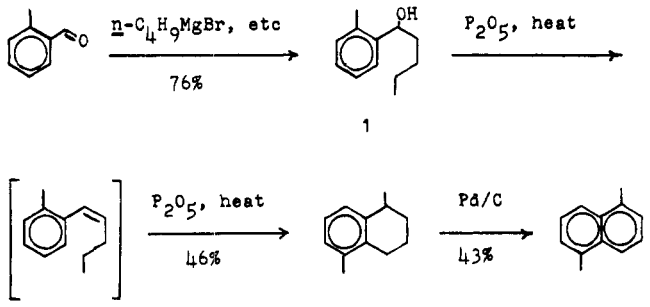
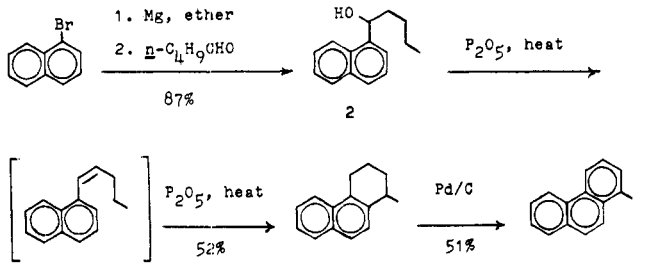
Scheme I



Scheme II



probably not as good as could be realized by use of chromatographic or chemical (picrate formation and regeneration, for example) methods of separation.

Experimental Section

1,5-Dimethylnaphthalene. The alcohol 1-(2-methylphenyl)-1-pentanol (1) was prepared in the usual way from freshly distilled 2-methylbenzaldehyde (53.6 g, 0.45 mol) and an ethereal solution of *n*-butylmagnesium bromide prepared from 14.7 g (0.60 mol) of magnesium and 95.6 g (0.698 mol) of 1-bromobutane. The yield of alcohol distilling at 119–132 °C (1.8 torr) was 60.3 g (75.5%) [lit.[4] bp 84 °C (0.6 torr)]. The alcohol 1 (56.3 g, 0.32 mol) was heated in a simple distillation apparatus with 3 g of phosphorus pentoxide until the distillation of water ceased. Some organic material that had distilled with the water was returned to the flask. The flask was fitted with an upright air-cooled reflux condenser, 2 g of fresh P_2O_5 was added, and the mixture was refluxed 2 h. It was then distilled under reduced pressure. A middle fraction, 27.2 g, distilling at 95–210 °C (23 torr), was redistilled and yielded 23.8 g (46.4% as $C_{12}H_{16}$) distilling at 119–126 °C (21 torr). Analysis by NMR spectroscopy and GLC[1] indicated a mixture containing 69.8% 1,5-dimethyl-1,2,3,4-tetrahydronaphthalene. The mixture (23.0 g) was heated with 1 g of 10% palladium on charcoal catalyst 4 h. The catalyst was removed by filtration with suction while the mixture was still hot. 1,5-Dimethylnaphthalene crystallized in the filtrate and was recrystallized from ethanol: yield 9.67 g (43.1%); mp 75 °C (lit.[2a] mp 77–78 °C).

α-Butyl-1-naphthalenemethanol[5] (2). A Grignard reagent was prepared in ether from 24.32 g (1 mol) of Mg and 217.8 g (1.05 mol) of 1-bromonaphthalene. To this was added an ethereal solution of 77.6 g (0.90 mol) of freshly distilled pentanal [bp 25–30 °C (24 torr)]. The semisolid reaction mixture was refluxed with stirring for 0.5 h. After workup in the usual way, an attempt was made to distill the product under reduced pressure. After removal of a small forerun distilling to 85 °C (5 torr), the distillate began to solidify in the side arm of the Claisen flask being used, and the attempt at distillation was abandoned. The residue in the flask solidified upon cooling and was used without further purification: yield 195.3 g; mp 56–62 °C (lit.[6] mp 65–66 °C). This crude product probably contained naphthalene and 1-bromonaphthalene as impurities.

1-Methylphenanthrene. The crude alcohol 2 (76.7 g) was heated with 7 g of phosphorus pentoxide. After removal of 4 mL of water, the mixture was boiled 2 h beneath an open air-cooled reflux condenser. It was then distilled under reduced pressure. There was obtained a middle fraction, 46.9 g, distilling at 135–180 °C (2.8 torr). After two redistillations to remove some solid, evidently P_2O_5, there was obtained 37.0 g of product distilling at 124–157 °C (1.9 torr); yield 52.6% as $C_{15}H_{16}$. Examination by GLC and NMR spectroscopy[1] indicated it was 79.5% 1-methyl-1,2,3,4-tetrahydrophenanthrene.[7] The presence of other components accounts for the wide range in the boiling point.

Of the cyclization product so obtained, 35.9 g was dehydrogenated by boiling for 2 h with 3.4 g of a 10% Pd/C catalyst with magnetic stirring. The mixture was filtered with suction while hot. The semisolid filtrate was redissolved in hot 95% ethanol and recrystallized. In a second recrystallization, Norit was used in an effort to remove some color but was not very effective. There was obtained 18.0 g (51.2%) of 1-methylphenanthrene as a golden brown microcrystalline solid, mp 118–120 °C (lit.[3] mp 118 °C).

Acknowledgment. David L. West[1] carried out the steps in the preparation of 1-methylphenanthrene from the alcohol 2. For financial aid here and in connection with the first paper of this series,[1] we are indebted to The City College's Department of Chemistry, its Faculty Senate Committee on Research and Publication, and its Biomedical Sciences Research Support Program.

Registry No. 1, 73178-44-6; 2, 3042-60-2; 1,5-dimethylnaphthalene, 571-61-9; 2-methylbenzaldehyde, 529-20-4; butyl bromide, 109-65-9; 1,5-dimethyl-1,2,3,4-tetrahydronaphthalene, 21564-91-0; 1-bromonaphthalene, 90-11-9; pentanal, 110-62-3; 1-methylphenanthrene, 832-69-9; 1-methyl-1,2,3,4-tetrahydrophenanthrene, 1559-81-5.

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(5) Name used by *Chemical Abstracts*.
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Conversion of L-Tyrosine to L-Phenylalanine. Preparation of L-[3',5'-$^{13}C_2$]Phenylalanine

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The importance of specifically ^{13}C-enriched amino acids, peptides, and proteins for a variety of chemical, physical, and biological studies related to structure, dynamics, metabolism, etc., has been increasingly recognized. However, at present, only a limited number of studies have been done due to the unavailability or high cost of these compounds.

In continuation of our program[1] to develop simple high yield syntheses of specifically ^{13}C-labeled amino acids and peptide hormones, we now report the preparation of L-[3',5'-$^{13}C_2$]phenylalanine.

We recently synthesized and resolved DL-[3',5'-$^{13}C_2$]-tyrosine[1b] (90% ^{13}C enriched) in a 10-step synthesis with an overall yield of 22%, using [1,3-$^{13}C_2$]acetone as the source of label. The ready availability of labeled tyrosine prompted us to attempt its conversion to [3',5'-$^{13}C_2$]-phenylalanine.

(1) (a) V. Viswanatha, B. Larsen, and V. J. Hruby, *Tetrahedron*, 35, 1575–80 (1979); (b) V. Viswanatha and V. J. Hruby, *J. Org. Chem.*, 44, 2892–6 (1979); (c) V. J. Hruby, V. Viswanatha, and Y. C. S. Yang, *J. Labelled Compd. Radiopharm.*, in press; (d) M. Blumenstein, V. J. Hruby, and D. M. Yamamoto, *Biochemistry*, 17, 4971–7 (1978); (e) M. Blumenstein and V. J. Hruby, *Biochemistry*, 16, 5169–77 (1977).

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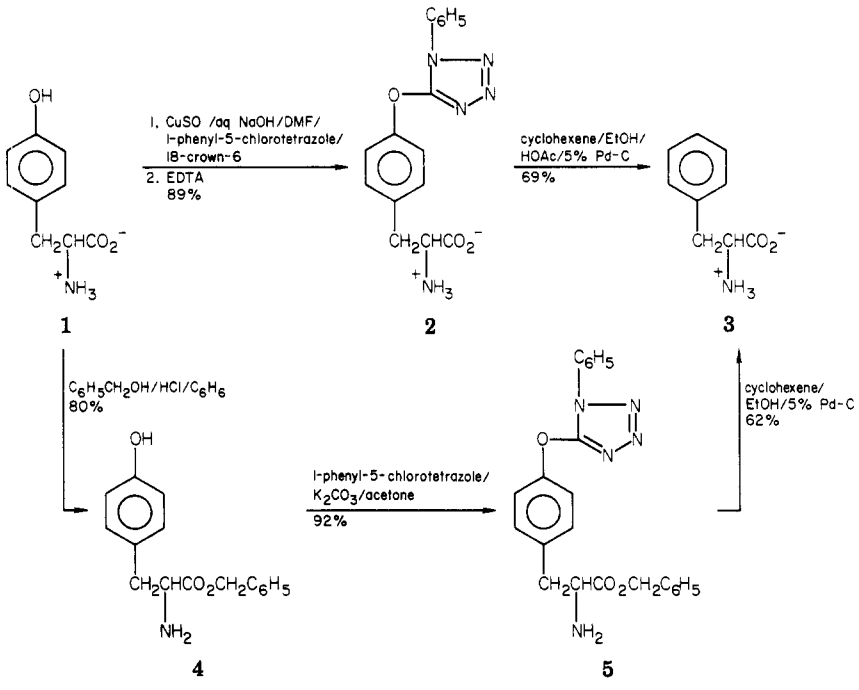
Scheme I



Although attempts[2] have been reported for the conversion of tyrosine to phenylalanine, these methods give phenylalanine either in racemized form or in low yield. It has been shown that the 1-phenyltetrazolyl ethers of phenols can be hydrogenolyzed to the corresponding deoxygenated aromatic compounds in good yields,[3] but the reduction proceeds slowly. However, we have found that the catalytic transfer hydrogenation method,[4] a method of proven utility for the removal of benzyl and nitro groups from amino acid derivatives and peptides, can be used to deoxygenate the tetrazolyl ether of tyrosine to phenylalanine within 2 h. As has been observed in other cases,[4b,c] the above method works best in the presence of an excess of catalyst.

The copper salt of L-tyrosine (see Scheme I) was allowed to react with 1-phenyl-5-chlorotetrazole in the presence of a catalytic amount of 18-crown-6 to give the copper salt of 1-phenyltetrazolyl ether of tyrosine from which the free base **2** was liberated by treatment with EDTA. Hydrogenolysis of **2** in the presence of cyclohexene and 5% Pd–C in ethanol and acetic acid yielded L-phenylalanine in an overall yield of 61%. Similarly L-[3',5'-$^{13}C_2$]tyrosine was converted to L-[3',5'-$^{13}C_2$]phenylalanine in an overall yield of 56%. The labeled phenylalanine was readily converted to *N*-*tert*-butyloxycarbonyl-L-[3',5'-$^{13}C_2$]phenylalanine (**6**). The ^{13}C NMR spectra of the tetrazolyl ether of L-[3',5'-$^{13}C_2$]tyrosine and L-[3',5'-$^{13}C_2$]phenylalanine contained sharp single peaks which indicated that either the 3'- and 5'-carbon atoms have the same chemical shifts or they are different but are in fast exchange on the NMR time scale.

In an alternative procedure, the tetrazolyl ether of L-tyrosine benzyl ester (**5**) (see Scheme I) was also hydrogenolyzed to phenylalanine in an overall yield of 46%. The benzyl ester of L-tyrosine (**4**) was prepared by the azeotropic removal of water, using hydrochloric acid as a catalyst. The reaction **1** → **4** in the presence of *p*-toluenesulfonic acid[6] gave **4** but in lower yield.

These simple transformations of L-tyrosine to L-phenylalanine can be used to convert other ^{13}C-, ^{14}C-, or ^{15}N-tyrosine derivatives to phenylalanine derivatives without going through separate synthetic schemes and resolution steps for each amino acid. We are currently engaged in the preparation of labeled phenylalanine derivatives and their incorporation into various peptide hormones.

Experimental Section

Melting points were determined on a Thomas-Hoover capillary melting point apparatus and are uncorrected. ^1H NMR were measured with a Varian T-60 spectrometer with Me_4Si as the internal reference except in the case of the labeled phenylalanine where the signal corresponding to H_2O (δ 5.00) is taken as the reference. ^{13}C NMR spectra were measured on a Bruker WH-90 FT NMR spectrometer with dioxane as reference. The chemical shifts are in parts per million with reference to Me_4Si. Optical rotations were measured at the mercury green line (546 nm) with a Perkin-Elmer 241-MC polarimeter. Elemental analyses were performed by Chemalytics, Inc. TLC was performed on silica gel G plates with the following solvent systems: (A) 1-butanol–acetic acid–water (4:1:5 upper phase only); (B) 1-butanol–acetic acid–pyridine–water (15:3:10:12); (C) ethyl acetate–pyridine–acetic acid–water (5:5:1:3). The compounds were detected on the TLC plates with iodine vapor and ninhydrin.

1-Phenyltetrazolyl Ether of Tyrosine (2). To a slurry of L-tyrosine (2.0 g) in 2 N NaOH (5.6 mL) was added a solution of $CuSO_4 \cdot 5H_2O$ (1.44 g) in water (16 mL). The blue mixture was heated at 50–55 °C for 30 min. It was then cooled in ice and treated with 5.6 mL of 2 N NaOH and *N*,*N*-dimethylformamide (DMF, 80 mL) to give a green precipitate. NaOH (2 N, 2 mL) was added followed by 1-phenyl-5-chlorotetrazole (3.0 g) in DMF (20 mL) and 18-crown-6 (0.16 g). The mixture was stirred overnight, diluted with acetone (100 mL), stirred for 15 min, and filtered. The light blue precipitate was washed with acetone (100

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mL), water (2 × 30 mL), and then acetone. The precipitate was dried overnight and heated with a solution of 4 g of EDTA in 120 mL of water on a steam bath for 30 min. The mixture was left overnight. The tan colored solid was filtered off and washed with water to give 3.2 g (89%) of **2**: mp 218–222 °C; ^{1}H NMR (TFA) δ 3.45–3.85 (m, 2 H), 4.50–5.05 (m, 1 H), 7.20–8.20 (br d, 9 H); $[\alpha]^{25}_{546}$ −14.95° (*c* 2.0, HOAc). Anal. Calcd for $C_{16}H_{15}N_5O_3 \cdot H_2O$: C, 55.97; H, 4.96; N, 20.41. Found: C, 55.62; H, 5.05: N, 20.73.

Similarly L-[3',5'-$^{13}C_2$]tyrosine (1.5 g) gave 2.2 g (82%) of the corresponding tetrazolyl ether: ^{1}H NMR (TFA) δ 3.40–3.75 (m, 2 H), 4.50–5.00 (m, 1 H), 5.88–6.25 (m, 1 H), 7.30–8.00 (m, 7 H), 8.83–9.20 (m, 1 H); ^{13}C NMR (TFA–D_2O) 102.47.

L-Phenylalanine (3). **2** (1.0 g) was dissolved in absolute ethanol (70 mL) and acetic acid (40 mL) by warming on a steam bath. The solution was cooled to room temperature, and cyclohexene (30 mL) was added followed by 5% Pd–C (2.0 g). The mixture was refluxed for 2.0 h at 100 °C. It was then evaporated to dryness, and the residue was stirred with water (3 × 100 mL) and filtered. The filtrate was evaporated to dryness, and the residue was crystallized from water–ethanol to yield 0.35 g (69%) of L-phenylalanine, $[\alpha]^{25}_{546}$ −39.2° (*c* 2.0, H_2O) [lit.[5] $[\alpha]^{25}_D$ −34.5° (*c* 2, H_2O)]. TLC behavior in solvent systems A, B, and C was identical with that of authentic L-phenylalanine.

Similarly the tetrazolyl ether of L-[3',5'-$^{13}C_2$]tyrosine (1 g) furnished 0.34 g (68%) of L-[3',5'-$^{13}C_2$]phenylalanine: ^{1}H NMR (D_2O) δ 3.35–3.60 (m, 2 H), 4.20–4.60 (m, 1 H), 5.00 (H_2O), 6.10–6.60 (m, 1 H), 7.45–7.80 (br d, 3 H), 8.68–9.24 (m, 1 H); ^{13}C NMR (D_2O) 129.95.

***N*-(*tert*-Butyloxycarbonyl)-L-[3',5'-$^{13}C_2$]phenylalanine (6).** The filtrate from the crystallization of labeled phenylalanine was evaporated to dryness, and the residue (0.1 g) was stirred with S-Boc-4,6-dimethyl-2-mercaptopyrimidine[7] (0.2 g), dioxane (0.5 mL), water (0.5 mL), and triethylamine (0.25 mL) overnight. The solution was diluted with water (5 mL) and extracted with ethyl acetate (3 × 20 mL). The aqueous solution was cooled to 0 °C and the pH was brought to 2.5 with 4% hydrochloric acid cooled to 0 °C. The mixture was saturated with sodium chloride and extracted with ethyl acetate (3 × 30 mL). The ethyl acetate solution was washed with 4% ice-cold hydrochloric acid (3 × 10 mL) and water (3 × 10 mL) and dried over anhydrous sodium sulfate, and the solvent was removed. The residue was crystallized from ether–petroleum ether to give 0.035 g of **6**: mp 83–85 °C; ^{1}H NMR ($CDCl_3$) δ 1.40 (s, 9 H), 2.90–3.25 (m, 2 H), 4.20–4.80 (m, 1 H), 4.80–5.45 (m, 1 H), 5.65–6.20 (m, 1 H), 7.00–7.50 (br d, 3 H), 8.32–8.81 (m, 1 H), 11.49 (br s, 1 H).

L-Tyrosine Benzyl Ester (4). A mixture of L-tyrosine (1.4 g), concentrated hydrochloric acid (4 mL), and benzyl alcohol (40 mL) was heated at 100 °C for 15 min to give a clear solution. Benzene (20 mL) was added, the solution was heated at 100–105 °C, and water was removed azeotropically for 2.5 h. More concentrated hydrochloric acid (1 mL) was added, and heating was continued for another 30 min. The mixture was cooled and diluted with ether (50 mL). The mixture was extracted with dilute hydrochloric acid (5 × 20 mL) and water (2 × 20 mL). The combined aqueous solution was washed with ether, and the pH was brought to 8 with dilute NH_4OH. The mixture was left overnight and the solid was filtered, washed with water, and dried. The white solid was stirred with acetone (100 mL) and filtered to give 50 mg of tyrosine. The filtrate was evaporated to give a gum which solidified to yield 1.6 g (80%) of **4**: mp 110–114 °C (lit.[6] mp 120 °C); ^{1}H NMR ($CDCl_3$–Me_2SO-d_6) δ 2.80–3.00 (m, 2 H), 3.6–4.2 (m, 4 H), 5.16 (s, 2 H), 6.75 (d, 2 H), 6.95 (d, 2 H), 7.35 (s, 5 H).

1-Phenyltetrazolyl Ether of L-Tyrosine Benzyl Ester (5). A mixture of 0.75 g of L-tyrosine benzyl ester (**4**), 1-phenyl-5-chlorotetrazole (0.5 g), and anhydrous K_2CO_3 (2.5 g) in acetone (80 mL) was refluxed overnight. The mixture was evaporated to dryness. The residue was treated with water and filtered to give 1.05 g (92%) of **5**, mp 90–95 °C. An analytical sample was obtained by recrystallization from ethyl acetate and hexane: mp 92–95 °C; ^{1}H NMR ($CDCl_3$) δ 1.70–1.90 (m, 2 H), 2.95–3.15 (m, 2 H), 3.60–3.90 (m, 1 H), 5.10 (s, 2 H), 7.30–7.80 (m, 14 H). Anal. Calcd for $C_{23}H_{21}N_5O_3 \cdot {}^1/_2H_2O$: C, 65.09; H, 5.18; N, 16.51. Found: C, 65.21; H, 5.36; N, 16.71.

L-Phenylalanine (3). A mixture of 1 g of **5** and 5% Pd–C (2 g) in ethanol (15 mL) and cyclohexene (10 mL) was refluxed for 2 h at 100 °C. L-Phenylalanine, 0.24 g (62%), $[\alpha]^{25}_{546}$ −37.0° (*c* 2, H_2O), was isolated as before. TLC behavior in solvent systems A, B, and C was identical with that of authentic L-phenylalanine.

Acknowledgment. We are grateful for the labeled acetone from the Stable Isotope Resource, which is supported by the U.S. Department of Energy and the National Institute of Health, Grant RR-00962-01, which made this work possible. We thank the U.S. Public Health Service, Grant AM-17420, and the National Science Foundation for financial support.

Registry No. 1, 60-18-4; 1 (3',5'-$^{13}C_2$), 70479-98-0; 2, 73198-07-9; 2 (3',5'-$^{13}C_2$), 73198-08-0; 3, 63-91-2; 3 (3',5'-$^{13}C_2$), 73198-09-1; 4, 42406-77-9; 5, 73198-10-4; 6, 73198-11-5; 1-phenyl-5-chlorotetrazole, 14210-25-4; S-Boc-4,6-dimethyl-2-mercaptopyrimidine, 41840-28-2.

Effect of C-21 Substituents on the Elimination of a 17α-Acyloxy Group from 17α-(Acyloxy)-20-oxo Steriods

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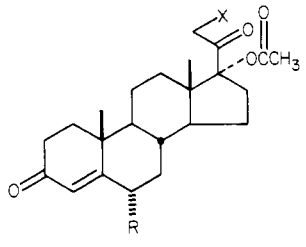
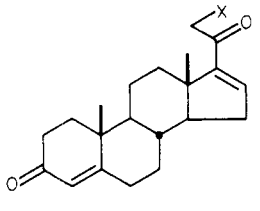
In 1970, Salce, Hazen, and Schoenewaldt[1] reported that heating 17α-acyloxy derivatives of 20-oxo steroids at 105 °C in dimethylformamide containing potassium acetate results in elimination of the acyloxy function to form 16-dehydro-20-oxo steroids. We noted that while the above procedure was described as a general method, all of the compounds cited as starting materials actually were 17α,21-bis(acyloxy)-20-oxo steroids. We report here a study of the effect of the C-21 substituents on the course of the above reaction.

When submitted to reaction conditions similar to those described by Salce et al.[1] 17α,21-diacetoxy-4-pregnene-3,20-dione (**1**) was converted to 21-acetoxy-4,16-pregnadiene-3,20-dione (**2**) in 62% yield. In contrast, 17α-acetoxy-6α-methyl-4-pregnene-3,20-dione (**3**) was recovered unchanged under similar conditions. If the latter compound was heated overnight in refluxing dimethylformamide containing potassium acetate, it decomposed to a complex mixture which did not contain an appreciable amount of the expected product.



1, R = H; X = OC(O)CH
3, R = CH_3; X = H
4, R = H; X = OH
5, R = H; X = Cl
6, R = H; X = OTHP



2, X = $OC(O)CH_3$

When 17α,21-dihydroxy-4-pregnene-3,20-dione 17-acetate (**4**) was heated under conditions similar to those of Salce et al.[1] it reacted by a transacylation to afford the

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