combined and streaked on silica gel (PF 254 and 366 plates). The new metabolite, a combination band, containing three additional metabolites, as well as other material resulted. The band with R_f of ~0.4 was scraped and eluted with chloroform–methanol (1:1). The extract was evaporated to dryness. The mass spectrum of this residue showed peaks at *m/e* 200 (M^+), 183, 171, 147, 145, 129, 117, and 115. This spectrum was identical in every respect with that of authentic 3-hydroxymethyl-*s*-triazolo[3,4-*a*]phthalazine.

The combination band with R_f value 0.5 was also scraped; it was eluted with acetone. The acetone extract was concentrated and streaked on another silica gel plate and chromatographed with acetone–cyclohexane (1:1).[6] These bands appeared at R_f values 0.7, 0.8, and 0.9. These bands were scraped off individually and eluted with acetone. Evaporation of the acetone extracts provided the metabolites **2**, **5**, and **6** as confirmed by their mass spectra as follows: metabolite **2** (*m/e*) 183 (M^+), 169, 145, 117, 115; metabolite **5** (*m/e*) 170 (M^+), 129, 117, 115; and metabolite **6** (*m/e*) 146 (M^+), 115, and 90.

(B) **Chemical Studies.** (1) **Synthesis of 3.** Hydralazine, 4.0 g (0.022 mol), in 15 ml of 70% glycolic acid was refluxed for 24 hr. The cooled solution was washed with 250 ml of chloroform into a separatory funnel and the excess acid neutralized with saturated sodium bicarbonate solution. The liquids were separated from a solid by filtration and gave 2.4 g of product. The chloroform and water were separated, the chloroform was dried (K_2CO_3) and stripped off, and an additional 1.85 g of product precipitated upon addition of 50 ml of toluene: total yield 4.25 g (94%). Recrystallization from toluene gave mp 208–209°: ir spectrum (KBr) 3200 (OH), 2925, 2850 (CH), 1620, 1520, 1460 cm^{-1} (Ar); NMR spectrum (Me_2SO-d_6) singlet δ 9.0 (1 H), multiplet 8.7–7.7 (4 H), singlet 5.0 (2 H); uv spectrum (MeOH) sh 235, 240, 247, 264, 273 nm. Anal. ($C_{10}H_8N_4O$) C, H, N.

(2) **Oxidation of 3-Hydroxymethyl-*s*-triazolo[3,4-*a*]phthalazine.** The oxidation was attempted by three different methods in order to obtain *s*-triazolo[3,4-*a*]phthalazine-3-carboxylic acid.

(a) **Permanganate.** To 100 ml of distilled water in a 250-ml round-bottom flask was added 1 g (0.005 mol) of **3**. The pH of the solution was adjusted to 10.0 with Na_2CO_3 and a solution containing 0.80 g of $KMnO_4$ in 10 ml of distilled water was added. A brown precipitate of MnO_2 appeared immediately. The mixture was permitted to stir at room temperature for 2 hr. Excess permanganate was destroyed by addition of a small quantity of 2-propanol. The MnO_2 precipitate was filtered off.

The pH of the clear filtrate was adjusted to 3.0 with 10% sulfuric acid; no precipitate formed. The pH was readjusted to 10.0 and continuously extracted with chloroform, followed by evaporation to dryness. The solid residue was recrystallized from ethanol. The spectral properties indicate that this compound was *s*-triazolo[3,4-*a*]phthalazine: yield 0.5 g (59%); mass spectrum 170, 129, 117, and 115; NMR ($CDCl_3$) multiplet δ 7.80–8.51 (4 H), singlet 8.69 (1 H), singlet 8.82 (1 H); ir (KBr) 3090 (CH, Ar), 3040 (CH, Ar), 3000 (CH, aliphatic), 2950 (CH, aliphatic), 2900, 2220 cm^{-1}; uv spectrum (MeOH) [λ_{max} (log ϵ)] sh 232 (3.40), 237 (3.80), 243 (3.3), sh 262 (1.06), sh 272 (1.00), sh 282 nm (0.83).

(b) **Molecular Oxygen.** The alcohol (1 g) and 0.4 g of Pt black were suspended in 500 ml of distilled water in a 1000-ml round-bottom flask. The pH was adjusted to 6.7 with a phosphate buffer.[9] The mixture was warmed on a water bath and stirred vigorously. Oxygen gas was bubbled through two interconnected gas dispersion tubes into the reaction mixture for 6 hr. Only starting material was recovered.

(c) **Dichromate.** In a round-bottom flask, equipped with a mechanical stirrer, dropping funnel, and reflux condenser, was placed 1.0 g (0.005 mol) of the title compound. Potassium dichromate, 0.5 g (0.0017 mol) in 50 ml of water, was added. The flask was heated on a water bath while a mixture of 0.25 g of H_2SO_4 in 10 ml of water was added dropwise for a period of 1 hr. When complete, the reaction mixture was cooled and nearly neutralized with 50% NaOH and then completely neutralized with saturated sodium carbonate. The chromium hydroxide precipitate was filtered off and the clear filtrate was acidified with 10% H_2SO_4. No precipitate formed and the solution was readjusted to a pH of about 10 and extracted with chloroform. The spectral properties of the product obtained indicate it to be identical with **5** (vide supra).

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Preparation and Analgesic Activity of 3,6-Diacetylnormorphine and 6-Acetylnormorphine

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3,6-Diacetylnormorphine (norheroin) and 6-acetylnormorphine have been prepared in excellent yield through the 3,*N*-bis(*tert*-butoxycarbonyl) derivative of normorphine via acetylation and selective removal of protecting groups. This general procedure would be applicable to the preparation of various 3,6-diesters or 6-monoesters of normorphine. The analgesic potency of norheroin was found to be the same as that of 6-acetylnormorphine, about 0.05 that of heroin. The onset, peak, and duration of action of these compounds were nearly identical and comparable with morphine.

Only a few secondary amines show good in vivo analgesic potency. One of these, nordesomorphine, has an ED_{50} of 2.21[1] (compared with its parent, desomorphine, which has an ED_{50} of 0.09[1,2]). Normorphine and norcodeine do not display much in vivo analgesic activity when the common routes of administration are used,[3] presumably because they are too polar to pass through a "blood brain barrier", or their more rapid metabolism causes facile elimination from the animal.

Insofar as we are aware, only normorphine and norco-

deine have been shown to display analgesic receptor binding similar to their *N*-methyl parents in the guinea pig ileum (which is known to have receptors for analgesics and their antagonists).[4] Norketobemidone, norpethidine, noroxymorphone, normetazocine, and norbemidone have much less agonist potency on these receptors than their corresponding *N*-methyl derivatives.[4] Similar observations have been made[5] with normetazocine and 1,2,3,4,5,6-hexahydro-8-hydroxy-6-methyl-*N*-nor-3-benzazocine using an in vitro analgesic receptor (from rat brain) binding technique; that is, the nor compounds do not bind as well to the receptors as their *N*-methyl derivatives.

Thus, when it was brought to our attention that norheroin (**5**) had never been prepared[6] we thought that it, and 6-acetylnormorphine (**7**), would have considerable theoretical interest. It is conceivable that the acetyl groups might facilitate the transport of these compounds to the narcotic receptor sites in vivo, assuming that these acetyl groups are not metabolically cleaved during transport.

Chemistry. To prepare norheroin we used the carbo-*tert*-butoxy protecting group derived from *tert*-butyl azidoformate, which has previously been utilized to block the nitrogen in amino acids for peptide syntheses.[7] The use of this N-blocking reagent also allowed us to prepare 6-acetylnormorphine. The latter compound was prepared by Pohland and Sullivan[8] through the use of diethyl azodicarboxylate in 43% yield, but its biological activity was not recorded.

The use of *tert*-butyl azidoformate should generally allow the formation of any 3,6-diester or 6-monoester of normorphine which might be used for further N-derivatization if desired.

The starting material, normorphine (**1**), was prepared using our hydrazine N-demethylation procedure.[9] The reaction of **1** with *tert*-butyl azidoformate gave 3,*N*-bis(*tert*-butoxycarbonyl)normorphine (**2**) in 94% yield. Base hydrolysis of **2** gave the N-protected normorphine **3**, which could be acetylated by the usual procedure to the 3,6-diacetyl analog of the N-protected normorphine **4**. Acid cleavage of the *N*-*tert*-butoxycarbonyl group in the presence of acetic acid assured the survival of the 3,6-diacetyl groups and gave norheroin (**5**) in 88% overall yield (from **1**) (Scheme I).

Although we had hoped that the *tert*-butyl azidoformate might act like a chloroformate ester[10] and yield **2** or **3** directly, in fact it does not appear to quaternize morphine. Thus, the initial N-demethylation of morphine was necessary.

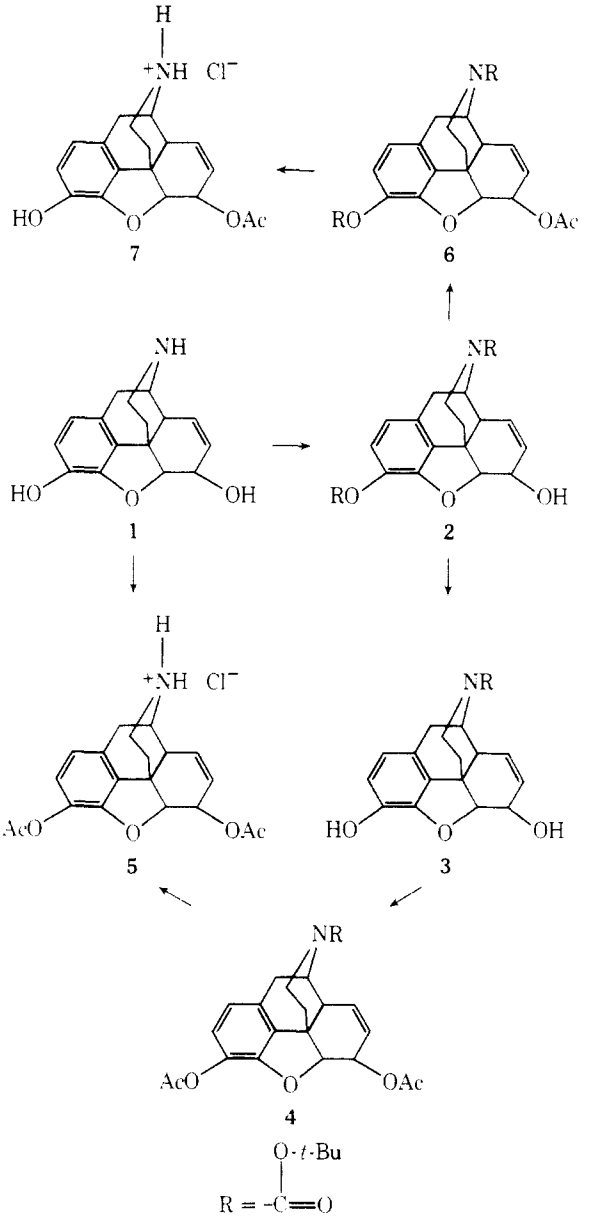
Compound **2** was directly acetylated to give the 3,N-protected 6-acetylnormorphine (**6**) which, again in the presence of acetic acid, could be acid cleaved to 6-acetylnormorphine (**7**) in an overall yield of 87% (from **1**). If **6** was treated with HCl gas in nitromethane solution, a basic nonphenolic product resulted which was not fully characterized but which appeared, by NMR, to be 6-acetyl-3-*tert*-butoxycarbonylnormorphine.

Norheroin could also be prepared directly from **1** by acetylation in HCl/Ac_2O–HOAc media, in 92% yield, at room temperature. However, this is not as general a route to the various esters of normorphine as the aforementioned procedure.

Both **5** and **7** were hydrolyzed back to **1** to assure the stereochemical integrity of the molecule.

Biological Results. In the Eddy hot-plate test for analgesic activity[2,11] (subcutaneous injection, mice, mg/kg) **5** had an ED_{50} = 10.1 (7.1–14.5) and **7** an ED_{50} = 10.6 (7.1–15.8).[12] Compounds **5** and **7** have a potency about 0.05 that of their *N*-methyl derivatives,[2,3,13] similar to the loss of potency shown by nordesomorphine. The onset, peak, and duration of action of these compounds (3.9, 38.6, and 139 for **5**, and 3.5, 40, and 135 for **7**) are very much alike and are more similar to morphine (4, 28, and 145) than to heroin (8, 11, and 69).

Scheme I



Conclusion

Secondary amines in this series appear to be too polar to allow facile transport to their receptor sites even though the hydrophilic hydroxyl groups were esterified.

It is interesting to note that the analgesic potencies of both **5** and **7** and their onset, peak, and duration of action were similar. It is possible that the 3-acetyl group in **5** is rapidly metabolized in vivo to give **7**. The 3-acetyl group in heroin is known to be labile in aqueous solution.[3] It is also possible that the observed in vivo potencies of **5** and **7** accurately reflect their binding to narcotic receptors, although this is, a priori, doubtful because of the known in vitro binding of normorphine to receptors.

Thus, it should be interesting to investigate the in vitro binding of **5** and **7** to the narcotic receptors. This work is presently being undertaken and will be reported in future communications.

Experimental Section

Melting points were determined in open capillary tubes using a Thomas-Hoover apparatus and are corrected. Microanalyses were performed by the Laboratory's Section on Microanalytical Services and Instrumentation and are within 0.3% of the calculated values. Ir (Perkin-Elmer 21), NMR (Varian A-60 or HA-100), and mass (Hitachi Perkin-Elmer RMU6E or Finnigan 1015D with Model 6000 data collection system) spectra were consistent with the expected structure.

3,*N*-Bis(*tert*-butoxycarbonyl)normorphine (2). To dry dimethylformamide (100 ml) was added normorphine (1)·$2H_2O$ (3.90 g, 12.7 mmol),[9] anhydrous K_2CO_3 10.0 g, 72.2 mmol), and *tert*-butyl azidoformate (5.0 g, 34.8 mmol). The mixture was stirred overnight at 25° and filtered and the inorganic material washed well with $CHCl_3$. The filtrate and washings were evaporated and the residue was dissolved in Et_2O (200 ml) and H_2O (25 ml). The Et_2O was washed successively with cold 10% citric acid, cold 0.1 *N* KOH, and brine and then dried ($MgSO_4$). The residue was crystallized from EtOAc–hexane to give 5.66 g (94%) of nearly pure **2** in two crops. Recrystallization from isopropyl ether gave **2**, mp 169.5–171° dec. Anal. ($C_{26}H_{33}NO_7$) C, H, N.

***N*-*tert*-Butoxycarbonylnormorphine (3).** Crude **2** (ca. 6 g, from 3.9 g of 1) was treated with KOH (1.80 g), in MeOH (100 ml), heated to 55° (10 min), and allowed to cool during 1 hr. The solution was evaporated, Et_2O (200 ml) and H_2O (30 ml) were added, and the mixture was acidified with 10% citric acid. The oil that separated was shaken into the Et_2O. The Et_2O was washed with H_2O (30 ml), 10% $NaHCO_3$ (2 × 30 ml), and brine (30 ml), dried ($MgSO_4$), and evaporated to a foam (4.84 g). Crystallization from *i*-PrOH–hexane gave **3**·*i*-PrOH, 5.02 g (91.5%), in two crops. Recrystallization from *i*-PrOH gave pure **3**·*i*-PrOH, mp 172–174° dec. Anal. ($C_{24}H_{33}NO_6$) C, H, N.

***N*-*tert*-Butoxycarbonyl-3,6-diacetylnormorphine (4).** To dry pyridine (50 ml) was added **3** (6.03 g, 13.95 mmol) and Ac_2O (11 ml, 117.5 mmol). The solution was heated on the steam cone 0.5 hr and cooled and MeOH (15 ml) added. After evaporation to a syrup in vacuo (<50°), the residue was dissolved in Et_2O (150 ml) and washed successively with H_2O (2 × 50 ml), 10% citric acid (2 × 50 ml), 10% $NaHCO_3$ (25 ml), and brine (25 ml). The resulting Et_2O solution was dried ($MgSO_4$) and evaporated to give **4** as a foam (6.54 g) which resisted crystallization from a variety of solvents but which was nearly homogenous on TLC.

Norheroin (5) Hydrochloride. Method A. The noncrystalline **4** (6.54 g, 13.95 mmol) was dissolved in HOAc (20 ml) and added slowly to a saturated solution of HCl gas in HOAc (40 ml) at 25°, while passing HCl gas through the solution. After standing 15 min, the solution was concentrated, in vacuo, to a light syrup that contained crystalline material. Me_2CO (60 ml) was added and crystallization proceeded rapidly. The solid was filtered, washed with Me_2CO and then Et_2O, and dried to give 5.24 g (96%) of **5**·HCl, mp 257–258.5° dec. Recrystallization from HOAc–Me_2CO gave stout prisms, mp 257–258° dec. Anal. ($C_{20}H_{22}NO_5Cl$) C, H, N.

Method B. 1·$2H_2O$ (500 mg, 1.63 mmol) was dissolved in HOAc (30 ml) and the solution was saturated with HCl gas at 25°. Ac_2O (2.5 ml, 26.4 mmol) was added and the solution allowed to stand overnight at 25°. The solution was evaporated and the residue triturated with Me_2CO. The material crystallized almost immediately and was filtered, washed with Me_2CO, and dried to give **5**·HCl (585 mg, 92%). Recrystallization from HOAc (3 ml) gave **5**·HCl, mp 256–257.5° dec, which was identical with norheroin prepared as described in method A.

When this acetylation mixture was refluxed overnight, a large amount of nonbasic material was isolated.

6-Acetyl-3,*N*-bis(*tert*-butoxycarbonyl)normorphine (6). Acetylation of **2** (4.52 g, 9.6 mmol) was carried out using Ac_2O (5 ml) in dry pyridine (50 ml) as described for the acetylation of **3** to yield 5.08 g of **6**, a foam, that resisted crystallization from a variety of solvents but which was essentially homogeneous on TLC.

6-Acetylnormorphine (7) Hydrochloride. The 5.08 g (9.6 mmol) of **6** in glacial HOAc (20 ml) was added, during 5 min, to a saturated solution of HCl gas in glacial HOAc (30 ml). A stream of HCl gas was continuously passed through the solution during the addition. The solution (after 5 min) was evaporated, in vacuo, to a crystalline mass. This residue was recrystallized from hot HOAc to give 3.13 g (93.5%) of **7** in two crops. Recrystallization gave needles, mp 317–318° dec (lit.[8] mp 313–315°).

Normorphine (1) from Norheroin (5) Hydrochloride and 6-Acetylnormorphine (7) Hydrochloride. Either **5** or **7**·HCl (200 mg) was refluxed several minutes with 1 *N* NaOH (4 ml) until the solution had become homogeneous. It was allowed to cool during 0.5 hr, acidified with 12 *N* HCl, and evaporated to a crystalline solid. The solid was dissolved in a small amount of hot H_2O made alkaline with 12 *M* NH_4OH, and the crystalline material that separated was filtered, washed, and dried to give 1 (base). The 1·base obtained from norheroin by methods A and B, and 6-acetylnormorphine had mp 277–279° dec, 277.5–279.5° dec, and 277–279.5° dec, respectively (lit.[14] mp 276–277°), and each gave a hydrochloride salt identical with an authentic sample.

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