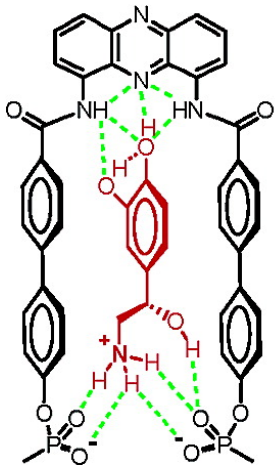
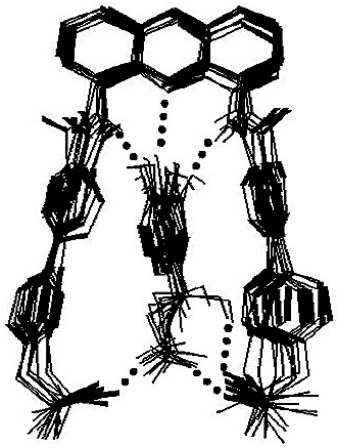
Communication

A Selective Biomimetic Tweezer for Noradrenaline

O. Molt, D. Rbeling, and T. Schrader

J. Am. Chem. Soc., **2003**, 125 (40), 12086-12087• DOI: 10.1021/ja035212l • Publication Date (Web): 12 September 2003





More About This Article

Additional resources and features associated with this article are available within the HTML version:

- Supporting Information
- Links to the 3 articles that cite this article, as of the time of this article download
- Access to high resolution figures
- Links to articles and content related to this article
- Copyright permission to reproduce figures and/or text from this article

View the Full Text HTML

J|A|C|S
COMMUNICATIONS

Published on Web 09/12/2003

A Selective Biomimetic Tweezer for Noradrenaline

O. Molt, D. Rübeling, and T. Schrader*

Philipps-Universität Marburg, Fachbereich Chemie, Hans-Meerwein-Strasse, 35032 Marburg

Received March 18, 2003; E-mail: schradet@mailer.uni-marburg.de

Catecholamines such as (nor-)adrenaline or dopamine are important hormones and neurotransmitters. Through agonistic interaction with adrenergic receptors a broad range of vital body functions are influenced, and these receptors are still hot targets for pharmaceutical research.[1] Many groups have been inspired to design artificial host molecules for catecholamines. However, most of these systems are monotopic and nonspecific.[2]

We intended to shape a highly preorganized receptor molecule, similar to those in Nature, for a more favorable complexation entropy and improved desolvation of the included guest. Tweezer molecules with stiff elements and connections have served this purpose very successfully in recent years.[3]

Figure 1 demonstrates the biomimetic design of this new host: Similar to the natural example, the ammonium alcohol is bound by salt bridges and π−cation interactions, reinforced by a network of ionic hydrogen bonds. The catechol itself is buried in an aromatic cleft capable of π-stacking, and the phenolic hydroxyls are all extra hydrogen-bonded.[4]

In contrast to macrocyclic hosts which require a multistep synthesis with the critical macrocyclization in the end, the new receptor is accessible in analytically pure form by a short and convergent synthesis: biphenylic side walls are linked to an aromatic diaminophenazine headgroup by standard amide coupling, followed by deprotection of the other end and attachment of both phosphonate moieties.[5] Apart from the rotations of the biphenyls around their own axis, this scaffold should exist in a quite immobile conformation. Furthermore, the well-known formation of intramolecular hydrogen bonds between both amide protons and the phenazine ring−nitrogen fixes the scaffold at an aromatic distance of 7.0 Å and orientates the amide protons inward, suitable for catechol recognition.[6] At the opposite part of the biphenyls two phosphonate anions guarantee the strong and essential recognition of the ammonium alcohol moiety.

To validate the chosen building blocks we synthesized three different tweezers and examined their complexes with noradrenaline hydrochloride. Job plots revealed that in all cases 1:1 complexes are formed, and the association constants in methanol determined by ^{1}H NMR titrations confirmed our strategy (Figure 2).[7] While host **2** with much too long tolane side walls does not bind noradrenaline at all, the biphenylic receptor **3** forms a weak complex ($K_a = 120$ M^{-1}). Replacing the flexible *m*-xylylene headgroup with the rigid phenazine moiety in receptor **1** strongly improves the affinity for the desired guest ($K_a = 1800$ M^{-1}).

In complexes of adrenaline derivatives with **1**, all catecholamine protons undergo complexation-induced shifts, demonstrating the close proximity of host and guest.[8]

The effective 1:1 complex formation between **1** and noradrenaline could also be monitored by ESI-MS, producing clean mass spectra with host and aggregate ion peaks, exclusively (Figure 3).[9] In the FT-IR, a strong shift (40 cm^{-1}) of the asymmetric P=O valence to lower wavenumbers demonstrates the amino alcohol recognition.

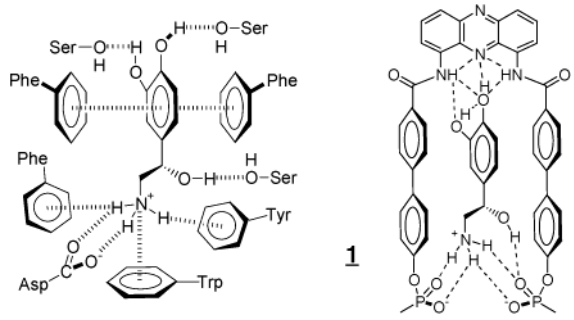


Figure 1. Schematic binding pattern of noradrenaline in the natural β-adrenergic receptor (left) and in the rigid biomimetic host **1** (right).

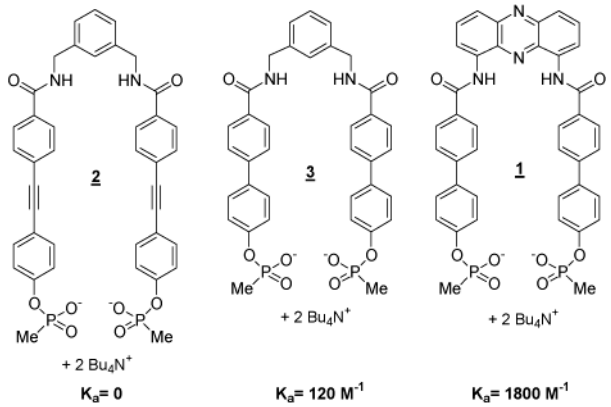


Figure 2. Lewis-structures of tweezers **1**−**3** and association constants of their complexes with noradrenaline hydrochloride in d_4-methanol at 20 °C.

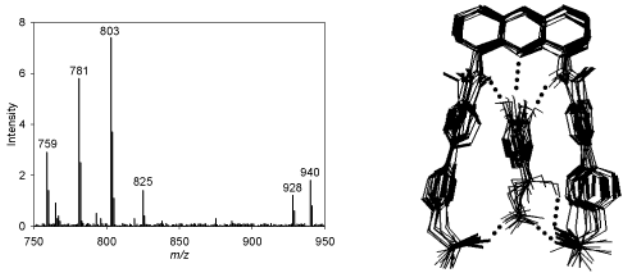


Figure 3. (Left) ESI mass spectrum of a solution of receptor **1** and noradrenaline hydrochloride (NA) in methanol. Host peaks: $m/z = 759$ [**1** + 3H]$^+$, 781 [**1** + 2H + Na]$^+$, 803 [**1** + H + 2Na]$^+$, 825 [**1** + 3Na]$^+$. Complex peaks: $m/z = 928$ [NA + **1** + 2H]$^+$, 940 [NA + **1** + 2Li]$^+$. Right: Complex between receptor **1** and noradrenaline according to Monte Carlo simulations in water with subsequent molecular dynamics (right).

To our disappointment, no shifts were found in the UV/vis spectra of **1** on addition of noradrenaline.

Molecular modeling studies strongly support our design (Figure 3).[10] Furthermore, molecular dynamics calculations confirm the rigidity of the upper part of the host and the flexibility of the phosphonates. Even inside the cavity the guest has some mobility, indicating that it is large enough to include the guest.

10.1021/ja035212l CCC: $25.00

Table 1. Association Constants [M^{-1}] from NMR Titrations of **1** with Neurotransmitters and Related Guests in d_4-Methanol at 25 °C[a]

entry	guest hydrochloride	K_a [M^{-1}]	entry	guest hydrochloride	K_a [M^{-1}]
1	(*R*/*S*)-noradrenaline	1800	7	L-tryptophan methyl ester	240
2	(*R*/*S*)-adrenaline	260	8	L-tyrosine methyl ester	170
3	dopamine	340	9	R-propranolol	130
4	phenethylamine	<1[b]	10	GABA	<1
5	2-aminoethanol	<1	11	glycine	<1
6	catechol	<1			

[a] Errors in K_a are standard deviations from the nonlinear regressions and were estimated at ±10−45%. [b] Lowest detection limits.

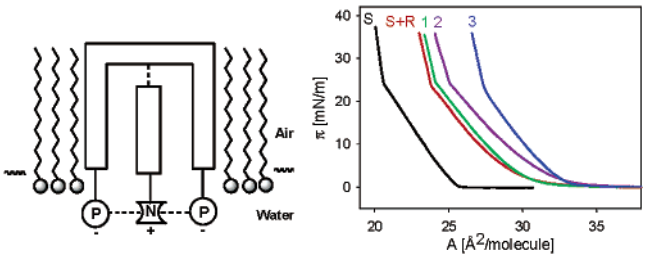


Figure 4. (Left) Schematic representation of the tweezer **1** embedded in a stearic acid monolayer at the air/water interface. The cavity is open to the water sub-phase and received a catecholamine guest molecule. (Right) Pressure−area isotherms of stearic acid (S) and receptor in a monolayer over water (S + R), dopamine (1), adrenaline (2), or noradrenaline (3).

A thorough examination of the tweezer's binding selectivity was carried out by titrating structurally related guests with **1** (Table 1). Representative guests are hydrochlorides of amines, amino alcohols, and amino acids with only marginal structural deviations from noradrenaline.[11]

Systematic alterations of noradrenaline's structure in minute steps (single methyl and OH groups) lead to a noticeable decrease in K_a at every step, demonstrating the specificity of receptor **1** for this guest (entries 1−4). Cutting the guest in two halves completely eliminates any affinity toward **1** (entries 5,6). Only replacement of the catechol by larger electron-rich π faces as in aromatic amino acid esters (entries 7,8) or the adrenergic receptor antagonist propranolol (entry 9) restores a weak attraction by host **1**. Molecular modeling studies reveal a distorted complex geometry for these cases, because their additional substituents prevent the inclusion of amino acids and β-blockers inside the cavity of **1**. None of the ammonium-based neurotransmitters listed in entries 10−11 showed any interaction with **1**. These investigations characterize **1** as an artificial receptor molecule with high noradrenaline specificity.

The highly amphiphilic structure of **1** prompted us to incorporate the receptor molecule in a stearic acid monolayer at the air/water interface. In the Langmuir film balance, substantial shifts are produced in the pressure/area diagram, indicating rapid incorporation of **1** into the monolayer.[12] Subinjection of various analytes found in Table 1 into the aqueous subphase (10^{-4} M) leads to strong effects reflecting the interaction with the embedded receptor molecule (no effects are produced with stearic acid alone). By far the largest shift is obtained from noradrenaline, followed by much smaller shifts from adrenaline and dopamine. All other guests produce negligible shifts, which beautifully supports the high selectivity of **1** for noradrenaline (Figure 4).

We are currently developing highly selective sensor devices in joint projects. To this end, host **1** will be incorporated in lipid/polydiacetylene assemblies for a photometric detection of noradrenaline.[13] Alternatively, a titanium dioxide matrix including **1** as receptor site will be immobilized on electrodes (ITO) or field-effect transistors for electrochemical catecholamine detection.[14]

Acknowledgment. This work was supported by the Deutsche Forschungsgemeinschaft.

Supporting Information Available: Synthetic details, NMR binding experiments, Langmuir film experiments, and molecular modeling data (PDF). This material is available free of charge via the Internet at http://pubs.acs.org.1

References

(1) (a) Ligett, S. B. *Pharmacology* **2000**, *61*, 167−173. (b) Bock, M. G.; Patane, M. A. *Annu. Rep. Med. Chem.* **2000**, *35*, 221−230.
(2) For an overview, see: Schrader, T.; Herm, M.; Molt, O. *Chem. Eur. J.* **2002**, *8*, 1485−1499.
(3) (a) Zimmerman, S. C. *Top. Curr. Chem.* **1993**, *165*, 71−102. (b) Brown, S. P.; Schaller, T.; Seelbach, U. P.; Koziol, F.; Ochsenfeld, C.; Klärner, F.-G.; Spiess, H. W. *Angew. Chem., Int. Ed.* **2001**, *40*, 717−720. (c) Krebs, F. C.; Jorgensen, M. *J. Org. Chem.* **2001**, *66*, 6169−6173.
(4) For complexation through charged hydrogen bonds, see: Echavarren, A.; Galan, A.; Lehn, J.-M.; de Mendoza, J. *J. Am. Chem. Soc.* **1989**, *111*, 4994−4995; Snowden, T. S.; Anslyn, E. V. *Curr. Opin. Chem. Biol.* **1999**, *3*, 740−746; Berger, M.; Schmidtchen, F. P. *J. Am. Chem. Soc.* **1999**, *121*, 9986−9993. For catechol binding with cleft-type hosts, see: Sijbesma, R. P.; Nolte, R. J. *Top. Curr. Chem.* **1995**, *175*, 25−26.
(5) All synthetic details are found in the Supporting Information.
(6) Safarowsky, O.; Nieger, M.; Fröhlich, R.; Vögtle, F. *Angew. Chem., Int. Ed.* **2000**, *39*, 1616−1618; Zimmerman, S. *Bioorg. Med. Chem.* **1996**, *4*, 1107.
(7) Wilcox, C. S. In *Frontiers in Supramolecular Chemistry*; Schneider, H. J., Ed.; Verlag Chemie: Weinheim, 1991; p 123.
(8) The tendency of ***1*** to self-associate in methanol was determined at $K_{sa} \leq 50\ M^{-1}$ by dilution titration.
(9) No molecular ion peaks were found for host dimers or higher aggregates.
(10) MacroModel 7.0, Schrödinger Inc., Force-Field: Amber*, Monte Carlo simulations in water: 3000 steps.
(11) Due to the low methanol solubility of **1**, the NMR titrations had to be carried out at millimolar concentrations, and the amount of added host did often not surpass 2 equiv. At these conditions, however, the complexation-induced shifts remained rather small, and only in the case of noradrenaline was 80% saturation reached. In all other cases, saturation did not exceed 50%, owing to their weak affinity towards 1. To secure the high K_a value found for noradrenaline, we repeated the respective binding experiment and obtained similar association constants from two independent proton signals.
(12) For a recent review, see: Ariga, K.; Kunitake, T. *Acc. Chem. Res.* **1998**, *31*, 371−378.
(13) Kolusheva, S.; Shahal, T.; Jelinek, R. *J. Am. Chem. Soc.* **2000**, *122*, 776−780.
(14) Lahav, M.; Kharitonov, A. B.; Willner, I. *Chem. Eur. J.* **2001**, *7*, 3992−3997.

JA035212L