

# Studies on Amino-Acids and Related Compounds. XIII. Electrolyses of some Amino-Acids in Nitric Acid.

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In part XII<sup>(1)</sup> the mechanism of the electrolysis of phenylalanine in nitric acid has been discussed. In the present paper, the electrolyses of tyrosine, glycine, alanine, and glutamic acid in nitric acid are also reported.

It was found that tyrosine, in comparison to phenylalanine, was profoundly attacked by the electrolysis in nitric acid, and, except oxalic acid as the only crystalline substance, a small quantity of resinous matter with a little of phenolic substance was obtained.

The electrolyses of glycine, alanine, and glutamic acid were performed in 4 N- or 8 N-NO<sub>3</sub>H at 100°C., and from the electrolysates there were isolated:

from *glycine*, oxalic acid (34%)\*\*; from *alanine*, formaldehyde and oxalic acid (27%); and from *glutamic acid*, succinic acid and oxalic acid.

These data do not indicate whether the oxidation was due to electrolysis or merely to the reaction of nitric acid. However, Mörner<sup>(2)</sup> has pointed out that the formation of oxalic acid from tyrosine occurred by heating with 25% or 60% nitric acid, but that oxalic acid was not obtained from glycine, alanine, phenylalanine, and glutamic acid by heating them even with 60% or 90% nitric acid, and that only when they were heated with fuming nitric acid (sp. gr. 1.50), oxalic acid was obtained from these amino-acids.

These facts ascertain that the formation of oxalic acid from these amino-acids is due to the electrolysis in nitric acid. In the previous paper,<sup>(1)</sup> it was confirmed that the formation of oxalic acid from phenylalanine by electrolysis originates principally from the side chain of phenylalanine. In the same way, it may be considered that the formation of oxalic acid is chiefly from the group -CH(NH<sub>2</sub>)COOH in case of the above-mentioned amino-acids.

In the case of glutamic acid, the yields of oxalic- and succinic-acids by the electrolysis carried out under the same conditions, were as follows:

The table shows that, within the range of 10 to 20 F/mol, the yield of succinic acid did not change but that of oxalic acid greatly

F/mol	Oxalic acid (%)	Succinic acid (%)
10	8.4	8.7
20	2.7	8.3

\* Translated by the author from *J. Chem. Soc. Japan*, **62**(1941), 31.

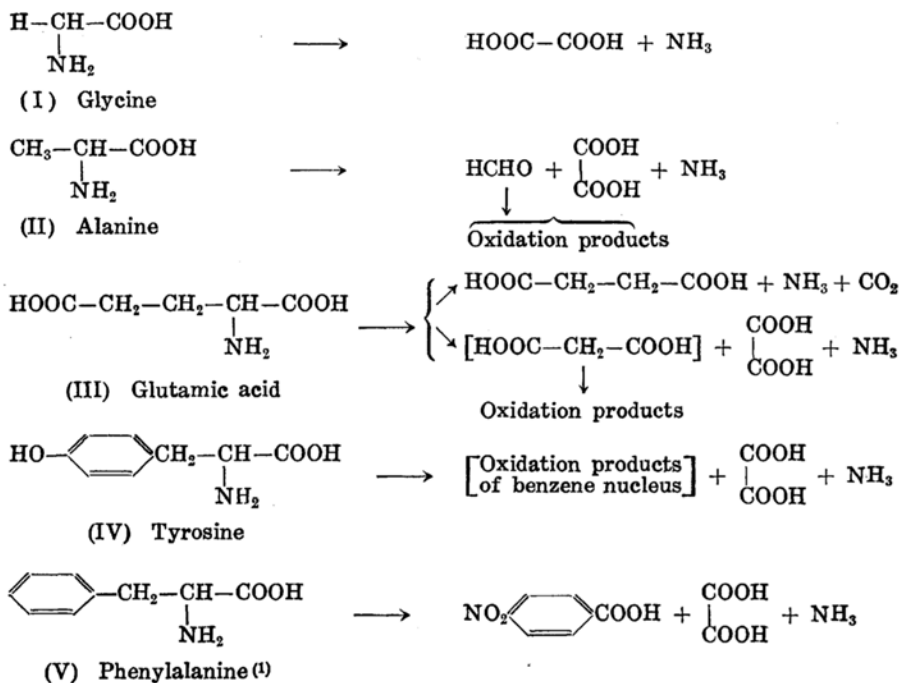
(1) Part XII, this Bulletin, **17**(1942), 45.

(2) C. Th. Mörner, *Z. physiol. Chem.*, **95**(1915), 263.

\*\* The percentages in this paper were expressed in molar per cent.

decreased. It is known that in the electrolysis of succinic acid or oxalic acid in nitric acid, the former is stable, whereas the latter is unstable.<sup>(1)</sup> The fact described above, therefore, may be interpreted as follows: although the formation of oxalic acid from the group  $-\text{CH}(\text{NH}_2)\text{COOH}$  was the principal reaction, yet oxalic acid thus formed was destroyed successively during the electrolysis, consequently it gave small yields.

From these facts it may be quite reasonable to consider that the electrolytic oxidation of glycine, alanine, glutamic acid, tyrosine, and phenylalanine in nitric acid proceeds as follows:



**Experimental.** (1) *Electrolysis of glycine.* 0.7545 g. (10 m. mols) of glycine (Kahlbaum) was dissolved in 30 c.c. of 8 N- $\text{NO}_3\text{H}$  and electrolysed under the conditions: 100°C., 4 amps./dm<sup>2</sup>, 5 F/mol. The electrolysate was neutralised with sodium carbonate solution and acidified with acetic acid. Calcium oxalate was precipitated with the addition of 20% calcium chloride solution; yield 0.5 g. (34%), purity 77.50% (determined by the titration with N/10- $\text{KMnO}_4$ ). The precipitate was decomposed with hydrochloric acid, and extracted with ether. From the ethereal extract crystals were obtained and recrystallised from water; m.p. 101.5–102°C., anhydride m.p. 186–187°C. (decomposed). It was identified as oxalic acid (Found: 0.009345 N- $\text{NaOH}$ , 5.62 c.c. Calc. for  $\text{C}_2\text{O}_4\text{H}_2\cdot 2\text{H}_2\text{O}$ : 0.009345 N- $\text{NaOH}$ , 5.62 c.c.; Found: N/10- $\text{KMnO}_4$ , 2.64 c.c. Calc. for  $\text{C}_2\text{O}_4\text{H}_2\cdot 2\text{H}_2\text{O}$ : N/10- $\text{KMnO}_4$ , 2.63 c.c.).

(2) *Electrolysis of alanine.* 0.9025 g. (101 m. mols) of alanine (Kahlbaum) was dissolved in 30 c.c. of 8 N- $\text{NO}_3\text{H}$  and electrolysed: 100°C., 4 amps./dm<sup>2</sup>, 5 F/mol. During the electrolysis, the volatile oxidation

product was led to two washing bottles containing water, by means of the delivery tube which was connected to the condenser of the cell. The volatile substance caught in water was identified as formaldehyde by the test with fuchsin-sulphurous acid solution and by the mercurous chloride reaction as well as the precipitation of *p*-nitrophenylhydrazone.

The electrolysate was neutralised with sodium carbonate solution and acidified with acetic acid. Oxalic acid was precipitated as in the case of glycine. Calcium oxalate 0.4 g. (27%), purity 94.2%. Oxalic acid was confirmed by the melting point test and by the titrations (Found: 0.009345 N-NaOH, 5.50 c.c. Calc. for  $C_2O_4H_2 \cdot 2H_2O$ : 0.009345 N-NaOH, 5.52 c.c. Found: N/10-KMnO<sub>4</sub>, 2.64 c.c. Calc. for  $C_2O_4H_2 \cdot 2H_2O$ : N/10-KMnO<sub>4</sub>, 2.56 c.c.).

(3) *Electrolysis of glutamic acid.* 3.011 g. (20.5 m. mols) of glutamic acid (N, 9.46%) was dissolved in 30 c.c. of 4N-NO<sub>3</sub>H and submitted to electrolysis: 100°C., 4 amps./dm<sup>2</sup>, 10 F/mol. The electrolysate was neutralised with sodium hydroxide solution and calcium oxalate was precipitated as usual. Calcium oxalate 0.155 g. (air dried), purity 78.8%. Pure calcium oxalate 0.84 m. mol. Free oxalic acid 0.07 g., m.p. 101–102°C. Volatile base NH<sub>3</sub> 0.1 m. mol. After distilling off the volatile base with barium hydroxide, the residue was acidified with sulphuric acid and extracted with ether. 0.3 g. of crystals was obtained from the ethereal extract. It was recrystallised from water and melted at 184°C. It was identified as succinic acid by the mixed melting point test and by the determination of acid equivalent (Found: N/20-NaOH, 6.62 c.c. Calc. for C<sub>4</sub>H<sub>6</sub>O<sub>4</sub>: N/20-NaOH, 6.57 c.c.).

Glutamic acid (3.012 g., 20.5 m. mols) was electrolysed in 8N-NO<sub>3</sub>H (30 c.c.) under the same conditions as in the case of 4N-NO<sub>3</sub>H. From the electrolysate the following substances were obtained. Calcium oxalate 0.3 g. (2.67 m. mols), purity 67.2%. Free acid melted at 101–102°C. (Found: N/10-KMnO<sub>4</sub>, 9.20 c.c. Calc. for  $C_2O_4H_2 \cdot 2H_2O$ : N/10-KMnO<sub>4</sub>, 9.24 c.c.). Succinic acid 0.21 g. (1.76 m. mols). After recrystallising from water it melted at 183°C. (Found: N/10-NaOH, 13.09 c.c. Calc. for C<sub>4</sub>H<sub>6</sub>O<sub>4</sub>: N/10-NaOH, 13.06 c.c.). NH<sub>3</sub> 0.08 m. mol.

Glutamic acid (3.003 g. 20.4 m. mols) was electrolysed in 8N-NO<sub>3</sub>H (30 c.c.) under the conditions: 100°C., 4 amps./dm<sup>2</sup>, 20 F/mol. Calcium oxalate 0.10 g. (0.68 m. mol), purity 79.4%. Succinic acid 0.20 g. (1.6 m. mols), m.p. 181–182°C. It was recrystallised from water and melted at 183°C. (Found: N/10-NaOH 7.24 c.c. Calc. for C<sub>4</sub>H<sub>6</sub>O<sub>4</sub>: N/10-NaOH 7.19 c.c.). NH<sub>3</sub> 0.05 m. mol.

(4) *Electrolysis of succinic acid in nitric acid.* 2.284 g. (19.3 m. mols) of succinic acid was dissolved in 30 c.c. of 8N-NO<sub>3</sub>H and submitted to electrolysis: 100°C., 4 amp./dm<sup>2</sup>, 5 F./mol. When the electrolysate was cooled the colourless crystals were separated. 1.15 g. of crystals were obtained and identified as the unchanged succinic acid (m.p. 184°C.) by the mixed melting point test and by the determination of acid equivalent. From the mother liquor 1.16 g. of succinic acid were recovered by the extraction with ether. Thus the succinic acid was not at all attacked and was recovered quantitatively.

(5) *Electrolysis of tyrosine.* Natural tyrosine (3.612 g., 19.95 m.

mols) was electrolysed in 8N- $\text{NO}_3\text{H}$  (30 c.c.) at  $100^\circ\text{C}$ ., 4 amps./ $\text{dm}^2$ ., 5 F./mol. The electrolysis was undertaken twice (expt. 1 and 2) in the same way. The electrolysate was colourless, but it became greenish yellow when neutralised with sodium hydroxide.

Calcium oxalate 0.33 g., purity 77.2% (1); 0.35 g., purity 82.1% (2). Free oxalic acid m.p.  $101\text{--}102^\circ\text{C}$ . (Found: N/10- $\text{KMnO}_4$  8.00 c.c. (1); 7.44 c.c. (2). Calc. for  $\text{C}_2\text{H}_2\text{O}_4 \cdot 2\text{H}_2\text{O}$ : N/10- $\text{KMnO}_4$  8.07 c.c. (1); 7.38 c.c. (2)). The filtrate of the calcium oxalate was extracted with ether. From the ethereal solution a small quantity of resinous and phenolic substances were obtained.  $\text{NH}_3$  0.2 m. mol.

Tyrosine (20 m. mols) was electrolysed in 8N- $\text{NO}_3\text{H}$  under the conditions:  $100^\circ\text{C}$ ., 2 amp./ $\text{dm}^2$ ., 0.65–3 F./mol. From the electrolysate, picric acid (0.2–0.4 m. mol) and oxalic acid (8.9–4.1 m. mols) were obtained. The formation of the picric acid and the oxalic acid thus obtained were not due to the electrolysis, but to the action of nitric acid upon the tyrosine. The mechanism of the formation of picric acid will be reported in the next paper.

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