376. *The Constitution of Ascorbic Acid. Action of Sodium Hypochlorite on α-Methoxy-acid Amides.*

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The constitutional formula of ascorbic acid which was advanced and fully supported from these laboratories (*J. Soc. Chem. Ind.*, 1933, 52, 221; *Nature*, 1933, **131**, 617) appears to have found universal acceptance. Confirmatory details have since been published by us (J., 1933, 1270) and additional data have been contributed by other workers. Nevertheless, the manner of its final acceptance by certain authors merits comment, and in this connexion attention may be drawn to the publications of Micheel (*Angew. Chem.*, 1934, **47**, 550). Micheel and Kraft (*Nature*, 1933, **131**, 274) had earlier advanced a formula for ascorbic acid which we in these laboratories had been unable to accept (*Nature*, 1933, **131**, 402). After the publication of our results Micheel (*Z. physiol. Chem.*, 1933, **218**, 280) abandoned his formula in favour of the one we had proposed and supported, but in the same paper he advanced what he considered to be important arguments against it. Despite his acceptance of the formula he declared that our method of proof was invalid and he has since endeavoured to provide an alternative proof of the same structure. We should naturally welcome such an alternative proof if it appeared well grounded, but in point of fact no rigid alternative proof has yet been provided by Micheel, who continues, however, to support the constitutional formula which we have developed.

The point initially at issue was Micheel's disbelief in the evidence we provided for the ring structure of ascorbic acid. When it became clear that on oxidation in acid solution ascorbic acid gave rise to a neutral lactone (*J. Soc. Chem. Ind.*, *loc. cit.*), it appeared that ascorbic acid itself contained a lactone ring. Proof of this was furnished by a study of the tetramethyl ether of ascorbic acid which, after ozonisation, gave rise to oxalic acid and 3 : 4-dimethyl *l*-threonic acid. The presence in the latter of a free hydroxyl group at position 2 (the α-position) necessarily gave us the evidence for the presence of the lactone ring connecting carbon atoms 1 and 4 in the original ascorbic acid molecule. The manner of this proof which we furnished was objected to by Micheel. We had shown that the amide of the 3 : 4-dimethyl *l*-threonic acid reacted readily with sodium hypochlorite to give sodium *iso*cyanate in good yield (Weerman reaction). Our deductions from the results were accurate, but Micheel has claimed that other acid amides not possessing a free hydroxyl group in the α-position also give this reaction and he has quoted the example of the methyl ether of mandelamide, which gives benzaldehyde on treatment with sodium hypochlorite. It is important to note, however, that the Weerman reaction for an α-hydroxy-amide requires that sodium *iso*cyanate should be produced. The formation of an aldehyde is immaterial and indeed the production of benzaldehyde from *O*-methylmandelamide can easily follow an ordinary Hofmann degradation, which is the normal course of the reaction in the absence of an α-hydroxy-group.

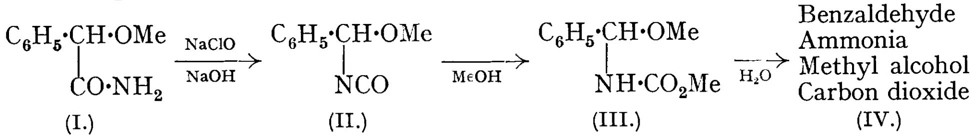
In a subsequent paper (*Ber.*, 1934, **67**, 841) Micheel appears to have realised this, and has proceeded to test for sodium *iso*cyanate in a number of comparative experiments with α-hydroxy- and α-methoxy-acid amides. He has now reached the conclusion, contrary to his previous statement, that it is possible to distinguish between these two classes of amides by the Weerman reaction. He indicates that the yield of sodium *iso*cyanate from α-methoxy-amides is very low, contrasting strongly with the yield from the α-hydroxy-amides, and he now employs methods which conform exactly to the conditions we originally laid down in carrying out this test, which we have frequently found valuable in structural determinations. The validity of the method rests, however, on much stronger foundations than may appear to be the case from Micheel and Kraft's experimental observations. We have repeated their experiments in a number of test cases and we have been unable to substantiate their claim concerning even the partial formation of sodium *iso*cyanate from α-methoxy-amides. We have carried out the Weerman reaction with *O*-methylmandelamide, *O*-methyl-lactamide, 2 : 4-dimethyl *d*-erythronamide, and 2 : 3 : 5-trimethyl *l*-arabonamide and in none of these cases was any trace of sodium *iso*cyanate detectable under the conditions described by Micheel and Kraft. These results exactly confirm our previous experience with the Weerman reaction, namely, that sodium *iso*cyanate is not encountered unless the α-hydroxy-group is present. There seems, therefore, no need to discuss the mechanism which these authors have suggested for the course of the Weerman reaction on α-methoxy-amides, although there are other points of interest which this class of amide presents.

With *O*-methylmandelamide (I), both benzaldehyde and ammonia are formed in quantity. Accompanying the formation of benzaldehyde, carbon dioxide is liberated (ultimately yielding sodium carbonate), but there is no trace of sodium *iso*cyanate. The course of the reaction appears to be that of a normal Hofmann degradation (A) except that in place of the α-methoxy-amine, which would normally be expected as the end product, benzaldehyde and ammonia are obtained.

Micheel and Kraft have now observed that in the presence of methyl alcohol the organic cyanate (II) produced at one stage of the Hofmann degradation reacts additively with the alcohol, giving a urethane to which they ascribe the formula (III). This urethane has been encountered in our experiments also and we find that it readily decomposes, giving benzaldehyde, carbon dioxide, and ammonia (IV). Probably in aqueous solution, water, in a similar way to methyl alcohol, reacts with the organic *iso*cyanate and gives an unstable intermediate compound which breaks down to give ammonia, methyl alcohol, and benzaldehyde. On this view the normal course of the Hofmann degradation will be represented by the series (A) with the possible inclusion of the bracketed formula of a carbamic acid. An exactly similar behaviour was shown in the case of *O*-methyl-lactamide, and these facts are in agreement with the observation (Irvine and Pryde, J., 1924, **125**, 1045; Pryde, J., 1931, 1298) that methylated hexonic acid amides possessing free hydroxy-groups in the γ- or δ-position react with sodium hypochlorite, yielding cyclic urethanes. No loss of methoxyl from the α-methoxyl group occurs and no sodium *iso*cyanate is formed.

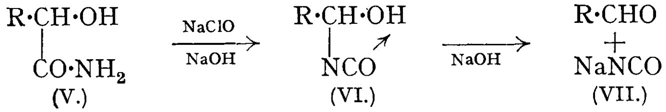
(A) $R{\cdot}CO{\cdot}NH_2 \longrightarrow R{\cdot}NCO \longrightarrow (R{\cdot}NH{\cdot}CO_2H) \longrightarrow R{\cdot}NH_2 + CO_2$

(B)



$$\begin{array}{c}C_6H_5{\cdot}CH{\cdot}OMe\\ |\\ CO{\cdot}NH_2\\ \text{(I.)}\end{array} \xrightarrow[\text{NaOH}]{\text{NaClO}} \begin{array}{c}C_6H_5{\cdot}CH{\cdot}OMe\\ |\\ NCO\\ \text{(II.)}\end{array} \xrightarrow[\text{MeOH}]{} \begin{array}{c}C_6H_5{\cdot}CH{\cdot}OMe\\ |\\ NH{\cdot}CO_2Me\\ \text{(III.)}\end{array} \xrightarrow[H_2O]{} \begin{array}{l}\text{Benzaldehyde}\\ \text{Ammonia}\\ \text{Methyl alcohol}\\ \text{Carbon dioxide}\\ \quad\text{(IV.)}\end{array}$$

(C)



$$\begin{array}{c}R{\cdot}CH{\cdot}OH\\ |\\ CO{\cdot}NH_2\\ \text{(V.)}\end{array} \xrightarrow[\text{NaOH}]{\text{NaClO}} \begin{array}{c}R{\cdot}CH{\cdot}OH\\ |\ \nearrow\\ NCO\\ \text{(VI.)}\end{array} \xrightarrow[\text{NaOH}]{} \begin{array}{c}R{\cdot}CHO\\ +\\ NaNCO\\ \text{(VII.)}\end{array}$$

The Weerman modification of the Hofmann change differs essentially from either of these in that the reaction proceeds with the formation in large amount of sodium *iso*cyanate (Series C). It may be suggested that stage (VI), indicating the formation of a co-ordinated intermediate product in place of a carbamic acid, is responsible for the marked difference in the character of the end products.

EXPERIMENTAL.

O-Methylmandelamide.—Mandelic acid was mixed with an excess of methyl iodide, and silver oxide was added in small quantities to the mixture cooled in ice. The reaction was completed by boiling for 3 hours. The methylation was repeated and the product distilled, giving methyl *O*-methylmandelate, b. p. 92°/0·03 mm. From this the amide was prepared by digestion with methyl-alcoholic ammonia at 0° for a few hours. The *O*-methylmandelamide was recrystallised from alcohol–ether; m. p. 113—114° (yield, 90%).

Sodium Hypochlorite and O-Methylmandelamide.—(*a*) (Compare Micheel and Kraft, *Ber.*, 1934, **67**, 841). *O*-Methylmandelamide (2·20 g.), dissolved in water (185 c.c.), was treated at 0° with a standard solution of sodium hypochlorite (18·5 c.c.). [The sodium hypochlorite solution used throughout these investigations was prepared according to the instructions of Weerman (*Rec. trav. chim.*, 1917, **37**, 16). The hypochlorite present was estimated by titration, and the solution was found to contain 64 g. of sodium hypochlorite in 1000 c.c. of solution.] After 2 hours the solution became milky owing to liberation of benzaldehyde, and the reaction was almost complete in 6 hours. The solution was filtered from a little unchanged amide (0·05 g.) and traces of sodium hypochlorite were removed by the addition of *N*/10-sodium thiosulphate (10 c.c.). Semicarbazide hydrochloride (5·50 g.), dissolved in water (20 c.c.), was added to the solution, and after 3 hours at 0° the crystalline deposit was filtered off and dried. M. p. 220—221°. Yield 1·90 g. (almost theoretical). The crystals dissolved readily in hot absolute alcohol (60 c.c.), leaving no residue, and on cooling, pure benzaldehydesemicarbazone, m. p. 222°, was deposited. The aqueous filtrate was kept at 0° for 2 days, but no further solid matter was deposited. It was shown that hydrazodicarbonamide is insoluble in boiling absolute alcohol and can be separated in the presence of a large excess of benzaldehydesemicarbazone. One part of hydrazodicarbonamide mixed with 250 parts of benzaldehydesemicarbazone is readily detectable.

(*b*) The action of sodium hypochlorite on *O*-methylmandelamide was repeated at 25°, the same quantities as before being used. The reaction was complete in about 4 hours. The deposit of benzaldehydesemicarbazone (1·95 g.) was entirely soluble in hot absolute alcohol (50 c.c.) and no trace of hydrazodicarbonamide was found.

(*c*) *O*-Methylmandelamide (0·70 g.) was treated at 0° with water (4 c.c.) and sodium hypochlorite solution (5·0 c.c.) and after 6 hours any free sodium hypochlorite was exactly decomposed with the requisite amount of sulphurous acid. After filtration, the benzaldehyde was removed from the alkaline filtrate by exhaustive extraction with ether. The aqueous solution was treated with an excess of solid sodium acetate and a saturated solution of semicarbazide hydrochloride (0·5 g.) in water (2 c.c.); a marked effervescence then occurred (carbon dioxide liberated from the carbonate formed during the Hofmann reaction). The clear solution was kept at 0° for several hours, but no solid was deposited. The semicarbazide was present unchanged in the solution, since, after the addition of two drops of benzaldehyde, an almost quantitative yield of benzaldehydesemicarbazone, m. p. 222—223°, was obtained.

It was shown in the following way that extraction with ether did not remove any sodium *iso*cyanate from the reaction solution. *l*-Galactonamide (0·50 g.) was treated at 0° with water (3 c.c.) and sodium hypochlorite (3·0 c.c.). The reaction was complete after 30—40 minutes; the solution was then extracted seven or eight times with a large excess (100 c.c.) of ether and treated with solid sodium acetate and a saturated solution of semicarbazide hydrochloride in water. No effervescence occurred on the addition of these reagents. Crystalline hydrazodicarbonamide separated in a few moments and was recrystallised from aqueous alcohol. M. p. 254—255° (decomp.). Yield, 0·17 g.

It was also demonstrated that the presence of benzaldehyde does not in any way affect the formation of sodium cyanate and its isolation as hydrazodicarbonamide. *l*-Galactonamide (0·50 g.) was treated with sodium hypochlorite as described above and after 5 minutes 3 or 4 drops of benzaldehyde were added to the solution. The mixture was kept at 0° for 3 hours and then extracted with a large excess of ether. Solid sodium acetate and a saturated solution of semicarbazide hydrochloride were added to the solution (no effervescence), and the crystalline deposit of hydrazodicarbonamide filtered off and recrystallised. M. p. 254—255°. Yield, 0·16 g.

Formation of Ammonia during the Action of Sodium Hypochlorite on O-*Methylmandelamide.*—*O*-Methylmandelamide (0·70 g.) was treated at 0° with the requisite amount of sodium hypochlorite and after 6 hours the solution was extracted thoroughly with ether. The aqueous solution was concentrated to dryness and the solid residue was shown to be sodium chloride containing traces of nitrite. The presence of the latter salt was proved by obtaining positive results with the Griess–Ilosvay test (pink coloration) and the Griess test (brown coloration). The ethereal extract containing benzaldehyde was treated with a small amount of hydrogen chloride and the white solid which was precipitated was removed by shaking the ethereal solution with a little water. The aqueous solution was concentrated, the free hydrochloric acid removed by repeated distillation with water, and on evaporation to dryness a white crystalline residue was obtained which was shown to be pure ammonium chloride.

A quantitative investigation into the formation of ammonia in this reaction was carried out in the following way. *O*-Methylmandelamide (0·620 g.) was allowed to react at 0° for 20 hours with the requisite amount of sodium hypochlorite. The reaction was then complete, only a trace of free hypochlorite being present (removed by the addition of 0·1*N*-sodium thiosulphate solution). The solution was made more alkaline with sodium hydroxide, and the ammonia distilled off into a known amount of acid (15·3 c.c. of 1·128*N*-sulphuric acid), the excess of which was titrated with alkali (14·4 c.c. of 0·985*N*-sodium hydroxide). Yield of ammonia, 81·6% of the theoretical. The deficiency is probably due to the oxidation of some ammonia by sodium hypochlorite.

Sodium Hypochlorite and O-*Methylmandelamide in the Presence of Methyl Alcohol* (compare Micheel and Kraft, *loc. cit.*).—*O*-Methylmandelamide (2·19 g.), dissolved in methyl alcohol (25 c.c.), was treated at 0° with sodium hypochlorite (15·0 c.c.). After 4 hours (hypochlorite present, 2·5% of original amount) the crystalline deposit was filtered off; m. p. 68—69°, yield 1·10 g. (45% theory) (Found: C, 61·6; H, 6·8; N, 7·2; OMe, 31·8. Calc. for $C_{10}H_{13}O_3N$: C, 61·5; H, 6·7; N, 7·2; OMe, 31·7%). The aqueous filtrate was treated with sodium acetate and semicarbazide hydrochloride, and after 1 hour at 0° the benzaldehydesemicarbazone was filtered off; m. p. 223°. Yield, 0·96 g. (47% theory). The semicarbazone dissolved completely in hot absolute alcohol, no residue remaining. No trace of hydrazodicarbonamide was present and no sodium *iso*cyanate was formed during the reaction.

The crystalline urethane derivative was readily soluble in most of the common solvents, but only moderately soluble in light petroleum, and was insoluble in water. It was most readily recrystallised from a cold aqueous alcoholic solution by cooling, but successive recrystallisations left the m. p. 68—69° unchanged. The crystals were unaffected by boiling in aqueous solution for a short time. On treating the substance with *N*-sodium hydroxide in the cold (0°) decomposition occurred with quantitative formation of benzaldehyde and ammonia. On testing the solution with sodium acetate and semicarbazide hydrochloride, no trace of hydrazodicarbonamide could be detected. The decomposition of the urethane proceeded, therefore, without the formation of sodium cyanate.

Sodium Hypochlorite and 2 : 3 : 5-*Trimethyl Arabonamide.*—2 : 3 : 5-Trimethyl arabonamide (0·30 g.), dissolved in water (1 c.c.), was treated at 0° with sodium hypochlorite (1·80 c.c.). After 6 hours the solution still contained much free hypochlorite and the reaction was not complete after 2 days. The small quantity of sodium hypochlorite was exactly decomposed with the requisite amount of sulphurous acid, and solid sodium acetate and semicarbazide hydrochloride (0·16 g.) in water were added to the reaction solution. No trace of solid matter could be detected and no crystalline deposit formed after several hours at 0°. The semicarbazide hydrochloride was present unchanged in the solution, as shown by the instant formation of benzaldehydesemicarbazone on the addition of one spot of benzaldehyde. Precisely similar results were obtained with 2 : 4-dimethyl *d*-erythronamide (compare Avery, Haworth, and Hirst, J., 1927, 2316).

Sodium Hypochlorite and O-*Methyl-lactamide.*—Silver lactate was allowed to react with methyl iodide and the ester so produced was methylated in the usual way with silver oxide and methyl iodide, giving methyl *O*-methyl-lactate. The yield was poor and it was not appreciably increased when the preliminary stages of the methylation were carried out with the aid of methyl sulphate and sodium hydroxide. By the action of methyl-alcoholic ammonia on the ester, *O*-methyl-lactamide was obtained. After several recrystallisations from alcohol–ether (which removed traces of lactamide), the m. p. was 84°. The value recorded in the literature is 79—81° (Found : OMe, 29·7. Calc. for $C_4H_9O_2N$: OMe, 30·1%).

(*a*) (According to the directions of Micheel and Kraft.) *O*-Methyl-lactamide (0·70 g.), dissolved in water (3 c.c.), was treated at 0° with sodium hypochlorite (8·4 c.c.). A moderately vigorous reaction took place and after 30 minutes only a small amount of sodium hypochlorite remained. The reaction was practically complete after 5 hours, the final traces of sodium hypochlorite being then removed with 0·1*N*-sodium thiosulphate (1 c.c.). Semicarbazide hydrochloride (1·60 g.) in water (3 c.c.) was added to the solution, causing a brisk effervescence (carbon dioxide, from carbonate produced during the Hofmann reaction). Although the solution was kept at 0° for several hours, no trace of any solid material could be detected. Semicarbazide was present unchanged in the solution, as shown by the instant formation of benzaldehydesemicarbazone on the addition of a little benzaldehyde. Precisely similar results were obtained when the experiment was carried out under the conditions specified in our former papers (semicarbazide added after addition of an excess of sodium acetate to the reaction mixture).

(*b*) *O*-Methyl-lactamide (0·20 g.), dissolved in water (1 c.c.), was treated at 0° with sodium hypochlorite (2·40 c.c.) and after 6 hours the last traces of hypochlorite were removed with a few drops of 0·1*N*-sodium thiosulphate. The presence of acetaldehyde (recognised by its characteristic smell) was demonstrated by testing the solution with (i) Schiff's reagent (red colour immediately restored), (ii) an aqueous solution of sodium nitroprusside containing piperidine (deep blue coloration), (iii) a dilute aqueous solution of benzidine hydrochloride (yellow coloration), (iv) ammoniacal solution of silver (immediate reduction in the cold with formation of a mirror). The tests were simultaneously carried out with a solution of acetaldehyde (0·25 g.) in water (15 c.c.), and from a comparison of the colorations produced in the two series it was evident that the concentration of acetaldehyde in both solutions was of the same order.

(*c*) *O*-Methyl-lactamide (0·30 g.), dissolved in water (1 c.c.), was treated at 0° with sodium hypochlorite (3·60 c.c.) and after 6 hours the trace of free hypochlorite was removed by the addition of one drop of sulphurous acid. The solution was made more alkaline with sodium hydroxide and distilled in steam. Any volatile products were swept by a slow stream of nitrogen into a receiver containing dilute hydrochloric acid. The distillate was concentrated at 40°, hydrochloric acid being removed by repeated distillation with water. A white crystalline residue was obtained which was easily recrystallised from aqueous alcohol. Yield, 0·13 g. This was definitely ammonium chloride (and not an amine hydrochloride) (Found : 0·0527 g. required 9·8 c.c. of 0·1*N*-sodium hydroxide. Calc. for ammonium chloride, 9·9 c.c.).

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