

# Acetylcholine Receptors (Nicotinic)

## Key References

- Bannon, A.W. et al. "Broad-spectrum, non-opioid analgesic activity by selective modulation of neuronal nicotinic acetylcholine receptors." *Science* **279**, 77-81 (1998).
- Changeux, J.P. et al. "Brain nicotinic receptors: Structure and regulation, role in learning and reinforcement." *Brain Res. Rev.* **26**, 198-216 (1998).
- Daly, J.W. et al. "Alkaloids from frog skin: The discovery of epibatidine and the potential for developing novel non-opioid analgesics." *Nat. Prod. Rep.* 131-135 (2000).
- Dani, J.A. "Overview of nicotinic receptors and their roles in the central nervous system." *Biol. Psychiatry* **49**, 166-174 (2001).
- Elgoyhen, A.B. et al. " $\alpha$ 10: A determinant of nicotinic cholinergic receptor function in mammalian vestibular and cochlear mechanosensory hair cells." *Proc. Natl. Acad. Sci. USA* **98**, 3501-3506 (2001).
- Holladay, M.W. et al. "Neuronal nicotinic acetylcholine receptors as targets for drug discovery." *J. Med. Chem.* **40**, 4169-4194 (1997).
- Leonard, S. et al. "The role of nicotine and nicotinic receptors in psychotherapy." in *Neuronal Nicotinic Receptors: Pharmacology and Therapeutic Opportunities* Eds. Arneric, S.P., Brioni, J.D., Wiley-Liss, New York, pp. 307-322 (1998).
- Lloyd, G.K., Williams, M. "Neuronal nicotinic receptors as novel drug targets." *J. Pharmacol. Exp. Ther.* **292**, 461-467 (2000).
- Lukas, R.J. et al. "Nicotinic acetylcholine receptors." in: *The IUPHAR Compendium of Receptor Characterization and Classification, 2nd edition*, pp. 64-77, IUPHAR Media, London, UK (2000).
- Potter, A. et al. "Acute effects of the selective cholinergic channel activator (nicotine agonist) ABT-418 Alzheimer's disease." *Psychopharmacology* **142**, 334-342 (1999).
- Ramirez-Latorre, J. et al. "Molecular composition and biophysical characteristics of nicotinic receptors." In *Neuronal Nicotinic Receptors: Pharmacology and Therapeutic Opportunities* Eds Arneric, S.P., Brioni, J.D., pp. 43-64, Wiley-Liss New York (1998).
- Wang, H.-Y. et al. "Amyloid peptide  $A\beta_{1-42}$  binds selectively with picomolar affinity to  $\alpha$ 7 nicotinic acetylcholine receptors." *J. Neurochem.* **75**, 1155-1161 (2000).

## Overview

Nicotinic acetylcholine receptors (nAChRs) are a family of ligand-gated cation channels classified on the basis of their activation by the alkaloid, nicotine, with acetylcholine (ACh) being the endogenous ligand. These pentameric conductance channels for calcium, potassium and sodium are formed from subunits; ten  $\alpha$  ( $\alpha$ 1 -  $\alpha$ 10) and four  $\beta$  ( $\beta$ 1 -  $\beta$ 4) cloned from mammalian and avian sources.  $\alpha$ 1,  $\beta$ 1,  $\gamma$ ,  $\delta$  and  $\epsilon$  subunits are associated with the neuromuscular junction nAChR. Combinations of these various subunits offer a considerable potential for diversity in both structure and associated function. Each subunit has four transmembrane spanning domains, M1 - M4, of which M2 lines the channel. The ACh binding site is formed between  $\alpha$  and adjoining subunits; thus, nAChR homomers have five putative binding sites while heteromers may typically have only two. The predominant CNS forms of nAChR, also known as neuronal nicotinic receptors (NNRs), are the  $\alpha$ 4 $\beta$ 2 and  $\alpha$ 7. The latter is presumed to form a functional pentameric homomer. The possible presence of additional unidentified subunits may be denoted by an asterisk (e.g.  $\alpha$ 4 $\beta$ 2\*). There is also evidence for an  $\alpha$ 3 $\beta$ 2\* combination. In the autonomic ganglia in the peripheral nervous system,  $\alpha$ 3 $\alpha$ 5 $\beta$ 4,  $\alpha$ 3 $\alpha$ 2 $\beta$ 4,  $\alpha$ 3 $\beta$ 2 $\beta$ 4 $\alpha$ 5 and  $\alpha$ 3 $\beta$ 2\* combinations may exist as well as  $\alpha$ 7 and  $\alpha$ 9 homomers. The  $\alpha$ 9 and  $\alpha$ 10 subunits have a restricted distribution to sensory tissues (ear, tongue and skin). The  $\alpha$ 10 subunit is only functional as a heteromer with  $\alpha$ 9.

The frog alkaloid, epibatidine, is a potent agonist at the  $\alpha$ 4 $\beta$ 2 site labeled by [ $^3$ H]-cytisine ( $K_i$  37-100 pM) and at other NNRs. This compound is 100-200-fold more potent than morphine as an analgesic, but


its interactions with the ganglionic and neuromuscular nAChRs make it toxic at doses close to its efficacious range. Anti-sense and mouse nAChR subunit knock-outs have provided definitive evidence for a role of  $\alpha$ 4 and  $\beta$ 2 subunit containing receptors in nociception, particularly at the level of the raphe nucleus.

Synthetic chemistry efforts have focused on ligands selectively acting at the  $\alpha$ 4 $\beta$ 2 and also at  $\alpha$ 7 NNRs. Newer ligands for the  $\alpha$ 4 $\beta$ 2 NNR, while having similar affinity for this receptor, have distinct pharmacological activities in animals, suggesting that the  $\alpha$ 4 $\beta$ 2 nAChR is a surrogate for a number of NNRs. ABT-418, an isoxazole isostere of nicotine, exhibits cognition enhancing activities and shows activity in adults with attention deficit hyperactivity disorder. ABT-594, an azetidine analog of epibatidine, is a potent analgesic with substantially reduced side effects in animal models as compared with epibatidine, while SIB-1508Y is a nicotine analog that shows activity in animal models of Parkinson's disease. All three compounds have potent interactions with  $\alpha$ 4 $\beta$ 2 nAChRs, but have very distinct functional profiles. At the  $\alpha$ 7 NNR, GTS-21 is a partial agonist that has cognition enhancing activity, while AR-R 17779 is a full agonist and the  $\alpha$ -conotoxin IM1 is an antagonist. SIB-1553A is selective for the  $\alpha$ 3 $\beta$ 4 subunit combination. The  $\alpha$ -conotoxins, AulB and MII, are antagonists for the  $\alpha$ 3 $\beta$ 4 and  $\alpha$ 3 $\beta$ 2/ $\alpha$ 3 $\beta$ 3\* NNRs, respectively, although their effects are somewhat species specific.

In addition to the ACh binding site, nAChRs have associated modulatory sites, including those for neurosteroids and some acetylcholinesterase inhibitors (e.g. physostigmine and galanthamine). Other

sites are located in the channel, for example, those activated by anesthetics such as lidocaine, phencyclidine and MK-801. The anthelmintic, ivermectin, potently modulates the  $\alpha$ 7 nAChR. The neurotoxic peptide,  $A\beta_{1-42}$ , is a picomolar non-competitive, use-dependent blocker. An association of  $\alpha$ -synuclein with  $\alpha$ 7 subunits has been proposed.  $\alpha$ 7 NNR modulators may thus be novel targets for Alzheimer's and Parkinson's disease therapy. The  $\alpha$ 7 NNR has also been implicated in the pathophysiology of schizophrenia.

## Acetylcholine Receptors (Nicotinic)

CURRENTLY USED NAME	Neuronal (CNS) ( $\alpha$ -Bungarotoxin insensitive)	Neuronal (CNS) ( $\alpha$ -Bungarotoxin sensitive)	Neuronal (autonomic ganglia)	Muscular
<b>SUBUNITS (ARRANGED AS PENTAMERS)</b>	$\alpha$ 4 $\beta$ 2 (major) $\alpha$ 3 $\beta$ 4 $\alpha$ 2? $\alpha$ 3 ?	$\alpha$ 7 homomers $\alpha$ 8 ? $\alpha$ 9 ? $\alpha$ 9/ $\alpha$ 10	$\alpha$ 7 homomers $\alpha$ 3 $\alpha$ 5 $\beta$ 4 $\alpha$ 3 $\alpha$ 5 $\beta$ 2 $\beta$ 4	$\alpha$ 1 $\beta$ 1 $\delta$ $\gamma$ ( $\epsilon$ )
<b>RECEPTOR SELECTIVE AGONISTS</b>	Cytisine <sup>a</sup> ( <a href="#">C 2899</a> ) RJR-2403 ( <a href="#">R-130</a> ) Epibatidine ( <a href="#">E 1145</a> ) Anatoxin-a ( <a href="#">A-224</a> ) ABT-418 A-85380 ( <a href="#">A-251</a> ) DBO-83 ( <a href="#">D 7938</a> )	Anatoxin-a ( <a href="#">A-224</a> ) DMAC GTS-21 <sup>a</sup> AR-R 17779	Epibatidine ( <a href="#">E 1145</a> ) SIB-1553A DMPP ( <a href="#">D 5891</a> )	Epibatidine ( <a href="#">E 1145</a> ) Anatoxin-a ( <a href="#">A-224</a> ) TMA ( <a href="#">T 3411</a> )
<b>RECEPTOR SELECTIVE ANTAGONISTS</b>	Mecamylamine ( <a href="#">M 9020</a> ) Dihydro- $\beta$ -erythroidine ( <a href="#">D-149</a> ) Erysodine $\alpha$ -Conotoxin AulB ( $\alpha$ 3 $\beta$ 4) $\alpha$ -Conotoxin MII ( $\alpha$ 3 $\beta$ 2)	Methyllycaconitine ( <a href="#">M-168</a> ) $\alpha$ -Bungarotoxin ( <a href="#">T 3019</a> ) $\alpha$ -Conotoxin IMI ( <a href="#">C 2461</a> )	Hexamethonium ( <a href="#">H 0879</a> ) Chlorisondamine ? Mecamylamine ? ( <a href="#">M 9020</a> ) $\kappa$ -Bungarotoxin	$\alpha$ -Bungarotoxin ( <a href="#">T 3019</a> )
<b>SIGNAL TRANSDUCTION MECHANISMS</b>	Modulation of cation channel conductance/permeability properties 			
<b>RADIOLIGANDS OF CHOICE</b>	[ <sup>3</sup> H]-Nicotine [ <sup>3</sup> H]-Epibatidine [ <sup>3</sup> H]-Cytisine	[ <sup>125</sup> I]- $\alpha$ -Bungarotoxin [ <sup>3</sup> H]-Methyllycaconitine	[ <sup>3</sup> H]-Epibatidine [ <sup>125</sup> I]- $\alpha$ -Bungarotoxin [ <sup>3</sup> H]-Methyllycaconitine	[ <sup>125</sup> I]- $\alpha$ -Bungarotoxin

### ABBREVIATIONS

**A-85380:** 3-(2[S]-Azetidinylmethoxy)pyridine  
**ABT-418:** (S)-3-Methyl-5-(1-methyl-2-pyrrolidinyl)isoxazole  
**AR-R 17779:** (-)-spiro[1-Azabicyclo[2.2.2]octane-3,5'-oxazolidin-2'-one (4a)  
**DBO-83:** 3-(6-Cl-3-yl)-Diazabicyclo(3.2.1)octane  
**DMAC:** 3-(4)-Dimethylaminocinnamylidene anabaseine  
**DMPP:** N,N-Dimethyl-N'-phenyl-piperazinium iodide  
**GTS-21:** [3-(2,4-Dimethoxybenzylidene)-anabaseine  
**RJR-2403:** N-Methyl-4-(3-pyridinyl)-3-buten-1-amine  
**SIB-1553A:** 4-[2-(1-Methyl-2-pyrrolidinyl)ethyl]thio]phenyl hydrochloride  
**TMA:** Tetramethylammonium