BNC375, A Novel Positive Allosteric Modulator of the α7 Nicotinic Acetylcholine Receptor, Exhibits Cognitive Enhancement in Rodent Behavioural Models

Cholinergic neurotransmission has long been implicated in processes of attention, cognition, and learning and memory. The α7 acetylcholine nicotinic receptor (α7 nAChR) is a promising drug target for diseases involving cognitive impairment such as Alzheimer’s and schizophrenia. α7 nAChR positive modulators (α7 PAMs) are a novel class of drugs offering advantages over α7 full/partial agonists. In contrast to α7 agonists, α7 PAMs do not affect α7 nAChRs by themselves and do not desensitize them. Instead, α7 PAMs work via amplification of responses induced by intrinsic agonist which preserves spatio-temporal signalling patterns.

1. BNC375 is a potent and efficacious α7 nAChR positive allosteric modulator.

2. Effects of BNC375 on ACh concentration-response curve

3. Kinetics of α7 nAChR-mediated currents and re-activation of desensitized receptors compared between BNC375 and a classical type II α7 PAM PNU-120596

4. BNC375 demonstrates in vivo cognitive enhancing properties in rodent behavioural models

METHODS

Electrophysiology:
GH4C1 cells stably expressing rat α7 nAChRs were patch-clamped in the recording chamber of 16-channel re-usable Dynaflow® ReSolve chips using EPC10 USB amplifier (HEKA Electronic, Germany). Extracellular solution contained NaCl - 117 mM, KCl - 5 mM, CaCl2 - 2.5 mM, MgCl2 - 1 mM, HEPES - 10 mM, D-Glucose - 15 mM, pH ~7.4. Thin wall borosilicate glass electrodes (Harvard Apparatus) were pulled to a resistance of 2-4 MΩ when filled with intracellular solution (0.5 M glucose - 120 mM KCl - 5 mM HEPES - 10 mM EETG - 10 mM MgCl2 - 1 mM ATP - 2 mM pH ~7.2). Cells were held at ~70°C. Cells with series resistance below 15 MΩ were kept and 40% compensation was applied routinely.

The recording protocol consisted of obtaining two control ACh responses (Epeak, peak, 250 ms pulse) prior to 30 s pre-incubation with a tested compound (3 µM) followed by 250 ms co-application of 3 µM compound plus EC20 ACh. Dose-responses for selected compounds were obtained by a continuous compound series resists below 15 MΩ were kept and 40% compensation was utilized routinely.

Current amplitudes along with net charge carried (area under curve, AUC) were measured in Patchmaster software (HEKA Elektronik, Germany) and percentage of peak current and AUC potentiation by test compounds was calculated using the above mentioned formula. Dose-responses for selected compounds were fitted and plotted in Prism4/5 (GraphPad Software, Inc., CA).

Statistics:
Statistical analyses were performed using the student’s t-test. P values indicating significant difference to scopolamine treatment: ^p≤0.05, ^^ p ≤ 0.01, ^^^ p ≤ 0.001, ^^^^^ p ≤ 0.0001. P values representing significant difference to vehicle treatment only: * p ≤ 0.05, ** p ≤ 0.01, *** p ≤ 0.001, **** p ≤ 0.0001.

CONCLUSIONS

- BNC375 is an α7 nAChR PAM
- Effective across a wide range of agonist concentrations (from subthreshold to saturating).
- Favourable type I-like kinetics mainly affecting the peak α7 current with little effect on current desensitization.
- Demonstrates efficacy in animal models of episodic and working memory with a broad therapeutic window (100 fold).
- Matches performance of Donepezil.