in 25 ml of EtOH, was added to 2.8 g (40 mmoles) of $HONH_2 \cdot HCl$ in 50 ml of EtOH containing 15 ml of pyridine. The reaction mixture was refluxed for 2 hr and then left at room temperature overnight. Dilution with a large volume of H_2O and filtration gave 4.52 g of the title compound **5** (59%), mp 154–156°, λ_{max}^{KBr} 3.10 (OH) and 6.17 μ (w, >C=N). The analytical sample (from MeOH) melted at 160–161°. *Anal.* ($C_{18}H_{33}NO_2$) C, H, N.

***dl*-7β-*t*-Butoxy-1,2,3,4,4aα,4b,5,6,7,8,8aα,9,10,10aβ-tetradecahydro-4bβ-methylphenanthryl-2α- and -2β-amines (6 and 7).**—A solution of 9.4 g (0.029 mole) of the oxime **5** in 70 ml of absolute EtOH was heated under reflux while 10 g (0.44 g-atom) of Na was added portionwise over a 2.5-hr period. At the end of this time more EtOH and some ice water were added. The mixture was extracted (Et_2O) and the ether was washed (saturated NaCl). The aqueous layers were washed with another portion of ether. The combined ether layers were dried (Na_2SO_4) and the solvent was removed by warming *in vacuo* to afford 10 g of an oil. The oil was chromatographed on 1 kg of silica gel using Et_2O–pentane–MeOH–*i*-$PrNH_2$ (50:44:3:3) for elution. Fractions were combined on the basis of tlc analysis. The main fraction afforded 7.2 g of a mixture of amines.

Partial separation of the isomers was obtained by using a method of partition chromatography described by Brown and Kupchan.[8] The solvent system employed was a 12:1:2:0.2 mixture of hexane–$(ClCH_2)_2$–MeOH–H_2O. Supercel (400 g) was wetted with 300 ml of the polar phase containing 100 mg of bromocresol purple, the color of the indicator mixture was adjusted to a pale creamy yellow (faintly acid) by gaseous HCl, and the solid was packed into a column 9 cm in diameter. A sample of the mixture of amines (6.7 g) was dispersed on approximately 50 g of Supercel and placed on top of the column. Elution of the column with the nonpolar phase of the solvent mixture developed the column; the position of the basic amines was revealed by two bands.

The more polar band was obtained by slicing the column and eluting the component with Et_2O-*i*-$PrNH_2$, (20:1). The oil obtained (2.3 g) gave a major single spot with an R_f of 0.19 and a trace of impurity at R_f 0.31 (silica gel, Et_2O–MeOH–*i*-$PrNH_2$, 94:3:3). The less polar band afforded 4.2 g of oil which was a mixture of the two amines by tlc. The latter material was chromatographed again on 14 silica gel coated plates (Brinkmann PF_{254} silica gel, 20 × 40 cm) having a 1-mm coating. The plates were developed with Et_2O–*i*-$PrNH_2$–MeOH, (94:3:3). The major more polar band afforded another 3.3 g of oil having a single spot by tlc identical with the material of R_f 0.19. The less polar material afforded 0.4 g of product having a major spot on a tlc analysis (silica gel, Et_2O–MeOH–*i*-$PrNH_2$, 94:3:3) at R_f 0.31 with a trace at R_f 0.19.

Structure **6** was assigned to the material of R_f 0.19 (62%) and **7** to the material at R_f 0.31 (4.5%). These amines could not be crystallized.

2-Chloroethyl *dl*-N-(7β-*t*-Butoxy-1,2,3,4,4aα,4b,5,6,7,8,8aα,-9,10,10aβ-tetradecahydro-4bβ methylphenanthr-2α-yl)carbamate (8).—The amine **6** (4.4 g, 15 mmoles) was suspended in 50 ml of ice-water. While the flask was being swirled in an ice bath, 4 g (28 mmoles) of chloroethyl chloroformate was added portionwise. When half of the reagent had been added, 10 ml of 2 *N* NaOH was added to the reaction mixture followed by the remainder of the chloroethyl chloroformate. More 2 *N* NaOH (10 ml) was added. The reaction mixture was swirled occasionally for another 15–30 min while in the ice bath. The oily contents of the flask solidified and were filtered. The solid residue was dissolved in ether. The ether solution was washed (dilute HCl, saturated NaCl) and dried (Na_2SO_4). Concentration of the solvent *in vacuo*, filtration, and recrystallization of the precipitate afforded 2.8 g (47%) of **8**. The analytical sample (from Et_2O) melted at 153–154°. A similar sample had λ_{max}^{KBr} 3.05 (ms) and 6.50 (NH), 5.94 μ (s) (>C=O). *Anal.* ($C_{22}H_{38}ClNO_3$) C, H, Cl.

2-Chloroethyl *dl*-N-(7β-*t*-Butoxy-1,2,3,4,4aα,4b,5,6,7,8,8aα,-9,10,10aβ-tetradecahydro-4bβ-methylphenanthr-2β-yl)carbamate (9).—In a procedure similar to that used in the above experiment, 0.4 g (1.3 mmoles) of amine **7** afforded 0.4 g (73%) of **9**, mp 132–134°. The analytical sample from cyclohexane melted at 134–135°. *Anal.* ($C_{22}H_{38}ClNO_3$) C, H, N.

2-Dimethylaminoethyl *dl*-N-(7β-*t*-Butoxy-1,2,3,4,4aα,4b,5,6,-7,8,8aα,9,10,10aβ-tetradecahydro-4bβ-methylphenanthr-2α-yl)carbamate (10).—A solution of 2.42 g (6.0 mmoles) of **8** in 80 ml of Me_2NH was heated in a sealed glass tube on a steam bath for 24 hr. The tube was cooled and opened and the excess Me_2NH was evaporated. Ether and 2 *N* NaOH were added and the layers were separated. The ether was washed with saturated NaCl. The aqueous layers were washed again with a portion of ether. The combined ether layers were dried (Na_2SO_4) and the solvent was removed by warming *in vacuo* to afford **10**. In the usual manner, 2.17 g (81%) of crude hydrochloride salt was obtained, mp 115–125° (gelatinous melt). Two recrystallizations from acetone–ether afforded 1.58 g of **10 hydrochloride**, mp 150–180°. *Anal.* ($C_{24}H_{44}N_2O_3 \cdot HCl$) C, H, N.

2-Dimethylaminoethyl *dl*-N-(1,2,3,4,4aα,4b,5,6,7,8,8aα,9,10,-10aβ-Tetradecahydro-7β-hydroxy-4bβ-methylphenanthr-2α-yl)-carbamate 7-Trifluoroacetate (11).—A solution of 1.3 g (3.1 mmoles) of **10** (free base) in 18 ml of CF_3CO_2H stood in an ice bath for 2.5 hr. The solvent was removed at 40° (maximum). CH_2Cl_2 and saturated $NaHCO_3$ were added. The layers were separated and the organic phase was washed with saturated NaCl and dried (Na_2SO_4), and the solvent was removed *in vacuo*. Recrystallization of the residue from ether and then from acetone afforded 0.85 g (61%) of **11**, mp 196–197°. The analytical sample from acetone melted at 199–200°. *Anal.* ($C_{22}H_{35}F_3N_2O_5$) N, F, N_{AE}.

2-Dimethylaminoethyl *dl*-N-(1,2,3,4,4aα,4b,5,6,7,8,8aα,9,10,-10aβ-Tetradecahydro-7β-hydroxy-4bβ-methylphenanthr-2α-yl)-carbamate (2).—Compound **11** (680 mg, 1.5 mmoles) in 50 ml of MeOH containing 10 ml of concentrated NH_4OH sat at room temperature for 1 hr. The mixture was poured into some ice and the aqueous mixture was extracted with ether. The ether was washed with saturated NaCl. The aqueous layers were washed with a fresh portion of ether. The combined ether layers were dried (Na_2SO_4). Concentration *in vacuo* afforded 400 mg of **2** (87%), mp 133–134°. The analytical sample obtained from ether melted at 134–135°. *Anal.* ($C_{20}H_{36}N_2O_3$) C, H, N.

(8) K. S. Brown and S. M. Kupchan, *J. Chromatog.*, **9**, 71 (1962).

Silicon-Substituted Medicinal Agents. Parasympatholytic Activity of 3,3-Dimethyl-1-butanol Carbamate and 2-Trimethylsilyl-1-ethanol Carbamate[1]

RALPH J. FESSENDEN[2] AND ROGER RITTENHOUSE

Department of Chemistry, San Jose State College, San Jose, California

Received May 31, 1968

In a previous study[3] it was observed that 3,3-dimethyl-1-butanol carbamate (**1**) was a convulsant and ten times more toxic than its silicon isostere, 2-trimethylsilyl-1-ethanol carbamate (**2**), a muscle relaxant at high doses.

$(CH_3)_3CCH_2CH_2OCONH_2$ **1** $(CH_3)_3SiCH_2CH_2OCONH_2$ **2**

$(CH_3)_3NCH_2CH_2OCONH_2$ **3**

Both **1** and **2** are similar in structure to carbachol (**3**), and it was of interest to determine if these compounds would elicit a muscarinic response in a guinea pig ileum assay.

After standardization of the test conditions (see Experimental Section), **1** and **2** were assayed and found to be void of muscarinic activity. However, both compounds were found to be antagonists of the muscarinic

(1) This research was supported by Public Health Service Research Grant GM10122 from the National Institute of General Medical Sciences.

(2) (a) Alfred P. Sloan Fellow, 1965–1969. (b) To whom inquiries should be addressed: Department of Chemistry, University of Montana, Missoula, Montana 59801.

(3) R. J. Fessenden and M. D. Coon, *J. Med. Chem.*, **8**, 604 (1965).

activity of both acetylcholine and carbachol. The dose–response curves of acetylcholine (10^{-7} *M*) antagonized by **1** or **2** were identical within experimental error. The ED_{50} for both **1** and **2** was 0.07 mg/ml (*ca.* 4×10^{-4} *M*).

Previous workers have studied acetylcarbocholine[4] and acetylsilicocholine[5] and have concluded that these two compounds act as indirect, nonnicotinic, cholinergic agents. This study implies that both **1** and **2** have an affinity for the muscarinic site but do not possess intrinsic activity.

Experimental Section

The procedure described by Turner[6] was followed for bioassay purposes. The bath temperature was maintained at 37 ± 1°, and oxygenated Tyrode's solution was used as the perfusing fluid. Hexamethonium bromide (10 mg/l.) was added to the Tyrode's solution to prevent ganglionic response. The final bath volume for each run was 30 ml and chemical concentrations were calculated accordingly.

The test compounds could only be dissolved in Tyrode's solution with the aid of ethanol. Delivery of 1 ml of the test solution to 29 ml of bath gave a final concentration of 1.6% ethanol. This did not have a significant influence on the muscle response induced by 10^{-7} *M* acetylcholine.

A typical run was carried out in the following manner. The ileum was dissected, then washed with Tyrode's solution. Approximately 5 cm of the muscle was attached to a transducer in the muscle bath (29 ml) and allowed to stand until the spontaneous contractions had subsided. The transducer was connected to a strip chart recorder. Acetylcholine chloride solution (1 ml) (final bath concentration 10^{-7} *M*, 1.6% ethanol) was delivered to the bath. The amplitude of the strip chart recorder was adjusted so that the resulting contraction gave a 70–90% pen deflection. The muscle was then washed continuously with Tyrode's solution until the muscle had relaxed. The wash solution was drained, 29 ml of fresh Tyrode's solution was added to the bath, and the system was allowed to equilibrate. After the pen deflection was adjusted, the calibration was repeated at least five times during and at the end of the assays of the test compounds. The average of these values was taken as the 100% contraction value for 10^{-7} *M* acetylcholine for the particular muscle.

The test compounds were assayed in essentially the same manner. The initial bath volume was 28 ml. To this was added 1.0 ml of the test solution followed within about 15 sec by 1.0 ml of the acetylcholine solution (final bath concentration, 10^{-7} *M* ACh). The process was repeated three times and the average of the pen deflections was taken as the value for that particular concentration of test compound. This value was then expressed as a per cent of the contraction value for 10^{-7} *M* ACh. At least four animals were used for each compound. The data that were obtained are summarized in Figure 1.

The test compounds alone gave no muscle response in the dose range of 0.30 to 2.5×10^{-5} mg/ml.

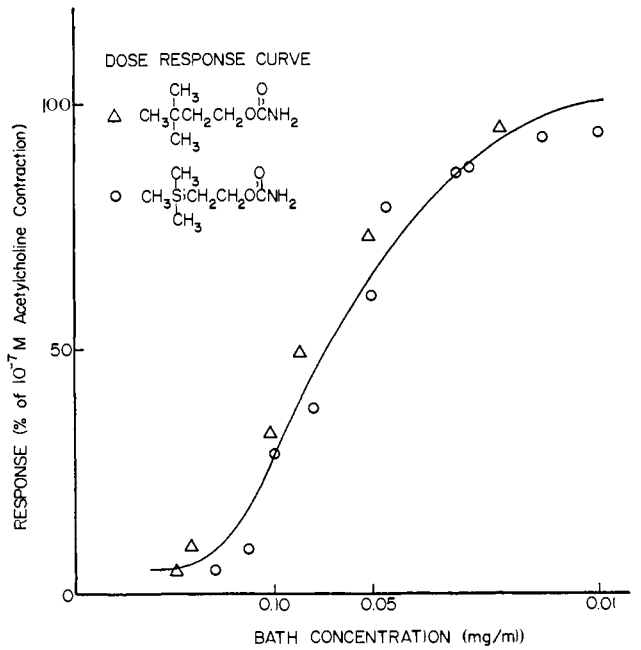


Figure 1.

(4) (a) J. Banister and V. P. Whittaker, *Nature*, **167**, 605 (1951); (b) A. S. V. Burgen, *Brit. J. Pharmacol.*, **25**, 4 (1965).

(5) P. Th. Henderson, E. J. Ariens, B. W. J. Ellenbrock, and A. M. Simonis, *J. Pharm. Pharmacol.*, **20**, 26 (1968).

(6) R. A. Turner, "Screening Methods in Pharmacology," Academic Press Inc., New York, N. Y., 1965, pp 42–46.

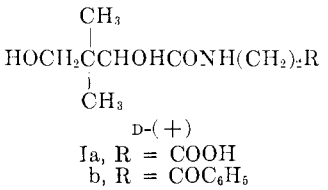
Potential Antimalarial Substances. Antimetabolites of Pantothenic Acid[1]

EDWARD F. ELSLAGER, MARLAND P. HUTT, AND LESLIE M. WERBEL

Research Laboratories, Parke, Davis and Company, Ann Arbor, Michigan 48106

Received March 25, 1968

In 1943 it was observed that the survival of the erythrocytic stages of *Plasmodium lophurae*, maintained intracellularly in duck red cell suspensions *in vitro*, was favored by the addition of calcium pantothenate to the medium.[2] Moreover, the development of *Plasmodium gallinaceum* in chickens is inhibited by a deficiency in pantothenic acid (Ia).[3] These observations stimulated a search for antimetabolites that might interfere with the utilization of pantothenic acid by plasmodia.[3–7] Among the antipantothenates that were synthesized and tested earlier,[3–7] D-(+)-phenylpantothenone (Ib) was equiactive with quinine against *P. gallinaceum* and twice as potent as quinine against *P. lophurae* in the chick.[5,7] In man, Ib exhibited slight activity against blood-induced vivax malaria and was tolerated well in doses of 2 g daily for 4 days.[5] The pantoyltaurine derivatives VIa and VIb were the



most active compounds synthesized; both were approximately ten times as potent as quinine against *P. gallinaceum* in chicks.[5,6]

More recently it was shown that pantothenic acid has no effect on the erythrocytic stages of *P. lophurae*

(1) This investigation was supported by U. S. Army Medical Research and Development Command Contract DA-49-193-MD-2754. This is Contribution No. 360 to the Army Research Program on Malaria.

(2) W. Trager, *J. Exp. Med.*, **77**, 411 (1943).

(3) S. Brackett, E. Waletzky, and M. Baker, *J. Parasitol.*, **32**, 453 (1945).

(4) J. F. Mead, M. M. Rapport, A. E. Senear, J. T. Maynard, and J. B. Koepfli, *J. Biol. Chem.*, **163**, 465 (1946).

(5) F. Y. Wiselogle, "A Survey of Antimalarial Drugs, 1941–1945," Vol. I, J. T. Edwards, Ann Arbor, Mich., 1946, pp 138–140.

(6) R. Winterbottom, J. W. Clapp, W. H. Miller, J. P. English, and R. O. Roblin, Jr., *J. Am. Chem. Soc.*, **69**, 1393 (1947).

(7) R. E. Lutz, J. W. Wilson, III, A. J. Deinet, G. H. Harnest, T. A. Martin, and J. A. Freek, *J. Org. Chem.*, **12**, 96 (1947).