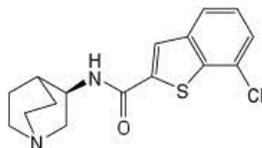


EVP-6124

EVP-6124 is a novel synthetic molecule that was found to bind with high affinity to $\alpha 7$ nAChRs. Recent animal data indicate that EVP-6124 acts as a potent partial and selective agonist at $\alpha 7$ nAChRs. EVP-6124 has an excellent brain to plasma exposure ratio and has shown excellent efficacy and potency in a number of animal models of cognition.

(R)-7-chloro-N-(quinuclidin-3-yl)benzo[b]thiophene-2-carboxamide



Source Data:

Prickaerts J, van Goethem NP, Chesworth R, Shapiro G, Boess FG, Methfessel C, Reneerkens OA, Flood DG, Hilt D, Gawryl M, Bertrand S, Bertrand D, König G. EVP-6124, a novel and selective $\alpha 7$ nicotinic acetylcholine receptor partial agonist, improves memory performance by potentiating the acetylcholine response of $\alpha 7$ nicotinic acetylcholine receptors. Neuropharmacology. 2012 Feb;62(2):1099-110. Epub 2011 Nov 10. PubMed PMID: 22085888.

Pharmacokinetics and Brain Penetration

EVP-6124 was found to bind moderately to rat plasma proteins. After a single dose of 0.3 mg/kg, p.o., T_{max} was at 4 h in plasma and 2 h brain, although the brain concentrations remained similar between 2 and 8 h. EVP-6124 demonstrated good brain penetration after oral administration, with brain:plasma ratios were 1.7–5.1 between 1 and 8 h, which supported further study of the compound in cognitive tests. The corresponding brain free drug concentrations were between 0.14 and 0.24 nM up to 8 h after treatment with EVP-6124. The unbound concentration of EVP-6124 was assumed to reflect the concentration available for binding to brain $\alpha 7$ nAChRs.

Receptor Binding Profile

EVP-6124 bind with high affinity to $\alpha 7$ nAChRs in rat brain membranes and displaced two radioactive ligands specific for $\alpha 7$ nAChRs, [3 H]-MLA ($K_i = 9.98$ nM) and [125 I]- α -bungarotoxin ($K_i = 4.33$ nM) binding in rat brain homogenates. EVP-6124 was approximately 300 fold more potent than the natural agonist ACh ($K_i = 3$ μ M).

The high affinity of EVP-6124 for $\alpha 7$ nAChRs, with an EC_{50} in the range of 0.16–0.39 μ M, suggested that therapeutic concentrations should be expected in the high nanomolar or low micromolar range, if the compounds were to act solely as receptor agonists that required near to full receptor occupancy.

The specificity of EVP-6124 for $\alpha 7$ nAChRs was confirmed by the absence of displacement of [3 H]-cytisine from $\alpha 4\beta 2$ nAChRs by 10 μ M EVP-6124, suggesting that the affinity of EVP-6124 for these heteromeric receptors was at least 1000 fold lower than the affinity of nicotine ($K_i = 8$ nM) for the $\alpha 4\beta 2$ nAChRs labeled by [3 H]-cytisine.

EVP-6124 inhibited the 5-HT₃ receptor by 51% at 10 nM, the lowest concentration tested (data not shown). This antagonist activity of EVP-6124 at 5-HT_{3A} receptors, homologues of $\alpha 7$ nAChRs, is a common feature of some of the recently characterized compounds, such as AZD0328 and RG3487 and could prove beneficial in reducing the potential emetic effects of nicotinic agonists.

Selectivity of EVP-6124 was further confirmed using electrophysiological characterization of the rat $\alpha 3\beta 4$, $\alpha 4\beta 2$, and muscle $\alpha 1\beta 1\gamma \delta$ receptors. There was no detectable agonist activity at concentrations

up to 100 μM . Antagonist activity was, however, observed at the rat $\alpha 3\beta 4$ receptor with an IC_{50} of 16 μM (data not shown).

EVP-6124 at a 10 μM concentration also lacked appreciable interactions with more than 60 molecular targets in a selectivity screening panel that included receptors, ion channels, and amine transporters.

Memory and Cognition

The memory enhancing effects of EVP-6124 *in vivo* were demonstrated in the object recognition test in a test of short-term memory impairment caused by scopolamine (0.1 mg/kg) and in a test of natural forgetting, with a 24 h retention interval between the two trials. EVP-6124 (0.1, 0.3, and 1.0 mg/kg) improved memory in a dose-dependent manner, reaching a peak of activity at brain concentrations in the low nanomolar range.

EVP-6124 (0.1, 0.3, and 1.0 mg/kg) also improved recognition of the familiar object and prevention of natural forgetting. In addition, the effect of 0.3 mg/kg of EVP-6124 in the natural forgetting test was blocked by administration of the selective $\alpha 7$ nAChR antagonist MLA (0.3 mg/kg, i.p. or 10 μg , i.c.v.). These studies indicated that the pro-cognitive effect of EVP-6124 was specifically mediated via brain $\alpha 7$ nAChRs.

EVP-6124 improved memory consolidation when administered immediately after first trial in an object recognition test.

Since treatment with $\alpha 7$ nAChR agonists may benefit AD patients, and they are often treated with an acetylcholine esterase inhibitor (AChEI), the potential beneficial interaction between AChEIs and EVP-6124 were investigated. The combined treatment of sub-efficacious doses of EVP-6124 (0.03 mg/kg, p.o.) and AChEI (donepezil; 0.1 mg/kg, p.o.) completely reversed the scopolamine-induced deficit in the object recognition test.

Co-agonist Mechanism - *Interaction at the cellular level*

Sustained exposure to a low concentration of EVP-6124 in the sub-nanomolar range (0.3 nM) potentiated ACh-evoked currents (40 μM) indicating the probable co-agonistic behaviour of EVP-6124 and ACh at $\alpha 7$ nAChRs that can account for the pro-cognitive effects observed in animals. While EVP-6124 in the low nanomolar range (3 nM) reduced ACh-evoked currents that was attributable to receptor desensitization.

Activation of $\alpha 7$ nAChRs by exposure to a low agonist concentration of EVP-6124 utilizing this co-agonist mechanism, is expected to increase the drug safety margin, to minimize undesired interactions with other receptors, and to open new and promising therapeutic avenues in combination with classical acetylcholine esterase inhibitors at lower than typically prescribed doses.

Clinical Trials

About 1,700 people had taken the drug in the company's trials, including those targeting schizophrenia and Alzheimer's disease, with no safety problems that the company considers significant.

Four clinical studies in >125 **healthy normal human subjects** have been completed with EVP-6124, including a single-ascending-dose study, a 14-day multiple-ascending-dose study, a 21-day, multiple-dose study, and a single-dose relative bioavailability, food and gender effect study.

EVP-6124 exhibited linear kinetics over the range of 1 to 180 mg and demonstrated a half-life suitable for once daily dosing. EVP-6124 appears to be safe and well-tolerated for up to 21 days as measured by adverse events, vital signs, continuous cardiac monitoring, physical examination, and clinical laboratory evaluations. In addition, in normal volunteers, EVP-6124 demonstrated pro-cognitive effects (CogState testing) in various cognitive domains including executive function.

Alzheimer's disease

Phase 1b Study

The safety and efficacy of EVP-6124 was assessed in a Phase 1b study of 48 mild to moderate Alzheimer's disease patients 60-80 years of age, on stable donepezil or rivastigmine therapy. Patients were dosed with placebo or two different doses of EVP-6124 (0.3 or 1.0 mg/d) for 28 days. Safety was evaluated by adverse events, ECG, and clinical laboratory measures. Cognitive effects were measured by CogState computerized cognitive testing and a subset of NTB scales.

EVP-6124 appeared to be safe and well tolerated with no significant adverse events reported more frequently in treated versus placebo patients; there were no SAEs reported. Subjects exposed to EVP-6124 in addition to donepezil or rivastigmine showed an increase in cognitive function, primarily in the domains of non-verbal learning, memory, and executive function. These data suggest that EVP-6124 administered to Alzheimer's disease patients on stable cholinesterase inhibitor therapies, may have potential benefit and that further study in this patient population is indicated.

Source Data:

Hilt D, Safirstein B, Hassman D, Apter J, Thein S, Maruff P, Harrison J, Gawryl M, Koenig G. (2009). *EVP-6124 - Safety, Tolerability and Cognitive Effects of a Novel alpha7 Nicotinic Receptor Agonist in Alzheimer's Disease Patients on Stable Donepezil or Rivastigmine Therapy. ICAD 2009, Vienna, Austria, July 11-16, 2009.*
http://www.envivopharma.com/pdf/Hilt_etal_A7_ICAD2009.pdf

Phase 2b Clinical Trial

The six-month, double-blind, placebo-controlled study enrolled 409 patients and evaluated three doses of EVP-6124 taken once per day - 0.3 mg, 1.0 mg and 2.0 mg - against placebo in patients with mild to moderate Alzheimer's disease. The trial included patients taking acetylcholinesterase inhibitor (AChEI) treatments (donepezil and rivastigmine) as well as patients not taking AChEIs. The 2.0 mg dose group (after 23 weeks of dosing) met both of the trial's primary endpoints with statistically significant positive effects on cognition ($p=0.0189$), as measured by the Alzheimer's Disease Assessment Scale-Cognitive subscale-13 (ADAS-Cog-13) and clinical function ($p=0.0253$) as measured by the Clinical Dementia Rating Scale Sum of Boxes (CDR-SB).

The data also showed statistically significant results across secondary endpoints of other cognitive and clinical measures including the cognition composite ($p=0.0037$), memory composite ($p=0.0088$) and executive function composite ($p=0.0427$).

The 2.0 mg group also showed positive effects versus placebo, as measured by the Mini-Mental State Examination (MMSE) ($p=0.0955$), and the Alzheimer's Disease Cooperative Study-Activities of Daily Living (ADCS-ADL) ($p=0.092$). The 0.3 mg and 1.0 mg doses showed positive trends, but not statistically significant improvements over placebo.

EVP-6124 was generally safe and well-tolerated over the trial's six-month dosing period with some mild to moderate gastrointestinal side effects (mild nausea and constipation) reported in a minority of patients in both the 1.0 mg and 2.0 mg dose groups.

*EnVivo will continue to advance EVP-6124 and plans to initiate a **Phase 3 Clinical Trials** in Alzheimer's disease in 2013.*

Source Data:

EnVivo Pharmaceuticals Announces Statistically Significant Improvement in Cognition and Clinical Function in Phase 2b Clinical Trial in Alzheimer's Disease
Data Presented at Alzheimer's Association International Conference (AAIC) 2012, July 14-19, 2012 in Vancouver, British Columbia.
<http://www.envivopharma.com/news-item.php?id=40>

Schizophrenia

Phase 2b Clinical Trial

This randomized, double-blind, placebo-controlled, parallel, 12-week Phase 2b trial evaluated the safety and efficacy of two doses of EVP-6124 (0.3 mg and 1.0 mg per day) versus placebo in chronic schizophrenia patients on stable second-generation antipsychotic drugs (except clozapine). Each patient was treated for three months and a total of 319 patients were enrolled in the U.S., Russia, Ukraine and Serbia. The trial's primary endpoint was overall cognition as measured by the CogState overall cognitive index and important secondary endpoints included cognition as assessed by the MCCB battery, clinical function (as measured by the SCoRS) and positive and negative symptoms (measured by PANSS). Safety and tolerability were also assessed.

The data showed that EVP-6124 had a clinically meaningful and statistically significant impact on patients' overall cognition - the trial's pre-specified primary endpoint - when taken in combination with second-generation antipsychotics and as measured by the full CogState overall cognitive index, or "OCI" ($p=0.05$ for all patients treated with EVP-6124 versus placebo.) This positive effect on the OCI was supported by a strong positive trend for improved cognition on the MCCB Battery of cognition tests, which were conducted on all U.S. patients in the trial.

Additionally, results from this Phase 2b trial demonstrated that patients treated with EVP-6124 (1.0 mg) showed clinically meaningful and statistically significant effects in key secondary endpoints: improvement in clinical function ($p=0.011$; as assessed by the Schizophrenia Cognition Rating Scale (SCoRS)) and reduction of the negative symptoms of schizophrenia ($p=0.028$; as measured by the Negative Symptom Scale of the Positive and Negative Symptoms Scale (PANSS)).

Importantly, EVP-6124 was generally safe and well-tolerated over the trial's three-month dosing period. The most commonly reported adverse events were headache, nausea and nasopharyngitis (all less than four percent), there were no drug-related serious adverse events.

*En-Vivo is encouraged by the results across both dose arms and the broad range of primary and secondary endpoints. They are now working with leading clinicians and researchers to finalize the design of their **Phase 3 Clinical Trials** to maximize these learning.*

Source Data:

<http://www.envivopharma.com/news-item.php?id=32>

No Study Results Posted on ClinicalTrials.gov for this Study
<http://clinicaltrials.gov/ct2/show/NCT00968851>

EnVivo Pharmaceuticals on April, 2009 announced an agreement with Mitsubishi Tanabe Pharma Corporation (MTPC). Under the terms of the agreement, MTPC has obtained exclusive rights to develop and commercialize EnVivo's EVP-6124 in Japan, Korea, Taiwan, Indonesia and several other Asian markets.