Classical Molecular Dynamics Simulations Toward the Understanding of Nitroamine Binding within the NR1 S1S2 Domain

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Nitroamine compounds arising from explosive materials have proven to be harmful reagents, of which their biological activities trigger a cascade deleterious health within the nervous system of various model species. In conjuction with their highly conserved biologic responses these compounds continue to persist within various mediums of contaminated ecosystems. Thus, the study of the chemical end-points within mammalian neural networks may shed light upon nitroamine- binding pathways and preferred routes therein. This understanding may also be applied to biomonitoring rationals for the detection of exposives-contamination within within exposed organisms.

It has been shown that after 48 hours of exposure 24mg/kg of the explosive-contaminant 1,3,5-Trinitro-1,3,5-triazacyclohexane (RDX) preferentially upregulates the transcripiton of the GRIN1 and GRIN2 genes. This highly conserved effect produces large amounts of the major, possibly minor, N-methyl-D-Aspartate receptor (NMDAR) subtypes NR1 and NR2 to position themselves on the post-synaptic neurilemma. Since these subtypes arrange in themselvese in an obligatory dimer-of-dimers stoichometry, it is of major interest for us to first study the direct binding routes of the natural inhibitors glycine (NR1) and L-glutamate (NR2) within the S1S2 main binding region. These sets of understandings will provide insight into direct, competitive, and allosteric binding within the major binding domains and the tetrameric NMDAR.

Due to lack of protein crystal data regarding non-S1S2 binding domains on the NR1, we have carried out sequence homology studies with the program SwissModel's "First Approach Mode" with a BLAST E-value limit of 1.0×10^{-5}. After a 3-D model was constructed we applied the ff99 force field within the Amber8 package and compared various models of simulation to detect domain movement within the S1S2 model i.e. ~76 ps simulation in the Generalized Born (GB) solvent model @350K, 50 ps gas phase Simulated Annealing (SA) simulation @370K, and 50 ps SA simulation in the GB model @370K. Root Mean Square Deviations (RMSDs) of 30 amino acid segment positions were calculated and visualized with VMD molecular viewing software.

The results show in SwissModel that the top five homologous protein sequences to the NR1 S1S2 are the holo-forms of the NR1 S1S2 binding domain complexed with various inhibtors from respective crystal studies. Our model was built from top candidate pdbid: 1y20 and showed 2.38e -173 e-value, and a 98% Sequence identity with our target sequence. Simulation efforts showed that the first 150 amino acids in the NR1 S1S2 model confer the main mode of the S1S2 domain opening (apo-form), while the last seventy amino acid segements are quite stationary after changing simulation conditions (moderately to severe). The 50 ps SA simulation in GB solvent with the ff99 force field proved to be the most severe treatment for domain opening. This method showed degradation of secondary character within the NR1 S1S2 model. A ~14 Å gap distance was achieved when we improved to the ff03 force field. Metadynamics approaches will be further employed.