Table I. Antiprotozoal Activity of 1-(3-Chloro-2-hydroxypropyl)nitroimidazoles and Derivatives

| Compd no. | *T. vaginalis* μg/ml sc | *T. vaginalis* mg/kg po | *T. foetus*, mg/kg po | *E. histolytica*, mg/kg po | $LD_{50}$ (mouse) mg/kg ip | $LD_{50}$ (mouse) mg/kg po |
|---|---|---|---|---|---|---|
| **2** | >1000 | >250 | 85 | >300 | >500 | >500 |
| **3** | 86 | 37 | 3 | 10 | >2000 | >2000 |
| **4** | 211 | 75 | 13 | 433 | 707 | >2000 |
| **7** | 21 | 76 | 46 | 29 | 1000 | 1414 |
| **8** | 37 | 21 | 9 | 137 | 354 | 595 |
| **9** | >1000 | >250 | >250 | >300 | 354 | 901 |
| **10** | >1000 | >250 | >250 | >300 | 420 | 707 |
| **11** | 0.4 | 17 | 6 | 25 | 158 | 330 |
| Metronidazole | 100 | 16 | 17 | 49 | >2000 | >2000 |

the various protozoal infections. Compound **3**, previously reported,[11] showed the same spectrum of activity as metronidazole and was as well tolerated. Compounds **4**, **7**, **8**, and **11**, while showing the same spectrum of activity as compound **3** and metronidazole, were overall more toxic. Compounds **2**, **9**, and **10** were without antiprotozoal activity except that **2** exerted an effect against *T. foetus*.

## Experimental Section‡

**1-(3-Chloro-2-hydroxypropyl)-2-methyl-4-nitroimidazole (2).** A mixture of 15 g (0.12 mol) of 2-methyl-4- (or 5-) nitroimidazole (**1**) and 0.5 g of anhydrous $K_2CO_3$ in 100 ml of epichlorohydrin was stirred and refluxed for 10–15 min and cooled. The crystals were collected and recrystallized from EtOH to give 12.5–15 g (50–60%) of **2**: mp 151°; uv $\lambda_{max}$ 300 nm (ε 7120); $pK_a = 0.3 \pm 0.1$. *Anal.* ($C_7H_{10}ClN_3O_3$) C, H, Cl, N.

**1-(3-Chloro-2-hydroxypropyl)-2-methyl-5-nitroimidazole (3).** To a solution of 127 g (1.0 mol) of **1** in 1.17 l. of 85% $HCO_2H$ at 5–10° was added 460 ml of epichlorohydrin over 5 hr. The mixture was stored at 5–10° for 48 hr and 20–25° for 24 hr, and the volatiles were evaporated under reduced pressure. To the residual oil 200 g of ice–water was added and the solution adjusted to pH 7–7.5 with concentrated $NH_4OH$. After addition of 100 g of $(NH_4)_2SO_4$, the mixture was repeatedly extracted with $C_6H_6$. The crystals that formed in the aqueous layer were filtered to give 25 g of **1**. The benzene layers were extracted with 10 *N* $H_2SO_4$ and saturated with $(NH_4)_2SO_4$ and the resulting aqueous layer was then neutralized with $NH_4OH$ to give an oil which slowly crystallized upon seeding. Recrystallization from toluene gave 92 g (42%) of **3**: mp 78°; uv $\lambda_{max}$ 228 nm (ε 3720), 312 (9150); $pK_a = 2.4 \pm 0.1$. *Anal.* ($C_7H_{10}ClN_3O_3$) C, H, Cl, N.

**1-(2,3-Dichloropropyl)-2-methyl-5-nitroimidazole (4).** Addition of 5 g (0.022 mol) of **3** to 2 g (0.013 mol) of $POCl_3$ and 6 g (0.024 mol) of $PCl_5$ generated heat and HCl and the mixture liquified. The reaction was cooled; ice–water was slowly added and neutralized with aqueous NaOH. The resulting precipitate was filtered, dried, and crystallized from EtOH to give 4.1 g (75%) of **4**: mp 123–124°. *Anal.* ($C_7H_9Cl_2N_3O_2$) C, H, Cl, N.

Alternatively, to a solution of 20 g (0.12 mol) of 1-allyl-2-methyl-5-nitroimidazole[7] (**5**) in concentrated HCl at 50–70° was slowly added 30% $H_2O_2$. The reaction mixture was diluted with $H_2O$, charcoaled, and neutralized to give 15 g of **4**, identical in mixture melting point with **4** obtained from **3**.

**1-(3-Chloroacetonyl)-2-methyl-5-nitroimidazole (6).** To a stirred solution of 12 g (0.055 mol) of **3** in 60 ml of 10 *N* $H_2SO_4$ was added dropwise a warm solution of 15 g (0.057 mol) of $Na_2Cr_2O_7$ in 50 ml of $H_2O$. The reaction mixture was extracted with EtOAc, the extracts were evaporated, and the residue was crystallized from EtOH to give 5 g (42%) of **6**: mp 111°; uv $\lambda_{max}$ 228 nm (ε 3350), 311 (9000); $pK_a = 1.9 \pm 0.1$. *Anal.* ($C_7H_8ClN_3O_3$) C, H, Cl.

**1-(2,3-Epoxypropyl)-2-methyl-5-nitroimidazole (7).** A solution of 5 g (0.22 mol) of **3** in 3 *N* NaOH was heated at 60–70° for 10–15 min and cooled, and the crystals were collected and recrystallized from EtOAc to give 3.4–3.6 g (80–85%) of **7**: mp 111°. *Anal.* ($C_7H_9N_3O_3$) C, H, N.

**1-(3-Chloro-2-hydroxypropyl)-2-isopropyl-5-nitroimidazole (8).** In a manner similar to the procedure given for **3**, 15.5 g (0.1 mol) of 2-isopropyl-4- (or 5-) nitroimidazole[12] and 50 g of epichlorohydrin in 140 ml of 85% $HCO_2H$ afforded 12.4 g (50%) of **8**: mp 103° (from $H_2O$); uv $\lambda_{max}$ 230 nm (ε 3400), 314 (9180); $pK_a = 2.1 \pm 0.1$. *Anal.* ($C_9H_{14}ClN_3O_3$) C, H, Cl, N.

**1-(3-Chloro-2-hydroxypropyl)-5-iodo-2-methyl-4-nitroimidazole (9) and 1-(3-Chloro-2-hydroxypropyl)-4-iodo-2-methyl-5-nitroimidazole (10).** A mixture of 4- (or 5-) iodo-2-methyl-5- (or 4-) nitroimidazole[6] (101 g, 0.4 mol) and 80 ml of epichlorohydrin in 100 ml of EtOH was refluxed for 12 hr, cooled, and diluted with 1.5 l. of $H_2O$. The semicrystalline precipitate was collected, slurried with a mixture of EtOAc and $Et_2O$, filtered, and dried to give 44 g (32%) of **9**: mp 142°; uv $\lambda_{max}$ ($H_2O$) 307 nm (ε 7400); $pK_a = 0.7 \pm 0.1$. *Anal.* ($C_7H_9ClIN_3O_3$) C, H, N.

The above aqueous mother liquor was extracted with EtOAc and the combined organic layers were evaporated. The residual syrup was dissolved in 100 ml of MeOH and addition of 100 ml of concentrated HCl precipitated 12 g of **9** as the hydrochloride. The filtrate was neutralized with $NH_4OH$, and the crystals were filtered and recrystallized from EtOH to give 22 g (16%) of **10**: mp 136–137°; uv $\lambda_{max}$ ($H_2O$) 272 nm (ε 4320), 340 (7800); $pK_a = 0.8 \pm 0.1$. *Anal.* ($C_7H_9ClIN_3O_3$) C, H, N.

**1-(3-Chloro-2-hydroxypropyl)-2-nitroimidazole (11).** By the procedure given for the preparation of **10** and **11**, 11.3 g (0.1 mol) of azomycin[8] and epichlorohydrin in ethanol gave 13.3 g (65%) of **11**: mp 158° (EtOH).§ *Anal.* ($C_6H_8ClN_3O_3$) C, H, Cl, N.

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‡All new compounds gave acceptable analytical data. The ultraviolet spectra were measured in 2-propanol unless otherwise noted.

§First prepared in these laboratories by Drs. A. G. Beaman and R. Duschinsky.

# Conformationally Restricted Analogs of Histamine $H_1$ Receptor Antagonists. 2-Phenyl- and 2-Benzyl-1,2,3,4-tetrahydro-4-dimethylaminoisoquinoline

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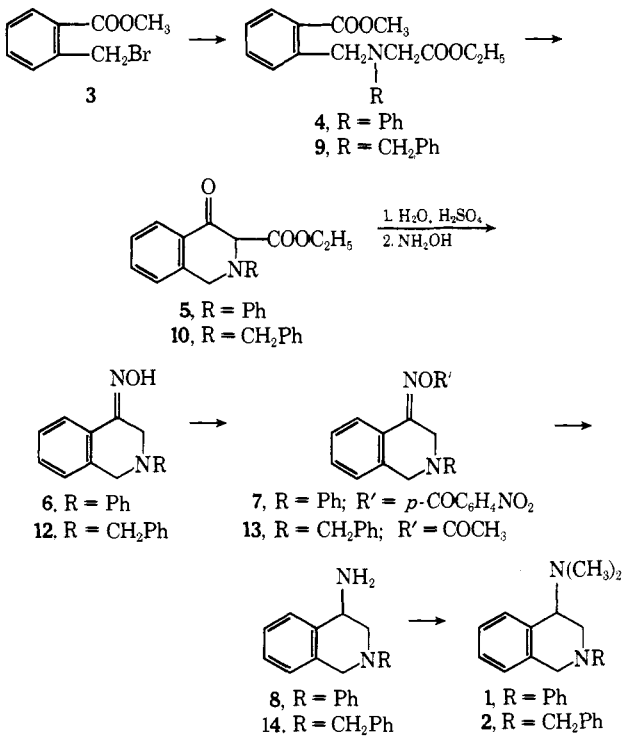
Previous reports from this laboratory have described the use of conformationally restricted analogs of ethylenediamine histamine antagonists as model compounds for as-

sessing the importance of steric and spatial relationships to histamine antagonist activity.[1,2] Our previous results indicated that a trans N-C-C-N conformation is not essential for $H_1$ receptor blockade by ethylenediamine antihistamines and evidence was presented in support of the conformational model of Casy and Ison.[3] In an attempt to further define the conformational aspects of the interaction of antagonists with $H_1$ receptors, the semirigid tetrahydroisoquinoline analogs 1 and 2 were prepared. Compounds 1 and 2 contain the essential structural features of the ethylenediamine histamine antagonists[4] and, at the same time, spatial restrictions are imposed upon the aromatic nuclei and the dimethylamino group. The 2-phenyl group of 1 and the 2-benzyl group of 2 are capable of assuming a position 5–6 Å from the dimethylamino group; this distance has been observed to be favorable for antagonist activity.[4] On the basis of previously established structure–activity relationships, the 2-phenyl substituent should be more favorable to antagonist activity than the 2-benzyl group.[4]

**Synthesis.** The approach to the preparation of the desired tetrahydroisoquinolines was fashioned after that of Grethe and coworkers who described the preparation of several 2,3-dihydro-4(1*H*)-isoquinolones.[5] The benzyl bromide 3 (Scheme I) was prepared by treatment of methyl *o*-toluate with *N*-bromosuccinimide. Compound 3 was then allowed to react with either ethyl *N*-phenylglycinate or ethyl *N*-benzylglycinate to afford 4 or 9, respectively. Dieckmann cyclization of 4 was accomplished in refluxing benzene in the presence of sodium hydride; however, the *N*-benzyl derivative 9 could not be cyclized under these conditions and sodium ethoxide was employed for its conversion to 10. Hydrolysis and decarboxylation of 5 and 10 was carried out in sulfuric acid solution to yield the expected isoquinolones. Because the isoquinolones were somewhat unstable, they were transformed directly to their oxime derivatives 6 and 12. The primary amines 8 and 14 were prepared by diborane reduction of the oxime esters 7 and 13 according to previously reported procedures.[6-8] The primary amines, particularly 8, appeared to undergo decomposition upon standing and therefore were used without further purification. The sodium cyanoborohydride–formaldehyde method of Borch and Hassid was used to convert 8 and 14 to the desired dimethylamino derivatives 1 and 2.[9]

Scheme I




**Biological Results.** Compounds 1 and 2 were evaluated for histamine antagonist activity on the isolated guinea pig ileum and both were found to exhibit a low order of activity.[10] In addition, neither exhibited a significant degree of specificity for the histamine $H_1$ receptor. At a concentration of $10^{-5}$ *M*, 1 and 2 caused slight inhibition of the contractions induced by $4 \times 10^{-6}$ *M* histamine. At a higher concentration ($10^{-4}$ *M*) compounds 1 and 2 blocked the effects of histamine and acetylcholine almost completely. By comparison, the potent ethylenediamine antihistamine tripelennamine ($10^{-7}$ *M*) caused a 66% inhibition of histamine-induced contractions and had no effect on the action of acetylcholine. In the presence of $4 \times 10^{-6}$ *M* histamine compounds 1 and 2 had $ED_{50}$ values of $3.287\ (\pm 0.058) \times 10^{-5}$ *M* and $1.672\ (\pm 0.067) \times 10^{-5}$ *M*, respectively. Antagonism was readily reversed by washing the tissue.

Although the weak activity and lack of specificity of 1 and 2 indicate that these dimethylaminoisoquinolines are not promising structures for studies of $H_1$ receptor antagonism, it is of interest to analyze their activity in light of recently published structure–activity relationships.[2,3,11] Compounds 1 and 2 have approximately the same order of potency as L-(*S*)- and D-(*R*)-3-ethylamino-1-phenylpyrrolidine.[11] The latter analogs contain a single phenyl ring which is located 5 Å from the aliphatic amino function. Addition of a phenyl ring to the 5 position of the pyrrolidine ring afforded antagonists with potent *in vitro* activity.[2] Similarly, replacement of the 1-phenyl substituent of L-(*S*)- or D-(*R*)-3-ethylamino-1-phenylpyrrolidine with a diphenylmethyl or dihydrobenzothiepin group afforded potent antagonists whereas the absolute configuration of the 3 position of the pyrrolidine ring contributed little to potency; the suggestion was made that alteration of the dihedral angle in N-C-C-N should afford compounds having relatively small differences in potency.[12] Indeed, *cis*- and *trans*-1,5-diphenyl-3-dimethylaminopyrrolidine had been found to differ primarily in duration of action rather than potency.[2]

Compounds 1 and 2 differ from the several pyrrolidines discussed above in that one of the phenyl rings is bonded in the ortho position to the carbon $\alpha$ to the dimethylamino group. The 1,2-diphenyl-4-amino-2-butenes upon which Casy and Ison predicated their $H_1$ antagonist model[3] are sufficiently flexible that one of the aromatic rings can approach the aliphatic amino group in the same orientation required by the ortho bonding in 1 and 2. However, the modest activity of 1 and 2 and the high activity of the various conformationally restricted pyrrolidines indicate that the spatial arrangement imposed by the ortho bonding in 1 and 2 is detrimental to antagonist activity. It must also be noted that conformational restrictions such as the ones incorporated in compounds 1 and 2 also necessarily cause changes in the physicochemical characteristics of the molecules and such changes may influence activity.

## Experimental Section

Melting points were obtained on a Mel-Temp apparatus and are uncorrected. Nmr spectra were recorded on a Varian Associates A-60D spectrometer in $CDCl_3$ using tetramethylsilane as the internal standard; results are expressed in parts per million. Ir spectra were recorded on Perkin-Elmer 237B and Perkin-Elmer Infracord spectrophotometers. Mass spectra were recorded using a Finnigan 1015 quadrupole mass spectrometer. The samples were introduced into the ion source *via* the solid probe (ambient tem-

perature); an ionization current of 200 μA and an electron energy of 70 eV were used. Microanalyses were performed by Midwest Microlab, Inc., Indianapolis, Ind., and agreed with the theoretical values to within 0.4%.

**2-Carbomethoxy-α-bromotoluene (3).** A mixture of methyl *o*-toluate (30.0 g, 0.2 mol), *N*-bromosuccinimide (35.6 g, 0.2 mol), and 1.0 g of benzoyl peroxide in carbon tetrachloride (500 ml) was stirred under reflux for 4 hr and allowed to stand overnight at room temperature. Succinimide was removed by filtration and washed with carbon tetrachloride, and the combined filtrates were washed with 5% sodium hydroxide (300 ml) and water (300 ml) and were dried ($Na_2SO_4$). Evaporation *in vacuo* afforded a liquid which was diluted with ether and cooled in a Dry Ice–methanol bath. A white crystalline solid (17.1 g) was collected by filtration and washed with cold ether. The crystallization process was repeated with the combined filtrate and ether washings to yield an additional 13.7 g (67%) of **3** which was used without further purification. *Caution:* compound **3** is a lachrymator and skin irritant.

**Ethyl *N*-(2-Carbomethoxybenzyl)-*N*-phenylglycinate (4).** A mixture of ethyl *N*-phenylglycinate (16.34 g, 0.098 mol), 2,6-lutidine (10.48 g, 0.098 mol), and **3** (22.41 g, 0.098 mol) was heated on a steam bath for 6 hr and cooled to room temperature, and the solid mass was diluted with ether and water. The layers were separated and the water layer was extracted with ether. The combined ether solutions were washed with water, filtered to remove insoluble particles, and dried ($MgSO_4$). Evaporation *in vacuo* afforded **4** as an orange oil which was pure enough (nmr) for use in the next reaction. A sample was purified for elemental analysis by column chromatography on silica gel. The compound was eluted with petroleum ether (bp 60–70°)–ethyl acetate. *Anal.* ($C_{19}H_{21}NO_4$) C, H, N.

**2-Phenyl-3-carboethoxy-2,3-dihydro-4(1*H*)-isoquinolone (5).** Sodium hydride (7.68 g of a 50% dispersion, 0.16 mol) was washed with toluene and suspended in 700 ml of anhydrous benzene. To the suspension was added dropwise **4** (13.1 g, 0.04 mol) in 200 ml of benzene followed by 1 ml of anhydrous ethanol. The mixture was heated under reflux in a nitrogen atmosphere for 5 hr, cooled to room temperature, and slowly diluted with water (200 ml). The layers were separated; the benzene was washed with three 250-ml portions of water, dried ($MgSO_4$), and evaporated to afford 8.74 g of an orange oil which crystallized upon standing at room temperature. Dilution with cold ethanol followed by filtration afforded **5** (5.65 g, 48%) as a yellow crystalline solid: mp 130–134°; nmr 11.83 (s, enolic OH), 6.66–7.95 (m, 9 aromatic H), 4.75 (s, C-1 methylene), 5.86 (q, $COOCH_2$), 1.05 (t, $CH_3$); a singlet at 3.70 which integrated for less than 1 H was assigned to the C-3 H in partially enolized **5**. An analytically pure sample of **5** was obtained by recrystallization from petroleum ether (bp 60–70°): mp 132–134°. *Anal.* ($C_{18}H_{17}NO_3$) C, H, N.

**2-Phenyl-2,3-dihydro-4(1*H*)-isoquinolone Oxime (6).** The keto ester **5** (11.81 g, 0.04 mol) was added to a solution of concentrated sulfuric acid (30 ml) in water (49 ml) and the resulting mixture was heated under reflux for 5 hr, cooled in an ice bath, and made basic with 20% sodium hydroxide. The basic mixture was extracted with ether and the ether solution was washed with water and dried ($MgSO_4$). Evaporation afforded 8.05 g of a reddish brown viscous oil, ir 1680 $cm^{-1}$, which was dissolved in 185 ml of ethanol and added to a solution of hydroxylamine hydrochloride (20.1 g) in 121 ml of water and 81 ml of 10% sodium hydroxide. The resulting solution was heated on a steam bath for 90 min, filtered while hot, and allowed to stand overnight in the cold. A precipitate (5.50 g) was collected and recrystallized from cyclohexane to afford **6** (5.16 g, 54% from **5**), mp 143–146°. An analytically pure sample was obtained by recrystallization from cyclohexane: mp 147°. *Anal.* ($C_{15}H_{14}NO$) C, N, H.

***O*-*p*-Nitrobenzoyl-2-phenyl-2,3-dihydro-4(1*H*)-isoquinolone Oxime (7).** To the oxime **6** (4.76 g, 0.02 mol) in 150 ml of ether was added a solution of *p*-nitrobenzoyl chloride (3.71 g, 0.02 mol) in 50 ml of ether. The resulting mixture was stirred while cooling in an ice bath for 30 min and evaporated *in vacuo*. The residue was taken up in 500 ml of dichloromethane, washed with two 200-ml portions of 5% sodium bicarbonate and with water, and was dried ($MgSO_4$). Evaporation afforded 6.90 g of a solid, mp 135–141°, which was recrystallized from ethyl acetate to yield **7** (4.42 g, 50%), mp 147–151°. A sample was prepared for elemental analysis by recrystallization from ethanol: mp 151–153°. *Anal.* ($C_{22}H_{17}N_3O_4$) C, H, N.

**2-Phenyl-1,2,3,4-tetrahydro-4-aminoisoquinoline (8).** To the oxime ester **7** (9.96 g, 0.026 mol) in 184 ml of anhydrous tetrahydrofuran was added 214 ml of a 1 *M* solution of borane in tetrahydrofuran while stirring in an ice bath. After the addition was complete, the solution was stirred at room temperature for 72 hr, cooled in an ice–salt bath, and acidified by careful dropwise addition of 215 ml of 5% hydrochloric acid. The tetrahydrofuran was removed by evaporation and the aqueous residue was washed with ether, cooled in an ice bath, and made basic (pH 11) by dropwise addition of 10% sodium hydroxide. Solid sodium chloride was added to the basic mixture which was then extracted with ether. The ether solution was washed with saturated sodium chloride, dried ($MgSO_4$), and evaporated to afford **8** (4.45 g, 79%) as an oil which darkened upon exposure to air and was used without further purification.

A sample of **8** was converted to its crystalline acetamide by the following procedure. The amine **8** (0.030 g, 0.001 mol) was added to 5% hydrochloric acid (15 ml) and filtered, and 5% sodium hydroxide was added until the solution became slightly turbid. A few drops of 5% hydrochloric acid were added to clarify the solution and this was followed by immediate addition of ice chips, 3 ml of acetic anhydride, and 3 g of sodium acetate in 3 ml of water. A precipitate was collected (0.33 g) which was dissolved in boiling ethanol and treated with activated charcoal. After removal of the charcoal and concentration of the ethanol, sufficient water was added to cause turbidity and the solution was cooled in a freezer. The acetamide (0.22 g, 80%) was collected as a white crystalline solid: mp 166–167°; ir (KBr) 1650 $cm^{-1}$. *Anal.* ($C_{17}H_{17}N_2O$) C, H, N.

**2-Phenyl-1,2,3,4-tetrahydro-4-dimethylaminoisoquinoline (1).** To a stirred solution of the amine **8** (0.80 g, 0.0033 mol) in 10 ml of acetonitrile was added 1.5 ml of 37% aqueous formaldehyde followed by 0.38 g of sodium cyanoborohydride. The mixture was stirred 15 min at room temperature. The pH was then adjusted to 7 by dropwise addition of glacial acetic acid. Stirring was continued for 45 min with glacial acetic acid being added as required to maintain the pH near 7. The mixture was evaporated to dryness and 20 ml of 2 *N* potassium hydroxide was added to the residue which was then extracted with several portions of ether. The ether extracts were combined and washed with 20 ml of 0.5 *N* potassium hydroxide and with several portions of 5% hydrochloric acid. The acid extracts were combined, made basic with solid potassium hydroxide, and extracted with ether. The ether extracts were dried ($MgSO_4$) and evaporated to yield crude **1** (0.60 g) as a brown oil which was purified by column chromatography on 50 g of basic aluminum oxide (activity grade I). Elution with petroleum ether, benzene–petroleum ether, and ethyl acetate–benzene afforded 0.31 g (36%) of **1** as a tan liquid. The nmr spectrum showed the expected integration although it could not be analyzed by first-order principles: nmr 6.58–7.60 (m, 9 H, aromatic), 2.40 (s, $NC_2H_6$), 2.16–4.71 (discontinuous m, C-1 $H_2$, C-3 $H_2$, and C-4 H); mass spectrum *m/e* (rel intensity) 207 (70), 206 (55), 194 (7), 146 (100), 132 (50). *Anal.* ($C_{17}H_{20}N_2$) C, H, N.

**Ethyl *N*-(2-Carbomethoxybenzyl)-*N*-benzylglycinate (9).** Ethyl *N*-benzylglycinate (15.48 g, 0.08 mol) was dissolved in 2,6-lutidine (8.56 g, 0.08 mol) and this solution was added dropwise at room temperature to **3** (18.32 g, 0.08 mol). After stirring 30 min at room temperature the reaction mixture was heated on a steam bath for 5 hr, cooled to room temperature, and diluted with 100 ml of water and 100 ml of ether. The layers were separated and the water layer was extracted with two 100-ml portions of ether. The combined ether fractions were washed with water, dried ($MgSO_4$), and evaporated to yield 27.65 g (100%) of **9** as a golden liquid. This material was used in the next reaction without further purification.

The hydrobromide of **9** was prepared for elemental analysis by dropwise addition of an ethanol solution of **9** to ethanolic hydrogen bromide and evaporation *in vacuo* followed by trituration of the residue with ether. The resulting precipitate was recrystallized from acetone: mp 118–120°. *Anal.* ($C_{20}H_{24}BrNO_4$) C, H, N.

**2-Benzyl-3-carboethoxy-2,3-dihydro-4(1*H*)-isoquinolone Hydrobromide (10).** Anhydrous ethanol (15.5 ml) was added dropwise to small pieces of sodium wire (10.7 g) and this mixture was warmed and stirred for 30 min. After cooling to room temperature, a solution of **9** (10.58 g, 0.03 mol) in 265 ml of anhydrous benzene was added dropwise in a nitrogen atmosphere. After the addition was complete, the solution was refluxed under nitrogen with stirring and slow distillation of an azeotropic mixture of ethanol–benzene. During the course of the reaction the solution became turbid and this was followed by the appearance of a precipitate. After 5 hr of heating, the mixture was cooled to room temperature and diluted by dropwise addition of 25% acetic acid (300 ml). The aqueous phase was separated and extracted with benzene. The combined benzene portions were washed with 5% sodi-

um bicarbonate and water and were dried ($Na_2SO_4$). Evaporation afforded 7.23 g of an oil which was dissolved in 18 ml of dry ethanol and added dropwise to 18 ml of ethanolic hydrogen bromide. The solution was diluted with benzene and evaporated and the residue was crystallized from ethanol–ether to afford 6.74 g (56%) of **10**, mp 144°. *Anal.* ($C_{19}H_{20}BrNO_3$) C, H, N.

**2-Benzyl-2,3-dihydro-4(1*H*)-isoquinolone (11).** The keto ester **10** (6.80 g, 0.022 mol) (as the free base) was dissolved in 40 g of 60% sulfuric acid. The resulting solution was stirred under reflux for 4 hr, cooled to room temperature, diluted with 200 ml of water, and cooled in an ice bath and the pH was adjusted to 9 by dropwise addition of 20% sodium hydroxide. The basic mixture was extracted with three 150-ml portions of ether which were combined, washed with water, and dried ($MgSO_4$). Evaporation afforded 3.70 g (68%) of **11** as a golden oil: ir (liquid film) 1680 $cm^{-1}$; nmr 8.05 (m, 1 H, aromatic), 7.40 (m, 8 H aromatic), 3.73 and 3.78 (2 H each, s, $CH_2NCH_2$), 3.40 (s, C-3 methylene).

**2-Benzyl-2,3-dihydro-4(1*H*)-isoquinolone Oxime (12).** The ketone **11** (3.70 g, 0.015 mol) was dissolved in 70 ml of ethanol and added to a solution of hydroxylamine hydrochloride (8.89 g) in 50 ml of water and 32 ml of 10% sodium hydroxide. The resulting solution was refluxed on a steam bath for 1 hr, filtered while hot, and allowed to stand overnight at room temperature. The oxime **12** (1.80 g, 47%) was collected by filtration and recrystallized from cyclohexane: mp 161°. *Anal.* ($C_{16}H_{16}N_2O$) C, H, N.

**2-Benzyl-2,3-dihydro-4(1*H*)-isoquinolone Oxime Acetate (13).** The oxime **12** (7.52 g, 0.03 mol) was dissolved in 47 ml of anhydrous pyridine along with 68 ml of acetic anhydride. The solution was heated on a steam bath for 20 min, cooled to room temperature, poured onto ice, and made basic (pH 8) by dropwise addition of 10% sodium carbonate. The cold, basic mixture was extracted immediately with ether and the ether solution was washed with water and dried ($MgSO_4$). Thorough evaporation *in vacuo* afforded **13** (8.30 g, 93%) as a yellow solid: mp 97–99°; nmr 8.11 (m, 1 H, aromatic), 7.16 (m, 8 H, aromatic), 3.70 and 3.80 (2 H each, s, $CH_2NCH_2$), 3.63 (s, C-3 methylene), 2.18 (s, $CH_3$).

**2-Benzyl-1,2,3,4-tetrahydro-4-dimethylaminoisoquinoline (2).** To a cold stirring solution of the oxime acetate **13** (6.35 g, 0.215 mol) in 110 ml of tetrahydrofuran was added dropwise 121 ml of a 1 *M* solution of borane in tetrahydrofuran. After the addition was complete, the mixture was stirred 70 hr at room temperature. The mixture was again cooled in an ice–salt bath and 84 ml of 5% hydrochloric acid was cautiously added. The solvent was evaporated and the aqueous residue was washed with ether and made basic (pH 12) by the addition of 10 *N* potassium hydroxide. Solid sodium chloride was added and the basic mixture was extracted with several portions of ether which were combined, washed with saturated sodium chloride, and dried ($MgSO_4$). Evaporation afforded 3.45 g (67%) of **14** as a brown viscous oil. The nmr spectrum of the crude amine gave the expected integration and exhibited a two proton singlet which disappeared upon treatment of the solution with deuterium oxide.

The crude amine **14** (3.45 g, 0.0145 mol) was dissolved in 41 ml of acetonitrile to which was added 5.7 ml of 37% formaldehyde and 1.35 g of sodium cyanoborohydride. After 15 min of stirring, sufficient glacial acetic acid was added to adjust the pH to 7. Stirring was continued for 45 min with glacial acetic acid being added as required to maintain a pH of 7. The acetonitrile was evaporated *in vacuo* and 50 ml of 2 *N* potassium hydroxide was added to the residue. The basic mixture was extracted with several portions of ether which were combined, washed with 0.5 *N* potassium hydroxide, and extracted with 10% hydrochloric acid. The acid extracts were made basic (pH 10) with solid potassium hydroxide and were extracted with ether. The ether extracts were combined, dried ($MgSO_4$), and evaporated to afford 3.05 g of crude **2** as a brown oil. The crude product was chromatographed on a column of 150 g of basic aluminum oxide (activity grade I). Elution with petroleum ether (bp 60–70°) followed by benzene–petroleum ether and benzene–ethyl acetate afforded **2** (2.20 g, 57%) as a tan oil. A sample of **2** was rechromatographed as described above for pharmacological testing and elemental analysis: nmr 6.70–7.66 (m, 9 H, aromatic), 3.88 (m, C-4 H), 3.63 and 3.51 (2 H each, s, $CH_2NCH_2$), 2.75 (m, C-3 methylene), 2.30 (s, $NC_2H_6$); mass spectrum *m/e* (rel intensity) 221 (66), 220 (38), 146 (100), 132 (62), 91 (22). *Anal.* ($C_{18}H_{22}N_2$) C, H, N.

**Pharmacology.** Testing was carried out on the isolated guinea pig ileum which was prepared according to a standard method.[10] The ileum was bathed in Tyrodes solution at 37° and bubbled with air. Tissues were allowed to stabilize for a minimum of 15 min before introduction of agonists. Histamine was then added at 3-min intervals until reproducible contractions were obtained. Antagonists were dissolved in 0.1 *N* hydrochloric acid and an aliquot was dissolved in Tyrodes solution for pharmacological evaluation. Antagonists were allowed to remain in contact with the ileal tissue for 15 min prior to the addition of $4 \times 10^{-6}$ *M* histamine or $4 \times 10^{-7}$ *M* acetylcholine. The $ED_{50}$ values cited in the discussion are the results of three determinations at three dose levels. The values in parentheses are standard errors.

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## Quinazolines as Inhibitors of Dihydrofolate Reductase. 2[1,†]

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During the past decade a wide variety of quinazoline derivatives has been synthesized and evaluated as potential chemotherapeutic agents. Noteworthy examples of classical quinazoline antifolates are chlorasquin and methasquin which are highly potent inhibitors of dihydrofolate reductase. These compounds have also been studied extensively as potential antineoplastic agents.[2-5] On the other hand, the discovery of the potent antiprotozoan activity of 2,4-diamino-6-(benzylamino)quinazolines, **1a–c**, has stimulated the development of a wide variety of potent nonclassical quinazoline antifolates.[6-9] These efforts have culminated in the selection of two 2,4-diamino-6-

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