Experimental Section

The melting point was determined in a "Mel-Temp" apparatus and is uncorrected. Elemental analyses, indicated by symbols of the elements, were within ±0.2% of the theoretical values. The ir spectrum was consistent with the proposed structure.

4-[3-(2-Trifluoromethyl)phenothiazin-10-yl]propyl-1-piperazineethyl *N*-(1-Adamantyl)carbamate Dihydrochloride (Fluphenazine Adamantylcarbamate, 1). A mixture of fluphenazine[9] [4-[3-(2-trifluoromethyl)phenothiazin-10-yl]-propyl-1-piperazineethanol, 3.06 g, 7 mmol], 1-adamantanyl isocyanate[10] (1.25 g, 7 mmol), and 150 ml of xylene was stirred and heated at reflux for 36 h. The reaction mixture was allowed to cool to room temperature and filtered to remove suspended solids, and the xylene was removed in vacuo. The residue, dissolved in the minimum of benzene, was placed on a silica gel column (2.5 × 35.5 cm). The carbamate was then eluted with $CHCl_3$. The elution was followed by silica gel TLC ($CHCl_3$–EtOH, 18:1). The fractions containing the carbamate were combined and the $CHCl_3$ was removed in vacuo leaving 2.13 ml of yellow oil. A solution of the oil in absolute EtOH was added to ether saturated with HCl. The resulting precipitate was filtered and dried to give 1.64 g of 1 as a white powdered dihydrochloride salt: mp 220–223° dec; yield, 42% based on recovered fluphenazine dihydrochloride (0.7 g). Anal. ($C_{33}H_{43}Cl_2F_3N_4O_2S$) C, H, N.

Spontaneous Locomotor Activity. Eighteen male Sprague–Dawley rats housed individually were divided into three groups. Two groups of animals were injected subcutaneously with either 0.1 mmol of 1·2HCl (70 mg/kg) or fluphenazine dihydrochloride (50 mg/kg) in sesame oil; the third group received the vehicle only (sesame oil). After the initial injections, 2 mg/kg of *d*-amphetamine in saline was administered on the first, second, fourth, and seventh day to all three groups of animals, and the motor activity of the rats was measured immediately after each injection for 30 min with an electronic motility meter (Motron-Produkter, Stockholm, Sweden, Model 40Fc). Results are expressed as the percentage counts of the experimental group in relationship to the vehicle control group. Each animal's motility was measured at the same time of day to minimize the diurnal effect.

Shock Avoidance Responses. A shuttle box was used for behavioral testing. Each trial began with the onset of light and tone (conditioned stimulus) followed in 5 s by an electronic shock (unconditioned stimulus). The electric shock of 0.8 mA (ac, 60 cycle) was delivered through a scrambler to the grid floor, while the light and tone continued without interruption. A crossing by the animal from one end to the other tilted the box and terminated the shock; crossing before the onset of shock constituted the avoidance response. The onset of light and tone was on a variable interval 60-s schedule. Termination of the trial also resulted if the animal did not respond to the shock within 30 s of its onset.

Fifty rats in groups of five were used for the study. Each group of animals were pretreated subcutaneously with 0.1 mmol of 1·2HCl or fluphenazine dihydrochloride 1, 2, 4, or 7 days prior to the administration of 2 mg/kg of *d*-amphetamine sulfate. A control group of animals received only amphetamine with no pretreatment with the test drugs. The animal was placed on one end of the shuttle box 30 min after the injection of amphetamine. None of the animals were used for more than one session. All sessions were conducted at the same time of day and counter-balanced according to pretreatment time. The number of avoidance responses in a total of 50 trials was recorded, and the percentage of avoidance responses for the experimental group in relationship to the amphetamine control group was calculated.

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2-Azabicyclo[2.2.2]octane Analogues of the Prodine Analgetics

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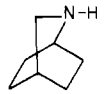
The synthesis and analgetic activity of analogues of prodine-type analgetics in which the conformation of the piperidine ring is restricted in the boat form using the 2-azabicyclo[2.2.2]octane nucleus are reported. One of these analogues, 2-methyl-6-*trans*-phenyl-6-*cis*-propionoxy-2-azabicyclo[2.2.2]octane (**3**), showed significant analgetic activity (ED_{50} = 3.1 mg/kg).

Conformational requirements at the analgetic receptor have been studied extensively and theories regarding the nature of the receptor have been postulated. By using the morphine structure as a mirror, Beckett and Casy[1] published a detailed description of the analgetic receptor. An analysis of the meperidine molecule in relation to this receptor led to the conclusion that an axial-phenyl arrangement was necessary for this drug to interact with the receptor. These postulations have stimulated a great deal of research and newer theories regarding the nature of the analgetic receptor have been developed.[2] The use of rigid drug models to examine the stereochemical requirements in 4-phenylpiperidine analgetics has led to conflicting results concerning the conformational requirements for the phenyl group.[3,4] Casy[5] has used the results of the studies on the preferred conformation of various piperidine analgetics in water solution to predict the conformational requirements for analgetic activity. It was concluded that the preferred conformation of the piperidine ring in analgetics of this class is the skew-boat form. Further examination of the prodine-type derivatives of 1,2,5-trimethylpiperidine indicated that the isomers best able to

assume this skew-boat form are the most active analgetics.

The 2-azabicyclo[2.2.2]octane ring (1) system can be considered a "conformationally restricted" analogue of the boat conformation of the piperidine ring. It can further be considered as an analogue of *cis*-1,2,5-trimethylpiperidine in which the 2- and 5-methyl groups are connected through a carbon–carbon bond. Thus, the 2- and 5-methyl groups are fixed in a cis-diaxial relationship. In the 4-phenyl-4-propionoxy derivatives of this series (promedols) the most potent isomer (equipotent with betaprodine) has a cis relationship of the 2-methyl, 4-phenyl, and 5-methyl groups.[6] Thus, the target compounds of this study (**2–5**) can be considered as conformationally restricted boat conformers of the most potent promedol



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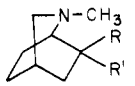
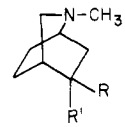
analogues. Isomers **2** and **3** represent restricted boat forms of 3-substituted piperidines while isomers **4** and **5** represent derivatives of the more potent 4-substituted piperidines. This positional variation should allow an examination of the interatomic distance requirements within the boat conformation of piperidine analgetics. Additionally in isomers **2** and **4** the phenyl group is cis to the nitrogen bridge while the phenyl group is trans to this bridge in **3** and **5**. This allows an opportunity to examine configurational requirements of the propionoxy and phenyl groups for receptor interactions.



2, R = Ph; R′ = O_2C-C_2H_5
3, R = O_2C-C_2H_5; R′ = Ph



4, R = Ph; R′ = O_2C-C_2H_5
5, R = O_2C-C_2H_5; R′ = Ph

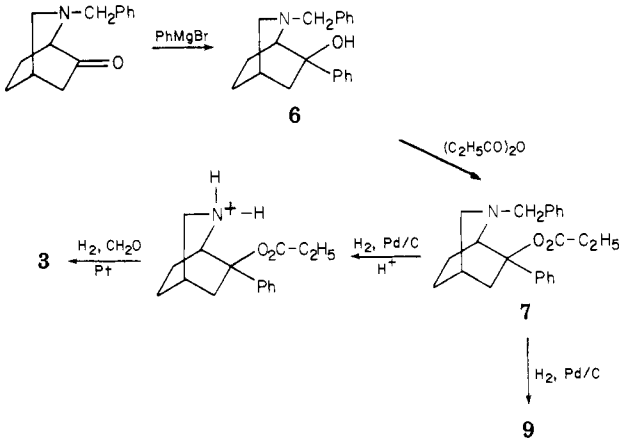
Chemistry. We have recently reported[7] the results of our studies regarding the stereoselective addition of phenylmagnesium bromide to various *N*-substituted 2-azabicyclo[2.2.2]octan-5- and -6-ones. These additions proceeded to give only one phenyl alcohol in each case examined, the direction of attack depending upon the nature of the substituent attached to nitrogen. We were unable to isolate or to detect the phenyl alcohol precursor to **5**.

Analogues **2** and **4** were prepared by treating the phenyl alcohol obtained from phenylmagnesium bromide addition to the *N*-methyl 5- or 6-ketone with propionic anhydride. Analogue **3** was more difficult to obtain (Scheme I). Treating *N*-benzyl-2-azabicyclo[2.2.2]octan-6-one with phenylmagnesium bromide gave only **6**. Since we had previously found that debenzylation of **6** followed by reductive alkylation gave only tricyclic products,[7] **6** was acylated with propionic anhydride prior to debenzylation. Debenzylation of **7** under strongly acidic conditions followed by reductive alkylation with formaldehyde under neutral conditions gave **3**. Debenzylation of **7** under neutral conditions gave only the product resulting from O to N migration of the propionyl group (**9**).

Results and Discussion

The analgetic activity of **2–4** and **7** was determined by the D'Armour–Smith tail flick method.[8] Compounds **2**, **4**, and **7** possessed no analgetic activity at dose levels of 100 mg/kg ip. However, **3** was found to possess analgetic activity with an ED_{50} of 3.1 mg/kg (1.9–5.0 mg/kg).

Scheme I



Compound **3** was also very toxic possessing an LD_{50} of 12.3 mg/kg (9.3–16.2 mg/kg). Although **3** is less potent than alphaprodine [ED_{50} = 1.4 mg/kg (1.1–1.6 mg/kg)] and more toxic (LD_{50} = 22 mg/kg[9]) its activity is interesting since one can consider **3** to be an analogue of a 3-substituted piperidine. The unavailability of isomer **5** at the present time would seem to make generalizations regarding the importance of boat conformations in piperidine-type analgetics unwarranted. Efforts to synthesize **5** by other routes are presently under investigation.

Experimental Section

General. Melting points were determined on a Thomas-Hoover Unimelt or Mel-Temp apparatus and are corrected. Infrared spectra were recorded on a Perkin-Elmer spectrometer and NMR spectra on a Jeolco C-60-HL spectrometer (all values are reported in parts per million from Me_4Si as internal standard).

Elemental analyses were performed by Chemalytics, Inc., Tempe, Ariz. Where analyses are indicated only by symbols of the elements, analytical results obtained for those elements were within ±0.4% of the theoretical values.

2-Methyl-6-*cis*-phenyl-6-*trans*-propionoxy-2-azabicyclo[2.2.2]octane (2). A solution of 2-methyl-6-*cis*-phenyl-6-hydroxy-2-azabicyclo[2.2.2]octane[7] (1.5 g, 0.007 mol) and 10 ml of propionic anhydride in 20 ml of dry C_6H_6 was added to a 100-ml three-necked flask equipped with a mechanical stirrer and a reflux condenser with a drying tube and refluxed for 96 h. The solvent was evaporated and the residue treated with 10% K_2CO_3 and then extracted with $CHCl_3$ (3 × 50 ml). The extracts were combined, dried ($MgSO_4$), and evaporated to yield a red oil (1.3 g). The oil was chromatographed on 20 g of silica gel using Et_2O–petroleum ether (1:9) as eluent to yield the desired ester (0.4 g, 20%): ir (CCl_4) 1740 cm^{-1} (C=O, ester); NMR ($CDCl_3$) δ 1.0 (t, 3, CH_2CH_3), 1.35–3.0 [broad signals, 15, bicyclic envelope, CH_2CH_3, and NCH_3 (s, 2.23)], 7.2–8.0 (m, 5, aromatic). The picrate was prepared in the normal manner: mp 177–179°. Anal. ($C_{23}H_{26}N_4O_9$) C, H, N.

2-Benzyl-6-*trans*-phenyl-6-*cis*-propionoxy-2-azabicyclo[2.2.2]octane (7). A solution of 2-benzyl-6-*cis*-hydroxy-6-*trans*-phenyl-2-azabicyclo[2.2.2]octane[7] (7.0 g, 0.024 mol) and 25 ml of propionic anhydride in 50 ml of dry C_6H_6 was refluxed for 24 h and then cooled to room temperature and the solvent was evaporated. The resulting oil was treated with 10% K_2CO_3 and extracted with $CHCl_3$ (3 × 50 ml). The $CHCl_3$ extracts were combined, dried ($MgSO_4$), and evaporated to yield an orange oil (8.3 g). The oil was chromatographed on 70 g of silica gel using Et_2O–petroleum ether (1:9) as eluent to give a clear oil (4.6 g, 55%): $n^{22}D$ 1.5616; ir (CCl_4) 1745 cm^{-1} (C=O, ester); NMR ($CDCl_3$) δ 0.9–3.2 [broad signals, 15, bicyclic envelope, CH_2CH_3 (t, 1.1), and CH_2CH_3 (q, 2.4)], 4.0 (s, 2, NCH_2Ar), 7.2–7.8 (m, 10, aromatic). Anal. ($C_{23}H_{27}NO_2$) C, H, N.

6-*cis*-Hydroxy-6-*trans*-phenyl-2-propionyl-2-azabicyclo[2.2.2]octane (9). A solution of **7** (3.0 g, 0.009 mol) in 75 ml of EtOH was added to a Parr flask and hydrogenated (3.15 kg/cm^2) over 0.3 g of 10% Pd/C for 4 h. The catalyst was removed by

filtration through Celite and the EtOH evaporated to yield an oil (2.6 g). The oil was taken up in $CHCl_3$ (100 ml) and the resulting solution washed with 10% HCl (3 × 30 ml), dried ($MgSO_4$), and evaporated to yield a clear oil which solidified on standing. The solid was recrystallized (C_6H_6–*n*-hexane) to yield white crystals (2.0 g, 83%): mp 108–110°; ir ($CHCl_3$) 3600 (OH), 1660 cm^{-1} (C=O, amide); NMR ($CDCl_3$) δ 1.17 (t, 3, CH_2CH_3), 1.5–3.0 (broad signals, 9, bicyclic envelope and CH_2CH_3), 3.4–3.9 (broad signals, 4, OH, H at C_1 and H at C_3), 7.3–7.9 (m, 5, aromatic). Anal. ($C_{16}H_{21}NO_2$) C, H, N.

The HCl extracts were combined, made basic with K_2CO_3, and extracted with $CHCl_3$ (2 × 40 ml). The $CHCl_3$ was dried ($MgSO_4$) and evaporated to yield no basic products.

2-Methyl-6-*trans*-phenyl-6-*cis*-propionoxy-2-azabicyclo-[2.2.2]octane (3). A solution of 7 (2.0 g, 0.005 mol) in 50 ml of EtOH was added to a Parr flask and the pH adjusted to approximately 2 with concentrated HCl. The solution was then hydrogenated (3.15 kg/cm^2) over 0.3 g of 10% Pd/C for 4 h and filtered through Celite. To the filtrate was added 2 ml of 37% CH_2O and the pH was readjusted to approximately 7 with 10% NaOH. The resulting solution was hydrogenated (3.15 kg/cm^2) over 0.3 g of 10% Pd/C for 4 h and then filtered through Celite. The filtrate was evaporated and the residue taken up in 50 ml of $CHCl_3$. The $CHCl_3$ mixture was extracted with 10% HCl (3 × 30 ml) and the extracts were combined and made basic with K_2CO_3 and extracted with $CHCl_3$ (3 × 50 ml). The $CHCl_3$ was combined, dried ($MgSO_4$), and evaporated to yield a clear oil (1.2 g). The oil was chromatographed on 8.0 g of silica gel using Et_2O–petroleum ether (1:4) as eluent to yield the desired ester (0.9 g, 64%) as a clear oil: ir (CCl_4) 1745 cm^{-1} (C=O, ester); NMR ($CDCl_3$) δ 1.03 (t, 3, CH_2CH_3), 1.2–3.4 [broad signals, 15, bicyclic envelope, CH_2CH_3, and NCH_3 (s, 2.65)], 7.3–7.8 (m, 5, aromatic); *m/e* calculated 273, found 273. Anal. ($C_{17}H_{23}NO_2$) C, H, N.

2-Methyl-5-*cis*-phenyl-5-*trans*-propionoxy-2-azabicyclo-[2.2.2]octane (4). A solution of 2-methyl-5-*cis*-phenyl-5-*trans*-hydroxy-2-azabicyclo[2.2.2]octane[7] (0.8 g, 0.004 mol) and 10 ml of propionic anhydride in 20 ml of dry C_6H_6 was refluxed for 96 h. The C_6H_6 was evaporated and the residue treated with 10% K_2CO_3 and extracted with $CHCl_3$ (3 × 50 ml). The extracts were combined, dried ($MgSO_4$), and evaporated to yield a clear oil (0.6 g, 60%): ir ($CHCl_3$) 1735 cm^{-1} (C=O, ester); NMR ($CDCl_3$) δ 1.0 (t, 3, CH_2CH_3), 1.2–3.1 [broad signals, 15, bicyclic envelope, CH_2CH_3, and NCH_3 (s, 2.28)], 7.3–7.9 (m, 5, aromatic). The picrate was made in the normal manner: mp 171–174°. Anal. ($C_{23}H_{26}N_4O_9$) C, H, N.

Pharmacological Methods. Analgetic potency was determined by the D'Armour–Smith tail-flick method[8] using male albino rats weighing 100–130 g. Each subject served as its own control and was used only once. Three control response times were determined at 15-min intervals. Groups of five rats were used for each dose of drug. After control responses were determined, a solution of the hydrochloride salt of the drug was administered ip using normal saline as the vehicle. The response was determined at 15-min intervals postinjection and a reaction time greater than the average control value was considered to indicate a state of analgesia. The ED_{50} for this study was defined as that dose of the drug which, in 50% of the animals tested, increased the reaction time by 50% at 15 min postinjection. The LD_{50} (ip) of **3** was determined by the method of Litchfield and Wilcoxin.[10] Numbers in parentheses following ED_{50} and LD_{50} data represent 95% confidence limits.

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Biological Activity of C-Terminal Partial Sequences of Substance P

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Substance P (Arg-Pro-Lys-Pro-Gln-Gln-Phe-Phe-Gly-Leu-Met-NH_2) and the C-terminal partial sequences down to the tripeptide were synthesized by a solid-phase method. These peptides were assayed for vasodilator, spasmogenic, and venoconstrictor properties using three preparations, viz. the hind limb blood flow of the dog, isolated guinea pig ileum, and the isolated rabbit ear vein. The tripeptide and tetrapeptide possessed weak vasodilator properties only but no activity was detected on the other less sensitive preparations. The pentapeptide produced appreciable spasmogenic and vasoactive effects. Sequences of six or more C-terminal amino acids were able to elicit activity at comparable doses to that of the parent endecapeptide; however, the activity did not increase regularly with the chain length. In each assay preparation the octapeptide was the most potent peptide. It was twice as potent as substance P on a molar basis in the guinea pig ileum but the enhancement of activity beyond that of substance P was less pronounced in the vascular preparations.

The amino acid sequence of substance P was determined by Chang et al.[1] to be Arg-Pro-Lys-Pro-Gln-Gln-Phe-Phe-Gly-Leu-Met-NH_2. This peptide has been synthesized by the solid-phase method[2,3] and was found to produce identical pharmacological effects to the naturally occurring peptide.

In order to study the relationship between the biological activity and chain length, the C-terminal partial sequences of substance P down to the tripeptide were synthesized and assayed for spasmogenic, vasodilator, and venoconstrictor properties.

Synthesis and Purification. Using techniques of a similar nature to those employed by Fisher et al.[3] we have also synthesized substance P on a benzhydrylamine resin[4] (0.06 mmol of amine/g) obtained from I.C.I., Australia. The carboxyl-terminal partial sequences were obtained by