# *Aryl-2-halogenoalkylamines. Part XIII.* Chloroethylamino-derivatives of Some Phenoxyalkanoic Acids and of Some Substituted α-Amino-acids.*

By W. Davis, J. J. Roberts, and W. C. J. Ross.

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The compounds $(Cl\cdot CH_2\cdot CH_2)_2N\cdot C_6H_4\cdot O\cdot[CH_2]_n\cdot CO_2H$, where $n = 1$—4, and $(Cl\cdot CH_2\cdot CH_2)_2N\cdot C_6H_4\cdot[CH_2]_n\cdot CH(NH_2)\cdot CO_2H$, where $n = 0$, 2, and 3, have been prepared, and the reactivities of their chlorine atoms have been assessed by determining the rates of hydrolysis in aqueous acetone. The tumour-growth inhibitory activity of a number of derivatives is briefly discussed.

In continuation of the programme outlined in Part XII,* namely, the preparation of compounds having a more selective action on neoplastic tissues, further derivatives of the "aromatic nitrogen mustard," *NN*-di-2-chloroethylaniline, have been prepared. The preparation and properties of some new carboxy-substituted derivatives and of some related α-amino-acids are described in the present communication.

The four *p*-di-2-chloroethylaminophenoxy-acids (I; R = $CH_2\cdot CH_2Cl$, R′ = H; $n$ = 1—4) were prepared by hydroxyethylation of the appropriate aminoester (I; R = H, R′ = Me or Et) by reaction with an excess of ethylene oxide in aqueous acetic acid followed by conversion of the hydroxyethyl ester into the dichloro-ester (I; R = $CH_2\cdot CH_2Cl$, R′ = Me or Et) by treatment with phosphorus oxychloride, and subsequent hydrolysis with concentrated hydrochloric acid (cf. Part XII).

*p*-Acetamidophenoxyacetic acid was prepared by reaction of *p*-acetamidophenol with chloroacetic acid in alkaline solution (Howard, *Ber.*, 1897, **30**, 546) and contrary to the experience of Jacobs and Heidelberger (*J. Amer. Chem. Soc.*, 1917, **39**, 2188) it was found possible to prepare β-*p*-acetamidophenoxypropionic acid in a similar manner by using β-bromopropionic acid. Heating these acetamido-acids with alcoholic hydrogen chloride caused esterification and simultaneous removal of the acetyl group.

1 : 3-Dibromopropane condensed with *p*-acetamidophenol to give *p*-acetamidophenoxypropyl bromide which was converted successively into the nitrile and γ-*p*-aminophenoxybutyric acid (I; R = R′ = H, $n$ = 3) (Jacobs and Heidelberger, *loc. cit.*).

(I) *p*-$R_2N\cdot C_6H_4\cdot O\cdot[CH_2]_n\cdot CO_2R'$ *p*-$(Cl\cdot CH_2\cdot CH_2)_2N\cdot C_6H_4\cdot[CH_2]_n\cdot CO_2H$ (II)

(III) *p*-$(Cl\cdot CH_2\cdot CH_2)_2N\cdot C_6H_4\cdot[CH_2]_n\cdot CH(NH_2)\cdot CO_2H$ *p*-$O_2N\cdot C_6H_4\cdot[CH_2]_n\cdot C(NHAc)(CO_2Et)_2$ (IV)

(V) *p*-$R_2N\cdot C_6H_4\cdot[CH_2]_n\cdot C(NHAc)(CO_2Et)_2$ $O_2N\cdot C_6H_4\cdot[CH_2]_3\cdot\overset{+}{N}C_5H_5\}Br^-$ (VI)

δ-*p*-Aminophenoxyvaleric acid (I; R = R′ = H, $n$ = 4) was similarly prepared starting from 1 : 4-dibromobutane. The yield of *p*-acetamidophenoxydecyl bromide from 1 : 10-dibromodecane was insufficient for further study.

β-*p*-(Di-2-chloroethylamino)phenylpropionic acid (II; $n$ = 2) is a moderately effective cytotoxic agent (Part XII) and recently Bergel and Stock (*J.*, 1954, 2409) have shown that the introduction of an α-amino-group giving the phenylalanine derivative (III; $n$ = 1) leads to considerable enhancement of biological activity. γ-*p*-(Di-2-chloroethylamino)-phenylbutyric acid (II; $n$ = 3) is a very effective compound and it was thought that the introduction of an α-amino-group into this structure might lead to a corresponding increase in activity. In view of the considerable variation in biological activity encountered in the homologous series (I) and (II) it was of interest to prepare compounds of structure (III) where $n$ = 0, 2, and 3.

In exploratory experiments acetamidomalonic ester was condensed with phenethyl bromide in the presence of sodium ethoxide, and hydrolysis of the resulting diethyl *C*-acetamido-*C*-phenethylmalonate gave α-amino-γ-phenylbutyric acid. *p*-Nitrophenylethyl bromide similarly gave α-amino-γ-*p*-nitrophenylbutyric acid. Hydrogenation of the

* Part XII, *J.*, 1953, 2386.

intermediate diethyl acetamido-*p*-nitrophenylethylmalonate over palladium–calcium carbonate gave the corresponding *p*-amino-derivative (V; R = H, $n = 2$) which with ethylene oxide yielded diethyl acetamido-*p*-(di-2-hydroxyethylamino)phenylethylmalonate (V; R = $CH_2 \cdot CH_2 \cdot OH$, $n = 2$). This was converted into the dichloroethyl ester which with hot concentrated hydrochloric acid gave the required α-amino-γ-*p*-(di-2-chloroethylamino)phenylbutyric acid (III; $n = 2$).

Considerable difficulty was encountered in the preparation of *p*-nitrophenylpropyl bromide which was required for the synthesis of the homologue (III; $n = 3$). Leffler and Volwiler (*J. Amer. Chem. Soc.*, 1938, **60**, 896) claim that when phenylpropyl bromide was nitrated by the somewhat ill-defined method used by Braun and Deutsch (*Ber.*, 1912, **45**, 2504) for the corresponding chloride an 82% yield of *p*-nitro-compound was obtained. In our experience all methods of nitration have given approximately equal amounts of *o*- and *p*-isomers and these can only be separated with difficulty by slow fractional distillation. In an attempted chromatographic separation of the isomers on activated alumina considerable amounts of the bromide were converted into the alcohol. The higher-boiling isomer was shown to be *p*-nitrophenylpropyl bromide by the formation of *p*-nitrobenzoic acid when it was oxidised with chromic acid. The *o*- and the *p*-isomer were characterised by the preparation of their respective pyridinium salts (VI).

The oily diethyl acetamido-*p*-(di-2-hydroxyethylamino)phenylpropylmalonate (V; R = $CH_2 \cdot CH_2 \cdot OH$, $n = 3$), which was prepared as above by way of the nitro-diester (IV; $n = 3$) and the amino-diester (V; R = H, $n = 3$), was converted directly into the amorphous di-2-chloroethyl ester (V; R = $Cl \cdot CH_2 \cdot CH_2$, $n = 3$) which on acid hydrolysis gave the crystalline α-amino-δ-*p*-(di-2-chloroethylamino)phenylvaleric acid (III; $n = 3$).

The first member of the series (III; $n = 0$) could not be prepared in the usual manner since *p*-bromonitrobenzene failed to condense with diethyl acetamidomalonate. Another route explored involved the preparation of *p*-aminophenyl-α-hydroxyiminoacetic ester (VII; R = H, X = $NH_2$). Whilst hydrogenation over Raney nickel of the nitro-oxime (VII; R = H, X = $NO_2$) effected reduction of both nitro- and hydroxyimino-groups, giving the diamino-ester (VIII; R = Et), hydrogenation over platinum gave the required amino-oxime (VII; R = H, X = $NH_2$). Acid hydrolysis of the diamino-ester (VIII; R = Et) gave the diamino-acid (VIII; R = H) which had previously been obtained by Grant and Pyman (*J.*, 1921, **119**, 1893).

*p*-X·$C_6H_4$·C(:N·OR)·$CO_2$Et (VII) *p*-$NH_2$·$C_6H_4$·CH($NH_2$)·$CO_2$R (VIII) *p*-(Cl·$CH_2$·$CH_2$)$_2$N·$C_6H_4$·$CH_2$·$CO_2$Et (IX)

*p*-(Cl·$CH_2$·$CH_2$)$_2$N·$C_6H_4$·CHO (X) *p*-(Cl·$CH_2$·$CH_2$)$_2$N·$C_6H_4$·CH($NH_2$)·CN (XI)

*p*-(Cl·$CH_2$·$CH_2$)$_2$·$C_6H_4$·O·$CH_2$·$CH_2$·C(NHAc)($CO_2$Et)$_2$ (XII)

No useful product was obtained by the action of ethylene oxide on the amino-hydroxyimino-ester and it was considered that this might be due to partial reaction with the hydroxyimino-group. Accordingly the *O*-acetate (VII; X = $NH_2$, R = Ac) was prepared by catalytic hydrogenation of the nitro-*O*-acetate (VII; X = $NO_2$, R = Ac). The amino-hydroxyimino-ester and its *O*-acetate gave the diacetate (VII; X = NHAc, R = Ac) on treatment with acetic anhydride. It was not possible to obtain the desired hydroxyethylamino-compound by treating the *O*-acetate (VII; X = $NH_2$, R = Ac) with ethylene oxide.

An attempt to prepare the α-hydroxyimino-derivative from ethyl *p*-(di-2-chloroethylamino)phenyl acetate (IX) was unsuccessful. The required amino-acid was eventually obtained by carrying out a Strecker reaction on *p*-(di-2-chloroethylamino)benzaldehyde (X). The amino-nitrile thus obtained was quantitatively converted into α-amino-*p*-(di-2-chloroethylamino)phenylacetic acid (III; $n = 0$) by heating it with concentrated hydrochloric acid.

*p*-(Di-2-hydroxyethylamino)phenoxyethyl bromide condensed with acetamidomalonic ester to give a product which was not isolated but was converted directly into acetamido-*p*-(di-2-chloroethylamino)phenoxyethylmalonic ester (XII).

Since a correlation between the chemical reactivity of aryl-2-halogenoalkylamines and their cytotoxic activity has been established (Ross, *Adv. Cancer Res.*, 1953, **1**, 397) the rates of hydrolysis of a number of the new compounds under standard conditions have been determined (cf. Ross, *J.*, 1949, 183, and subsequent papers). The annexed Table also indicates the effectiveness of the compounds as inhibitors of the growth of the transplanted

*The extent of hydrolysis of 2-chloroethylamino-acids in 50% acetone at 66°. Concn. of amino-acid derivative* 0·02M. *Time,* 30 *min.*

| | Free acid | Ester | Sodium salt | | |
|---|---|---|---|---|---|
| $n$ | H or Cl (%) | H or Cl (%) | H (%) | Cl (%) | Biological activity † |
| | *p*-(Cl·CH$_2$·CH$_2$)$_2$N·C$_6$H$_4$·O·[CH$_2$]$_n$·CO$_2$H | | | | |
| 1 | 53 | 44 | 52 | 68 | + |
| 2 | 57 | — | 52 | 70 | +++ |
| 3 | 62 | 63 | 52 | 71 | ++ |
| 4 | 64 | — | 54 | 70 | +++ |

| | Free acid | Ester | Sodium salt | | |
|---|---|---|---|---|---|
| $n$ | H or Cl (%) | H or Cl (%) | H (%) | Cl (%) | Biological activity † |
| | *p*-(Cl·CH$_2$·CH$_2$)$_2$N·C$_6$H$_4$·[CH$_2$]$_n$·CH(NH$_2$)·CO$_2$H | | | | |
| 0 | 10 | — | — | — | ‡ |
| 1 * | 22 | — | — | — | +++ |
| 2 | 30 | — | — | — | +++ |
| 3 | 34 | — | — | — | +++ |

* Bergel and Stock, *J.*, 1954, 2409; see also British Empire Cancer Campaign Report for 1953, p. 6.
† As inhibitors of the growth of the transplanted Walker rat carcinoma.
‡ Still under test.

Walker rat carcinoma (for techniques see Badger, Elson, Haddow, Hewett, and Robinson, *Proc. Roy. Soc.*, 1942, *B*, **130**, 255; Haddow, Harris, Kon, and Roe, *Phil. Trans. Roy. Soc.*, 1948, *A*, **241**, 147).

*Biological Results* (Personal communication from Professor A. Haddow).—All four acids (I; $n$ = 1—4) exhibit tumour-growth inhibitory activity, and of these the phenoxypropionic acid derivative (I; $n$ = 2) is outstanding. It is interesting that this compound is isosteric with the most active member of the series (II), namely, the phenylbutyric acid derivative ($n$ = 3) (Part XII). This supports the view expressed there that the especial activity of the phenylbutyric acid derivative was connected with its molecular structure. Activity in these compounds is still dependent on the reactive chlorine atoms since *p*-aminophenylbutyric acid and its *NN*-di-2-hydroxyethyl derivative are ineffective.

In the series of α-amino-acid derivatives the biological activity of the DL-forms of three members have so far been compared (see, also, Bergel and Stock, *loc. cit.*). Preliminary investigations indicate that the amino-acids (III; $n$ = 1—3) are equally effective at a dose level of 1 mg. per rat. It is thus apparent that the insertion of an α-amino-group into the phenylbutyric acid derivative (II; $n$ = 3) does not result in an increase in activity of the magnitude encountered by the insertion of such a group into the phenylpropionic acid derivative (II; $n$ = 2).

## Experimental

p-(*Di-2-chloroethylamino*)*phenoxyacetic Acid.*—Ethylene oxide (25 g.) was added to a stirred suspension of ethyl *p*-aminophenoxyacetate (25 g.) in N-acetic acid (50 ml.). Stirring was continued for 4 hr. and next day the excess of ethylene oxide was removed under reduced pressure and the product was extracted with ether. The ether solution was washed with 2N-sodium hydrogen carbonate and water and then dried and evaporated. *Ethyl* p-(*di-2-hydroxyethylamino*)*phenoxyacetate* (18 g.) formed prisms, m. p. 57·5°, from benzene–light petroleum (b. p. 40—60°) (Found: C, 59·3; H, 7·6. $C_{14}H_{21}O_5N$ requires C, 59·3; H, 7·5%). This ethyl ester was converted into *ethyl* p-(*di-2-chloroethylamino*)*phenoxyacetate*, rhombs, m. p. 55·5°, from light petroleum (b. p. 40—60°) (Found: C, 52·9; H, 6·2. $C_{14}H_{19}O_3NCl_2$ requires C, 52·5; H, 6·0%), and p-(*di-2-chloroethylamino*)*phenoxyacetic acid*, needles, m. p. 112°, from *cyclo*hexane (Found: C, 49·4; H, 5·4. $C_{12}H_{15}O_3NCl$ requires C, 49·3; H, 5·2%), by methods described in Part XII.

*Methyl* β-p-(*di-2-hydroxyethylamino*)*phenoxypropionate*, needles, m. p. 68°, from benzene (Found: C, 58·9; H, 7·5. $C_{14}H_{21}O_5N$ requires C, 59·3; H, 7·5%), *methyl* β-p-(*di-2-chloroethylamino*)*phenoxypropionate*, rhombs, m. p. 51°, from pentane (Found: C, 52·3; H, 6·2. $C_{14}H_{19}O_3NCl$ requires C, 52·5; H, 6·0%), β-p-(*di-2-chloroethylamino*)*phenoxypropionic acid*,

needles, m. p. 93°, from *cyclo*hexane (Found : C, 51·2; H, 5·9. $C_{13}H_{17}O_3NCl_2$ requires C, 51·0; H, 5·6%), *methyl* γ-p-(*di-2-hydroxyethylamino*)*phenoxybutyrate*, rhombs, m. p. 37°, from benzene–*cyclo*hexane (Found : C, 61·0; H, 7·7. $C_{15}H_{23}O_5N$ requires C, 60·6; H, 7·8%), *methyl* γ-p-(*di-2-chloroethylamino*)*phenoxybutyrate*, small prisms, m. p. 65°, from carbon tetrachloride–light petroleum (b. p. 40—60°) (Found : C, 54·6; H, 6·3. $C_{15}H_{21}O_3NCl_2$ requires C, 53·9; H, 6·3%), and γ-p-(*di-2-chloroethylamino*)*phenoxybutyric acid*, needles, m. p. 85·5°, from *cyclo*hexane (Found : C, 52·8; H, 6·2. $C_{14}H_{19}O_3NCl_2$ requires C, 52·5; H, 6·0%), were similarly prepared from the appropriate amino-ester.

δ-p-(*Di-2-chloroethylamino*)*phenoxyvaleric Acid.*—1 : 4-Dibromobutane (Wilson, *J.*, 1945, 48) was condensed with *p*-acetamidophenol (40 g.) giving p-*acetamidophenoxybutyl bromide* (42 g.), m. p. 102·5°, flattened needles from carbon tetrachloride (Found : C, 50·8; H, 5·8. $C_{12}H_{16}O_2NBr$ requires C, 50·4; H, 5·6%), which was converted into p-*acetamidophenoxybutyl cyanide* (89%), small prisms, m. p. 105·5°, from benzene (Found : C, 66·7; H, 7·0. $C_{13}H_{16}O_2N_2$ requires C, 67·2; H, 6·9%) (cf. Jacobs and Heidelberger, *loc. cit.*). Heating the nitrile with concentrated hydrochloric acid gave δ-p-*aminophenoxyvaleric acid hydrochloride*, rhombs, m. p. 188° (decomp.), from concentrated hydrochloric acid (Found : C, 53·5; H, 6·6. $C_{11}H_{16}O_3NCl$ requires C, 53·8; H, 6·6%). *Methyl* δ-p-*aminophenoxyvalerate*, platelets, m. p. 65°, from benzene–light petroleum (b. p. 60—80°) (Found : C, 64·4; H, 7·7. $C_{12}H_{17}O_3N$ requires C, 64·6; H, 7·7%), was converted into the *NN*-di-2-hydroxyethyl and *NN*-di-2-chloroethyl derivatives in the usual manner and as these did not crystallise the latter was hydrolysed to δ-p-(*di-2-chloroethylamino*)*phenoxyvaleric acid*, needles, m. p. 87·5°, from light petroleum (b. p. 60—80°) (Found : C, 53·8; H, 6·7. $C_{15}H_{21}O_3NCl_2$ requires C, 53·9; H, 6·3%).

p-*Acetamidophenoxydecyl Bromide.*—Decamethylene dibromide (30 g.) and *p*-acetamidophenol (5 g.) were heated under reflux in a solution of sodium hydroxide (1·33 g.) in ethanol (30 ml.) and water (1·5 ml.). After 4 hr. the mixture was steam-distilled to remove unchanged dibromide. The non-volatile material was dissolved in benzene–light petroleum (b. p. 60—80°) (1 : 1), and the solution was passed through a short column of activated alumina. The eluates contained a very small amount of the *phenoxydecyl bromide* which formed small prisms, m. p. 122·5—125°, from pentane (Found : C, 58·9; H, 8·0; N, 3·7. $C_{18}H_{28}O_2NBr$ requires C, 58·4; H, 7·6; N, 3·8%).

*Diethyl Acetamidophenylethylmalonate.*—Diethyl acetamidomalonate (5·4 g.) was added to a solution prepared by dissolving sodium (0·6 g.) in ethanol (50 ml.). After the further addition of phenethyl bromide (6 g.) the mixture was heated under reflux with stirring for 16 hr. The filtered solution was evaporated under reduced pressure and a solution of the resulting oil in benzene was washed with water, then dried and evaporated. *Diethyl* C-*acetamido*-C-*phenethylmalonate* (5·1 g.) formed needles, m. p. 114—115°, from light petroleum (b. p. 60—80°) (Found : C, 63·5; H, 7·3. $C_{17}H_{23}O_5N$ requires C, 63·5; H, 7·2%). When this ester (1 g.) was heated for 3 hr. in 3N-hydrochloric acid and then sodium acetate was added to the concentrated solution α-amino-γ-phenylbutyric acid separated as prisms, m. p. 295—296° (cf. Knoop and Hoessli, *Ber.*, 1906, **39**, 1478).

*Diethyl Acetamido*-p-*nitrophenylethylmalonate.*—The p-*nitro-ester* (39 g.), prepared in a similar manner by condensing *p*-nitrophenylethyl bromide (40 g.) (Foreman and McElvain, *J. Amer. Chem. Soc.*, 1940, **62**, 1436) with acetamidomalonic ester (33·2 g.), formed fine needles, m. p. 117·5°, from benzene–light petroleum (b. p. 40—60°) (Found : C, 55·9; H, 6·2; N, 7·6. $C_{17}H_{22}O_7N_2$ requires C, 55·7; H, 6·1; N, 7·7%). On hydrolysis with 3N-hydrochloric acid the *p*-nitro-ester yielded α-*amino*-γ-p-*nitrophenylbutyric acid*, pale yellow needles, m. p. 233°, from water (Found : C, 53·6; H, 5·7; N, 12·3. $C_{10}H_{12}O_4N_2$ requires C, 53·6; H, 5·4; N, 12·5%). Potentiometric titration of this acid in aqueous solution indicated that the $pK_a$ of the amino-group was 9·1 and that of the carboxyl group was 3·05.

*Diethyl Acetamido*-p-*aminophenylethylmalonate.*—The *p*-nitro-ester (30 g.), dissolved in ethanol (500 ml.) containing Raney nickel catalyst, was shaken at 40—50° in hydrogen until the theoretical amount had been taken up. The p-*amino-ester* formed small prisms, m. p. 113°, from benzene–*cyclo*hexane (Found : C, 60·6; H, 7·3; N, 8·7. $C_{17}H_{24}O_5N_2$ requires C, 60·7; H, 7·2; N, 8·3%).

*Diethyl Acetamido*-p-(*di-2-chloroethylamino*)*phenylethylmalonate.*—The *p*-amino-ester was treated with ethylene oxide in 2N-acetic acid in the usual manner. *Diethyl acetamido*-p-(*di-2-hydroxyethylamino*)*phenylethylmalonate* formed small prisms, m. p. 122°, from benzene–light petroleum (b. p. 60—80°) (Found : C, 59·1; H, 7·4; N, 6·4. $C_{21}H_{32}O_7N_2$ requires C, 59·4; H, 7·6; N, 6·6%). When this dihydroxyethyl ester (0·7 g.) was heated with phosphorus oxychloride (1 ml.) in dry benzene (10 ml.) for 0·5 hr., the corresponding *di-2-chloroethyl derivative*,

small needles, m p. 79—80°, from light petroleum (b. p. 60—80°), was obtained (Found : C, 54·7; H, 6·6; N, 6·1. $C_{21}H_{30}O_5N_2Cl_2$ requires C, 54·7; H, 6·6; N, 6·1%).

α-*Amino*-γ-p-(*di*-2-*chloroethylamino*)*phenylbutyric Acid.*—Heating the di-2-chloroethyl ester under reflux with concentrated hydrochloric acid for 1 hr. caused hydrolysis and decarboxylation, with the formation of the α-*aminobutyric acid*, small prisms, m. p. 174—176°, from ethanol (Found : C, 52·6; H, 6·9; N, 8·6. $C_{14}H_{20}O_2N_2Cl_2$ requires C, 52·7; H, 6·3; N, 8·8%).

p-(*Di*-2-*hydroxyethylamino*)*phenoxyethyl Bromide.*—To *p*-aminophenoxyethyl bromide (Jacobs and Heidelberger, *J. Amer. Chem. Soc.*, 1917, **39**, 2442) (5·0 g.) in 2N-acetic acid (15 ml.), ethylene oxide (20 ml.) was added and the mixture was stirred for 4 hr. The *dihydroxyethylamino*-compound formed prisms, m. p. 82—83°, from benzene–light petroleum (b. p. 60—80°) (Found : C, 47·5; H, 6·2; Br, 26·2. $C_{12}H_{18}O_3NBr$ requires C, 47·4; H, 6·0; Br, 26·3%); it gave a *picrate*, prisms, m. p. 110°, from benzene (Found : C, 41·0; H, 4·25; N, 10·4. $C_{18}H_{21}O_{10}N_4Br$ requires C, 40·5; H, 4·0; N, 10·5%).

*Diethyl Acetamido*-p-(*di*-2-*chloroethylamino*)*phenoxyethylmalonate.*—*p*-(Di-2-hydroxyethylamino)phenoxyethyl bromide (9·0 g.) was condensed with acetamidomalonic ester (5·4 g.) as described above. The oily product (10 g.) was heated with phosphorus oxychloride (10 ml.) in benzene (30 ml.) for 1 hr. The benzene solution was poured on ice and then dried and passed through a column of activated alumina. Final elution of the column with ether afforded *diethyl acetamido*-p-(*di*-2-*chloroethylamino*)*phenoxyethylmalonate* which formed needles, m. p. 102—103°, from light petroleum (b. p. 60—80°) (Found : C, 52·4; H, 6·3; N, 6·2; Cl, 14·4. $C_{21}H_{30}O_6N_2Cl_2$ requires C, 52·8; H, 6·3; N, 5·9; Cl, 14·9%).

3-p-*Nitrophenylpropyl Bromide.*—3-Phenylpropyl bromide (100 g.) was added during 45 min. to a stirred mixture of nitric acid (200 ml.; *d* 1·42) and nitric acid (50 ml.; *d* 1·50). The temperature was kept at 0° during the addition and for a further 2 hr., after which the mixture was poured on crushed ice (1 kg.) and extracted with ether (2·5 l.). The ether layer was washed with an excess of saturated aqueous sodium hydrogen carbonate and finally dried ($CaCl_2$). The yellow oil which remained after evaporation of the ether was slowly distilled at 0·75 mm., the following fractions being collected : (*a*) b. p. 65—70° (37 g. of unchanged bromide), (*b*) b. p. 120—125°, $n_D^{23}$ 1·5691 (4·6 g.), (*c*) b. p. 125—128°, $n_D^{23}$ 1·5732 (18 g.), (*d*) b. p. 128—132°, $n_D^{23}$ 1·5745 (14 g.), (*e*) b. p. 132—134°, $n_D^{23}$ 1·5760 (11 g.), (*f*) b. p. 134—136°, $n_D^{23}$ 1·5775 (6 g.), (*g*) b. p. 136—138°, $n_D^{23}$ 1·5780 (5 g.), and (*h*) b. p. 138—140°, $n_D^{23}$ 1·5792 (20 g.). Fraction (*h*) was pure *p*-nitrophenylpropyl bromide, m. p. −2° to 0°, b. p. 130—136°/0·4 mm., 156—160°/2 mm., $n_D^{21}$ 1·5780 (Leffler and Volwiler, *loc. cit.*, gave b. p. 152—156°/2 mm., $n_D^{23}$ 1·576), which gave *p*-nitrobenzoic acid in high yield on oxidation with chromic–sulphuric acid. When the *p*-nitrobromide was heated with pyridine for 10 min. 1-p-*nitrophenylpropylpyridinium bromide*, prisms from ether–methanol, m. p. 177—180° (Found : C, 52·0; H, 4·7; N, 8·85. $C_{14}H_{15}O_2N_2Br$ requires C, 52·0; H, 4·7; N, 8·7%), was formed. On redistillation fractions (*b*), (*c*), and (*d*) gave *o*-nitrophenylpropyl bromide, m. p. 0°, b. p. 114°/0·75 mm., 138—144°/2 mm., $n_D^{21}$ 1·573, which was characterised by the preparation of its *pyridinium salt*, small prisms, m. p. 182—184°, from ether–methanol, depressed to 150° on admixture with the *p*-isomer (Found : C, 52·1; H, 4·8; N, 8·6%). The *o*-derivative gave no nitrobenzoic acid on oxidation with chromic–sulphuric acid.

α-*Amino*-δ-p-(*di*-2-*chloroethylamino*)*phenylvaleric Acid.*—*p*-Nitrophenylpropyl bromide was condensed with acetamidomalonic ester by the method described above, to give *diethyl acetamido*-p-*nitrophenylpropylmalonate*, m. p. 75—78°, fine needles from benzene–light petroleum (b. p. 60—80°) (Found : C, 57·0; H, 6·3. $C_{18}H_{24}O_7N_2$ requires C, 56·8; H, 6·4%). On hydrogenation in ethanol solution over palladium–calcium carbonate the nitro-ester (9·8 g.) gave *diethyl acetamido*-p-*aminophenylpropylmalonate* (8 g.), m. p. 74—75·5°, prismatic needles from ether–pentane or benzene–*cyclo*hexane (Found : C, 61·9; H, 7·6. $C_{18}H_{26}O_5N_2$ requires C, 61·7; H, 7·5%); it formed a *picrate*, m. p. 179—180°, prisms from benzene (Found : C, 49·8; H, 5·4. $C_{24}H_{29}O_{12}N_5$ requires C, 49·7; H, 5·0%). The amino-ester (6 g.) was treated with ethylene oxide in dilute acetic acid as described above, and the oily product was dissolved in dry chloroform (30 ml.). After addition of phosphorus oxychloride (10 ml.) the solution was heated under reflux for 0·5 hr. and then evaporated under reduced pressure. The residue was dissolved in concentrated hydrochloric acid (50 ml.) and the solution was heated under reflux for 4 hr. On the addition of saturated aqueous ammonium acetate to the ice-cooled solution a buff-coloured precipitate formed. This was collected, washed with ammonium acetate solution, then with water, and dried on porous tile (yield, 3·7 g.). The *amino-acid* was crystallised by dissolution in acetic acid (4 ml. per g.) followed by the addition of benzene (20 ml. per g.), small flattened needles containing acetic acid of crystallisation being slowly deposited. The acetic

acid was slowly lost on exposure to air; a specimen dried at 80°/5 mm. for 3 hr. had m. p. 180—184° (decomp.; dependent on the rate of heating) (Found: C, 54·0; H, 6·6; N, 8·4; Cl, 21·0. $C_{15}H_{22}O_2N_2Cl_2$ requires C, 54·1; H, 6·7; N, 8·4; Cl, 21·3%).

*α-Amino*-p-(*di-2-chloroethylamino*)*phenylacetic Acid.*—Potassium cyanide (2 g.) and ammonium chloride (1·6 g.) were dissolved in the minimum quantity of water and added to a solution of *p*-(di-2-chloroethylamino)benzaldehyde (2·5 g.) (Anker and Cook, *J.*, 1944, 489) in methanol (50 ml.). After 2 days at room temperature the turbid solution was diluted with water and extracted with ether. The ethereal extract was extracted three times with 2N-hydrochloric acid. The oil which separated from the acid extract on the addition of anhydrous sodium carbonate was dissolved in saturated ethanolic hydrogen chloride and when this was diluted with dry ether small crystals of *α-amino*-p-(*di-2-chloroethylamino*)*benzyl cyanide dihydrochloride* (1·5 g.), m. p. 110—115° (decomp.), were precipitated (Found: N, 12·6. $C_{12}H_{17}N_3Cl_4$ requires N, 12·2%). A solution of the amino-nitrile hydrochloride (0·5 g.) in concentrated hydrochloric acid (5 ml.) was heated under reflux for 3 hr. On saturation of the cooled solution with sodium acetate *α-amino*-p-(*di-2-chloroethylamino*)*phenylacetic acid* was precipitated. It formed prisms, m. p. 182°, from methanol (Found: C, 49·2; H, 5·8; N, 9·5; Cl, 24·1. $C_{12}H_{16}O_2N_2Cl_2$ requires C, 49·5; H, 5·5; N, 9·6; Cl, 24·4%).

*Ethyl α-Amino*-p-*aminophenylacetate.*—A solution of α-hydroxyimino-*p*-nitrophenylacetic ester (9 g.) (Borsche, *Ber.*, 1909, **42**, 3597) in methanol (1·5 l.) was hydrogenated over Raney nickel. The filtered solution was concentrated to 200 ml. and saturated with hydrogen chloride, and then dry ether was added. The precipitated *dihydrochloride* (5·35 g.) formed stout prisms, decomp. *ca.* 250°, from ether–methanol (Found: C, 44·4; H, 5·9; N, 10·2; Cl, 26·8. $C_{10}H_{14}O_2N_2$,2HCl requires C, 44·9; H, 6·0; N, 10·5; Cl, 26·5%). The ester formed a *diacetate*, prisms, m. p. 175—177°, from water (Found: C, 59·9; H, 6·5; N, 10·4. $C_{14}H_{18}O_4N_2$ requires C, 60·4; H, 6·5; N, 10·1%). α-Amino-*p*-aminophenylacetic acid dihydrochloride, which was obtained by heating the diamino-ester (1 g.) with concentrated hydrochloric acid (3 ml.) for 3·5 hr., formed prisms, m. p. >280° from ether–ethanol (Grant and Pyman, *loc. cit.*, record m. p. >280° for this dihydrochloride) (Found: C, 40·4; H, 5·3; N, 11·8. Calc. for $C_8H_{12}O_2N_2Cl_2$: C, 40·2; H, 5·1; N, 11·7%).

*Ethyl* p-*Aminophenyl-α-hydroxyiminoacetate.*—Ethyl α-hydroxyimino-*p*-nitrophenylacetate (1·7 g.) was hydrogenated over Adams platinum catalyst in methanol (250 ml.). Concentration of the filtered solution gave *ethyl* p-*aminophenyl-α-hydroxyiminoacetate*, which formed prisms, m. p. 141—142°, from benzene–light petroleum (b. p. 40—60°) (Found: C, 57·9; H, 5·9; N, 13·5. $C_{10}H_{12}O_3N_2$ requires C, 57·7; H, 5·8; N, 13·5%). The amino-hydroxyimino-ester formed a *hydrochloride*, prisms, m. p. 205°, from ether-ethanol (Found: C, 48·5; H, 5·7; N, 11·4; Cl, 14·9. $C_{10}H_{12}O_3N_2$,HCl requires C, 49·1; H, 5·4; N, 11·5; Cl, 14·5%), and a *diacetate*, prisms, m. p. 127—128°, from benzene–light petroleum (b. p. 40—60°) (Found: C, 57·8; H, 5·7; N, 9·9. $C_{14}H_{16}O_5N_2$ requires C, 57·5; H, 5·5; N, 9·6%).

*Ethyl α-Acetoxyimino*-p-*aminophenylacetate.*—Ethyl α-hydroxyimino-*p*-nitrophenylacetate was converted into its O-*acetate*, m. p. 105°, prisms from benzene–light petroleum (b. p. 40—60°) (Found: C, 51·3; H, 4·4; N, 10·3. $C_{12}H_{12}O_6N_2$ requires C, 51·4; H, 4·3; N, 10·0%), by treatment with acetic anhydride, and this acetate afforded *ethyl α-acetoxyimino*-p-*aminophenylacetate*, m. p. 218—219°, prisms from aqueous ethanol (Found: C, 57·2; H, 5·6; N, 11·3. $C_{12}H_{14}O_4N_2$ requires C, 57·6; H, 5·6; N, 11·2%), on hydrogenation over a platinum catalyst. The reduction product formed the diacetate, m. p. 127—128°, described above, when treated with acetic anhydride.

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THE CHESTER BEATTY RESEARCH INSTITUTE, THE INSTITUTE OF CANCER RESEARCH:
ROYAL CANCER HOSPITAL, FULHAM ROAD, LONDON, S.W.3. [*Received, November 19th*, 1954.]