Updating RoNBio molecular modelling system to support *in silico* investigation of AMP activity on membrane models

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Romanian Node for Computational Biology (RoNBio)

- Integrated system based on a grid of HTC and HPC resources dedicated to the modeling and simulation of macromolecular structures, accessible through a graphical frontend (SIMBAGRAN portal).

- The system automates procedures for the investigation of current research topics, such as bacterial drug resistance, drug discovery, and bioinformatics by means of programmable and reusable Taverna workflows in order to simplify the user's tasks.

DISTRIBUTED SYSTEM OF HTC & HPC RESOURCES

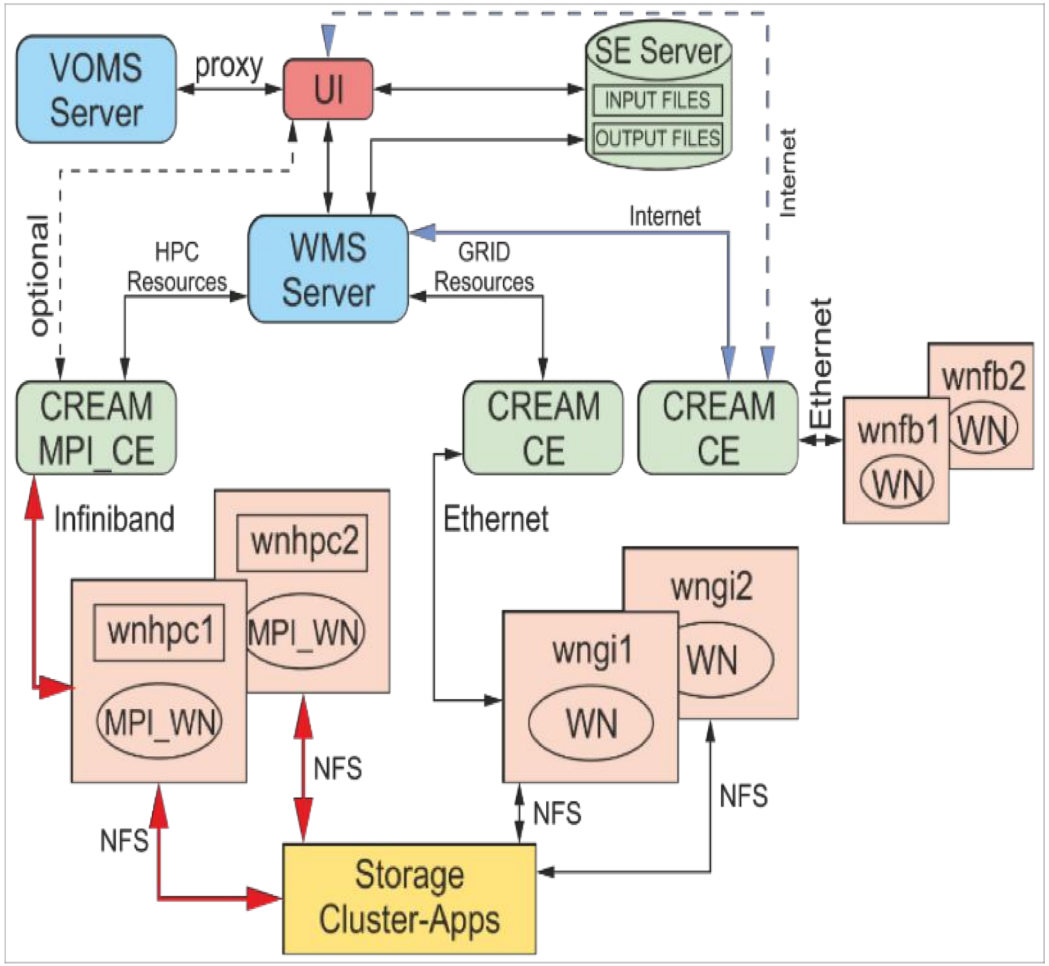
The system is currently using the resources of the GRIDIFIN site

Grid jobs containing Taverna workflows are submitted and executed on the HTC/HPC clusters.

GRIDIFIN site comprised of:

- One HPC cluster
- Two HTC WNs
- Storage

GRIDIFIN employs two CREAM-CE service (Computing Resource Execution And Management) - one used for distributed and the other for parallel computing.



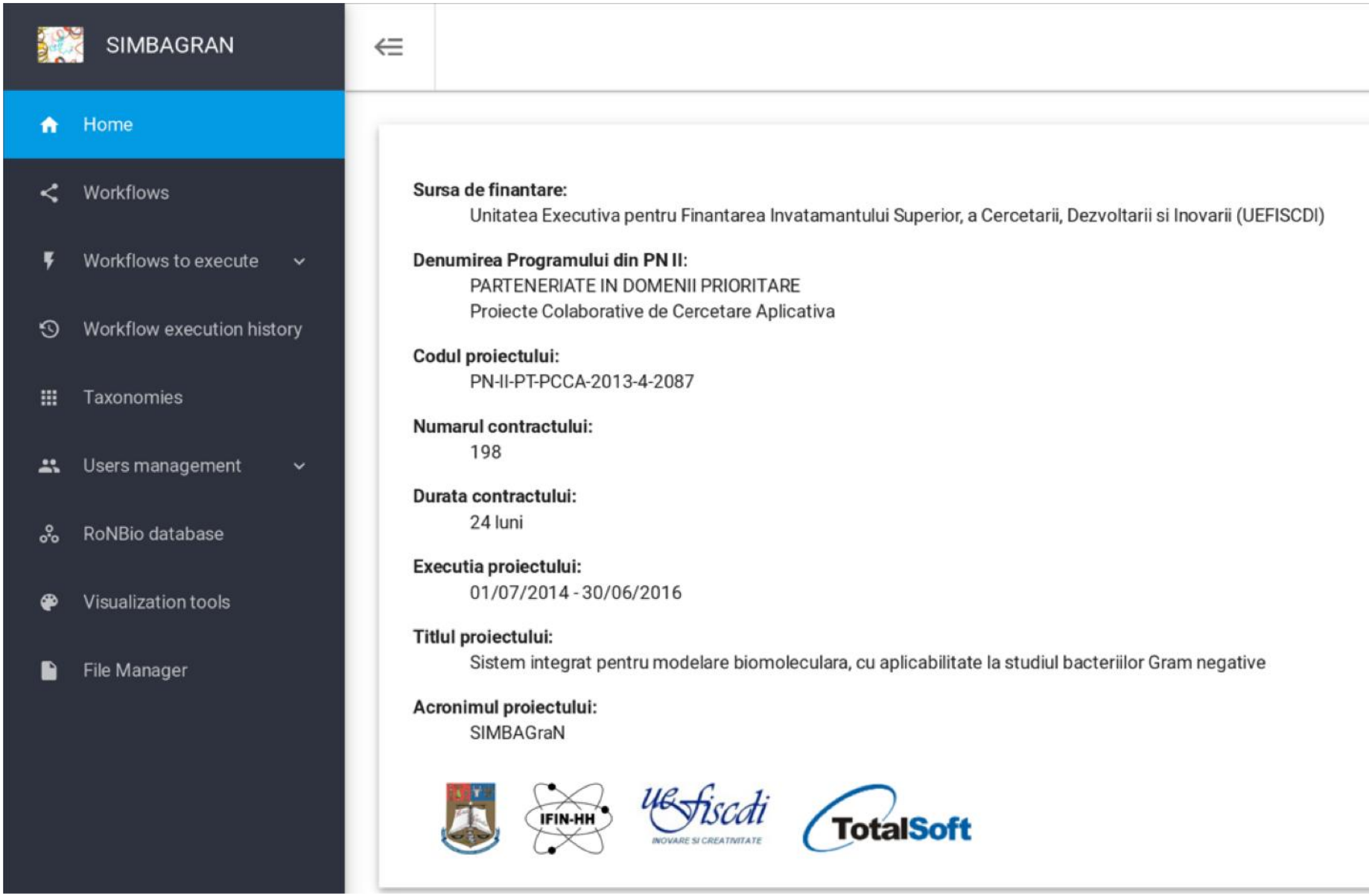
SIMBAGRAN portal

- The SIMBAGRAN portal integrates workflows for *in silico* investigation of Gram-negative bacteria resistance to β-lactam antibiotics by outer membrane permeability.

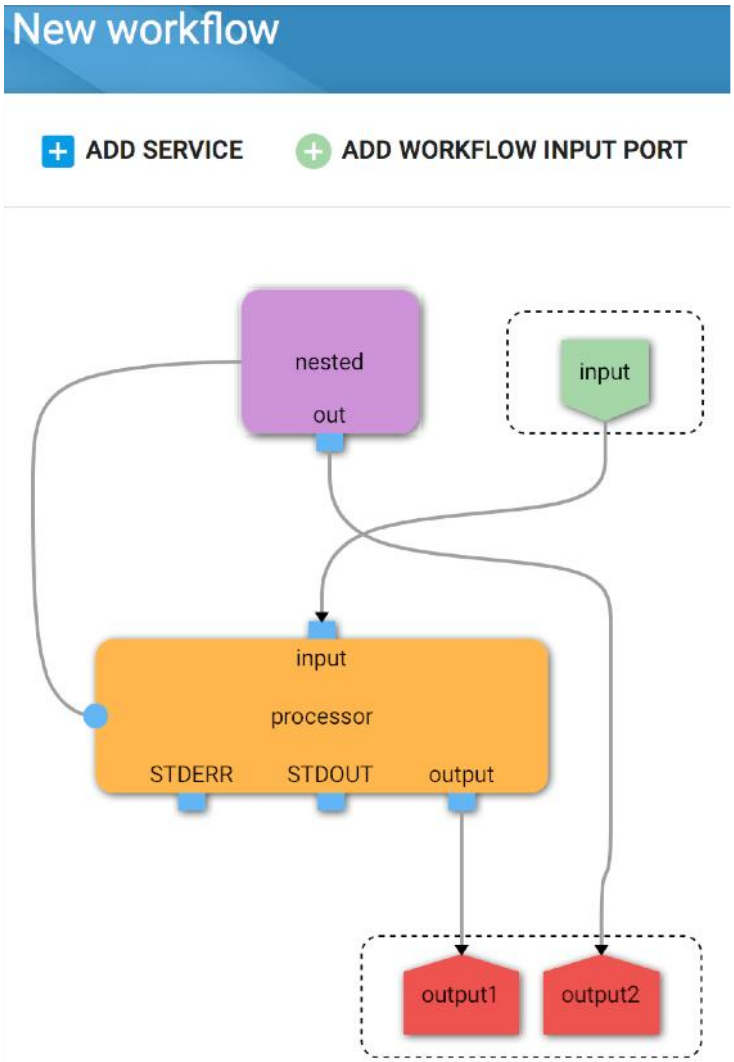
- Bioinformatics for next generation sequencing analysis: *de novo* assembly, mapping, indel analysis, SPNs and variant identification, and other bioinformatics tools.

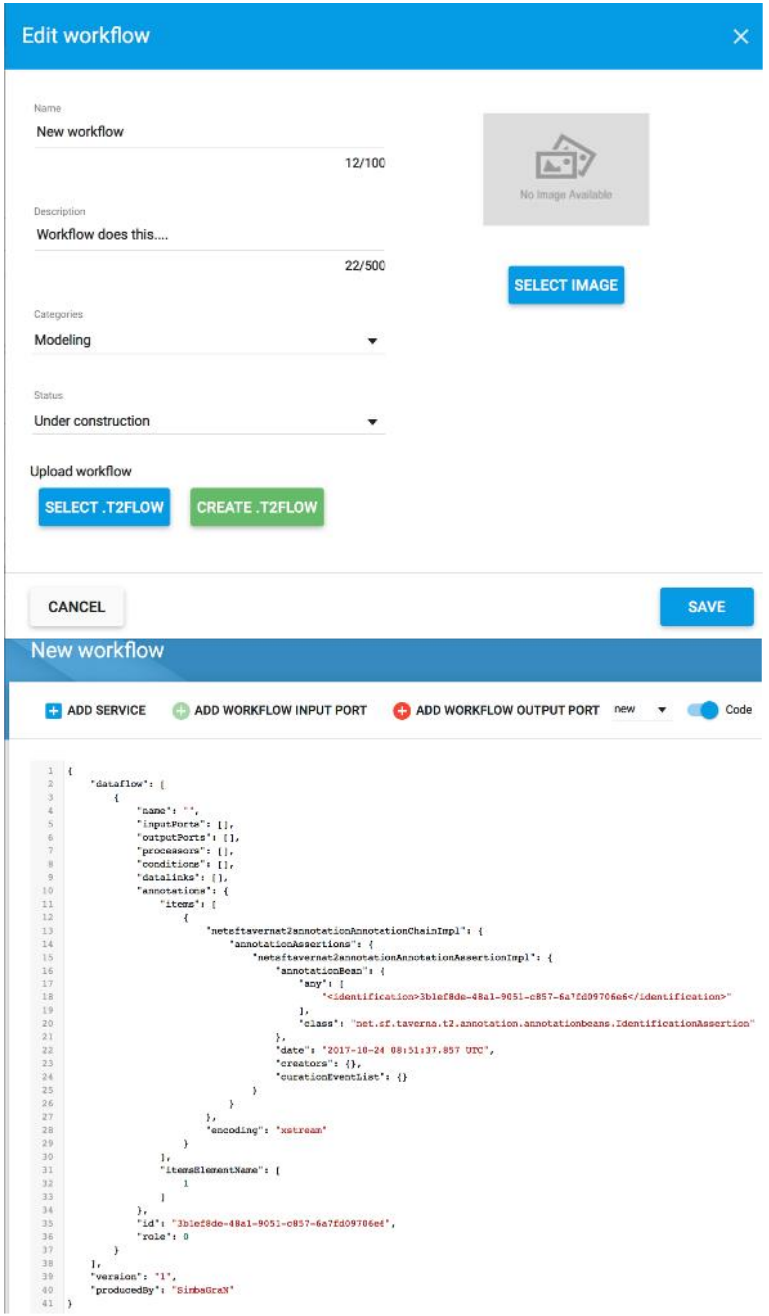
- The recently added workflows to RoNBio automates the analysis of molecular dynamics simulations meant to investigate the *in silico* activity of antimicrobial peptides (AMP) on lipid bilayers, including: membrane thickness, area per lipid (APL), AMPs insertion in the bilayer surface, and estimation of the AMPs free binding energy to the membrane.

SIMBAGRAN portal web interface

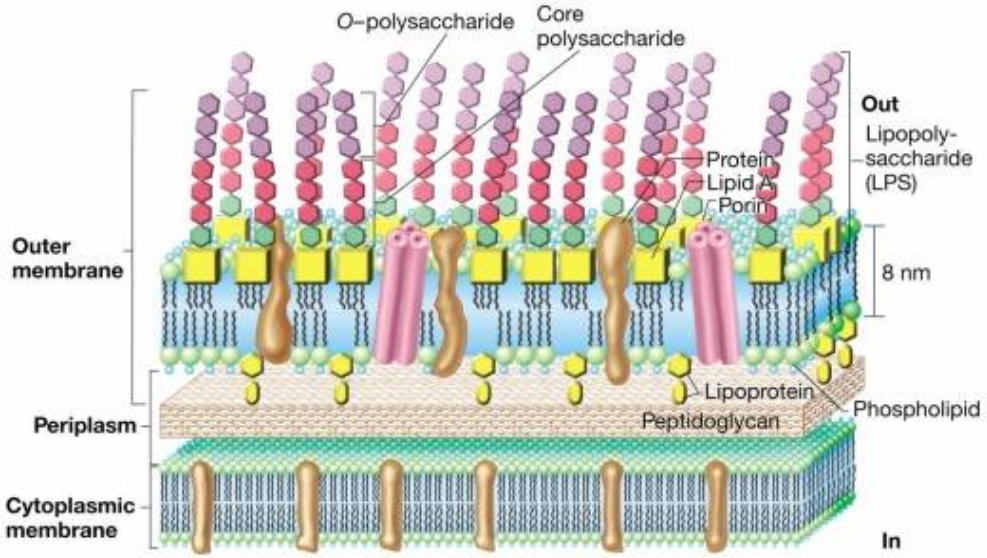


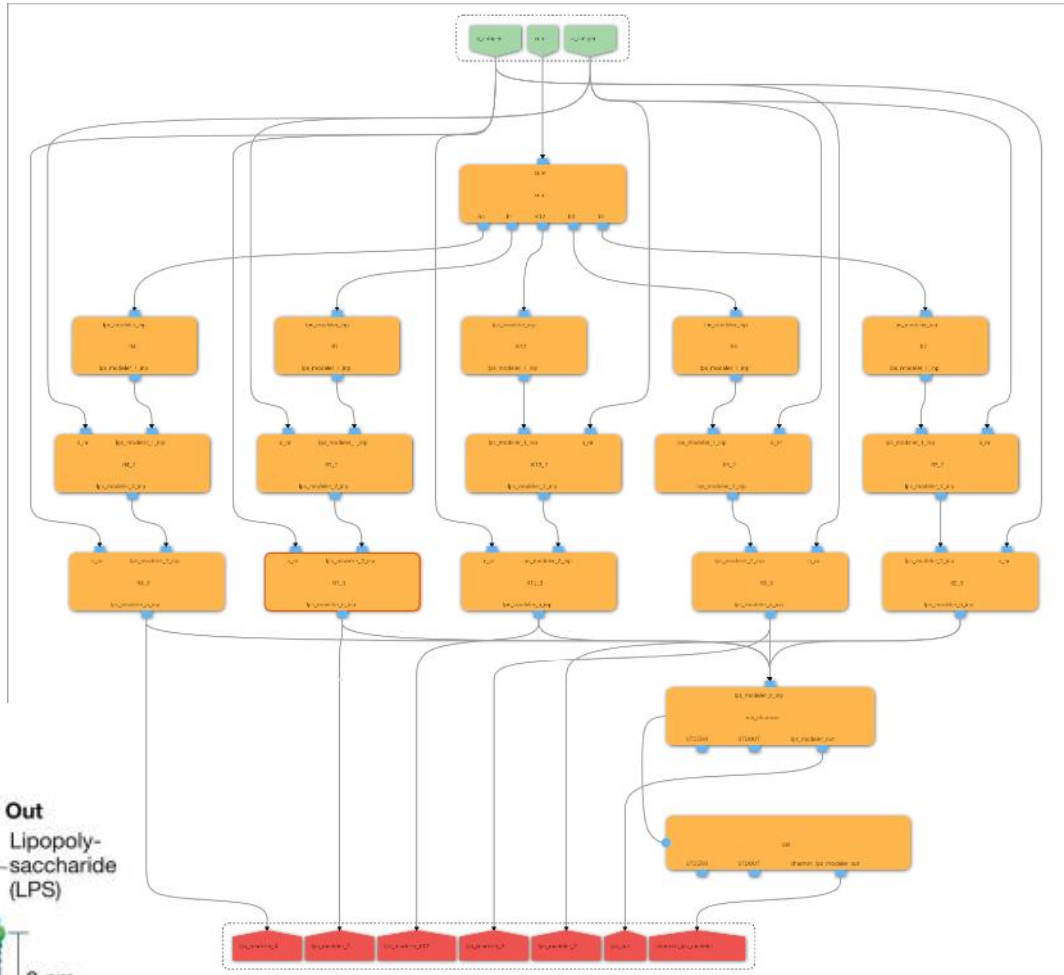
WORKFLOW CREATION



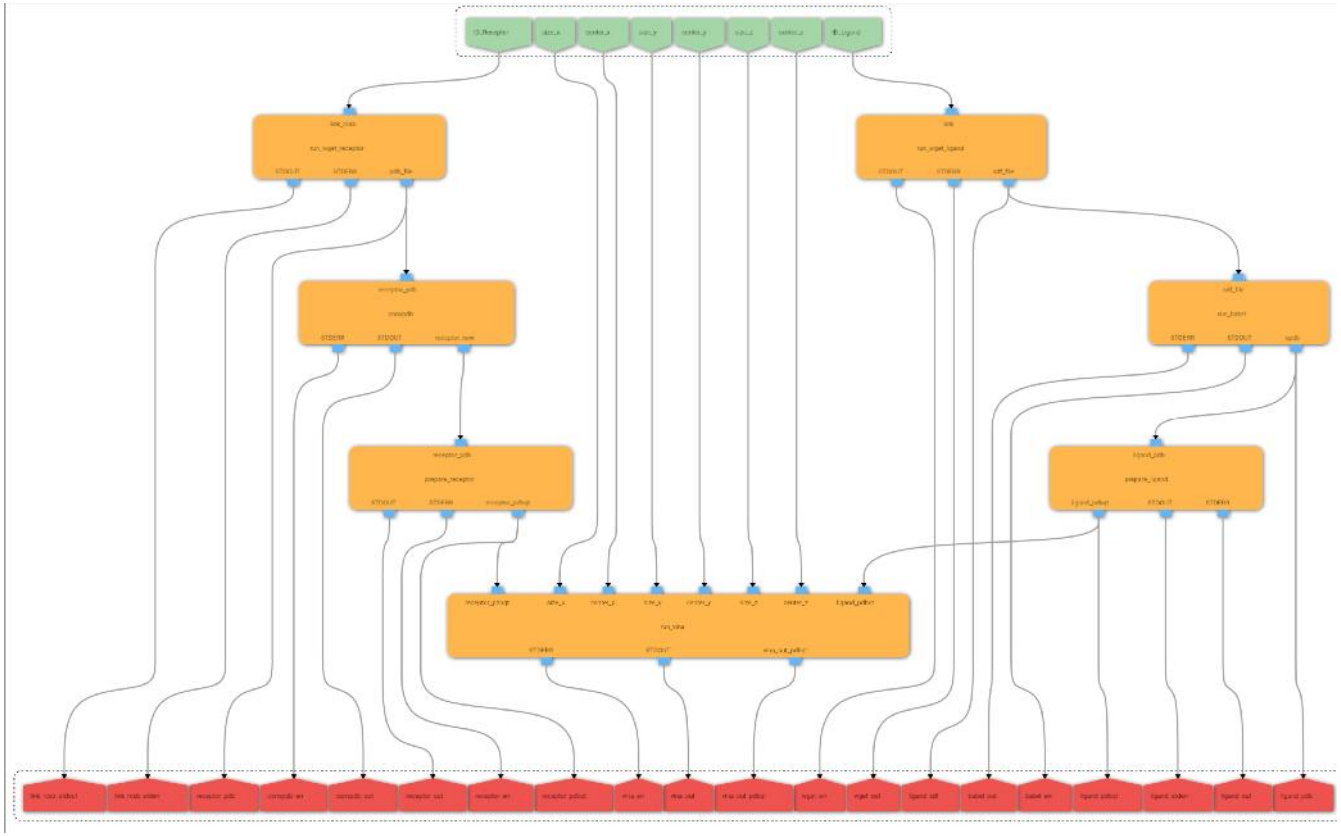


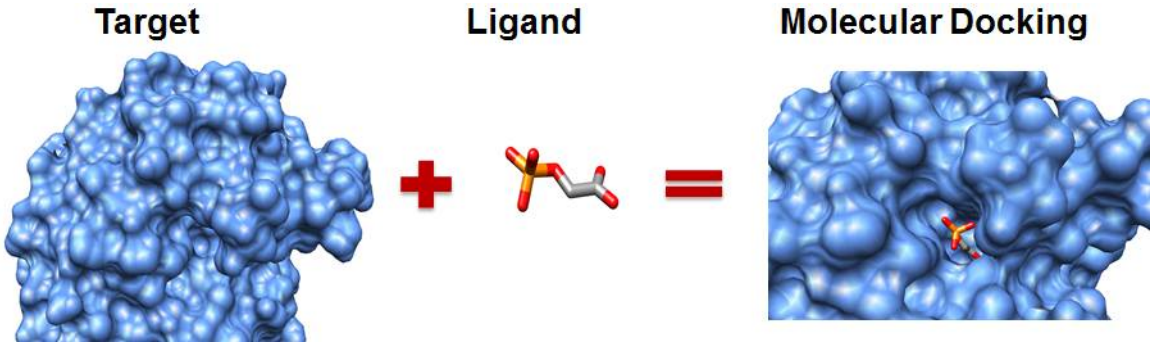
Parametrization and modeling of LPS molecules from *Escherichia coli*





Molecular docking workflow with Autodock Vina for drug discovery





Alejandra Hernández-Santoyo et al., 2013

Antimicrobial peptides

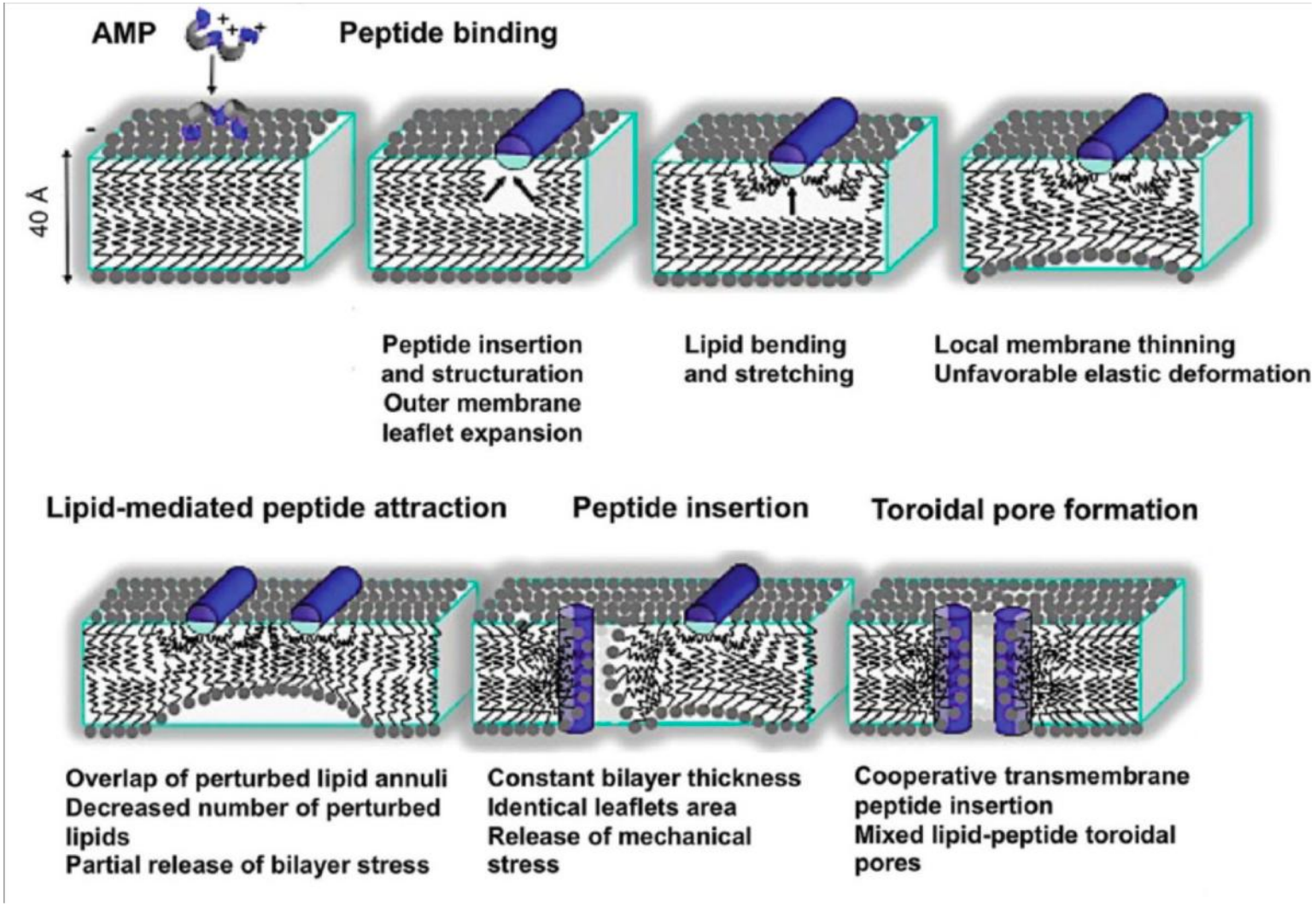
- Antimicrobial peptides (AMP) are oligopeptides with variable number of amino acids (typically between 5 and over 100)

- The interaction of AMPs with plasmatic membrane determines membrane permeabilization and consequently cell death rapidly after coming into contact with the cell membrane

- They can also act synergistically with antibiotic molecules and may reduce the inhibitory concentration of some antibiotics in some cases hundreds of times.

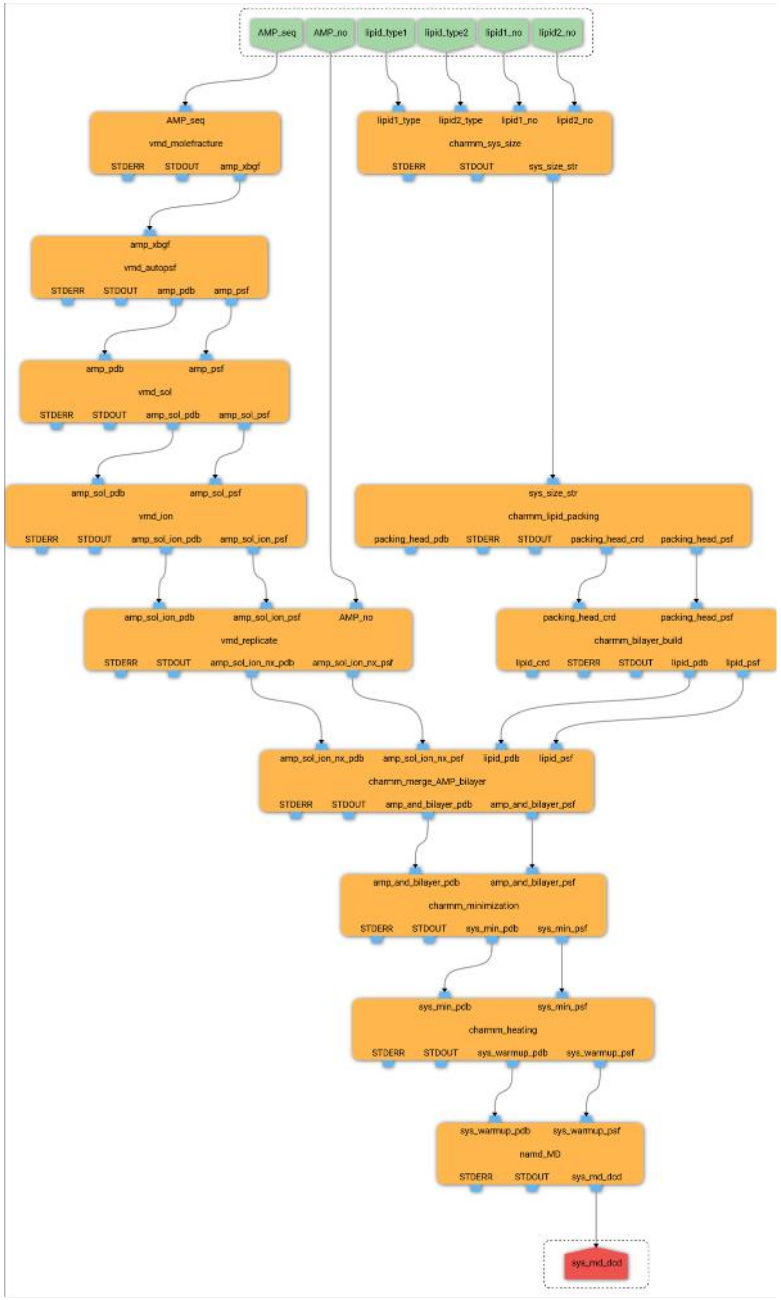
- Scientific interest due to their potential as alternatives to antibiotics that have low propensity to be susceptible to bacterial resistance mechanisms

Antimicrobial peptides interaction with membrane model



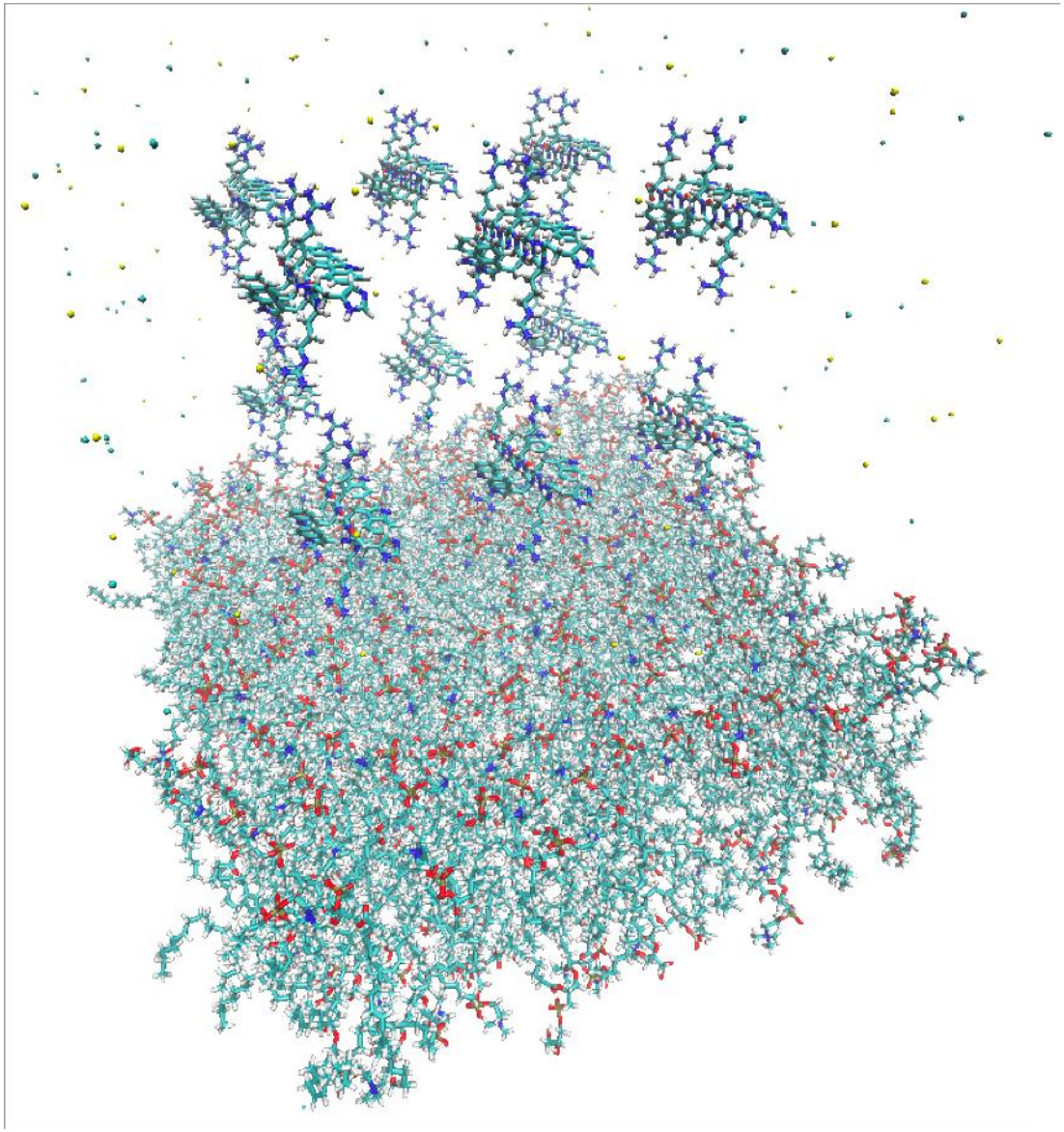
A. Ladram and P. Nicolas, Frontiers in Bioscience, Landmark, 21, 1341-1371, 2016]

Workflow for building molecular dynamics systems and simulation



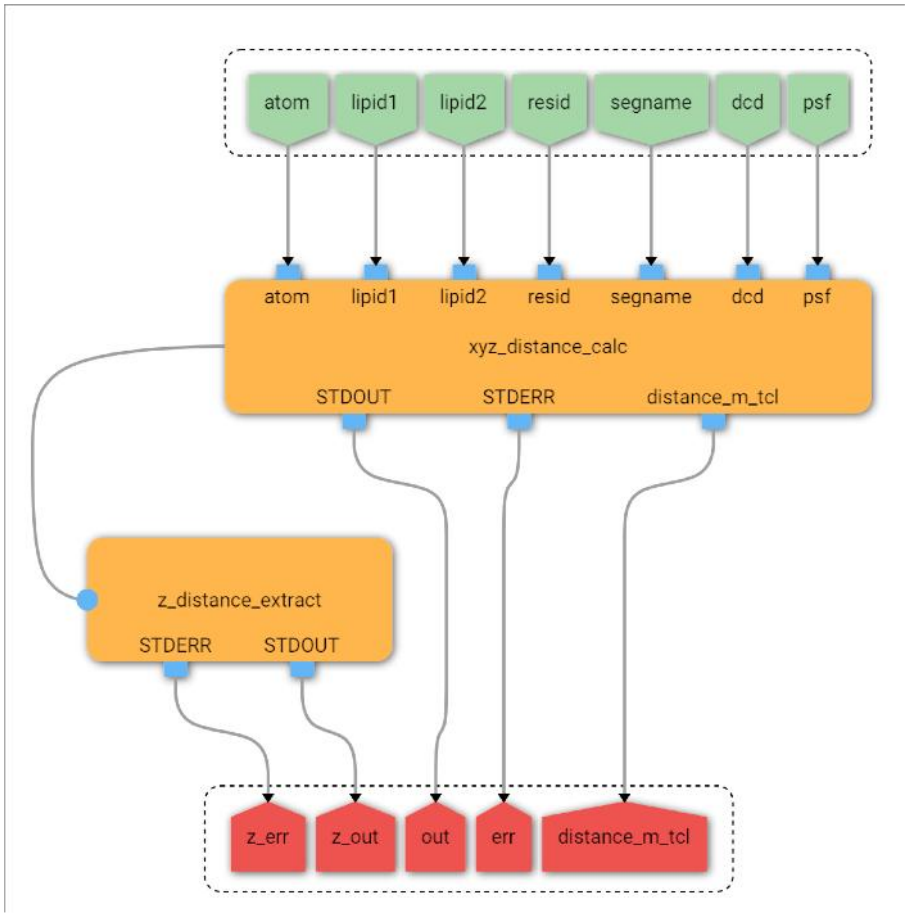
- The workflow employs VMD for building the AMP component of the system; CHARMM for building the lipid bilayer and NAMD for minimization, system warmup and NPT molecular dynamics.
- Two branches intended for separate modelling of AMPs and lipid bilayer
- Single letter amino acidic sequence e.g. RRWWRWWRR
- Solvates the AMP molecule with TIP3P molecular model and ionize the solvated molecule with NaCl to a concentration of 150 mM/L
- Builds configurations of AMP structures with various membrane models (e.g. DOPC, DOPG, POPC, etc.)
- Merges the results of the two branches (AMPs and membrane model), minimizes the structure, followed by gradual heating to a temperature of 300 K, and performs molecular dynamics.

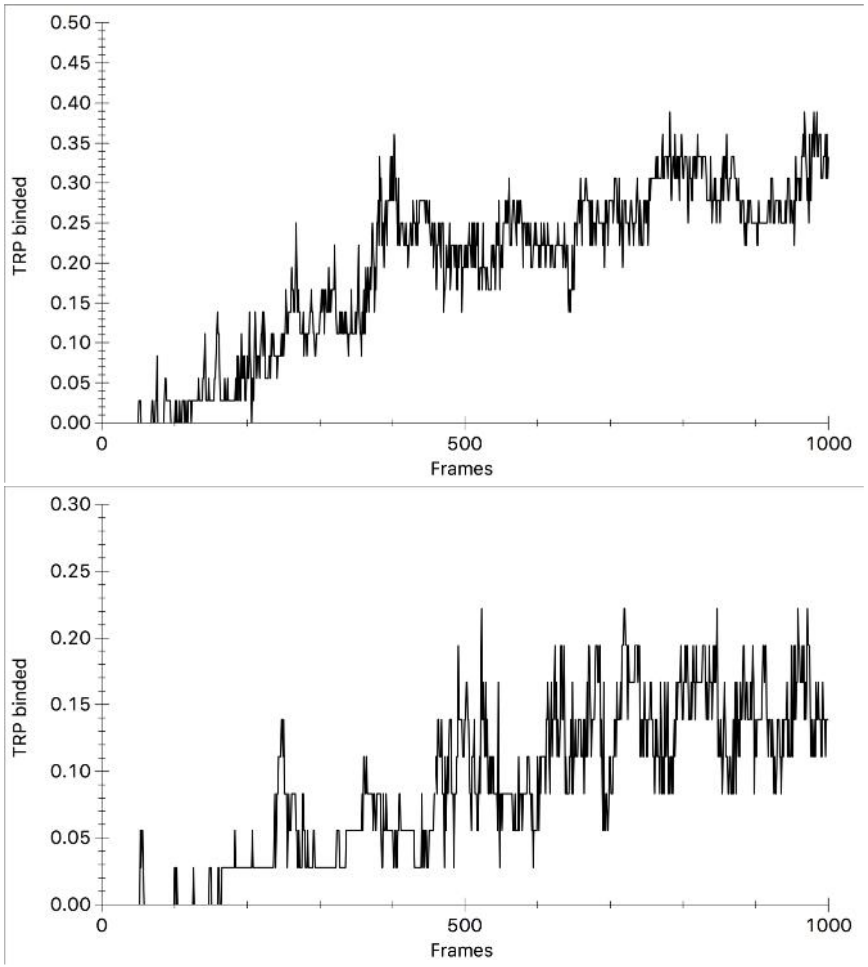
End result of workflow for building molecular dynamics systems and simulation



- MD simulation performed on a small (6 node) HPC cluster running CentOS 7.3, each fitted with two 14 core Intel Xeon E5-2660 v4 processors and 128GB RAM.
- NAMD NPT MD simulation was run on two NVIDIA Tesla K80 GPUs.
- 250 ns molecular dynamics with 12 AMPs DOPC:DOPG (molar ratio 85:15) membrane models.

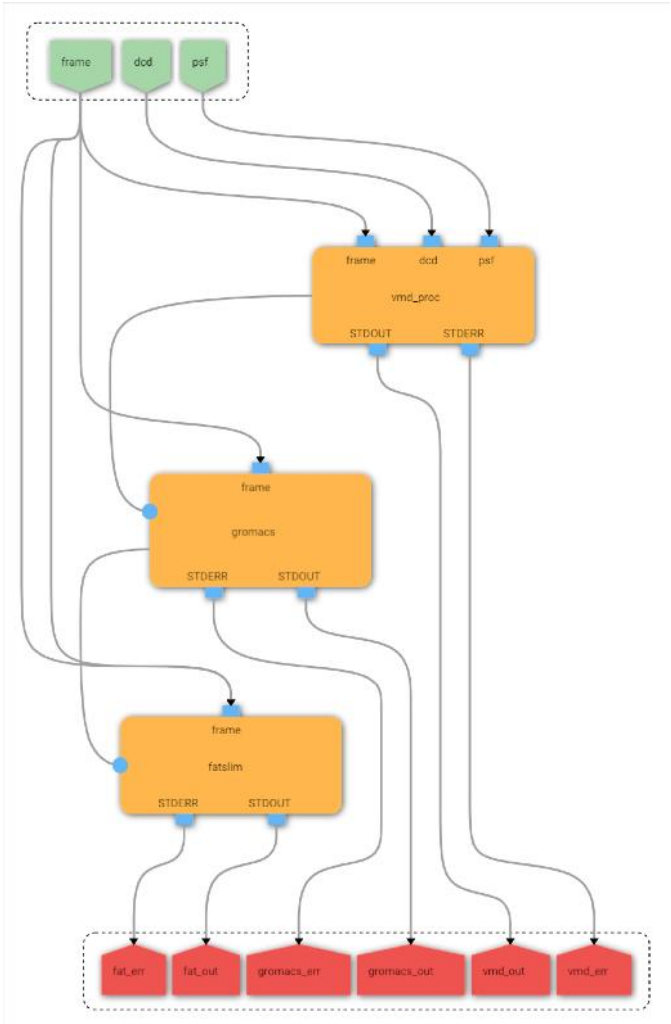
Workflow for calculation of Trp residues insertion in the membrane model

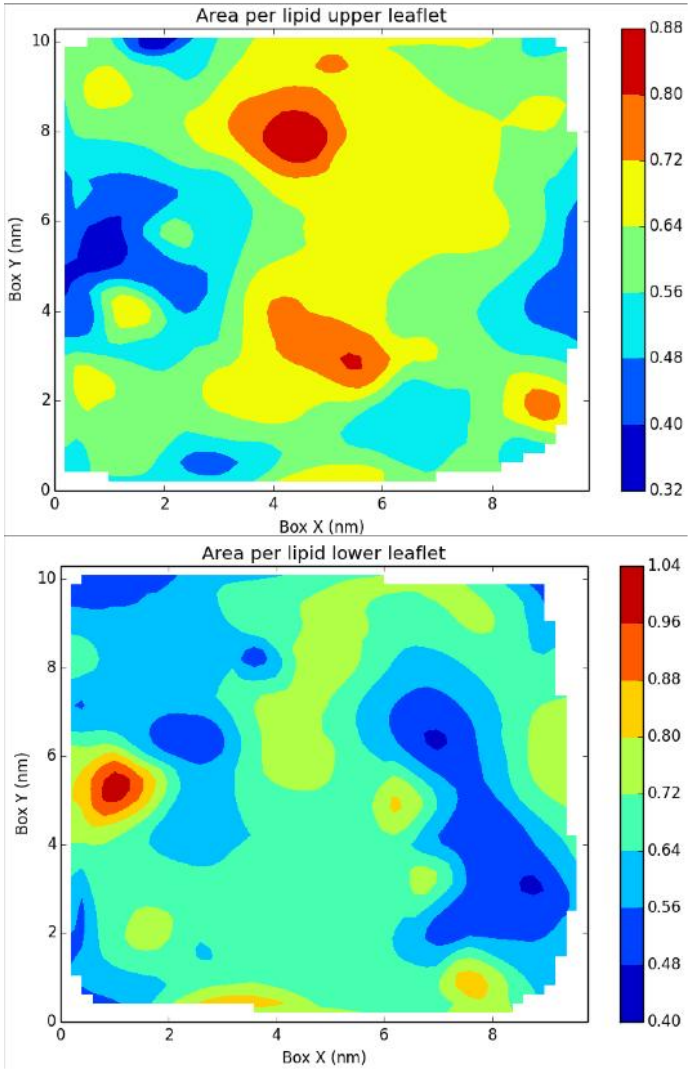




The workflow is comprised of two modules:
The first module loads the structural and trajectory files in VMD and submits a .tcl script that calculates the distance on spatial axes between the two references (e.g. P atom and center of mass of the residue) for each loaded frame of the trajectory.
The second module extracts the values on the Z axe and performs a scatter plot.

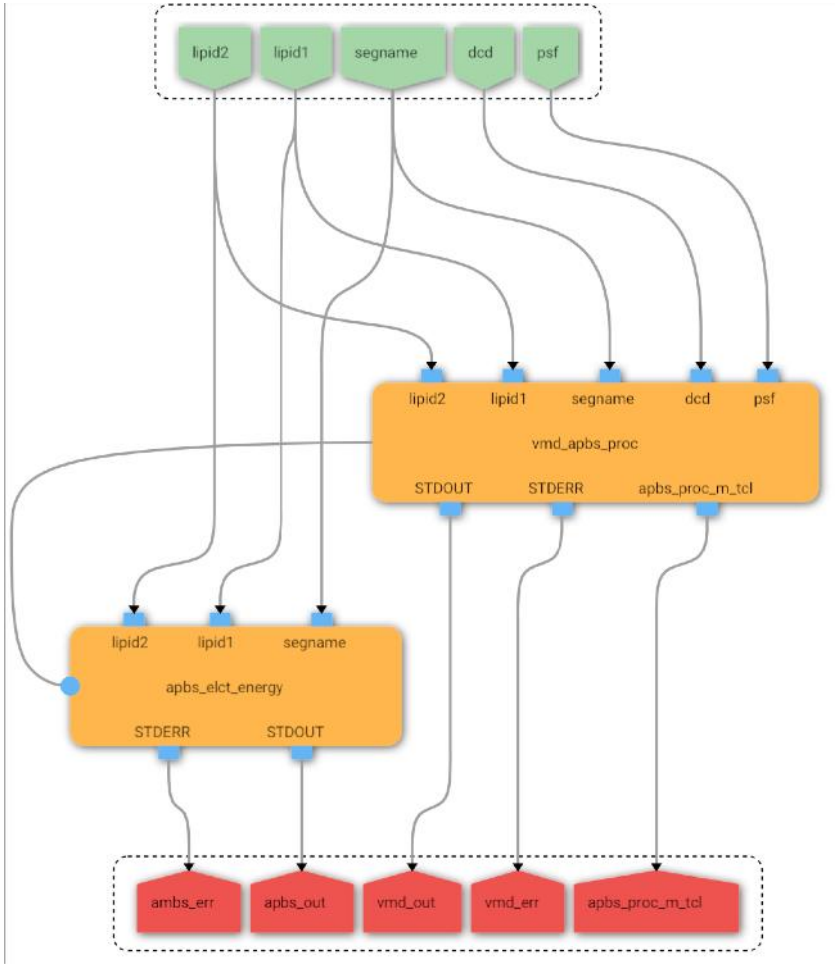
Workflow for automatic generation of area per lipid (APL) analysis

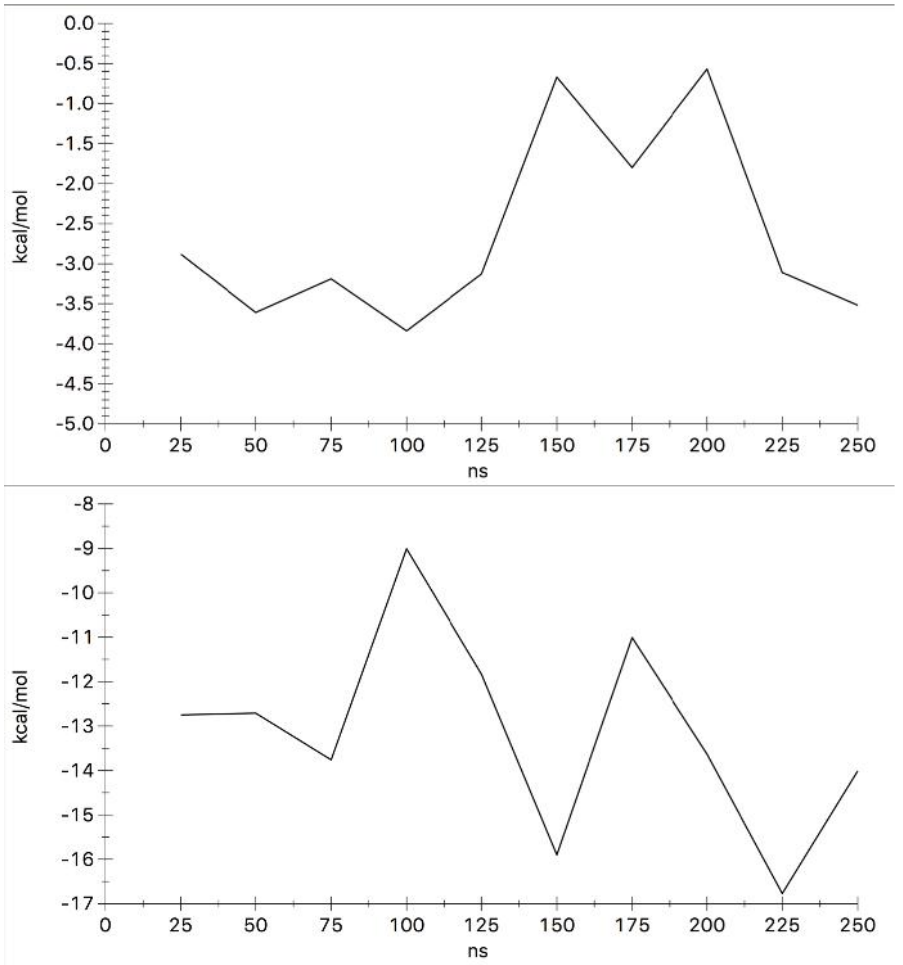




The workflow is comprised of three modules:
The first loads the structure files, and saves the desired structure frame in Gromacs.
The second module employs Gromacs program to index the atoms.
The third module employs the program FatSlim to produce an APL map.

Workflow calculating electrostatic binding energy between AMPs and membrane model





The workflow calculates the electrostatic binding energy between the AMP and the membrane lipids within 9 Å of the AMP for a total of 10 frames using APBS.

The method uses Linearized Poisson-Boltzmann equation for calculation of electrostatic binding energies using a solvent dielectric constant of 78 and solute dielectric constant set to 4, at a temperature of 298.15 K.

Conclusions

- We have developed and successfully tested workflows that automates the analysis of molecular dynamics simulations meant to investigate the activity of AMPs on lipid bilayers and we hope they will provide a real benefit to the scientific community.
- The workflows automates molecular modelling procedures that are not very complex but are very time consuming
- Other workflows that are available on the platform for analysis of molecular dynamics data are for RMSD, hydrogen bond analysis, and membrane thickness.
- The updated RoNBio molecular modelling system is available online at http://ronbio.ifin.ro

Acknowledgements

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Thank you!