Molecular Orbital Calculations on the Preferred Conformation of Neostigmine

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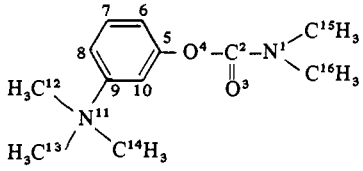
Neostigmine (I), a potent inhibitor of acetylcholinesterase, also displays direct nicotine-like activity.[1] If this direct activity of neostigmine is seriously to be considered a consequence of acting *via* the nicotinic receptor at the neuromuscular junction, it is reasonable that its active centers sterically approximate those of nicotine when each assumes its minimum energy state.



I

The preferred conformation of nicotine as described by Kier[2] predicts at least 2 active centers—a quaternary N and a partially negative site approximately 4.8 Å removed. This hypothesis is supported by a similar study on ACh[3] and on phenylcholine ether.[4,5]

Pauling and Petcher[6] described a preferred conformation for neostigmine bromide by X-ray diffraction analysis. A distance of approximately 6.5 Å is reported between the quaternary N and the C=O—a value significantly different from Kier's 4.8-Å distance. To further clarify this matter we have completed a molecular orbital study on neostigmine using Hoffmann's Extended Hückel Technique.[7]

Methods

All calcns were in the Extended Hückel approximation as described by Hoffmann.[7] The input required was detd from a program supplied by the University of Indiana's Quantum Chemistry Program Exchange. This program calcs precise 3-dimensional atomic coordinates necessary for Extended Hückel Theory (EHT). All bond angles and bond lengths were of standard magnitude as described by Sutton.[8] The EHT parameters which include a choice of K, calcn of resonance integrals, all coulomb integrals, and Slater exponents, are chosen consistent with Kier.[3] Due to the extensively delocalized character of the urethane system, centers N(1), C(2), O(3), O(4), C(15), C(16) are assigned a planar configuration—leaving C(2)-O(4)-C(5)-C(6) and C(8)-C(9)-N(11)-C(12) as the only variable rotomers. The latter torsion angle is assumed to be in a staggered conformation with respect to the Ph ring. C(2)-O(4)-C(5)-C(6) is varied from 0° to 360° in increments of 60°. Two addl conformations were obtd at 150° and at 210°.

The computations were made on the IBM 360 Model 75. All calcns were done in double precision arithmetic. PL/I and Fortran IV were the programming languages used.

Results and Discussion

The torsion angle C(2)-O(4)-C(5)-C(6) in the minimum energy state was found to be $\theta = 120°$ and 240°. The distance values are represented in Table I.

A calcd energy barrier of 2 eV for $\theta = 120°$ to $\theta = 150°$ was found. The least preferred conformation was calcd at $\theta = 0°$ and 180° with an energy barrier of 5.6 eV. It should be noted that EHT tends to overestimate this parameter.

Table I. Interatomic Distances Found in the Preferred Conformation of Neostigmine

	Å
d (1, 2)[a]	1.32
d (1, 3)	2.20
d (1, 4)	2.32
d (1, 11)	6.33
d (2, 11)	5.12
d (3, 11)	4.52
d (4, 11)	4.88
d (3, 4)	2.24

[a] d (i, j) represents the atomic distance between atoms i and j in Å.

Pauling and Petcher,[6] in their X-ray diffraction analysis, predict a minimum energy conformation in neostigmine when the torsion angle C(2)-O(4)-C(5)-C(6) is 150° and −150° (210°). However, d(3, 11) = 6.47 Å as calcd by Pauling and Petcher[6] is significantly different from the d(3, 11) = 4.5 Å predicted in EHT where the torsion angle of 120° is predicted for a minimum energy state.

Kier[2] postulated that the quaternary N and the pyridine N were 2 important active sites in the nicotine molecule. Using EHT on neostigmine d(3, 11) = 4.5 Å agrees reasonably well with the distance necessary for maximum direct nicotine-like activity as put forth by Kier and thus supports the idea that neostigmine indeed presents the C=O and the quaternary N to the nicotine receptor in such a way as to produce a direct stimulation.

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Acylpyruvates As Potential Antifungal Agents

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During the course of another investigation we found reason to believe that ethyl 2,4-dioxovalerate (**1**) might possess antifungal activity. A survey of the literature revealed that although numerous acylpyruvate analogs have been prepared, none has been reported to demonstrate antifungal activity.

A sample of ester **1** was prepared by the base condensation of acetone with diethyl oxalate according to the procedure of Marvel and Dreger.[1] When tested against *Candida albicans* and *Microsporum canis* by the agar diffusion-cylinder cup method,[2] ester **1** was found to inhibit the