Derivatives of the Muscarinic Agent N-Methyl-N-(1-methyl-4-pyrrolidino-2-butynyl)acetamide

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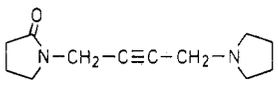
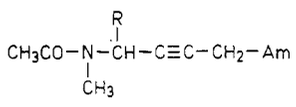
A series of tertiary and quaternary analogues (acyclic imides, sulfonimides, N-acetyl sulfonamides, and trifluoroacetamides) of the selective partial muscarinic agonist N-methyl-N-(1-methyl-4-pyrrolidino-2-butynyl)acetamide (BM 5, **35**) was synthesized. The compounds were found to be muscarinic agonists, partial agonists, or antagonists in the isolated guinea pig ileum. Replacement of the acetyl group or the N-methyl group of **35** and its analogues by a methanesulfonyl group abolished efficacy and decreased affinity at ileal muscarinic receptors. Trifluoroacetamide analogues of **35** also had lower affinity and efficacy than **35**. Substitution of an acetyl group for the N-methyl group in compounds related to **35** decreased efficacy, but had no appreciable effect on affinity. Most of the tertiary amines showed central antimuscarinic activity as they antagonized oxotremorine-induced tremors in mice.

The oxotremorine analogue N-methyl-N-(1-methyl-4-pyrrolidino-2-butynyl)acetamide (BM 5, **35**)[1] has been reported to be a presynaptic antagonist and postsynaptic agonist at muscarinic receptors in vitro[2] and in vivo.[3] Racemic **35**,[1] as well as its pure enantiomers,[4] was found to block oxotremorine-induced tremors. The latter observation suggests that **35** acts mainly as an antagonist in brain regions that are responsible for the tremor response. In other regions of the brain, e.g., those involved in the production of analgesia and hypothermia, **35** acts as a muscarinic agonist.[5] Both enantiomers of **35** have been classified as partial muscarinic agonists in the guinea pig ileum, the *R* enantiomer being the more potent.[4] The partial agonist properties of **35** have been invoked to explain its selective actions in vivo[5] and in vitro.[6]



OXOTREMORINE



34: R = H, Am = NC_4H_8
35: R = CH_3, Am = NC_4H_8
36: R = CH_3, Am = $N(CH_3)_2$
37: R = CH_3, Am = $N^+(CH_3)_3$

Compounds with pharmacological profiles similar to that of **35** have potential for the therapy of diseases associated with deficiences in cholinergic transmission, e.g., Huntington's chorea and Alzheimer-type dementia.[2,7]

Previous structure–activity relationship studies of flexible oxotremorine analogues have demonstrated that the muscarinic activity is modified by changes of the nature or size of the amide substituents.[8] To investigate further the effects of structural modifications in the amide moiety in compounds related to **35**, we have now synthesized compounds **16–33** (Table II).

The new compounds were investigated for tremorogenic and tremorolytic activity in mice and for muscarinic and antimuscarinic activity on the isolated guinea pig ileum (Table III). Some compounds were partial agonists in the ileum and others were relatively potent tremorolytic agents, but none showed both of these effects (cf. **35**). However, **23**, **26**, and **30** were fairly potent antagonists to carbachol in the guinea pig ileum.

Chemistry

Synthesis. Standard methods starting from propargylamine or 1-methyl-2-propynylamine were used to obtain target compounds **16–33** (see the Experimental Section). The final step in the preparation of the tertiary amines involved a Mannich condensation (method H) between an acetylenic intermediate, the appropriate secondary amine, and paraformaldehyde in the presence of a catalytic amount of cuprous chloride. The quaternary ammonium derivatives were prepared by methylation of the corresponding tertiary Mannich bases (method I).

Various reaction conditions were investigated for the preparation of the N,N-diacetylated intermediates N-(2-propynyl)diacetamide (**10**) and N-(1-methyl-2-propynyl)-diacetamide (**11**). Attempts to prepare **10** by use of the method described by Dahlbom et al.[9] for the preparation of N-propargylsuccinimide (diacetamide, propargyl bromide, NaOEt in EtOH) were unsuccessful, probably due to decomposition of the diacetamide. However, **10** could be prepared by alkylation of diacetamide with propargyl bromide in the presence of potassium carbonate in acetonitrile (method G). Compound **11** was prepared from **9** by using acetyl chloride and triethylamine in a large excess (method F). It is noteworthy that attempts to prepare **11** from 1-methyl-2-propynyl *p*-toluenesulfonate by method G were unsuccessful.

In general, the acyclic imides appear to be considerably less stable than their previously reported[9,10] cyclic analogues; partial decomposition was observed when crude **10** was passed through an alumina column eluted with

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(1) Resul, B.; Dahlbom, R.; Ringdahl, B.; Jenden, D. J. *Eur. J. Med. Chem.-Chim. Ther.* **1982**, *17*, 317.
(2) Norström, Ö.; Alberts, P.; Westlind, A.; Undén, A.; Bartfai, T. *Mol. Pharmacol.* **1983**, *24*, 1.
(3) Casamenti, F.; Cosi, C.; Pepeu, G. *Eur. J. Pharmacol.* **1986**, *122*, 288.
(4) Dahlbom, R.; Jenden, D. J.; Resul, B.; Ringdahl, B. *Br. J. Pharmacol.* **1982**, *76*, 299.
(5) Ringdahl, B.; Roch, M.; Jenden, D. J. *J. Pharmacol. Exp. Ther.* **1987**, *242*, 464.
(6) Ringdahl, B. *Mol. Pharmacol.* **1987**, *31*, 351.
(7) Hershenson, F.; Moos, W. H. *J. Med. Chem.* **1986**, *29*, 1125.
(8) For reviews, see: Dahlbom, R. In *Cholinergic Mechanisms: Phylogenetic Aspects, Central and Peripheral Synapses, and Clinical Significance*; Pepeu, G., Ladinsky, H., Eds.; Plenum: New York, 1981; p 621. Ringdahl, B.; Jenden, D. J. *Life Sci.* **1983**, *32*, 2401.
(9) Dahlbom, R.; Karlén, B.; George, R.; Jenden, D. J. *J. Med. Chem.* **1966**, *9*, 843.
(10) Karlén, B.; Lindeke, B.; Lindgren, S.; Svensson, K.-G.; Dahlbom, R.; Jenden, D. J.; Giering, J. E. *J. Med. Chem.* **1970**, *13*, 651.

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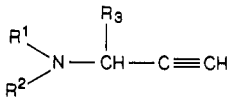
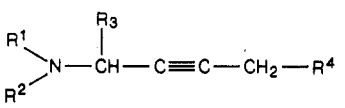
Table I. Physical Data of the Intermediates



compd	R^1	R^2	R^3	prepn meth[a]	yield, %	bp (mmHg) or mp, °C	formula
1	CH_3SO_2	H	H	A	71	55.5–58[b]	$C_4H_7NO_2S$
2	CH_3SO_2	H	CH_3	A	90	46–48[b]	$C_5H_9NO_2S$
3	CH_3SO_2	CH_3	H	C	60	76–78 (0.3)	$C_5H_9NO_2S \cdot {}^1/_4H_2O$
4	CH_3SO_2	CH_3	CH_3	C	97	73–74[c]	$C_6H_{11}NO_2S$
5	CH_3SO_2	CH_3SO_2	H	D	55	111.5–112.5[c]	$C_5H_9NO_4S_2$
6	CH_3SO_2	CH_3SO_2	CH_3	D	29	96–99[c]	$C_6H_{11}NO_4S_2$
7	CH_3SO_2	CH_3CO	H	E	91	45–46[d]	$C_6H_9NO_3S$
8	CH_3SO_2	CH_3CO	CH_3	E	80	73–75 (0.3)	$C_7H_{11}NO_3S$
9	CH_3CO	H	CH_3	*e*	81	91–92[c]	
10	CH_3CO	CH_3CO	H	G	94	136–137 (48)	$C_7H_9NO_2$[f]
11	CH_3CO	CH_3CO	CH_3	F	65	94–95 (10)	$C_8H_{11}NO_2$[g]
12	CF_3CO	H	H	B	65	*h*	
13	CF_3CO	H	CH_3	B	74	70 (18)	$C_6H_6F_3NO$[i]
14	CF_3CO	CH_3	H	C	56	57 (15)	$C_6H_6F_3NO$[j]
15	CF_3CO	CH_3	CH_3	C	72	50–51 (10)	$C_7H_8F_3NO$[k]

[a] See the Experimental Section. [b] From ether–petroleum ether. [c] From ether. [d] From ether–cyclohexane. [e] Previously described (ref 1), mp 92.5–93 °C. [f] Anal. Calcd for $C_7H_9NO_2$: C, 60.4; N, 10.1. Found: C, 59.2; N, 9.5. MS (12 eV) m/z 97 ($M^+ - CH_2CO$). [g] Anal. Calcd for $C_8H_{11}NO_2$: C, 62.7. Found: C, 62.2. MS (12 eV) m/z 111 ($M^+ - CH_2CO$). [h] Previously reported (ref 30), bp 95 °C (11 mmHg). [i] Anal. Calcd for $C_6H_6F_3NO$: C, 43.6. Found: C, 42.0. MS (70 eV) m/z 150 ($M^+ - CH_3$). [j] Anal. Calcd for $C_6H_6F_3NO$: N, 8.5. Found: N, 9.4. MS (70 eV) m/z 165 (M^+). [k] Anal. Calcd for $C_7H_8F_3NO$: C, 46.9; N, 7.8. Found: C, 45.3; N, 7.3. MS (70 eV) m/z 165 ($M^+ - CH_3$).

Table II. Physical Data for the Compounds Tested



compd	R^1	R^2	R^3	R^4[a]	yield, %	recrystn solvents[b]	mp, °C	formula[c]
16	CH_3SO_2	CH_3	H	NC_4H_8	65	A	76–79	$C_{10}H_{18}N_2O_2S \cdot C_2H_2O_4$
17	CH_3SO_2	CH_3	CH_3	NC_4H_8	64	A	112–114.5	$C_{11}H_{20}N_2O_2S \cdot C_2H_2O_4$
18	CH_3SO_2	CH_3SO_2	H	NC_4H_8	80	B	145.5–146.5	$C_{10}H_{18}N_2O_4S_2 \cdot C_2H_2O_4$
19	CH_3SO_2	CH_3SO_2	CH_3	NC_4H_8	84	B	162–163	$C_{11}H_{20}N_2O_4S_2 \cdot C_2H_2O_4$
20	CH_3SO_2	CH_3CO	H	NC_4H_8	90	C	124–126	$C_{11}H_{18}N_2O_3S \cdot C_2H_2O_4$
21	CH_3SO_2	CH_3CO	CH_3	NC_4H_8	70	A	120–122	$C_{12}H_{20}N_2O_3S \cdot C_2H_2O_4$
22	CF_3CO	CH_3	H	NC_4H_8	75	A	83–84	$C_{11}H_{15}F_3N_2O \cdot C_2H_2O_4$
23	CF_3CO	CH_3	CH_3	NC_4H_8	40	A	60–62.5	$C_{12}H_{17}F_3N_2O \cdot C_2H_2O_4 \cdot {}^1/_4H_2O$
24	CF_3CO	CH_3	CH_3	$N(CH_3)_2$	28	A	117–119	$C_{10}H_{15}F_3N_2O \cdot C_2H_2O_4$
25	CF_3CO	CH_3	CH_3	$N(CH_3)_3^+$	54	C	163–165	$C_{11}H_{18}F_3N_2O \cdot I$
26	CH_3CO	CH_3CO	H	NC_4H_8	85	C	70–72	$C_{12}H_{18}N_2O_2 \cdot C_2H_2O_4$
27	CH_3CO	CH_3CO	H	$N(C_2H_5)_2$	65	B	120–121.5	$C_{12}H_{20}N_2O_2 \cdot C_2H_2O_4$
28	CH_3CO	CH_3CO	H	$N(CH_3)_2$	46	A	108–110	$C_{10}H_{16}N_2O_2 \cdot C_2H_2O_4$
29	CH_3CO	CH_3CO	H	$N(CH_3)_3^+$	38	D	109–111	$C_{11}H_{19}N_2O_2 \cdot I$
30	CH_3CO	CH_3CO	CH_3	NC_4H_8	43	E	78.5–79.5	$C_{13}H_{20}N_2O_2 \cdot C_2H_2O_4 \cdot {}^1/_4H_2O$
31	CH_3CO	CH_3CO	CH_3	$N(C_2H_5)_2$	55	A	81.5–82.5	$C_{13}H_{22}N_2O_2 \cdot C_2H_2O_4 \cdot {}^1/_2H_2O$
32	CH_3CO	CH_3CO	CH_3	$N(CH_3)_2$	66	A	87.5–89.5	$C_{11}H_{18}N_2O_2 \cdot C_2H_2O_4$
33	CH_3CO	CH_3CO	CH_3	$N(CH_3)_3^+$	87	D	109–110.5	$C_{12}H_{21}N_2O_2 \cdot I$

[a] NC_4H_8 = pyrrolidino. [b] Recrystallization solvents: A, EtOH–ether; B, EtOH; C, MeOH–ether; D, acetone–ether; E, acetone. [c] All compounds were analyzed for C, H, and N. The analytical results obtained for those elements were within ±0.4% of the theoretical value.

CH_2Cl_2, and aminolysis occurred when excess of the secondary amine was used in the Mannich reactions of compounds **10** and **11**. For example, a 6:1 mixture of imide **27** and the corresponding secondary amide was formed when compound **10** was subjected to a Mannich reaction in the presence of 1.25 equiv of diethylamine.

Physical data of the synthetic intermediates **1–15** and of the target compounds **16–33** are presented in Tables I and II, respectively.

Spectroscopy. NMR spectroscopic data of the compounds are given in the supplementary material. Some compounds gave informative spectra that allowed determination of certain conformational characteristics.

It was evident from ^1H, ^{13}C, and ^{19}F NMR spectroscopy that the trifluoroacetamides exist as an equilibrium mixture of *E* and *Z* conformations in solution. Resonances could be assigned to *E* or *Z* conformations on the basis of the magnitude of the fluorine–proton couplings observed in the ^1H and ^{19}F NMR spectra and on the magnitude of the carbon–fluorine couplings in the ^{13}C NMR spectra. The relative proportions of the two conformations were determined by integration over appropriate resonances in the ^1H and ^{19}F NMR spectra (information obtained from ^{19}F NMR spectroscopy is collected in Table IV). For example, integration of the *N*-Me resonances due to the *Z* (δ 3.17, $^5J_{H,F}$ = 1.7 Hz) and *E* (δ 3.05; $^5J_{H,F} < 1$ Hz) conformations of the oxalate of **23** in CD_3OD indicated that the *Z* conformation predominated (4:1) at 20 °C. Corroborating results were obtained by integration of the quartet (δ −69.7; *Z* conformation) and the broad singlet (δ −68.5; *E* conformation), which were observed as the sole resonances in the ^{19}F NMR spectrum of **23**. The *N*-Me resonance due to the *Z* conformation appeared as a well-resolved quartet (δ 30.7; $J_{C,F} \approx 4$ Hz) in the proton de-

Table III. Muscarinic and Antimuscarinic Effects of Some Oxotremorine Analogues[a]

compd	guinea pig ileum					tremorolytic dose in mice,[f] μmol/kg
	EC_{50},[b] μM	E_{max}[c]	EPMR[d]	K_D, μM	relative efficacy[e]	
16				68.0 ± 8.6 (4)		174 ± 51
17				13.0 ± 3.5 (4)		82.0 ± 17.6
18				15.9 ± 4.3 (5)		>200
19				5.4 ± 0.06 (4)		157 ± 44
20				17.9 ± 2.0 (5)		>200
21				3.1 ± 0.3 (5)		99.7 ± 23.0
22	16.9 ± 5.2 (5)	0.16 ± 0.03 (5)	145 ± 38 (5)	15.6 ± 3.4 (4)	0.0030 ± 0.0004 (4)	150 ± 41
23		0		0.57 ± 0.03 (7)		21.4 ± 2.5
24		<0.1		95.4 ± 12.6 (5)		>200
25	27.1 ± 3.5 (7)	0.89 ± 0.04 (7)	214 ± 24 (7)	91.9 ± 22.1 (5)	0.026 ± 0.004 (5)	
26		0		0.44 ± 0.06 (5)		2.9 ± 0.5
27		0		7.2 ± 0.6 (5)		107 ± 19
28	38.8 ± 10.1 (6)	0.84 ± 0.07 (6)	320 ± 12 (6)	65.6 ± 11.9 (5)	0.013 ± 0.003 (5)	>200
29	2.0 ± 0.3 (8)	1.00 ± 0.02 (8)	19.3 ± 1.8 (8)	8.2 ± 2.1 (7)	0.034 ± 0.008 (7)	
30		0		0.28 ± 0.04 (7)		3.2 ± 1.0
31		0		1.6 ± 0.2 (6)		39.0 ± 5.3
32	5.5 ± 0.7 (6)	0.99 ± 0.03 (6)	57.9 ± 2.9 (6)	21.6 ± 6.5 (5)	0.030 ± 0.006 (5)	109 ± 11.3
33	1.2 ± 0.1 (8)	1.01 ± 0.02 (8)	10.7 ± 0.7 (8)	8.6 ± 1.9 (7)	0.055 ± 0.01 (7)	
34	0.069 ± 0.002 (7)	1.01 ± 0.01 (7)		2.2 ± 0.3 (6)	0.15 ± 0.02 (6)	*g*
35	0.19 ± 0.03 (4)	0.83 ± 0.003 (4)		0.24 ± 0.07 (4)	0.013 ± 0.0002 (4)	0.6
38	1.0 ± 0.03 (8)	0.98 ± 0.01 (8)		5.0 ± 0.5 (5)	0.027 ± 0.002 (5)	23
40	2.5 ± 0.1 (9)	1.01 ± 0.01 (9)		208 ± 30 (6)	0.35 ± 0.07 (6)	*g*
42				0.15 ± 0.01 (4)[h]		1.2[i]
carbachol	0.11 ± 0.01 (6)	1.00	1.00	15.4 ± 3.1 (6)	1.00	

[a] Values represent means plus or minus standard errors. [b] The concentration of an agonist or a partial agonist that elicits 50% of its own maximum contractile response. [c] The maximum contractile response relative to that elicited by carbachol. [d] Equipotent molar ratio relative to that of carbachol, which equals 1.00. [e] Efficacy relative to that of carbachol; calculated from EC_{50} and K_D values as described in ref 6 and 15. [f] Dose required to double the dose of oxotremorine inducing a predetermined (grade 2) tremor intensity in mice. [g] Agonist. [h] Unpublished. [i] From ref 10.

Table IV. ^{19}F NMR Spectral Data of Some Trifluoroacetamides[a]

compd	solvent	chemical shifts, δ		% *Z*
		Z conformer[b]	*E* conformer[b]	
12	$CDCl_3$	−76.7		
13	$CDCl_3$	−76.3		
14	$CDCl_3$	−70.4	−69.6	72
15	$CDCl_3$	−70.2	−69.0	77
22	CD_3OD	−69.5	−68.6	74
23	CD_3OD	−69.7	−68.5	79
24	CD_3OD	−69.8	−68.6	81
25	CD_3OD	−69.7	−68.5	83

[a] Spectra were referenced to internal $CFCl_3$. [b] Assignment was based on the magnitude of the $^5J_{F,H}$ coupling constants.

coupled ^{13}C NMR spectrum of the oxalate of **23**. In the *E* conformation of **23**, this carbon was observed as a broad unresolved resonance (δ 30.0). The other trifluoroacetamides (**22**, **24**, and **25**) were found to have similar conformational preferences (*Z*/*E* around 4:1; compare Figure 1). In $CDCl_3$, the acetamide analogue (base) of **22** exists to about 60% in the *Z* conformation at 25 °C,[11] and the base form of acetamide **35** assumes a 3:1 ratio of the *Z* and *E* conformations at 37 °C in the same solvent.[1] Integration of the *N*-Me resonances in the spectrum of the oxalate of **35** in CD_3OD at 25 °C shows that the *Z* conformation is considerably more favored under these latter conditions (*Z*/*E* = 4:1).

Acyclic imides can assume *E*,*E*, *E*,*Z*, or *Z*,*Z* conformations.[12] In ^1H NMR spectra of CD_3OD solutions of the oxalates of **26–28** and **30–32**, the two acetyl groups appeared as a single resonance at 20 °C. These observations indicate either that **26–28** and **30–32** assume mainly one symmetric imide conformation or that the barrier to conformational interconversion is low. In contrast, ^1H NMR spectra of the methiodides **29** and **33** in CD_3OD at 20 °C exhibited two resonances (**29**, δ 1.97 and 2.02; **33**, δ 1.96 and 2.02) due to the acetyl groups that were of equal intensity and were assigned to the unsymmetrical *E*/*Z* conformation.

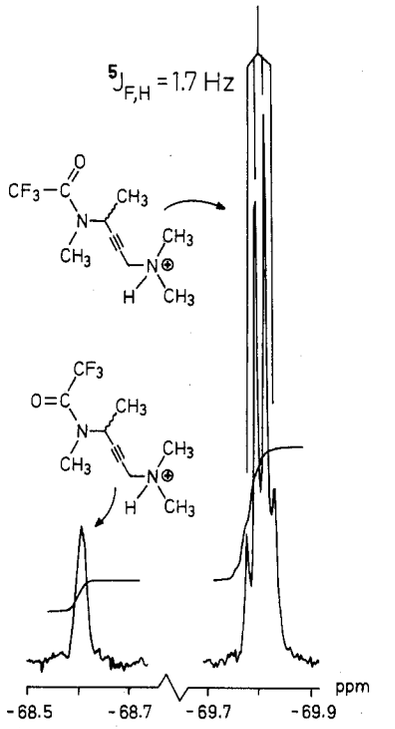


Figure 1. Partial ^{19}F NMR spectrum of **24** in CD_3OD (at 20 °C), which demonstrates the predominance of the *Z* conformation.

The IR spectra of **29** and **33** (KBr pellets) showed two carbonyl absorption bands, one of stronger intensity at ν 1715 cm^{-1} and one of medium intensity at ν 1695–1700

(11) Resul, B.; Ringdahl, B.; Hacksell, U.; Svensson, U.; Dahlbom, R. *Acta Pharm. Suec.* **1979**, *16*, 225.
(12) (a) Noe, E.; Raban, M. *J. Am. Chem. Soc.* **1973**, *95*, 6118; **1975**, *97*, 5811. (b) Tetu, G.; Duplan, J.-C.; Pellissier, N.; Laurent, E. *J. Chem. Res. Synopses* **1978**, 98.

cm^{-1}. Only a single strong carbonyl absorption band at ν 1710 cm^{-1} was observed for the bases of the corresponding tertiary amines (liquid film).

Pharmacology

Compounds **16–33** were tested for muscarinic and antimuscarinic activity in the isolated guinea pig ileum and in intact mice. The results are presented in Table III. For comparison, relevant data for **34**, **35**, **38**, **40**, and **42** are also included in the table. In Table III, E_{max} values are equivalent to intrinsic activities in the terminology of Ariens.[13] K_D is the equilibrium dissociation constant of the drug–receptor complex and is a measure of affinity.[14] K_D values were determined on the ileum by methods[15] that also provided reliable estimates of agonist efficacy.[16]

None of the *N*-methanesulfonyl-substituted analogues (**16–21**) showed spasmogenic activity on the guinea pig ileum. Instead, they were antagonists to carbachol on this tissue. Spasmolytic activity as well as tremorolytic activity among **16–21** increased with methyl substitution in position 1 of the butynyl chain (Table III).

The trifluoroacetamides **22** and **25** were about 100-fold less potent than carbachol on the ileum. They elicited contractile responses that were significantly ($P < 0.05$) lower than those produced by carbachol. Compound **24** also appeared to be a partial agonist, but its efficacy was too low to be measured. In contrast, **23** was a pure antagonist having relatively high affinity for muscarinic receptors.

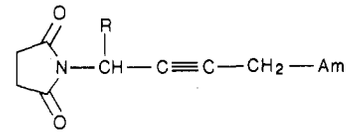
Among the acyclic imides (**26–33**), the dimethylamino (**28** and **32**) and trimethylammonium (**29** and **33**) derivatives were full or nearly full agonists on the ileum, but they were more than 10-fold less potent than carbachol. This was due primarily to lower efficacy, since the affinities of **28**, **29**, **32**, and **33** were similar to that of carbachol. The pyrrolidine derivatives **26** and **30** were potent antagonists to carbachol on the ileum and to the central effects of oxotremorine in vivo. Trifluoroacetamides and acyclic imides are known to be sensitive to hydrolysis.[17,18] The possibility that the compounds studied were broken down in the tissue bath was not investigated in detail. However, the high potency of **23**, **26**, and **30** in vitro suggests that hydrolysis was not extensive.

Among the tertiary amines, only **32** showed visible muscarinic effects in vivo; doses of **32** that produced tremor blockade also gave profuse salivation. The trimethylammonium salts were not tested in vivo.

Discussion

Replacement of the acetyl or *N*-methyl group (or both) of **35** by a methanesulfonyl group decreased affinity and abolished efficacy at muscarinic receptors in the guinea pig ileum as the resulting compounds (**17**, **19**, and **21**) were relatively weak antagonists (Table III). Similarly, substitution of a trifluoroacetyl group for the acetyl group of **35** (yielding **23**) abolished efficacy, but caused only a 2-fold reduction in affinity. The trifluoroacetamides **22** and **25** were considerably less potent in causing contractions of the ileum than the previously reported acetamide analogues **34**[19] and **37**.[4] Compounds **22–24** were relatively poor tremorolytic agents in vivo. This may be due partly to the hydrolytic instability of these compounds.[17] The trifluoroacetamides and the acetamides appear to have similar conformational preferences (see above). Thus, the lower in vitro potency of the trifluoroacetamides as compared to the analogous acetamides does not appear to be related to conformational differences. Instead, it may be related to the decreased oxygen-electron density (increased C=O stretching frequency; **23**, ν 1695 cm^{-1}, and **35**, ν 1640 cm^{-1}) in the fluorinated amides (compare ref 20).

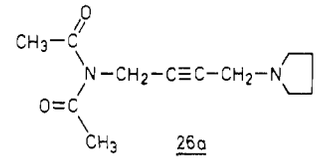
Substitution of an acetyl group for the *N*-methyl group of **34–37** decreased (**32** and **33**) or abolished (**26** and **30**) efficacy without having much effect on affinity. The acyclic imides **28** and **29** were about 15-fold less potent on the ileum than the corresponding succinimides **40** and **41**.[21]



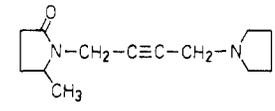
38: R = H, Am = NC_4H_8
39: R = H, Am = $N(C_2H_5)_2$
40: R = H, Am = $N(CH_3)_2$
41: R = H, Am = $N^+(CH_3)_3$
42: R = CH_3, Am = NC_4H_8

This was due entirely to lower efficacy of **28** and **29** as they had somewhat higher affinity than their succinimide analogues. Similarly, the antagonists **26** and **27** had 5- to 10-fold higher affinity than the succinimides **38** and **39**.[21]

It is noteworthy that **38** was an agonist, albeit of low efficacy,[21] whereas **26** was an antagonist, in agreement with the lower efficacy of the acyclic imides. It is tempting to suggest that the 10-fold higher affinity of **26** as compared to **38** is due to the presence of the *E*/*Z* conformer **26a** which resembles closely the potent antagonist **43**.[22] The



26a



43

10-fold higher affinity of **43** as compared to oxotremorine was explained by additional binding of the methyl group to the receptor.[23,24] It seems likely that the *Z*-methyl group of **26a** also can participate in such binding.

Methyl substitution in the butynyl chain of **26–29** had little effect on affinity and efficacy. In contrast, analogous methyl substitution in the succinimides **38–41** in general leads to substantial increase in affinity and sharp decrease in efficacy. For example, addition of a methyl group in position 1 of the butynyl chain of **38**, giving **42**, increased affinity for muscarinic receptors 30-fold and abolished efficacy (Table III).[10,25] The tremorolytic potencies of the acyclic imides were similar to or lower than those of the

(13) Ariens, E. J., Ed. *Molecular Pharmacology. The Mode of Action of Biologically Active Compounds* Academic: New York, 1964; Vol. 1.
(14) Furchgott, R. F. *Adv. Drug Res.* **1966**, *3*, 21.
(15) Ringdahl, B. *J. Pharmacol. Exp. Ther.* **1984**, *229*, 199.
(16) Stephenson, R. P. *Br. J. Pharmacol. Chemother.* **1956**, *11*, 379.
(17) Greene, T. W. *Protective Groups in Organic Synthesis*; Wiley: New York, 1981; p 254.
(18) Corey, E. J.; Sheehan, J. C. *J. Am. Chem. Soc.* **1952**, *74*, 4555.
(19) Bebbington, A.; Brimblecombe, R. W.; Shakeshaft, D. *Br. J. Pharmacol. Chemother.* **1966**, *26*, 56. Ringdahl, B.; Resul, B.; Jenden, D. J.; Dahlbom, R. *Eur. J. Pharmacol.* **1982**, *85*, 79.
(20) Bebbington, A.; Brimblecombe, R. W. *Adv. Drug. Res.* **1965**, *2*, 143.
(21) Ringdahl, B. *Eur. J. Pharmacol.* **1987**, *140*, 13.
(22) Ringdahl, B.; Muhi-Eldeen, Z.; Ljunggren, C.; Karlén, B.; Resul, B.; Dahlbom, R.; Jenden, D. J. *Acta Pharm. Suec.* **1979**, *16*, 89.
(23) Ringdahl, B.; Jenden, D. J. *Mol. Pharmacol.* **1983**, *23*, 17.
(24) Amstutz, R.; Ringdahl, B.; Karlén, B.; Roch, M.; Jenden, D. J. *J. Med. Chem.* **1985**, *28*, 1760.
(25) Ringdahl, B. *Proceedings of the 8th International Symposium on Medicinal Chemistry*; Dahlbom, R., Nilsson, J. L. G., Eds.; Swedish Pharmaceutical: Stockholm, 1985; Vol. 2, p 391.

corresponding succinimides with the exception of **26**, which was almost 10-fold more potent than **38**. It should be noted that acyclic imides are less stable toward solvolysis than cyclic imides[18] and that this may influence considerably the in vivo data.

In summary, the structural modifications of **35**, which are described herein, generally gave compounds with lower efficacies than that of **35**. Compounds showing agonist or partial agonist properties had lower affinity than **35**. However, some of the compounds were fairly potent antagonists.

Experimental Section

Chemistry. General Comments. Melting points (uncorrected) were determined in open glass capillaries on a Thomas-Hoover apparatus. IR spectra were recorded on a Perkin-Elmer 298 infrared spectrophotometer. ^1H, ^{19}F, and ^{13}C NMR spectra were recorded on a JEOL FX 90Q spectrometer at 90, 84.3, and 22.5 MHz, respectively. ^1H NMR and ^{13}C NMR spectra were referenced to internal tetramethylsilane. ^{19}F NMR spectra were referenced to internal $CFCl_3$. Mass spectra were obtained at 70 eV on a LKB 9000 spectrometer using a direct insertion probe. All spectra were in accordance with the assigned structures. The elemental analyses, which were performed by Mikro Kemi AB, Uppsala, Sweden, were within ±0.4% of the calculated values unless otherwise noted.

***N*-(2-Propynyl)methanesulfonamide (1). Method A.** A solution of methanesulfonyl chloride (4.0 g, 35 mmol) in dry ether was carefully added to a cooled (0 °C) solution of propargylamine (3.8 g, 69 mmol) in dry ether (60 mL) under N_2. The resulting mixture was stirred for 30 min, and the precipitate that formed was filtered off and washed with ether. Concentration of the combined ether layers afforded the crude sulfonamide, which was purified by recrystallization: ^1H NMR ($CDCl_3$) δ 5.30 (br s, NH), 3.96 (dd, *N*-CH_2), 3.11 (s, CH_3SO_2), 2.43 (t, J = 2.5 Hz, CCH); ^{13}C NMR ($CDCl_3$) δ 79.06 (C2), 73.32 (C3), 41.35 (CH_3SO_2), 32.55 (C1).

When triethylamine or pyridine was used as acid scavengers, the yield of **1** was lower, and some dimesylation occurred; for example, when 1 equiv of pyridine was used, the crude reaction product consisted of **1** and dimesylated product (**5**) in a 5:1 ratio.

***N*-(1-Methyl-2-propynyl)trifluoroacetamide (13). Method B.** A solution of trifluoroacetic anhydride (9.0 g, 43 mmol) in dry ether (30 mL) was added to a stirred solution of 1-methyl-2-propynylamine[26] (2.7 g, 39 mmol) and pyridine (3.1 g, 39 mmol) in dry ether (40 mL) under N_2 at 0 °C. After 30 min, the pyridinium trifluoroacetate was filtered off, and the ether was evaporated. The oily residue was purified by distillation: ^1H NMR ($CDCl_3$) δ 6.70 (br s, NH), 4.82 (m, *N*-CH), 1.51 (d, J = 6.9 Hz, CCH_3), 2.37 (d, J = 2.4 Hz, CCH); ^{13}C NMR ($CDCl_3$) δ 158.06 (q, $J_{C,F}$ = 38 Hz, CF_3CO), 116.06 (q, $J_{C,F}$ = 287 Hz, CF_3CO), 38.11 (C1), 21.37 (C1 Me), 82.00 (C2), 71.90 (C3).

***N*-Methyl-*N*-(2-propynyl)methanesulfonamide (3). Method C.** A solution of iodomethane (0.65 g, 4.6 mmol) in dry acetone was added to a stirred mixture of **1** (0.52 g, 3.9 mmol) and a large excess of anhydrous powdered K_2CO_3 (8.0 g, 58 mmol) in dry acetone (75 mL). The reaction mixture was heated under N_2 for 5 h. Ether (25 mL) was added, and the mixture was filtered. Concentration of the ether layer afforded **3** as an oil, which was purified by distillation: ^1H NMR ($CDCl_3$) δ 4.08 (d, J = 2.5 Hz, *N*-CH_2), 2.95, 2.92 (s's, CH_3SO_2, *N*-CH_3), 2.40 (t, J = 2.5 Hz, CCH); ^{13}C NMR ($CDCl_3$) δ 76.93 (C2), 75.17 (C3), 39.41, 36.16 (*N*-CH_3, CH_3SO_2), 34.25 (C1).

***N*-(1-Methyl-2-propynyl)dimethanesulfonamide (6). Method D.** Methanesulfonyl chloride (3.7 g, 32 mmol) was added dropwise to a mixture of *N*-(1-methyl-2-propynyl)sulfonamide (**2**; 1.0 g, 6.8 mmol), sodium hydroxide (0.31 g, 7.8 mmol), and water (7 mL). The mixture was stirred at room temperature for 12 h. NaOH (5 M, 10 mL) was added, and the solution was extracted twice with ether. The combined ether layers were dried (K_2CO_3) and concentrated to give crude **6**, which was purified by recrystallization: ^1H NMR ($CDCl_3$) δ 5.30 (dq, *N*-CH), 3.38 (s, 2 × CH_3SO_2), 2.57 (d, J = 2.4 Hz, CCH), 1.77 (d, J = 7.0 Hz, CCH_3); ^{13}C NMR ($CDCl_3$) δ 80.73 (C2), 73.84 (C3), 47.62 (C1), 44.75 (2 × CH_3SO_2), 22.30 (C1 Me).

The reaction affording **5** was complete in 30 min.

***N*-Acetyl-*N*-(2-propynyl)methanesulfonamide (7). Method E.** A solution of acetyl chloride (3.3 g, 42 mmol) in dry acetone (5 mL) was added to a mixture of **1** (2.3 g, 17 mmol) and anhydrous, powdered K_2CO_3 (30.0 g, 217 mmol) in dry acetone (100 mL). The reaction mixture was kept under N_2 at room-temperature for 24 h. Filtration followed by evaporation of volatiles yielded crude **7** as an oil, which solidified upon cooling. Pure **7** was obtained after recrystallization: ^1H NMR ($CDCl_3$) δ 4.57 (d, J = 2.4 Hz, *N*-CH_2), 3.33 (s, CH_3SO_2), 2.46 (s, CH_3CO), 2.42 (t, J = 2.4 Hz, CCH); ^{13}C NMR ($CDCl_3$) δ 170.23 (CH_3CO), 77.80 (C2), 73.23 (C3), 42.50 (CH_3SO_2), 35.30 (C1), 24.43 (CH_3CO).

***N*-(1-Methyl-2-propynyl)diacetamide (11). Method F.** A solution of acetyl chloride (7.7 g, 98 mmol) in dry ether (10 mL) was slowly added to a precooled (0 °C) mixture of **9** (1.0 g, 9 mmol) and triethylamine (4.5 g, 45 mmol) in dry ether (100 mL). The reaction mixture was allowed to reach room temperature and was then stirred under N_2 for 24 h. The resulting mixture was filtered, and the volatiles were evaporated. Ether (100 mL) was added to the residue, and the ether solution was extracted with five 15-mL portions of saturated aqueous $NaHCO_3$. Concentration of the dried (K_2CO_3) organic layer, followed by chromatography on silica gel using CH_2Cl_2 as eluant, afforded **11** as a colorless oil: ^1H NMR ($CDCl_3$) δ 5.61 (dq, *N*-CH), 2.48 (s, 2 × CH_3CO), 2.39 (d, J = 2.6 Hz, CCH), 1.57 (d, J = 7.0 Hz, CCH_3); ^{13}C NMR ($CDCl_3$) δ 172.89 (2 × CH_3CO), 82.40 (C2), 72.33 (C3), 40.80 (C1), 26.78 (2 × CH_3CO), 20.75 (C1 Me).

***N*-(2-Propynyl)diacetamide (10). Method G.** Propargyl bromide (3.3 g, 28 mmol) was added to a stirred mixture of diacetamide[27] (2.0 g, 20 mmol) and dry powdered K_2CO_3 (20 g, 145 mmol) in acetonitrile (40 mL). The mixture was stirred under N_2 for 3 days at room temperature. The mixture was filtered, and the volatiles were evaporated. The oily residue was purified by flash chromatography on silica gel using CH_2Cl_2 as eluant: ^1H NMR ($CDCl_3$) δ 4.49 (d, J = 2.4 Hz, *N*-CH_2), 2.50 (s, 2 × CH_3CO), 2.27 (t, J = 2.4 Hz, CCH); ^{13}C NMR ($CDCl_3$) δ 171.28 (2 × CH_3CO), 77.61 (C2), 70.91 (C3), 32.80 (C1), 25.23 (2 × CH_3CO).

***N*-Acetyl-*N*-[4-(diethylamino)-1-methyl-2-butynyl]acetamide (31). Method H.** A solution of **11** (0.30 g, 1.96 mmol) in dioxane (5 mL) was added to a stirred mixture of paraformaldehyde (0.07 g, 2.33 mmol), diethylamine (0.14 g, 1.91 mmol), glacial acetic acid (0.12 g, 2 mmol), and a few grains of cuprous chloride in dioxane (10 mL). The mixture was stirred under N_2 at room temperature for 20 h. The volatiles were evaporated, and the oily residue was dissolved in 1 M HCl (40 mL). The water solution was extracted with ether (3 × 30 mL). Saturated aqueous $NaHCO_3$ was added to render the aqueous layer alkaline (pH ~8). Extraction with CH_2Cl_2 (6 × 45 mL) and drying (K_2CO_3) and concentration of the organic layer yielded crude **31** as an oily residue. The base was converted into the oxalate and recrystallized: ^1H NMR (CD_3OD) δ 5.31 (m, *N*-CH), 4.11 (d, J = 1.7 Hz, CH_2N), 3.30 (q, J = 7.3 Hz, $N(CH_2C)_2$), 2.41 (s, 2 × CH_3CO), 1.59 (d, J = 7.0 Hz, CCH_3), 1.32 (t, J = 7.3 Hz, $N(CCH_3)_2$); ^{13}C NMR (CD_3OD) δ 174.40 (2 × CH_3CO), 90.12 (C2), 72.36 (C3), 48.92 (C4), 44.35 (C1), 41.69 ($N(CH_2CH_3)_2$), 26.81 (2 × CH_3CO), 20.41 (C1 Me), 9.61 ($N(CH_2CH_3)_2$).

Further Comments. Excess secondary amine (1.2 equiv) was added in the preparation of **16–24**. The reaction mixtures giving **16** and **17** were heated at 80 °C for 2.5–5 h. Aqueous NaOH was used to alkalinize the acidic water layer in the workup of **16–19**. Crude **27** was contaminated with around 15% of the corresponding secondary amide (^1H NMR); IR spectroscopy of crude **27** revealed absorptions at ν 3300 cm^{-1} (N—H stretching), 1650–1670 cm^{-1} (C=O stretching), and 1540–1550 cm^{-1} (N—H bending) due to the impurity. An IR spectrum of pure **27** (obtained after repetitive recrystallization from ethanol) showed a single carbonyl absorption at ν 1710 cm^{-1}.

(4-Diacetamidopent-2-ynyl)trimethylammonium Iodide (33). Method I. An excess of iodomethane (0.5 mL, 8 mmol) was added to the free base of **32** (generated from 0.26 g, 0.86 mmol,

(26) Marszak-Fleury, A. *Ann. Chim. (Paris)* **1958**, *13*, 656.

(27) Titherly, A. W. *J. Chem. Soc.* **1898**, *79*, 411.

of the oxalate) in dry acetone (8 mL). After 1.5 h at room temperature, ether was added, and the precipitate that formed was recrystallized: ^{1}H NMR (CD_3OD) δ 4.73 (m, *N*-CH), 4.37 (d, J = 1.7 Hz, CH_2N^+), 3.24 (s, $N^+(CH_3)_3$), 2.02 and 1.96 (2 s, CH_3CO), 1.44 (d, J = 7.1 Hz, CCH_3); ^{13}C NMR (CD_3OD) δ 173.32 and 172.18 (CH_3CO), 94.10 (C2), 70.60 (C3), 57.54 (C4), 53.58 ($N^+(CH_3)_3$), 38.02 (C1), 21.28 (C1 Me), 22.70 and 20.69 (CH_3CO).

Pharmacology. Guinea Pig Ileum. A standard guinea pig ileum preparation was set up in Tyrode solution (pH 7.4) at 37 °C as described previously.[15] The Tyrode solution contained hexamethonium (0.03 mM). Contractions were recorded isotonically at 1 g of tension. Concentration–response curves were constructed by the cumulative dose–response technique by increasing stepwise the concentration of agonist by a factor of 2.15. Densensitization during cumulative addition of drugs was unlikely to occur since preliminary experiments showed that there was no difference in agonist potency measured by cumulative and intermittent administration.

Spasmogenic activity (EC_{50} values) was estimated by interpolation at the 50% response level of each compound. Maximal contractile responses (E_{max}) and equipotent molar ratios (EPMR) were expressed with reference to carbachol. Statistical significance ($P < 0.05$) of differences between mean E_{max} and EMPR values was determined by the Student's t test.

Dissociation constants (K_D) and relative efficacies of **22**, **25**, **28**, **29**, **32**, and **33** at muscarinic receptors in the ileum were determined by comparison of their concentration–response curves with that of carbachol as described previously for compounds acting as partial agonists.[6,15] K_D values of antagonists were estimated by using carbachol as the agonist.[15,21] The antagonist was allowed to equilibrate with the tissue for 15 min before the addition of carbachol. At least four different concentrations of each antagonist were used.

Tremorolytic Activity in Mice. Male Swiss–Webster mice (24–32 g body weight) were used. The test compounds were administered ip at a dose of 200 μmol/kg to groups of six or more mice, while six control animals remained untreated. Twenty minutes after drug administration, the ED_{50} value of oxotremorine, injected iv, was estimated by the up-and-down method[28] with intermittent spontaneous (grade 2) tremor[29] as the end point. Compounds that showed no significant antagonism of oxotremorine under these conditions were not tested further. Test of significance ($P < 0.05$) of differences between ED_{50} values of oxotremorine in the presence and absence of antagonist was performed, assuming that the ratio of the difference to its standard error was normally distributed. Compounds that significantly blocked oxotremorine at a dose of 200 μmol/kg were tested at two additional doses. For the latter compounds, the ED_{50} value of oxotremorine was plotted against the dose of antagonist (including zero) used for premedication. That dose of antagonist that doubled the ED_{50} value of oxotremorine was estimated by linear regression analysis.[5,24] Under these conditions, atropine and **35** had tremorolytic doses of 0.9 and 0.6 μmol/kg, respectively.

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Registry No. **1**, 93501-84-9; **2**, 111903-20-9; **3**, 111903-21-0; **4**, 111903-22-1; **5**, 111903-23-2; **6**, 111903-24-3; **7**, 111903-25-4; **8**, 111903-26-5; **9**, 42105-25-9; **10**, 111903-27-6; **11**, 111903-28-7; **12**, 14719-21-2; **13**, 111903-29-8; **14**, 111903-30-1; **15**, 111903-31-2; **16**, 111903-33-4; **17**, 111903-35-6; **18**, 111903-37-8; **19**, 111903-39-0; **20**, 111903-41-4; **21**, 111903-43-6; **22**, 111903-45-8; **23**, 111903-47-0; **24**, 111903-49-2; **25**, 111903-50-5; **26**, 111903-52-7; **27**, 111903-54-9; **28**, 111903-56-1; **29**, 111903-58-3; **30**, 111903-60-7; **31**, 111903-62-9; **32**, 111903-64-1; **33**, 111903-65-2; butynylamine, 41282-40-0; dimethylamine, 124-40-3; trimethylamine, 75-50-3; diethylamine, 109-89-7; methanesulfonyl chloride, 124-63-0; propargylamine, 2450-71-7; trifluoroacetic anhydride, 407-25-0; 1-methyl-2-propynylamine, 30389-17-4; iodomethane, 74-88-4; acetyl chloride, 75-36-5; propargyl bromide, 106-96-7; diacetamide, 625-77-4; paraformaldehyde, 30525-89-4.

Supplementary Material Available: Four tables with ^{1}H and ^{13}C NMR spectral data of compounds **1**–**35** (6 pages). Ordering information is given on any current masthead page.

(28) Dixon, W. J. *J. Am. Stat. Assoc.* **1965**, *60*, 967.
(29) Cho, A. K.; Jenden, D. J. *Int. J. Neuropharmacol.* **1964**, *3*, 27.
(30) Pailer, M.; Hübsch, W. J. *Monatsh. Chem.* **1966**, *97*, 1541.