Available online at www.sciencedirect.com
ScienceDirect
Bioorganic & Medicinal Chemistry Letters 17 (2007) 6734–6738
Bioorganic & Medicinal Chemistry Letters

Bis-azaaromatic quaternary ammonium salts as antagonists at nicotinic receptors mediating nicotine-evoked dopamine release: An investigation of binding conformation

Guangrong Zheng, Zhenfa Zhang, Marharyta Pivavarchyk, Agripina G. Deaciuc, Linda P. Dwoskin and Peter A. Crooks*

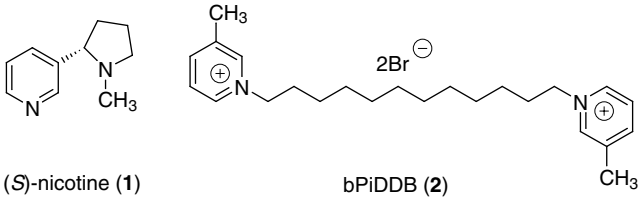
Department of Pharmaceutical Sciences, College of Pharmacy, University of Kentucky, Lexington, KY 40536-0082, USA

Received 17 August 2007; revised 15 October 2007; accepted 15 October 2007
Available online 18 October 2007

Abstract—A series of conformationally restricted bis-azaaromatic quaternary ammonium salts (**3** and **4**) have been designed and synthesized in order to investigate the possible binding conformations of *N*,*N*′-dodecane-1,12-diyl-bis-3-picolinium dibromide (bPiDDB; **2**), a compound which potently inhibits neuronal nicotinic acetylcholine receptors (nAChRs) mediating nicotine-evoked dopamine release. The preliminary structure–activity relationships of these new analogues suggest that bPiDDB binds in an extended conformation at the nAChR binding site, and that flexibility of the linker may be important for its high potency in inhibiting nAChRs mediating nicotine-evoked dopamine release.

Tobacco contains one of the most widely abused psychoactive substances in the world, that is, (*S*)-nicotine (**1**; Fig. 1), which is believed to be primarily responsible for tobacco dependence.[1,2] Like many abused drugs, nicotine addiction has been linked to the release of the neurotransmitter, dopamine (DA).[3–5] Nicotine-evoked DA release, which is thought to be responsible for reward, leading to addiction, is believed to result from activation of presynaptic neuronal nicotinic acetylcholine receptors (nAChRs).[6–12] Nicotine stimulates all known nAChR subtypes[13] and upon activation, these receptors modulate the release of various neurotransmitters.[14–16] The subunit composition of nAChR subtypes responsible for mediating nicotine-evoked DA release has not been elucidated conclusively.[17] In this regard, subtype-selective nAChR antagonists, which inhibit nicotine-evoked DA release, may have potential as novel treatments for nicotine addiction.[18–21]

Previous work in our laboratories has led to the discovery of *N*,*N*′-dodecane-1,12-diyl-bis-3-picolinium dibromide (bPiDDB; **2**; Fig. 1), which potently inhibited nAChR subtype(s) mediating nicotine-evoked [^{3}H]DA release from superfused rat striatal slices in vitro (IC_{50} = 5 nM), and did not interact at the ligand binding site of either α4β2* or α7* nAChRs.[22,23] In vivo microdialysis studies demonstrated that pretreatment with bPiDDB dose-dependently reduced the increase in extracellular DA in rat nucleus accumbens produced by acute or repeated nicotine treatment.[24] In addition, we have demonstrated that despite the cationic nature and polarity of the molecule, bPiDDB is brain bioavailable, due to its facilitated transport via the blood–brain barrier choline transporter.[25] Moreover, behavioral studies in rats showed that bPiDDB dose-dependently decreased intravenous nicotine self-administration, but not sucrose-maintained responding, suggesting a specific inhibition of nicotine reward.[26] Taken together, bPiDDB and its analogues represent lead compounds



(*S*)-nicotine (**1**) bPiDDB (**2**)

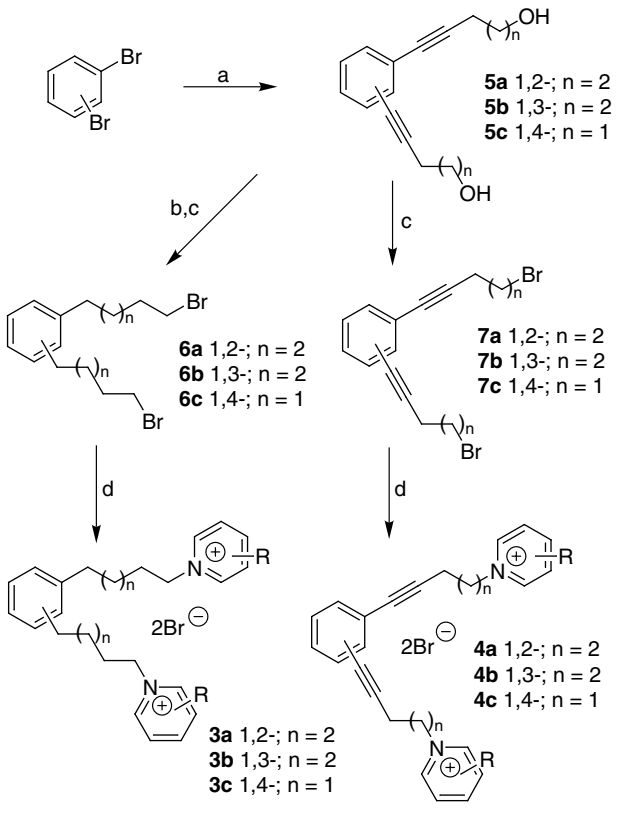
Figure 1. Structures of (*S*)-nicotine (**1**) and bPiDDB (**2**).

Keywords: Nicotinic acetylcholine receptor; Quaternary ammonium; Dopamine release; Nicotine addiction.
* Corresponding author. Tel.: +1 859 257 1718; fax: +1 859 257 7585; e-mail: pcrooks@email.uky.edu

0960-894X/$ - see front matter © 2007 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmcl.2007.10.052

for the development of a new class of therapeutic agents for the treatment of tobacco dependence.

bPiDDB (**2**) contains two 3-picolinium head groups connected via an *N*-*N*-12-methylene linker unit; thus, it is a highly flexible, di-cationic molecule. A better understanding of the potential binding conformation of bPiDDB at nAChRs responsible for mediating nicotine-evoked DA release may provide some insight into the nature of the pharmacophore requirements for optimal inhibition of this receptor. Energy minimization calculations reveal that the lowest energy conformation of bPiDDB is one in which the *N*-*N* alkyl linker is fully extended.[27] However, the low energy conformations of ligand molecules may not always reflect the conformation in which the molecule binds to the receptor. Utilizing this extended or linear low energy conformation, a QSAR model utilizing a back-propagation artificial neural network approach has been constructed and has been found to afford good predictivity for inhibition of nicotine-evoked DA release.[27] The hypothesis that bPiDDB binds to nAChR subtype(s) mediating nicotine-evoked DA release in a 'linear' or 'extended' conformation is supported by experimentally determined data[23] showing that analogues with shorter *N*-*N* alkyl linker units have decreased inhibitory potency compared to bPiDDB. In the present study, we designed a series of model compounds which mimic different binding conformations of bPiDDB to provide further information on the active conformation of this novel nAChR antagonist.



Scheme 1. Reagents and conditions: (a) 3-butyn-1-ol or 4-pentyn-1-ol, $Pd(PPh_3)_2Cl_2$, CuI, Et_3N, 80 °C; (b) H_2, 10% Pd/C, MeOH, 45 psi, rt; (c) PPh_3, CBr_4, CH_2Cl_2, 0 °C; (d) azaaromatic compounds, 60 °C.

The design of the model compounds incorporated a benzene ring into the middle of the *N*-*N* linker unit, allowing a variety of arrangements of the two methylene linker units around the aromatic ring (i.e., 1,2-, 1,3-, or 1,4-positions) (see **3a**, **3b**, **3c**, respectively, Scheme 1), and thereby, constraining these molecules into an 'extended' or 'angular' geometry. In this respect, rigidification has been used extensively in drug design to lock ligands into a desired conformation or geometry with the goal of increasing activity and selectivity of the analogues. In the current series of compounds, a triple bond also has been introduced into each of the linker units attached to the central phenyl ring to provide further restriction of conformational freedom and geometry (**4a**, **4b**, **4c**, Scheme 1).

The requisite compounds of general structure **3** and **4** were prepared by the route shown in Scheme 1. The synthesis was initiated with Sonogashira coupling[28] of 1,2-, 1,3-, or 1,4-dibromobenzene with 4-pentyn-1-ol (for the 1,2- and 1,3-isomers) or 3-butyn-1-ol (for 1,4-isomers) to afford compound **5a** (1,2-isomer), **5b** (1,3-isomer), or **5c** (1,4-isomer). In the synthesis of compound **5a**, the reaction required a prolonged heating time (at 80 °C) for 6 days for completion. Compounds **5a**–**5c** and their corresponding Pd/C-catalyzed hydrogenation products were transformed into dibromides **7a**–**7c** and **6a**–**6c**, respectively, by bromination using PPh_3/CBr_4.[29] Alkylation of the corresponding azaaromatic free bases, including 2-picoline, 3-picoline, 4-picoline, and nicotine, using the above obtained dibromide **6a**, **6b**, **6c**, **7a**, **7b**, or **7c**, afforded the corresponding bis-azaaromatic quaternary ammonium compound **3a**, **3b**, **3c**, **4a**, **4b**, or **4c**, respectively (Table 1). Among these analogues, the 1,2-isomers in both the **3** and **4** series (**3a** and **4a**, respectively) and 1,3-isomers in series **4** (i.e., **4b**) represent 'angular' conformations of bPiDDB, whereas the 1,3-isomers in series **3** (i.e., **3b**, see Fig. 2 for the difference between **3b** and **4b**) and the 1,4-isomers in both series **3** and **4** (i.e., **3c** and **4c**, respectively) represent 'extended' conformations.

These analogues were evaluated for their ability to inhibit nicotine-evoked [^{3}H]DA release from superfused rat striatal slices and to inhibit (*S*)-(−)-[^{3}H]nicotine ([^{3}H]NIC) binding (probing $\alpha4\beta2^*$ nAChRs) and [^{3}H]methyllycaconitine ([^{3}H]MLA) binding (probing $\alpha7^*$ nAChRs) to rat brain membranes. [^{3}H]NIC and [^{3}H]MLA binding assays were performed using 3 and 2.5 nM concentrations of radioligand, respectively, and 10 μM NIC and 10 μM MLA to assess nonspecific binding to whole brain membranes.[30] Analogues were evaluated at a probe concentration of 100 nM. The amount of inhibition is presented as a percentage of radioligand binding in the absence of analogue (control, Table 1). Analogue-induced inhibition of nicotine-evoked [^{3}H]DA release was determined using 10 μM nicotine and 100 nM analogue.[30] Inhibition is presented as a percentage of the response to nicotine under control conditions (in the absence of analogue) and the values are provided in Table 1.

All of the analogues containing 2-, 3-, or 4-picolinium head groups showed little or no affinity for either