It should be noted in this connection, however, that fluoropyruvic acid, which would be derivable from 3-fluoroalanine, has the relatively low toxicity to rats and mice of 80 mg./kg.[5] and evidence has been presented that this compound is not converted to fluoroacetic acid in the rat.[6]

EXPERIMENTAL[7]

Diethyl acetamido(3-fluoropropyl)malonate. Twenty-three grams (1 mole) of sodium was dissolved in 1 l. of absolute ethyl alcohol and to this solution were added 217 g. (1 mole) of diethyl acetamidomalonate and 141 g. (1 mole) of 1-bromo-3-fluoropropane.[8] The mixture was refluxed for 15 hr., filtered from sodium bromide, and evaporated. The crystalline residue was washed with water and recrystallized from ethyl alcohol. There was obtained 174 g. (63% yield) of diethyl acetamido(3-fluoropropyl)malonate in three crops; m.p. 75–77°.

Anal. Calcd. for $C_{12}H_{20}FNO_5$: F, 6.85; N, 5.05. Found: F, 6.85; N, 5.02 (K).

5-Fluoronorvaline. Diethyl acetamido(3-fluoropropyl)-malonate (140 g.), 325 g. of 48% hydrofluoric acid, and 100 ml. of water were heated at 105–112° for 5 hr. in a "Monel" pressure vessel. The vessel was cooled in ice, opened, and 300 g. of calcium hydroxide was added in portions to bring the *p*H to 3.5. The calcium fluoride was filtered off, washed with water, and the filtrate was evaporated to dryness under reduced pressure. The residue was extracted with ethyl alcohol to remove soluble material. The ethyl alcohol-insoluble residue was recrystallized from ethyl alcohol-water to give 18.5 g. of shining plates of 5-fluoronorvaline; m.p. 190°; yield 27%.

Anal. Calcd. for $C_5H_{10}FNO_2$: F, 14.06; N, 10.37. Found: F, 14.1; N, 10.14 (K).

Diethyl acetamido(4-fluorobutyl)malonate. This compound was prepared by the same procedure used for the 3-fluoropropyl derivative from 1-bromo-4-fluorobutane.[8] It was obtained as a sirup.

6-Fluoronorleucine. The crude diethyl acetamido(4-fluorobutyl)malonate (180 g.) was heated with 360 g. of 48% hydrofluoric acid in a polyethylene bottle in a steam bath for 15 hr. The solution was diluted with 1.5 l. of water, neutralized to *p*H 6 with calcium hydroxide, filtered from calcium fluoride, and evaporated to dryness. The residue was recrystallized 3 times from ethyl alcohol-water to give 43 g. (47% yield) of 6-fluoronorleucine. The product still contained a trace of inorganic material. This was removed by heating the amino acid in water with basic copper carbonate to form the insoluble copper salt, decomposing the copper salt with hydrogen sulfide, and again recrystallizing the amino acid which then melted at 244°.

Anal. Calcd. for $C_6H_{12}FNO_2$: F, 12.74; N, 9.39. Found: F, 12.1; N, 9.13 (K).

Diethyl acetamido(2-fluoroethyl)malonate. This compound was prepared from 1-bromo-2-fluoroethane[8] as described for the 3-fluoropropyl homolog. It was recrystallized from ethyl alcohol to give a 34% yield in 3 crops; m.p. 74–75°.

Anal. Calcd. for $C_{11}H_{18}FNO_5$: F, 7.22; N, 5.32. Found: F, 7.5; N, 5.24 (K).

In hydrolysis experiments, the amino acid produced contained from 8 to 39% of the theoretical amount of fluorine. It was not completely purified.

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DL-3-*p*-Aminophenylalanine. Nitrogen Mustard and Other Derivatives[1]

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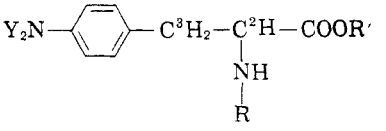
In 1956 Larionov[2] reported that DL-phenylalanine nitrogen mustard ("Sarkolysin") (I) caused the regression of a well established rodent tumor. The need for large amounts of this drug for further experimental tumor studies led us to develop an improved synthesis, since the previous synthetic routes of Bergel *et al.*[3] were not too satisfactory for preparative purposes.

After successful completion of our new synthesis (which is based on the well known azlactone approach to aromatic amino acids), we learned of a recent publication by Pedrazzoli,[4] which describes a similar synthesis of the L- and D- (but not the DL-) forms of phenylalanine nitrogen mustard. Our synthesis also provides a new route to DL-3-*p*-



(I, Y = CH_2CH_2Cl, R = R′ = H)
(II, Y = R = R′ = H)
(III, Y = O, R = C_6H_5CO, R′ = H, 2,3-didehydro, azlactone)
(IV, Y = O, R = C_6H_5CO, R′ = C_2H_5, 2,3-didehydro)
(V, Y = H, R = C_6H_5CO, R′ = C_2H_5)
(VI, Y = R′ = H, R = C_6H_5CO)

(1) Presented before the Organic Division at the San Francisco Meeting of the American Chemical Society, April 1958.

(2) (a) L. F. Larionov, E. N. Shkodinskaja, V. I. Troosh-silcina, A. S. Khokhlove, O. S. Vasina, and M. A. Novikova, (a) *Lancet*, **2**, 169 (1955); (b) *Brit. J. Cancer*, **10**, 26 (1956).

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aminophenylalanine[5] (II) itself; and similarly, Pedrazzoli describes a new preparation for the L- and D- (but not the DL-) forms of this diamino acid.

By reaction of *p*-nitrobenzaldehyde with hippuric acid we obtained the expected azlactone (III). By base-catalyzed ethanolysis this was converted to the ethyl *p*-nitrocinnamate derivative (IV). Hydrogenation of the latter caused simultaneous reduction of the nitro group and the carbon-carbon double bond, giving the monobenzoyl diamino ethyl ester (V). The latter was converted by basic hydrolysis to the monobenzoyl diamino acid (VI); and by acidic hydrolysis to DL-3-*p*-aminophenylalanine itself (II). The compound (V) was also characterized as its picrate. Finally (V) was converted to the desired nitrogen mustard (I), characterized as its monoethanolate.

Since Pedrazzoli's publication[4] is available, we have described below in detail only those compounds not prepared by him, namely: DL-I (monoethanolate), DL-II, DL-V (picrate), and DL-VI. For the remaining compounds, which were prepared in both laboratories, only deviations in procedure have been specified.

EXPERIMENTAL

All melting and boiling points have been corrected. Microanalyses by Drs. Weiler and Strauss, Oxford, England.

Ethyl 2-benzamido-p-nitrocinnamate. For ethanolysis of 5-(*p*-nitrobenzylidene)-2-phenyloxazolone-4,[6] we used sodium ethoxide catalyst in absolute ethanol–benzene, instead of the sulfuric acid catalyst previously employed.[4] An 87% yield of recrystallized product melting at 170–171° was obtained (reported[4] m.p. 168–169.5°). A sample was recrystallized three times more from ethanol for analysis;[7a] the melting range was not improved.

Anal. Calcd. for $C_{18}H_{16}N_2O_5$: C, 63.52; H, 4.74; N, 8.25. Found: C, 63.63, 63.71; H, 4.60, 4.59; N, 8.01, 8.06.

DL-*2-N-benzoyl-3-(p-aminophenyl)-alanine ethyl ester.* By hydrogenating the nitrocinnamate with palladium/charcoal in ethanol (instead of palladium/aluminum oxide[4] in methanol) we were able to complete the hydrogenation at 25° instead of the previously reported[4] 50°. An 80% yield of once-recrystallized (from benzene, not ethanol) product,[7b] m.p. 113–115°, was obtained (reported[4] m.p. 117–118°). A sample was recrystallized repeatedly for analysis, melting point not improved.

Anal. Calcd. for $C_{18}H_{20}N_2O_3$: C, 69.21; H, 6.45; N, 8.97. Found: C, 69.12, 69.26; H, 6.47, 6.35; N, 8.64, 8.89.

DL-*2-N-benzoyl-3-p-aminophenylalanine ethyl ester picrate.* Treatment of 310 mg. of the benzoyl ethyl ester with ethanolic picric acid in the usual manner gave 500 mg. of the crude picrate, m.p. 174.5–176.0°. The product was recrystallized thrice from 95% ethanol for analysis, m.p. 179.5–180.5°.

Anal. Calcd. for $C_{24}H_{23}N_5O_{10}$: N, 12.94. Found: N, 12.83.

DL-*2-N-benzoyl-3-(p-aminophenyl)alanine.* The ethyl ester (0.935 g., 0.003 mole) was suspended in 6.0 ml. of *M* sodium hydroxide and the mixture stirred for 3 hr. The resulting clear yellow solution was filtered from a small residue, diluted 1:1 with water, and adjusted to *p*H 7 with hydrochloric acid. The white precipitate which separated was collected, washed with water, and dried, giving 0.80 g. (94%) of colorless powder, m.p. 206–210° (dec.). This material was recrystallized from 95% ethanol (70 ml./g.) giving 0.70 g. of colorless micro-needles, m.p. 213–215° (dec.). The product was soluble in dilute aqueous hydrochloric acid, or sodium hydroxide.

Anal. Calcd. for $C_{16}H_{16}N_2O_3$: C, 67.59; H, 5.67; N, 9.86. Found: C, 67.58; H, 5.93; N, 9.94.

DL-*3-(p-aminophenyl)alanine.* To the DL-benzoyl ethyl ester (0.935 g., 0.003 mole) was added 10 ml. of 6*M* aqueous hydrochloric acid, and the mixture[8] boiled under reflux for 5 hr. The resulting colorless solution was cooled, and extracted with benzene to remove (precipitated and dissolved) benzoic acid. The aqueous phase was adjusted to *p*H 5–7 with powdered sodium carbonate. A colorless precipitate formed (dry weight, 0.35 g.). This was dissolved in 25 ml. of boiling 85% ethanol. On cooling the solution deposited 160 mg. (dry weight) of colorless needles, m.p. 240–245°, dec. (reported[5a] 245–250°, dec.).

In later experiments the benzene-extracted acidic aqueous solution was deionized with Amberlite IR-45 ion exchange resin, and the filtrate evaporated to dryness to give the crude diamino acid.

DL-*3-p-di-(2-chloroethyl)aminophenylalanine ("phenylalanine mustard").* The *N*-benzoyl ethyl ester was hydroxyethylated, chlorinated, and hydrolyzed by essentially the same procedure employed by Bergel *et al.*[3,9] with their phthalimido ethyl ester. A 93% yield of crude product, m.p. 174–176° (dec.) was obtained (reported[3] m.p. 180–181°; dec.). Recrystallization from ethanol-benzene raised the m.p. to 178–180° (dec.), but the recovery was only 42%. Recrystallization from a large volume (500 ml./g.) of boiling ethanol gave the previously unreported *monoethanolate,*[10] colorless leaflets, m.p. 179–181° (dec., rapid heating, preheat to 170°).

Anal. Calcd. for $C_{13}H_{18}Cl_2N_2O_2.C_2H_6O$: C, 51.29; H, 6.89; Cl, 20.12, N, 7.98. Found: C, 51.42, H, 6.64; Cl, 20.19; N, 8.13.

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(7) (a) The carbon and hydrogen analyses here given are the first which have been reported for this compound. (b) To facilitate certain isotopic studies, Mr. William Nye of this laboratory has converted the monobenzoyldiamino ethyl ester (V) to its 2,4-dinitrophenyl derivative, m.p. 184°, analysis correct; details will be reported elsewhere.

(8) After initial dissolution of the ester in hydrochloric acid, crystals (presumably of the hydrochloride) separated, but redissolved when the mixture was heated to boiling.

(9) The noncrystalline intermediates obtained after treatment with ethylene oxide, and with phosphoryl chloride, were not purified, but used directly for further operations.

(10) The L- (or D-) enantiomorph of this product reportedly forms a monomethanolate on treatment with methanol (See Ref. 3).