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Structural Comparison of the Potent Antimuscarinic Agent Azaprophen Hydrochloride with Aprophen Hydrochloride and Structurally Related Antimuscarinic Agents

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Abstract

A comparison of the crystalline structure of the potent azaprophen with the crystalline structures of aprophen and four other structurally related antimuscarinic agents reveals the potential for an ionic interaction of the cationic nitrogen atom and the carbonyl oxygen atom with the muscarinic receptor and an aromatic interaction with a phenyl group. 6-Methyl-6-azabicyclo[3.2.1]octan-3α-ol 2,2-diphenylpropionate hydrochloride (azaprophen hydrochloride), $C_{23}H_{28}NO_2^+.Cl^-$, $M_r = 385{\cdot}9$, monoclinic, $P2_1/c$, $a = 8{\cdot}490$ (1), $b = 14{\cdot}335$ (2), $c = 16{\cdot}847$ (2) Å, $\beta = 93{\cdot}63$ (1)°, $V = 2046{\cdot}2$ Å^3, $Z = 4$, $D_x = 1{\cdot}253$ g cm^{-3}, Cu $K\alpha$, $\lambda = 1{\cdot}54178$ Å, $\mu = 17{\cdot}86$ cm^{-1}, $F(000) = 824$, room temperature, final $R = 4{\cdot}25\%$ for 2460 reflections with $|F_o| > 3\sigma$. 2-Diethylaminoethyl 2,2-diphenylpropionate hydrochloride (aprophen hydrochloride), $C_{21}H_{28}NO_2^+.Cl^-$, $M_r = 361{\cdot}9$, orthorhombic, $Pbca$, $a = 15{\cdot}118$ (3), $b = 7{\cdot}488$ (2), $c = 36{\cdot}306$ (10) Å, $V = 4110{\cdot}8$ Å^3, $Z = 8$, $D_x = 1{\cdot}316$ g cm^{-3}, Cu $K\alpha$, $\lambda = 1{\cdot}54178$ Å, $\mu = 17{\cdot}45$ cm^{-1}, $F(000) = 1552$, room temperature, final $R = 7{\cdot}96\%$ for 1846 reflections with $|F_o| > 3\sigma$. Both azaprophen and aprophen were crystallized as tertiary amine salts. The overall conformation of both molecules is similar as demonstrated by space-filling models and superimposed stick drawings. Although the interatomic distance between the nitrogen atom and the carbonyl oxygen atom of azaprophen and aprophen is comparable at 5·41 and 5·07 Å, respectively, the nitrogen atoms of azaprophen and aprophen are 1·16 Å apart when the acyloxy portion (—O—C═O) of both molecules is superimposed. A conformational analysis of azaprophen, aprophen and the structurally similar antimuscarinic agents reveals a buried ether oxygen atom and an exposed carbonyl oxygen atom as well as the common placement of a phenyl group on the same side of the

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acyloxy plane as the cationic nitrogen atom. The varied orientations of the acid-derived portion and the diethylaminoethyl group of aprophen, adiphenine and benactyzine, in their respective crystalline structures, and an examination with hand-held CPK models suggests flexibility on both sides of the acyloxy group. A common receptor site for all six molecules must accommodate varying positions of the cationic nitrogen atom.

Introduction

Interest in the pharmacological development of antimuscarinic agents stems from their cholinolytic and antispasmodic activity. The new antimuscarinic agent azaprophen hydrochloride (Fig. 1) (Carroll, Abraham, Parham, Griffith, Ahmad, Richard, Padilla, Witkin & Chiang, 1987) was designed to contain an azabicyclo ring system isomeric to the azabicyclo ring system of atropine, one of the classical antimuscarinic agents, and a diphenylpropionate group identical to the acid-derived portion of aprophen, another potent antimuscarinic agent (Gordon, Padilla, Moore, Doctor & Chiang, 1983). This combination has led to an antimuscarinic agent which is more potent than atropine, aprophen and the other anticholinergic agents shown in Fig. 1 in the inhibition of acetylcholine-induced contraction of guinea-pig ileum and in the inhibition of carbachol-induced release of α-amylase from pancreatic acini cells (Carroll *et al.*, 1987; Witkin, Gordon & Chiang, 1987).

Despite extensive research, the optimal conformational requirements for the binding of antimuscarinic agents to receptor site(s) remain undefined. The three-dimensional conformation of the muscarinic receptor(s) is unknown. Thus, at the present time, information on the receptor site(s) for muscarinic antagonists must be derived from the three-dimensional conformation of known active and inactive antimuscarinic agents. Since (±)-azaprophen hydrochloride is a relatively rigid structure of high antimuscarinic potency, the determination of its three-dimensional structure and comparison of its structure with the potent, but less active, aprophen hydrochloride, and with the structures of (±)-adiphenine hydrochloride (Guy & Hamor, 1973) (±)-benactyzine hydrochloride (Petcher, 1974), (−)-atropine hydrobromide (Kussäther & Haase, 1972) and quinuclidinyl benzilate hydrobromide (Meyerhöffer & Carlström, 1969), should help to elucidate the geometric and electronic binding requirements of antimuscarinic agents to the muscarinic cholinergic receptor.

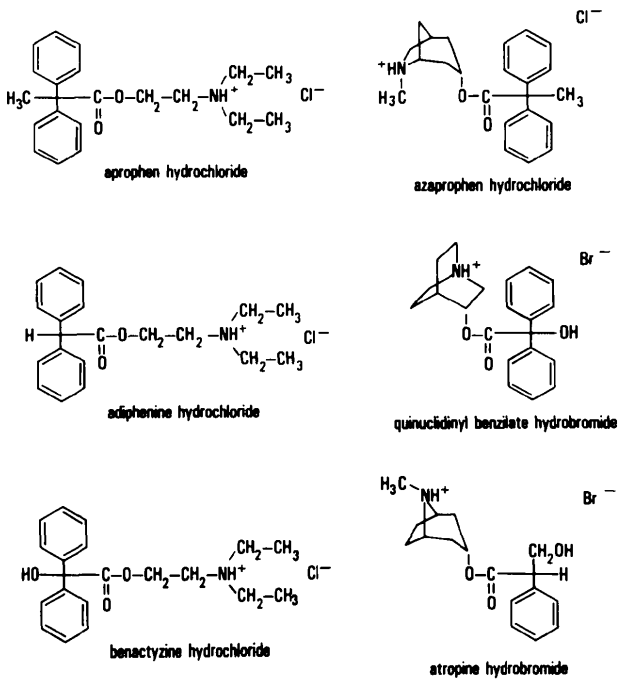

Fig. 1. Chemical structure of the title compounds and structurally related antimuscarinic agents.

Experimental

Azaprophen hydrochloride was synthesized at the Research Triangle Institute (Research Triangle Park, NC) (Carroll *et al.*, 1987) and was crystallized from ethyl acetate. Diffraction data were collected from a striated colorless irregular crystal, 0·2 × 0·7 × 0·8 mm, mounted diagonally to the striations, in the θ–2θ mode to a maximum 2θ value of 114° on a $R3m$/micro Nicolet four-circle diffractometer with a graphite monochromator. Range of indices: h 0→8, k 0→15 and l −18→18. The total number of independent reflections was 3133. The standard reflections 008, 400 and 060 were monitored after every 60 intensity measurements. The standards remained constant within 2·6%. The lattice parameters were based on 25 centered reflections with 2θ values between 35 and 55°. No correction for absorption or extinction was used. The structure was solved routinely by direct phase determination (Karle & Karle, 1966). All of the non-hydrogen atoms were found in the first E map. All of the hydrogen atoms were found in subsequent difference maps. Least-squares refinement was performed using 2460 reflections with $|F_o| > 3\sigma(F)$. Coordinates for all atoms except the hydrogen atoms were refined (on F) by a blocked-cascade program in the *SHELXTL* system (Sheldrick, 1980). Coordinates for the hydrogen atoms were kept fixed in idealized positions. Anisotropic thermal parameters for the C, N, O and Cl atoms and isotropic thermal parameters for hydrogen atoms were refined for a total of 244

Table 1. *Fractional coordinates* ($\times 10^4$) *and thermal parameters* U_{eq} ($Å^2 \times 10^3$) *for azaprophen with e.s.d.'s in parentheses*

$U_{eq} = \frac{1}{3}\sum_i\sum_j U_{ij} a_i^* a_j^* \mathbf{a}_i.\mathbf{a}_j$.

	x	y	z	U_{eq}
Cl	1997 (1)	5904 (1)	4200 (1)	56 (1)
N(1)	5103 (2)	6864 (1)	4137 (1)	53 (1)
C(1)	6172 (3)	6155 (2)	4555 (2)	50 (1)
C(2)	7927 (3)	6322 (2)	4471 (2)	52 (1)
C(3)	8578 (3)	7237 (2)	4816 (1)	44 (1)
O(4)	8518 (2)	7920 (1)	4170 (1)	43 (1)
C(5)	9698 (3)	8554 (2)	4183 (1)	39 (1)
O(6)	10736 (2)	8589 (1)	4699 (1)	53 (1)
C(7)	9546 (2)	9210 (1)	3464 (1)	37 (1)
C(8)	10506 (3)	10093 (2)	3687 (2)	52 (1)
C(9)	10293 (2)	8731 (2)	2756 (1)	37 (1)
C(10)	10424 (3)	9225 (2)	2054 (1)	46 (1)
C(11)	11115 (3)	8826 (2)	1415 (1)	55 (1)
C(12)	11686 (3)	7927 (2)	1465 (2)	58 (1)
C(13)	11564 (3)	7430 (2)	2153 (2)	55 (1)
C(14)	10872 (3)	7827 (2)	2800 (1)	44 (1)
C(15)	7796 (3)	9463 (2)	3288 (1)	41 (1)
C(16)	6936 (3)	9181 (2)	2610 (2)	55 (1)
C(17)	5343 (3)	9407 (2)	2481 (2)	72 (1)
C(18)	4610 (3)	9915 (2)	3034 (2)	78 (1)
C(19)	5431 (3)	10193 (2)	3717 (2)	74 (1)
C(20)	7020 (3)	9975 (2)	3849 (2)	57 (1)
C(21)	5081 (3)	7699 (2)	4675 (2)	56 (1)
C(22)	5935 (3)	7364 (2)	5449 (2)	51 (1)
C(23)	7691 (3)	7608 (2)	5506 (1)	51 (1)
C(24)	5750 (3)	6311 (2)	5396 (2)	54 (1)
C(25)	5363 (3)	7056 (2)	3293 (2)	73 (1)

Table 2. *Fractional coordinates* ($\times 10^4$) *and thermal parameters* U_{eq} ($Å^2 \times 10^3$) *for aprophen with e.s.d.'s in parentheses*

$U_{eq} = \frac{1}{3}\sum_i\sum_j U_{ij} a_i^* a_j^* \mathbf{a}_i.\mathbf{a}_j$.

	x	y	z	U_{eq}
Cl	5943 (1)	11818 (2)	427 (1)	76 (1)
N(1)	5751 (3)	7735 (6)	468 (1)	65 (2)
C(2)	6501 (4)	7700 (7)	736 (1)	50 (2)
C(3)	6765 (3)	5884 (7)	878 (1)	47 (2)
O(4)	6170 (2)	5390 (4)	1172 (1)	50 (1)
C(5)	6475 (4)	4132 (7)	1404 (1)	48 (2)
O(6)	7214 (3)	3539 (5)	1380 (1)	65 (2)
C(7)	5820 (3)	3596 (6)	1702 (1)	47 (2)
C(8)	5945 (4)	1567 (7)	1752 (2)	69 (2)
C(9)	6075 (3)	4591 (7)	2057 (1)	51 (2)
C(10)	5891 (4)	3888 (8)	2400 (2)	72 (2)
C(11)	6104 (5)	4814 (11)	2717 (2)	87 (3)
C(12)	6511 (5)	6436 (11)	2700 (2)	83 (3)
C(13)	6686 (4)	7169 (10)	2365 (2)	77 (3)
C(14)	6480 (4)	6264 (8)	2046 (2)	67 (2)
C(15)	4867 (4)	4022 (7)	1588 (1)	48 (2)
C(16)	4344 (4)	5217 (8)	1773 (2)	64 (2)
C(17)	3464 (4)	5511 (9)	1670 (2)	84 (3)
C(18)	3111 (4)	4597 (9)	1384 (2)	86 (3)
C(19)	3621 (4)	3404 (9)	1194 (2)	83 (3)
C(20)	4494 (4)	3125 (8)	1292 (2)	67 (2)
C(21)	5965 (6)	6768 (9)	112 (2)	101 (4)
C(22)	6771 (6)	7490 (13)	−76 (2)	143 (5)
C(23)	4911 (4)	7081 (9)	601 (2)	121 (5)
C(24)†	4226 (11)	8449 (21)	708 (8)	169 (15)
C(25)†	4543 (9)	8090 (18)	924 (4)	69 (6)

†Disordered CH_3 group. C(25) was weighted 47%.

parameters. Final $R = 4{\cdot}26\%$ and $wR = 4{\cdot}71\%$, $w = 1/[\sigma^2(|F|) + 0{\cdot}0003(F_o)^2]$. Final difference electron density $|\rho|_{max} = 0{\cdot}30$ and $|\rho|_{min} = 0{\cdot}24\,e\,Å^{-3}$. Atomic scattering factors were those incorporated in *SHELXTL* (Sheldrick, 1980). $(\Delta/\sigma)_{max} = 0{\cdot}04$.

Aprophen hydrochloride was synthesized by Brown *et al.* at the Walter Reed Army Institute of Research (Brown, Smejkal, Breuer, Doctor & Chiang, 1988) and was crystallized from ethyl acetate. Diffraction data were collected from a clear colorless needle, $0{\cdot}2 \times 0{\cdot}2 \times 0{\cdot}7$ mm, in the θ–2θ mode to a maximum 2θ value of 114° on a *R3m*/micro Nicolet four-circle diffractometer with a graphite monochromator. Range of indices: h 0→16, k −8→0 and l 0→39. The total number of independent reflections was 2543. The standard reflections 0,0,10, 400 and $0\bar{2}0$ were monitored after every 60 intensity measurements. The standards remained constant within 3·0%. The lattice parameters were based on 25 centered reflections with 2θ values between 23 and 53°. A correction for absorption was applied. The structure was solved routinely by direct phase determination (Karle & Karle, 1966). All of the non-hydrogen atoms were found in the first E map; however, the thermal factor for C(24) was large. Assigning two positions for C(24) at approximately half-weight provided a better fit of the data. Most of the hydrogen atoms were present in subsequent difference maps. Least-squares refinement was performed using 1846 reflections with $|F_o| > 3\sigma(F)$. Coordinates for all atoms except the hydrogen atoms were refined (on F) by a blocked-cascade program in the *SHELXTL* system (Sheldrick, 1980). Coordinates for all the hydrogen atoms were kept fixed in idealized positions. Anisotropic thermal parameters for the C, N, O and Cl atoms and isotropic thermal parameters for hydrogen atoms were refined for a total of 190 parameters. Final $R = 7{\cdot}96\%$ and $wR = 7{\cdot}55\%$, $w = 1/[\sigma^2(|F| + 0{\cdot}0003(F_o)^2]$. Final difference electron density $|\rho|_{max} = 0{\cdot}45$ and $|\rho|_{min} = 0{\cdot}40\,e\,Å^{-3}$. Atomic scattering factors were those incorporated in *SHELXTL* (Sheldrick, 1980). $(\Delta/\sigma)_{max} = 0{\cdot}34$.*

All stereodrawings were made using *MOGLI* (Evans & Sutherland, Salt Lake City, UT). All of the other computer drawings were drawn using the system of programs contained in *SHELXTL*.

Discussion

Coordinates and U_{eq} values for the non-hydrogen atoms and coordinates for the refined hydrogen atoms for azaprophen and aprophen are listed in Tables 1 and 2, respectively. Bond lengths, bond

* Lists of structure factors, anisotropic thermal parameters and H-atom parameters have been deposited with the British Library Document Supply Centre as Supplementary Publication No. SUP 52327 (22 pp.). Copies may be obtained through The Executive Secretary, International Union of Crystallography, 5 Abbey Square, Chester CH1 2HU, England.

Table 3. *Bond lengths* (Å), *bond angles* (°) *and selected torsion angles* (°) *of azaprophen with e.s.d.'s in parentheses*

N(1)—C(1)	1·507 (3)	N(1)—C(21)	1·503 (3)
N(1)—C(25)	1·479 (3)	C(1)—C(2)	1·525 (3)
C(1)—C(24)	1·501 (4)	C(2)—C(3)	1·525 (3)
C(3)—O(4)	1·461 (3)	O(4)—C(5)	1·352 (3)
C(5)—O(6)	1·199 (3)	C(5)—C(7)	1·532 (3)
C(7)—C(8)	1·539 (3)	C(7)—C(9)	1·546 (3)
C(7)—C(15)	1·540 (3)	C(9)—C(10)	1·389 (3)
C(9)—C(14)	1·385 (3)	C(10)—C(11)	1·382 (3)
C(11)—C(12)	1·378 (4)	C(12)—C(13)	1·369 (4)
C(13)—C(14)	1·393 (3)	C(15)—C(16)	1·376 (3)
C(15)—C(20)	1·395 (3)	C(16)—C(17)	1·394 (3)
C(17)—C(18)	1·363 (4)	C(18)—C(19)	1·367 (5)
C(19)—C(20)	1·388 (4)	C(21)—C(22)	1·529 (3)
C(22)—C(24)	1·520 (4)	C(3)—C(23)	1·521 (3)
C(22)—C(23)	1·528 (3)		

C(1)—N(1)—C(21)	106·6 (2)	C(1)—N(1)—C(25)	116·9 (2)
C(21)—N(1)—C(25)	116·1 (2)	N(1)—C(1)—C(2)	114·5 (2)
N(1)—C(1)—C(24)	99·7 (2)	C(2)—C(1)—C(24)	111·0 (2)
C(1)—C(2)—C(3)	115·6 (2)	C(2)—C(3)—C(23)	114·0 (2)
C(2)—C(3)—O(4)	107·4 (2)	C(3)—O(4)—C(5)	116·6 (2)
O(4)—C(5)—O(6)	123·4 (2)	O(4)—C(5)—C(7)	112·0 (2)
O(6)—C(5)—C(7)	124·6 (2)	C(5)—C(7)—C(8)	107·1 (2)
C(5)—C(7)—C(9)	108·6 (2)	C(8)—C(7)—C(9)	108·5 (2)
C(5)—C(7)—C(15)	109·2 (2)	C(8)—C(7)—C(15)	110·1 (2)
C(9)—C(7)—C(15)	113·2 (2)	C(7)—C(9)—C(10)	119·3 (2)
C(7)—C(9)—C(14)	122·4 (2)	C(10)—C(9)—C(14)	118·3 (2)
C(9)—C(10)—C(11)	120·9 (2)	C(10)—C(11)—C(12)	120·2 (2)
C(11)—C(12)—C(13)	119·6 (2)	C(12)—C(13)—C(14)	120·5 (2)
C(9)—C(14)—C(13)	120·4 (2)	C(7)—C(15)—C(16)	123·1 (2)
C(7)—C(15)—C(20)	119·0 (2)	C(16)—C(15)—C(20)	117·9 (2)
C(15)—C(16)—C(17)	121·3 (2)	C(16)—C(17)—C(18)	119·9 (3)
C(17)—C(18)—C(19)	120·0 (3)	C(18)—C(19)—C(20)	120·5 (3)
C(15)—C(20)—C (19)	120·4 (2)	N(1)—C(21)—C(22)	104·0 (2)
O(4)—C(3)—C(23)	109·8 (2)	C(21)—C(22)—C(23)	112·9 (3)
C(21)—C(22)—C(24)	102·7 (2)	C(23)—C(22)—C(24)	109·1 (2)
C(3)—C(23)—C(22)	113·7 (2)	C(1)—C(24)—C(22)	100·0 (2)

C(21)—N(1)—C(1)—C(2)	−80·9 (2)	C(−21)—N(1)—C(1)—C(24)	37·5 (2)
C(25)—N(1)—C(1)—C(2)	50·8 (3)	C(25)—N(1)—C(1)—C(24)	169·3 (2)
C(1)—N(1)—C(21)—C(22)	−9·2 (2)	N(1)—C(1)—C(2)—C(3)	61·7 (3)
C(24)—C(1)—C(2)—C(3)	−50·1 (3)	N(1)—C(1)—C(24)—C(22)	−50·8 (2)
C(2)—C(1)—C(24)—C(22)	70·3 (2)	C(2)—C(3)—O(4)—C(5)	−145·3 (2)
C(1)—C(2)—C(3)—O(4)	−93·8 (2)	C(1)—C(2)—C(3)—C(23)	28·1 (3)
C(23)—C(3)—O(4)—C(5)	90·2 (2)	C(2)—C(3)—C(23)—C(22)	−31·7 (3)
C(3)—O(4)—C(5)—O(6)	−1·8 (3)	C(3)—O(4)—C(5)—C(7)	178·0 (2)
O(4)—C(5)—C(7)—C(8)	159·0 (2)	O(4)—C(5)—C(7)—C(9)	−84·0 (2)
O(4)—C(5)—C(7)—C(15)	39·8 (2)	O(6)—C(5)—C(7)—C(8)	−21·3 (3)
O(6)—C(5)—C(7)—C(9)	95·7 (2)	O(6)—C(5)—C(7)—C(15)	−140·4 (2)
C(5)—C(7)—C(9)—C(10)	−174·3 (2)	C(5)—C(7)—C(9)—C(14)	4·3 (3)
C(8)—C(7)—C(9)—C(10)	−58·2 (2)	C(8)—C(7)—C(9)—C(14)	120·4 (2)
C(15)—C(7)—C(9)—C(10)	64·3 (2)	C(15)—C(7)—C(9)—C(14)	−117·2 (2)
C(5)—C(7)—C(15)—C(16)	−113·1 (2)	C(5)—C(7)—C(15)—C(20)	64·8 (3)
C(8)—C(7)—C(15)—C(16)	129·6 (2)	C(8)—C(7)—C(15)—C(20)	−52·6 (3)
C(9)—C(7)—C(15)—C(16)	8·0 (3)	C(9)—C(7)—C(15)—C(20)	−174·1 (2)
N(1)—C(21)—C(22)—C(23)	94·9 (2)	N(1)—C(21)—C(22)—C(24)	−22·5 (2)
C(21)—C(22)—C(23)—C(3)	−56·4 (3)	C(24)—C(22)—C(23)—C(3)	57·2 (3)
C(21)—C(22)—C(24)—C(1)	45·9 (2)	C(23)—C(22)—C(24)—C(1)	−74·1 (2)
O(4)—C(3)—C(23)—C(22)	88·9 (2)	C(25)—N(1)—C(21)—C(22)	−141·4 (2)

Table 4. *Bond lengths* (Å), *bond angles* (°) *and selected torsion angles* (°) *of aprophen with e.s.d.'s in parentheses*

N(1)—C(2)	1·494 (7)	N(1)—C(21)	1·518 (9)
N(1)—C(23)	1·445 (8)	C(2)—C(3)	1·508 (7)
C(3)—O(4)	1·444 (6)	O(4)—C(5)	1·346 (6)
C(5)—O(6)	1·205 (7)	C(5)—C(7)	1·519 (8)
C(7)—C(8)	1·542 (7)	C(7)—C(9)	1·538 (7)
C(7)—C(15)	1·534 (7)	C(9)—C(10)	1·379 (8)
C(9)—C(14)	1·395 (8)	C(10)—C(11)	1·383 (9)
C(11)—C(12)	1·363 (11)	C(12)—C(13)	1·362 (11)
C(13)—C(14)	1·377 (9)	C(15)—C(16)	1·370 (8)
C(15)—C(20)	1·387 (8)	C(16)—C(17)	1·399 (8)
C(17)—C(18)	1·354 (9)	C(18)—C(19)	1·366 (9)
C(19)—C(20)	1·383 (9)	C(21)—C(22)	1·497 (12)
C(23)—C(24)	1·507 (19)	C(23)—C(25)	1·502 (17)

C(2)—N(1)—C(21)	112·6 (5)	C(2)—N(1)—C(23)	116·4 (5)
C(21)—N(1)—C(23)	108·1 (5)	N(1)—C(2)—C(3)	116·1 (4)
C(2)—C(3)—O(4)	108·6 (4)	C(3)—O(4)—C(5)	115·4 (4)
O(4)—C(5)—O(6)	121·9 (5)	O(4)—C(5)—C(7)	114·1 (5)
O(6)—C(5)—C(7)	124·0 (5)	C(5)—C(7)—C(8)	105·3 (4)
C(5)—C(7)—C(9)	107·8 (4)	C(8)—C(7)—C(9)	110·4 (4)
C(5)—C(7)—C(15)	111·4 (4)	C(8)—C(7)—C(15)	110·5 (4)
C(9)—C(7)—C(15)	111·2 (4)	C(7)—C(9)—C(10)	121·3 (5)
C(7)—C(9)—C(14)	121·4 (5)	C(10)—C(9)—C(14)	117·3 (5)
C(9)—C(10)—C(11)	120·9 (6)	C(10)—C(11)—C(12)	121·0 (6)
C(11)—C(12)—C(13)	119·1 (7)	C(12)—C(13)—C(14)	120·7 (7)
C(9)—C(14)—C(13)	121·1 (5)	C(7)—C(15)—C(16)	123·0 (5)
C(7)—C(15)—C(20)	119·4 (5)	C(16)—C(15)—C(20)	117·5 (5)
C(15)—C(16)—C(17)	121·3 (6)	C(16)—C(17)—C(18)	120·0 (6)
C(17)—C(18)—C(19)	119·7 (6)	C(18)—C(19)—C(20)	120·5 (6)
C(15)—C(20)—C(19)	121·0 (5)	N(1)—C(21)—C(22)	112·9 (6)
N(1)—C(23)—C(24)	117·3 (8)	N(1)—C(23)—C(25)	114·5 (7)

C(21)—N(1)—C(2)—C(3)	−64·0 (6)	C(23)—N(1)—C(2)—C(3)	61·6 (6)
C(2)—N(1)—C(21)—C(22)	−56·8 (7)	C(23)—N(1)—C(21)—C(22)	173·2 (6)
C(2)—N(1)—C(23)—C(24)	101·7 (13)	C(2)—N(1)—C(23)—C(25)	60·4 (9)
C(21)—N(1)—C(23)—C(24)	−130·4 (13)	C(21)—N(1)—C(23)—C(25)	−171·8 (8)
N(1)—C(2)—C(3)—O(4)	−81·3 (5)	C(2)—C(3)—O(4)—C(5)	−159·0 (4)
C(3)—O(4)—C(5)—O(6)	4·1 (7)	C(3)—O(4)—C(5)—C(7)	−177·9 (4)
O(4)—C(5)—C(7)—C(8)	142·3 (5)	O(4)—C(5)—C(7)—C(9)	−99·8 (5)
O(4)—C(5)—C(7)—C(15)	22·5 (6)	O(6)—C(5)—C(7)—C(8)	−39·7 (7)
O(6)—C(5)—C(7)—C(9)	78·1 (6)	O(6)—C(5)—C(7)—C(15)	−159·6 (5)
C(5)—C(7)—C(9)—C(10)	−151·6 (5)	C(5)—C(7)—C(9)—C(14)	30·2 (7)
C(8)—C(7)—C(9)—C(10)	−37·1 (7)	C(8)—C(7)—C(9)—C(14)	144·8 (5)
C(15)—C(7)—C(9)—C(10)	86·0 (6)	C(15)—C(7)—C(9)—C(14)	−92·1 (6)
C(5)—C(7)—C(15)—C(16)	−117·0 (6)	C(5)—C(7)—C(15)—C(20)	65·4 (6)
C(8)—C(7)—C(15)—C(16)	126·3 (6)	C(8)—C(7)—C(15)—C(20)	−51·4 (7)
C(9)—C(7)—C(15)—C(16)	3·3 (7)	C(9)—C(7)—C(15)—C(20)	−174·4 (5)

angles and torsion angles for azaprophen and aprophen are listed in Tables 3 and 4, respectively. The bond length of all hydrogen atoms was kept fixed at 0·96 Å throughout the refinement procedure.

Both molecules were crystallized in ionic form containing a positively charged nitrogen atom as part of the tertiary amine salt (Figs. 2*a* and 3*a*). The six-membered ring portion of the azabicyclo ring system of azaprophen is in a chair conformation and the *N*-methyl group is in the *endo* conformation pointing towards rather than away from the ether oxygen atom. The conformation of the azabicyclo ring portion of azaprophen is similar to the lowest energy conformation of 6-methyl-6-azabicyclo-[3.2.1]octan-3α-ol predicted by molecular-modeling studies using the *MM*2 molecular-mechanics program of Allinger (Carroll *et al.*, 1987). Although the N(1) and the ether O(4) atoms of aprophen and azaprophen are separated by two and three carbon atoms, respectively, the interatomic N(1)···O(4) distances of 3·17 Å in aprophen and 3·27 Å in azaprophen are close. The interatomic distance between N(1) and the carbonyl O(6) atoms in both molecules is similar, being 5·07 and 5·41 Å in aprophen and azaprophen, respectviely.

As illustrated in Fig. 4(*a*), the diphenylpropionate portions of both molecules share nearly the same conformation. Even though the N(1)—O(4) distances in both molecules are similar, superimposing the acyloxy portions of the molecules, atoms O(4), C(5)

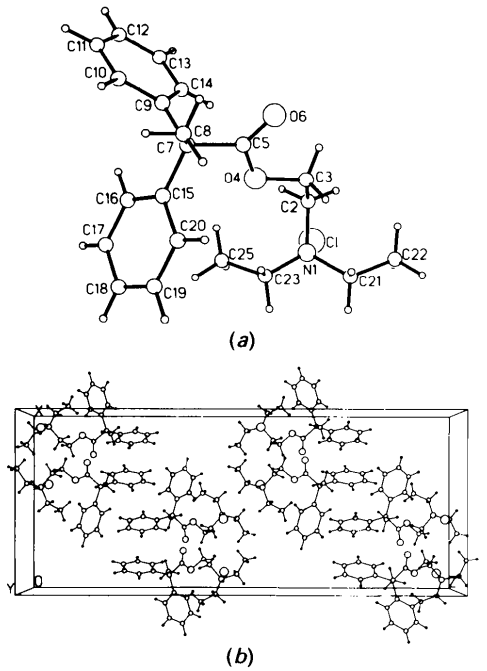

Fig. 2. (*a*) Conformation and numbering scheme of aprophen. (*b*) Packing diagram of aprophen hydrochloride viewed down the *b* axis. Atom C(24) of the disordered methyl group in aprophen is not illustrated. The size of the circles was arbitrarily chosen to correspond approximately to the atomic weight of the atom.

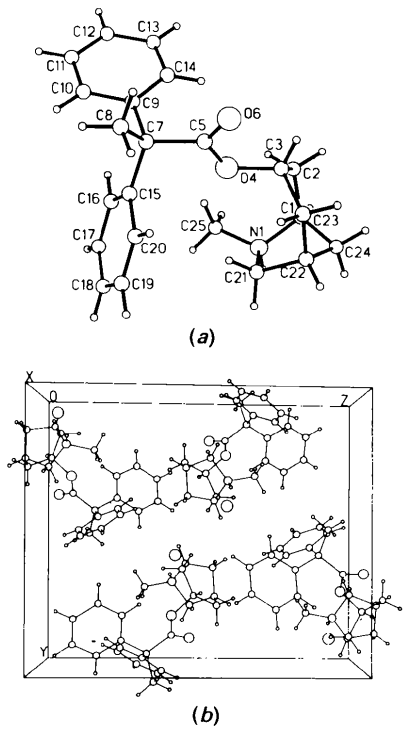

Fig. 3. (*a*) Conformation and numbering scheme of azaprophen. (*b*) Packing diagram of azaprophen hydrochloride viewed down the *a* axis. The size of the circles was arbitrarily chosen to correspond approximately to the atomic weight of the atom.

and O(6), does not superimpose the N(1) atoms of the two molecules. The angle between the C(5)=O(6) and the $N(1)^+$—H(1) bond vectors is 117° in aprophen and 138° in azaprophen. The N···N distance is 1·16 Å, and the Cl atoms are 3·7 Å apart in the superposition. Fig. 4(*b*) illustrates the superpositioning of the N(1) and O(6) atoms of aprophen and azaprophen such that the carbonyl oxygen atoms and the nitrogen atoms are each 0·17 Å apart. In this model the chlorine atoms are 3·0 Å apart. Hydrogen bonding from N(1) to a receptor could occur with both molecules from underneath. However, without a major change in the conformations of aprophen and azaprophen upon approach to the receptor, the anionic region of the muscarinic receptor's binding site would either need to be broad or in multiple locations. In addition, interaction of the carbonyl group in both azaprophen and aprophen with the receptor could occur from the top right side.

The terminal methyl group on one of the *N*-methyl chains of aprophen is disordered such that the methyl group is primarily located in two different positions approximately 50% of the time [see atoms C(24) and C(25) in Table 2]. However, the larger thermal factor associated with atom C(24) in comparison to the thermal factors associated with atom

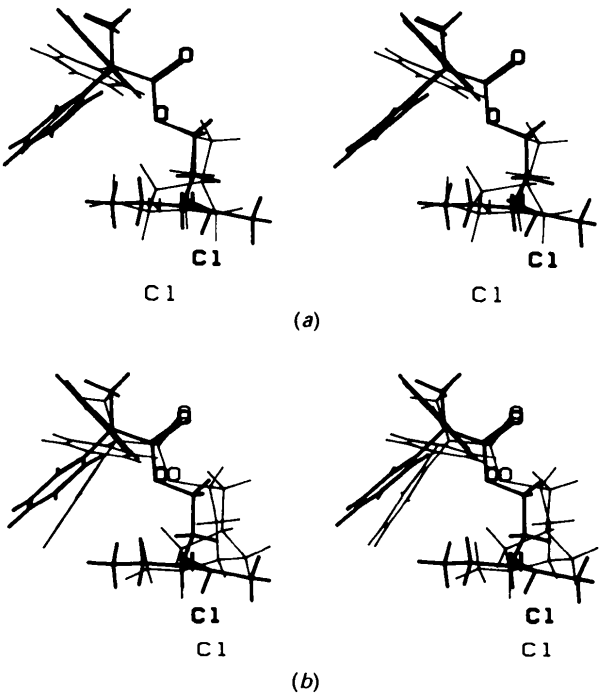

Fig. 4. Stereodiagrams of the superposition of aprophen (heavy lines) and azaprophen (light lines). (*a*) The acyloxy groups [atoms O(4), C(5), and O(6)] of each molecule are superimposed. (*b*) The N(1) and the carbonyl O(6) atoms of each molecule are superimposed. Atom C(24) of the disordered methyl group in aprophen is not illustrated.

C(25) suggests that the positioning of the methyl group is not fully accounted for and may also occur in some positions of lesser occupancy. The disordered methyl group probably contributed to the higher R value determined for aprophen. In the crystal, aprophen molecules are oriented such that the diethylamino groups from separate molecules border each other and the phenyl groups from separate molecules are stacked in a staggered manner (Fig. 2*b*). The closest contacts between atoms in the phenyl rings, C(10)···C(13′) = 3·89 Å, occur between phenyl rings in parallel stacks rather than between rings in the same stack. The packing of azaprophen does not display stacking of phenyl groups (Fig. 3*b*). No solvent molecules were found in either crystal. In both structures the N(1) atom is hydrogen bonded to the chlorine atom. The N(1)···Cl distance in azaprophen is 2·983 (3) Å and in aprophen is 3·075 (6) Å.

The chemical structure of aprophen is very similar to the antimuscarinic agents adiphenine and benactyzine differing only by the replacement of the methyl group of the diphenylpropionate group by a hydrogen atom or a hydroxyl group, respectively (Fig. 1). Carroll *et al.* (1987) found aprophen to be 2 to 3 times more potent than benactyzine and approximately 30 times more potent than adiphenine. The stereodiagrams in Fig. 5 have been drawn such that the acyloxy group is in an identical orientation for each molecule. These stereodiagrams and manipulation of hand-held CPK space-filling models suggest the presence of rotatable bonds in all three compounds. The acid-derived portion of the molecules containing the diphenyl groups is rotated differently in aprophen, benactyzine and adiphenine, even though the orientation of the diphenylpropionate portion of aprophen is very similar to the identical portion of azaprophen. Although the crystalline structures of aprophen, adiphenine and benactyzine are expected to represent low-energy conformations, CPK models of these molecules demonstrate that the C—O ether bond is rotatable. The diethylaminoethyl group of aprophen, benactyzine and adiphenine also occurs in different orientations in each crystalline structure and can be rotated about the C—C(carbonyl) bond using CPK models by concerted bond rotation of the diethylaminoethyl group. The varied orientations of the diethylaminoethyl group result in different nitrogen atom to ether oxygen atom distances of 3·17, 3·21 and 2·80 Å, and nitrogen atom to carbonyl oxygen atom distances of 5·07, 5·03 and 4·85 Å in aprophen, adiphenine and benactyzine, respectively. In all three molecules one of the phenyl groups is on the same side of the plane formed by the acyloxy group as the nitrogen atom. This generally common feature of antimuscarinic compounds was previously noted by Guy & Hamor (1974).

Fig. 6 illustrates the stereodiagrams of azaprophen, atropine and quinuclidinyl benzilate, all of which contain isomeric azabicyclo ring systems in which the nitrogen atom is placed at different positions. As mentioned in the *Introduction*, azaprophen is more potent than all of the other antimuscarinic agents shown in Fig. 1, demonstrating > 10-fold more potency than aprophen and atropine in inhibition of carbachol-induced release of α-amylase from pancreatic acini cells (Carroll *et al.*, 1987; Witkin *et al.*, 1987). In Fig. 6, the three compounds, azaprophen, atropine and quinuclidinyl benzilate, are drawn such that the acyloxy group is oriented in exactly the same position for all three molecules and exactly the same as for the acyclic molecules in Fig. 5. As for the acyclic compounds, all three molecules have a phenyl group which is oriented on the same side of the plane formed by the acyloxy group as the nitrogen atom. Each molecule is *trans* about C(3)—O(4)—C(5)—C(7) with dihedral angles of 178, 176 and 189° for azaprophen, quinuclidinyl benzilate and atropine, respectively. Although these dihedral angles cause the bicyclo ring systems to be slightly shifted from each other with respect to the acyloxy group, the backbone atoms of the bicyclo ring system for azaprophen and atropine assume nearly identical conformations and are superimposable. However, owing to the varied locations of the cationic nitrogen atom, the position of the cationic

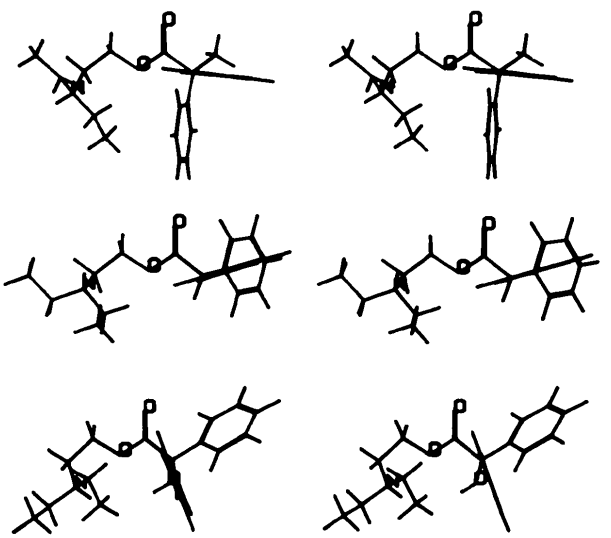

Fig. 5. Stereodiagrams of aprophen (top), adiphenine (middle) and benactyzine (bottom). The orientation of the acyloxy group (the —O—C═O group) is identical for each compound in this figure and in Fig. 6. The hydrogen atoms attached to carbon atoms in adiphenine and benactyzine were placed in idealized positions by the *MOGLI* software. Atom C(24) of the disordered methyl group in aprophen is not illustrated.

nitrogen atom in relation to the acyloxy group differs widely among the three molecules. The nitrogen atom to ether oxygen atom distances are 3·27, 3·55 and 3·75 Å and nitrogen atom to carbonyl oxygen atom distances are 5·41, 4·42 and 5·31 Å in azaprophen, quinuclidinyl benzilate and atropine, respectively. These distances are comparable to the distances found in the acyclic aprophen, adiphenine and benactyzine. Simple rotation, if allowable, of the azabicyclo groups will not superimpose the nitrogen atoms when the acyloxy groups are superimposed. The closest possible approach of the nitrogen atoms of azaprophen and quinuclidinyl benzilate is 2·2 Å, and of azaprophen and atropine is 1·6 Å. Since the cationic nitrogen atoms for the six antimuscarinic agents discussed in this paper cannot be readily superimposed, for these compounds to interact with a common receptor site, the receptor site must allow for variability in positioning of the cationic center.

The stereodrawings in Figs. 5 and 6 illustrate that all of the antimuscarinic agents are configured such that the ether oxygen atom is shielded and the carbonyl oxygen atom is always exposed for potential interaction with a receptor. The ether oxygen atom of azaprophen, the most potent of the six compounds, is the most surrounded by the remainder of the molecule and, therefore, the most shielded from the approach of a receptor. An interaction of the ether oxygen atom of azaprophen with a receptor could not take place without a major conformational change. The biological data of Flavin *et al.* (Flavin, Lu, Thompson & Bhargava, 1987) support a binding-site conformation of this class of antimuscarinic agents that contains an exposed carbonyl oxygen atom. They synthesized conformationally restricted analogues of benactyzine and found that analogues that mimicked a conformation of benactyzine in which the hydroxyl group is hydrogen bonded to the ether oxygen exhibited potent competitive antagonism towards acetylcholine at the muscarinic receptor. The analogues which mimicked a conformation of benactyzine in which the hydroxyl group is hydrogen-bonded to the carbonyl oxygen atom failed to demonstrate competitive inhibition and failed to displace [^{3}H]quinuclidinyl benzilate from the binding site.

In summary, the high potency of azaprophen suggests that azaprophen is capable of assuming a conformation close to optimal for maximum antagonist response towards the muscarinic receptor. The crystalline conformation of azaprophen suggests that the cationic nitrogen atom and the carbonyl oxygen are the potential sites of ionic interaction with the muscarinic receptor and the phenyl ring opposite the

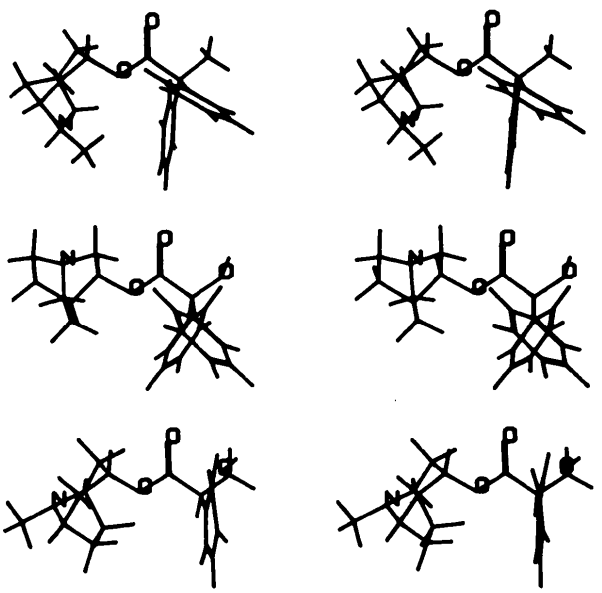

Fig. 6. Stereodiagrams of azaprophen (top), quinuclidinyl benzilate (middle) and atropine (bottom). The orientation of the acyloxy group (the —O—C=O group) is identical for each compound in this figure and in Fig. 5. The hydrogen atoms for quinuclidinyl benzilate and atropine were placed in idealized positions by the *MOGLI* software.

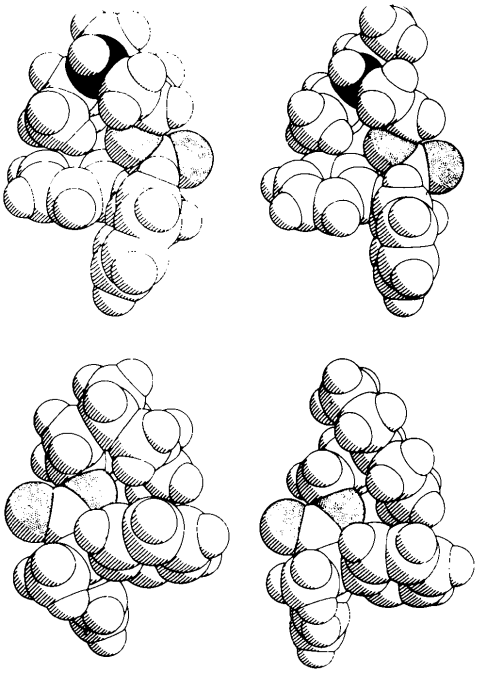

Fig. 7. Space-filled drawing depicting front and back views of azaprophen (left side) and aprophen (right side). The nitrogen atoms have been colored black, and the oxygen atoms have been dotted. The radii of the spheres are 75% of the van der Waals radii.

cationic head may be important for an interaction with an aromatic binding site. The crystalline conformation of aprophen and the other four antimuscarinic agents share these features. The space-filling drawings of azaprophen and aprophen in Fig. 7 demonstrate the high similarity of the overall geometric shape of these two compounds. Even the exposed surface area of approximately 488 $Å^2$ for azaprophen and 482 $Å^2$ for aprophen as estimated by *MOGLI* (Evans & Sutherland, Salt Lake City, UT) is nearly identical. However, the direction and spatial location of the N^+—H bond in comparison to the C=O bond differs in the crystal structures of azaprophen and aprophen as well as between azaprophen and atropine and quinuclidinyl benzilate. Since even rotation of the C—O(ether) bond will not superimpose the nitrogen atoms of the antimuscarinic agents containing an azabicyclo ring system when the acyloxy ring system is superimposed, the muscarinic receptor must allow for the distribution of geometries found in the cationic sites of antimuscarinic agents.

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Molecular-Dynamics Simulation of Crystalline 18-Crown-6:* Thermal Shortening of Covalent Bonds

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Abstract

Molecular-dynamics simulations of crystalline 18-crown-6 have been performed in a study of the apparent thermal shortening of covalent bonds observed in crystal structures. At 100 K, a shortening of 0·006 ± 0·001 Å for C—C and C—O bonds was obtained. This result was found to be independent of details of the force field and the simulation. There was agreement between computational and experimental values for the thermal parameters, as well as for the molecular geometry (bond and dihedral angles) of 18-crown-6. Some differences are attributed to the inability of the force field to reproduce hydrogen-bonding geometries. Simulation at 295 K resulted in an estimated shortening of 0·019 ± 0·005 Å. Thus at room temperature for C—C bonds (apparent) thermal shortening and (real) chemical shortening, resulting from the electronegative oxygen substituents, are of the same order of magnitude. In the simulation at 295 K occasional dihedral transitions were observed, which may reflect the proximity of the melting point (312 K).

Introduction

Short C—C bonds in macrocyclic polyethers have been observed in numerous instances with single-crystal X-ray diffraction methods (Dalley, 1978; Goldberg, 1980). The normal aliphatic C—C bond length is 1·54 Å (Sutton, 1965), and an average value of 1·53 Å is reported for Csp^3—Csp^3 bond lengths derived from diffraction experiments (Allen, Kennard, Watson, Brammer, Orpen & Taylor, 1987). The shortening of the C—C bonds to values in the range 1·46–1·52 Å has been called the 'macrocyclic C—C shortening effect' (Shoham, Lipscomb & Olsher, 1983). The same effect is found, however, in linear polyethers (Weber, Hirayama, Saenger &

* IUPAC name: 1,4,7,10,13,16-hexaoxacyclooctadecane.

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