Anal. Calcd. for $C_{13}H_5F_8O$: C, 66.67; H, 2.15; F, 24.34. Found: C, 66.40; H, 2.39; F, 24.16.

Acknowledgment.—We thank the Crown-Zellerbach Corp. for a generous supply of dimethyl sulfoxide.[8] We also thank Marylin E. Sanford for preparation of some of the starting materials, and Carol-Ann Cole for running infrared spectra.

The Synthesis of DL-*p*-(Hydroxymethyl)phenylalanine

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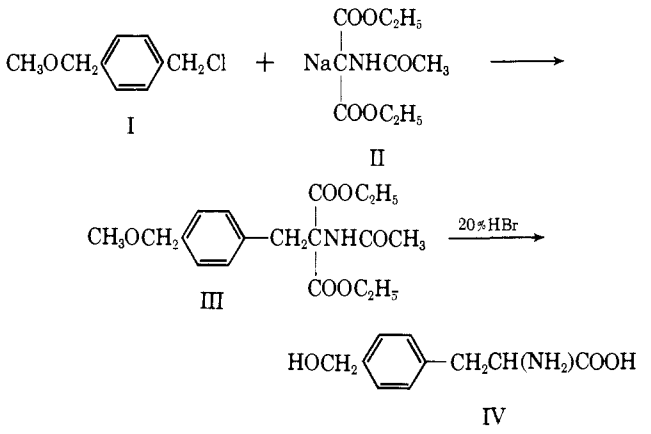
In the course of investigation of the metabolism of certain aromatic amino acids, a study of the properties of *p*-(hydroxymethyl)phenylalanine became of interest. A method for synthesis of this substance was devised (I–IV), patterned after the procedure for preparation of analogous aromatic amino acids used by Herr, *et al.*[2]

Phenylalanine hydroxylase preparations from liver, which form tyrosine from phenylalanine, did not convert *p*-(hydroxymethyl)phenylalanine to tyrosine under the conditions of Udenfriend and Cooper[3] or those of Kaufman.[4] Furthermore, *p*-(hydroxymethyl)phenylalanine in a 4×10^{-3} *M* concentration did not inhibit the oxidation of phenylalanine to tyrosine by the enzyme preparations from rat liver.

Experimental[5]

Diethyl 2-Acetamido-2-[(*p*-methoxymethyl)benzyl]malonate (III).—A solution of 9.8 g. (45 mmoles) of diethyl acetamidomalonate (Calbiochem, Los Angeles, Calif.; recrystallized from toluene; b.p. 96–98°, lit.[6] 96.5–98°) in 35 ml. of absolute ethanol



was added with stirring to a solution of 1.03 g. (44 mg.-atoms) of sodium in 35 ml. of absolute alcohol. *p*-(Methoxymethyl)-benzyl chloride[7] (41 mmoles)[8] was then added, and the mixture was boiled under reflux for 4 hr. The NaCl precipitate was removed by filtration and the filtrate was treated with 100 ml. of water. The oil which formed was separated from the mixture and washed for 1 hr. by suspending it in 650 ml. of water with vigorous stirring to remove residual diethyl acetamidomalonate. The oil was dried by dissolving it in 25 ml. of absolute alcohol and by removing alcohol and water from the solution in a flash evaporator. This treatment was repeated three times. The water-free oil was suspended in 400 ml. of boiling petroleum ether (b.p. 65–110°). The hot solution was filtered to remove a small undissolved residue and was then allowed to stand at room temperature. The white crystals formed were collected by filtration: 9.3 g., m.p. 74–79°. The material was recrystallized from 400 ml. of cyclohexane: yield 8.5 g. (24 mmoles), m.p. 83–85°. Two additional recrystallizations from cyclohexane afforded the analytical sample.

Anal. Calcd. for $C_{18}H_{25}NO_6$: C, 61.5; H, 7.2; N, 4.0; alkoxyl as OCH_3, 26.5. Found: C, 61.5; H, 7.0; N, 3.9; alkoxyl as OCH_3, 26.4.

DL-*p*-(Hydroxymethyl)phenylalanine (IV).—A mixture of diethyl 2-acetamido-2-[(*p*-methoxymethyl)benzyl]malonate (2 g., 5.7 mmoles) and 20% HBr (24 ml.) was heated under reflux for 8 hr. The solution was concentrated to a small volume; the crystalline precipitate formed was collected by filtration and dried at 50° under vacuum: yield 1.6 g., m.p. 198–205° dec.

The crude hydrobromide (1.6 g.) was dissolved in water (200 ml.) and passed through a column (2 × 2 cm.) of the anion-exchange resin Dowex AG 3 X4 (OH⁻ form) to remove HBr. The effluent, approximately pH 6, was concentrated to dryness. The residue was dissolved in 10 ml. of water, treated with 200 mg. of Darco G 60 for 10 min. on a steam bath, and filtered. The residual charcoal was washed with 10 ml. of hot water. The filtrates were combined and evaporated to dryness under reduced pressure. The residue was dissolved in 4 ml. of hot water, 10 ml. of ethanol was added to the heated solution, and the mixture was allowed to stand at 5°. The white precipitate which formed was collected by filtration and dried under vacuum: yield 640 mg. (58%), m.p. 231–237° dec.

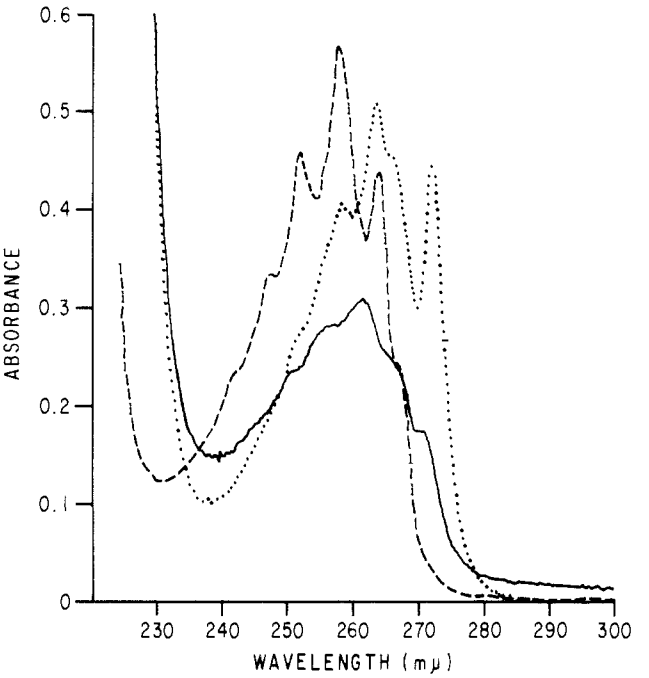


Figure 1.—Ultraviolet spectra of: *p*-(hydroxymethyl)phenylalanine, 0.96×10^{-3} *M*, ———; *p*-methylphenylalanine, 1.4×10^{-3} *M*, ... ; and phenylalanine, 3.0×10^{-3} *M*, – – – . All compounds were dissolved in water.

(1) (a) This investigation was aided by research grants from the National Institute of Arthritis and Metabolic Diseases, National Institutes of Health, U. S. Public Health Service, and a Research Career Award (GM-K6-1551) from the United States Public Health Service. (b) The competent technical assistance of Mr. T. Aogaichi is gratefully acknowledged.

(2) R. R. Herr, T. Enkoji, and J. P. Bailey, *J. Am. Chem. Soc.*, **79**, 4229 (1957).

(3) S. Udenfriend and J. R. Cooper, *J. Biol. Chem.*, **194**, 503 (1952); **196**, 227 (1952).

(4) S. Kaufman, "Methods in Enzymology," Vol. V, S. P. Colowick and N. O. Kaplan, Ed., Academic Press Inc., New York, N. Y., 1962, p. 809.

(5) All melting points are uncorrected. Microanalyses were performed by Australian Microanalytical Service, Parkville, Victoria, Australia. All operations at the boiling points of liquids were done at the prevailing atmospheric pressure (av. 640 mm.).

(6) S. G. Cohen and L. H. Klee, *J. Am. Chem. Soc.*, **82**, 6038 (1960).

(7) R. Quelet, *Bull. soc. chim. France*, **53**, 222 (1933).

(8) The preparation of *p*-(methoxymethyl)benzyl chloride of Quelet[7] also contains the dimethyl ether of *p*-xylene-α,α'-diol in the same fraction (b.p. 120–130° at 15 mm.). The amount of *p*-(methoxymethyl)benzyl chloride in the mixture was determined by estimation of the chlorine content.

Anal. Calcd. for $C_{10}H_{13}NO_3$: C, 61.5; H, 6.7; N, 7.2; O, 24.6; alkoxyl as OCH_3, 0.0. Found: C, 61.0; H, 6.8; N, 7.1; O, 24.8; alkoxyl as OCH_3, 0.0.

The material was assayed by the colorimetric ninhydrin method of Troll and Cannan[9] using phenylalanine as a standard. If one assumes that the color yield of *p*-(hydroxymethyl)phenylalanine is the same as that of phenylalanine, the product is calculated to be 98% pure.

The compound possesses the following spectral characteristics in water solution: λ_{min} 280, 239 mμ (ε 20, 141); λ_{max} 270 (sh), 265 (sh), 262, 257 (sh), 252, below 239 mμ (ε 160, 235, 292, 263, 217, end absorption). Its ultraviolet absorption spectrum is distinct from that of DL-phenylalanine and DL-*p*-methylphenylalanine (Figure 1). Its infrared spectrum[10] (Nujol mull) showed bands at 860, 1015, 1160, 1210, 1310, 1340, 1410 cm.$^{-1}$ in the 800–1500-cm.$^{-1}$ region. The peak at 1015 cm.$^{-1}$ is not found in spectra of DL-phenylalanine or DL-*p*-methylphenylalanine.

Paper chromatography of the compound in the system[11] 1-butanol–acetic acid–water, 200:30:75 (Whatman 3 MM paper ascending for 15 hr.), revealed a single ninhydrin staining spot (R_f 0.34). The migration of the compound in this system differs markedly from that of authentic DL-phenylalanine (R_f 0.52) or DL-*p*-methylphenylalanine (R_f 0.64).

(9) W. Troll and R. K. Cannan, *J. Biol. Chem.*, **200**, 803 (1953).

(10) These measurements were performed by Dr. C. H. Winestock.

(11) "Specifications and Criteria for Biochemical Compounds," Publication 719, National Academy of Sciences, National Research Council, Washington, D. C., 1960, p. AAi.

Coupling Reactions in the Synthesis of Amitriptyline

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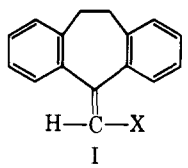
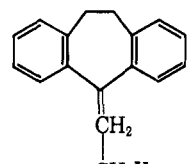
The readily available bromo compounds I and II (X = Br) provide new and direct routes to amitriptyline systems in fair to moderate yields by direct coupling of their Grignard derivatives with β-dimethylaminoethyl chloride and isobutoxymethyldimethylamine, respectively. Hydrocarbon dimers are by-products in each case.



I



II

Experimental

5-Bromomethylene-5H-dibenzo[*a,d*]-10,11-dihydrocycloheptene (I, X = Br).—A solution of 1.85 g. of 5-methylene-5H-dibenzo[*a,d*]-10,11-dihydrocycloheptene (I, X = H)[1] in 15 ml. of chloroform was brominated by dropwise addition of 1.45 g. of bromine dissolved in 9 ml. of $CHCl_3$. Bromine absorption was instantaneous, accompanied by copious evolution of HBr. The reaction mixture was evaporated to dryness and crystallized from petroleum ether (b.p. 30–60°); yield 2.5 g., m.p. 70–72°. A sample crystallized for analysis melted at 73–75°.

Anal. Calcd. for $C_{16}H_{13}Br$: C, 67.37; H, 4.56; Br, 28.08. Found: C, 66.98; H, 4.46; Br, 28.17.

5-(3-Dimethylaminopropylidene)-5H-dibenzo[*a,d*]-10,11-dihydrocycloheptene Hydrochloride (Amitriptyline; I, X = $CH_2CH_2N(CH_3)_2$·HCl). A. From I, X = Br.—The corresponding Grignard reagent was prepared by adding slowly under nitrogen a solution of 2.00 g. of the 5-bromomethylene compound I (X = Br) in 10 ml. of tetrahydrofuran to a stirred mixture of 170 mg. of magnesium turnings and 2 ml. of tetrahydrofuran. The reaction was started by initially adding a trace of methyl iodide. When addition of I (X = Br) was complete, the dark brown solution was refluxed for 90 min. It was then cooled to room temperature and a solution of 760 mg. of β-dimethylaminoethyl chloride[2] was added. The stirred mixture was refluxed for 2 hr., kept at room temperature overnight, and worked up by cooling to 10° and adding 20 ml. of 10% aqueous NH_4Cl and 10 ml. of 2.5 *N* HCl. Following ether extraction the acidic layer was made basic with 2.5 *N* NaOH and extracted with ether. From the latter extract 230 mg. of basic material was obtained which on treatment with HCl in ether and crystallization from ether gave 170 mg. (20% based on recovered I, X = H; see below) of amitriptyline (I, X = $CH_2CH_2N(CH_3)_2$·HCl), m.p. 175–190°, raised to 192–194° on one recrystallization from ether–2-propanol. On admixture with an authentic sample of m.p. 192–194°, there was no melting point depression and the respective infrared spectra were identical.

The neutral ether extract (1.150 g.) on alumina chromatography gave 810 mg. (56%) of reusable 5-methylene compound I (X = H), m.p. 60–63°, and 25 mg. of the dimer I [X = ( )$_2$] crystallized from acetone; m.p. 270–271°, $\lambda_{max}^{dioxane}$ 318 mμ (ε 33,000).

Anal. Calcd. for $C_{32}H_{26}$: C, 93.61; H, 6.38; mol. wt., 410. Found: C, 93.27; H, 6.33; mol. wt., 398.[3]

B. From II, X = Br.—A "cyclic reactor" based on a design by Greenlee[4] was employed in the Grignard reaction to minimize coupling. This reactor consisted, in ascending order, of a 100-ml. three-neck reservoir flask (two necks stoppered) of refluxing ether, a side-arm column containing magnesium turnings immersed in ether, a condenser, and a dropping funnel with a nitrogen inlet. The magnesium in the reactor column was amalgamated by standing overnight under a saturated ether solution of $HgBr_2$. After draining this solution, the reactor column was refluxed with ether for 1.5 hr.; and finally, the magnesium was further activated with 0.2 ml. of methyl iodide in 1 ml. of ether followed by refluxing in ether for 1.5 hr. In these and all subsequent steps the ether level in the reactor was maintained just above the surface of the magnesium, and the system was kept under dry nitrogen. The reservoir flask was charged with 15 ml. of dry ether and refluxing was begun. The dropwise return rate (from the reactor column to the reservoir flask) was adjusted such that the ether level in the reactor remained constant. One gram (3.34 mmoles) of 5-(β-bromoethylidene)-5H-dibenzo[*a,d*]-10,11-dihydrocycloheptene (II, X = Br)[5] dissolved in 30 ml. of dry ether was added to the reactor column at a steady rate of 1 drop/30 sec. (ratio of reflux-drop rate to addition-drop rate was 30:1). Addition was complete in 3.5 hr., after which time the reactor column was refluxed for an additional 30 min. The reaction mixture was cooled to 25°, the reactor column assembly on the reservoir flask was replaced by a stirrer, and a reflux condenser (with nitrogen inlet) and addition funnel were added to the previously stoppered necks of the reservoir flask. A solution of 0.433 g. (3.30 mmoles) of isobutoxymethyldimethylamine[6] in 10 ml. of dry ether was added dropwise with stirring to the Grignard solution over a period of 5 min. The resulting mixture was refluxed for 0.5 hr., kept overnight at room temperature, and refluxed an additional hour. The reaction mixture was chilled in an ice bath and treated with 20 ml. of saturated NH_4Cl solution. The aqueous layer was extracted with ether, and the ether extract was washed successively with 10 ml. of 2.5 *N* HCl and 15 ml. of water. The aqueous phase was made alkaline with concentrated NH_4OH and extracted with ether. The basic ether extract was washed with 15 ml. of saturated salt solution, dried ($MgSO_4$), and evapo-

(1) W. Treibs and H. J. Klinkhammer, *Ber.*, **83**, 367 (1950).

(2) Freshly prepared from the corresponding hydrochloride: see E. Knorr, *ibid.*, **37**, 3507 (1904); R. B. Burtner, *J. Am. Chem. Soc.*, **71**, 2578 (1949).

(3) The molecular weights were determined by means of a Mechrolab vapor pressure osmometer.

(4) K. W. Greenlee, D. C. Rowlands, C. E. Boord, Abstracts, 117th National Meeting of the American Chemical Society, Philadelphia, Pa., April 1950, p. 8L. We are grateful to Dr. Greenlee for communicating to us details of this method.

(5) R. D. Hoffsommer, D. Taub, and N. L. Wendler, *J. Med. Chem.*, **7**, 392 (1964).

(6) R. Robinson, *J. Chem. Soc.*, 532 (1923).