# Supplementary Material for

# Gene expression anti-profiles as a basis for accurate universal cancer diagnostics

Héctor Corrada Bravo[1], Vasyl Pihur[2], Matthew McCall[3], Rafael A. Irizarry[2] and Jeffrey T. Leek[2]

1. Center for Bioinformatics and Computational Biology, Department of Computer Science, University of Maryland, College Park, MD, USA.

2. Department of Biostatistics, Johns Hopkins Bloomberg School of Public Health, Baltimore, MD, USA.

3. Department of Biostatistics and Computational Biology, University of Rochester Medical Center, Rochester, NY, USA.

**Supplementary Table 1. Curated dataset of Affymetrix HGU133plus2 microarrays.**

| GEO accession number | Number of arrays | Number of normal samples | Number of tumor samples | Tissue/tumor types | Clinical annotation available |
|---|---|---|---|---|---|
| GSE10282 | 43 | 0 | 43 | skin | biopsy type;M_stage;Braf_status;Nras_status;F_stage |
| GSE10327 | 62 | 0 | 62 | medulloblastoma | Gender;Histology;Metastatic stage |
| GSE10406 | 8 | 8 | 0 | sinus_mucosa | disease state |
| GSE10445 | 62 | 0 | 62 | lung | pT2_status;age;gender;disease state |
| GSE10696 | 3 | 3 | 0 | a431_cells | Organ; Tissue; Disease; Age; Gender |
| GSE10714 | 3 | 3 | 0 | colon | |
| GSE10791 | 1 | 1 | 0 | sigmoid_colon | source |
| GSE10846 | 154 | 0 | 154 | lymph_node | Gender;Age;Tissue;Disease state;Individual;Clinical info |
| GSE10916 | 48 | 0 | 48 | skin | treatment;status;BRAF status;NRAS status |
| GSE10927 | 64 | 10 | 54 | adrenal_cortex | age;sex;side of body;Cinical characteristics;tumor diameter (cm);tumor weight (gm);Weiss Grade of tumor;Mitotic rate of tumor;Tumor stage;Adrenocortical Carcinoma cluster in our analysis;Years to last followup;dead or alive at last followup;First principal component for 33 ACCs; Conn's syndrome; |
| GSE11001 | 16 | 0 | 16 | breast | tumor cells;ER;PR;HER2;tumor stage;node stage;grade |
| GSE11045 | 3 | 3 | 0 | kidney | |
| GSE11151 | 64 | 5 | 59 | kidney | |
| GSE11281 | 3 | 3 | 0 | pbmc | |
| GSE11318 | 22 | 0 | 22 | lymph_node | Gender;Age;Tissue;Disease state;Individual;Clinical info |
| GSE11430;GSE11540 | 4 | 4 | 0 | monocytes | Age;Gender;Tissue;Cell type; |
| GSE11525 | 3 | 3 | 0 | pbmc | |
| GSE11882 | 42 | 42 | 0 | hippocampus | Individual;Brain region;Gender;Age |
| GSE12090 | 18 | 0 | 18 | kidney | |
| GSE12102 | 36 | 0 | 36 | ewing's_sarcoma | tumor status |
| GSE12141 | 1 | 0 | 1 | brain | metastasis status |
| GSE12172 | 57 | 0 | 57 | ovary | BRAF status; KRAS statusl Stage; Grade; implant history; metastatic status; |
| GSE12305 | 5 | 5 | 0 | astrocytes;u251_cells | |

**Supplementary Table 1. Curated dataset of Affymetrix HGU133plus2 microarrays.**

| | | | | | |
|---|---|---|---|---|---|
| GSE12366 | 3 | 3 | 0 | memory_b_cells | |
| GSE12460 | 28 | 0 | 28 | neuroblastoma | |
| GSE12460;GSE14880 | 28 | 0 | 28 | neuroblastoma | |
| GSE12606 | 4 | 4 | 0 | kidney | ;Gender;Date of birth |
| GSE12667 | 71 | 0 | 71 | lung | Race;Gender;TSP_Patient |
| GSE12763;GSE12790 | 29 | 0 | 29 | breast | tumor type;Her2 status |
| GSE12767 | 8 | 8 | 0 | placenta | race;karyotype status |
| GSE12875 | 1 | 1 | 0 | cd4+_t_cells | |
| GSE12902 | 17 | 0 | 17 | skin | |
| GSE12992 | 33 | 0 | 33 | medulloblastoma | Patient age at diagnosis |
| GSE13041 | 27 | 0 | 27 | glioblastoma | |
| GSE13067 | 73 | 0 | 73 | colon | tumor type |
| GSE13136;GSE13141 | 28 | 0 | 28 | neuroblastoma | Gender;Age;Stage;MYCN CNV status.;1p cnv status;11q cnv status;17q cnv status |
| GSE13294 | 127 | 0 | 127 | colon | tumor type |
| GSE13351 | 88 | 0 | 88 | lymphocyte | subtype |
| GSE13355 | 51 | 51 | 0 | skin | |
| GSE13471 | 8 | 4 | 4 | colon | |
| GSE13506 | 44 | 44 | 0 | adipose_tissue | |
| GSE13671 | 8 | 8 | 0 | breast_epithelial_cells | |
| GSE13732 | 20 | 20 | 0 | cd4+_t_cells | |
| GSE13738 | 3 | 3 | 0 | cd4+_t_cells | |
| GSE13787 | 23 | 0 | 23 | breast | Gender;subtype;Grade |
| GSE13887 | 4 | 4 | 0 | t_cells | |
| GSE13911 | 60 | 23 | 37 | stomach | tumor type |
| GSE14905 | 19 | 19 | 0 | skin | |

**Supplementary Table 1. Curated dataset of Affymetrix HGU133plus2 microarrays.**

| | | | | | |
|---|---|---|---|---|---|
| GSE2109 | 1919 | 0 | 1919 | endometrium;ovary;kidney;colon;uterus;bladder;prostate;breast;omentum;lung;renal_pelvis;thyroid;rectum;rectosigmoid;urinary_bladder;peritoneum;small_intestine;cervix;stomach;liver;esophagus;cervix_uteri;fallopian_tube;brain;pancreas;vulva;axillary_lymph_node;adrenal_gland;lymph_node;small_bowel;parotid_gland;pelvic_mass;sigmoid_colon;corpus_uteri;pelvic_lymph_node;cecum;cervical_lymph_node | Quality metric = 28S to 18S;Patient Age;Gender;Ethnic Background;Tobacco Use ;Alcohol Consumption?;Family History of Cancer?;Days from Patient Diagnosis to Excision;Pathological T;Pathological N;Pathological M;Pathological Stage;Pathological Grade;Pathological Multiple Tumors;Pathological Stage During or Following Multimodality Therapy;Primary Site;Histology;Years of Tobacco Use;Type of Tobacco Use;Presenting Symptoms;Clinical T;Clinical N;Clinical M;Clinical Stage;Clinical Grade;Clinical Multiple Tumors;Clinical Stage During or Following Multimodality Therapy;Diagnosis made by;Pathological Dukes Stage;Pathological Multiple Tumours;Pathological Staged During or Following Multimodality Therapy;PSA;# of years PSA Tested?;Cancer discovered by digital exam?;Clinical Gleason Score;Screening studies performed?;Prior Therapy;Mammogram;Number of years in which a mammogram was obtained?;Oral contraceptives?;Oophorectomy?;Fibrocystic disease?;Mammogram calcifications?;Pathological ER;Pathological PR;Pathological HER/2 Neu;Pathological Metastatic Sites;Workplace or household smokers?;Clinical Multiple Tumours;Clinical Staged During or Following Multimodality Therapy;Relapse Since Primary Treatment;Number of Years Until Relapse;Retreatment T;Retreatment N;Retreatment M;Retreatment Metastatic Sites;Retreatment Stage;Retreatment Grade;Hormonal therapy (;Clinical Question;Clinical Value;Pathological Question;Pathological Value;Have you ever had esophagitis/reflux?;Pathological Gleason Score;Retreatment ER;Retreatment PR;Retreatment HER/2 Neu;Number of years of use? (Oral contraceptives);Clinical Metastatic Sites;Have you ever been diagnosed as having HPV on your PAP smear?;Clinical Dukes Stage;Retreatment Multiple Tumors;Retreatment Stage During or |
| GSE2677 | 37 | 0 | 37 | blood | age |
| GSE2817 | 30 | 0 | 30 | glioma | |

**Supplementary Table 1. Curated dataset of Affymetrix HGU133plus2 microarrays.**

| | | | | | |
|---|---|---|---|---|---|
| GSE3526 | 133 | 133 | 0 | adipose_tissue_omental; bronchus;adipose_tissue; adrenal_gland_cortex;coronary_artery;cervix;cerebellum;heart_atrium;hippocampus;endometrium;kidney_cortex;esophagus;lung;myometrium;mammary_gland;kidney_medulla;lymph_node;pharyngeal_mucosa;ovary;oral_mucosa;stomach_cardiac;saphenous_vein;skeletal_muscle;pituitary;prostate_gland;spleen;stomach_fundus;stomach_pyloric;salivary_gland;tongue_main_corpus;thyroid;vagina;trigeminal_ganglia;tonsil;trachea;vulva;urethra | |
| GSE3744 | 47 | 7 | 40 | breast | |
| GSE4183 | 38 | 8 | 30 | colon | |
| GSE4237 | 8 | 0 | 8 | pituitary | |
| GSE4780 | 1 | 0 | 1 | brain | |
| GSE5040 | 4 | 4 | 0 | gm15851_cells | |
| GSE5081 | 5 | 5 | 0 | stomach | Age;Gender;Tissue |
| GSE5110 | 2 | 2 | 0 | skeletal_muscle | |
| GSE5460 | 95 | 0 | 95 | breast | ER;HER2;B-R grade;node status;LVI;tumor type;tumor size |
| GSE5563 | 10 | 10 | 0 | vulva | Gender |
| GSE5675 | 38 | 0 | 38 | pilocytic_astrocytoma | |
| GSE5764 | 4 | 0 | 4 | breast_lobular_cells | |

**Supplementary Table 1. Curated dataset of Affymetrix HGU133plus2 microarrays.**

| | | | | | |
|---|---|---|---|---|---|
| GSE5787 | 33 | 0 | 33 | cervix | FIGO;Histology;Grade;hp5;HGB;ifp;age;tumsizeMR;ResponseToTherapy |
| GSE5993 | 6 | 0 | 6 | cervix | Age |
| GSE6004 | 4 | 4 | 0 | thyroid | Gender; Age |
| GSE6283 | 6 | 6 | 0 | chorion_villus_cells | tissue;Age;gender;karyotype |
| GSE6532 | 80 | 0 | 80 | breast | |
| GSE6565 | 10 | 10 | 0 | fetal_cartilage | |
| GSE6573 | 2 | 2 | 0 | placenta;adipose_tissue | |
| GSE6764 | 23 | 1 | 22 | liver | |
| GSE6791 | 22 | 4 | 18 | cervix | Case;Anatomical sites;gender;age;Tumor Stage |
| GSE7224 | 9 | 9 | 0 | gingival_epithelium;tonsil_epithelium | |
| GSE7305 | 10 | 10 | 0 | endometrium | Menstrual phase - Follicular;Menstrual phase - Luteal |
| GSE7307 | 69 | 69 | 0 | ovary;breast;synovial_membrane;penis;colon;cerebellum;kidney;prostate_gland;placenta;salivary_gland;skeletal_muscle;testes;stomach;trachea;skin;thyroid;cecum;adipose_tissue;endometrium;myometrium;mammary_gland;hippocampus;urethra;deltoid_muscle | Tissue/Cell Line [C];Disease/Normal or Treatment [C];Gender;Disease type |
| GSE7508 | 3 | 3 | 0 | jurkat_cells | wild-type Jurkat cells;wild type Jurkat cells |
| GSE7553 | 22 | 4 | 18 | skin | tumor size |
| GSE7562 | 2 | 2 | 0 | a431_cells | |
| GSE7586 | 10 | 10 | 0 | placenta | Parity;StudyID;Inflammation;ChronicPM;PM status;%parasitemia;InfantSex;MaternalAge;Birthweight(kg) |
| GSE7696 | 70 | 0 | 70 | brain | patient; age; gender; treatment; survival status; survival time in months |
| GSE7753 | 25 | 25 | 0 | pbmc | |

**Supplementary Table 1. Curated dataset of Affymetrix HGU133plus2 microarrays.**

| | | | | | |
|---|---|---|---|---|---|
| GSE7832 | 5 | 5 | 0 | airway_epithelial_cells | Age;Sex;Ethnic group;Smoking status |
| GSE7832;GSE8545 | 1 | 1 | 0 | airway_epithelial_cells | Age;Sex;Ethnic group;Smoking status |
| GSE7835 | 3 | 3 | 0 | u251_cells | |
| GSE7904 | 1 | 0 | 1 | breast | |
| GSE8050 | 2 | 2 | 0 | kidney | age;gender;tissue mass |
| GSE8671 | 30 | 7 | 23 | rectum_mucosa;sigmoid_colon_mucosa | Tissue;Location;Size;Patient ID;CEL Filename |
| GSE8977 | 10 | 6 | 4 | breast_stroma | |
| GSE9089;GSE9196 | 5 | 5 | 0 | cd31+_cells | |
| GSE9090;GSE9196 | 5 | 5 | 0 | cd49a+_cells | |
| GSE9171;GSE9200 | 13 | 0 | 13 | glioblastoma | |
| GSE9195 | 68 | 0 | 68 | breast | samplename;series;age;grade;size;er;pgr;node;t.rfs;e.rfs;t.dmfs;e.dmfs;treatment |
| GSE9254 | 10 | 10 | 0 | sigmoid_colon;rectum;cecum | |
| GSE9438 | 31 | 0 | 31 | brain | ER;PR |
| GSE9489;GSE9493 | 13 | 13 | 0 | kidney | |
| GSE9599 | 35 | 0 | 35 | pancreas | |
| GSE9624 | 6 | 6 | 0 | omental_adipose_tissue | Tissue;Group;Gender;Age;z score BMI |
| GSE9829;GSE9843 | 48 | 0 | 48 | liver | BCLC_staging ;Age ;Sex ;Code ;Class ;AFP ;TP53 mutation ;CTNNB1 mutation ;Immunohistochemistry |
| GSE9834 | 3 | 3 | 0 | astrocytes | Cell strain;Passage;Growth Medium; Insulin, 1.25 ml; Ascorbic Acid, 0.5 ml; GA-1000, 0.5 ml, L-Glutamine, 5.0 ml; FBS, 15 ml.;Grown at 37C in 5%CO2, seeded at 5000 cells/cm2;Untreated |
| GSE9891;GSE9899 | 221 | 0 | 221 | ovary;peritoneum;fallopian_tube | Primary.Site ;Type ;Subtype ;StageCode ;Consolidated.Grade |
| GSE9984 | 12 | 12 | 0 | placenta | first trimester;second trimester;term placenta |
| total | 4826 | 688 | 4138 | | |

**Supplementary Table 2. Gene ontology category enrichment analysis of hyper-variable genes in cancer.**

| Gene ontology ID | Odds ratio | Expected count | Observed count | Category size | Adjusted P-value | Gene ontology term | Gene symbols |
|---|---|---|---|---|---|---|---|
| GO:0007275 | 1.80 | 99.44 | 155 | 1873 | 7.067E-06 | multicellular organismal development | NOX1;COL10A1;IBSP;SPRR1B;DCN;DCN;SOX11 ;SPRR1A;FN1;PCDHB2;ONECUT2;SFTPB;SOX11;SOX11;ONECUT2;CSPG5;FGF20;DCN;LPPR1;HES6;HMGA2;CXCL17;CTNND2;ODAM;HMGA2;EN2;MYT1;PCDHB3;COL8A1;ARX;MYT1L;HMGA2;S100A7;FGF14;LUM;SCN2A;DKK4;PCDHB6;C14orf39;CHL1;GABRA5;DCN;PLEKHB1;FN1;TNFRSF17;FGF14;KRT14;CTNND2;GRIN2A;NEFH;SLC1A3;SH3GL3;LY6H;CACNA1D;FGFR3;IGFBP5;PAX3;ITGB6;REG3A;GPR98;LY6D;UGT8;NGFRAP1;SHISA2;KRT5;COL3A1;SPRR1A;FREM2;FAM5C;BNC1;SEMA6A;ACSL6;ARSE;GART;TLL2;USH2A;C1S;FGF14;HTN3;COL10A1;INA;COL3A1;COL15A1;CHRM3;SULF1;GRIN2A;COL3A1;COL6A1;FHL1;PKP1;SLC1A3;NDN;KLK7;DOK5;SOHLH2;PROM1;KIF5C;KLK6;MST1R;SERPINB5;PCDHB5;LEFTY1;PTPN22;PCDHB16;KRT17;IGSF9;NHLH2;INSM1;COL6A3;NRTN;CALML5;ERBB3;ERBB3;OLFM3;COMP;KRT15;COL12A1;SOX3;PTN;TFAP2B;MMP2;COL1A2;COL9A3;SLITRK5;ERBB4;ASS1;ENAM;SEMA6A;FN1;NOX1;KLK5;ELAVL3;COL5A1;POSTN;JPH1;FN1;CELSR3;FN1;SFTPB;CBS;MPPED2;CRB1;SLITRK6;DDX25;PDZRN3;CD3D;APP;CBS;CHRNA1;TIMP3;FN1;ECE2;PTPN22;E2F5;CSTA;SULF1;MPPED2;TYR;OBSL1;SIX6;TFAP2B;SHOX2;OPCML;SULF2;COL5A1;NTM;NTM;CHL1;POSTN;L1CAM;PCSK9;OLFM3;NPNT;ITGB6;FUT9;LRP2;PTN;OBSL1;DCX;MAEL;DCX;UNC5A;NR2F1;KRT17;GPR65;FN1;SULF1;MYT1L;PTPRZ1;FMOD;CD72;GHR;DYX1C1;PCDHB10;VAV3;NDP;FGF13;NEFH;EPHB1;KLK7;VAV3;ANO1 |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| GO:0007155 | 2.66 | 21.29 | 50 | 387 | 1.752E-05 | cell adhesion | COL11A1;COL11A1;IBSP;COL11A1;FN1;LGALS4;MUC4; MUC5B;MUC16;MSLN;CLCA2;MUC4;CHL1;FN1;ITGB6;FERMT1;LY6D;CLC A2;MUC4;COL3A1;MEGF10;CLCA2;CXCL12;FERMT1;COL3A1;COL15A1;C OL3A1;COL6A1;PKP1;CCL11;MUC5AC;SLAMF7;COL6A3;TPBG;COMP;COL 12A1;FN1;MTSS1;COL5A1;POSTN;FN1;FN1;THBS2;CLCA2;APP;SPON1;FN 1;ITGBL1;HOXD3;FLRT3;OPCML;COL5A1;NTM;NTM;CHL1;POSTN;SELL;NP NT;ITGB6;PERP;NID2;EFS;WISP1;TRIP6;FLRT3;ANTXR1;ISLR;FN1;AZGP1;C LEC4A;CD72;MYBPC2 |
| GO:0051674 | 2.24 | 33.48 | 67 | 588 | 2.937E-05 | localization of cell | NOX1;NKX2-1;MMP1;FN1;ONECUT2;ONECUT2; DNAH5;S100A2;ARX;S100A7;ASCL1;SIX1;POU4F1;FOXD1;FN1;ASCL1;FOX E1;FAP;IGFBP5;GJA1;CALCA;LAMA3;FERMT1;SEMA3A;PTPRM;FOXD3;CX CL12;FERMT1;TGFB2;CDH2;NDN;POU3F2;CCL11;COL1A1;NRTN;TREM1;C ALCA;PDGFRA;NKX2-3;MSN;COL1A2;ERBB4;NKX2- 1;FN1;NOX1;COL5A1;MMP12;CAV1;FN1;SEMA3A;FN1;RARRES2;SFRP1;F N1;TNFSF11;CALCA;BAMBI;GAS6;IL8;SERPINE2;SRPX2;COL5A1;COL1A1;L 1CAM;CTHRC1;VIL1;SELL;RASGEF1A;ASTN1;POU4F1;SFRP2;SST;TRIP6;NR 2F1;FN1;CD48;PTPRM;S100A14;DYX1C1;VAV3;OLR1;PRSS3;VAV3;LBP;EN PP2;ARAP3 |
| GO:0009887 | 4.52 | 5.65 | 21 | 103 | 8.886E-05 | organ morphogenesis | NKX2-1;DCN;CALB1;DCN;CALB1;ONECUT2;SFTPB; ONECUT2;DCN;DCN;LY6H;PAX3;PAX6;SEMA6A;FOXA2;PAX6;FHL1;SYCP2; WT1;COMP;SOX3;NKX2- 1;SEMA6A;SFTPB;PITX2;E2F5;SIX6;PBX1;NOTCH2;IRX2 |
| GO:0040011 | 2.01 | 37.83 | 69 | 675 | 3.806E-04 | locomotion | NOX1;CXCL11;CXCL11;FOXG1;MMP1;FN1;DNAH5; CXCL17;CCL18;S100A2;CCL18;ARX;ASCL1;ETV4;CXCL6;JPH3;CHL1;SIX1;P OU4F1;FOXD1;FN1;ASCL1;FOXE1;FAP;CACNA1D;CXCL9;GJA1;CALCA;FER MT1;BMP7;COL3A1;FOXD3;SEMA6A;FERMT1;COL3A1;COL3A1;COL6A1;C DH2;NDN;POU3F2;KIF5C;COL6A3;NRTN;TREM1;CALCA;NKX2- 3;MSN;COL1A2;COL9A3;SEMA6A;FN1;NOX1;LHX2;COL5A1;MMP12;CAV 1;FN1;FN1;APP;FN1;TNFSF11;CALCA;BAMBI;GAS6;COL5A1;CHL1;L1CAM; ATP2B2;CCL28;CTHRC1;SELL;RASGEF1A;ASTN1;POU4F1;DCX;CXCL5;DCX; UNC5A;NR2F1;HOXA1;SERPIND1;FN1;CD48;CD72;DYX1C1;VAV3;OLR1;P RSS3;EPHB1;VAV3 |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| GO:0030199 | 9.86 | 1.54 | 10 | 27 | 4.918E-04 | collagen fibril organization | COL11A1;COL11A1;COL11A1;LUM;COL3A1;COL3A1; TGFB2;COL3A1;COL1A1;COL12A1;COL1A2;LOX;COL5A1;COL5A1;COL1A1; LOX;SFRP2 |
| GO:0071842 | 4.44 | 4.65 | 17 | 82 | 9.589E-04 | cellular component organization at cellular level | NOX1;COL11A1;COL11A1;IBSP;COL11A1;TNFRSF11B; LUM;TGFB2;TNFRSF11B;SERPINB5;MUC5AC;WT1;PDGFRA;COL12A1;ERB B4;NOX1;POSTN;PDZRN3;CHRNA1;POSTN;SFRP2;ABI3BP |
| GO:0006928 | 2.16 | 23.34 | 46 | 416 | 2.467E-03 | cellular component movement | NOX1;MMP1;FN1;DNAH5;S100A2;ARX;ASCL1;SIX1; POU4F1;FOXD1;FN1;ASCL1;FOXE1;FAP;DNALI1;GJA1;CALCA;FERMT1;FO XD3;FERMT1;CDH2;NDN;POU3F2;MST1R;SERPINB5;NRTN;TREM1;CALCA ;NKX2- 3;MSN;COL1A2;FN1;NOX1;MTSS1;COL5A1;MMP12;CAV1;FN1;FN1;FN1;T NFSF11;CALCA;BAMBI;GAS6;COL5A1;L1CAM;CTHRC1;SELL;RASGEF1A;AS TN1;POU4F1;NR2F1;FN1;CD48;DNALI1;DYX1C1;VAV3;OLR1;PRSS3;VAV3; DNAH9 |
| GO:0007156 | 3.34 | 6.89 | 20 | 121 | 3.585E-03 | homophilic cell adhesion | PCDHB2;DSG3;PCDHB3;DSC3;PCDHB6;CDH6;DSG3 ;FREM2;PTPRM;CDH6;DSC3;CDH2;PCDH8;PCDHB5;PCDHB7;PCDH19;PCD HB16;PCDH7;CELSR3;FAT1;L1CAM;PCDHA9;PTPRM;CDH6;PCDHB10 |
| GO:0009954 | 7.93 | 1.59 | 9 | 28 | 3.585E-03 | proximal/distal pattern formation | DLX2;DLX1;EN1;TP63;HOXC10;HOXA9;SP8;PBX1;IRX2 |
| GO:0007267 | 2.52 | 13.64 | 31 | 245 | 3.585E-03 | cell-cell signaling | CXCL11;SIX3;CXCL11;SOX2;PTHLH;NKX2-1;PTHLH; WISP3;TSHR;FOXA1;TSHR;FGF20;PTHLH;CCL18;CCL18;FGF14;CXCL6;FOX A1;SIX1;CEACAM6;FGF14;CEACAM6;CXCL9;GJA1;CALCA;SH2D1A;FGF14; TGFB2;PCDH8;STC2;CALCA;NKX2- 1;CRB1;FAT1;ECE2;SOX2;CALCA;AR;SH3KBP1;SFRP2;CXCL5;WISP1;TSHR; NDP;FGF13 |
| GO:0050910 | 27.78 | 0.46 | 5 | 8 | 5.995E-03 | detection of mechanical stimulus involved in sensory perception of sound | COL11A1;COL11A1;COL11A1;SOX2; GPR98;SOX2;CHRNA9;ATP2B2 |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| GO:0022008 | 1.92 | 30.60 | 54 | 552 | 6.170E-03 | neurogenesis | LHX8;ONECUT2;ONECUT2;FGF20;PCSK1;EN2;ARX; ETV4;EN1;CHL1;GRIN2A;SLC1A3;CACNA1D;FGFR3;GJA1;GPR98;COL3A1; PTPRM;SEMA6A;ACSL6;USH2A;COL3A1;GRIN2A;COL3A1;COL6A1;SLC1A3 ;PROM1;KIF5C;COL1A1;IGSF9;PAX7;COL6A3;NRTN;HOXC10;OLFM3;COL1 A2;COL9A3;SLITRK5;SEMA6A;LHX2;COL5A1;CELSR3;CRB1;SLITRK6;APP;O PCML;FZD8;COL5A1;COL1A1;NTM;NTM;CHL1;L1CAM;ATP2B2;PCSK9;CT HRC1;OLFM3;DCX;DCX;UNC5A;NR2F1;HOXA1;FZD8;PTPRM;PTPRZ1;CD7 2;DYX1C1;EPHB1 |
| GO:0006958 | 6.84 | 1.77 | 9 | 31 | 6.975E-03 | complement activation, classical pathway | IGKV4-1;IGKC;C1S;MBL2;CRP;C4BPB;C7;C2;CFI |
| GO:0016339 | 7.87 | 1.42 | 8 | 25 | 8.102E-03 | calcium-dependent cell-cell adhesion | PCDHB2;PCDHB3;PCDHB6;CDH2;PCDHB5;PCDHB7; PCDHB16;PCDHB10 |
| GO:0002253 | 5.58 | 2.28 | 10 | 40 | 9.482E-03 | activation of immune response | IGKV4-1;IGKC;CFHR5;C1S;MBL2;CRP;C4BPB;C7;C2;CFI |
| GO:0045665 | 6.02 | 1.94 | 9 | 34 | 1.281E-02 | negative regulation of neuron differentiation | SOX2;FOXG1;NKX2-2;ASCL1;ASCL1;BMP7;FOXA2;ID4; SOX2;PBX1;ASPM |
| GO:0048706 | 8.41 | 1.19 | 7 | 21 | 1.418E-02 | embryonic skeletal system development | DLX1;DLK1;COL1A1;PAX7;HOXA9;COL1A1; HOXD1;PBX1 |
| GO:0050982 | 11.12 | 0.85 | 6 | 15 | 1.418E-02 | detection of mechanical stimulus | COL11A1;COL11A1;COL11A1;SOX2;GPR98;TRPA1;SOX2;CHRNA9;ATP2B2 |
| GO:0072376 | 5.07 | 2.45 | 10 | 43 | 1.420E-02 | protein activation cascade | IGKV4-1;IGKC;CFHR5;C1S;MBL2;CRP;C4BPB;C7;C2;CFI |
| GO:0001501 | 3.44 | 5.01 | 15 | 90 | 1.420E-02 | skeletal system development | COL10A1;TNFRSF11B;EN1;COL3A1;DLL3;ARSE;COL10A1;COL3A1;COL3A1 ; TNFRSF11B;TP63;HOXC10;COMP;COL12A1;TEAD4;COL1A2;POSTN;SHOX 2;POSTN;ANKH |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| GO:0009605 | 1.65 | 48.13 | 74 | 856 | 1.420E-02 | response to external stimulus | CXCL11;CXCL11;NKX2-1;FOXG1;PCSK1;CXCL17;CCL18;CCL18;ARX;S100A7;TNFRSF11B;MMP3;ASCL1;ETV4;CXCL6;CHL1;FOXD1;PLEKHB1;ASCL1;CACNA1D;CXCL9;CALCA;BMP7;COL3A1;SDR16C5;PTPRM;FOXD3;SEMA6A;F12;TNFSF4;CXCL12;COL3A1;TGFB2;FOXA2;COL3A1;COL6A1;TNFRSF11B;CPB2;TSPAN8;CCL11;KIF5C;COL6A3;STC2;NRTN;CALCA;INHBB;COL1A2;COL9A3;CNR1;NKX2-1;ASS1;SEMA6A;LHX2;COL5A1;CBS;RARRES2;SCGB1A1;APP;SFRP1;CBS;SERPINA7;TNFSF11;CALCA;IL8;SERPINE2;COL5A1;CHL1;L1CAM;CCL28;PCSK9;HRK;LRP2;SFRP2;DCX;SST;CXCL5;DCX;UNC5A;HOXA1;SERPIND1;PTPRM;CD72;RBP1;S100A14;GHR;EPHB1;LBP;ENPP2;CCNE1 |
| GO:0007586 | 4.50 | 2.96 | 11 | 52 | 1.439E-02 | digestion | AKR1B10;CTSE;TFF3;CHRM3;AKR1C2;TFF2;MUC5AC;PRSS2;AKR1C2;SST;CYP39A1;PRSS3 |
| GO:0030574 | 7.79 | 1.25 | 7 | 22 | 1.585E-02 | collagen catabolic process | MMP1;MMP13;MMP3;KLK6;MMP2;PRSS2;MMP7 |
| GO:0048856 | 2.10 | 17.60 | 34 | 359 | 1.722E-02 | anatomical structure development | COL10A1;PCDHB2;CSPG5;LPPR1;HES6;MYT1;PCDHB3;MYT1L;FGF14;SCN2A;PCDHB6;FGF14;NEFH;SH3GL3;UGT8;DLK1;FAM5C;ACSL6;ARSE;C1S;FGF14;COL10A1;CHRM3;ARNT2;DOK5;PCDHB5;PCDHB16;NHLH2;COL12A1;ELAVL3;MPPED2;CD40;MPPED2;HOXD1;FUT9;MYT1L;DYX1C1;PCDHB10;FGF13;NEFH |
| GO:0022612 | 6.12 | 1.70 | 8 | 30 | 1.874E-02 | gland morphogenesis | CDKN2A;CDKN2A;PTHLH;PTHLH;PTHLH;ETV4;SFRP4;CCL11;ERBB3;ERBB3;NKX2-3;CAV1;SFRP4 |
| GO:0044259 | 4.52 | 2.68 | 10 | 47 | 2.377E-02 | multicellular organismal macromolecule metabolic process | MMP1;MMP13;MMP3;COL3A1;COL3A1;COL3A1;KLK6;COL1A1;MMP2;PRSS2;COL5A1;MMP7;COL5A1;COL1A1 |
| GO:0016540 | 22.19 | 0.40 | 4 | 7 | 2.795E-02 | protein autoprocessing | PCSK1;F12;KLK6;PCSK9 |
| GO:0032355 | 3.46 | 4.32 | 13 | 76 | 2.795E-02 | response to estradiol stimulus | TFF1;FOXA1;MMP3;FOXA1;BMP7;SFRP4;ARNT2;CCND2;PDGFRA;ASS1;SFRP4;SLC6A1;GHR;SOCS2;CCNE1 |

**Supplementary Table 5: Cancer-specific analysis of tissue specificity and hyper-variability.** We computed the proportion of hyper-variable genes that are specific to each of the seven tissues with sufficient samples of both normal and cancer. The vast majority of hyper-variable genes are not tissue-specific in these tissues (column 2). However, there is enrichment of hyper-variability in tissue-specific genes (columns 3 and 4). We repeated the same analysis for differentially expressed genes, but found no enrichment in the set of tissue-specific genes.

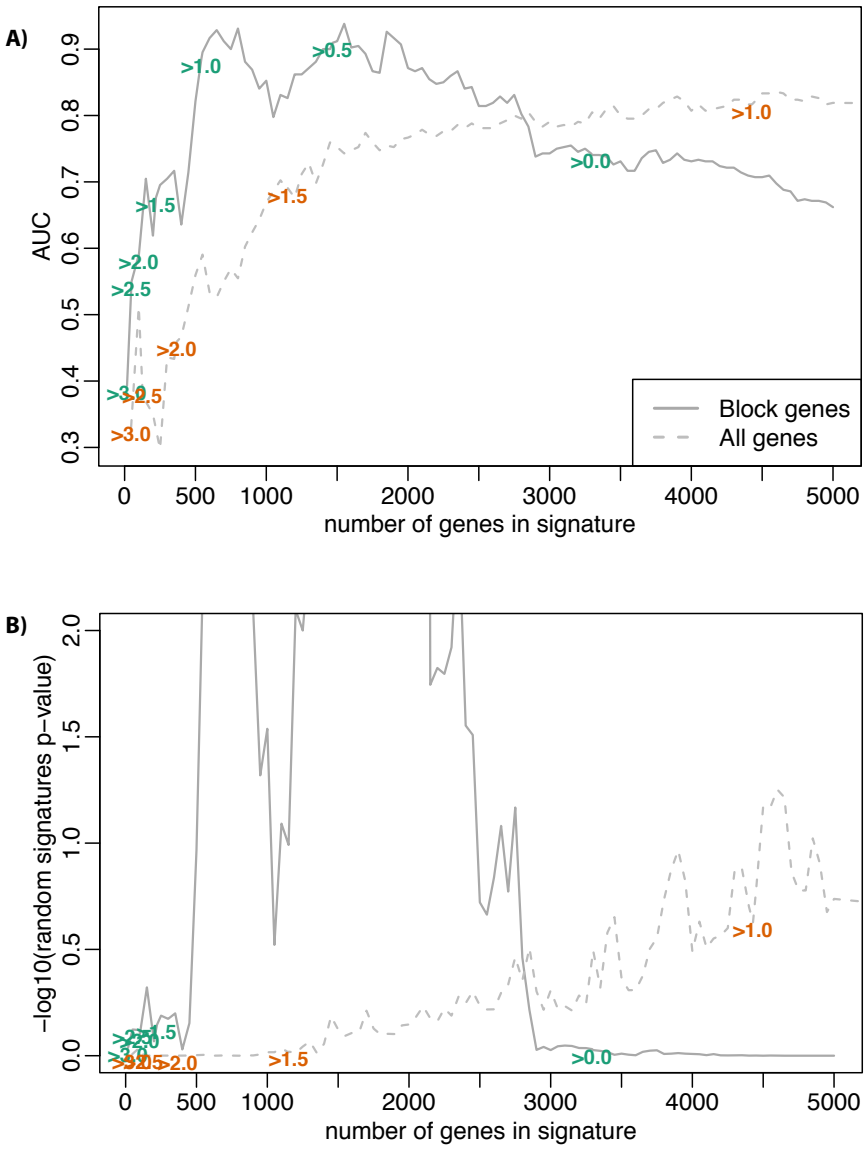
| | Number of hyper-variable genes | Pct. that are tissue-specific | OR | P-value |
|---|---|---|---|---|
| **adrenal cortex** | 14293 | 5.11 | 5.02 | 1.39E-156 |
| **colon** | 8882 | 3.25 | 4.23 | 5.89E-65 |
| **endometrium** | 5808 | 1.05 | 2.08 | 1.84E-06 |
| **kidney** | 4290 | 3.19 | 5.73 | 3.48E-48 |
| **skin** | 9590 | 1.89 | 2.96 | 7.04E-27 |
| **stomach** | 4906 | 1.35 | 2.39 | 7.67E-09 |
| **vulva** | 5323 | 3.57 | 5.33 | 5.19E-60 |

| | Number of down-regulated genes | Pct. that are tissue-specific | OR | P-value |
|---|---|---|---|---|
| **adrenal cortex** | 1790 | 2.46 | 1.17 | 3.16E-01 |
| **colon** | 4218 | 0.97 | 0.80 | 1.84E-01 |
| **endometrium** | 6560 | 0.55 | 0.97 | 9.30E-01 |
| **kidney** | 4826 | 0.64 | 0.81 | 3.03E-01 |
| **skin** | 7754 | 0.67 | 0.75 | 4.68E-02 |
| **stomach** | 2427 | 0.45 | 0.70 | 2.96E-01 |
| **vulva** | 5221 | 0.84 | 0.86 | 3.73E-01 |

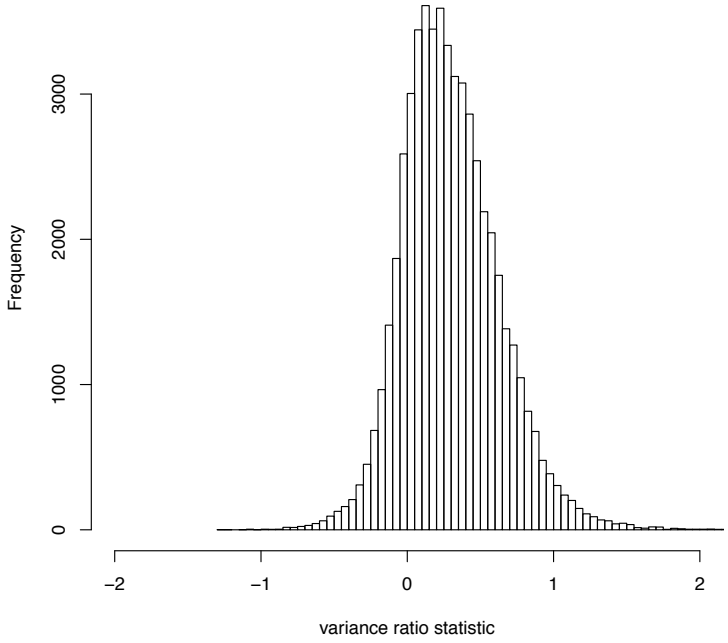
| | Number of up-regulated genes | Pct. that are tissue-specific | OR | P-value |
|---|---|---|---|---|
| **adrenal cortex** | 2543 | 1.93 | 0.90 | 5.26E-01 |
| **colon** | 7558 | 0.98 | 0.80 | 7.62E-02 |
| **endometrium** | 5437 | 0.59 | 1.05 | 7.75E-01 |
| **kidney** | 6509 | 0.66 | 0.83 | 2.92E-01 |
| **skin** | 8625 | 0.93 | 1.09 | 4.86E-01 |
| **stomach** | 5118 | 0.47 | 0.72 | 1.17E-01 |
| **vulva** | 4687 | 1.11 | 1.16 | 3.11E-01 |

**Supplementary Table 6. Anti-profile signatures are not explained by pathological tumor stage or grade heterogeneity.** For each of the leave-one-tissue-out experiments reported in Figure 4, we used available annotated pathological tumor stage or grade to find genes that are differentially expressed across clinical stages or grades. The first column in the table indicates the number and percentage of genes that show significant pathological stage or grade differences (FDR<0.1). The second column indicates the number and percentage of these genes included in the 100-gene anti-profile used to classify samples in the leave-one-tissue-out experiment. The last two columns are odds ratio and P-value for a Fisher exact test for enrichment of differentially expressed genes in the anti-profile. For adrenal cortex, stomach and vulva there are very few differentially expressed genes (22, 2 and 4 respectively).

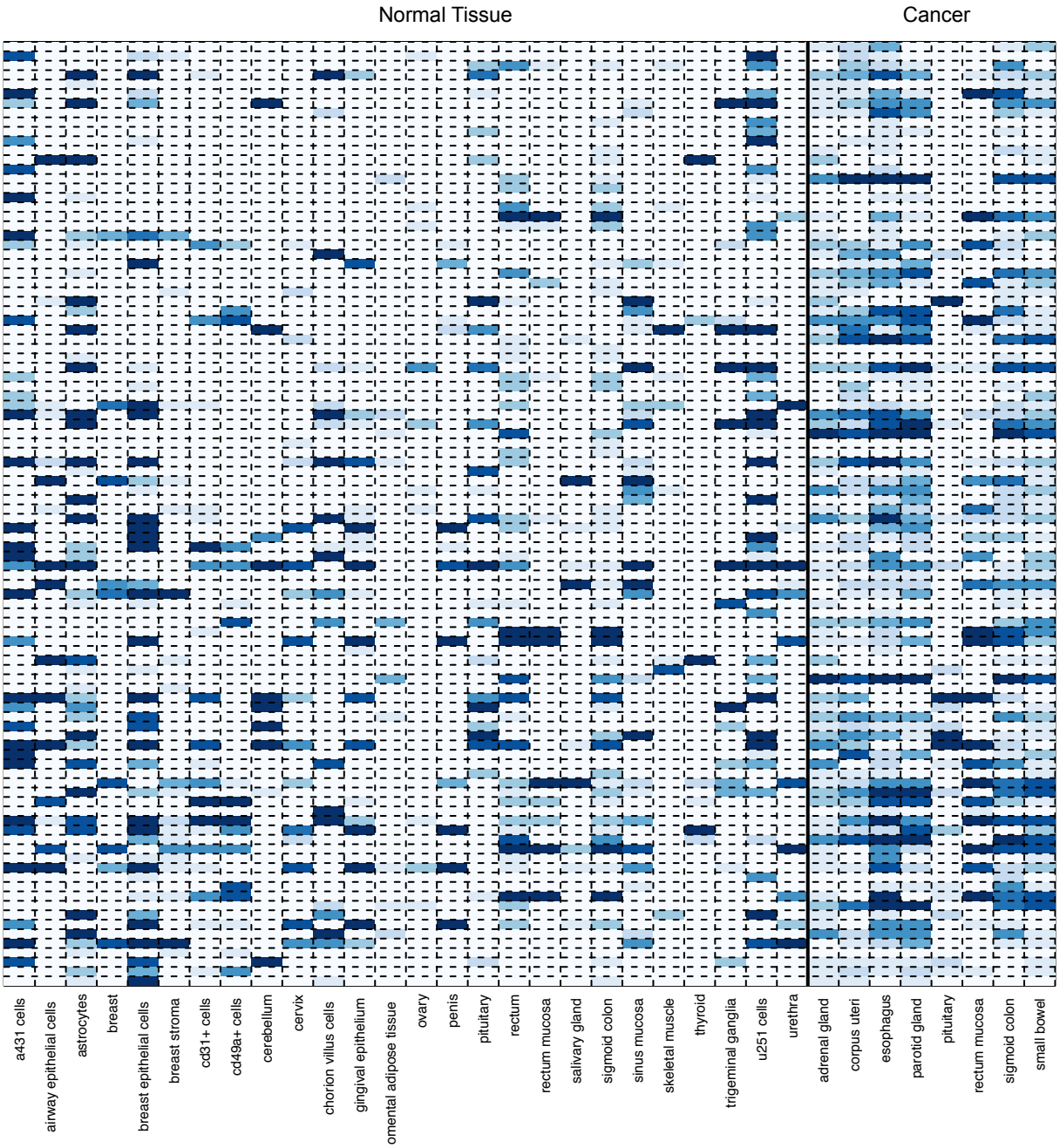
| | Number of differentially expressed genes | Number in anti-profile | OR | P-value |
|---|---|---|---|---|
| **adrenal cortex** | 22 (0.040%) | 1 (4.545%) | 26.2 | 0.040 |
| **colon** | 9271 (16.976%) | 29 (0.313%) | 2.0 | 0.003 |
| **endometrium** | 4377 (8.015%) | 15 (0.343%) | 2.0 | 0.016 |
| **kidney** | 8183 (14.984%) | 13 (0.159%) | 0.8 | 0.675 |
| **skin** | 11482 (21.024%) | 22 (0.192%) | 1.1 | 0.806 |
| **stomach** | 2 (0.004%) | 0 (0.000%) | 0.0 | 1.000 |
| **vulva** | 4 (0.007%) | 0 (0.000%) | 0.0 | 1.000 |



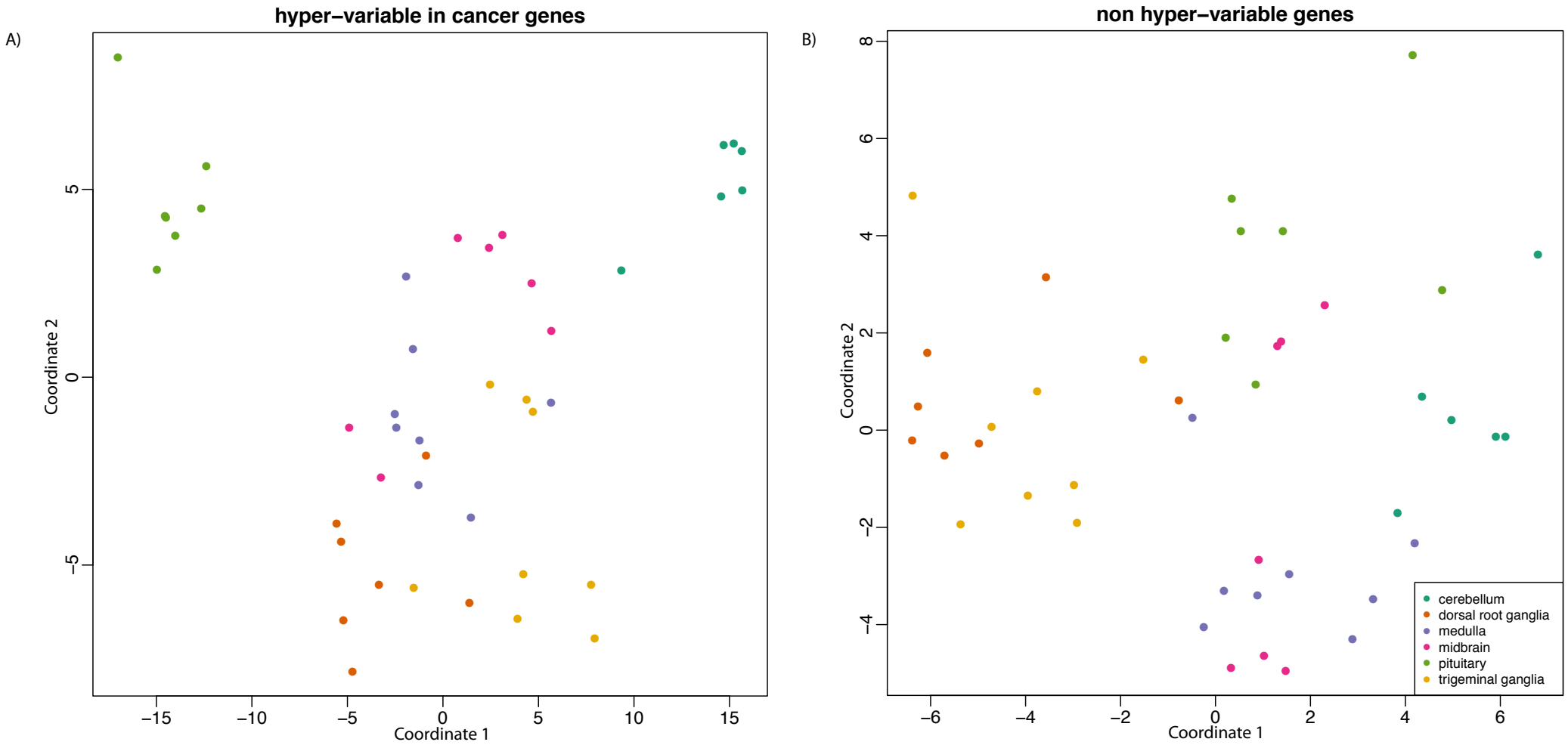

**Supplementary Figure 1. Sensitivity and significance of colon cancer peripheral blood signature.** (A) AUCs obtained by signatures of varying size. The solid line corresponds to signatures built using only genes inside blocks of cancer methylation changes [23], while the dashed line corresponds to all genes. Cutoffs for the log2 variance ratio statistic are indicated along the curve. The signature reported in the manuscript includes genes in blocks (solid line) with cancer variability at least twice that of normal (>1) which obtained AUC of 0.89. Similar AUCs are obtained with signatures with about 500-2000 genes inside blocks. This highlights the importance of including genes inside blocks of methylation changes in colon cancer. (B) The proportion of random signatures of corresponding size obtaining an AUC greater than or equal to the anti-profile. Results show that significantly high AUC was obtained by signatures that include about 500-2000 of the top hyper-variable genes inside methylation blocks and therefore the results discussed in the main text are not very sensitive to signature size. For signatures greater than 3000 genes, the anti-profiles built from all genes perform better than anti-profiles built from only genes inside blocks of cancer methylation changes. This is due to the fact that while hyper-variable genes are enriched in these blocks 2,047 of the 5,339 genes inside these blocks are not hyper-variably expressed, and their inclusion degrades anti-profile performance.



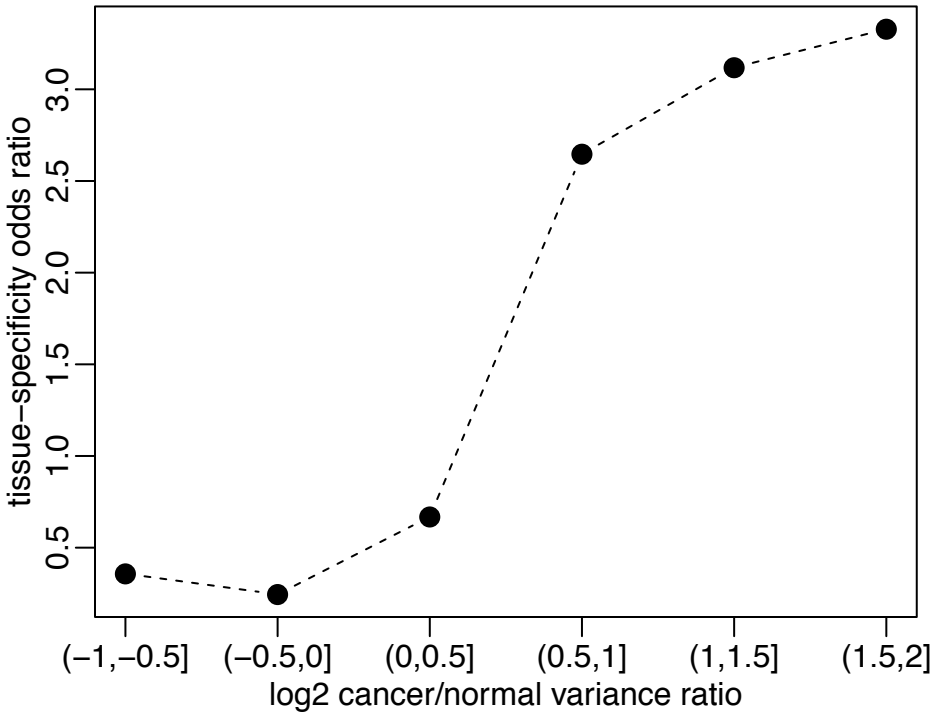

**Supplementary Figure 2. Histogram of log2 cancer/normal variance ratio.** We computed a statistic to determine general hyper-variability in cancer as discussed in Methods and Materials. Here we plot the histogram of this statistic across the ~55,000 probesets in the HGU133plus2 microarray. While a majority of probes show increased variability in cancer (log2 ratio >0), relatively few genes (1,456) show hyper-variability (log2 ratio >1).



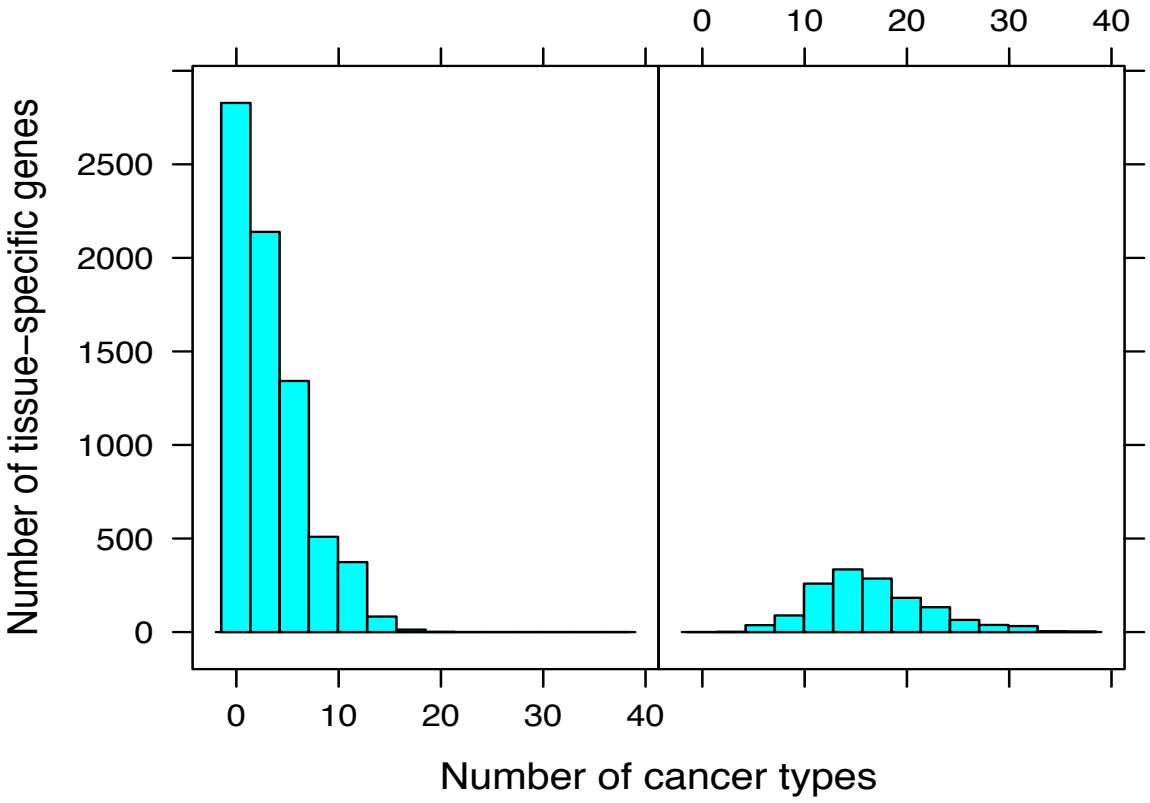

**Supplementary Figure 3. Genes with consistent hyper-variability across cancer types.** A plot of the 100 genes that most consistently show hyper variability across cancer types, similar to Figure 3A, on tissue types and cancers not used to define hyper-variability. As for the samples in Figure 3A, we observed that for the majority of genes, the percentage of samples in each normal tissue type outside normal range is close to either 0% (most tissues) or 100% (the small number of tissues for which the gene is tissue-specific). We also observe that in cancer, percentages are consistently away from 0% or 100%, indicating high variability.



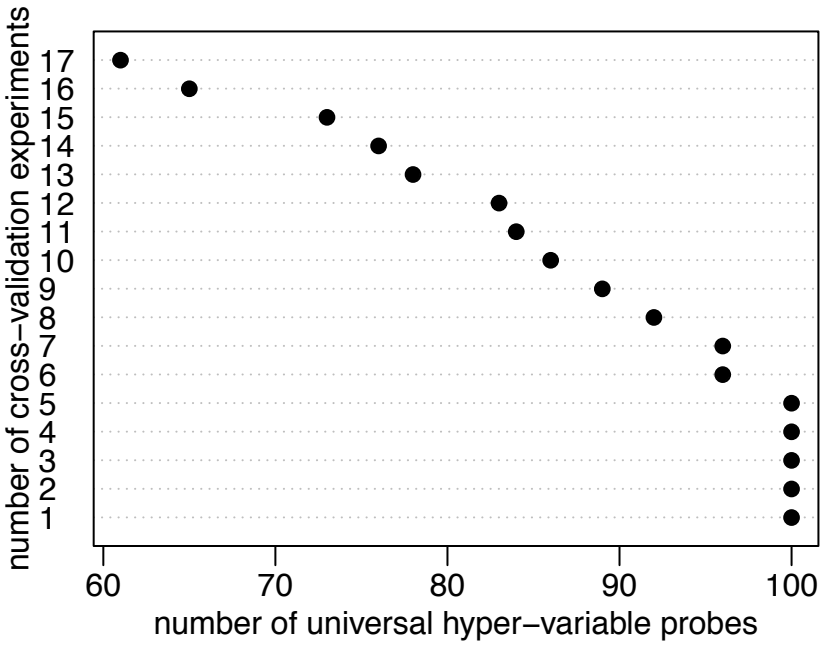

**Supplementary Figure 4. Hyper-variable genes in cancer distinguish normal samples from different brain regions.** (A) Multi-dimensional scaling of gene expression of hyper-variable genes in cancer from normal samples of different brain regions. (B) Multi-dimensional scaling of gene expression in 100 randomly selected non-hyper-variable genes in cancer for the same samples of different brain regions. For genes that are hyper-variable in cancer, samples cluster by brain region indicating their tissue specificity. However, the set of control genes (non-hyper-variable in cancer), lack tissue specificity and samples do not cluster by brain region. We computed ratios of between-cluster variance to within-cluster variance for both hyper-variable and control and observed that brain region clusters are tighter for hyper-variable genes than control genes (variance ratios of 60.5 and 4.2 for hyper-variable and control genes respectively).



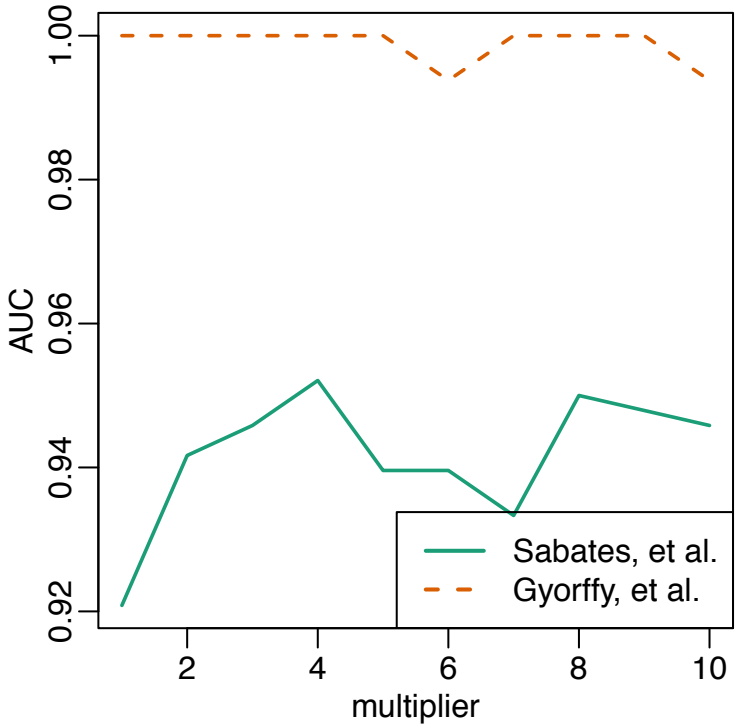

**Supplementary Figure 5. The set of tissue-specific genes is enriched for genes that are hyper-variable in cancer.** We plot the odds ratio for a hyper-variability enrichment Fisher exact test in tissue-specific genes as a function of the log2 variance ratio. Each point corresponds to genes with log2 variance ratio within the indicated range. Odds ratio increases as variability in cancer increases. All tests are significant at P<0.05 level.




**Supplementary Figure 6. Specificity of the universally hyper-variable tissue-specific genes.** For each tissue-specific gene, we calculate the number of cancer types for which it is hyper-variable by computing the log2 ratio between standard deviation for that cancer type and mean normal standard deviation across tissues. We plot on the left a histogram of the number of cancer types for tissue-specific genes that are not consistently hyper-variable in cancer. The histogram on the right corresponds to genes that are consistently hyper-variable. We see that the vast majority of tissue-specific genes show hyper-variability in a small number of cancer-types. This suggests that consistent hyper-variability in cancer cannot be ascribed entirely to cellular heterogeneity.




**Supplementary Figure 7. The anti-profile method produces robust signatures.** We plot the percentage of the top 100 universally hyper-variable genes selected in at least *n* cross-validation experiments. We observed that more than 73% of the universally hyper-variable genes are selected in 15 or more of the 17 cross-validation experiments performed, while all of the hyper-variable genes were selected in at least 5 cross-validation experiments, indicating the robustness and reproducibility of the anti-profile method.




**Supplementary Figure 8. Sensitivity of colon anti-profile to choice of normal expression region multiplier.** We repeated the cross-validation experiment reported in Figure 1B using different median absolute deviation multipliers to define normal regions of expression. Here we plot the AUC achieved in cross-validation against the choice of multiplier. We find that results are not very sensitive to the choice of multiplier except for narrow regions of normal expression (1-3) where performance drops in one of the datasets.