

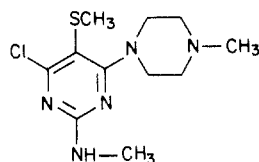
## 4-Amino-6-chloro-2-piperazinopyrimidines with Selective Affinity for $\alpha_2$ -Adrenoceptors

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A series of 4-amino-6-chloro-2-piperazinopyrimidines were synthesized and evaluated for their ability to interact with  $\alpha_1$ - and  $\alpha_2$ -adrenoceptors in vitro in binding assays using [ $^3$ H]WB-4101, [ $^3$ H]clonidine, and [ $^3$ H]idazoxan as radioligands. Some compounds were also tested as inhibitors of [ $^3$ H]spiroperidol binding. Several members of this series showed high and selective affinity for  $\alpha_2$ -adrenoceptors. The nature of the 4-amino substituent seems to be the most critical factor in determining the potency at these receptors.

A previous communication<sup>1</sup> described 2-amino-6-chloro-4-piperazinopyrimidines as inhibitors of spiroperidol binding. One of these derivatives, mezilamine, first reported by Mattioda et al.<sup>2</sup> as an antiemetic compound, was shown to be an atypical dopamine (DA) antagonist, having, in addition, agonistic properties at postsynaptic  $\alpha_1$ - and antagonistic properties at presynaptic  $\alpha_2$ -adrenoceptors.<sup>3</sup>



This result prompted us to synthesize compounds structurally related to mezilamine, in order to search for  $\alpha_2$ -antagonists devoid of substantial affinity for  $\alpha_1$  or DA receptors.

In this paper we describe the synthesis and preliminary evaluation of the 4-amino-6-chloro-2-piperazinopyrimidines 1-27 (Table I).

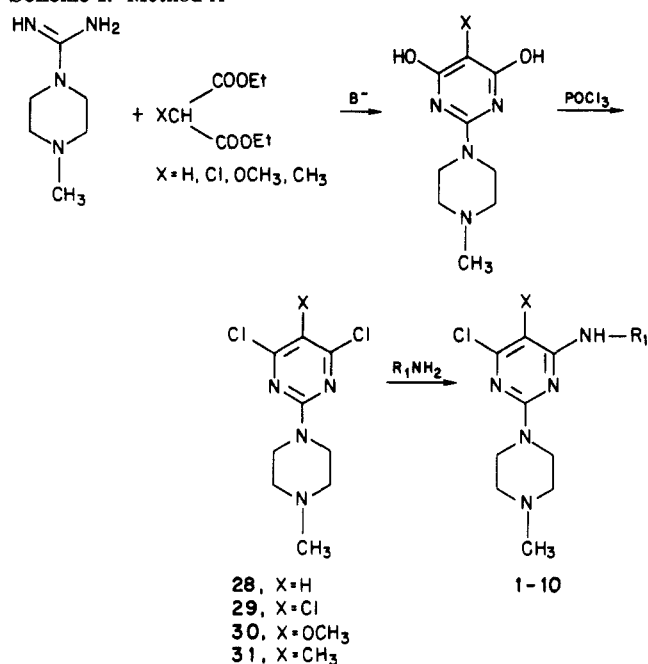
**Chemistry.** Scheme I shows the classical synthetic route<sup>4</sup> used for the preparation of 2-(N-methylpiperazino)pyrimidines 1-10, bearing the H, Cl, OCH<sub>3</sub>, or CH<sub>3</sub> group as a substituent in position 5 (method A). 1-Methyl-4-piperazinecarboxamide was condensed with the appropriate diethyl malonate derivative to give the corresponding 4,6-dihydroxy-2-(N-methylpiperazino)pyrimidine. The crude products were treated directly with POCl<sub>3</sub>, and the resulting dichloro derivatives, 28-31, were converted to the desired compounds, 1-10, by treatment with a primary amine, R<sub>1</sub>-NH<sub>2</sub>.

The pyrimidines 11-20, bearing the methylthio group in position 5, were prepared as outlined in Scheme II (method B).<sup>5</sup> Reaction of 1,4-dimethylpiperazine with 5-(methylthio)-2,4,6-trichloropyrimidine led to the formation of chloromethane and 32.

The assigned structure for 32 was based upon <sup>13</sup>C NMR data, C(4) and C(6) being found to be equivalent. The intermediate 32 was then reacted with the appropriate amine, R<sub>1</sub>-NH<sub>2</sub>, to give 11-20. The N-unsubstituted piperazines, 25 and 26, were obtained by the same method (Scheme II), starting from 5-(methylthio)-2,4,6-trichloropyrimidine or 5-methyl-2,4,6-trichloropyrimidine but using 1-methyl-4-formylpiperazine instead of 1,4-dimethylpiperazine. Condensation of the resulting dichloro derivatives, 33 and 34, with isopropylamine, followed by cleavage of the formyl group in an acidic medium, gave the desired products, 25 and 26.

The bromo derivatives, 21-24, were obtained from their methylthio analogues, 11, 12, 14 and 16, by treatment with BrONa in aqueous acetic acid, as previously described<sup>6</sup>

Scheme I. Method A



(method C). In a similar way 27 was prepared from 6-chloro-2-(N-formylpiperazino)-4-(isopropylamino)-5-(methylthio)pyrimidine, 36, by treatment with BrONa, followed by hydrolytic cleavage of the formyl group.

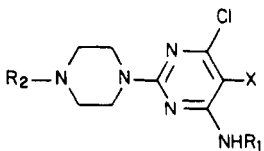
**Receptor Binding Studies in Vitro.** Compounds 1-27 were tested in vitro for  $\alpha_2$ -receptor affinity using a [ $^3$ H]-clonidine binding assay on rat cortical membranes. These compounds were evaluated also for their ability to interact with  $\alpha_1$ -receptors by measuring the inhibition of [ $^3$ H]WB-4101 binding to rat cortical membranes in vitro. We also examined the ability of some of the compounds to interact

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† Pharmuka Laboratoires.

Table I. Physicochemical Data for 4-Amino-6-chloro-2-piperazinopyrimidines

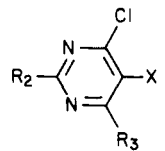


The chemical structure shows a piperazine ring attached at the 2-position to a pyrimidine ring. The pyrimidine ring has a chlorine atom at the 6-position and a substituent X at the 4-position. The piperazine ring has a substituent R<sub>2</sub> on one of its nitrogens. The pyrimidine ring also has an NHR<sub>1</sub> group at the 1-position.

compd	X	R <sub>1</sub>	R <sub>2</sub>	method	formula	mp, °C	recrystd solvent
1	H	CH <sub>3</sub>	CH <sub>3</sub>	A	C <sub>10</sub> H <sub>16</sub> ClN <sub>5</sub> ·2HCl	>260	EtOH
2	H	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	CH <sub>3</sub>	A	C <sub>12</sub> H <sub>20</sub> ClN <sub>5</sub> ·HCl·0.5H <sub>2</sub> O	225 dec	H <sub>2</sub> O
3	Cl	H	CH <sub>3</sub>	A	C <sub>9</sub> H <sub>13</sub> Cl <sub>2</sub> N <sub>5</sub> ·HCl	260	
4	Cl	CH <sub>3</sub>	CH <sub>3</sub>	A	C <sub>10</sub> H <sub>15</sub> Cl <sub>2</sub> N <sub>5</sub> ·HCl	>260	EtOH
5	Cl	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	CH <sub>3</sub>	A	C <sub>12</sub> H <sub>19</sub> Cl <sub>2</sub> N <sub>5</sub> ·HCl	252	
6	Cl	<i>c</i> -C <sub>6</sub> H <sub>5</sub>	CH <sub>3</sub>	A	C <sub>14</sub> H <sub>21</sub> Cl <sub>2</sub> N <sub>5</sub> ·HCl <sup>a</sup>	258	H <sub>2</sub> O
7	OCH <sub>3</sub>	CH <sub>3</sub>	CH <sub>3</sub>	A	C <sub>11</sub> H <sub>18</sub> ClN <sub>5</sub> O·HBr	not det	H <sub>2</sub> O
8	OCH <sub>3</sub>	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	CH <sub>3</sub>	A	C <sub>13</sub> H <sub>22</sub> ClN <sub>5</sub> O·HCl <sup>b</sup>	142	EtOH-H <sub>2</sub> O (1:1)
9	OCH <sub>3</sub>	<i>c</i> -C <sub>6</sub> H <sub>5</sub>	CH <sub>3</sub>	A	C <sub>15</sub> H <sub>24</sub> ClN <sub>5</sub> O·HCl	>260	H <sub>2</sub> O
10	CH <sub>3</sub>	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	CH <sub>3</sub>	A	C <sub>13</sub> H <sub>22</sub> ClN <sub>5</sub> ·2HCl	258	acetone
11	SCH <sub>3</sub>	H	CH <sub>3</sub>	B	C <sub>10</sub> H <sub>16</sub> ClN <sub>5</sub> S <sup>c</sup>	131	EtOH-H <sub>2</sub> O (1:1)
12	SCH <sub>3</sub>	CH <sub>3</sub>	CH <sub>3</sub>	B	C <sub>11</sub> H <sub>18</sub> ClN <sub>5</sub> S·HCl	222	EtOH
13	SCH <sub>3</sub>	C <sub>2</sub> H <sub>5</sub>	CH <sub>3</sub>	B	C <sub>12</sub> H <sub>20</sub> ClN <sub>5</sub> S·HCl	223	AcOEt-EtOH (4:1)
14	SCH <sub>3</sub>	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	CH <sub>3</sub>	B	C <sub>13</sub> H <sub>22</sub> ClN <sub>5</sub> S	68	EtOH-H <sub>2</sub> O (1:1)
15	SCH <sub>3</sub>	<i>t</i> -C <sub>4</sub> H <sub>9</sub>	CH <sub>3</sub>	B	C <sub>14</sub> H <sub>24</sub> ClN <sub>5</sub> S·HCl	246	H <sub>2</sub> O
16	SCH <sub>3</sub>	<i>c</i> -C <sub>6</sub> H <sub>5</sub>	CH <sub>3</sub>	B	C <sub>15</sub> H <sub>24</sub> ClN <sub>5</sub> S·HCl <sup>d</sup>	202	H <sub>2</sub> O
17	SCH <sub>3</sub>	<i>c</i> -C <sub>6</sub> H <sub>11</sub>	CH <sub>3</sub>	B	C <sub>16</sub> H <sub>26</sub> ClN <sub>5</sub> S·HCl	202	H <sub>2</sub> O
18	SCH <sub>3</sub>	C <sub>6</sub> H <sub>5</sub>	CH <sub>3</sub>	B	C <sub>16</sub> H <sub>20</sub> ClN <sub>5</sub> S·HCl	263	H <sub>2</sub> O
19	SCH <sub>3</sub>	CH <sub>2</sub> -C <sub>6</sub> H <sub>5</sub>	CH <sub>3</sub>	B	C <sub>17</sub> H <sub>22</sub> ClN <sub>5</sub> S·HCl <sup>e</sup>	210	AcOEt-EtOH (5:1)
20	SCH <sub>3</sub>	CH <sub>2</sub> -CH <sub>2</sub> -C <sub>6</sub> H <sub>5</sub>	CH <sub>3</sub>	B	C <sub>18</sub> H <sub>24</sub> ClN <sub>5</sub> S·HCl	214	EtOH
21	Br	H	CH <sub>3</sub>	B	C <sub>9</sub> H <sub>13</sub> BrClN <sub>5</sub> ·HCl	>260	EtOH
22	Br	CH <sub>3</sub>	CH <sub>3</sub>	B	C <sub>10</sub> H <sub>15</sub> BrClN <sub>5</sub> ·HCl	265 dec	EtOH-MeOH (2:1)
23	Br	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	CH <sub>3</sub>	B	C <sub>12</sub> H <sub>19</sub> BrClN <sub>5</sub> ·HCl·0.5H <sub>2</sub> O	229	Me <sub>2</sub> CO-H <sub>2</sub> O (1:1)
24	Br	<i>c</i> -C <sub>6</sub> H <sub>5</sub>	CH <sub>3</sub>	B	C <sub>14</sub> H <sub>21</sub> BrClN <sub>5</sub> ·HCl <sup>f</sup>	238	H <sub>2</sub> O
25	CH <sub>3</sub>	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	H	C	C <sub>12</sub> H <sub>20</sub> ClN <sub>5</sub> ·HCl	200	
26	SCH <sub>3</sub>	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	H	C	C <sub>12</sub> H <sub>20</sub> ClN <sub>5</sub> S·HCl	147	H <sub>2</sub> O
27	Br	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	H	C	C <sub>11</sub> H <sub>17</sub> BrN <sub>5</sub> ·HCl	250 dec	EtOH

<sup>a</sup>H: calcd, 6.05; found, 5.42. <sup>b</sup>O: calcd, 4.76; found, 4.33. <sup>c</sup>C: calcd, 43.87; found, 44.21. <sup>d</sup>H: calcd, 6.66; found 6.16. <sup>e</sup>C: calcd, 51.00; found, 51.46. <sup>f</sup>Br: calcd, 19.43; found 18.75.

Table II. Physicochemical Data for Pyrimidine Intermediates



The chemical structure shows a pyrimidine ring with substituents R<sub>2</sub> at the 2-position, R<sub>3</sub> at the 4-position, and a chlorine atom at the 6-position. There is also a substituent X at the 5-position.

compd	X	R <sub>2</sub>	R <sub>3</sub>	formula	mp, °C	recrystd solvent
28	H	<i>N</i> -methylpiperazino	Cl	C <sub>9</sub> H <sub>12</sub> Cl <sub>2</sub> N <sub>4</sub>	80	cyclohexane
29	Cl	<i>N</i> -methylpiperazino	Cl	C <sub>9</sub> H <sub>11</sub> Cl <sub>3</sub> N <sub>4</sub>	115	cyclohexane
30	OCH <sub>3</sub>	<i>N</i> -methylpiperazino	Cl	C <sub>10</sub> H <sub>14</sub> Cl <sub>2</sub> N <sub>4</sub> O <sup>a</sup>	101	cyclohexane
31	CH <sub>3</sub>	<i>N</i> -methylpiperazino	Cl	C <sub>10</sub> H <sub>14</sub> Cl <sub>2</sub> N <sub>4</sub>	68	cyclohexane
32	SCH <sub>3</sub>	<i>N</i> -methylpiperazino	Cl	C <sub>10</sub> H <sub>14</sub> Cl <sub>2</sub> N <sub>4</sub> S	82	EtOH-H <sub>2</sub> O (3:1)
33	CH <sub>3</sub>	<i>N</i> -formylpiperazino	Cl	C <sub>10</sub> H <sub>12</sub> Cl <sub>2</sub> N <sub>4</sub> O	216	toluene
34	SCH <sub>3</sub>	<i>N</i> -formylpiperazino	Cl	C <sub>10</sub> H <sub>12</sub> Cl <sub>2</sub> N <sub>4</sub> OS	136	EtOH
35	CH <sub>3</sub>	<i>N</i> -formylpiperazino	NH- <i>i</i> -C <sub>3</sub> H <sub>7</sub>	C <sub>13</sub> H <sub>20</sub> ClN <sub>5</sub> O	169	
36	SCH <sub>3</sub>	<i>N</i> -formylpiperazino	NH- <i>i</i> -C <sub>3</sub> H <sub>7</sub>	C <sub>13</sub> H <sub>20</sub> ClN <sub>5</sub> OS	92	EtOH-H <sub>2</sub> O (1:1)

<sup>a</sup>C: calcd, 43.32; found, 42.82.

with D<sub>2</sub> DA receptors by measuring the inhibition of [<sup>3</sup>H]spiroperidol binding to rat striatal membranes in vitro. Displacement by 10 compounds of [<sup>3</sup>H]idazoxan (RX-781094, 2-[2-(1,4-benzodioxanyl)]-2-imidazoline), a new selective α<sub>2</sub>-antagonist,<sup>7</sup> is also reported. These receptor binding data for compounds 1–27 and for reference compounds are listed in Table III. Mezilamine<sup>3</sup> and yohimbine<sup>8</sup> were used as the reference α<sub>2</sub>-antagonists and phenoxybenzamine<sup>9</sup> as the reference α<sub>1</sub>-antagonist.

## Results and Discussion

The most potent inhibitors of [<sup>3</sup>H]clonidine binding were the 4-isopropylamino derivatives 5, 10, 14, 23, 25, 26, and 27 and the 4-cyclopentylamino derivative 16, all being more potent than yohimbine. These results suggest that steric hindrance in the vicinity of the nitrogen atom of the 4-amino group could be a critical factor for α<sub>2</sub>-receptor

affinity. The importance of this factor was studied in the 5-methylthio series (compounds 11–20). Introduction of large substituents in the 4-amino group such as *tert*-butyl, cyclohexyl, phenyl, benzyl, or phenethyl significantly reduced the affinity. Moreover, the affinity was also lowered by even small substituents: *i*-C<sub>3</sub>H<sub>7</sub> > C<sub>2</sub>H<sub>5</sub> > CH<sub>3</sub> > H. *N*-Methylation of the piperazine nitrogen did not substantially affect the affinity.

The results obtained from the [<sup>3</sup>H]clonidine binding assay were confirmed for 10 of the compounds (Table III) by using [<sup>3</sup>H]idazoxan as the radioligand. There was a good correlation between the *K*<sub>i</sub> values for displacement of these two α<sub>2</sub> ligands, according to the following equation:

$$\log K_i (\text{idazoxan}) = 0.426 + 0.859 \log K_i (\text{clonidine})$$

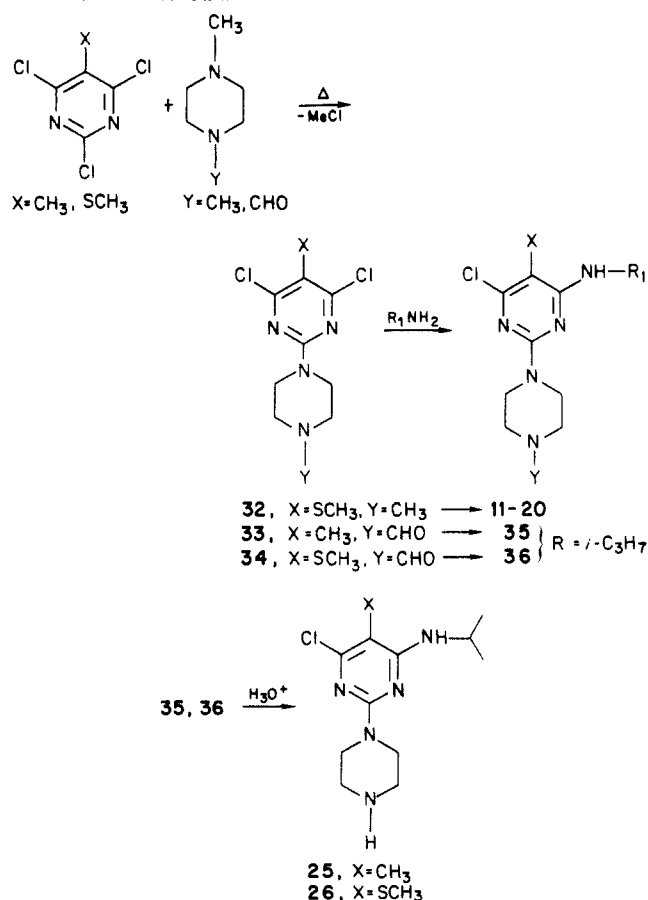
$$n = 10, r = 0.93, F_{1,8} = 55.8 (F_{1,8}/0.01 = 11.26)$$

Within this series, most of the compounds were found to

**Table III.** Receptor Binding Data for 4-Amino-6-chloro-2-piperazinopyrimidines

compd	X	R <sub>1</sub>	R <sub>2</sub>	inhibition of binding: K <sub>i</sub> , nM <sup>a</sup>			
				spiroperidol	WB-4101	clonidine	idazoxan
1	H	CH <sub>3</sub>	CH <sub>3</sub>	630	280	50	100
2	H	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	CH <sub>3</sub>		>180	130	200
3	Cl	H	CH <sub>3</sub>		67	50	
4	Cl	CH <sub>3</sub>	CH <sub>3</sub>	1 200	4	44	
5	Cl	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	CH <sub>3</sub>	2 400	42	5	14
6	Cl	<i>c</i> -C <sub>6</sub> H <sub>5</sub>	CH <sub>3</sub>		240	120	97
7	OCH <sub>3</sub>	CH <sub>3</sub>	CH <sub>3</sub>		1700	150	
8	OCH <sub>3</sub>	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	CH <sub>3</sub>		310	50	
9	OCH <sub>3</sub>	<i>c</i> -C <sub>6</sub> H <sub>5</sub>	CH <sub>3</sub>	670	1700	50	29
10	CH <sub>3</sub>	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	CH <sub>3</sub>	880	200	16	
11	SCH <sub>3</sub>	H	CH <sub>3</sub>	1 000	820	620	
12	SCH <sub>3</sub>	CH <sub>3</sub>	CH <sub>3</sub>		1800	360	
13	SCH <sub>3</sub>	C <sub>2</sub> H <sub>5</sub>	CH <sub>3</sub>		1800	90	
14	SCH <sub>3</sub>	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	CH <sub>3</sub>	640	360	26	
15	SCH <sub>3</sub>	<i>t</i> -C <sub>4</sub> H <sub>9</sub>	CH <sub>3</sub>	920	450	180	
16	SCH <sub>3</sub>	<i>c</i> -C <sub>6</sub> H <sub>5</sub>	CH <sub>3</sub>	1 200	76	22	
17	SCH <sub>3</sub>	<i>c</i> -C <sub>6</sub> H <sub>11</sub>	CH <sub>3</sub>		240	290	
18	SCH <sub>3</sub>	C <sub>6</sub> H <sub>5</sub>	CH <sub>3</sub>		140	240	500
19	SCH <sub>3</sub>	CH <sub>2</sub> -C <sub>6</sub> H <sub>5</sub>	CH <sub>3</sub>	14	100	1500	
20	SCH <sub>3</sub>	CH <sub>2</sub> -CH <sub>2</sub> -C <sub>6</sub> H <sub>5</sub>	CH <sub>3</sub>	1 400	180	910	
21	Br	H	CH <sub>3</sub>	10 000	820	1600	
22	Br	CH <sub>3</sub>	CH <sub>3</sub>	200	180	150	
23	Br	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	CH <sub>3</sub>	480	290	7	
24	Br	<i>c</i> -C <sub>6</sub> H <sub>5</sub>	CH <sub>3</sub>		250	140	230
25	CH <sub>3</sub>	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	H	2 900	860	6	13
26	SCH <sub>3</sub>	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	H	3 000	500	36	62
27	Br	<i>i</i> -C <sub>3</sub> H <sub>7</sub>	H	880	200	16	28
clonidine				>4 000	310	1.5	
yohimbine				2 000	360	45	48
phenoxybenzamine				120	1.8		
mezilamine				40	91	9	12

<sup>a</sup> K<sub>i</sub> values are the mean of triplicate determinations using four different membrane preparations. Standard deviations were always smaller than 15%.

**Scheme II. Method B**

have weak affinity for  $\alpha_1$ -adrenoceptors compared to phenoxybenzamine, but no obvious structure-activity re-

lationship emerges from the binding data concerning the  $\alpha_1/\alpha_2$  selectivity.

Among the compounds tested in the [<sup>3</sup>H]spiroperidol assay, only the 4-benzylamino derivative, **19**, was found to have high affinity for D<sub>2</sub> receptors. It is interesting to note that in the 2-amino-4-(*N*-methylpiperazino)pyrimidine series,<sup>1</sup> the position isomer of **19** also exhibited a high degree of affinity for the DA receptor.

In conclusion, the results obtained in the present study demonstrate that structural modification of mezilamine, a DA antagonist having agonist properties at  $\alpha_1$ - and antagonist properties at  $\alpha_2$ -receptors, can lead to compounds with selective affinity for  $\alpha_2$ -adrenoceptors.

The most selective compound was **25** ( $\alpha_2/\alpha_1 = 143$ ). This compound has been selected for in-depth pharmacological studies; it has been found to be a more potent and more selective  $\alpha_2$ -adrenoceptor antagonist than yohimbine in vivo.<sup>10</sup> Details of the pharmacology of **25** will be published elsewhere.

**Experimental Section**

**Chemistry.** Melting points were determined on a Kofler hot stage. NMR spectra were recorded on a Varian T-60 or JEOL-90 spectrometer; 70–230-mesh silica gel was used for column chromatography. The purity of compounds was checked by TLC analysis on silica gel GF plates, and components were visualized by UV fluorescence properties. All compounds exhibited proper spectral characteristics and were homogeneous by TLC analysis. Microanalysis results on new compounds are within  $\pm 0.4\%$  of the theoretical values unless otherwise indicated.

**Method A. 6-Chloro-4-(methylamino)-2-(*N*-methylpiperazino)pyrimidine Dihydrochloride (1).** **Step A. 4,6-Dichloro-2-(*N*-methylpiperazino)pyrimidine (28).** 1-Methyl-4-piperazinecarboxamide hydrogen sulfate<sup>11</sup> (47.7 g, 0.25 mol) and a solution of MeONa obtained by dissolving Na (11.5 g, 0.5 mol) in MeOH (270 mL) were refluxed for 0.5 h. Diethyl malonate (40 g, 0.25 mol) was added and reflux maintained for 5 h. The mixture was cooled, poured into H<sub>2</sub>O (500 mL), made

neutral with AcOH (45 mL), and evaporated in vacuo. The residue was stirred with warm  $\text{CHCl}_3$  (1 L), and the resultant solid was filtered off: crude 4,6-dihydroxy-2-(*N*-methylpiperazino)pyrimidine (70 g) was thus obtained, which was converted into its dichloro derivative by refluxing with  $\text{POCl}_3$  (180 mL) for 3 h. The mixture was poured into ice (1 kg), made alkaline with 10 N NaOH (700 mL), extracted with  $\text{CHCl}_3$  ( $3 \times 600$  mL), and evaporated under reduced pressure. The residue was chromatographed on silica gel by eluting with toluene-diethylamine (9:1) to give 28 (8.4 g, 13.6%). After crystallization from cyclohexane an analytical sample melted at 80 °C. Anal. ( $\text{C}_9\text{H}_{12}\text{Cl}_2\text{N}_4$ ) C, H, Cl, N.

**Step B. 6-Chloro-4-(methylamino)-2-(*N*-methylpiperazino)pyrimidine Dihydrochloride (1).** The product from step A (28) (7 g, 0.028 mol), toluene (140 mL), pulverized  $\text{K}_2\text{CO}_3$  (2.1 g, 0.015 mol), and a 33% solution of methylamine in EtOH (30 mL, 0.035 mol) were refluxed for 32 h. The mixture was filtered and evaporated in vacuo. The residue was purified by chromatography on silica gel by eluting with toluene-diethylamine (9:1). It was converted into the dihydrochloride and recrystallization from EtOH gave 1 (4.85 g, 55%), mp >260 °C. Anal. ( $\text{C}_{10}\text{H}_{16}\text{ClN}_5 \cdot 2\text{HCl}$ ) C, H, Cl, N.

**Method B. 4-Amino-6-chloro-2-(*N*-methylpiperazino)-5-(methylthio)pyrimidine Hydrochloride (11).** **Step A. 4,6-Dichloro-2-(*N*-methylpiperazino)-5-(methylthio)pyrimidine (32).** To a solution of 1,4-dimethylpiperazine (38 g, 0.33 mol) in toluene (300 mL) was added dropwise, at ca. 100 °C, 5-(methylthio)-2,4,6-trichloropyrimidine<sup>12</sup> (69 g, 0.30 mol) in toluene (300 mL), and the mixture was stirred at 90 °C for 2 h. After the mixture was cooled to room temperature, it was filtered and concentrated under reduced pressure. Chromatography on silica gel by eluting with toluene-MeOH (87:13) afforded 32 (69 g, 78.5%). After crystallization from EtOH-H<sub>2</sub>O (3:1) an analytical sample melted at 82 °C. <sup>13</sup>C NMR  $\delta$  158.5 ( $\text{C}_2$ ), 166.6 ( $\text{C}_4$ ,  $\text{C}_6$ ). Anal. ( $\text{C}_{10}\text{H}_{14}\text{Cl}_2\text{N}_4\text{S}$ ) C, H, Cl, N, S.

**Step B. 4-Amino-6-chloro-2-(*N*-methylpiperazino)-5-(methylthio)pyrimidine (11).** The product from step A (32) (30 g, 0.102 mol) and MeOH (330 mL) containing 60 g of  $\text{Na}_2\text{CO}_3$  were heated at 130 °C in an autoclave for 3 h. The mixture was evaporated in vacuo, and the residue was purified by chromatography on silica gel by eluting with  $\text{CHCl}_3$ -diethylamine (98:2) to give 11 (10 g, 35.8%). After crystallization from EtOH-H<sub>2</sub>O (1:1) an analytical sample melted at 131 °C. Anal. ( $\text{C}_{10}\text{H}_{16}\text{ClN}_5\text{S}$ ) C, H, N.

**Method B. 6-Chloro-4-(isopropylamino)-5-(methylthio)-2-piperazinopyrimidine Hydrochloride (26).** **Step A. 4,6-Dichloro-2-(*N*-formylpiperazino)-5-(methylthio)pyrimidine (34).** To a solution of 1-formyl-4-methylpiperazine (42.3 g, 0.33 mol) in toluene (300 mL) was added dropwise, at ca. 100 °C, 5-(methylthio)-2,4,6-trichloropyrimidine (69 g, 0.30 mol) in toluene (300 mL), and the mixture was stirred at 90 °C for 2 h. After the mixture was cooled to room temperature, it was filtered and evaporated in vacuo to dryness to give 34 (89.1 g, 96.7%), mp 136 °C. <sup>13</sup>C NMR  $\delta$  158.4 ( $\text{C}_2$ ), 166.8 ( $\text{C}_4$ ,  $\text{C}_6$ ). Anal. ( $\text{C}_{10}\text{H}_{12}\text{Cl}_2\text{N}_4\text{OS}$ ) C, H, Cl, N.

**Step B. 6-Chloro-2-(*N*-formylpiperazino)-4-(isopropylamino)-5-(methylthio)pyrimidine (36).** Compound 34 (77 g, 0.25 mol), toluene (500 mL), triethylamine (31 g, 0.3 mol), and isopropylamine (17.7 g, 0.3 mol) were heated at 100 °C for 4 h. The warm mixture was filtered, and the filtrate was washed with H<sub>2</sub>O and concentrated under reduced pressure. Chromatography of the residue on silica gel by eluting with toluene-diethylamine (95:5) afforded a product, two crystallizations of which from EtOH-H<sub>2</sub>O (1:1) yielded 36 (34.2 g, 41.5%), mp 92 °C. Anal. ( $\text{C}_{13}\text{H}_{20}\text{ClN}_5\text{OS}$ ) C, H, Cl, N, O, S.

**Step C. 6-Chloro-4-(isopropylamino)-5-(methylthio)-2-piperazinopyrimidine Hydrochloride (26).** Compound 36 (32.95 g, 0.10 mol), H<sub>2</sub>O (600 mL), and concentrated HCl (60 mL) were refluxed for 2 h, then 5 g of charcoal was added and the cooled mixture was filtered. The filtrate was made alkaline with 10 N NaOH (100 mL) and extracted with  $\text{CHCl}_3$  ( $2 \times 350$  mL). The organic layer was evaporated in vacuo. The residue was dissolved in EtOH and converted to the hydrochloride. Three crystallizations from H<sub>2</sub>O gave 26 (12.55 g, 37.1%), mp 147 °C. Anal. ( $\text{C}_{12}\text{H}_{20}\text{ClN}_5\text{S} \cdot \text{HCl}$ ) C, H, Cl, N, S.

**Method C. 4-Amino-5-bromo-6-chloro-2-(*N*-methylpiperazino)pyrimidine Hydrochloride (21).** To a solution of

11 (8.1 g, 0.0296 mol) in AcOH (35 mL) was added over 1 h a 1.2 N solution of sodium hypobromite (240 mL). After the mixture stood for 1 h at room temperature, it was cooled at 10 °C, made alkaline with 10 N NaOH (50 mL), and extracted with  $\text{CHCl}_3$  ( $2 \times 100$  mL); the organic layer was washed with H<sub>2</sub>O and concentrated under reduced pressure. Chromatography on silica gel by eluting with  $\text{CHCl}_3$ -diethylamine (92:8) yielded 2.8 g, which was converted into the hydrochloride in EtOH. Recrystallization from EtOH afforded 21 (2.1 g, 20.7%), mp >260 °C. Anal. ( $\text{C}_9\text{H}_{13}\text{BrClN}_5\text{HCl}$ ) C, H, Br, Cl, N.

**Method C. 5-Bromo-6-chloro-4-(isopropylamino)-2-piperazinopyrimidine Hydrochloride (27).** This compound was prepared as 21 but starting from 36 instead of 11. The resulting product was then hydrolyzed according to the method described for 26. From 36 (33 g, 0.1 mol) 5-bromo-6-chloro-4-(isopropylamino)-2-(*N*-formylpiperazino)pyrimidine (21.4 g) was obtained. Hydrolysis gave 27 after two recrystallizations from H<sub>2</sub>O (5.35 g, 14.4%), mp 250 °C dec. Anal. ( $\text{C}_{11}\text{H}_{17}\text{BrClN}_5\text{HCl}$ ) C, H, Br, Cl, N.

**Biological Methods.** The affinities of selected compounds for central  $\alpha$ -adrenergic ( $\alpha_1$  and  $\alpha_2$ ) and dopaminergic ( $\text{D}_2$ ) receptors were determined by using methods described previously.<sup>13-15</sup> Rat brain cortex ( $\alpha_1$  or  $\alpha_2$ ) or rat brain striatum ( $\text{D}_2$ ) was dispersed, using an ultraturax homogenizer, and the membranes were suspended in 20 vol of ice-cold 50 mM Tris-HCl buffer (pH 7.4). The homogenates were centrifuged at 50 000g for 10 min, and the pellets were washed twice with the same volume of buffer. The final pellets were resuspended either in the homogenization buffer ( $\alpha_1$  or  $\alpha_2$ ) or in supplemented 50 mM Tris-HCl buffer<sup>16</sup> ( $\text{D}_2$ ) at a final protein concentration of 2 ng/mL, for the binding assays.

The radioligands [<sup>3</sup>H]spiroperidol (29.5 Ci/mmol), [<sup>3</sup>H]clonidine (66.8 Ci/mmol), and [<sup>3</sup>H]WB-4101 (19 Ci/mmol) were obtained from New England Nuclear and [<sup>3</sup>H]idazoxan (40 Ci/mmol) from Commissariat à l'Energie Atomique. Radioligand concentrations were 0.3 nM [<sup>3</sup>H]spiroperidol ( $\text{D}_2$ ), 1 nM [<sup>3</sup>H]WB-4101 ( $\alpha_1$ ), 1 nM [<sup>3</sup>H]clonidine, or 3 nM [<sup>3</sup>H]idazoxan ( $\alpha_2$ ). Nonspecific binding was determined as that binding occurring in the presence of 10<sup>-6</sup> M butaclamol ( $\text{D}_2$ ), 10<sup>-4</sup> M noradrenaline ( $\alpha_1$ ), and 2.10<sup>-6</sup> M yohimbine ( $\alpha_2$ ). The reaction was started by addition of the membrane suspension (1 ng of protein for  $\alpha_1$  and  $\alpha_2$ , 0.2 ng for  $\text{D}_2$ ). The final incubation volume was 5 mL.

After incubation for 15 min at 25 °C, the membranes were harvested by vacuum filtration through GF/B glass fiber filters, which were then washed twice with 5 mL of buffer. Radioactivity was measured by liquid scintillation spectrometry.

$\text{IC}_{50}$  for these compounds were obtained from five-point displacement curves.  $K_i$  values were then calculated by using the formula

$$K_i = \frac{\text{IC}_{50}}{1 + [\text{C}]/K_D}$$

where [C] was the concentration of the radioligand. The equilibrium dissociation constant,  $K_D$ , had been determined previously from Scatchard plots as 0.3 nM for [<sup>3</sup>H]spiroperidol, 0.22 nM for [<sup>3</sup>H]WB-4101, 0.57 nM for [<sup>3</sup>H]clonidine, and 4.9 nM for [<sup>3</sup>H]idazoxan.

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**Registry No.** 1, 84727-45-7; 1·2HCl, 84727-46-8; 2, 102396-27-0; 2·HCl, 84727-47-9; 3, 84727-50-4; 3·HCl, 84727-51-5; 4, 84727-52-6; 4·HCl, 84727-53-7; 5, 84727-54-8; 5·HCl, 84727-55-9; 6, 84727-56-0; 6·HCl, 84727-57-1; 7, 102396-28-1; 7·HBr, 102396-40-7; 8, 84727-

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58-2; 8-HCl, 84727-59-3; 9, 84727-62-8; 9-HCl, 84727-63-9; 10, 84727-70-8; 10-2HCl, 84727-71-9; 11, 84727-84-4; 12, 102396-30-5; 12-HCl, 102396-41-8; 13, 102396-31-6; 13-HCl, 102396-42-9; 14, 61973-58-8; 15, 84727-88-8; 15-HCl, 84707-98-2; 16, 84708-05-4; 16-HCl, 84708-06-5; 17, 84708-07-6; 17-HCl, 84708-08-7; 18, 83199-77-3; 18-HCl, 83199-78-4; 19, 102396-32-7; 19-HCl, 102396-43-0; 20, 102396-33-8; 20-HCl, 102396-44-1; 21, 102396-34-9; 21-HCl, 102396-45-2; 22, 102396-35-0; 22-HCl, 102396-46-3; 23, 102396-36-1; 23-HCl, 61973-50-0; 24, 102396-37-2; 24-HCl, 102396-47-4; 25, 102396-29-2; 25-HCl, 84727-77-5; 26, 84708-09-8; 26-HCl, 84727-80-0; 27, 102396-39-4; 27-HCl, 102396-48-5; 28,

84727-44-6; 29, 84727-49-1; 30, 84727-61-7; 31, 84727-69-5; 32, 84727-83-3; 33, 84727-78-6; 34, 84727-81-1; 35, 84727-79-7; 36, 84727-82-2; 36 (X = Br), 102396-38-3; 5-methyl-2,4,6-trichloropyrimidine, 1780-36-5; 5-methylthio-2,4,6-trichloropyrimidine, 24795-76-4; 1,4-dimethylpiperazine, 106-58-1; 4-methyl-1-piperazinecarboxaldehyde, 7556-55-0; 4,6-dihydroxy-2-(N-methylpiperazino)pyrimidine, 81746-24-9; 5-chloro-4,6-dihydroxy-2-(N-methylpiperazino)pyrimidine, 84727-48-0; 4,6-dihydroxy-5-methoxy-2-(N-methylpiperazino)pyrimidine, 84727-60-6; 4,6-dihydroxy-5-methyl-2-(N-methylpiperazino)pyrimidine, 84727-68-4; 1-methyl-4-piperazinecarboxamide, 45798-01-4.

## Synthesis and Class III Antiarrhythmic Activity of (Phenylbut-2-enyl)ammonium Salts. Effect of Conformation on Activity

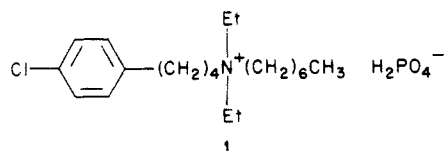
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The syntheses of seven 4-(substituted phenyl)but-2-en(or yn)yl quaternary ammonium salts and four related tertiary amines are described. The Meerwein arylation reaction was the preferred synthetic method for the required intermediate 1-aryl-4-halo-2-butenes (15a-c, 18). In the case of 18, the trans stereochemistry of the Meerwein adduct of 2,3-dimethylbutadiene was established unambiguously by 2D NMR and X-ray studies. The title compounds represent conformationally restricted analogues of the class III antiarrhythmic agent clofilium (1) and exhibit comparable potency and efficacy in the in vitro evaluation using isolated canine Purkinje fibers. These results suggest that the alkylene chain in 1 is extended in the active conformation. Computer-aided conformational analysis (MM2) supports this conclusion. Selective catalytic hydrogen conditions were developed for the conversion of the unsaturated analogue 2 to clofilium (1) with minimal hydrogenolysis of the allylic quaternary ammonium moiety, thus completing a novel and efficient synthesis of this substance.

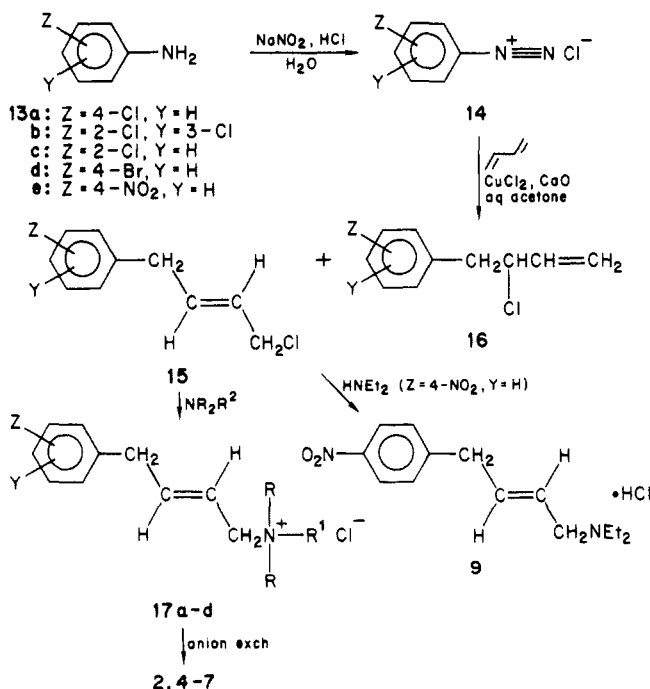
Sudden cardiac death is a major public health problem in the United States today.<sup>1</sup> Ventricular arrhythmias that progress to ventricular tachycardia and fibrillation are the most common cause of sudden cardiac death. We have been interested in the development of pure class III antiarrhythmic agents that are expected to be effective in ventricular arrhythmias caused by reentry mechanisms. According to the designation of antiarrhythmic agents defined by Vaughan Williams, a class III compound prolongs the action potential duration of the cardiac cell without depressing conduction in cardiac tissue.<sup>2</sup> Relatively few compounds of this type have been reported to date, and most, such as sotalol and amiodarone, possess other activities as well.<sup>3</sup>

One compound that appears to exhibit pure class III activity is clofilium (1).<sup>4</sup> In this study the effect on class



III activity of introducing conformational restrictions between the phenyl ring and the quaternary nitrogen atom of the clofilium molecule was examined. The fully saturated four-carbon connecting chain of 1 allows the molecule to assume a variety of spatial orientations of the aromatic ring and the ammonium group including, in principle, a

**Scheme I.** Synthesis of Ammonium Salts and Amines Employing the Meerwein Arylation Reaction of Butadiene



folded conformation in which the quaternary nitrogen interacts with the electron-rich  $\pi$  cloud of the aromatic

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