

**P012** Structure of TCP-binding sites on resting and desensitized nicotinic acetylcholine ion channels

Hugo R. Arias(1), James R. Trudell(2)  
and Michael P. Blanton(3)

(1) *Western University of Health Sciences, Pomona,  
CA 91766, USA*

(2) *Stanford University School of Medicine, Stanford,  
CA 94305, USA*

(3) *Texas Tech University Health Sciences Center, Lubbock,  
TX 79430, USA*

Thienylcyclohexylpiperidine (TCP) is a noncompetitive antagonist (NCA) of the nicotinic acetylcholine receptor (AChR) that binds with high-affinity to the resting and desensitized ion channel. A series of amantadine derivatives along with very well characterized NCAs were used to probe the structure of the TCP locus. Inhibition experiments and thermodynamic analysis yielded the following results: (1) the rank order of potencies for inhibition of [3H]TCP (or [3H]ethidium) binding to the desensitized AChR is: memantine > adamantylethylamine > adamantanemethylamine > 1- > 2-adamantanamine; (2) a similar rank order was observed for the resting AChR; (3) with exception of adamantane, these derivatives enhanced [14C]amobarbital binding and [125I]TID labelling to the resting AChR. We conclude that: (a) positively charged derivatives bind to the TCP site on the resting AChR close to position M2-20, maybe interacting with the negatively charged residue alpha-Glu262; (b) neutral adamantane prefers the barbiturate/TID domain (M2-9/13); (c) the hydrophobic environment and size of the TCP locus are nearly identical in both states; (d) the TCP site in the resting state is larger (accommodates molecules with volumes > 257 +/- 64 Å<sup>3</sup>) than the TID locus (cutoff = 333 +/- 45 Å<sup>3</sup>). Supported by Western University Intramural Grant (to HRA).