

design_auto_xsd.xml

<ROSETTASCRIPTS>

<SCOREFXNS>

```
    <ScoreFunction name="talaris_full" weights="ref2015">
      <Reweight scoretype="coordinate_constraint"
weight="0.4"/>
      <Reweight scoretype="res_type_constraint"
weight="0.4"/>
    </ScoreFunction>
```

```
    <ScoreFunction name="soft_rep_full"
weights="soft_rep">
      <Reweight scoretype="coordinate_constraint"
weight="0.4"/>
      <Reweight scoretype="res_type_constraint"
weight="0.4"/>
    </ScoreFunction>
```

```
    <ScoreFunction name="tal_no_pssm" weights="ref2015">
      <Reweight scoretype="coordinate_constraint"
weight="0.4"/>
    </ScoreFunction>
```

```
    <ScoreFunction name="talaris_pure" weights="ref2015"/>
  </SCOREFXNS>
```

<TASKOPERATIONS>

```
    <RestrictIdentitiesAtAlignedPositions
name="RIAAP_design" source_pdb="4zstA.pdb.gz"
resnums="267,196,167,23,21,135" keep_aas="HDKE"
design_only_target_residues="1"/>keep cat arg
    <RestrictIdentitiesAtAlignedPositions
name="RIAAP_design_glu" source_pdb="2vc5B_ppk.pdb.gz" resnums="166"
keep_aas="HDKE" design_only_target_residues="1"/>
```

</TASKOPERATIONS>

<RESIDUE_SELECTORS>

```
    <Task name="RIAAP_design" packable="1"
designable="1" task_operations="RIAAP_design"/>RIAAP_design_glu />
    <Not name="No_RIAAP_design"
selector="RIAAP_design"/>
```

</RESIDUE_SELECTORS>

<TASKOPERATIONS>

```
    <ProteinInterfaceDesign name="no_repack_ligand"
repack_chain2="0" />
```

```

        <RestrictIdentitiesAtAlignedPositions
name="no_repack_cat_Res" source_pdb="4zstA.pdb.gz"
prevent_repacking="1" resnums="267,196,167,23,21,135,134,136"
keep_aas="HDKE"/>keep cat arg
        <RestrictIdentitiesAtAlignedPositions name="no_repack_cat_glu"
source_pdb="2vc5B_ppk.pdb.gz" prevent_repacking="1" resnums="166"
keep_aas="HDKE"/>

```

```

        <OperateOnResidueSubset name="No_RIAAP_design"
selector="No_RIAAP_design" >
        <RestrictToRepackingRLT/>
        </OperateOnResidueSubset>

        <InitializeFromCommandline name="init"/>
        <ReadResfile name="read_resfile" filename="resfile/
resfile.dat"/>
        <RestrictToRepacking name="rtr"/>
        <SeqprofConsensus name="seqprofcons"
min_aa_probability="1" probability_larger_than_current="0"
ignore_pose_profile_length_mismatch="1" keep_native="1" chain_num="0"
debug="1"/>

```

</TASKOPERATIONS>

<MOVERS>

```

        <RotamerTrialsMinMover name="sc_min"
scorefxn="talaris_full" task_operations="RIAAP_design,seqprofcons"/>
        <PackRotamersMover name="restore_cat_res"
scorefxn="talaris_full" task_operations="seqprofcons,No_RIAAP_design"/
>
        <AtomCoordinateCstMover name="chelating_res"
reference_pdb="4zstA.pdb.gz" func_groups="1"
task_operations="RIAAP_design" coord_dev="0.01"/>
        <Subroutine name="splice_constraints"
xml_fname="splice_constraints_xsd.xml"/>
        <AddConstraintsToCurrentConformationMover
name="add_CA_cst" />
        <PackRotamersMover name="soft_design"
scorefxn="soft_rep_full"
task_operations="init,read_resfile,seqprofcons,no_repack_cat_Res,no_re
pack_cat_glu"/>
        <PackRotamersMover name="hard_design"
scorefxn="talaris_full"
task_operations="init,read_resfile,seqprofcons,no_repack_cat_Res,no_re
pack_cat_glu"/>
        <RotamerTrialsMinMover name="RTmin"
scorefxn="talaris_full"
task_operations="rtr,no_repack_cat_Res,no_repack_cat_glu"/>
        <TaskAwareMinMover name="soft_min"
scorefxn="soft_rep_full" chi="1" bb="1" jump="0"

```

```

task_operations="no_repack_cat_Res,no_repack_cat_glu" />
    <TaskAwareMinMover name="hard_min"
scorefxn="talaris_full" chi="1" bb="1" jump="0"
task_operations="no_repack_cat_Res,no_repack_cat_glu"/>
    <SaveAndRetrieveSidechains name="sars"/>
    <ParsedProtocol name="design_block">
        <Add mover_name="soft_design"/>
        <Add mover_name="soft_min"/>
        <Add mover_name="soft_design"/>
        <Add mover_name="hard_min"/>
        <Add mover_name="hard_design"/>
        <Add mover_name="hard_min"/>
        <Add mover_name="hard_design"/>
        Add mover_name=sars/>
        <Add mover_name="RTmin"/>
        <Add mover_name="RTmin"/>
        <Add mover_name="hard_min"/>
    </ParsedProtocol>
    <LoopOver iterations="4" mover_name="design_block"
name="iter4"/>
    </MOVERS>

    <FILTERS>
        <ScoreType confidence="0" name="stability_score_full"
score_type="total_score" scorefxn="talaris_full" threshold="0"/>
        <ScoreType confidence="0"
name="stability_without_pssm" score_type="total_score"
scorefxn="tal_no_pssm" threshold="0"/>
        <ScoreType confidence="0" name="stability_pure"
score_type="total_score" scorefxn="talaris_pure" threshold="0"/>
        <PackStat name="packstat" confidence="0"/>
        <Time name="timer"/>
        <DesignableResidues name="designable"
task_operations="No_RIAAP_design" designable="1" packable="0"/>
    </FILTERS>

    <PROTOCOLS>
        <Add mover_name="FSP"/>
        <Add mover_name="restore_cat_res"/>
        Add filter_name="designable"/>
        <Add mover_name="add_CA_cst"/>
        <Add mover_name="iter4"/>
        <Add filter_name="stability_score_full"/>
        <Add filter_name="stability_without_pssm"/>
        <Add filter_name="stability_pure"/>
        <Add filter_name="packstat"/>
    </PROTOCOLS>

</ROSETTASCRIPTS>

```

filterscan_auto_xsd.xml

<ROSETTASCRIPTS>

<SCOREFXNS>

<ScoreFunction name="talaris_full" weights="ref2015">

<Reweight scoretype="coordinate_constraint"
weight="0.4"/>

<Reweight scoretype="res_type_constraint"
weight="0.8"/>

</ScoreFunction>

</SCOREFXNS>

<RESIDUE_SELECTORS>

<Chain name="chA" chains="A"/>

<Chain name="lig" chains="B"/>

<InterfaceByVector name="interF" grp1_selector="chA"
grp2_selector="lig" cb_dist_cut="15" nearby_atom_cut="15"
vector_angle_cut="75" vector_dist_cut="9"/>

</RESIDUE_SELECTORS>

<TASKOPERATIONS>

<InitializeFromCommandline name="init"/>

<SeqprofConsensus name="pssm_cutoff"

min_aa_probability="0" probability_larger_than_current="0"
convert_scores_to_probabilities="0" keep_native="1" debug="1"/>

<RestrictIdentitiesAtAlignedPositions

name="no_repack_cat_Res" source_pdb="4zstA.pdb.gz"
prevent_repacking="1" resnums="267,196,167,23,21" keep_aas="HDKE"/

>keep cat arg

<RestrictIdentitiesAtAlignedPositions

name="RIAAP_design" source_pdb="4zstA.pdb.gz"
resnums="135,267,196,167,23,21" keep_aas="HDKE"

design_only_target_residues="1"/>keep cat arg

<RestrictIdentitiesAtAlignedPositions name="no_repack_cat_glu"
source_pdb="4qsfa_ppk.pdb.gz" prevent_repacking="1" resnums="176"
keep_aas="HDKE"/>keep cat arg

<DesignAround name="test" design_shell="1" resnums="2"
repack_shell="8" allow_design="1" resnums_allow_design="1"/>

</TASKOPERATIONS>

<MOVERS>

```

        <PackRotamersMover name="restore_cat_res"
scorefxn="talaris_full" task_operations="RIAAP_design"/>
        <AddConstraintsToCurrentConformationMover
name="add_CA_cst" />
        <FavorSequenceProfile name="FSP" scaling="none"
weight="1" pssm="%%pssm%%" scorefxns="talaris_full"/>
        <MinMover name="min_all" scorefxn="talaris_full"
chi="1" bb="1" jump="0"/>
        <Subroutine name="design"
xml_fname="design_auto_xsd.xml"/>
        <RotamerTrialsMinMover name="sc_min"
scorefxn="talaris_full" task_operations="RIAAP_design"/>
        <AtomCoordinateCstMover name="chelating_res"
reference_pdb="4zstA.pdb.gz" func_groups="1"
task_operations="RIAAP_design" coord_dev="0.01"/>
        <Subroutine name="splice_constraints"
xml_fname="splice_constraints_xsd.xml"/>
    </MOVERS>

    <FILTERS>
        <DesignableResidues name="designable"
task_operations="init,pssm_cutoff,no_repack_cat_Res,no_repack_cat_glu"
designable="1" packable="1"/>

        <ScoreType name="stability_score_full"
scorefxn="talaris_full" score_type="total_score" threshold="0.0"/>
        <Delta name="delta_score_full"
filter="stability_score_full" upper="1" lower="0" range="0.5"/>

        <Time name="timer"/>
        <FilterScan name="filter_scan" scorefxn="talaris_full"
relax_mover="min_all" keep_native="1"
task_operations="init,pssm_cutoff,no_repack_cat_Res,no_repack_cat_glu"
delta_filters="delta_score_full" delta="true" resfile_name="resfile/
resfile.dat" report_all="1" delta_filter_thresholds="-0.75"
score_log_file="%%pdb%%_score_full.log" dump_pdb="0"/>
    </FILTERS>

    <PROTOCOLS>
        <Add mover_name="restore_cat_res"/>
        <Add mover_name="add_CA_cst"/>
        <Add mover="splice_constraints"/>
        <Add filter="filter_scan"/>
        <Add mover="design"/>
    </PROTOCOLS>

</ROSETTASCRIPTS>

```

splice_constraints_xsd.xml

<ROSETTASCRIPTS>a script for prepacking, redocking and minimizing a
pdb structure. The idea is to
relieve minor clashes that are often observed in PDBs.

Recommended command-line:

rosetta_scripts -s PDB -parser:protocol refine_two_chain_pdb.xml -ex1
-ex2 -use_input_sc -database ~/minirosetta_database -
docking:fake_native

<TASKOPERATIONS>

<RestrictToRepacking name="rtr"/>

SampleRotamersFromPDB name=template_chelating_res

add_rotamer=1 aligned_positions=22,24,137,170,198,255 debug=0/> //

These definitions are used for the E chelating residue of 1bf6

</TASKOPERATIONS>

<SCOREFXNS>

<ScoreFunction name="talaris14" weights="ref2015">

<Reweight scoretype="res_type_constraint"

weight="0.5"/>

</ScoreFunction>

</SCOREFXNS>

<FILTERS>

<Ddg name="ddg" confidence="0"/>

<Sasa name="sasa" confidence="0"/>

<Rmsd name="rms" confidence="0"/>

</FILTERS>

<MOVERS>

<Splice name="splice_seqconstraints" scorefxn="talaris14"

add_sequence_constraints_only="1">tolrance=10>

<Segments current_segment="blade4">

<Segment name="frm1"

pdb_profile_match="pdb_profile_match" profiles="2vc5B:frm1_2vc5B.PSSM,
2vc5B:frm1_2vc5B.PSSM"/>

<Segment name="blade4"

pdb_profile_match="pdb_profile_match" profiles="%%blade4%%"/>

<Segment name="blade5"

pdb_profile_match="pdb_profile_match" profiles="%%blade5%%"/>

<Segment name="blade6"

pdb_profile_match="pdb_profile_match" profiles="%%blade6%%"/>

<Segment name="blade7"

pdb_profile_match="pdb_profile_match" profiles="%%blade7%%"/>

<Segment name="frm2"

pdb_profile_match="pdb_profile_match" profiles="2vc5B:frm2_2vc5B.PSSM,
2vc5B:frm2_2vc5B.PSSM"/>

</Segments>

</Splice>

```

    </MOVERS>
    <APPLY_TO_POSE>
    </APPLY_TO_POSE>
    <PROTOCOLS>
        <Add mover_name="splice_seqconstraints"/>
    </PROTOCOLS>
</ROSETTASCRIPTS>

```

splice_in_xsd.xml

```

<ROSETTASCRIPTS>
    <TASKOPERATIONS>
        <RestrictToRepacking name="rtr"/>

        <InitializeFromCommandline name="init"/>
        <SeqprofConsensus name="seqprofcons"
min_aa_probability="1" probability_larger_than_current="0"
ignore_pose_profile_length_mismatch="1" keep_native="1" debug="0"/>

        <RestrictIdentitiesAtAlignedPositions
name="RIAAP_no_des" source_pdb="4zstA.pdb"
design_only_target_residues="1" resnums="267,196,167,23,21"
keep_aas="HDGARNY"/>keep cat arg
        <RestrictIdentitiesAtAlignedPositions name="RIAAP"
source_pdb="4zstA.pdb" design_only_target_residues="0"
resnums="267,196,167,23,21" keep_aas="HD"/>

    </TASKOPERATIONS>
    <SCOREFXNS>

        <ScoreFunction name="soft_rep_res_type_cst"
weights="soft_rep">
            <Reweight scoretype="res_type_constraint"
weight="0.2"/>
        </ScoreFunction>

        <ScoreFunction name="TalarisCal_coordcst"
weights="ref2015">
            <Reweight scoretype="res_type_constraint"
weight="0.2"/>

```

```

        <Reweight scoretype="coordinate_constraint"
weight="1"/>
        <Reweight scoretype="chainbreak" weight="10"/>
    </ScoreFunction>

    <ScoreFunction name="soft_rep_coordcst"
weights="soft_rep">
        <Reweight scoretype="coordinate_constraint"
weight="0.06"/>
    </ScoreFunction>

    <ScoreFunction name="TalarisReg" weights="ref2015"/>
    <ScoreFunction name="talaris14" weights="ref2015">
        <Reweight scoretype="res_type_constraint"
weight="0.5"/>
        <Reweight scoretype="dihedral_constraint"
weight="10"/>
        <Reweight scoretype="coordinate_constraint"
weight="10"/>
        <Reweight scoretype="chainbreak" weight="10"/>
    </ScoreFunction>

    <ScoreFunction name="talaris2014_chainbreak"
weights="ref2015">
        <Reweight scoretype="chainbreak" weight="1"/>
    </ScoreFunction>
</SCOREFXNS>

<FILTERS>
    <ScoreType name="chainbreak_val"
scorefxn="talaris2014_chainbreak" score_type="chainbreak"
threshold="0.2" confidence="1"/>
    <PackStat name="packstat" threshold="0.63" repeats="3"
confidence="0"/>
    <ScoreType name="total_score" score_type="total_score"
scorefxn="TalarisReg" threshold="1000000"/>
</FILTERS>

<MOVERS>
    <RotamerTrialsMinMover name="rtmin"
task_operations="rtr,init" scorefxn="TalarisCal_coordcst"/>

    <Splice name="splice_blade4"
torsion_database="2vc5B_blade4.db" use_sequence_profiles="1"
scorefxn="talaris14" ccd="0" thread_ala="0" repack_shell="100"
design_shell="100" template_file="%%start_pdb%%"
design_task_operations="init,seqprofcons" design="1" CG_const="0"
ignore_chain_break="1" debug="0" rtmin="1" min_seg="1"
delta_lengths="-1,0,1" equal_length="1" dbase_iterate="1">
        <Segments current_segment="blade4">

```

```

        <Segment name="frm1"
pdb_profile_match="pdb_profile_match" profiles="2vc5B:frm1_2vc5B.PSSM,
2vc5B:frm1_2vc5B.PSSM"/>
        <Segment name="blade4"
pdb_profile_match="pdb_profile_match" profiles="%%blade4%%"/>
        <Segment name="blade5"
pdb_profile_match="pdb_profile_match" profiles="%%blade5%%"/>
        <Segment name="blade6"
pdb_profile_match="pdb_profile_match" profiles="%%blade6%%"/>
        <Segment name="blade7"
pdb_profile_match="pdb_profile_match" profiles="%%blade7%%"/>
        <Segment name="frm2"
pdb_profile_match="pdb_profile_match" profiles="2vc5B:frm2_2vc5B.PSSM,
2vc5B:frm2_2vc5B.PSSM"/>
    </Segments>
</Splice>

```

```

    <Splice name="splice_blade5"
torsion_database="2vc5B_blade5.db" use_sequence_profiles="1"
scorefxn="talaris14" ccd="0" thread_ala="0" repack_shell="100"
design_shell="100" template_file="%%start_pdb%%"
design_task_operations="init,seqprofcons" design="1" CG_const="0"
ignore_chain_break="1" debug="0" rtmin="1" min_seg="1"
delta_lengths="-1,0,1" equal_length="1" dbase_iterate="1">
        <Segments current_segment="blade5">
            <Segment name="frm1"
pdb_profile_match="pdb_profile_match" profiles="2vc5B:frm1_2vc5B.PSSM,
2vc5B:frm1_2vc5B.PSSM"/>
            <Segment name="blade4"
pdb_profile_match="pdb_profile_match" profiles="%%blade4%%"/>
            <Segment name="blade5"
pdb_profile_match="pdb_profile_match" profiles="%%blade5%%"/>
            <Segment name="blade6"
pdb_profile_match="pdb_profile_match" profiles="%%blade6%%"/>
            <Segment name="blade7"
pdb_profile_match="pdb_profile_match" profiles="%%blade7%%"/>
            <Segment name="frm2"
pdb_profile_match="pdb_profile_match" profiles="2vc5B:frm2_2vc5B.PSSM,
2vc5B:frm2_2vc5B.PSSM"/>
        </Segments>
    </Splice>

```

```

<Splice name="splice_blade6" torsion_database="2vc5B_blade6.db"
use_sequence_profiles="1" scorefxn="talaris14" ccd="0" thread_ala="0"
repack_shell="100" design_shell="100" template_file="%%start_pdb%%"
design_task_operations="init,seqprofcons" design="1" CG_const="0"
ignore_chain_break="1" debug="0" rtmin="1" min_seg="1"
delta_lengths="-1,0,1" equal_length="1" dbase_iterate="1">
    <Segments current_segment="blade6">
        <Segment name="frm1"

```

```

pdb_profile_match="pdb_profile_match" profiles="2vc5B:frm1_2vc5B.PSSM,
2vc5B:frm1_2vc5B.PSSM"/>
        <Segment name="blade4"
pdb_profile_match="pdb_profile_match" profiles="%%blade4%%"/>
        <Segment name="blade5"
pdb_profile_match="pdb_profile_match" profiles="%%blade5%%"/>
        <Segment name="blade6"
pdb_profile_match="pdb_profile_match" profiles="%%blade6%%"/>
        <Segment name="blade7"
pdb_profile_match="pdb_profile_match" profiles="%%blade7%%"/>
        <Segment name="frm2"
pdb_profile_match="pdb_profile_match" profiles="2vc5B:frm2_2vc5B.PSSM,
2vc5B:frm2_2vc5B.PSSM"/>
        </Segments>
    </Splice>

<Splice name="splice_blade7" torsion_database="2vc5B_blade7.db"
use_sequence_profiles="1" scorefxn="talaris14" ccd="0" thread_ala="0"
repack_shell="100" design_shell="100" template_file="%%start_pdb%%"
design_task_operations="init,seqprofcons" design="1" CG_const="0"
ignore_chain_break="1" debug="0" rtmin="1" min_seg="1"
delta_lengths="-1,0,1" equal_length="1" dbase_iterate="1">
    <Segments current_segment="blade6">
        <Segment name="frm1"
pdb_profile_match="pdb_profile_match" profiles="2vc5B:frm1_2vc5B.PSSM,
2vc5B:frm1_2vc5B.PSSM"/>
        <Segment name="blade4"
pdb_profile_match="pdb_profile_match" profiles="%%blade4%%"/>
        <Segment name="blade5"
pdb_profile_match="pdb_profile_match" profiles="%%blade5%%"/>
        <Segment name="blade6"
pdb_profile_match="pdb_profile_match" profiles="%%blade6%%"/>
        <Segment name="blade7"
pdb_profile_match="pdb_profile_match" profiles="%%blade7%%"/>
        <Segment name="frm2"
pdb_profile_match="pdb_profile_match" profiles="2vc5B:frm2_2vc5B.PSSM,
2vc5B:frm2_2vc5B.PSSM"/>
    </Segments>
    </Splice>

    <ClearConstraintsMover name="clear_constraints"/>
    <Subroutine name="splice_constraints"
xml_fname="splice_constraints_xsd.xml"/>
        Subroutine name="splice_blade4"
xml_fname="splice_in_blade4_db_iterate.xml"/>
        Subroutine name="splice_blade5"
xml_fname="splice_in_blade5_db_iterate.xml"/>
        Subroutine name="splice_blade6"
xml_fname="splice_in_blade6_db_iterate.xml"/>

```

```

        <AtomCoordinateCstMover name="chelating_res"
reference_pdb="4zstA.pdb" func_groups="1"
task_operations="RIAAP_no_des" coord_dev="0.01"/>
        <PackRotamersMover name="ppk" scorefxn="talaris14"
task_operations="seqprofcons" />
        <ParsedProtocol name="splice_menu"
mode="single_random"> Sampling blade_7-8 more because it's more
variable
                <Add mover="splice_blade4"/>
                <Add mover="splice_blade5"/>
                <Add mover="splice_blade6"/>
                <Add mover="splice_blade7"/>
        </ParsedProtocol>

        <GenericSimulatedAnnealer name="annealer"
mover_name="splice_menu" filter_name="total_score" trials="30"
preapply="0" />
        <PackRotamersMover name="design_talarisCal"
scorefxn="TalarisCal_coordcst" task_operations="init,seqprofcons"/>
        <PackRotamersMover name="design_softrep"
scorefxn="soft_rep_coordcst" task_operations="init,seqprofcons"/>

        <TaskAwareMinMover name="soft_min"
scorefxn="soft_rep_coordcst" bb="0" jump="0" chi="1"
task_operations="rtr,init"/> do not minimize bb
        <TaskAwareMinMover name="soft_min_all"
scorefxn="soft_rep_coordcst" bb="1" jump="0" chi="1"/> minimize all
but don't minimize chain2 backbone because it could contain tails and
holes
        <TaskAwareMinMover name="hard_min"
scorefxn="TalarisCal_coordcst" bb="0" jump="0" chi="1"
task_operations="rtr,init"/>

        <ParsedProtocol name="post_splice_refine">
                <Add mover="clear_constraints"/> //
remove any residual constraints from splice mover
                <Add mover="splice_constraints"/> //re-
institute the sequence constraints
                <Add mover="chelating_res"/>
                <Add mover="soft_min"/>
                <Add mover="hard_min"/>
                <Add mover="design_softrep"/>
                <Add mover="soft_min"/>
                <Add mover="hard_min"/>
                <Add mover="design_talarisCal"/>

```

```
                <Add mover="rtmin"/>
                <Add mover="hard_min"/>
            </ParsedProtocol>
        </MOVERS>

        <PROTOCOLS>
            <Add mover="chelating_res"/>
            <Add mover="annealer"/>
            <Add mover="post_splice_refine"/>
            <Add filter="packstat"/>

        </PROTOCOLS>
    </ROSETTASCRIPTS>
```