: From June 2006 up to December 2008 (31 months), 162 patients were first diagnosed with HIV disease (mean 5.2 novel cases per month), and 78 of them were judged to have a recent infection (as established on the ground of a specific "avidity" serologic testing). Males greatly prevailed over females (93 *versus* 69 cases), while homosexual exposure (53 cases) was prevalent over heterosexual one (38 patients), and only two novel cases were registered among i.v. drug users. The median age at diagnosis was 36.2 years, while the main laboratory parameters showed a mean CD4+ count of 502 cells/μL, and a mean HIV-RNA load of 8.21×10^4 copies/mL. Although subtype B of HIV greatly prevailed (141 cases: 87%), also subtypes A-A1, and recombinant HIV virions were found among newly infected patients. When conducting a genotypic resistance assay including all available antiretroviral agents, the overall prevalence of primary mutations accounted for 13% of newly infected patients: 11.1% of them had one or more mutations of the reverse transcriptase gene, and 9.3% of subjects had one or more mutations of the protease inhibitor gene (including one case of multiple mutations, probably conferring resistance extended to the third-generation protease inhibitor tipranavir). The majority of patients with recent infection (50 out of 78: 64.1%) were asymptomatic or paucisymptomatic, while a full-blown AIDS or a symptomatic disease were present in 18 and 10 cases respectively.
Discussion: Nothwistanding the massive prevention campaigns of the last two decades, HIV infection continues to spread predominantly via sexual route, and may increasingly involve immigrants. A delayed-missed recognition of HIV infection poses patients at a very high risk to develop HIV-related disorders (since these subjects could not take advantage from antiretroviral therapy). Non-subtype B viruses, recombinant viruses, and HIV strains already encoding for resistance against different antiretroviral compounds are of significant concern. A permanent, active monitoring of this phenomenon and its correlates is strongly needed.

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Men who have sex with men(MSM) and HIV epidemic in China: a web-based study on MSM

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Background: To explore MSM population's contribution to HIV epidemic in China through learning AIDS-related knowledge, attitude and behavior among the hidden population-MSM.
Methods: A web-based cross-sectional study on MSM population, recruiting Men who had sex with men in the previous 3 years in mainland of China through a variety of available ways, such as Internet, mass media etc. and collecting data by online questionnaires and in-depth interview through chat-software QQ or MSN.
Results: A total of 5710 eligible participants from 30 provinces (municipalities and autonomous regions) except Tibet, and 97.2% aged between 18 and 44 yrs, 63.3% thinking that they are likely to be infected by HIV, and 88.7% knowing counseling hotline on HIV whereas only 8.1% of them dialed, and 59.8% knowing at least one site for HIV test whereas of which 40.1% did, and the number increasing year by year, and in the past 6 months,90.3% dating male partners via Internet, and 87.3% having sex with men and 18.1% with both men and women and 12.8% reporting STD-related symptoms. Of 5442 participants, 99.6% told one

could be infected HIV through anal sex with HIV cases and 90.3% through oral sex, and 95.0% knew it was more easily infected HIV having unprotected sex with more men. Among 4916 participants who had sex with men, 67.2% had more than one male partner. The proportions of 100% condom use when having anal sex with fixed partners, occasional partners, Money-Boys and customers in the past 6 months were 30.0%, 42.7%, 66.7%, 57.9%, respectively; when having oral sex were 7.0%, 8.8%, 6.2%, 4.4%, respectively. The proportion of 100% using waterbased lubricants was 45.8%.
Discussion: Most of MSM are at risk for HIV/STD infection. With increasing HIV prevalence in MSM population, MSM population would make a strong impact on HIV epidemic in China.

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Abstract withdrawn
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Killer immunoglobulin-like receptor genes and heterosexual HIV-1 transmission
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Background: Killer immunoglobulin-like receptor (KIR) genes regulate natural killer cell function. KIR gene content has been reported to influence HIV-1 acquisition and progression, but consensus is lacking. We investigated the impact of KIR and KIR/HLA genes on heterosexual transmission in an African cohort.
Methods: Between 1995 and 2006, 566 HIV-1 serodiscordant couples in Zambia, were followed for counseling and serologic testing for a minimum of nine months. KIR genes and HLA alleles were detected by standard typing methods. We tested the association of KIR genes and KIR gene/HLA ligand combinations with HIV-1 transmission and with index partner viral load (VL). All analyses of VL by linear regression were adjusted for age, sex, and time from enrollment. Covariates in proportional hazards models of HIV-1 transmission included VL in all index partners and genital ulcers in all partners.
Results: In the index partners KIR2DS4*001, the only *KIR2DS4* allele to encode a full length receptor, was associated with higher rates of HIV-1 transmission (OR = 2.40, 95% CI = 1.31-4.39, $p = 0.003$) by logistic regression. Survival analysis for KIR2DS4*001 demonstrated accelerated transmission of HIV-1 (RH = 1.72, $p = 0.005$). This allele in the seronegative partners was not associated with acquisition. The KIR2DS4*001 allele was also associated with a high VL (0.17 ± 0.08 $\log_{10}$, $p < 0.05$). No association was observed with its ligand, HLA-Cw*04, or with the HLA-Cw*04/KIR2DS4*001 combination. Previously reported findings on other KIR genes could not be confirmed.
Discussion: We observed an association of KIR2DS4*001 carriage by a seropositive partner with an increased hazard of HIV-1 transmission and with high VL. Whatever the significance of this KIR allele association in Zambian couples, it did not depend on epistatic interaction with HLA-Cw*04. The association will require confirmation in functional assays.

P106

Non-injected illicit drugs and alcohol and HIV-related high-risk sexual behaviors in a street-recruited sample of non-injecting drug users in New York City
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Background: Unlike injecting drug use, non-injecting drug use has not been definitively shown to be a risk factor for the sexual transmission of Human Immunodeficiency Virus (HIV) infections. HIV-related high-risk sexual behaviors would be more prevalent among non-injecting cocaine users than non-users and among multidrug users than single drug users.
Methods: Among the Non-Injecting Drug Users (NIDUs) in the South Bronx and Harlem, New York City questionnaires were administered about the past use and within last three months use of crack cocaine and powder cocaine, alcohol, marijuana/hashish, and heroin and HIV-related sexual behaviors. Sexual behavior was summarized as a single variable, the Vaginal Equivalent Episode (VEE) score; VEE scores above and below the median were categorized as high and low sexual risk respectively.
Results: Among 264 NIDUs who completed the questionnaires, 132(84 males, 48 females) used crack cocaine, 139 (101 males, 38 females) powder cocaine, 194 (140 males, 54 females) marijuana/hashish, and 110 (71 males, 39 females) heroin in the last three months. In a model that controlled for age and sex, only powder cocaine was associated with HIV-related sexual behaviors (recent use AOR = 1.87; 95% CI: 1.19-2.93; past use AOR = 2.50; 95% CI: 1.08-5.81). High-risk sexual behaviors were also more common among the users of all four drugs studied than among users of only one (OR = 1.50; 95% CI: 1.05-2.13).
Discussion: Like some earlier studies, the present study found cocaine to be strongly correlated with high-risk sexual behaviors. Future research is needed on the mechanisms of that association and on the association of the use of other non-injecting illicit drugs with HIV-related sexual behaviors. The results also support the promotion of HIV preventive measures, such as condoms, among sexually active NIDUs.

P107

A validated stigma scale measures decreased HIV-related stigma among men in a community-based HIV prevention services program in rural Maharashtra, India
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Retrovirology 2010, **7(Suppl 1):**P107

Background: To validate an HIV stigma scale among men in a high HIV-prevalent rural Indian district, and measure six-month changes in stigma after introducing an HIV prevention services program.
Methods: Between August 2006 and April 2007, a community HIV-related behavioural change and HIV testing services program was initiated in rural Aurangabad district, Maharashtra, to increase HIV services knowledge, reduce HIV stigma, and increase testing utilization. A questionnaire was administered to random cross-sectional samples of 400 adult men age 18-49 at Baseline and in the Post-Intervention and a separate post-Control community. Principle component factor analysis was used to develop a 14-item stigma scale. Stigma indices were calculated and dichotomized.
Results: Factor analysis consistently identified 3 HIV stigma subscales in each sample population. From Baseline to Post-Intervention, high stigma levels significantly decreased Overall (42% vs. 20%), for Fear of HIV Transmission (33% vs 14%), and for Perception of Enacted Stigma (34% to 14%) ($p < 0.001$). High stigma levels increased for Moral Judgements (79% vs 86%, $p < 0.05$). High stigma correlated independently with low education (AOR 2.7, 95%CI 1.6-4.3), and low HIV knowledge (AOR 3.5, 95%CI 2.2-5.5). Odds of high stigma reduced over 45% with participation in the Intervention program (AOR 0.54, 95%CI (0.36-0.82)).
Discussion: This HIV stigma scale, identifying three stigma subscales consistent with other studies, was validated in 3 community-based samples of rural men in. The HIV prevention services program reduced high HIV stigma overall and in HIV knowledge-related stigma domains. The program's current efforts include addressing Moral Judgment attitudes and longer-term study for impact on HIV testing utilization. Measurably reducing HIV-related stigma is essential for HIV prevention in rural.
This HIV stigma scale, identifying three stigma subscales consistent with other studies, was validated in 3 community-based samples of rural men in India. The HIV prevention services program reduced high HIV stigma overall and in HIV knowledge-related stigma domains. The program's current efforts include addressing Moral Judgment attitudes and longer-term study for impact on HIV testing utilization. Measurably reducing HIV-related stigma is essential for HIV prevention in rural India.

P108

Difficulties of routine rapid HIV screening in emergency department

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Background: To evaluate the feasibility of routine rapid HIV screening in an emergency department of a general hospital.
Methods: From January 2008 to August 2008, all patients admitted from 8 AM to 4 PM to the emergency unit of the hospital were offered a rapid screening for HIV infection. After inform consent, HIV rapid test was performed by the biology laboratory. Result of the test was transmitted to the clinicians who informed the patient of the result of his test. An ELISA was systematically performed later to confirm the results of the rapid test.
Results: During the period of the study, 16 024 patients were admitted to the emergency department. Routine screening was proposed to 420 patients. 19 patients denied the test. So, only 401 HIV tests have been performed (2,5% of the total number of eligible patients). 1 test was positive (0,25%). No discordance between rapid tests and ELISA were observed.
Discussion: Despite a high acceptance of screening by the patients and high sensibility and specificity of rapid tests, these results seem disappointing. Only a very low minority of patients were offered an HIV screening. The main explanation seems to be a reluctance of clinicians to propose the test to their patients. Arguments advanced by physicians to explain this were various: lack of time, reluctance to obtain inform consent, questions about the interest of routine HIV screening in emergency department... So, reflections must be conducted to increase the acceptance of routine screening by physicians. One of the promising options would be to use an opt-out procedure.

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Abstract withdrawn

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P110

Subacute and rehabilitation care facilities « Soins de Suite et de Réadaptation (SSR) », dedicated to HIV patients in Marseilles, France

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Background: To describe sociodemographic and clinical characteristics of HIV inpatients of Polyclinique La Feuilleraie in Marseilles. Five beds of SSR are dedicated to HIV people in this clinic.
Methods: Local database from January 2003 to June 2009.
Results: Admission criteria were wide, including patients ongoing opioid substitution treatment or still intravenous drug users. Patients were mostly transfered from acute medical care units in Marseilles geographic area (88%). Therapeutic objectives vary from monitoring somatic recovery, monitoring newly prescribed HAARTand VHC therapy, seeking for supportive housing, palliative care...
There were few patients who had long and repeated stays in the unit (103 pts with 199 stays). Length of stay exceeded 31 days for 42% of the patients. Most of them have low socioeconomic status (63% COTOREP-AAH,31% CMU,7% RMI). Disability profile was rather severe - measured by impairment in activities of daily living, ADL, and instrumental activities of daily living, IADL - with one third of patients with ADL<3 and mean IADL score of 2.
Immune status was low, with 49% of the patients who had less than 200 CD4/mul and 42% < 500 CD4/mul. 56% had AIDS and 60% were co-infected by VHC virus. 40% of patients had psychotropic medications on admission. Discharge destination was not adequate for 21% of the patients (patients with neurocognitive disorders, and/or psychiatric disorders and/or housing problems, who needed housing in special structures). Therapeutic objectives were not reached in one third of the stays.
Discussion: SSR dedicated to HIV inpatients in Marseilles remains a pilot experience in the region. Patients have growing inadequate discharge orientation, and growing lengths of stays, which are not compatible with the definition of SSR. New payment-by-the-act funding system should consider the specificity of HIV care in SSR.

P111

Identifying practices and strategies to increase HIV testing among Haitian migrant communities

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Background: Quebecers of Haitian origin (QHO) are disproportionately infected with HIV/AIDS as a result of socio-economic, cultural, and migratory factors. The Public Health Department of Montreal (PHDM) sought to identify practices and strategies to increase HIV testing in Haitian migrant communities.
Methods: Key informant interviews were conducted with representatives of community-based organizations providing HIV/AIDS services to the largest Haitian communities in the USA (New York, Boston, and Miami). A thematic analysis was completed to categorize qualitative data subsequently grouped into themes.
Results: • Rapid HIV testing had a significant role in decreasing the likelihood of Haitians not returning for their results due to stigma. It was also the preferred method of HIV testing for community level interventions due to its flexibility, mobility, and ease in carrying out.
• Group level interventions (GLI) targeting demographic groups were effective in increasing HIV/AIDS education, promoting behavior change, and decreasing stigma in Haitian migrant communities. Based on social cognitive learning theory, GLIs use curricula that are non-judgmental, culturally sensitive, and linguistically appropriate.
• Community level interventions (CLI) in Haitian migrant communities engaged large audiences and increased HIV testing among populations who do not access conventional testing. CLIs included the use of mobile HIV testing vans, health conferences and fairs providing HIV testing, and outreach workers to conduct street outreach.
• Key informants advocated partnerships with clinics, hospitals, churches, and the business community. Many interventions to increase HIV/AIDS education and testing were conducted with stakeholders due to their unique capacity to mobilize and influence Haitian migrant communities.
• Collaborating with local Haitian media to design culturally competent public service announcements were seen as a valuable and cost-effective method to provide HIV/AIDS education, promote HIV testing, and recruit participants for GLIs and CLIs.
Discussion: Increasing HIV testing among QHO requires a comprehensive strategy designed and implemented by a multitude of stakeholders. Strategies to address stigma and low HIV/AIDS education are best tackled via GLIs and CLIs implemented by organizations trusted by QHO. The PHDM should also re-examine current HIV testing policy in Quebec and consider increasing access to rapid HIV testing.

P112

HIV/AIDS epidemic in Belarus

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Background: Main objective of our work is the estimation of an epidemiological situation on HIV/AIDS in Belarus at the present stage.

Methods: EIA, Western blot, nested RT-PCR, PCR, sequencing, statistical
Results: The HIV/AIDS epidemic in Belarus can be divided into two stages. The first stage (1986-1996) was characterized by low prevalence and incidence of HIV-1. The total number of HIV-1-infected individuals in this period was 117, of whom 43 were foreigners who have arrived for study or work in Belarus. Since July 1996, the epidemiological situation in Belarus has changed. An HIV-1 outbreak in Svetlogorsk and Zhlobin in 1996-1997 has caused 1021 new HIV-1 infections within a six-month period. This first stage of the epidemic was characterized by:
1. Young men being mainly affected, with >60% of new infections being registered among individuals of 14-24 years old;
2. Parenteral route of infection (92% of all transmissions);
3. Epidemiological process being largely limited to the cities of Svetlogorsk and Zhlobin;
4. The outbreak was caused by a subtype A HIV-1 strain.
For October, first, 2009 10428 cases of a HIV-infection or 107,8 cases per 100 thousand population are officially registered in Belarus. HIV/AIDS Cases are registered more, than in 190 administrative territories.
The current stage of the epidemic is characterized by:
1. The epidemic still affecting young population (individuals of 19-29 years old - around 66% of new infections);
2. Heterosexual route of transmission becoming dominant (2009 - 77% of new infections);
3. Geographical expansion - the epidemic is registered in 198 administrative territories with the highest prevalence in Svetlogorsk (3, 100 cases per 100,000 inhabitants);
4. About 95% of new HIV-1 cases are caused by the "Svetlogorsk" subtype A strain.
With two-three new HIV-1 cases being registered daily in Belarus, mostly among the young generation. Such situation leads to that the quantity of children born to HIV-infected mothers annually increases. Despite of spent preventive therapy, 11% of such children are HIV-infected.
Discussion: Thus, at the present stage epidemic VICH/SPID in Belarus is in the concentrated stage, however gradually falls outside the limits risk groups. The risk group includes now young men of sexually active age: 19-29 years. For changing such situation at carrying out of preventive actions it is necessary to give more attention to the given age group. It is necessary to make epidemiological investigations of cases of infection for an establishment of a source of infection, definition to what group of risk it concerns.

P113

Cervical dysplasia in postmenopausal patients infected by HIV

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Background: An increasing number of patients infected with HIV reach the age of menopause. HIV infection is associated with a higher incidence of cervical dysplasia. The aim of our study was to describe cervical abnormalities observed in these patients during menopause.
Methods: Retrospective study at Louis Mourier Hospital, comprising all cervical cytologic and colposcopic examinations in post-menopausal HIV-infected women from 1995 to 2008.
Results: In 18 postmenopausal patients, median age 54 years (range 43 to 63) diagnosed with HIV-1 infection for 7,5 years (range 2 to 25), 51 samples were analyzed. There were 27 (50.98%) abnormalities, including 4 (7,84%) ASC-US, 15 (29.41%) low-grade lesions, and 7 (13.73%) high-grade lesions. Ten of these patients underwent surgery (laser, conization, hysterectomy) during this period. During follow-up, the lesions were stable in 40.48%, regressed in 35,71%, and progressed in 23.81%. In our study, the median onset of high-grade dysplasia during menopause is 5 years.
Discussion: Cervical lesions were frequent in postmenopausal HIV-infected women, mostly low grade and persistant. Women with HIV infection should continue to have gynaecological follow-up, with baseline HPV typing and a yearly Pap tests.

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Abstract withdrawn

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Knowledges, attitudes and practices in university students about sexual and reproductive health and HIV/AIDS In Bogotá D.C., Colombia

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Background: To identify protection and risk factors, behavioral and cognitive, in university students about sexual and reproductive health, including STDs and HIV infection.
Methods: An epidemiological descriptive cross-sectional study was carried out in students selected for multistage cluster method. Participants were surveyed through KAP questionnaire on web-based platform after having received informed consent. The survey was constructed based on similar surveys and was previously validated (α = 0.73) and collected data on knowledge and attitudes regarding sexual behaviors. Data analysis was descriptive on SPSS.
Results: 72 students from two universities (mean age 22, 70% female) were surveyed. 50% considered knowing enough about sexuality, but 33% doesn't recognize mechanisms of STDs/HIV transmission rightly and 57% doesn't associate healthy behaviors with prevention, although 82% adequately recognize the symptoms of STDs. In addition, 25% expressed stigma attitudes towards HIV-diagnosed people and 31% showed low self-efficacy for protection decisions in their sexual practices. Only 13% reported using condom always and 39% used it in their last sexual intercourse; 50% reported sexual intercourse under alcohol effect and 10% under other psychoactive substances effect. 33% expressed having been tested for HIV.
Discussion: Risky behaviors and attitudes about sex in university students make this study relevant. Prevention programs are not enough for education and awareness focused to young population and this issue contributes to continuing increase in STDs and HIV incidence. These results are an invitation for institutions to develop more appropriate prevention programs and work on psychological and social component of youth sexuality.

P116

Transition to adult clinics in youth living with HIV since birth

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Background: For youth living with HIV, transition from childhood to adulthood is also a transition from child to adult HIV health care clinics. Related studies show the more youth are spending time in pediatric services the more difficult the transition is likely to be. It appears many factors can facilitate or cause difficulties in the transition process; including individual factors and environmental factors. In this study we explore the transition process from child to adult HIV health care clinics through personal experiences of young adults infected by perinatal transmission.
Methods: Within a qualitative chronological (longitudinal) research framework, 18 youth aged from 13 to 22 years participated twice in individual semi-directed interviews within a three year interval at the Centre Maternel et Infantile sur le Sida du CHU Sainte-Justine in Montreal, Canada. At the second interview, 8 participants had gained

their majority (18 years or more). Among those, 6 participants were then attending an adult HIV clinic and 2 had abandoned their medical followup.
Results: Youth for whom the medications remained meaningful three years later reported continuous trajectories in terms of treatment plan and medical follow-up. In contrast, youth with more troublesome relationships with the medication reported trajectories that were marked by setbacks: they underwent treatment changes involving heavier dosage regimens and more serious side effects, and in many cases they stopped treatment for significant lengths of time. They felt that taking medication was a daily reminder of their HIV status, which differentiated them from their peers. In addition to the meaning attributed to their treatment, relations with caregivers tend to play an important role in the clinical transition process. Youth who reported good treatment adherence and uninterrupted follow up described the same trusting relationship and closeness with their new physician and health care team.
Discussion: Particular attention should be paid to youth encountering difficulties of adherence during their critical transition period to adult clinics. Interventions aimed at helping youth through the transition process should focus on the meaning attributed to their medical follow up and allow them to explore affective issues along their journey.

P117

Knowledge and attitudes on HIV/AIDS and sexual behaviour among male soldiers in Sri Lanka Army

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Introduction: HIV/AIDS can pose a threat to military preparedness and cohesion. Military in Sri Lanka too experience a very favourable environment for risk behaviours.
Objectives: To assess knowledge and attitudes on HIV/AIDS and sexual behaviour among male soldiers in Sri Lanka Army.
Methods: A cross sectional descriptive study was conducted among 600 soldiers selected using multi stage random sampling and a self administered questionnaire was used to collect information during the month of August 2007. Level of knowledge was decided using inter quartile range (25^{th} centile was considered as poor knowledge) and attitudes were assessed on a likert scale.
Results: Response rate was 99.5% (n = 597). A majority (88.6%; n = 447) had completed education up to GCE O/L. Level of knowledge on HIV/AIDS was satisfactory in 57.1% (n = 341). A statistically significant association of satisfactory knowledge on HIV/AIDS was observed with an educational level of ≥GCE A/L ($p < 0.001$) and period of military service of >10 years ($p = 0.03$) but not with area of service and age.
A majority of soldiers (55.7%; n = 333) had desirable attitudes towards HIV/AIDS which was significantly associatedwith an educational level of ≥GCE A/L ($p < 0.001$) but not with age, period of military service or area of service.
Ninety percent (n = 498) of them were currently sexually active. Nearly half (47.7%; n = 259) had their sex debut before 20 years of age A majority (80.2%; n = 481) have had premarital sex and 45% (n = 238) of currently sexually active soldiers reported having non regular sexual partners during last 12 months. Prevalence of consistent condom use with a non regular partner was only 21.8% (n = 52) where as only 37% (n = 88) had used a condom at the most recent sexual contact with a non regular partner. Reasons for not using were mainly perception of invulnerability (62.7%; n = 94) and non availability (24%; n = 36). Lifetime homosexual experience was 19.6% (n = 99). About one third (32.1%; n = 190) had not had any educational exposure on HIV/AIDS.
Discussion: Marginal level of knowledge (57.1%) and risky sexual behaviour existing among soldiers would create a conducive environment to trigger a possible epidemic endangering the general population too.

P118

Female condom in HIV/AIDS prention among female commercial sex workers in Nigeria: a geographical perspective

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Background: One of the primary concerns of the Millennium Development Goals is to combat and reverse the spread of the HIV/AIDS. An estimated three million people in Nigeria live with AIDS, and current research suggests that unprotected sex accounts for about 90 per cent of the infections among adolescent girls, economically disadvantaged women and commercial sex workers who constitute the high risk group. The paper considers more effective and equitable ways to contain the spread of the virus, especially among the socially excluded group of female commercial sex workers in Nigeria.
Methods: The paper is based on a survey of about 1,500 brothel based commercial sex workers who responded to a questionnaire designed to elicit information on the extent of infection among them, and on current attitudes and measures to control the spread of the pandemic in four major geographical zones of Nigeria selected for the study. Government officials and health representatives were also interviewed on how existing laws protect the rights and welfare of CSWs, and the programmes of government and its development partners to promote the use of female condoms and on other measures to control the AIDS pandemic. Secondary sources were consulted for theoretical and comparative insights on the spatial and behavioral aspects of disease and health, and on the merits and limitations of condom use in Southern Africa and elsewhere. Maps and charts are used where necessary to illustrate spatial variations.
Results: The study confirms that female commercial sex workers suffer discrimination and neglect, and have limited access to information on the new devises and other resources available to reduce and treat HIV infections. It argues that since women appear to be in a subordinate position in sexual relations with men, the female condom, over which the women have greater control, is for them the only safe-sex method available, and should therefore constitute an essential component of any strategy for contraception, microbicide and AIDS prevention. I calls for a well designed and properly targeted government intervention that would subsidize the high cost of female condoms, and promote its accessibility and use among commercial sex workers.
Discussion: The paper concludes with some general reflections on the implications of the study for public policy, and on how the geographer can contribute to a better understanding of the spatial incidence and spread of disease, and the optimal and equitable location of health programmes and interventions.

P119

Human immunodeficiency virus viral markers seroperevalence in first-time healthy blood donors refered to transfusion centers of bushehr province, South of Iran (April 2004 to March 2008)

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Background: AIDS is one of blood-transmitted diseases so that WHO recommends carrying out the HIV screening test on all donated blood samples. In this study we analyzed seroprevalence, demographic and epidemiologic characteristics of positive HIV cases in first time blood donors of Bushehr province.
Methods: In this cross-sectional study, 66873 first time donors who were referred for blood donation, during 5 years (April2004 to March2008), according to the records registered in Bushehr province's blood transfusion organization were studied. Donors had been passed the initial screening (ELISA) and confirmatory test (Western Blot) for HIV Ags.

Results: A total of 51884 people out of 66873 volunteers were able to blood sampled. 24 of them were HIV positive with respect to initial screening test. The results verified in confirmatory test for 6 donors (Prevalence = 0.011%). All of them were undergraduate men, below diploma, 4 married and 2 singles and all younger than 35 years old except one. It is notable that 5 people out of 24 people who were HIV positive with respect to initial screening test, were self-rejected their bloods out of the cycle secretly and confirmatory test were also positive for these 5 individuals. (Self reject = 83.33%)
Discussion: HIV-contaminated bloods make a few percentage of all donated bloods. These contaminated bloods mostly belong to undergraduate young men whom don't belong to high social levels. At the same time, due to awareness of their high-risk behavior they don't want their blood to be injected to others. According to this fact that most of infected persons were young and undergraduate married men, this segment of society should be more considered.

P120

Correlates of HIV knowledge and sexual risk behaviors among female military personnel

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Background: Uniformed services personnel are at an increased risk of HIV infection. We examined the HIV/AIDS knowledge and sexual risk behaviors among female military personnel to determine the correlates of HIV risk behaviors in this population.
Methods: The study used a cross-sectional design to examine HIV/AIDS knowledge and sexual risk behaviors in a sample of 346 females drawn from two military cantonments in Southwestern Nigeria. Data was collected between 2006 and 2007. Using bivariate analysis and multivariate logistic regression, HIV/AIDS knowledge and sexual behaviors were described in relation to socio-demographic characteristics of the participants.
Results: Multivariate logistic regression analysis revealed that level of education and knowing someone with HIV/AIDS were significant [$p < 0.05$] predictors of HIV knowledge in this sample. Condom use self-efficacy was significantly [$p < 0.05$] predicted by annual income and ethnicity. Condom use attitudes were also significantly [$p < 0.05$] associated with number of children, annual income, and number of sexual partners.
Discussion: Data indicates the importance of incorporating these predictor variables into intervention designs.

P121

Ethnological inquiry into sexual behaviours at risk to homosexuals in Portugal

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Background: In Portugal, since 1982 homosexuality is not any more a crime. However, the population remains profoundly homophobe. Marriage isn't allowed between persons of the same sex and crimes or attacks against homosexuals, transsexual or transgender punctuate actuality. The condition of oppressed minority predominates in their group identity but also in their own identity, in their self-definition. As a result, these practices are considered twice more transgressive. The socially transgressive context in which take place these sexual exchanges sets the individuals in a territory considered "out of limits".
Methods: As part of a research on sexual behaviours in the Portuguese population led by the Institute of Social sciences of the University of Lisbon, I realized ten interviews with both homosexual and bisexual men, from 20 to 65 years, in the areas of Lisbon and Porto.
Results: The most part of the interviewed is having sexual relations with occasional partners, notably in meeting places and saunas. The questioned persons define these relations as sexual exchanges dominated by the only and simple quest of pleasure, anonymity and freedom. These moments, governed by transgression, are lived as parentheses in daily life. In this special context, knowing that prevention takes more the path of duty (having to protect oneself and having to protect partners) than free choice, desire and pleasure, men are breaking occasionally the safer sex rules.
Discussion: Without forgetting that the difficulty in maintaining long-term prevention sexual behaviours, the increase of the longevity of the hiv positive and regarding trivialization of illness led by HAART, all aspects have to be considered to explain the relapse (the slackening of precautionary behaviours), the question of transgression of the actors and their practices must be added to the dynamics of risky behaviours.

P122

Stigma and HIV risk behaviors of transgender women in Nepal: implications for HIV prevention

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Background: The growing HIV epidemic among Metis (i.e. transgender women) in Nepal has important implications for this at-risk population and the country overall. In order to develop interventions targeting this population, researchers must better understand the unique cultural context within which risk behavior occurs. This study was conducted to explore the social context of HIV risk behavior among Metis in Kathmandu, Nepal.
Methods: Qualitative data were collected using in-depth interviews with fourteen Metis. These data were taken from a larger study with a purposeful convenience sample of men who have sex with men in Kathmandu, Nepal. Seven Metis reported currently being sex workers, while seven reported not currently engaging in sex work but having a history of providing sex for money.
Results: Utilizing a phenomenological approach, we found that stigma towards Metis resulting in discrimination by family members, law enforcement, and employers had an important effect on HIV risk for Metis. Specific HIV-related risks identified were rape and abuse by law enforcement officers leading to inconsistent condom use due to fear of carrying condoms in public. Low access and ability to carry condoms paired with high reported numbers of sexual partners revealed an environment ripe for the spread of HIV among Metis and their partners.
Discussion: These data suggest the imminent need for interventions to reduce law enforcement violence towards Metis in order to decrease the risk for HIV among Metis who are raped and to increase access to and the ability to safely carry condoms. Sensitivity trainings and strict enforcement of existing legal protections for Metis should be considered. Interventions targeting risk reduction among Meti sex workers are also needed, in addition to programs that focus on providing alternative forms of employment for Metis that face discrimination in the workplace and from families. Further research to inform anti-stigma campaigns and foster a better understanding by family members of Meti identity may also have an important impact on reducing discrimination, subsequently having an impact on HIV risk behavior within this at-risk population.

P123

Seroprevalence of Hepatitis B - surface antigen among non-professional blood donors in selected hospital in Dhaka city

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Background: The purpose of this study was of collect information about prevalence and socio-demographic features of hepatitis B virus. Also to see the prevalence of HBsAg in relation to age, sex, marital status, educational status, income and occupation, possible route of transmission, knowledge about hepatitis B virus infection and with frequency of blood donation.
Methods: A cross sectional study was carried out on two hundreds and seven non-professional blood donors who attended at the department of transfusion medicine, in Dhaka medical hospital, with predonation consent, blood samples were collected in specially covered disposable test tube. The collected samples were carried in a vaccine carrier up to pathology, Dhaka. There blood samples were tested for HBs Ag by ELISA test.
Results: Majority of the respondents were in the age group 20 to 29 years age group Majority of the donors were male which is 91.8 percent and remaining 8.2 percent were female. 97.1 percent of the total donors were literate and 2.9 percent were illiterate. Regarding occupation highest number occupied by others were 54.1 percent followed by students 16.9 percent, lower class employee 11.6 percent and unemployed were 11.6 percent. Among total 207 samples 16 HBsAg cases 4 had the history of jaundice and 12 did not have any history of jaundice. Fifty three point one percent do not have knowledge about hepatitis B virus infection. Among 207 respondents only 12 (5.8 percent) took vaccine against hepatitis B virus and 94.2 percent were not vaccinated 44.9 percent had the history of salon shaving 1.9 percent had the history of high-risk sexual practice. 9.2 percent had the history of travel abroad. Among total of 207 donors, 58 percent donated blood for the first time and 42 percent donated blood for more than one time. 7.7 percent of the non-professional; blood donors found HBsAg positive.
Discussion: In the respect of public health standpoint these findings are alarming. Wide spread transmission of this disease is a great hazard to the mass population. So, collection of blood from low risk donors by screening test and wide spread vaccination is of paramount importance.

P124

HIV prevalence among TB patients attending DOTS centres in rural Haryana, India

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Retrovirology 2010, **7(Suppl 1):**P124

Background: This study was done to estimate the prevalence of HIV infection among TB patients; and the presence of HIV related risk behavior among these TB patients attending DOTS centres in Ballabgarh, Haryana, India.
Methods: The study was carried out in two DOTS centres of Ballabgarh Tehsil of district Faridabad, Haryana. It was a health facility based cross sectional survey. Study subjects were all TB patients (Pulmonary and Extra Pulmonary) who were registered for DOTS. Data was collected from January 2007 to June 2008. We enrolled 413 patients. Eligible subjects were informed about the study objectives and written consent was obtained. Interview schedule was used for data collection. All patients were offered free Liver Function Test. Unlinked anonymous testing on aliquot of LFT blood samples was performed after removing all identifiers. HIV testing was done using three E/R/S.
Results: Four hundred and thirteen TB patients were interviewed and blood samples could be obtained from 368 patients (89.1%). Among them four samples got contaminated and labels of ten samples was lost during the transportation. Finally off the 354 samples tested two were found HIV sero reactive. Prevalence of HIV among TB was 0.56% (95% CI 0.068-2.02).
Of the 315 (76.3%) who ever had sexual intercourse, 289 (91.7%) reported sex with spouse/regular partner, 20 (6.4%) with non regular partner, five with female sex worker and one had with multiple partner. Three (11.5%) subjects used condoms regularly, eight (30.8%) used occasionally and 15 (57.7%) never used condom (n = 26). Among all married patients (n = 293), 13 (4%) had sex outside marriage. Six patients had a paid sex (n = 13). One patient each reported of having anogenital ulcer and intravenous drug usage in last one year.
Discussion: HIV prevalence was low and similar to the rate observed among pregnant women (0.13%) who could be considered as proxy to general low risk population. HIV risk behavior among TB patients was low and comparable to general population rate observed in a state wide behaviour survey. Low risk behaviour contributed to low HIV prevalence among the study population.

P125

Factor associated with getting HIV tested for infant whose born from HIV infected mothers, Thailand: 2008

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Background: Thai National PMTCT program was started since 2001 to provide antiviral drug for pregnant women to cutting HIV transmission from mother in new born children. 2004 PMTCT policy was revised again to launch a new regimen by adding Nevirapine, and High Active Anti Retroviral Therapy for one whose CD4 count less than 200 cell/microlitre. In late 2006 tail end regimen was added to prevent Nevirapine resistant. Antibody and antigen test were done to confirm HIV infection status of children. This study was designed to find out the factor associated with getting HIV tested of children who was born from infected mother.
Methods: This cross-sectional survey was conducted from March to July 2008. Study population was children whose born from HIV infected mother during October 2006 to December 2007 in 12 provinces with the highest number of HIV- positive delivered from 11 Public Health Regions of Thailand. The total number of sample was 187 to conduct indepth interview.
Results: Mothers of new born child had ANC performance as 95%. From that statistics 96% received counseling about HIV testing of their child, 97% wanted know their child status, 83% willing to bring their child for testing, 56% knew their child result and 94% were recommended to bring their child for testing at the age of 2, 4, 6, 12, 18 month. Totally of new born children have HIV Testing is 68% (85% of by PCR and 15% by ELISA) only 54% of them can diagnose their status. The reason of did not receive testing because of did not met criteria for testing(47%), miss an appointment, no information available, sicked or died before testing (2%), lack of money for commutation, did not want to get testing, could not draw blood, loss to follow up(22%), children did not live with their parents. The important things were their parents did not getting to continuous care services and did not want to disclosure their status.
Discussion: Although mothers reported high pre-post test counseling uptake(96%), high HIV & PMTCT knowledge scores(8.6/10 points), high uptake of ARV for PMTCT(94%) and access to HIV care services. But some of them need more HIV information and confidentiality protection concerns. In case of lower coverage of early infant HIV testing the potential setting should be improved especially surveillance system for these children and Laboratory unit should be covered overall health care unit to support for PCR or ELISA testing. Not only that Information, Education and Communication are important as well to bring the children come to get testing and prevent of miss or delay testing. Finally to increase coverage of HIV testing in new born their mothers should have a plan to follow up and mobile testing should be prepared for this situation.

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Condom use and sexual partnerships among truck drivers in Southern Brazil

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Retrovirology 2010, **7(Suppl 1):**P126

Background: The truck drivers are a social group that is characterized by two things: be predominantly male and their itinerant character. These

features provide a particular context to the experiences of gender and sexuality. This paper aims to demonstrate condom use and its relationship to the sexual partners of truck drivers.
Methods: The data analyzed are the result of a study conducted in five cities that concentrate a large number of truck drivers in southern Brazil. In the quantitative phase, 854 were interviewed truck drivers and qualitative phase were conducted semi-structured interviews with truck drivers and participant observation at gas stations.
Results: The truck mention the use of condoms, however there is a continued use, it depends on the partnership and the type of relationship. Thus, 68.8% of respondents claim to use condoms, always (36.5%) or sometimes (32.3%), but only 24.9% of them reported condom use at last intercourse. Those who used condoms in their relationship, did so with partners classified as sex worker (98.2%) or possible (84.4%). Condom use falls significantly with a primary partner (wife or girlfriend) to 14%. Everyone recognizes that it is common to use the services of prostitutes in night parades. Approximately 57.4% of the subjects reported having used such services. Only 46.2% had condoms in the truck during the interview.
Discussion: Truck drivers are a very vulnerable group of STD/AIDS referred for engaging in risk associated with known infection, such as multiple sexual partners not associated with condom use and high use of alcohol and stimulants. While the high use of condoms, it occurs irregularly, depending on the type of partnership considered. Preventive actions should consider the character predominantly male and this itinerant population, with specific actions to stimulate prevention.

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Abstract withdrawn

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HIV infection and the immigration in Italy. Consequences on inpatient hospitalizations and Day-Hospital admissions at a a metropolitan hospital, during the last nine years

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Retrovirology 2010, **7(Suppl 1):**P128

Background: Immigration is a recent phenomenon in Italy, mainly caused by the sudden and unexpected arrival of wawes of foreign citizens, refugees, and individuals escaping from war. This phenomenon is of great concern, due to its serious social-economic and health care impact.
Methods: A prospective survey of all charts of patients (p) hospitalized or followed on day-hospital (DH) basis at our Infectious Disease ward until end-2008, allowed us to assess the frequency of admission of immigrants from extra-Western Europe (eWE), and to analyze multiple variables related to multiple epidemiological and clinical features.
Results: The rate of p immigrated from eWE showed a significant increase among our inpatients, and at a lesser extent and later for DH admissions: 7.7% and 3.1% during the year 2000, 10.1% and 4.6% in 2001, 13.2% and 6.2% in 2002, 17.9% and 7.9% in 2003, 21.3% and 8.9% in 2004, 17.7% and 10,8% in 2005, 17.9% and 11.3% in the year 2006, 17.3% and 10.9% in the year 2007, up to 17,7% and 11.4% in the year 2008 ($p < .0001$ for inpatients; $p < .001$ for DH p). Over 60% of p came from Africa, followed by Eastern Europe, Asia, and Central-Southern America. When comparing the admission features of WE citizens with those of p coming from abroad, no differences were found as to duration and intensity of assistance, with HIV disease prevailing among regular admissions (33.6%), and DH access (30.2%), followed by acute-chronic hepatitis, pulmonary or other-site tuberculosis, central nervous system and respiratory tract infection, and sexually-transmitted diseases. HIV-infected immigrants were frequently (>60% of cases) "AIDS presenters", and less than 5% of them were already on an antiretroviral therapy upon admission. While the frequency of HIV-associated admissions did not show differences in the considered 9-year period, p from eWE had an increasing frequency of tuberculosis, skin-soft tissue infection, infectious exanthems, gastroenteric-parasitic diseases, and malaria ($p < .05$ to <.0001).
Discussion: A continued monitoring of this phenomenon is strongly warranted, in order to improve a sustainable social-cultural network, to plan health resource allocation for the next future, and to define adequate and well-targeted prevention and public health measures.

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The implementation of EPP software using HIV prevalence data from studies conducted in Greece

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Background: In Greece, HIV/AIDS surveillance is based mainly on the case reporting system, which, however, is affected by patterns of testing and reporting. Specific information on the prevalence and incidence of HIV disease is required for an effective and well-planned response to the epidemic. The Estimation and Projection Package (EPP) is a software developed by UNAIDS and partners, which assists countries to map their HIV epidemic on the basis of available HIV prevalence data. The aims of the present study include: i) the systematic review of Greek and international literature in order to retrieve relevant HIV prevalence studies carried out in Greece, and ii) the use of the extracted prevalence estimates in EPP to produce the Greek epidemic curve.
Methods: HIV prevalence data were extracted from studies conducted in Greece among high risk behavior groups [Injecting Drug Users (IDUs) and Men who have Sex with Men (MSM)] and the general population. The EPP software was employed to generate an epidemic curve.
Results: According to EPP data, since the beginning of the epidemic the HIV prevalence was steadily increasing and reached 0.13% in 2009. Concerning IDUs, a secular decrease was observed and the current prevalence estimate approximates 0.4%. After a sharp increase during the 90 s, the HIV prevalence among MSM has been stabilized to the considerably high number of 6.5%.
Discussion: EPP software incorporating data from HIV prevalence studies, indicated that HIV/AIDS epidemic in Greece is concentrated on MSM, with an estimated prevalence above the 5% threshold used by WHO for concentrated epidemics. The results are consistent with estimates derived from the National HIV/AIDS reporting system and highlight the need for intensified interventions in the sub-population of MSM.

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HIV/AIDS related knowledge and attitude among female sex workers in Tehran/Iran

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Background: To assess the knowledge and attitude of female sex workers regarding HIV/AIDS and utilization of health services prepared for them in Tehran capital city of Iran.
Methods: This study includes two phases. Phase one: A cross-sectional survey was conducted among 280 female sex workers in various urban areas in Tehran. During the survey a 114 item researcher designed questionnaire was used. The validity and reliability of questionnaire was assessed during a pilot study. Phase two: Following the quantitative survey a qualitative study was done which 39 in-depth interviews were conducted to gain insights into the context of their risky sexual behaviors and suggestions for feasible HIV primary prevention interventions.

Results: The mean age of the respondents was 27.6 years and majority of them were literate. Among study population 43.1% were born in another province besides Tehran. The mean years of sex work was reported as 5.8 years by participants in this study. About 97.5% of them reported that they have heard about HIV/AIDS and 98.5% reported that they have heard about condom. Among respondents 70% believed that there is no feasibility for confidential HIV test in Tehran. The findings revealed that 30.3% of participants in this study were forced to take an HIV/AIDS infectious test exam against their personal will.
Discussion: Although majority of female sex workers in Tehran were familiar with HIV/AIDS and condom but their knowledge was superficial. Most of study group had serious concerns about referring to health clinics for HIV/AIDS tests and they had negative attitudes in this regard. It seems that peer group education, providing secure stations for HIV/AIDS testing and providing easy access to condom would be useful to address this problem.

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Problem of nosocomial infection with Hepatitis B, C viruses and HIV in Russian dental institutes: review
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Background: The problem of nosocomial infection with hepatitis B, C viruses and HIV is important for the Russian Dental Health Service. It is connected with the unfavourable epidemic situation in respect of these viral infections, limited availability of dental safe technologies (DST), means of postcontact prevention etc. in public dental practice.
The purpose of the review is the problem reflection of nosocomial HIV & Viral hepatitis infection for dental service in scientific domestic publications.
Methods: The review of the scientific literature on this problem for last 5 years.
Results: According to Platoshina O. et al (2004), among patients of the health care institutions in Saint-Petersburg (S.-Pb.) carriers of HBV and HCV are 3-7 times more frequent, than in the general population. Their longitudinal three-year survey detected 56.6% a prevalence of hepatitis B virus antigen (HBsAg) among the dental staff in S.-Pb. Interrogation of S.-Pb. medical persons has shown that 24% do not use the safety equipment (SE). In other survey the surgeons-dentists at municipal clinics in S.-Pb. reported regularly changing gloves in only 36.7% of cases, and 63.3% of applied antiseptic to the gloves without changing them after the treatment. This is perhaps accounted for by the insufficient provision within municipal medical institutions of SE.
A high prevalence of HIV-infection has been observed in Moscow during the latest decade. Based on those general population statistics, the Probability of Infection (PI) of a dentist with HIV in Moscow would be 0.000013, while in providing dental aid to Injection Drug Users, the PI wouldbe 0.00078, which is **60** times higher. Retrospective epidemiological analysis in Moscow shows that the PI of the dentist for HIV has increased during 10 years (1995-2005) as much as **65** times [Goliusov A., 2008].
Discussion: Considering current conditions concerning viral hepatitis and HIV-infection in Russia, it is important to develop in dental personnel a better understanding of the potential risks, mechanisms of infection with blood-borne viruses and to draw up protocols with precise information about various occupational exposures. This should be enhanced by regular training on the use of DST for all members of the dental team.

P134
Feto-maternal outcomes in HIV positive pregnant women: Pakistan PPTCT experience
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Background: To determine the feto-maternal outcomes in sero-positive pregnant women.
Methods: Study population: All HIV positive pregnant women referred to seven PPTCT (Prevention of Parent-to-Child Transmission) Centers of Pakistan.
Study time: March 2007-Oct 2009.
Study design: Descriptive study.
During the study period, thirty seven HIV positive pregnant women were referred to PPTCT Centers for antenatal care and delivery. All women were given ARV prophylaxis (CD4>350 mm) or HAART (CD4<350 mm) and AFASS criteria was used to select a safer infant feeding option. Outcomes in terms of pregnancy complications, mode of delivery and peri-natal transmission of HIV are observed.
Results: Among thirty seven women registered with PPTCT centres, thirty delivered at term. Mean gestational age at time of delivery was 39 weeks. Two women had miscarriages at nineteen and twenty one week. Five women have ongoing pregnancies. The preferred mode of delivery at hospital was caesarean section. Through AFASS, twenty eight women opted for formula milk for their babies while two opted exclusive breast feeding. Twenty three babies tested, so far, with HIV PCR viral load were negative, showing effectiveness of PPTCT interventions.
Conclusion: Though Pakistan has limited PPTCT experience, availability of ARVs and AFASS criteria has proved quite effective, giving hope to people living with HIV (PLHIV) to wish for and have HIV negative babies.

P135
An analysis of non response in HIV testing in India: evidences from National Family Health Survey-3
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Retrovirology 2010, **7(Suppl 1):**P135

Background: This paper aims to analyze biases in the community based estimate of HIV prevalence in India due to non-response.
Methods: It uses NFHS-3 data, where community based HIV prevalence has been derived by testing 103,000 adults as part of national level household survey.
Results: Analysis of sexual behavior of the respondents in terms of number of sexual partners in the last 12 months, condom use with the last sexual partner and pattern of alcohol consumption portrays relatively lower non response among those reported to indulge in risky sexual behavior. Women and men living in urban areas, completed atleast 10 years of schooling and coming from wealthier households are significantly more likely than their respective counterparts to refuse for HIV testing after participating in the interview. The odds of refusals are 0.5 times less likely among rural women and men ($p < 0.001$). Adult men reporting two or more sexual partners in the last 12 months and women reporting not using condom in their last sexual encounter are significantly less likely to refuse HIV testing. HIV prevalence is predicted among those who *were not* tested based on a model of HIV for those who *were* tested. Separate predictions are made for the "interviewed, not-tested" and "eligible, but not interviewed, not-tested" groups. *Adjusted* HIV prevalence is calculated as a weighted average of the observed prevalence in the tested group and predicted prevalence in the two non-tested groups. The missed population analysis uses reasonable assumptions about HIV prevalence in the non-household population. Given the low HIV prevalence in the general population, it is highly unlikely that the prevalence in the missed population would exceed 10%. Even if we assumed that the prevalence rate in the missed population was as high as 10%, HIV prevalence in the total population would increase from 0.3% to only 0.4%.

Discussion: On the basis of the adjusted values of these predictors, the hypothesis of relationship between risky sexual behaviour and non response in HIV testing is rejected with 95% confidence interval and hence the non response to HIV testing in NFHS-3 is not likely to under estimate the HIV prevalence in India.

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The evaluation of peer education workshops efficacy on knowledge of non medical students about HIV/AIDS, Bushehr, South of Iran

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Background: Some researches have shown that lack of information in high-risk communities, especially among young people about ways of transmission and protection of HIV/AIDS increase risk of disease. Different strategies regarding the risk of AIDS awareness have been used up to now. The present study is done to evaluate peer education workshops efficacy on knowledge of non medical student (of Khalij-e-fars university and Bushehr payam-e-nour university) about HIV/AIDS.
Methods: In this interventional study, rate of awareness of 144 students participated in peer education HIV/AIDS workshop in 2009 as a pre-test and post test questions in the form of a questionnaire 7 is assessed. Data are analyzed using SPSS version 13.0 and student T Test. $P < 0.05$ indicated a statistically significant difference between groups.
Results: 144 non-medical students from two Persian Gulf University (63) and Bushehr payame noor University (81) participate in this study. There is a significant difference between rate of correct responses of all students in pre test and post test questions, test 1 (P value < 0.001), test 4 (P value = 0.001), test 5 (P value < 0.001) and test 6 (P value = 0.004). The mean of pre test score was 4.73 with standard deviation 1.10 and the mean of post test score was 5.31 with standard deviation 0.94. This increase in terms of knowledge is statistically significant (P value < 0.001).
Discussion: In this study students awareness about HIV/AIDS after workshops teached by the peer education group had a significant increase and regards to effectiveness of this method, students' cooperation and financial benefits of such trainings using this method can have a large share in student education.

P137

Determinants of the willingness-to-pay for HIV/AIDS prevention: the case of mother-to-child transmission in selected hospitals in Ghana

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Background: Mother-to-child transmission (MTCT) is the primary route of HIV infection in children under 15 years of age. In Ghana PMTCT programme was piloted in two hospitals in Atua government hospital and St Martin de Porres Hospital in 2002 and later expanded to other health centres. In Ghana Prevention of Mother to Child Transmission (PMTCT) services are free. It is however worth noting that, with the ever increasing public health expenditure in recent years, the government cannot foot the bill on PMTCT all alone. There is therefore the need for individual contributions to ensure sustainability of HIV/AIDS and other health care interventions, hence the study to determine how much individuals will be willing to pay to prevent MTCT of HIV/AIDS.
Methods: The study employs the contingent valuation method to investigate the willingness-to-pay for the PMTCT of HIV/AIDS, as well as the determinants of pregnant women's willingness-to-pay for PMTCT of HIV/AIDS in three antenatal care centres in Ghana (Atua Government Hospital, St. Martins Deporres Hospital-Agormanya and the Central Regional Hospital-Cape Coast) using logit and OLS regression.
Results: About 91 percent of the respondents were willing to pay some amount of money for PMTCT of AIDS. The average willingness-to-pay by all the respondents was $4 (GH¢4.20). Results from both the logit and the OLS regressions indicate that income is the most significant factor that affects the willingness-to-pay for PMTCT of HIV/AIDS. Other factors such as HIV/AIDS status, distance to antenatal clinic, age and marital status were also shown to have effect on the WTP for HIV/AIDS, though the results were not robust.
Discussion: The study indicate that pregnant women place high value on the PMTCT of HIV/AIDS programme in Ghana, hence their willingness to pay some amount of money for PMTCT.

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Knowledge and attitudes of students in an offshore Caribbean medical school towards HIV/AIDS

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Background: Human Immunodeficiency Virus/Acquired Immunodeficiency Syndrome (HIV/AIDS) is seen is increasingly become one of the most pressing public health dilemma in the Caribbean.
This study is aimed at assessing the level of knowledge, attitudes and misconceptions of the medical students in an Offshore Caribbean Medical School towards HIV/AIDS.
Methods: Semi-structured questionnaires were used to collate information. Verbal Voluntary consents were obtained fromall the respondents.
Results: Out of 150 questionnaires distributed, 130 were filled, showing a response rate of 87%. The ages of the respondents ranged from 19-45 years with mean age of 26. Our finding revealed that the knowledge of HIV/AIDS is high: 97.7%. HIV/AIDS associated beliefs: 22.3% sees the disease as a punishment for unfaithfulness or immorality. A negative attitude towards HIV patients was seen in 3.8% of the respondents.
Discussion: Risky behaviours were identified but the prevalence of such behaviours is low. Religion was identified to play an important role in the conception and misconception about condom usage and possibly transmission of the virus. Behaviour modification course is recommended to be incorporated in the school curriculum to correct misconceptions among medical students.

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Creating a supportive environment is very important to reduce STI, HIV/AIDS and infectious disease

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Background: The aim is to create a supportive environment for HIV, AIDS and STI prevention program among hotel based sex worker in Chittagong, Bangladesh through social mobilization.
Methods: With a view to achieve the aim we form a Project Facilitation Team (PFT). The member was selected as representatives from hotel owners, Chittagong City Corporation, the Department of Police, Department of Health, Social Workers, Media Professionals, Law enforcement agencies, Department of Women Affairs, Muslim religious leader and members from other NGO working in the city.
Results: The Project Facilitation Team (PFT) member has been seating quarterly basis to review program progress. The PFT members advise and cooperate for reducing barriers and mobilizing social support for project activities take place. Especially they have been Facilitating regular contact with key stakeholders, including sex workers, member of law enforcement agencies, hotel owners and workers. As a result police is not harassing our Peer Educator and they are allowing to conduct the Behavior Change Communication (BCC) session at hotel and the hotel owner and hotel management are giving the free access to work with the sex worker in their hotel i.e. conduction of BCC session, distribution of condom and lubricant.
Discussion: Formation and functioning of Project Facilitation Team is very effective to create a supportive environment for reducing the STI, HIV/AIDS and other infectious dissease because the members of the team

can support the project's implementation with their local knowledge and influence. So, it can be a best model for other country.

P140

Civic educating rural pregnant mothers to take full nevaripine dose a tool to reducing HIV prevalence in Malawi

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Background: It is estimated that 600,000 women get pregnant every year in Malawi. Primary health care plays a vital role. Prevention to Child Transmission of HIV education is important in a country like Malawi where prevalence of HIV positive mothers exceeds 20% than normal country prevalence of 12%. In the ultra-partum period Nevaripine has proved to be an effective drug in reducing transmission from mother to Child. Accessibility of essential medicine like nevaripine drug to women and neonates is especially a problem to rural areas where many women deliver at home due to difficulties in accessing health facilities.
Objectives: This civic education intervention aimed to increase rural HIV+ pregnant mothers uptake of Nevarapine tablets and syrup for infants' dose for Prevention of mother to child transmission of HIV and to promote hospital delivery.
Methods: Global Hope Mobilization is providing education and counselling to women in rural setting to access basic medicines such as nevaripine, as well as to promote hospital delivery in Lilongwe district rural setting. Households with pregnant women were included through a random sample and a total of 700 pregnant women were included in a programme to provide civic education materials, including leaflets; local meetings, counselling sessions, and home visits. Nevaripine tablets and syrup were also administered, ANC data were collected and used from the nearest health facility. The intervention was assessed through one to one interviews, focus group discussion, and questionnaires.
Results: 100% of the pregnant women were sensitized on taking nevaripine, counselled and referred for HIV testing. To date 98% of women have attended pre and post VCT counselling testing and ANC at health facilities and at private clinics referred to. Of these women 66% were found to be HIV positive. Nevaripine tablets and instructions were issued to them when they reached 32 to 34 weeks of gestation. 60% Infants received Nevaripine syrup within 3 days after birth. Of these 6% are not known to have taken syrup and were lost to follow up, while 60% returned for post counselling. The major barriers of the intervention were home delivery, traditional beliefs and lack of knowledge of HIV status and on the course of nevaripine.
Discussion: The main risk factor for lack of full take up of Nevaripine for HIV+ women is home delivery and lack of knowledge on their HIV status. A civic education on Nevaripine and hospital delivery among HIV positive pregnant women in this rural population setting can reduce HIV transmission from mother to child.

P141

A study of HIV/AIDS related knowledge and attitudes amongst the engineering college students

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Background: The present study designs to find out the knowledge and attitudes of the students of engineering colleges. The information obtained in this study will be used to demonstrate the need for development and integration of an HIV IEC training module.
Methods: One hundred seventy four randomly selected students studying in the various engineering colleges, studying in Uttar Pradesh were surveyed to assess their knowledge on HIV/AIDS. Pre tested, pre designed and preformed questionnaire was used to collect data.
Results: Response rate of 87% was obtained (174 out of 200). Overall, females showed less knowledge pertaining to issues related to human sexuality and HIV transmission, As compared to their male peers. Anal intercourse was observed as a risk for HIV transmission by 3% of females as compared to 20% of males. In general, there were considerable misconceptions regarding the spread and risk of HIV transmission among all engineering students. Attitudes of most of the students toward HIV-infected individuals could be best described as ambivalent. Interesting to note that female students showed more positive attitude towards HIV infected people than their male peers. Findings suggest the need of integrating IEC activities and BCC activities promotion in the community starting from the initial stages mainly concentrating on teenagers and youngsters.
Discussion: IEC materials and media have the potential to facilitate the development of positive behaviors and attitudes among engineering students as they relate to HIV. Studies over the past decade among health professionals in India identify the gaps in their knowledge concerning risks and transmission of HIV.

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Services integration for injection drug users on antiretroviral therapy for management of HIV epidemic in Estonia

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Background: To assess coverage of harm reduction services for HIV-infected patients in Estonia and their integration with HIV care; to identify barriers to integrated services provision.
Methods: We reviewed HIV surveillance documents, administrative data and scientific literature, and interviewed key informants providing treatment and harm reduction services in North-Eastern county of Estonia.
Results: Estonia had the highest incidence of HIV infection in Europe with 472 cases per million and an adult HIV prevalence of 1,3% in 2007, the second-highest in Europe. Majority of new cases occur in the North-Eastern county, with incidence exceeding 4 times that of Estonia as a whole (183 *vs* 47 per 100 000). The Estonian epidemic is driven by injection drug use, and local studies have revealed a 40-90% prevalence of HIV among injection drug users (IDUs). HIV treatment is provided by state-funded public health-care system. At end of 2008 1006 patients were receiving HAART. Harm reduction services are provided by non-profit organizations (usually state funded). There are 7 centers for methadone substitution therapy (649 patients treated at end of 2008); 36 syringe exchange programs; 2 centers for free and anonymous STI services. Yet in different regions only 6-28% of IDUs report currently receiving treatment for drug use and 5-12% receiving HAART. The main challenge and strategic goal of the National HIV/AIDS Prevention Strategy remains coverage of IDUs by treatment and harm reduction services. The major barrier identified was failure to provide well integrated services to those in need. The underlying factors leading to the barrier were substantial misperceptions in regards to the utility of methadone substitution therapy among HIV medical care providers.
Discussion: In the current resource constrained environment improvement and integration of already existing services would ensure sustainability of the national HIV prevention strategy. Barriers to integration need to be delineated and addressed.

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The international physical activity questionnaire overestimates moderate and vigorous physical activity in Human Immunodeficiency Virus compared with accelerometry

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Aim: The study aimed to evaluate the validity of the last 7-day, self administered version of the International Physical Activity Questionnaire long form in an human immunodeficiency virus-infected population, using accelerometry as the objective criterion.

Materials and methods: Thirty male participants (mean age 53.2 years (SD = 10.2)) took part in the study. The ActiGraph GT1M accelerometer was worn during all waking hours for seven days and the questionnaire was completed on day 7. Agreement between measures was assessed using correlations and modified Bland-Altman analysis.
Results: The total number of MET-minutes per week reported on the questionnaire correlated modestly with the main criterion measure of total weekly activity counts measured by the accelerometer (r = 0.41, p = 0.023). However, time spent in both moderate and vigorous physical activity was over reported on the questionnaire. The mean difference compared to accelerometer was 546.63 minutes per week (95% CI 217.1 to 871.2 minutes) for moderate and 295.33 minutes per week (95% CI 88.08 to 502.6 minutes) for vigorous activity. The tool's sensitivity to detect individuals with insufficient physical activity to derive a health benefit was low (9.5%), however specificity was high (100%).
Discussion: We conclude that self-reported physical activity measured by this questionnaire correlates with the objective criterion of accelerometry, but substantial over-reporting occurs. The tool may be useful in screening physical activity levels but should not be used to determine precise amounts of activity.

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The perception of taxi drivers in southern part of Saint Lucia, West Indies towards HIV/AIDS and condom use

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Background: The prevention and control of HIV/AIDS remains a challenge to health care providers in the Caribbean. Despite the threat posed by the rapid spread of the virus, many subgroups of the populations continue to engage in risky sexual behaviours. The taxi drivers belong to an occupation whose lifestyles while on duty have made them to be identified as highly susceptible. This study was conducted with the aim of understanding HIV-related knowledge and sexual behaviour with respect to condom use. The prevention and control of HIV/AIDS remains a challenge to health care providers in the Caribbean. Despite the threat posed by the rapid spread of the virus, many subgroups of the populations continue to engage in risky sexual behaviours. The taxi drivers belong to an occupation whose lifestyles while on duty have made them to be identified as highly susceptible. This study was conducted with the aim of understanding HIV-related knowledge and sexual behaviour with respect to condom use.
Methods: This study was conducted in May 2009 among male taxi drivers in the Hewanorra International Airport, St Lucia. This was a descriptive study using a semi-structured close-ended questionnaire. The data were collated and analysed using the Statistical Package for Social sciences 13.0 (SPSS 13.0) data base. Statistical significance was set at $p < 0.01$.
Results: Eighty questionnaires were distributed, sixty were correctly filled, showing a response rate of 75%. The knowledge of HIV/AIDS was high among the drivers (78.3%). More than 50% of the respondents do not use condom consistently for sexual intercourse.
Discussion: Some misconceptions about the mode of transmission of HIV/AIDS were observed. Also constraints to the affordability and availability were identified. Behaviour modification programmes should be taught periodically to this subgroup. The government should endeavour to make condoms more accessible especially in public places.

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Interventions and strategy to mitigate HIV related risk behavior among young men in India

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Background: Adolescents present both a challenge as well as an opportunity while exploring the linkages between HIV related risk behaviour across different sub populations. Adolescents are an unprecedented large and growing population, especially in developing world like INDIA. With sexual activity often initiated in adolescence -within or outside of marriage - the risk relating to their sexual behaviour is often underestimated.
Therefore, this paper focuses at the extent of comprehensive knowledge among young men age 15-24 and the socio-economic and contextual correlates of HIV related risk behaviors in India, which may have top programmatic priorities for designing evidence based interventions.
Methods: The basic data used in this paper has been taken from the third round of National Family Health Survey (NFHS-3), where it is the first time when information on HIV related risk behavior have been collected from a nationally representative sample of men age 15-54 and women age 15-49 irrespective of their marital status. Bi-variate, multivariate and logistic modeling statistical techniques are used for the analysis.
Results: Only one-third (36 percent) of the young men in India have "comprehensive knowledge" about HIV/AIDS- a pre requisite for effectiveness in prevention and control programme in any country. Overall 6 percent of the young men reported to have an "intention to have sex before marriage" and the study clearly reveals that among the youth who are consuming alcohol and who have frequent mobility are more likely to have an intention for the pre marital sex. Data depict that, though 15 percent of the youth reported to be used condom during their first sexual intercourse, only 7 percent of the never married men reported to use condom shows that the involvement in safe sexual practices among the young men. Overall nearly 4 percent of youth in India are involved in risky sexual behavior.
Discussion: Thus the need to focus on young men and their involvement in the existing HIV/AIDS intervention programs is essential. Intervention programs should focus at enhancing not only the "comprehensive knowledge" but also "condom promotion" and "safer sexual practices". While deciding the research priorities and also designing programmatic response to the epidemic.

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Increased antenatal HIV testing among rural pregnant women: a community-based HIV prevention services program in rural Maharashtra, India

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Background: To characterize changes in antenatal HIV testing utilization, six months after the introduction of a targeted HIV prevention and testing services intervention program.
Methods: Between August 2006 and April 2007, a community-based HIV prevention campaign and antenatal clinic-integrated voluntary HIV testing program was initiated in 52 villages in rural Aurangabad district, Maharashtra, India. Questionnaires assessing HIV risk factors and HIV testing utilization were administered to random cross-sectional community samples of 400 adult women who had given birth in the prior 12 months, at Baseline and, after six months, to 400 women each in the Post-Intervention and a separate Control community.
Results: During the study period, a total of 1200 recently-pregnant women were surveyed. Sociodemographic characteristics including >90% antenatal care utilization were similar for all sample groups. From Baseline to Post-Intervention, HIV testing facility awareness significantly increased (6% to 16%, $p < 0.05$) and independently correlated only with participation in the community-based campaign (AOR 2.1, 95%CI (1.3-3.5)). Antenatal HIV testing utilization increased (3.3% vs 7.5%, $p < 0.05$), and correlated with discussing HIV in antenatal care (AOR 10.2, 95%CI (4.7 - 22.4)) but did not vary with STI symptom history. Of Post-Intervention women reporting antenatal HIV testing (n = 30), 43% sought testing at known voluntary HIV testing facilities, including Intervention clinics and the District-level hospital.
Discussion: The community- and clinic-integrated HIV prevention services program significantly raised women's HIV testing awareness and the number of clinic-integrated facilities for voluntary HIV testing, influencing increased community-level antenatal HIV testing utilization. In all rural

communities, it is necessary for current antenatal services to identify and counsel the high-HIV risk subgroup of women with STI symptom history, and to ensure voluntary informed consent, for all clients.

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Effect of materials on HIV/AIDS by the different segment of population in a selected community area

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Background: On the effect of IEC materials on HIV/AIDS by the different segment of based on mainly popular ongoing IEC campaign i.e. poster, billboard, advertisement on TV and radio, drama etc. People's choice, preference for effective media was sought for future pragmatic endeavour.
Methods: This study is a descriptive cross-sectional. Respondents from different segment of population were used as a sample. Two hundred people selected as sample size from a community of Gazipur district and using a semi-structured questionnaire data was collected on the effect of Information, Education and communication (IEC) materials on HIV/AIDS by the different segment of was collected.
Results: People admit that 69% get the information about AIDS from television, contrast with that from both TV and radio 25% among the respondents 54% has acquaintance about AIDS for five and more years, 41% has more than one year. Significantly, 97% respondents admitted that there is enough importance of IEC materials or mass media to prevent HIV/AIDS. About 63% respondent told about the route of transmission of AIDS causes for illegal sexual relationship, 26% told about sex with several partners. And only 3% indicated that repeated use of injection syringe may cause AIDS. In the field of sigma and discrimination alarmingly 98%expressed that people show negative attitude towards HIV positive people. The respondents suggest (69%) that media can play role by making drama, discussion etc. to aware people. From bi-variate analysis it has been explored that there is strong association with the respondent's education and knowledge of HIV/AIDS ($p < 0.05$). The marital status of the respondent and knowledge on transmission of HIV/AIDS are statistically associated ($p < 0.05$).
Discussion: The study revealed that level of education had positive impact on prevention of HIV/AIDS.

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Assessing risk criteria and HIV prevalence in antenatal clinic clients from 2 urban tertiary hospitals in Pakistan

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Background: Pakistan is in the concentrated phase of HIV epidemic for the last five years. The HIV prevalence among injecting drug users and hijra sex workers has been reported by second generation surveillance to be more than 5%. Considering the risk of HIV transmission to the spouses and children of the high risk groups including migrant workers, the National AIDS Control Programme established six comprehensive prevention of parent to child transmission (PPTCT) sites linked with HIV treatment centres in tertiary hospitals. The objective of this study was to determine the HIV prevalence among antenatal clinic (ANC) attendees at two tertiary care hospitals and correlate risk assessment questionnaire with HIV diagnosis.
Methods: This cross-sectional study was conducted simultaneously at two PPTCT centers; Lady Wallingdon Hospital (LWH)-Lahore and Pakistan Institute of Medical Sciences (PIMS)-Islamabad from September 2008 to August 2009. A sample of 2589 at PIMS and 2983 at LWH were taken using convenience sampling technique. A risk assessment questionnaire was used to screen the at-risk group and then rapid HIV test was done. The positive cases were confirmed using ELISA.
Results: The total number of pregnant women screened through risk assessment questionnaire was 5572 (both sites). All of them consented for biological testing and were provided pre- and post-test counseling. Only three cases at PIMS were reactive on rapid HIV test, yet tested negative on ELISA. None of the cases from LWH was reactive on rapid HIV test.
Discussion: Based on the study findings, it is recommended that a revised strategy should be used for ANC screening in situations of low prevalence general population as in Pakistan. Moreover, the prevention strategies for general population should be strengthened. Referral linkages of PPTCT centers with NGOs working for people living with HIV/AIDS and most at risk population groups is critical for identifying HIV positive pregnant women and spouses of HIV positive men to access PPTCT services.

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Develop IEC Material By PLHA to lead a positive life

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Background: There is a scarcity of IEC materials necessary in disseminating information about HIV/AIDS to the most at risk and hard to reach populations, in particular information regarding stigma and discrimination faced by PLWHA. Furthermore, involvement of PLWHA in designing such IEC is virtually non existent.
Methods: Family Health International (FHI) and Oxygen Research and Development Forum (ORDF) conceptualized a process of Care and Support IEC material development involving local PLWHA consultants. Two groups of four consultants each worked for six months and adapted booklets 1 through 5 developed in Cambodia by FHI/USAID to the Nepali context. The booklets provide information along with sketches about HIV/AIDS, nutritional needs of PLWHA, stigma and discrimination, social and family support, taking care of one's body and health, income generation, OIs and ARVs etc. The users of the booklet are individual outreach educators who work with illiterate vulnerable communities across Nepal.
Results: IEC materials development is a technical process that needs a lot of careful planning and orientation. Those involved in the process should have real knowledge of the issues that are of utmost importance to the targeted users. Care and attention needs to be given to language, social norms, and depiction. Involvement of local PLWHA is of utmost importance when developing IEC and other materials. In addition to creating booklets that are representative of the local needs and situation, exposure to working in an office environment as consultants making steady income and gaining much needed skills in computer, translation, editing and sketching helps develop capacities of the involved consultants significantly.
Discussion: For ensuring the design of effective IEC materials related to HIV/AIDS, it is imperative that local PLWHA be involved in the planning, design, pretest and launch phases. Involvement of community PLWHA will also contribute towards capacity development and skills enhancement.

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Service integration of blood borne viral infections in HIV/AIDS prevention sites

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Background: Outbreaks of acute hepatitis C virus (HCV) in HIV infected men who have sex with men (MSM) were recently reported in Europe, Australia, and New York City (Vogel & Rockstroh, 2009). Acute HCV infection is defined as a newly identified viral HCV antibody with either jaundice, serum alanine amino-transverse (ATL) levels >400 IU/L (CDC, 2007). In addition to known acute hepatitis C cases, an at risk population may be defined as men who have sex with men (MSM), who did not already have chronic hepatitis C and who reported sexual and/or drug-related risk behaviors within the prior 6 months (Taylor, 2009). A

comprehensive strategy is needed to identify and treat populations at risk for blood borne viral infections.
Methods: Although a recent survey of local health officers showed that 87 percent of city and county health departments provide education about HIV/AIDS and 77 percent provide HIV testing, less than 50 percent provide hepatitis C counseling and only 23 percent provide HCV testing (CDC, 2001). Direct service workers have limited experience with combining counseling, testing, prevention immunization and treatment services for these diseases in HIV/AIDS prevention sites, STD clinics, drug treatment sites, and correctional health programs (CDC, 2001).
Results: Integration of services to prevent blood borne viral infections is a fairly new prevention strategy. HIV, HBV, and HCV present unique opportunities to provide service delivery at a single client visit. Treatment may include PEGYLATED INTEFERON and RIBAVIRIN. Persons with HCV-related liver disease should be vaccinated against diseases that may produce further complications or increase their risk of death.
Discussion: Data from several demonstration projects indicate that integration of HCV counseling and testing into existing public health programs [including AIDS Service Organizations, STD clinics, drug treatment sites, and correctional health programs] is feasible and may enhance identification of persons with risk behaviors for other blood borne virus infections, such as HIV and HBV (CDC, 2001).
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HBV and HCV viral markers seroperevalence in first-time healthy blood donors refered to transfusion centers of bushehr province, South of Iran (April 2004 to March 2008)

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Background: The risk of infection by transfusion-transmitted viruses has been reduced remarkably. However, a zero-risk blood supply is still desirable. Hepatitis B (HBV) and Hepatitis C (HCV) viruses are transmitted mainly by parenteral route, following which, a remarkable proportion of infected cases, may progress to chronic hepatitis. In this study we analyzed seroprevalence, demographic and epidemiologic characteristics of positive HBV and HCV cases in first time blood donors of Bushehr province.
Methods: In this cross-sectional study, 66873 first time donors who were referred for blood donation, according to the records registered in Bushehr province's blood transfusion organization during 5 years (April2004 to March2008), were studied. Donors had been passed the initial screening (ELISA) and confirmatory test (Western Blot) for HBs Ag and HCV Ab.
Results: A total of 51884 people out of 66873 volunteers were able to blood sampled. We determine seroprevalence of HBV 0.47% (245 persons), HCV 0.33% (174 persons) and HBV-HCV co-infection 0.013% (7 persons).
In those who were infected by HBV: 94.7% (232 persons) were male and 84.9% (208 persons) were married. The majority of them were young, less than 30 years old, (37.60%) and undergraduate, below diploma, (43.67%) people.
In HCV infected persons: 97.7%(170 persons) were male and 71.84% (125 persons) were married. Furthermore the majority of these persons were undergraduate (62.64%) and young (42.19%) people.
Discussion: We compared our results with those of other studies in near Bushehr provinces and concluded that the prevalence rate of HBV and HCV in our area is less than most of them and now we are in low prevalence state. In attention to Bushehr geographical situation it is an important note. In addition HBV-HCV co-infection is uncommon in our area. According to this fact that most of infected persons were young and undergraduate married men, these segments of society should be more considered.

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Protecting at risk cadres of health workers from medical transmission of HIV and Hepatitis B and C through injection safety interventions

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Background: The Making Medical Injection Safer (MMIS) project funded by the President's Emergency Plan for AIDS Relief implemented by the Ministries of Health and John Snow Inc. aimed to improve injection safety and healthcare waste management practices in eleven countries between 2004 and 2009. MMIS engaged in training and capacity building for healthcare workers, including waste handlers, as well as commodity and logistics support, behavior change communications, and advocacy for safe injection and waste management policies. Waste handlers are considered a highly vulnerable and somewhat ignored group in the infection control chain. Injuries from contaminated needles are a primary route through which blood borne pathogens such as HIV, hepatitis B and C are transmitted in healthcare settings. Percent of reported needlestick injuries, immunization against hepatitis B, and knowledge of disease transmission are key indicators related to occupational safety for waste handlers.
Methods: Cross-sectional studies were carried out to evaluate progress in these areas. Through baseline and follow-up surveys at the intervention sites, observations of waste management and interviews were carried out with waste handlers.
Results: Waste handlers were asked if they are aware of diseases transmitted through needle stick injuries. Surveys across countries showed that majority are aware of HIV, but not Hepatitis B or C. During the 2008 survey in Kenya for example, 91% of waste handlers mentioned HIV, but only 33% mentioned Hepatitis B, and 5% Hepatitis C. Only three waste handlers reported receiving all three doses of the Hepatitis B vaccination in Kenya while in Uganda, only one waste handler interviewed was fully vaccinated at follow-up. The percent of waste handlers who reported having personal protective equipment varied widely between countries, with 55% in Kenya reporting having heavy duty gloves and boots, while only 7% of waste handlers reporting the same in Haiti.
Discussion: As waste handlers are exposed to blood-born pathogens, there is an urgent need to promote hepatitis B immunization programs which will ensure proper inoculation. Protecting waste handlers through training on proper waste management techniques and provision of personal protective equipment to handle medical waste helps reduce needle stick injuries. All of these are vital for preventing transmission of blood-born pathogens.

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Awareness about hepatitis B infection among the grass root level health and family planning workers in a selected health complex at Dhaka district

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Background: Regarding their awareness about Hepatitis B infection.
Methods: This descriptive cross sectional study was carried out amongst 145 health and family planning workers of Tejgaon Thana complex. Mohakhali, Dhaka. The health workers were selected randomly and a structured questionnaire was used as research instrument. A score sheet was prepared to assess the level of awareness.
Results: The mean age of workers was 22.12 years with standard deviation of 4.2 years. Majority of them were H.S.C (39.60%) and S.S.C (42.76%) passed. Nine of them (6.21%) were found graduates and two of them (1.38%) were found post graduates. It was observed that 79.3% were married and remaining 19.31% were unmarried and 1.38% was widow. Majority of them were found Muslims 91.72%). The mean length of service was 3.23 years with standard deviation of 2.25 years. More than four fifth (86.21%) knew infected blood is the source of Hepatitis B

infection followed by contaminated syringe and infected person. Regarding the mode of transmission majority opined infected blood transfusion (86.21%), use of contaminated syringe (67.59%) and sexual contact (59.31%) were the principal way. But only few (16.55%) opined about transplacental transmission. The highest percentage of high risk group was identified as close relatives (78.62%), followed by professional blood donors (71.03%), doctors (67.59%), medical technologist (51.03%), nurses (50.34%), prostitutes (44.83%) and dental surgeons. Majority were unaware (77.24%) about complication of hepatitis B infection. It was found in this study that only 28.28% had tested their blood for HbsAg and only 8.27% took vaccine against. Hepatitis B. regarding knowledge on preventive measures against Hepatitis B, 91.03% opined healthful environment sanitation, 70.34% vaccination, 63.45% use of disposable syringe, 63.45% washing hands after handling infected patient and only 31.72% safe sexual habit. Majority (70.94%) opined the necessity of health education regarding lowering the incidence of hepatitis B infection. Bi-variate analysis revealed that younger age had poor awareness regarding Hepatitis B infection ($p > 0.05$). Similarly higher level of education had good awareness than lower level of education ($p > 0.05$). However as per point score only 20% had shown good awareness and majority were found average (46.9%) and poor awareness (33.60%).
Discussion: Research finding concluded that level of education and good practice can prevent infectious disease.

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Seroprevalence of Hepatitis B-surface antigen among selected group of population

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Background: The purpose of this study was to collect information about prevalence and socio-demographic features of hepatitis B virus carrier. Also to see the prevalence of HBsAg in relation to age, marital status, education, intravenous injection, surgical treatment and drug addiction.
Methods: A analytical cross sectional study was carried out on one hundred seventeen Rick show Puller and Auto Rick show driver in Kuril area Dhaka. Blood samples were collected in specially covered disposable test tube. The collected samples were properly carried out of Pathology, Dhaka. There blood samples were tested for HBsAg by enzyme linked immunosorbent assay (EISA) method.
Results: Majority of the respondents were in the age group 20 to 29 year's age group that is 41 percent of total subject. The mean age of the respondents was 30.9 years with standard deviation 7.7. Out of 117 respondents 66 were Richshow puller and 51 were Auto rickshow driver. Among them 30.8 percent were illiterate. The majority of the respondents were Muslim 96.6 percent and 3.4 percent were non-Muslim out of 117 samples. Nine HBsAg positive cases 4 had the history of jaundice and 5 did not have any history of jaundice. 27.4 percent had the history of dental surgery and 72.6 percent do not have the history of dental surgery. 10.3 percent had history of blood transfusion and 89.7 percent do not have history of blood transfusion. Among the 117 respondents, 50.4 percent had the history of taking intravenous injection, 10.3 percent had history of surgical treatment, 6 percent had history of drug addiction, 7 percent had the history of travel abroad and 31.6 percent had extramarital sexual practice. 7.7 percent of the respondents found HBsAg positive.
Discussion: In respect of public health stand point these findings are alarming, wide spread transmission of this disease is a great hazards to the population.

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Home visiting to an HIV positive patient prior to initiation of HAART has Lasting impact on their adherence. TASO Experience

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Background: Adherence to life saving Anti-retroviral drugs (ARVs) is important to the success of antiretroviral therapy (ART) programs. In order to achieve optimum viral suppression an adherence level of >95% is required.
Readiness of the patient to take medication for life during the initiation process of ART is one of the key criteria used. If this criterion is under looked it has got adverse consequences on maintaining high level of adherence.
The AIDS Support organization (TASO) put emphasis on patients' readiness and presence of family support as one of the criteria for ART initiation.
Methods: The AIDS Support Organization (TASO) is a national non governmental organization in Uganda, whose mission is preventing HIV infection, restoring hope and improving the quality of life of individuals, families and communities infected and affected by HIV and disease.
Patients are usually screened for ART eligibility by CD4 and WHO staging. When the patient is found to be eligible a home visit is arranged for with the patient consent to. During the home visit, which is done by a counselor/field staff, psychological and social preparation is done. This involves seeking for long lasting family support to the patients, identification and addressing barriers to adherence in the home, and identification of a medicine supporter. Also during the home visit adherence plan is made by the patient and this is routinely reviewed during the subsequent counseling sessions.
Results: Retrospective analysis of the data at one of the centers (TASO Masindi). 78% of patients on ART were assessed for readiness for ART through home visit prior to initiation of ART. Out of these 85% had adherence level >95% while 78% of the patients not home visited had adherence level of <95%.
Other findings were that, the proportion of patients on ART disclosing sero-status to family members has increased. Follow-up of Patient's in case of non adherence has been made easy since it is easy to review the adherence plans that have been made.
Discussion: Addressing psychological and social factors during a home visit contributes significantly to good adherence for people living with HIV/AIDS.

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Prevention by care and treatment of HIV-positive pregnant woman in Côte d'Ivoire

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Background: Côte d'Ivoire is a west Africa country. With a HIV prevalence of 8.6% among pregnant women and 661,000 births per year, Cote d'Ivoire has an estimated 55,000 HIV-infected women delivering per year who need PMTCT services. ACONDA's extension of decentralized prevention and care for pregnant women and PLWHA is based on a health district approach.
Methods: Health workers were trained. After, the program strategy consisted in coaching the care providers at the sites in VCT techniques with rapid HIV testing for women with unknown HIV status in ANC, labor-and-delivery rooms and Family Planning unit also.
Drawing up and spreading simple technical procedures helped the care providers in the implementation of PMTCT.
The combined prophylaxis was offered to HIV-infected pregnant women and their newborns systematically, as recommended by national program, and then she got initial biological exams. Those who were eligible received a readjusted treatment. Those who were ineligible continued the current disease prevention. A psychosocial supports for treatment adherence, was provided by counselors and Nutritional advices also. A child's early HIV diagnosis by PCR is made after 6 weeks of postnatal follow up.
Results: From January through November 2008, PMTCT services were integrated into 70 ANC clinics in urban areas and 20 in rural areas, covering 23 districts, with 100 trained health workers. Of 54,876 pregnant

women using antenatal services, 45,730 (83.33%) received HIV counseling and testing; 3100 (6.77%) were HIV-positive; and 3,000 infected pregnant women (96%) received their test results. 78% of HIV-infected women received the mother and child combined prophylaxis against 68% in 2007. Among the HIV-infected women, 520 were eligible for ART according to the WHO criteria.
Discussion: Providing the combined prophylaxis from the disclosure of test results is essential if we noticeably want to reduce the Mother to child HIV Transmission for the scaling up. Without intervention in our country, the rate of transmission is between 30-40 per cent.

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A model to determine effective HIV/AIDS and Multi-Drug Tuberculosis (MDRTB) treatment policies: a case study from the Russian Federation

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Background: The explosive increase in the number of people infected with tuberculosis, multi drug resistant tuberculosis (MDRTB), and injecting drug users (IDU) HIV/AIDS has become a serious public health challenge in Russia. The World Health Organization (WHO) is recommending policies including simultaneous use of highly active antiretroviral therapy (HAART) to treat HIV/AIDS and second line drugs to treat MDRTB. However, it is not clear what would be the impact of implementing these recommendations on tuberculosis and HIV/AIDS mortality. In this context, the aim of this research is to quantify the consequences of adopting these policies in terms of deaths reduction.
Methods: A System Dynamics (SD) computer simulation model was developed to represent the dynamic transmission of tuberculosis, MDRTB, and HIV/AIDS. The model represented explicitly the complex interactions between these diseases and how these link to their transmission and spread in the population. The model simulated scenarios, over a 20 years period, regarding MDRTB cure rate and the fraction of HIV/AIDS patients covered by HAART.
Results: The results over a 20 year period indicate that reduction in tuberculosis and HIV associated tuberculosis deaths would be negligible for HAART coverage up to 50%. The reduction is only significant for HAART coverage of 70% and above. Similarly, high MDRTB cure rate reduces significantly deaths from tuberculosis and MDRTB and this reduction is more important as the HAART coverage is increased.
Discussion: This research demonstrates, through a computer simulation model, that policies recommended by the WHO will not be effective unless HAART coverage is ramped up to include a sizeable fraction of HIV/AIDS patients. This will have to be coupled with an extensive use of second line drugs to address MDRTB. It is only through combination of these policies that tuberculosis and HIV/AIDS mortality could be reduced significantly.

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Sexual behaviours and drug use among the street children

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Background: The aim of the study was to describe the HIV and STD risk behaviours among the street children.
Methods: This is a descriptive cross sectional study in which convenient sampling technique was adopted.
Results: Among the 100 conveniently chosen street children, 74 were Indians and 26 were Nepalese.
Ninety-one children were Hindu, 80 were above 12 years of age with the mean age and SD 14.74 ± 2.77, 72 had some education, 54 children at present worked as hawkers and servants at shops and 69 had a daily income between Rs. 30-90.
Forty-two subjects had sexual exposure at various age with either girlfriend or prostitute in which two subjects were homosexual. Seventy subjects were substance/s abusers, which included 49 alcohol abusers, 48 smokers, 42 tobacco chewers, 3 injectable drug abusers and 51 various other types of substance/s abusers. Majority of the children were uncertain about the amount and frequency of the substance/s they abuse except the tobacco chewers where majority i.e. 28 chew one or two packets of tobacco per week.
Discussion: It is concluded that the street children are in high risk for HIV and STD infection. The study was supported by various other studies conducted in Asia, Africa and Europe.

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Hospitalization potential at an infectious diseases division of a metropolitan Hospital of Northern Italy; persisting limitations and related problems

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Retrovirology 2010, **7(Suppl 1):**P159

Background: Notwithstanding the deep modifications of the natural history of HIV/AIDS, the hospitalization potential of Infectious Diseases (ID) wards remains largely inadequate in Italy, according to the continued modification of epidemiology and disease spectrum.
Methods: A surveillance study of patients (p) needing hospitalization at our inpatient ward (located at S. Orsola Hospital, Bologna, Italy) and their outcome, was prospectively performed.
Results: From January 2000 to May 2002 our inpatient unit could rely on 16 beds, while since June 2003 (after joining with the other ID unit of our 800,000-inhabitant metropolitan area), the available beds rose to 35. The ID Specialist must act as a consultant for every p with a suspected ID, to assess need of hospitalization and/or isolation measures, and eventually search an adequate place (the so-called "bed service"), should room is not available at our ward. The rate of p admitted elsewhere dropped from the year 2000 (34.3%), to 2001 (26.9%), and 2002 (12.9%), but reached a stabilization during years 2003-2008 (12.1%; $p < .0001$ *versus* year 2000). Among the 771 p who could not be admitted by us, no epidemiologic differences were found during time, and HIV disease prevailed (311 p: 40.3%). When infectious (but not diffusive) illnesses are of concern, p may be accepted by other city Hospitals, while the event of diffusive ID required a transfert to the closest ID ward. Until May 2002, the other city ID Unit accepted over 30% of p, but the unification into a single ward partially reduced the need of transferts 40-115 Km far from our city, stabilizing the rate around 12-13% in the last six years. Among the 311 HIV p not accepted at our ward, 26% had a place at the other ID ward, 61% at our Hospital, 3% at Hospitals of the Bologna province, while 10% needed a transfert to other cities.
Discussion: Still in the third millennium, ID wards play a key role in health care inpatient assistance, although a continuous fitting to prevailing ID and available resources is needed. The lack of suitable beds for p needing admission remains a striking problem, especially when p with acute-severe illness are of concern, and a long-distance transfert may led to potentially severe risks for p health, and breakdown of isolation/protection measures.

P160

HIV/AIDS and human rights in legislation of the Republic of Serbia

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Background: The aim of the abstract is to present a short overview of the legislation of the Republic of Serbia related to HIV/AIDS and suggestions for improvement in accordance with the international regulations and practices.
Methods: Analysis of the laws and by-laws of the Republic of Serbia in connection with human rights of persons living with HIV in comparation to the key international treates and other international documents pertaining to HIV/AIDS issues.

Results: Protection and advancement of the human rights are of crucial importance for prevention of the HIV epidemic. The best efforts of the international and national communities aiming at promotion of the human rights and prevention of discrimination, provision of adequate health care for the persons living with HIV and dissemination of information to the general public, have not yet succeeded in overcoming of the stereotypes and securing adequate protection of human rights of the persons living with HIV.
Discussion: Analysis of the current legislation in Serbia indicates that there are ways, though mostly indirect, to protect the interests of the vulnerable persons. However, most of these provisions were originally drafted with other vulnerable groups in mind and can apply to the group under discussion only if the courts and other relevant bodies show willingness to interpret the existing regulations in a way that is most favorable for this group.
It would be a much better solution to have special regulations that would directly protect those human rights of the AIDS patients and the persons living with HIV that are violated most frequently. These are the rights that are in general protected by the Constitution of the Republic of Serbia and international covenants signed by the Republic of Serbia, most important among them being the International Covenant on Civil and Political Rights, the International Covenant on Economic and Social Rights and the European Convention on Human Rights. These are the right to life, the right to privacy, the right to personal dignity and reputation, nondiscrimination, the right to work under equal conditions, the right to the highest attainable standard of living, the right to health care, the right to equal reward for equal work, the right to social insurance, the right to exercise parental rights, the right to education, the right of movement and settlement, the right of expression, freedom of thought and conscience and freedom of participation in social and political life. Some of these are so-called solidarity rights and cannot be exercised by means of court decisions. They must be guaranteed by an appropriate social policy and the best efforts of government bodies. Therefore they cannot be defined solely as individual rights, but as obligations of government bodies.
The general recommendation, agreed upon by the legal experts is that the best solution available is to pass a special law (*lex specialis*) that will regulate most of the issues in connection with HIV/AIDS. This recommendation is based on the finding that there are numerous regulations that indirectly pertain to HIV/AIDS, but they are scattered over too many different laws and by-laws and contain no specific references to HIV. That might explain the small number of the HIV/AIDS related cases before the courts in Serbia. A special law on HIV/AIDS could replace or amend the provisions contained in the existing laws and clearly regulate the legal relations between different parties as required, taking into consideration the specific requirements of the HIV/AIDS related cases.

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Supporting homeless people living with HIV

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Retrovirology 2010, **7(Suppl 1):**P161

Background: Homelessness impacts on all areas of a person's life. However for homeless people living with HIV the stigma associated with the disease can lead to reluctance to seek any support. This paper reports findings from an evaluation of a 'Housing Support and Outreach and Referral' service developed to support people living with HIV. The paper highlights the importance of coordinating services across agency boundaries as a means of addressing the complexity of need experienced by homeless people living with HIV.
Methods: Two sources of data collection were used: quarterly project evaluation reports and interviews with professionals and service users. The evaluation reports recorded progress against aims and objectives, for example the number of clients for whom a tenancy had been arranged, whether or not these tenancies were sustained; the number of clients registered with health care services; whether or not they maintained contact with services and how service users perceived their own health to be.
Results: Over the course of the evaluation the service accepted 27 referrals. Of these, 15 people received tenancy support of who 12 had previously been rough sleeping or living in insecure accommodation. At the end of the evaluation period all of these tenancies had been maintained. At referral only 9 of the 27 were registered with a General Practitioner however at the end of the period all were registered. Additionally 12 people were helped to register with an HIV clinic and a further 5 people were supported to re-engage with HIV services. Monitoring data from the services showed that CD4 counts for 5 service users had improved. Interviews with service users suggested real improvements in their own perceptions of their health status and in their experiences of healthcare services.
Discussion: The evaluation of this small scale service demonstrates significant improvements for individual service-users in terms of their housing status, their contact with specialist and generic healthcare services and their own perceptions of their health status. The evaluation illustrates the importance of collaborative working as a health promotion strategy to meet the complex needs of people living with HIV who were homeless.

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Difficulties to implement a support group for HIV patients

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Background: The project was to establish a support group for HIV patients, animated by a psychologist.
Our step was to allow patients to learn how to live better with their sickness by sharing their experiments of HIV.
Methods: All the 550 patients followed in our unit were informed.
Before integrating the group the patients met the psychologist individually in order to better determine their waitings.
Seven patients were met including six finally interested for the participation in the group. Among the six, two of them finally desisted.
It had been well specified with the first group that it would take place only with the presence of at least three people and thus to inform the psychologist in case of absence.
Results: The first group took place with three participants only, the second 15 days after with three participants also, and the third (and the last in fact). The last had to take place but was cancelled for lack of participants.
Discussion: Several assumptions can be proposed to explain the failure of this project:
- the very variable request according to participants: quasi absent for some, coming more for one group of meeting being quasi in the refusal of their disease, and others, on the contrary, very petitioning to exchange on the difficulties of living with this sickness where the probable disappointment of certain participants who came only once and note that there were difficulties to divide in-depth with others wishing to remain on surface.
- the discouragement, for some, to have moved for nothing since finding itself to two (some had not prevented of their desistance), the group did not take place.
- past lapse of time between preliminary maintenance and the effective beginning of the group, (we needed a minimum number of interested patients to start the group) and finally, the schedule which returned it non accessible to people working.

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Allosteric regulation by non peptidic, low molecular weight compounds of CCR5 coupling to g-proteins and interaction with Gp120 - consequences on inhibition of R5 HIV-1 infection

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Background: Low molecular weight CCR5 ligands inhibit R5-tropic HIV-1 entry into cells. They bind to regions of CCR5 separate from the viral envelope gp120 binding site and would act by an allosteric mechanism, *i.e.* by inducing CCR5 conformational changes, which in turn might reduce CCR5 affinity for gp120. Indeed, these compounds block allosterically chemokine (CHK) binding to CCR5. Some of them are inverse agonists for CCR5, and stabilize G-protein uncoupled, inactive CCR5. But, whether all of them are inverse agonists and to what extent inverse agonism (*i.e.* G protein uncoupling) contributes to antiviral activity is unclear.
Methods: Standard protocols reported elsewhere were used. ^{35}S-gp120 from the Bx08 strain was produced using a SFV type vector in BHK cells. Viral progeny with the renilla-luciferase gene was used to infect U87 cells or PBMCs.
Results: The inhibitors Maraviroc (MVC) and TAK779 are weak and full inverse agonists for CCR5, respectively, and stabilize distinct receptor conformations. TAK and MVC promote CHK dissociation from the receptor with an efficiency correlating with their inverse agonist efficacy. However, we found that gp120 is a CCR5 antagonist, so that its dissociation does not depend on CCR5 uncoupling from G-proteins. Kinetic studies showed that gp120 dissociation from CCR5 (k_{off} = 0.59 h^{-1}) is enhanced in the presence of TAK (5.4 h^{-1}), and to a lesser extent by MVC (1.6 h^{-1}). However, in displacement experiments of ^{35}S-gp120 binding, affinities of MVC and TAK for CCR5 are in the same range (IC_{50}~7 *vs* 21 nM), although MVC is 100-fold more potent than TAK for inhibiting HIV infection.
Discussion: Our results imply that gp120 has a lower affinity for TAK- than for MVC-bound CCR5, although TAK has a weaker antiviral activity. Thus, blocking of infection by these compounds does not solely rely on their ability to reduce affinity of CCR5 for gp120.

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Modulations of human placental transfer of lopinavir, ritonavir and enfuvirtide

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Background: Lopinavir boosted by ritonavir is one of the most prescribed HAART during pregnancy. This study used the human cotyledon-perfused model to investigate different elements (albumin concentrations, High-molecular weight antiviral (enfuvirtide, 4491 g/mol) and P-glycoprotein inhibitor drugs (ciclosporin, 1202 g/mol)) that modulate its placental transfer.
Methods: Thirteen human cotyledons were perfused with different concentrations of albumin (10, 20, 30, 40 g/L). Ciclosporin A (n = 11) and enfuvirtide (n = 2) were respectively coperfused at middle time of the perfusion. Clearance index (CI) of lopinavir and ritonavir were compared at different albumin concentrations and between control and ciclosporin or enfuvirtide phases.
Results: The CI of lopinavir was significantly lower at physiologic (30 and 40 g/L) albumin concentrations in control phases ($p < 0.0001$). When adding ciclosporin A at physiological albumin concentrations, the CI of lopinavir increased significantly 10.3 fold ($p = 0.046$) and became positive for ritonavir. The mean CI of lopinavir was 0.184 ± 0.169 in the control phase and 0.253 ± 0.239 following the addition of enfuvirtide, wich is 1.4 times higher but not statistical significant (p= 0.39). There was no placental transfer of ritonavir at baseline and no variation after adding enfuvirtide. Even at supraphysiologic concentration, enfuvirtide did not cross the placenta.
Discussion: This study suggests that the most influent event for placental transfer of highly bound drug like lopinavir during pregnancy is the physiologic variation of serum albumin. P-glycoprotein expression on human placenta has also a role but seems to be slighty. Also the competition with another antiviral highly bound drugs like enfuvirtide (92%) has a little but not significant effect on placental transfer of lopinavir.

P165

Abstract withdrawn

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Duration of antiretroviral regimens in treatment-experienced patients in clinical practice

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Background: Antiretroviral regimens (ART) with few changes due to side effects or treatment failure are preferred. Therefore, duration of treatment and persistence (defined as continuing therapy or not) are useful measures of success of ART. We studied duration of ART in treatment-experienced patients (TEP) and analyzed how a history of non-adherence affects it.
Methods: In September 2009, we conducted a retrospective, observational study of adult TEP whose ART was switched between 01/05/2008 and 30/04/2009. We used pharmacy records to select all patients who switched an ART containing darunavir, raltegravir, maraviroc, and/or etravirine and patients who had switched regimens not containing these drugs (1:1) on the same day. The primary endpoint was duration of treatment from inclusion until the last refill, or until the first refill of a new regimen. Patients were classified as non-adherent if they had collected less than 90% of the doses needed during the year before inclusion. Control variables were viral load (VL), CD4 count at inclusion, and time since first ART regimen (tART). Persistence was estimated using Kaplan-Meier plots. Groups were compared using the log-rank test and a Cox regression model was adjusted for control variables.
Results: We included 146 patients (66.4% men); mean age 45.4 years. Baseline clinical characteristics (median [IQR]) were VL = 50 (50-7331) copies/mL, CD4 count = 345 (184-540) cells/μL, tART = 10.0 (4.2-10.7) years. Etravirine, maraviroc, and/or raltegravir were administered to 45.9% of patients, and efavirenz to 21.9%.
Persistence (95% CI) at 6 and 12 months was 77.9% (70.9%-84.9%) and 65.9% (55.5%-76.5%), respectively. Mean duration of treatment was 342 (15) days. Non-adherence was observed in 29.3% (21.2%-37.3%). Persistence at 12 months for adherent and non-adherent patients was 71.7% (58.1%-85.3%) vs 59.0% (32.2%-84.8%), respectively ($p = 0.342$). There were no differences in persistence adjusted for control variables (HR, 0.774 [0.343-1.750]; $p = 0.539$).
After a median 281 days of follow-up, 24.7% (17.7-31.6) had stopped or changed ART because of toxicity (52.8%), treatment failure (19.4%), or simplification (8.3%).
Discussion: Our results for persistence are similar to published data for naïve patients and toxicity is also the main reason for switching treatment. A history of non-adherence has less effect on persistence than expected.

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A model using adherence: house compliance center Oasis AAS

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Background: Putting in processing a large number of people receiving ARVs needed to enhance adherence to ARV treatment. But this development should be fixed on a new vision. The activities of aid Adherence: being punctual, our profit were delayed or even absent in ARV treatment because they did not have enough information ARVs and even on the resistors. They did not meet the appointment aid activities Adherence: while each visit has a theme. It has not happened has touched all the meetings, that is how the idea of creating a house of adherence is born.

Methods: The House of adherence is a community center for patient education, promotion of health and readiness to return to active social life of people living with HIV/AIDS on ARV therapy. In contrast with brief and timely aid to compliance, the specificity of home monitoring is to provide a temporary withdrawal of the living environment which can involve deeper and more lasting quality lives of PLWHA. The House of observance takes a comprehensive approach to compliance that puts decision-treatment against AIDS in the context of psychological, social and economic life of people and their environment.
Results: This project allowed people on ARV treatment to integrate into their lifestyle, to help restore equity in access to ARV treatment for people most vulnerable and preparing for expanding access to treatment ARVs. Our approaches were based on psychosocial determinants, medical, economic, political etc.. This house observance is open to all people on ARV therapy, starting treatment or changing treatment. In addition the person must have difficulty taking ARVs, be in good health, does not present a contagious infection, be motivated and committed to participate. It should be noted that 108 people who stayed and after a period of 07 months, 87% saw their CD4 counts increased by more than 60.8% under 50CD4, 5% saw their CD4 dropped by over 80% took more 5 kg, 14% less than 5 kg, 4% did not change weight and 2% decreased weight.
Discussion: This house has had much impact because more than 80% of persons conducting activities and who was abandoned due to the disease resumed its activities 06 months after their passage home observance. We believe expanding the house of compliance has other associations and replicate within the association.

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Scorpion model of influenza A/H1N1: Hemagglutinin (HA) contains a scorpion toxin, binding to voltage-gated sodium Na+ channel: Na+ channel inhibitors as therapy

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Background: Despite therapeutic progress in receptor binding inhibition, some deaths continue to occur, especially in young people. We try to find new approaches to fight against Influenza virus resistance. We were interested in Guillain-Barré syndrome (GBS) during the 1976 US vaccination campaign, because Breurec JY in France described a case of GBS after a scorpion sting by a Centruroides noxius species.
Methods: We compared amino acid sequences of Influenza virus A/H1N1 with scorpion venom toxins.
Results: The cysteine-rich region (56-109) with C59, C72, C84, C107 was aligned in 3 dimensions with scorpion toxin Figure 1.
a) HA of pandemic Influenza virus A/H1N1 2009 Mexico (466-VKEYI-462) [ACY77964], Canada-AB (56-109), Japan (246-YYWKLV-251)
b) scorpion toxin (AaH II/Cn II-13 VKEGYI) (AaH IT4 YFWKLA)
Discussion: The finding of a three dimensional scorpion toxin in HA means that Influenza virus binds to the scorpion toxin receptor, i.e. the voltage-gated sodium Na^+ channel; in fact, flecainide, a sodium channel ligand, can alleviate experimental auto-immune neuritis induced by P2 myelin protein in Lewis rat (Bechtold DA, 2005). Many drugs act on the sodium channel: Local anaesthetics, antiarythmics, antiepileptics, antimalarials, fatty acid omega 3, Tacrine. Quinine was used as an antipyretic against Influenza, but it may by serenpidity be an antiviral by inhibiting the sodium channel. Omega 3 is particularly interesting, as it is very well tolerated even at high doses and can also be given I.V. (Omegaven).

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Development of a test system for simultaneous detection of HIV RNA and Hepatitis C virus (HCV) and DNA of Hepatitis B virus (HBV) in blood samples

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Background: The aim of this work was to create a PCR test systems for simultaneous detection of HIV RNA, HCV RNA and DNA of HBV, which will have high sensitivity and high bandwidth.
The issue of viral safety of blood products is of particular relevance in relation to the epidemiological situation with regard to viral hepatitis and HIV infection. The use of serological tests for screening of blood products has significantly reduced the risk of transmission of these infections in the transfusion. But from the moment of infection until the appearance of antibodies to the virus may be a considerable period, known as «serological window». In this situation, great care may have tests to detect the virus itself or its components. Using molecular genetics methods of diagnosis helped reduce the period of «serological window» and improve the safety of transfusion.
The actual development of low-cost highly sensitive tests, allowing both to detect HIV and hepatitis viruses B and C, increase the capacity of blood transfusion stations in the molecular genetic studies. This involves the basic requirements for test systems for screening of donated blood - the highest analytical sensitivity.
Methods: To increase the sensitivity and specificity of multiplex PCR, a novel Dual Priming Oligonucleotide (DPO) technology was used. This system is structurally and functionally different from the primer system currently in wide-spread use blocked extension of non-specially primed templates, and thereby generates consistently high PCR specificity even under less than optimal PCR conditions. The DPO primer system includes a poly(I) linker between two unequal segments of primer sequences. The poly(I) linker forms a bubble-like structure that separates a single primer into two functional regions, thereby increasing specificity. Furthermore, because the bubble like structure of its poly(I) linker efficiently prevents primer-dimer and hairpin structure formation, the DPO system is more accurate for multiplex PCR applications. The test system developed for the simultaneous detection of nucleic acids of HIV, HCV and HBV, involves extraction of RNA/DNA from individual samples (100-1000 µl). The selected RNA/DNA is added to tubes containing the reaction buffer for RT-PCR combined with reverse transcription. Detection of amplification products was done by using the TaqMan technology.
Results: To determine the analytical sensitivity and the inhibitory influence of concomitant diseases, a number of experiments on model samples containing all three infectious agent in different concentrations were done. The test system allows both to identify nucleic acids of HIV, HCV and HBV with a sensitivity of 50 RNA copies/ml (for HIV), 10 IU/ml (for HCV) and 20 copies of DNA/ml (for HBV).
Discussion: To use a molecular diagnostic method in the service of blood in order to increase the viral safety should be developed test-system with high sensitivity and high bandwidth. In this regard, we develop a

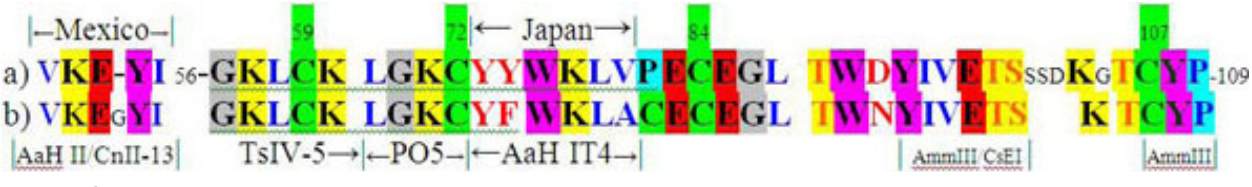

Figure 1 (abstract P168)

DPO-multiplex PCR, for simultaneous detection of HIV nucleic acids and viruses of hepatitis B and C. The proposed format is a low cost and high throughput. We also showed that the DPO-PCR method was successfully utilized for multiplex amplification with a little amount of test samples with high sensitivity and specificity because of high accuracy of priming during PCR.

P170

Analyses reveal a novel avirulent *Streptococcus suis* Serotype 2 strain that induces protective immunity against challenge with the highly virulent strains

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Background: By comparative analysis of the bacterial genomesm of *Streptococcus suis* serotype 2 both virulent and avirulent strains are instrumental in the development of vaccines and the functional characterization of important of genetic determinants.
Methods: We isolated from a clinically healthy pig an avirulent strain, 05HAS68.
Results: By comparing the genomes of the virulent and avirulent strains, we observed massive genome rearrangements that may result in alterations in gene expression and, in turn, causing enormous single gene gain and loss for the *S. suis* 2 species. It is most interesting that both virulent and avirulent species feature a similarly structured genomic island (GEI) which carries different idiosyncratic systems as an adoptive evolutionary response. Strikingly, all of the animals vaccinated with the avirulent strain 05HAS68 were protected from challenge infection with the most virulent *S. suis* 2 strain, 05ZYH33, and, furthermore, the protective immunity could be transferred with T cells and plasma from the vaccinated pigs to unimmunized animals. Increased production of tumor necrosis factor alpha (TNF-alpha) and gamma interferon (IFN-gamma) in peripheral blood of the immunized animals may be attributed to the protective immunity elicited by the live vaccine.
Discussion: Since the *S. suis* 2 strains command large genetic diversity, it is almost impossible to achieve global protection for all *S. suis* 2 pathogenic strains by depending on one, or even several, virulence-related substances as vaccines. In view of our results, both humoral and cellular immunities induced by live 05HAS68-based vaccine are required for the specific protection against virulent *S. suis* strains; therefore, we suggest reconsideration of research strategy in terms of model strains to test for vaccine design.

P171

A prospective project of microbiological surveillance at a teaching Hospital in Italy; evolving epidemiological features, and in vitro antimicrobial sensitivity trends

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Background: An active bacteriological surveillance project is part of the mandatory knowledge on the local microbial isolation and their antimicrobial resistance pattern.
Methods: The trend of microbial isolations from patients admitted in the last year 2008 (January 1, 2008-December 31, 2008) at our Hospital is reported on quarterly basis, together with updated antimicrobial sensitivity testing.
Results: As a whole, Gram-negative agents showed an increasing trend of isolation, regardless of the examined clinical specimens, while the epidemiology of Staphylococci remained somewhat unchanged, and their methicillin resistance rate remained under control (around 39% of overall isolates from blood cultures). When considering Enterococci, the active surveillance of VRE strains successfully acted against the potential nosocomial spread of these organisms: the only 7 cases of "Van A" *Enterococcus faecalis* strains were not related with each other, and glycopeptide resistance remained limited to less than 3% of overall Enterococci. An increasing number of ESBL-producing Enterobacteriaceae was noticed (with a 29-36% quarterly rate for *Escherichia coli*, and up to 41% for other organisms, as a whole). The overall resistance rate against fluoroquinolones is on steady increase: the last quarterly report shows a 47.3% rate for *E. coli*, 30.8% for Enterobacteriaceae, and 42.6% for *Pseudomonas aeruginosa*.
Discussion: An active, prospective microbiological monitoring may significantly add to the knowledge of local epidemiological figures and antimicrobial sensitivity trends, and plays a role of paramount importance when selecting and planning chemoprophylaxis and therapeutic strategies, on a local and regional basis.

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A prospective microbiological surveillance in a teaching Italian Hospital; microbial isolations and in vitro antimicrobial susceptibility levels, and their modifications over time

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Background: A prospective microbiological surveillance of micro-organisms isolated at a reference hospital and their antimicrobial susceptibility, is of paramount importance in the awareness of evolving local epidemiology.
Methods: The trend of microbial isolations from patients (p) hospitalized in the last year 2008 (January 1, 2008-December 31, 2008), is reported on quarterly basis, together with the related antimicrobial sensitivity testing. Surveillance cultures and multiple isolations of the same organism(s) from one p within one month of hospitalization, were excluded.
Results: 4,906 overall evaluable pathogens were isolated in the last 12 mo, with *E. coli* (1,279), *E. faecalis* (596), *P. aeruginosa* (430), and *S. aureus* (365), as prevailing organisms. Among blood cultures (1,421 isolates), a major role was played by Staphylococci as a whole (688), followed by *Enterobacteriaceae* (253), and Enterococci (77). With regard to the overall susceptibility rates, methicillin-resistant *S. aureus* accounted for a mean 42%, while methicillin resistance was greater for *S. epidermidis* (>70%), with a slight reduction compared with the year 2007 figures. Substantially ameliorated sensitivity levels were found among Enterococci, with only 7 "VRE" strains recorded (*versus* 17-21 strains per year, during 2005-2007); a slight increase of resistance towards glycopeptides was found, reaching 3.1% of isolates in the last 3 mo. A 25-38% resistance rate to macrolides was found among streptococci, without appreciable temporal variations. Among Gram-negative organisms, extended spectrum beta-lactamase production regarded 31.8% of overall *E. coli* isolates, and 39.6% of other Enterobacteriaceae, with a slight increase compared with the year 2007. *P. aeruginosa* showed an stable resistance pattern to penicillins-cephalosporins (including those protected by beta-lactamase inhibitors), carbapanems, fluoroquinolones, and aminoglycosides (ranging from 55% to 75% of strains), but remained full susceptible to colistin. *Stenotrophomonas maltophilia* confirmed its extensive resistance spectrum, but remained 90-100% sensitive to cotrimoxazole and colistin, as well as *Acinetobacter* spp., which showed a favorable susceptibility rate (60-100%) to aminoglycosides, piperacillin-tazobactam, and colistin.
Discussion: An active bacteriological surveillance may notably add to the knowledge of local epidemiological figures and antimicrobial sensitivity trends, and plays a major role when planning surveillance measures, chemoprophylaxis, and empiric antimicrobial treatment, on both local and regional basis.

P173

Auto-immune thrombocytopenia after Measles Mumps Rubella MMR vaccination: molecular mimicry of measles virus phosphoprotein with platelet gpIIb

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Retrovirology 2010, **7(Suppl 1):**P173

Tabele 1 (abstract P173)

platelet gpIIb 782	RGNSFP
Measles virus P protein	199-RGNSFP-204

Background: MMR vaccination is complicated by rare cases of auto-immune purpura or thrombocytopenia in a chronological delay of about 10-24 days, which corresponds to the rise of antibodies (Autret E, 1996; Vlacha V, 1996). We have demonstrated that in the cases of HIV-1, Parvovirus B19, Chikungunya virus, Leptospirosis, the culprit is an epitope localized on the platelet gpIIIaIIb, centred by a phenylalanine-proline (Tran MKG, ISHEID Toulon 2002); these auto-antibodies are very powerful and induce in mice a thrombocytopenia; allo-immunisation from mother to the newborn has the same epitope on gpIIIaIIb (Brouk H, 2009). We continue the same direction of research for MMR vaccine.
Methods: Comparison of amino acid sequences between gpIIIaIIb and viruses (measles, mumps and rubella).
Results: There is a molecular mimicry between gpIIb (P08514) and measles virus (strain MVi/Victoria.Aus/12.99) Phosphoprotein P (ABV24494) (Bankamp B, 2008), Table 1.
Discussion: The occurrence of thrombocytopenia 2 or 3 weeks after MMR vaccination is an auto-immune phenomenon, on a peculiar genetic background prone to make auto-antibodies against phenylalanine-proline containing epitopes. MMR vaccine must be avoided in these patients with idiopathic thrombocytopenia (Drachtman RA, 1994). The chronological argument is by itself convincing and confirmed here by the biological finding of the causal epitope on platelet. Thus we must be very cautious in presence of a MMR vaccine clinical auto-immune complication (such as autism) (Wakefield AJ) and not discard it as a simple coincidence, but rather try to elucidate its mechanism and genetics (HLA-DR4). The gpIIIaIIb epitope may serve as a chelating hapten for treatment.

P174

Urinary tract pathogens among inpatients at a large Italian tertiary care Hospital; a prospective monitoring study

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Background: A standardized, prospective microbiological surveillance study of urinary tract infection is ongoing at our Hospital.
Methods: The temporal trend of microbial isolates from urines of inpatients hospitalized during the last available calendar year (2008), was evaluated quarterly according to the main bacterial and fungal isolates. The same pathogens cultured more than once from the same patient within one month, have been considered only once.
Results: Of 2,327 overall episodes (as defined above), 983 (42.2%) were determined by *Escherichia coli*, followed by *Enterococcus fecalis* (443 episodes: 19.0%), *Proteus mirabilis* (156 cases: 6.7%), *Klebsiella* spp. (152 episodes: 6.5%), *Pseudomonas aeruginosa* (117 cases: 5.0%), *Enterococcus faecium* (56 episodes: 2.4%), while the first fungal pathogen was *Candida albicans* (70 episodes: 3.0%). During the proportionally short observation period, no significant variations occurred in the frequency of isolation of each mentioned urinary tract pathogen, when excluding a mild increased frequency of *Enterococcus faecium* and *Enterobacter cloacae* ($p < .07$).
Discussion: A prospective microbiological observation study significantly adds to the knowledge of local epidemiological figures and antimicrobial susceptibility pattern of hospital-associated infections, including urinary tract infections, which are responsible for considerable morbidity among inpatients. During the still limited (one year) observation time, modest variations occurred in the frequency of isolation of the most frequent microorganisms, with *Escherichia coli* representing over 40% of cultured organisms, followed by *Enterococcus faecalis* (which proved responsible of around 19% of overall episodes). The tendency towards an increased incidence of *Enterococcus faecium* and *Enterobacter cloacae* is of concern, given the unpredictable antibiotic sensitivity profile of these last microorganisms.

P175

Agents causative of sepsis-bacteremia in a four-year prospective surveillance study carried out at a teching Italian Hospital

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Background: A prospective microbiological surveillance study of bacteremias is ongoing at our Hospital since the year 2005.
Methods: The temporal trend of microbial isolates from blood cultures of inpatients hospitalized during the last four calendar years (2005 to 2008), was evaluated according to the main bacterial and fungal isolates. The same pathogens cultured more than once from the same patient within one month, have been considered only once.
Results: Of 4,168 overall episodes, *Staphylococcal epidermidis* remained the leading organism (761 cases: 18.3%), but a dramatic drop in its frequency occurred during the observation time (from 26.1% of cases in 2004, to 18.3% in 2008; $p < .0001$). The second causative agent of bacteremia was *Escherichia coli* (465 episodes: 11.2%), followed by *Staphylococcus aureus* (309 cases: 7.4%), *Enterococcus faecalis* (223 episodes: 5.4%), *Pseudomonas aeruginosa* (179 cases: 4.3%), *Klebsiella* spp. (143 episodes: 3.4%), and *Enterococcus faecium* (104 cases: 2.5%). When excluding the above-mentioned changes is staphylococcal isolations, significant time-based modifications occurred only for *Pseudomonas aeruginosa* (temporal increase: $p < .04$), and *Klebsiella* spp. (temporal increase: $p < .01$). Among fungi, *Candida albicans* was the most represented organism, with 104 episodes (2.5%), without changes in its frequency in the 2005-2008 period.
Discussion: A prospective microbiological monitoring is expected to significantly add to the awareness of local epidemiological figures and antimicrobial sensitivity profile of hospital infections, including bacteremias, which are responsible for considerable morbidity and mortality rates among inpatients. Although the main ethiological agents of inpatient bacteremias are still represented by coagulase-negative Staphylococci, these microorganisms significantly declined during the four-year study period, thus confirming a positive trend toward a progressively reduced incidence of contaminated blood cultures. On the other hand, an appreciable increased frequency occurred over time for *Pseudomonas* and *Klebsiella* spp. A major, persisting role as agents of hospital bacteremic episodes is still exerted by *Escherichia coli* among Gram-negative pathogens, and *Staphylococcus aureus* among Gram-positive ones.

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The emerging and the spread of the fifth Plasmodium responsible of human malaria: *Plasmodium knowlesi*

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Background: On the ground of the most recent evidences regarding the role of some malaria Plasmodia which have primates as natural reservoirs, the Authors focused their attention on the emerging species *Plasmodium knowlesi*.
Methods: Based on the international literature reports, *P. knowlesi* infectious foci have been identified in the past decade in Malaysia, and in particular in the States of Sarawak-Sabah (Malaysia Borneo), and in the Pahang region (peninsular Malaysia).
Results: The valuable role of molecular biology assays (polymerase chain reaction), performed with specific primers for *P. knowlesi* is underlined, since the traditional hemoscopic examination does not allow to distiguish specific features, especially when *P. knowlesi* is compared with protozoa belonging to the species *Plasmodium malariae*. Furthermore, malaria caused by *P. knowlesi* may be burdened by a severe and sometimes fatal course, since the clinical presentation and course are more severe

compared with those of *P. malariae*, especially due to the extremely elevated levels of parasitemia. The most effective vector for *P. knowlesi* transmission is represented by the mosquito *Anopheles latens*, which may parasite both humans and primates. Among primates, the natural hosts of *P. knowlesi* are represented by the species *Macaca fascicularis*, *M. nemestina*, *M. inus*, and *Saimiri scirea*.

Discussion: After remarking the possible, severe prognosis of *P. knowlesi* malaria, we underline the paramount importance of a timely diagnosis and therapy, especially when patients gain access to Western Hospitals, after retrurning from South-Eastern Asia regions, where they eventually practiced excursions and trekking activities in the rain forest. When signs-symptoms suggestive of malaria are recognized in subjects coming back from endemic areas, a rapid diagnosis and treatment are crucial in the management of *P. knowlesi* malaria. In the light of the most recent epidemiological issues, certainly *P. knowlesi* may be added to the list of the known human malaria parasites, which until now included *P. vivax*, *P.*ovale, *P. malariae*, and *P. falciparum*, as the fifth potential human pathogenic Plasmodium. In the next future, an extensive surveillance system and an improved epidemiological control will become needed. Paralleling epidemiological studies and public health implications, a more accurate survey of the clinical features of *P. knowlesi* will be strongly needed, since preliminary data indicate an increased disease severity, associated with a greater level of parasitemia, probably secondary also to the progressive increase of the number of interhuman "passages" of this emerging species of malaria Plasmodium.

P177

Perceived quality of clinical Care provided by nurses to people living with HIV at Four TASO sites in Uganda

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Background: The AIDS Support Organization (TASO) started an ART program in 2004. With the increasing client load, it was inevitable that clinical care had to be shifted to the nurses. This study aimed at assessing the quality of clinical HIV care provided by nurses to TASO clients.

Methods: A cross-sectional survey employing qualitative and quantitative study designs was conducted at 4 purposively selected TASO sites. 400 exit interviews were conducted; 28 questionnaires administered to nurses, 56 clinical sessions observed; 51 medical charts reviewed and 4 key informant interviews conducted. Data was analyzed with the aid of Excel version 2000 and STATA version SE/10. Qualitative data was analyzed with the aid of Nvivo.

Results: About 92% of the clients expressed satisfaction with services received from TASO nurses. Peasant farmers were twice more likely to be satisfied with staff availability than the unemployed (OR= 2.08; 95% CI: 1.01 – 4.26; p-value = 0.046*). Married clients were three times more likely to be satisfied with staff availability than the co-habiting (OR = 2.64; 95%CI: 1.20- 5.82; p-value = 0.016*). Clients who had attained primary education were 1.5 times more likely to be satisfied with staff availability than the uneducated (OR = 1.52; 95% CI: 0.91 – 2.55; p-value = 0.113). Nurses needed technical support to manage complex disease conditions. Fifty six percent of the nurses were able to write accurate ARV prescriptions. Key informants noted that comprehensive nurses had good clinical skills and positive attitudes.

Discussion: TASO Nurses exhibit positive attitudes. Comprehensive nurses have better clinical skills than the other nurse cadres. However, the overall nurses' technical competence as regards clinical HIV care provision requires improvement. They should be permitted to provide clinical care, under the supervision of a medical officer. They should be given protocols to follow during clinical sessions. Nursing schools should incorporate clinical HIV/AIDS care training into their curricula. Gender biases among clients should be addressed and a national dialogue on task shifting held.

P178

An increasing pathomorphism of pulmonary tuberculosis. Is there a therapeutic role for novel antimicrobial compounds effective on *Mycobacterium tuberculosis*?

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Background: During recent y, a progressive emerging of tuberculosis (T) occurred, related to the overall increased age of general population, primary-secondary immunodeficiencies (including frequent-prolonged exsposure to immunosuppressive-cytotoxic treatments), surgical and intensive care supports, bone marrow-solid organ transplantation, and recent immigration of people coming from T-endemic areas.

Methods: Since Jan 2006, we hospitalized 78 cases of pulmonary T, in over 75% of cases occurred in patients (p) immigrated from developing countries.

Results: In only 4 p resistant or multiresistant (MDR) T strains were found, while 2 more p had a multi-resistant (XDR) T. Although enforcing all possible measures to increase p'adherence to treatments (empowerment, use of i.v. formulation whenever possible, delivery of oral drugs under direct control), over one third of p had a very slow clinical-radiological ameliorement (2-4 mo) (19 p of 78 even experienced an initial worsening during early treatment), with persistence of sputum and/or bronchoalveolar lavage (BAL) fluid positive for *Mycobacterium tuberculosis* for over 2-4 mo (mean 2.3 ± 0.7 mo), during apparently adequate treatment. When excluding XDR-MDR p, which had very prolonged admissions (sometimes over one y), and always deserved second-third-line drugs, in 8 more p we observed that off-label linezolid (L) adjunct together with at least 3 drugs with residual activity against T, led to a rapid clinical-radiological improvement and negative microbiological search, with consequent possibility to achieve a protected discharge, supported by a sequential, oral therapy. L was also successfully employed in all p with XDR-MDR T, when a temporarily negativization of respiratory secretions was achieved always and only after the L adjunct to a combination therapy selected on the ground of residual *in vitro* susceptibility assays.

Discussion: Notwithstanding the maintained, extensive microbiological susceptibility of *M. tuberculosis* strains responsible of the great majority of cases of pulmonary T, an unexpected tendency of p to have a persistingly positive sputum/BAL and experience prolonged hospitalization for cure and isolation, has been recognized in the last y. No particularly suggestive chest imaging seems predictive of a so prolonged course, so that we presently lack of clinical-radiological elements predictive of this slow treatment response. The oxazolidinone L has an affordable activity against *M. tuberculosis*, and an extremely elevated intracellular concentration in respiratory tissues. The increasing microbiological, pharmacological, and clinical evidences may recommend the use as an off-label salvage L treatment of pulmonary T refractory to treatment, although not necessarily determined by resistant (MDR-XDR) strains. To rely on controlled data, randomized clinical trials including initially p with chemioresistant T, are needed.

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Symptomatic, prolonged Parvovirus B19 infection and accompanying illness in otherwise healthy adults

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Background: Parvoviridae are part of air-, parenteral- and perinatal-transmitted ubiquitous viruses, whose associated signs and symptoms strongly depend on patient's age and immune defence.

Methods: All cases of symptomatic Parvovirus B19 infection in otherwise healthy adults which came to our attention since spring 2006 were prospectively investigated and followed-up.

Results: In a 21-month period, 11 patients (7 females and 4 males), with a mean age of 36.9 (range 27-46) years with a symptomatic Parvovirus B19 infection were recorded and followed-up. Intrafamiliar exposure and occupational (health care) exposure were identified in two cases each. Clinical signs and symptoms included fever (100% of cases), polyarthralgia (90%), followed by headache (80%), anemia (70%), and rash. A mild-to-moderate myelosuppression of all hematological lines characterized 8 cases of 11 (72.7%), while increased serum transaminases were associated in 63.6% of patients. Three patients of ours deserved hospitalization (mean 10.8 days of admission), and five more cases were followed on Day-Hospital basis (for a median 75-day period); in four patients a treatment with i.v. high-dosage human immunoglobulins was performed. Elevated levels of specific serum anti-Parvovirus B19 IgM antibodies were detected in all cases. In a 33-year-old female a severe anemia and a persisting headache, vomiting, and neck stiffness, led to RBC transfusion and a diagnosis of meningoencephalitis, with positive search of IgM antibodies and Parvovirus B19 viremia (detected by RT-PCR) in the cerebrospional fluid which lasted up to three months, despite treatment with i.v. serum immunoglobulins.
Discussion: Parvovirus B19 infection may play a significant role also in adult, immunocompetent subjects, and the disease sometimes is not mild and self-limiting, requiring admission and/or frequent outpatient interventions in a significant number of cases. The causes supporting a persistant infection in immunocompetent subjects have not been investigated to date, as well as the pathogenesis of myelosuppression and severe polyartrhalgia. Symptomatic Parvovirus B19 infection is still an underestimated clinical condition, and therapeutic perspectives are extremely limited until now.

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An Infectious Disease Day-Hospital service in a metropolitan area of Northern Italy. Evolving assistance features in the last fifteen years (1994-2008)

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Background: Aim of our study is to evaluate the frequency and features of admissions performed at an Infectious Diseases Day-Hospital service at S. Orsola Hospital, Bologna, Italy.
Methods: A retrospective evaluation of all admissions of the last 15 years (1994-2008), was performed.
Results: Before the introduction of potent, combination antiretroviral treatments (cART) (years 1994-1996), the proportionally low mean number of admissions (110/year), was linked to the elevated prevalence of HIV disease, which accounted for 89.4% of Day-Hospital hospitalizations, their recurrence, and their prolonged duration. Immediately after cART introduction, the number of Day-Hospital admissions showed a significant increase, from 171 (year 1997), to 318 (2002),338 (2003),347 (2004),331 (2005),356 (2006), 341 (2007), and 378 (2008) ($p < .0001$ *versus* the pre-cART era), although this phenomenon paralleled a drop of percentage of HIV-related admissions (from 59.1% of 1997, to a minimum of 23.8% of the year 2005; $p < .0001$). While HIV-associated hospitalizations decreased, a temporal increase of admissions due to chronic liver disease occurred ($p < .0001$). The reduction of admission duration allowed an increase of overall number of hospitalizations of each examined year ($p < .0001$), and the mean bed occupation rate showed a continued rise (8.2 in the year 2000, to maximum value of 12.0 reached in the year 2006 ($p < .0001$).
Discussion: The modifications occurred at our Infectious Diseases Day-Hospital service during the last 15 years are largely attributable to the significant changes occurred in the spectrum of infectious disorders which came to our attention: from a low number of prolonged hospitalizations typical of patients with advanced HIV disease, the cART era led to a progressive broadening of the spectrum of disease, and a notable reduction of admission time. Notwithstanding this situation, no significant modification was observed as to mean weight of diagnosis-related group (DRG) features:from a mean 1.03 rate per patient of the year 2000, to a mean 1.33 figure in the year 2008. The evolution of assistance features in a Day-Hospital setting, seems strictly linked to the modification of prevailing disorders. A permanent monitoring of the features of health care provision at an Infectious Disease Day-Hospital service may allow to consider significant temporal modifications, and contribute to ensure adequate assistential planning, including the eventual revision of structural, professional, technical, and funding resources.

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Is there pre-existing cross-reactivity to influenza A (H1N1)2009 in a tropical population?

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Background: With the advent of the 2009 influenza pandemic, we sought to determine if there was pre-existing cross-reactive response and potential protection against the novel influenza A (H1N1) pandemic virus in our population in tropical Singapore.
Methods: 246 archived sera collected between January 2008 and March 2009 from persons aged 0 – 93 years were tested, in serial dilution from 10 to 1280, for haemagglutination inhibiting (HI) antibody against pandemic influenza A/Auckland/1/2009(H1N1)v and seasonal influenza A/Brisbane/59/2009(H1N1).
Results: 12.6% had detectable antibody titres against A/Auckland/1/2009 (h1N1)v. Of these, only 2.9% had titres ≥ 40; such titres were most frequently seen in persons aged at least 70 years (9.4%; 3 of 32), followed by those aged 20 – 29 years (6.5%; 2 of 31) and 30 – 39 years (5.9%; 2 of 34). The peak titre was 160, in 2 individuals aged 91 and 30 years. None from the other age groups showed any sero-reactivity.
Comparatively, antibody reactive to A/Brisbane/59/2002(H1N1) was detectable in 52.7% of all individuals and 20.8% of these had titres ≥ 40. Those aged 10 – 19 years, followed by 20 – 29 years, had the highest percentage of titres ≥ 40 of 51.5% and 45.2%, respectively. The peak titre detected was ≥ 1280.
Discussion: We found minimal cross-reactive antibody response to the 2009 H1N1 virus in our study group. The very elderly (70 years old and above) had the highest frequency of titres of ≥ 40, at a mere 9.4%. They probably had been infected in the distant past with strains more antigenically related to the present virus. Four relatively young and mobile adults between 20 to 39 years old had titres ≥ 40 against 2009 H1N1, possibly through travel-associated exposure in a globally connected world. With only 2.9% overall having titres ≥ 40, it would appear that virtually the entire population would need the pandemic influenza vaccine and/or adherence to infection control practices for protection.

P182

Molecular epidemiology of chikungunya strains in Singapore in 2008

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Background: Strain characterization and genotyping of chikungunya virus (CHIKV) using sequencing for epidemiological study.
Methods: Blood samples were inoculated into *A. albopictus* C6/36 cells and Vero cells. Positive cultures for CHIKV were confirmed by RT-PCR targeting the gene that codes for CHIKV viral envelope protein E2. Viral RNA was extracted from the supernatant of viral infected Vero cells using QIAamp Viral RNA Mini Kit (Qiagen). RT-PCR was performed using a one-step RT-PCR kit (SuperScript™ III One-Step RT-PCR System with Platinum® *Taq* DNA Polymerase, Invitrogen). Primers 9648F, 10403R, 10145F, 11158R, 10959F and 11690R were used to amplify and sequence the E1 gene. DNA fragments were purified using QIAquick PCR Purification Kit (Qiagen) and sequenced using the same primers. The sequences obtained were

aligned using ClustalX programme. The phylogenetic tree was constructed based on the 1,044-nt region within the E1 gene from codons 91-438 by neighbor-joining method with Phylip, Version 3.5 and the statistical significance estimated by bootstrap analysis using 1,000 pseudoreplicate data sets.
Results: Three CHIKV strains were successfully isolated. These samples were all collected in August 08. The molecular epidemiological study revealed that all viruses were related to East, Central, and South Africa (ECSA) phylogroup. All isolates had alanine replaced by valine at aa residue 226 (A226V) of the E1 gene. Besides the nonsynonymous mutation, these isolates possessed 2 nucleotide mutations, C300T and A363G of the E1 gene. One of the isolates showed another synonymous mutations at nucleotide position 1030 (A1030G) of the E1 gene.
Discussion: Phylogenetic analysis suggests that the circulating chikungunya strains in Singapore in August 08 belong to the genogroup ECSA, which has caused large CHIKV outbreaks in several countries worldwide and beyond the African continent, like in the Indian Ocean Islands and India during 2005-2006. C300T, A363G and C677T (A226V) found in all CHIKV isolates in Singapore were also identified in CHIKV strains isolated in Malaysia from April to December 2008. Notably, C300T was unique to CHIKV strains isolated in Malaysia. Our study suggests that the concurrent chikungunya outbreaks in Singapore and Malaysia, where two countries are of close geographical proximity, were interconnected.

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Synthesis of the studies about the transmission cycle of malaria in an area of very low incidence outside the Amazon Region in Brazil

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Background: To describe malaria transmission cycle in an area of residual occurence in Brazil.
Methods: Survey of cases reported from April 2001 to March 2004. Blood sampling of inhabitants (two kilometers around the cases). ELISA for antibodies against the Circunsporozoite protein (CSP) of *Plasmodium vivax* and its variants, and *Plasmodium malariae*. Blood smears. Indirect Fluorescent Antibody (IFA) test for crude blood stage antigens of *P. vivax*, *P. malariae* and *Plasmodium falciparum*. Protein chain reaction (PCR) for amplification of the DNA of *P. vivax*, *P. malariae* and P. falciparum (Rubio's protocol). Mosquitoes captures with CDC-CO2 traps placed in the canopy and in the ground inside the forest and at its margin, as in the open field. Shannon light traps at the margin of the forest. PCR of the macerated mosquitoes for DNA of *Plasmodium* (Kimura's protocol) (thorax and abdomen tested separately).
Results: Sixty-five patients and 1,777 inhabitants evaluated. Patients were 35.11 +/- 16 years old in average and most of them were males (78.5%). There were no case clusters. *P. vivax* (morphology and PCR) in 47 of 48 patients, and *P. malariae* in the remainder. Anti-CSP antibodies for *P. vivax,* its variants and *P. malariae* in high frequency in the patients and in the inhabitants. IFA positive for IgM against *P. malariae* in 40 of 253 samples from the inhabitants (15.8%) and in 113 of 253 for IgG antibodies (44.6%). Figures for *P. vivax* were 105 of 1,701 (IgM) (6.2%) and 641 of 1,701 (37.7%) (IgG). PCR from the inhabitants revealed *P. vivax* in 23, *P. malariae* in 15, *P. falciparum* in 9 and *P. falciparum* plus *P. malariae* in one. *Anopheles cruzii* had an acrodendrophylic behavior (90% of the specimens captured in the canopy). DNA of *Plasmodium* was amplified in several species of anopheline mosquitoes.
Discussion: Predominance of males and absence of vectors nearby the houses favor an extradomiciliary transmission. Absence of clusters and distance among the cases point out to the existence of a silent reservoir. Two possibilities: asymptomatic human reservoir (not important in our survey) and simian reservoir (close relation between cases and the forest; acrodendrophylic beahvior of the main vector).

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Clinical presentation of influenza a (H1N1) infection in a local emergency department in Greece

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Background: After the global spread of the new swine-origin influenza virus A (H1N1) the World Health Organization raised the level of influenza pandemic alert up to phase 6, thus declaring a public health emergency of major importance. In response to this declaration many countries, including ours, organized a network for the reporting, diagnosis, and treatment of influenza A (H1N1) infection. Our aim is to report the epidemiological and clinical characteristics of H1N1 infected patients, who presented to our local emergency department and compare their clinical features with patients with common influenza-like symptoms who tested negative for the H1N1 virus.
Methods: Our study included forty-five patients with influenza-like symptoms who presented to the emergency department of General Hospital of Argos from July to August 2009 and whose medical history was suggestive Clinical examination, blood and x-ray tests were conducted in all patients. In addition, pharyngeal swabs were obtained and tested for detection of H1N1 virus by real-time reverse transcription–PCR (RT-PCR) of H1N1 infection.
Results: We examined 26(57.8%) males and 19(42.2%) females, with an average age of 30 years old. Eighteen cases (40%) were positive for the H1N1 virus. The patients clinical and laboratory data are presented in Table 1. Forty-one percent of the infected patients came in contact with a verified case of H1N1 infection, 18% had recently traveled abroad and 41% had no whatsoever obvious cause of infection. This study included 3 families of whom 8 members out of 9 were found positive. Statistical analysis showed that the number of white blood cells were significantly higher in patients who tested negative for the H1N1 infection (8.910 ± 4.250 vs 6.670 ± 2.340, $P < 0.05$) compared to the H1N1 infected patients. No significant differences were found between H1N1 positive and negative patients with respect to the patients' age and gender, duration of illness, presence of fever and serious complications.
Discussion: The clinical characteristics of the new influenza A (H1N1) appeared mild and resembled those of common influenza-like illness. The patients who tested negative for the H1N1 virus had a higher mean value of white blood cells; nonetheless their number remained within the normal reference range of our laboratory. Further studies are necessary in order to verify these results.

P185

Comparison of a novel real-time RT-PCR, NS1 antigen detection and serology in early diagnosis of dengue in travelers

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Background: The increased traveling to dengue endemic regions and the numerous epidemics have led to a rise in imported dengue. The laboratory diagnosis of acute dengue requires several types of tests and often paired samples are needed for obtaining reliable results. Although several diagnostic methods are available, comparative data on their performance are lacking. The aim of the study was to compare the performance of novel methods including a novel pan-DENV real-time RT-PCR and a commercially available NS1 capture-EIA in regard to IgM detection for optimizing the early diagnosis of DENV in travelers.
Methods: A panel of 99 selected early phase serum samples of dengue patients was studied by real-time RT-PCR, NS1 antigen ELISA, IgM-EIA, IgG-IFA and cell culture virus isolation.

Table 1 (abstract P184). Patients' clinical and laboratory data

	H1N1(+) N = 18	H1N1(-) N = 27	P VALUE
Age (mean ± SD)	28.3 ± 12.8	31.2 ± 13.5	0.468
White Blood Cells (mean ± SD)	6.670 ± 2.340	8.910 ± 4.250	*0.049*
Duration of Illness (days) (mean ± SD)	5.22 ± 1.99	5.22 ± 1.99	1.000
Gender (male/female)	9/9 (50%/50%)	17/10 (63%/37%)	0.388
White Blood Cells			0.095
Normal	15 (83.3%)	22 (81.5%)	
High	0 (0%)	4 (14.8%)	
Low	3 (16.7%)	1 (3.7%)	
Neutrophilles			0.172
Normal	13 (72.2%)	17 (63%)	
High	1 (5.6%)	7 (25.9%)	
Low	4 (22.2%)	3 (11.1%)	
Lymphocytes			0.816
Normal	8 (44.4%)	13 (48.1%)	
High	4 (22.2%)	4 (14.8%)	
low	6 (33.3%)	10 (37.1%)	
Monocytes			0.393
Normal	7 (38.9%)	14 (51.9%)	
High	11 (61.1%)	13 (48.1%)	
Fever			0.420
≥38	14 (77.8%)	18 (66.7%)	
<38	4 (22.2%)	9 (33.3%)	
Hospital admission (no/yes)	15/3 (83.3%/16.7%)	26/1 (96.3%/3.7%)	0.134
Complications (no/yes)	17/1	25/2	0.807
Bronchitis	1(5.6%)	-	
Pneumonia	-	2(7.4%)	

Results: The novel real-time RT-PCR was shown specific and sensitive for detection of DENV-1-4 RNA and suitable for diagnostic use. The diagnostic rates using combination of RNA and IgM detection were higher than when using NS1 and IgM detection. The results of RNA and NS1 antigen detection disagreed in some samples that had only RNA or NS1 antigen detected.
Discussion: The diagnostic rates of early samples are higher when either RNA or NS1 antigen detection is combined with IgM-detection. Additionally, DENV RNA and NS1 antigen detection methods provide accurate diagnosis as they are DENV specific diagnostic methods unlike serological methods that are voulnerable to cross-reactions caused by related flaviviruses. Besides the differences in the RNA and NS1 detection assays, the observed discrepancy of the results in some samples that had only RNA or NS1 antigen detected, could suggest individual variation or differences in timing of these markers in patient serum.

P186

Influence of water on the circulation of the West Nile Virus in horses in Southern France

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Background: West Nile Virus (WNV) affects humans and horses, potentially causing severe neurological manifestations. Recent outbreaks of West Nile fever in horses were reported in Camargue (2000, 2004), Var (2003) and Pyrénées Orientales (2006). The circulation of this virus is strongly influenced by environmental conditions. This study aimed at explaining the circulation of WNV in horses by quantifying water bodies around equine stables using Landsat images.
Methods: A total of 135 stables were selected in three French departments (Hérault, Gard, Bouches-du-Rhône) and 1161 horses were tested by serological analysis between 2007 and 2008.
15 Landsat images (August 2006 to August 2008) were classified into 3 classes: open water, flooded vegetation and other. Surface areas of the first two classes were calculated for buffers of 2 to 5 km around each stable and for each date.
Two multivariate analyses were conducted: GLMs to identify which environmental variables were involved in the viral circulation in stables and GRMs to identify the horse variables linked to WNV circulation after retrieving the effect of the environment.
Results: The best model distinguishing 46 negative stables (no positive horse considering an error threshold of 0.15) from 61 positive stables (at least 1 positive horse) used 2 km buffers and included mean area of flooded vegetation, total number of horses present in the stable, mean area of open water and X and Y geographic coordinate. The first two variables had a positive effect and the other three a negative effect. The model predicted correctly 73% of positive stables and 71% of negative ones. At the horse level, breed, activity and age were significant. See Figure 1.
Discussion: These results can be used to target the surveillance of this human and equine disease in Southern France.

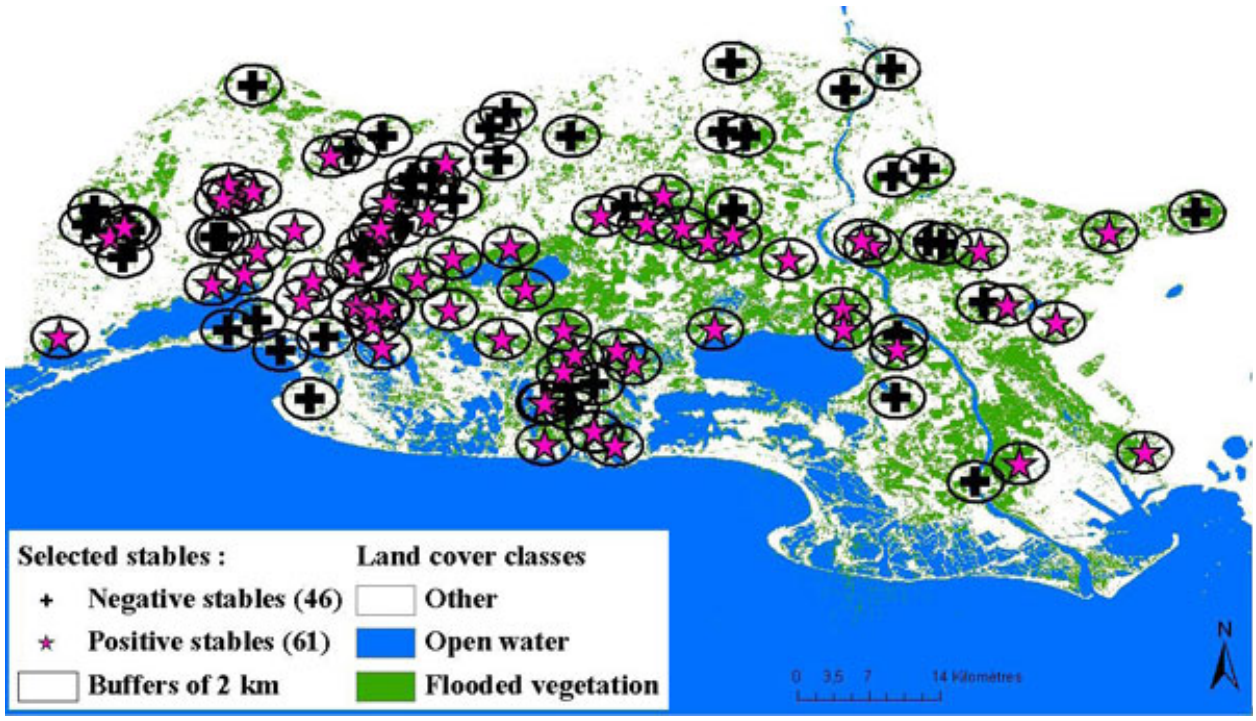

Figure 1 (abstract P186). Localisation of stables in the study zone (classification of a Landsat image of September 8th 2007).

P187

An outbreak of Influenza A (H1N1) 2009 at Mae Fah Luang University, Chiang Rai Province, northern Thailand

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Background: Since the first outbreak of a novel influenza A (H1N1) was reported in Thailand in late April 2009, new cases have gradually increased nationwide. On 2 July 2009, Mae Fah Luang University officials were notified of many suspected cases among students who sought care at the university hospital. Severe cases were referred to a district hospital and a regional hospital. On 3 July 2009, 3 positive H1N1 PCR results were reported and immediate actions were taken to investigate and control this outbreak.
Methods: At the early morning of July 3, 2009, the epidemiologist was called for the rapid increasing of the number of patients who attended at MeaFahLuangUniversityHospital, with the signs and symptoms related to the respiratory tract infection. This might been the first epidemic of the Influenza A (H1N1) virus which looks like an outbreak on the large of infected people at the same area and the same time. The investigator went to the field and conducted disease investigation and control.
On 4 July 2009, we began to investigate the outbreak by closely monitoring patients and seeking suspected cases on the University campus where 1,115 students occupied in 32 dormitories. We developed questionnaires based on MOPH ones to collect demographical and epidemiological data. Following control measures were implemented. Patients were treated with proper care (intensive care/isolated rooms). Tamiflu and influenza vaccine were stockpiled. Face masks were distributed. Housekeepers and students were provided with AVANTA® to disinfect areas of disease contamination in buildings and dormitories. Knowledge on influenza and personal hygiene were emphasized. Large social gatherings were canceled. Necessary activities were conducted in well-ventilated areas. Air conditioners were not operated. Patients and suspected cases were asked to wear face masks at all times and rest at dormitories; they were closely monitored after returning to classes.
Results: During 2-7 July 2009, 165 patients (16, 33, 36, 54, 24 and 2 cases on consecutive days) were reported with signs and symptoms related to a novel influenza A (H1N1). Major signs included fever (70.3%), cough (83.9%), muscle pain (28.9%) and diarrhea (4.2%). Mean age was 19.6 years (range 16-46 years); 68.5% were females; 58.1% and 22.9% were the 1st and 2nd year students, respectively. Most patients were students in LawSchool (16.25%) and LiberalArtsSchool (12.50%). Six severe cases were admitted at ChiangRaiRegionalHospital, PCR positive with H1N1, and administered with Tamiflu. Attack rate was 89.7/1000 population.
Discussion: Influenza A (H1N1) were rapidly transmitted in air conditioned classrooms. Urgent investigation and effective control measures could stop an outbreak within 10 days with no death reported.

P188

Adherence to the treatment of individuals with the HIV/tuberculosis co-infection: integrative review

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Background: The study it deals with integrative review whose objective was to evaluate the available evidences in literature on the factors associate with adherence to the treatment of patients with the HIV/Tuberculosis (TB) co - infection.
Methods: Were collected articles published in the period of 2002 to 2008, through LILACS and MEDLINE databases, which contained questions related to the proposed objective; the articles had been categorized in accordance with the year of publication, periodic, local of the study and factors related to the adhesion. The final sample was composed for eight articles.

Results: Five studies were developed in Brazil and others in Peru, USA and France. The factors associates to the adherence to the treatment of co-infection HIV/TB were grouped into 3 categories:
1. **Related to the individual and its style of life** – previous treatment of TB, distrust of stigma, chemical substance use, depression, social support;
2. **Related to the illness and treatment** – type of medicines regimen and complexity, collateral effect, difficulty of diagnosis of TB in patients with aids;
3. **Related to the health services** – operational problems to follow the treatment, the cost of drugs, training of the professionals, supervision of treatment (DOTS), doctor -patient relationship, distinct places for HIV and TB attendance.
Discussion: Adherence to treatment of the co - infection HIV/TB isn't a theme very explored in the cientific literature. Co - infection HIV/TB brings a strong impact on the epidemic behavior of both diseases and is responsible for increased rates of mortality, making it a challenge to public health. The patient compliance issues is influenced by multiple factors; the health professionals should be trained and be alert for the presence of life's situations that can increased his vulnerability and cause disruptions in the adherence to the treatment.

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HIV & emerging infectious diseases in mobile populations in Europe

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Background: Despite the significant progress that has been made in diagnosing and treating people with HIV in most European countries, little is known about HIV and its implications for co-infections such as TB among mobile populations in Europe, in particular among undocumented migrants and Roma. Both these groups tend to fall outside the main health care delivery networks and pathways of EU countries. Thus despite the growing statistical importance of undocumented migrants in the EU and the high internal mobility of Roma people, little is known about their vulnerability to HIV and TB and how they could best be reached with prevention and treatment interventions. In order to shed light on this emerging problem and explore possible solutions to it, we assessed HIV and TB co-infection incidence and prevalence data in mobile populations, especially undocumented migrants and Roma people in EU and EEA countries, and how countries are responding to it. In addition we investigated if and how culturally defined attitudes and behavior as well as the socio-economic and legal status of undocumented migrants and Roma influences their vulnerability to HIV-TB co-infection and their access to diagnosis, treatment and likelihood of adherence. We further examined if and to what extent this might be related to emerging patterns of multi-drug resistant TB.
Methods: We conducted a triangulation of current knowledge on this theme, including a comprehensive literature (published and unpublished) search using medical and social science journals, national and NGO reports, and we also conducted interviews of key knowledgeable people in selected high in-migration countries.
Results: Despite the growing threat of HIV-related TB and MDR-TB in the context of mobile populations, especially (but not only) clandestine migrants and Roma, there is little evidence that countries and health care organizations have taken it upon themselves to focus on these groups and develop the type of tailored interventions that are needed. Undocumented migrants and Roma are not being reached by currently available services and there is little if any evidence of HIV-TB surveillance of their disease situation.
Discussion: Unless specifically structured interventions are made available to national health care providers working in settings with a high movement of undocumented migrants and Roma, there is a danger that TB and MDR-TB will become more problematic within these groups and by extension, elsewhere as well. To this end much more emphasis should be placed on alerting and preparing primary health care personnel for this challenge and providing them with guidelines and facilities needed to reach and sustain the interest of undocumented migrants and Roma. Growing poverty will continue to drive both legal and undocumented migration, and could make TB and HIV more difficult to identify and control. Growing poverty in European countries could meanwhile extend the parameters of vulnerability within the host population.

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Serum levels of inflammatory cytokines in leprosy patients

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Background: Measure the serum cytokine levels in untreated leprosy with different parts of the spectrum of leprosy.
Methods: Concentrations of IL-4, IL-6 and IL-12 in the serum of patients were determined with a commercially available enzyme-linked immunosorbent assay (ELISA) kit (R&D Systems) according mannufactors instructions.
Results: We observed that mean of IL-4 and IL-6 were higher in lepromatous pole and IL-12 and TNF-α were higher in tuberculoid pole, but only IL-12 were statistic significantly. Our data also demonstrated that most of the patients present very lows or absent levels of TNF-α, IL-4 and IL-6.
Discussion: This is probably because leprosy is a chronic infectious disease and this profile cytokines is increase acute infections. Thus, studies of cytokine in leprosy have given us a more detailed description of the immunological parameters of the polar types of leprosy and play a significant role in classification and prognosis of this disease.

P191

Detection of ovine herpesvirus-2 in swine semen, Brazil

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Background: Malignant Catarrhal Fever (MCF) is an often lethal viral disease of susceptible biungulates from *Bovidae, Cervidae and Suidae* subfamilies. Cases of porcine MCF have been associated with direct or indirect contact with sheep, the main reservoir of ovine herpesvirus-2 (OvHV-2). Pigs are terminal hosts and not considered in virus spread. Herein, we described the possible venereal transmission by infected semen in a pig herd.
Methods: In a breeding herd, it has been observed the occurrence of neurologic signs represented by excitation associated with high fever and abortion in pregnant sows and gilts for the last seven years. The mortality rate of disease ranged up to 5% of total sow breeding stock per year. Due the elevated biosecurity pattern, the pigs have no previous contact with sheep or any other ruminant. The histopathologic findings from brainstem samples of 5 sows collected from 2004 to 2008 outbreaks were consistent with MCF-like lesions, such as severe lymphocytic meningoencephalitis and necrotizing vasculitis. DNA was extracted from these samples and PCR was performed to detect ovine herpesvirus 2 (OvHV-2), pseudorabies and porcine cytomegalovirus.
Results: The OvHV-2 DNA was detected in 4 paraffin sections of brainstem. No amplification was observed for specific PCR assays for Aujeszky's disease and porcine cytomegalovirus. To confirm the member of MCFV group, one amplicon was sequenced and showed an identity of 99% with nucleotide sequences of OvHV-2 deposited in the GenBank. In order to find possible virus carrier, blood samples were collected from pregnant sows and breeding boars, being the viral DNA detection in three boars. To investigate the potential mode of OvHV-2 transmission, nasal swabs and semen samples were collected from these infected boars. The OvHV-2 DNA was detected only in semen samples. Two out of 3 semen had > 350 copy number/2 μg of total DNA, suggesting possible shedders. Important to mention that through out this period all infected boars remained clinically healthy.

Discussion: The combination of these results together with the absence of direct or indirect contact with sheep suggests the possibility of venereal transmission from infected semen to susceptible sows and gilts.

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Multiple, concurrent or subsequent dysreactive and autoimmune disorders. Potential clinical-patogenetic correlations, and systemic infectious complications

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Background: Since mid-sixties, the association between miasthenia gravis, tymectomia for the disease control, and development of autoimmune disorders (i.e. systemic lupus erythematosus, ulcerative cholitis, rheumatoid arthritis, lichen planus), is known, while the relationship with the occurrence of systemic infectious complications is less lnown (underlying immunodeficiency, jatrogenic immunosuppression?).
Methods: A 26-y-old female patient (p), with a post-thyroiditis hypotireodism, polycystic oophoritis, and a diagnosis of myasthenia gravis posed 4 y before, developed an ileal-colonic Chron's disease treated since 2 mo with steroids.
Results: When moved to our Infectious Disease Division due to septic hyperpyrexia, not responsive to an initial empiric antimicrobial therapy, and with a presumed allergic-toxic rash, underwent a further workup. An evident leukocyotosis (WBC 23,550/μL, 88.3% neutrophils), was associated with increased ESR (86), mild hepatocytolisis, hemorrhagic conjunctivitis, and nodular erythema at lower limbs. An ultrasonographic-CT scan suggested a multifocal pyelonephritis, comfirmed by the isolation of *E. coli* at urinalysis. Combined, full-dose i.v. cefotaxime-metronidazole, was changed upon discharge with ciprofloxacin, and with the reintroduction of steroideal therapy for Chron's disease.
Discussion: A large number of predisposing conditions make subjects suffering from autoimmune/dysreactive disorders prone to develop even severe infectious complications, including the frequent immunosuppressive therapies, and the multiple immunodeficiencies detected at the time of diagnosis of these conditions. In the reported p, an autoimmune thryroiditis, a myasthenia gravis, a Crohn's disease, and an erytema nodosum were disclosed in a young female p aged 26. Consultants requested of diagnosis and management of complications, should take into consideration the heterogeneic, systemic background of each disease presentation, and their possible complications, with a proportionally elevated risk of infectious diseases, which take advantage from the chronic administration of steroids or other immunosuppressive drugs, and the unbalanced immune system, usually shifted towards a Th1 response, which tends to balance the increased Th2 activity typical of autoimmune disorders. As known, p with chronic inflammatory bowel diseases suffer from myasthenia gravis with a greater frequency *vs* the general population. From a pathogenetic point of view, the intrathymic maturation process of T-lymphocytes is altered during myasthenia gravis, while intyrathymic B-lymphocyte abnormalities may contribute to the onset of autoimmune disorders. In cases like ours, the concurrence of multiple disorders may complicate the differential diagnosis, and hamper a prompt recognition and management of potentially severe infectious complications.

Cite abstracts in this supplement using the relevant abstract number, e.g.: Manfredi: Multiple, concurrent or subsequent dysreactive and autoimmune disorders. Potential clinical-patogenetic correlations, and systemic infectious complications. *Retrovirology* 2010, **7(Suppl 1)**:P192