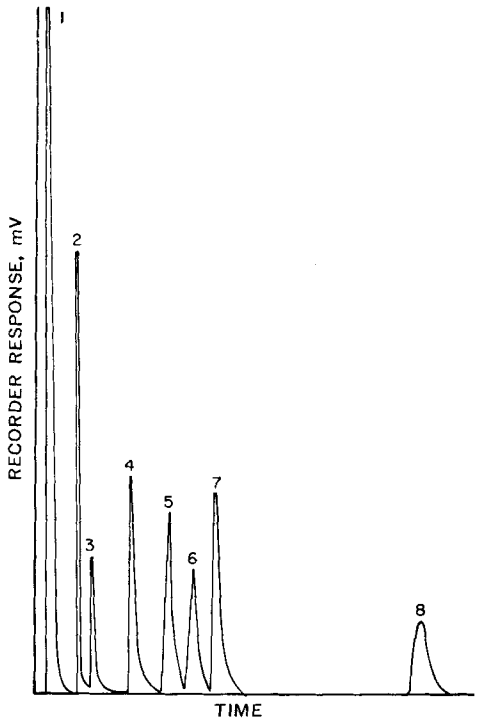

Figure 1—*Gas chromatograph showing:* (1) *solvent peak; derivatives of;* (2) *benzoic acid;* (3 *and* 4) *salicylic acid;* (5) *acetylsalicylic acid;* (6) *2,5-dihydroxybenzoic acid;* (7) *2,3-dihydroxybenzoic acid; and* (8) *2,3,4-trihydroxybenzoic acid.*

persisted, after which approximately a 20% excess by volume of ethereal diazomethane was added. The reactions were allowed to proceed at room temperature for 48 hr. in stoppered flasks. Additional ethereal diazomethane was added periodically if the resulting solutions became clear. After the reaction was completed, the solvent was evaporated at room temperature under a stream of dry nitrogen. The residual yellow viscous methyl ester–methyl ether derivatives were transferred quantitatively to 10-ml. volumetric flasks and brought to volume with pyridine.

An Aerograph 200, model 2041B (Wilkens Instrument and Research Inc., Walnut Creek, Calif.) equipped with a hydrogen-flame ionization detection system was used. Separations were carried out at 155° with a 1.524 m. (5 ft.) × 1.75 mm. i.d. stainless steel column packed with 5% SE-30 on diatomaceous earth[1] 60-80 mesh. Injector and detector temperatures were maintained at 205°. The helium carrier gas flow rate was 25 ml. per minute and the hydrogen pressure was maintained at 10 p.s.i.

RESULTS AND DISCUSSION

The relative retention times of the respective derivatives, as compared with the retention time of the methyl ester of benzoic acid, are reported in Table I. Figure 1 represents a chromatograph of a mixture of the respective compounds. Salicylic acid yielded two peaks with the same relative retention times following three separate derivative preparations. The purity of the salicylic acid was confirmed by TLC. 2,3,4-Trihydroxybenzoic acid was used as a model for the behavior of the 2,3,5-trihydroxybenzoic acid metabolite. It is not unreasonable to assume that the two compounds would demonstrate similar behavior toward the GLC system used. Since the retention time of the trihydroxy derivative is much longer than any of the other hydroxy derivatives, one would not expect the 2,3,5-trihydroxy metabolite to interfere with the analysis of the other compounds. The derivatives were found to be stable for a period of several weeks when stored at 4°.

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[1] Chromosorb W, Applied Science Laboratories Inc., State College, Pa.

New Synthesis of *rac.* Anhalonidine and *rac.* Pellotine

M. TAKIDO, K. L. KHANNA, and A. G. PAUL

Abstract □ A new synthesis of *rac.* anhalonidine and *rac.* pellotine is reported. The procedure, a modification of the method of Bobbitt *et al.* (5) for the synthesis of 1,2,3,4-tetrahydroisoquinolines, is simpler and gives better yields than those previously reported.

Keyphrases □ Anhalonidine, racemic—synthesis □ Pellotine, racemic—synthesis □ IR spectrophotometry—structure □ NMR spectroscopy—structure

The need for quantities of anhalonidine and pellotine for use in the biosynthesis of the peyote alkaloids and the inability to obtain them commercially or in sufficient quantities from the peyote cactus required that they be synthesized. Reported syntheses (1–4) are either lengthy or the yields are low. The method of Bobbitt *et al.* (5) for the synthesis of related tetrahydroisoquinolines was modified for the synthesis of these alkaloids as shown in Fig. 1.

3,4-Dimethoxygalloacetophenone (I), prepared by the method of David and Kostanecki (6), was condensed with aminoacetaldehyde diethylacetal to give Schiff's base (II) which was conveniently reduced at room temperature with sodium borohydride to yield *N*-[2-(3′,4′-dimethoxy-2′-hydroxyphenyl)ethyl]aminoacetaldehyde diethylacetal (III). Compound III was cyclized in 8 *N* HCl at room temperature to yield 4,8-dihy-

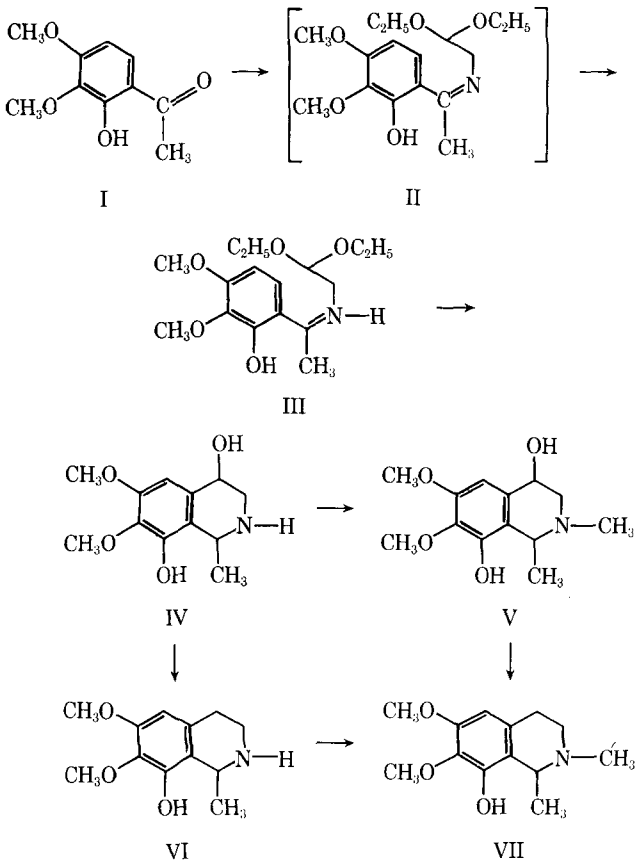

Figure 1—*Synthesis of* rac. *anhalonidine and* rac. *pellotine.*

droxy-6,7-dimethoxy-1-methyl-1,2,3,4-tetrahydroisoquinoline hydrochloride (IV). Reduction of IV in 6 *N* HCl with palladium-on-charcoal as the catalyst yielded anhalonidine hydrochloride (VI). Alternatively, III was mixed with 6 *N* HCl and palladium-on-charcoal and reduced to give VI directly. Yields by both methods were approximately equal.

Pellotine (VII) was synthesized by adding sodium borohydride to a mixture of IV and formaldehyde in methanol to give 4,8-dihydroxy-6,7-dimethoxy-1,2-dimethyl-1,2,3,4-tetrahydroisoquinoline (V). Reduction of V in 6 *N* HCl with palladium-on-charcoal as the catalyst yielded VII as the hydrochloride. Alternatively, VII was prepared by methylation of VI using formaldehyde in methanol in the presence of sodium borohydride.

In addition to presenting new methods for the synthesis of VI and VII in better yields then previously reported, III, IV, and V represent new compounds not previously reported in the literature. Proof of structure of these compounds is based upon elemental analyses, IR and NMR spectral data, and analogy with similar types of compounds synthesized by Bobbitt and Sih (7).

EXPERIMENTAL

***N*-[2-(3′,4′-dimethoxy-2′-hydroxyphenyl)ethyl]aminoacetaldehyde Diethylacetal (III)**—A solution of I (3.96 g.) and aminoacetaldehyde (2.66 g.) in 80 ml. of absolute ethanol was allowed to stand at room temperature for 24 hr. The solution was stirred at room temperature and sodium borohydride (1.0 g.) was added in small portions over a period of 0.5 hr. The solution was then added to 80 ml. of water and extracted with ether (3 × 200 ml.). The ether solution was concentrated and extracted with 1 *N* HCl. The acid extract was basified with 2 *N* Na_2CO_3 solution and extracted with ether. The ether was removed and the oil (4.5 g., 71%) was purified by distillation, b.p. 116–117°/0.03 mm. $\nu_{max.}^{film}$: 3275 cm.$^{-1}$ (—NH—); 3125 cm.$^{-1}$ (—OH). NMR[1] ($CDCl_3$), p.p.m.: 6.46 d [1], J = 8 c.p.s. (ar. H); 6.13 d [1], J = 8 c.p.s. (ar. H); 4.45 tr [1], J = 5 c.p.s. [—CH_2—$\underline{CH}(OC_2H_5)_2$]; 3.91 s [3], ($OCH_3$); 3.82 s [3], ($OCH_3$); 3.60 m [4], (2 × —O—$\underline{CH_2}$—$CH_3$); 2.53 d [2], J = 5 c.p.s. [—$\underline{CH_2}$—CH(OC_2H_5)$_2$]; 1.19 m [9], (2 × —OCH_2—CH_3; —NH—CH—CH_3).

Anal.—Calcd. for $C_{16}H_{27}NO_5$: C, 61.32; H, 8.68. Found: C, 61.74; H, 8.48.

4,8-Dihydroxy-6,7-dimethoxy-1-methyl-1,2,3,4-tetrahydroisoquinoline (IV) Hydrochloride (4-Hydroxyanhalonidine Hydrochloride)—Compound III (3.0 g.) was dissolved in 75 ml. 8 *N* HCl and the solution was permitted to stand for 10 hr. at room temperature. The acid solution was then adjusted to pH 6.5 with aqueous NaOH and washed with chloroform. The aqueous solution was filtered, freeze-dried, and the solid residue extracted with chloroform–methanol (9:1). The extract was evaporated to dryness *in vacuo* at <40° and the residue crystallized from methanol–ethylacetate as colorless plates, m.p. 217–218° (yield, 2.5 g., 87%). IR (KBr): 3420 cm.$^{-1}$ (—NH—); 3100 cm.$^{-1}$ (—OH); 1178 cm.$^{-1}$ (phenolic —OH); 1070 cm^{-1} (sec. alcohol —OH).

Anal.—Calcd. for $C_{12}H_{18}ClNO_4$: C, 52.27; H, 6.58; Cl, 12.86; N, 5.08. Found: C, 52.10; H, 6.70; Cl, 12.84; N, 5.03.

The base, prepared from the hydrochloride, crystallized from benzene–ethylacetate as colorless plates, m.p. 195–196° (dec.). IR (KBr): 3510 cm.$^{-1}$ (—NH—); 3070 cm.$^{-1}$ (—OH); 1190 cm.$^{-1}$ (phenolic —OH); 1060 cm.$^{-1}$ (sec. alcohol —OH). NMR (DMSO), p.p.m.: 6.49 s [1], (ar. H); 4.17, 4.05, 3.95 complex [2], [CH_3—$\underline{CH}$—NH—, —$\underline{CH}$(OH)—CH_2—]; 3.77 s [3], (—OCH_3); 3.68 s [3], (—OCH_3); 2.91 q [2], J = 2.5 c.p.s. [—CH(OH)—$\underline{CH_2}$]; 1.26 d [3], J = 6.5 c.p.s. ($\underline{CH_3}$—CH—NH—).

The picrate, prepared from the hydrochloride, recrystallized from benzene–ethanol as yellow plates, m.p. 206–208°.

4,8-Dihydroxy-6,7-dimethoxy-1,2-dimethyl-1,2,3,4-tetrahydroisoquinoline (V) (4-Hydroxypellotine)—Compound IV (200 mg.) and formaldehyde (120 mg., 37%) were dissolved in methanol (10 ml.). Sodium borohydride (80 mg.) was added to the stirred solution in small portions over a period of 0.5 hr. The solution was then added to water (10 ml.) and extracted with chloroform (3 × 20 ml.). The chloroform extract was evaporated to dryness and the residue washed with ether. The crystalline solid remaining was crystallized from benzene to give colorless needles, m.p. 177–178° (yield 180 mg., 86%). IR (KBr): 3130 cm.$^{-1}$ (—OH); 1185 cm.$^{-1}$ (phenolic —OH); 1080 cm.$^{-1}$ (sec. alcohol —OH). NMR ($CDCl_3$), p.p.m.: 6.54 s [1], (ar. H); 4.40 s, 4.09 s, 3.98 complex [2], [—$\underline{CH}$(OH)—CH_2—, —NH—$\underline{CH}$—CH_3]; 3.88 s [3], (—OCH_3); 3.86 s [3], (—OCH_3); 2.90 d [2], J = 13 c.p.s. [—CH(OH)—$\underline{CH_2}$]; 2.50 s [3], (>N—CH_3), 2.17 s [1], [—CH($O\underline{H}$)—CH_2—]; 1.17 d [3], J = 6.5 c.p.s. [—CH(CH_3)—N<].

Anal.—Calcd. for $C_{13}H_{19}NO_4$: C, 61.64; H, 7.56; N, 5.53. Found: C, 61.84; H, 7.66; N, 5.44.

Anhalonidine (VI) Hydrochloride—*Method A*—Compound III (1.2 g.) was dissolved in 8 *N* HCl (25 ml.) and the solution was permitted to stand at room temperature for 9 hr. The solution was then adjusted to approximately 6 *N* by the addition of water (10 ml.) and additional 6 *N* HCl (10 ml.) added. Palladium-on-charcoal (1.0 g., 5%) was added and the mixture was reduced at atmospheric pressure and room temperature for approximately 24 hr. The mixture was filtered and the filtrate concentrated *in vacuo* at <50° using a film-flash evaporator. Ethanol–benzene was added from time to time to azeotrope the water. The solid residue obtained was crystallized from methanol–ether to give colorless needles (yield 0.75 g., 73%).

Method B—Compound IV (100 mg.) was dissolved in 6 *N* HCl (20 ml.), palladium-on-charcoal (0.2 g., 5%) added, and the mixture

[1] s = singlet; d = doublet; tr = triplet; q = quartet; m = multiplet; ar. = aromatic.

was reduced and treated as described above (yield, 70 mg., 74%).

The free base, m.p. 160° [lit. m.p. (1) 160°], and the salicylate, m.p. 223–225° [lit. m.p. (4) 223.5–224.5°], prepared from VI hydrochloride showed no melting point depression when mixed with authentic samples. The IR spectra of the hydrochloride and the salicylate were identical with authentic samples.

Pellotine (VII) Hydrochloride—*Method A*—Sodium borohydride (20 mg.) was added in small portions over a period of 0.5 hr. to a stirred solution containing VI (38 mg.) and formaldehyde (2 drops, 37%) in methanol (5 ml.) at room temperature. The solution was then added to water (5 ml.) and extracted with chloroform (3 × 10 ml.). The chloroform extract was dried over anhydrous sodium sulfate and then evaporated to dryness. The residue was dissolved in a small quantity of methanol–chloroform–ether, and dry HCl gas was passed through the solution. Colorless needles crystallized (yield 35 mg., 75%).

Method B—A mixture of V (90 mg.), 6 *N* HCl (20 ml.), and palladium-on-charcoal (200 mg., 5%) was reduced at atmospheric pressure and room temperature for approximately 24 hr. The mixture was filtered and the filtrate concentrated *in vacuo* at <50° using a film-flash evaporator. Ethanol–benzene was added from time to time to azeotrope the water. The solid residue was crystallized from methanol–ether (yield 87 mg., 89%).

The free base, m.p. 116° [lit. m.p. (1) 111–112°], showed no melting point depression when mixed with authentic pellotine. The IR spectra of the base and hydrochloride were identical with that of authentic samples.

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Pentaerythritol Tetranitrate Sustained-Release Tablets: Relation of *In Vitro* Release of the Drug to Blood Pressure Changes after Administration to Anesthetized Cats

SACHCHIDANANDA BANERJEE, ARUN KUMAR MUKHERJEE, and ARUP KUMAR HALDER

Abstract □ The *in vitro* release of pentaerythritol tetranitrate (PET) from a sustained-action PET tablet was determined using USP tablet disintegration apparatus with the modification that 100-mesh stainless steel wire cloth replaced the usual 10-mesh. The immersion medium was simulated gastric fluid for the first 2 hr. and simulated intestinal fluid for the subsequent 10 hr. There was a gradual release of the drug and 80 to 100% of the drug was dissoluted at the end of 12 hr. The tablet when administered by mouth to anesthetized cats produced a 28 to 30% fall in arterial blood pressure which persisted for 12 hr. A good correlation between the rate of release of the drug *in vitro* and the *in vivo* effect in sustained lowering of the blood pressure was obtained. The *in vitro* method described is proposed for the quality control of sustained-action PET tablets.

Keyphrases □ Pentaerythritol tetranitrate sustained-release tablets—drug release □ Blood pressure changes—pentaerythritol tetranitrate release □ Dissolution rates—pentaerythritol tetranitrate tablets □ *In vivo-in vitro* release rates correlation—pentaerythritol tetranitrate tablets

Difficulties were experienced in testing the quality of sustained-release tablets of pentaerythritol tetranitrate using procedures usually adopted for the quality control of sustained-release preparations. In the dissolution test the concentration of the drug released in the bath fluid from the tubes containing tablets is estimated at various intervals. In the assay of pentaerythritol tetranitrate such a procedure could not be adopted as the analysis is based on the estimation of nitrates. To estimate nitrates in the bath fluid, large volumes of the solution had to be evaporated to dryness before extraction with glacial acetic acid (1). In the *in vivo* test the determination of the rate of absorption of the drug would involve the collection of large volumes of blood at frequent intervals and estimation of the drug in these blood samples. Both procedures were impractical.

In the dissolution test, if the remnants of the tablets in the disintegration apparatus and not the bath fluid were collected at different intervals, the estimation of pentaerythritol tetranitrate would be possible and the *in vitro* rate of release of the drug from the tablet could be calculated. The drug lowered blood pressure and a sustained fall in blood pressure could be observed when the sustained-release tablet was administered to cats. A study, therefore, was undertaken to establish a relation, if any, between the *in vitro* release of the drug and the lowering of blood pressure in cats after administration of sustained-release pentaerythritol tetranitrate tablets. A good correlation was observed between the *in vitro* release of the drug and the fall in