CXV. NOTE ON THE ERLENMEYER AMINO-ACID SYNTHESIS.

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THE series of reactions, constituting a general synthesis of α -amino-acids, devised by Erlenmeyer [1893] and since employed by many other workers, can be conducted, in most cases, with little trouble and excellent yields except at one stage. The condensation of an aromatic aldehyde with hippuric acid and the conversion of the resulting azlactone into the corresponding α -benzoylaminoacrylic acid proceed almost always in yields of 70 % or over. The ease with which the acrylic acid derivative may be reduced to the corresponding propionic acid varies considerably, however, in different cases. Moreover, even in those cases in which the ultimate yield obtainable is fairly high, the reaction is apt to be somewhat troublesome to carry out. In order to bring about this reduction, Erlenmeyer employed sodium amalgam, in which he has been followed by the majority of subsequent workers, although occasionally [cf. Barger and Ewins, 1917] sodium in alcohol has been used with success.

In the synthesis of thyroxine [Harington and Barger, 1927] where it was desired to utilise Erlenmeyer's method for the preparation of β -3 : 5-diiodo-4-[4'-(hydroxyphenoxy)phenyl]- α -aminopropionic acid, the use of any alkaline reducing agent was precluded by the presence of iodine atoms in the molecule. We attempted at first to surmount the difficulty by using hydrogen and palladium in neutral or acid solution, but our efforts in this direction were entirely without success. It then occurred to us that the desired end might be achieved by the use of hydriodic acid and red phosphorus, since we had previously obtained evidence that the iodine atoms, in the compounds with which we were working, were stable towards these reagents. This experiment was successful, and we were able, by boiling the α -benzoylamino-3 : 5-diiodo-4-(4'-methoxyphenoxy)cinnamic ester with hydriodic acid and red phosphorus, to obtain a 25 % yield of the desired amino-acid.

Later it became necessary, for further work, to prepare larger quantities of this amino-acid, and experiments were therefore undertaken with the object of improving the yield. Without going into the various unsuccessful variations that were tried, it may be said at once that we have succeeded in

improving the above-mentioned yield of 25 % to one of 82 % by the simple modification of substituting for the constant boiling hydriodic acid originally employed a mixture of equal parts of the latter and of acetic anhydride. This mixture is an excellent solvent, and, by its use, the reaction could be made to proceed smoothly and to yield a clean product which could be worked up without difficulty. It may further be remarked that the whole reaction, *i.e.* the reduction and the simultaneous hydrolytic removal of the benzoyl group, was complete after boiling for $1\frac{1}{2}$ hours with the hydriodic acid-acetic anhydride mixture, so that the saving of time effected, as compared with Erlenmeyer's original method, even supposing the latter to have been applicable in this case, was very considerable.

The surprising success of this method in the case under discussion, and its great rapidity, led us to investigate its applicability to the synthesis of some other amino-acids. In particular were we interested to try to prepare in this way 3 : 4-dihydroxyphenylalanine, the synthesis of which by Erlenmeyer's method has been carried out by Funk [1911], but with unsatisfactory yields. Using vanillin as our starting point we were indeed able to prepare this amino-acid in yields which were satisfactory, whilst not being so good as those which we obtained in other cases. We have further applied the method to the synthesis of phenylalanine, tyrosine, and desiodothyroxine, the results in all cases being superior to those obtainable by Erlenmeyer's original method.

It will be noticed in the experimental part that we have employed in some instances the benzoylaminoacrylic acid and in others the ester; we have observed slight but definite differences in the yields of the different amino-acids obtainable from the acid and the ester; it appears to be a matter of experiment to determine, in any given case, which is the more suitable.

EXPERIMENTAL.

β -[3 : 5-Diiodo-4-(4'-hydroxyphenoxy)phenyl]- α -aminopropionic acid.

α -Benzoylamino-3 : 5-diiodo-4-(4'-methoxyphenoxy)cinnamic acid. The azlactone prepared by the condensation of 3 : 5-diiodo-4-(4'-methoxyphenoxy)-benzaldehyde with hippuric acid [cf. Harington and Barger, 1927] was dropped into 100 parts of a boiling solution of 1 % sodium hydroxide in 70 % alcohol; the substance passed rapidly into solution and the reaction was complete after boiling for 5 minutes. The hot solution was acidified with hydrochloric acid, whereupon the acid began immediately to crystallise; after standing a few hours in the ice-chest, the acid was filtered off; the first crop so obtained amounted to 60 % of the theoretical yield; a further 30 % was obtained by heating the filtrate to boiling, diluting with an equal volume of hot water, and again cooling; the total yield was therefore almost quantitative. The acid was moderately soluble in hot alcohol, and sparingly soluble in water; on recrystallisation from 90 % alcohol it formed colourless needles, m.p. 239–241° (decomp.).

Analysis. 18.3 mg. gave 0.38 mg. N (micro-Kjeldahl).

Calculated for $C_{23}H_{17}O_5NI_2$ 2.2 % N; found 2.1 % N.

β -[3 : 5-Diiodo-4-(4'-hydroxyphenoxy)phenyl]- α -aminopropionic acid. 5 g. of the foregoing acid were boiled for $1\frac{1}{2}$ hours under a reflux condenser with a mixture of 25 cc. hydriodic acid (Sp. Gr. 1.7) and 25 cc. acetic anhydride together with 3 g. red phosphorus. The solution was filtered hot through asbestos into a Claisen flask, the phosphorus being washed with acetic acid. The filtrate was evaporated to dryness *in vacuo*; some water was added and the evaporation repeated. The residue was dissolved in about 40 cc. boiling water; the solution was cooled under the tap, the flask being well shaken, and the precipitate, consisting of the hydriodide of the amino-acid mixed with benzoic acid, was filtered off and washed thoroughly with ether; the aqueous portion of the filtrate was extracted twice with ether, and warmed to remove dissolved ether. The first crop of hydriodide was added to the boiling aqueous solution, a little hydrochloric acid being added, if necessary, to obtain complete solution, and the free amino-acid precipitated by the cautious addition of ammonia; 3.35 g. of the pure amino-acid, or 82 % of the theoretical amount, were obtained.

Phenylalanine.

10 g. of α -benzoylaminocinnamic acid, obtained by the method of Erlenmeyer [1893], were boiled under a reflux condenser with 100 cc. of the hydriodic acid-acetic anhydride mixture and 10 g. of red phosphorus for $1\frac{1}{2}$ hours; the solution was filtered and evaporated to dryness *in vacuo*; water was added and the evaporation repeated; the residue was shaken up with water and ether; the aqueous layer was separated, and, after a second extraction with ether, was heated to boiling and carefully neutralised with ammonia; on cooling there separated, in colourless plates, analytically pure phenylalanine. The yield was 88 % of the theory.

Analysis. 8.8 mg. gave 0.74 mg. N (micro-Kjeldahl).

14.19 mg. gave 2.0 cc. moist N_2 at 19° and 766 mm. (Van Slyke).

	Total N	NH_2-N
Calculated	8.5 %	8.5 %
Found	8.4	8.1

Tyrosine.

In a precisely similar manner there was obtained from ethyl α -benzoyl-amino-*p*-methoxycinnamate a 60 % yield of tyrosine.

Analysis. 15.5 mg. gave 1.2 mg. N (micro-Kjeldahl).

14.11 mg. gave 1.83 cc. moist N_2 at 19° and 766 mm. (Van Slyke).

	Total N	NH_2-N
Calculated	7.7 %	7.7 %
Found	7.7	7.4

3 : 4-Dihydroxyphenylalanine.

Azlactone from vanillin and hippuric acid. An intimate mixture of vanillin (15.2 g.), hippuric acid (17.9 g.), and freshly fused sodium acetate (15 g.) was treated with 30 cc. acetic anhydride and heated on the water-bath for

15 minutes. The reaction product was ground up with water, filtered, and the precipitate well washed. The crude product was crystallised from glacial acetic acid, and formed yellow needles, m.p. 189°. The yield was 75 %.

Analysis. 15.2 mg. gave 0.646 mg. N (micro-Kjeldahl).

Calculated for $C_{19}H_{15}O_5N$ 4.15 % N; found 4.25 % N.

Ethyl α -benzoylamino-3-methoxy-4-hydroxycinnamate. 10 g. of the above azlactone were dissolved in 100 cc. alcohol to which were added 10 cc. concentrated sulphuric acid, and the solution was boiled under a reflux condenser for 15–20 minutes; the greater part of the alcohol was then distilled off under reduced pressure, and the residue was poured into a dish and allowed to stand. After some days crystallisation began and, at the end of a week, was complete; the ester was filtered off and recrystallised from dilute alcohol. It formed colourless prisms, m.p. 128–129°. The yield was 65 % of the theoretical amount.

Analysis. 0.0862 g. gave 0.2109 g. CO_2 ; 0.0450 g. H_2O .

15.7 mg. gave 0.63 mg. N (micro-Kjeldahl).

	C	H	N
Calculated for $C_{19}H_{15}O_5N$	66.8 %	5.6 %	4.1 %
Found	66.7	5.8	4.0

It will be noted that the analysis indicates that the acetyl group, which was present in the azlactone, was split off in the process of preparation of the ester.

3 : 4-Dihydroxyphenylalanine. 5 g. of the ester were boiled for 1½ hours with 25 cc. hydriodic acid, 25 cc. acetic anhydride and 5 g. red phosphorus, in an atmosphere of hydrogen; the solution was filtered and evaporated to dryness under diminished pressure, hydrogen being led into the capillary tube of the Claisen flask; the aqueous solution of the residue, after removal of the benzoic acid by ether extraction, was neutralised with ammonia and evaporated to dryness under the same precautions to avoid access of air; the product was dissolved in a little water and the solution treated with a large excess of alcohol which precipitated the greater part of the pigment; the latter was rapidly filtered off and the filtrate once more taken to dryness. The residue was dissolved in hot water containing a little sulphur dioxide, the solution was boiled with charcoal and filtered, and the filtrate allowed to crystallise in a vacuum desiccator. In this way the amino-acid was obtained almost colourless and in well-formed crystals. The yield was 50 % of the theoretical. The substance melted at 269° (decomp.) and agreed in its properties with the product described by Funk [1911].

Analysis. 14.3 mg. gave 0.98 mg. N (micro-Kjeldahl).

21.3 mg. gave 2.55 cc. moist N_2 at 17° and 760 mm. (Van Slyke).

	Total N	NH_2 -N
Calculated	7.1 %	7.1 %
Found	6.9	6.9

Desiodothyroxine.

The azlactone was prepared in the usual manner from 4-(4'-methoxyphenoxy)benzaldehyde [cf. Harington, 1926] and hippuric acid; there was obtained 70 % of the theoretical amount of a substance which, after crystallisation from glacial acetic acid, formed yellow needles, m.p. 141°.

Analysis. 16.0 mg. gave 0.575 mg. N (micro-Kjeldahl).

Calculated for $C_{23}H_{17}O_4N$ 3.8 % N; found 3.6 % N.

The above azlactone was converted into the acid by boiling for 5 minutes with 100 parts of 1 % sodium hydroxide in 45 % alcohol; the acid, on recrystallisation from dilute alcohol, formed colourless branched needles, m.p. 192.5°. The yield was 95 % of the theory.

Analysis. 15.8 mg. gave 0.554 mg. N (micro-Kjeldahl).

Calculated for $C_{23}H_{19}O_5N$ 3.6 % N; found 3.5 % N.

The preceding compound was converted into the amino-acid in a precisely similar manner to that described for tyrosine and phenylalanine. The yield of amino-acid was 61 % of the theoretical. The product melted at 253–254° (decomp.), and gave a hydrochloride with m.p. 239–240°; it was therefore in all respects identical with the substance previously prepared by Harington [1926].

Analysis. 19.9 mg. gave 1.02 mg. N (micro-Kjeldahl).

26.8 mg. gave 2.22 cc. moist N_2 at 17° and 760 mm. (Van Slyke).

	Total N	NH_2 -N
Calculated for $C_{18}H_{18}O_4N$	5.1 %	5.1 %
Found	5.1	4.9

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