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New analogues of agmatine with higher affinity to imidazoline receptors

Adam P. Treder [a], Ryszard Andruszkiewicz [a,*], Włodzimierz Zgoda [b], Celeste Ford [c], Alan L. Hudson [c]

[a] Department of Pharmaceutical Technology and Biochemistry, The Faculty of Chemistry, Gdańsk University of Technology, Narutowicza St. 11/12, 80-952 Gdańsk, Poland
[b] Department of Medicinal Chemistry, Medical University of Gdańsk, Dębinki St. 1, 80-952 Gdańsk, Poland
[c] Department of Pharmacology, 947 Medical Sciences Building, University of Alberta, Edmonton, Canada T6G 2R3

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ABSTRACT

Compilation of agmatine structure and imidazoline ring leads to a new family of imidazoline/α_2-adrenoceptor ligands, 4(5)-(2-aminoethyl)imidazoline derivatives. Constraining of the guanidine moiety into heterocyclic ring improved the affinities of the resultant fusion compounds in comparison to agmatine itself. In this work, the synthetic approach and results for I_1, I_2, and α_2-adrenoceptors affinities are reported.

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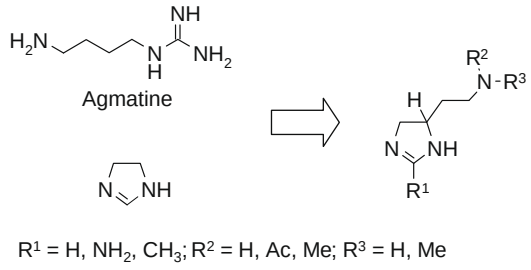
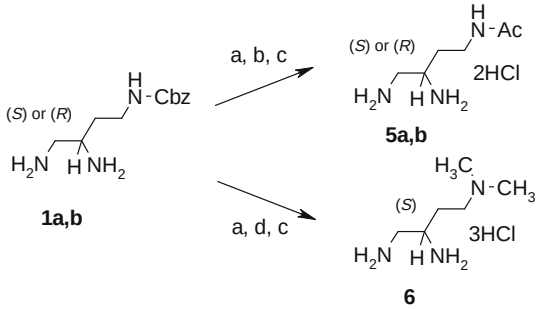
Imidazolines belong to a numerous and long-known family of compounds with wide therapeutic applications. They are active ingredients of many antiallergic, antihypertensive, antidiabetic, sympatholytic, analgetic and anxiety-relieving drugs. The therapeutic potential of these agents is mainly mediated by two types of receptors: α_2-adrenoceptors and imidazoline (I) receptors. According to the most recent hypothesis three types of imidazoline receptors (I_1, I_2, and I_3) and α_2-adrenoceptors mediate the desired and unwanted actions of the imidazoline drugs. I_1 receptors in Rostral Ventrolateral Medulla mediate the sympathetic system responses of clonidine-like drugs. The second major binding sites, I_2 receptors, are mainly located on monoamine oxidase (MAO) A and B on the outer membrane of mitochondria and are widely distributed in CNS and in peripheral tissues. This type of imidazoline receptors has an influence on endogenic amines/neurotransmitters metabolism. The third type of binding sites, I_3-receptors, are located on pancreatic β-cells and affect insulin secretion. Moreover, the hypotensive effect of imidazoline ligands is attributed to both α_2-adrenoceptors and I_2 receptors.[1]

There are several reported proposals of natural imidazoline receptors ligands including β-carbolines[2] and imidazoleacetic acid ribotide.[3] In early 90s, agmatine, a decarboxylated metabolite of arginine, that was long known to be synthesised in plants, bacteria, many invertebrates and fish,[4] was discovered in mammalian tissues.[5,6] This natural polyamine was proposed as an endogenous ligand of the I-receptors and a novel neurotransmitter in mammalian brain.[5,7,8] Agmatine also affects several other receptors (NMDA, nicotinic, 5-HT, and opioid receptors)[7,9–11], and enzymes (NOS, ODC, and SSAT).[13] This diversity of molecular targets is responsible



Scheme 1. 4(5)-(2-Aminoethyl)imidazolines as a compilation of agmatine structure and imidazoline ring.



Scheme 2. Synthesis of N^4-acetyl and N^4,N^4-dimethyl-1,2,4-triaminobutanes. Reagents and conditions: (a) Boc_2O, Et_3N, MeOH, then 5% Pd/C H_2; (b) Ac_2O, AcOH; (c) HCl/dioxane; (d) 37% HCHO/H_2O, $NaBH_3CN$, CH_3CN.

* Corresponding author. Tel.: +48 58 3471493; fax: +48 58 3471144.
E-mail address: ryszarda@chem.pg.gda.pl (R. Andruszkiewicz).

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for various biological properties of agmatine:[14] antihypertensive,[15] antidiabetic,[16] anticonvulsant,[17] antidepressant,[18] antinociceptive,[9,10,19,20] anti- or proapoptotic.[11,12,21]

We have developed a working hypothesis, that compilation of the imidazoline ring and the structure of agmatine (the natural ligand of imidazoline receptors) into one formula, may improve biological properties of the resultant compounds (Scheme 1). In this study, we describe a new group of previously unpublished agmatine/imidazoline analogues and I receptors ligands, 4(5)-(2-aminoethyl)imidazoline derivatives.

The synthesis of 4(5)-(2-aminoethyl)imidazoline derivatives was performed using the series of reactions shown in Schemes 2 and 3. (2*S*)- **1a** or (2*R*)-N^4-benzyloxy-1,2,4-triaminobutane **1b** as well as the (2*S*)- and (2*R*)-N^1,N^2-di-*tert*-butoxycarbonyl-1,2,4-triaminobutanes were obtained from L- or D-glutamic acid as we described previously.[22] N^4-Acetyl **5a and 5b** and N^4,N^4-dimethyl-1,2,4-triaminobutanes **6** were synthesised from **1a and 1b** via N^1,N^2-di-*tert*-buthoxycarbonyl-1,2,4-triaminobutane using standard acylation and *N*,*N*-dimethylation[23] methods (Scheme 2).

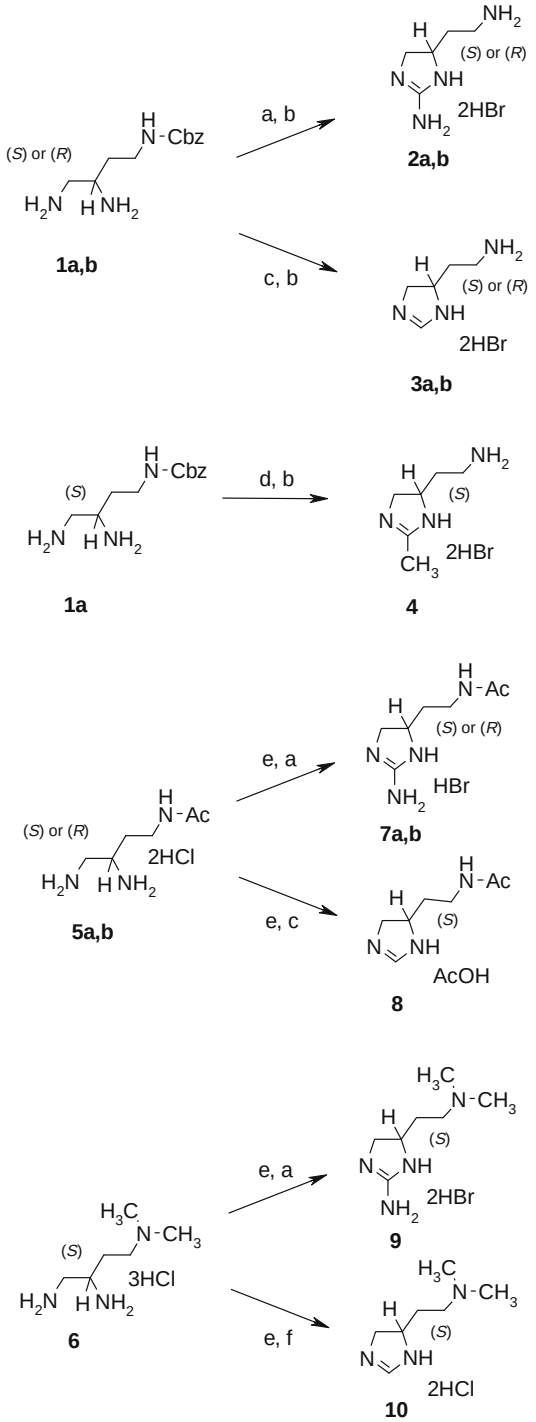
The cyclisation reactions were performed in accordance with reported imidazoline ring synthetic procedures using formamidine acetate,[24] ethyl orthoformate,[25,26] cyanogen bromide[24,27] and iminoesters.[28–31] The benzyloxycarbonyl groups protecting the aminoethyl moiety of imidazolines were cleaved with HBr in glacial acetic acid.

Binding affinity and selectivity of the newly prepared imidazoline compounds toward the α_2-adrenoceptors and imidazoline (I_1 and I_2) receptors were evaluated in in vitro assays. Clonidine hydrochloride was enclosed as reference compound. The affinity studies were performed using kidney and brain male Sprague–Dawley rats membranes as was reported recently.[32] The results are presented in Table 1.

Affinity of agmatine to the imidazoline/α_2-adrenoceptors is rather low, but this has not prevented this common polyamine being considered as a natural ligand of imidazoline receptors and a functional neurotransmitter. The distributions and levels of agmatine found in various brain areas correlate with its role as a neuromodulator.

According to the reported results,[26,28–30,33] substitution in position 4(5) of the imidazoline ring lead usually to inactive or at least less active compounds. Most of the obtained analogues of agmatine showed very low or no affinity to imidazoline and α_2-adrenoceptors but in two cases the results are very interesting (**2a and 4**).

Compound **2a** ([4(5)*S*]-4(5)-(2-aminoethyl)imidazoline)[34] is the closest analogue of agmatine, obtained by the constraining of guanidine moiety into heterocyclic ring. Compound **2a** showed almost 100 times higher affinity to I_1 receptors than agmatine. It is remarkable that in comparison to Clonidine, an established and well-known I_1 and α_2-adrenoceptors' ligand, **2a** possesses almost similar affinity to I_1 receptors and much higher selectivity against I_2 and α_2-adrenoceptors. Compound **4** possesses some, rather low affinity to I_2 receptors but this result is over 200 times higher than obtained for agmatine. It is also worth being noticed, that the *N*-acetyl and *N*,*N*-dimethyl 4(5)-(2-aminoethyl)imidazoline derivatives (**7a, 7b, 8, 9 and 10**) in the aminoethyl moiety, lost their properties in comparison to compounds **2a, 2b** and **3a, 3b**.



Scheme 3. Synthesis of 2-substituted 4(5)-(2-aminoethyl)imidazolines. Reagents and conditions: (a) 3 M BrCN/DCM, DCM, 0° C–rt; (b) HBr/AcOH; (c) $H_2NCH(NH)$·AcOH, EtOH; (d) $CH_3C(NH)OEt$·HCl, EtOH; (e) EtONa/EtOH 2 equiv, EtOH; (f) $HC(OEt)_3$, EtOH, Δ.

Table 1
Binding affinities to I_1, I_2 and α_2-adrenoceptors for compounds **2a**, **2b**, **3a**, **3b**, **4**, **7a**, **7b**, **8**–**10**, agmatine and clonidine

Compound (configuration)	I_1 IC_{50} (nM)	I_2 K_i (nM)	α_2 K_i (nM)
2a (*S*)	477	54,950	11,770
2b (*R*)	>100,000	>100,000	18,820
3a (*S*)	>100,000	65,830	>100,000
3b (*R*)	16,500	>100,000	62,070
4 (*S*)	115,100	1844	76,440
7a (*S*)	235,000	>1,000,000	>1,000,000
7b (*R*)	382,600	>1,000,000	>1,000,000
8 (*S*)	213,400	>1,000,000	>1,000,000
9 (*S*)	44,850	>1,000,000	>52,860
10 (*S*)	64,000	>1,000,000	>1,000,000
Clonidine	366.2	364.5	8.7
Agmatine[a]	36,532 ± 3080	416,700 ± 118,800	31,700 ± 11,000

[a] Result drawn from literature,[2] obtained in similar experimental conditions using Wistar rats tissues.

In conclusion, the constraining of the guanidine moiety of agmatine into heterocyclic imidazoline ring leads to a new group of imidazoline receptors ligands with much higher affinities to imidazoline receptors than agmatine itself. For the closest analogue of agmatine, [4(5)*S*]-4(5)-(2-aminoethyl)imidazoline **2a**, the affinity to I_1 is comparable to clonidine and moreover, the selectivity of **2a** against I_2 and α_2-adrenoceptors is much higher. This results may lead to a new family of imidazoline receptors ligand, 4(5)-(2-aminoethyl)imidazoline derivatives.

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Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.bmcl.2008.11.055.

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34. *[4(5)S]-2-amino-4(5)-(2-aminoethyl)imidazoline dihydrobromide (**2a**):* Mp 175–177 °C: ^{1}H NMR (200 MHz DMSO-d_6): 1.65–2.10 (m, 2H, CHCH_2CH$_2$), 2.65–3.00 (m, 2H, CH$_2$CH_2NH), 3.21 (dd, J_1 = 6.6, J_2 = 9.5, 1H, HNCH_2CH), 3.65 (t, J = 9.5, 1H, HNCH_2CH), 3.85–4.20 (br s, 3H, CH$_2$C*H*(NH)CH$_2$, N*H*), 7.78 (s, 1H, NH), 7.85–8.10 (br s, 3H, N$\underline{H}_3^+$), 8.25 (s, 1H, N*H*). ^{13}C NMR (200 MHz, MeOD): 34.1, 37.6, 49.5, 54.5, 161.3. MSFAB MH$^+$ 129 (Calcd for $C_5H_{12}N_4$ M = 128.2).