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## ACTIONS OF IONIZING RADIATIONS ON NITROGEN COMPOUNDS IN AQUEOUS MEDIA

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## ABSTRACT

Most of the nitrogen-containing compounds that have been studied in radiation chemistry may be formally classified as derivatives of ammonia. The actions of ionizing radiations on this system of nitrogen compounds in aqueous media have been related in terms of the initial reaction $H_2O \rightsquigarrow H_2, H_2O_2, H^+, OH, e^-_{aq}$. Substances that have been treated range in complexity from ammonia on the one hand to the N-heterocyclics on the other. Emphasis has been placed on the reactions of $e^-_{aq}$ with the various types of nitrogen function.

# ACTIONS OF IONIZING RADIATIONS ON NITROGEN COMPOUNDS IN AQUEOUS MEDIA†

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May 1963

Most of the nitrogen-containing compounds that have been studied in radiation chemistry—and particularly those that have been studied because of their biological interest—may be formally classified as organic derivatives of ammonia i.e., as $RNH_2$, $R_2NH$, $R_3N$. The radiation chemistry of these compounds in aqueous media is frequently determined by the nature of the substituent groups R. However, under certain conditions, the reactivity of the nitrogen locus is manifested as the characteristic radiation-chemical property. Hence, we consider here first the radiation-induced reactions of the parent compound, ammonia, and its closely related inorganic derivatives, hydrazine and hydroxylamine.

It is assumed in accord with current theory[1-3] that the earliest chemically detectable products of the decomposition of liquid water by ionizing radiations are given by

$$(0.1)\quad H_2O \rightsquigarrow H_2, H_2O_2, H^+, OH, e^-_{aq}$$

where $e^-_{aq}$ represents the hydrated electron. The formation of H atoms in irradiated water is then attributed to the secondary reaction

$$(0.2)\quad H_3O^+ + e^-_{aq} \rightarrow H + H_2O$$

In developing the present discussion, an effort was made to treat separately all of the possible reactions of OH, H and $e^-_{aq}$ with each nitrogen

compound in its various possible forms in aqueous solution. However, in most cases the available data are not complete enough to permit such a uniform and symmetrical treatment. The reactions, therefore, have been grouped arbitrarily simply from the standpoint of facilitating discussion.

(1.) Ammonia

(1.1) $NH_3 + OH \rightarrow NH_2 + H_2O$

(1.2) $NH_4^+ + OH \rightarrow NH_3^+ + H_2O$

Riggs, Stein and Weiss[4] established in 1952 that ammonia undergoes a radiation-induced oxidation to yield nitrite ion in oxygenated solution. Through interrelated studies of the effects of pH and solute concentration it was established that $G(NO_2^-)$ is dependent on the concentration of free base as given by the equilibrium $NH_3 + H_2O \rightarrow NH_4^+ + OH^-$. For example, it was found that the production of nitrite ion in a dilute solution of ammonium sulfate ($5 \times 10^{-4}$ M) at pH 9.62 follows practically the same course as in a more concentrated solution ($10^{-1}$ M) at pH 6.90; the concentration of free base in these two solutions is approximately the same. Removal of OH radicals, then, in the radiolysis of ammonia solutions apparently occurs exclusively through hydrogen abstraction from the free base via reaction (1.1). The mechanism proposed for the radiolysis of oxygenated solutions of ammonia includes reaction (1.1) and the steps

$$NH_2 + O_2 \rightarrow NH_2O_2$$

$$NH_2O_2 + O_2 \rightarrow HNO_2 + HO_2$$

$$H(e^-) + O_2 \rightarrow HO_2(O_2^-)$$

$$HO_2 \rightleftarrows O_2^- + H^+$$

$$2HO_2 \rightarrow H_2O_2 + O_2$$

The conclusion by Riggs et al. that the ammonium ion is inert toward the hydroxyl radical is in accord with recent studies which give $D(NH_3^+-H) \simeq 130$ kcal[5] as compared to $D(HO-H) \simeq 116$ kcal.[6]

(1.3) $NH_3 + H \rightarrow NH_2 + H_2$

(1.4) $NH_4^+ + H \rightarrow NH_3^+ + H_2$

The available experimental evidence indicates that neither $NH_3$ nor $NH_4^+$ reacts with atomic hydrogen in aqueous solution. Collinson and Dainton[7] concluded that H atom removal via reaction (1.3) does not occur even in liquid ammonia. Reaction (1.4) is estimated from the work of Tal'roze and Frankevich[5] to be endothermic by about 27 kcal.

(1.5) $NH_4^+ + e_{aq}^- \rightarrow NH_3 + H$

Jortner et al.[8] investigated the effects of $NH_4^+$ on hydrogen production in the $\gamma$-radiolysis of oxygen-free solutions of formic acid at pH values above 7. They found $G(H_2) \simeq 3.0$ for $10^{-2}$ M ammonium formate as compared to $G(H_2) \simeq 1.8$ for $10^{-2}$ M sodium formate under identical conditions. This increase in $G(H_2)$ is attributed to the formation of H atoms via reaction (1.5) followed by

$$H + HCOO^- \rightarrow H_2 + COO^-$$

The rate of conversion of $e_{aq}^-$ to H by ammonium ion is estimated by Jortner et al. to be about $10^{-4}$ times the rate of conversion of $e_{aq}^-$ by the hydronium ion in reaction (0.2).

(2) Hydrazine

(2.1) $N_2H_5^+ + OH \rightarrow N_2H_4^+ + H_2O$

(2.2) $N_2H_5^+ + H \rightarrow N_2H_4^+ + H_2$

Dewhurst and Burton[9] in 1955 found that the radiolysis of oxygen-free solutions of hydrazine could be interpreted over a wide range of pH in terms of reactions (2.1) and (2.2) followed by the steps

$$N_2H_4^+ \rightarrow N_2H_3 + H^+$$

$$2N_2H_3 \rightarrow N_2 + 2NH_3$$

Product yields with $Co^{60}$ $\gamma$-rays were found to be strictly independent of pH through the range 1 to 8 with $G(H_2) \simeq 2.5, G(N_2) \simeq 2.5, G(NH_3) \simeq 5.2$. These results were in reasonable agreement with the theory of water decomposition generally accepted at that time—namely that H and OH were the primary products of water radiolysis.[10] It is interesting, in retrospect, to observe that of the various solutes then known to react with H via

$$RH + H \rightarrow R + H_2$$

only hydrazine failed to show the characteristic decrease in $G(H_2)$ with increasing pH above values of 3 to 4.

(2.3) $N_2H_5^+ + e^-_{aq} \rightarrow N_2H_4 + H$

(2.4) $N_2H_5^+ + e^-_{aq} \rightarrow N_2H_3 + H_2$

The data of Dewhurst and Burton are consistent with the present concepts of the primary process in water if the assumption is made that reactions (2.3), (2.4) are in competition with reaction (0.2) in the removal of $e^-_{aq}$. The detailed reaction scheme otherwise remains the same. Since the basic dissociation constant of hydrazine corresponds[11] to $K \simeq 0.5 \times 10^{-7}$ it is clear that the concentration of $N_2H_5^+$ is not greatly affected by pH below a value of ~7. The observed decrease in product yields at higher pH values

occurs over the pH range in which $N_2H_5^+$ is neutralized to give the free base.

(2.5) $N_2H_5^+ + e^-_{aq} \rightarrow NH_2 + NH_3$

(2.6) $N_2H_4 + e^-_{aq} \rightarrow NH_2 + NH_2^-$

If reactions (2.5), (2.6) (followed by $NH_2 + N_2H_5^+ \rightarrow NH_3 + N_2H_4^+$) also occurred in competition with reaction (0.2) and (2.3) then $G(NH_3)$ would gradually increase with pH at the expense of $G(H_2)$ and $G(N_2)$. The data of Dewhurst and Burton show no indication of such a trend.

(3.) Hydroxylamine.

(3.1) $NH_3OH^+ + OH \rightarrow NH_2OH^+ + H_2O$

(3.2) $NH_3OH^+ + H \rightarrow NH_3^+ + H_2O$

Lefort and Tarrago[12] have shown that the production of $N_2$ and $NH_3$ in dilute aqueous solution of hydroxylamine under $\gamma$-rays increases abruptly with increasing pH above ~3 and then levels off at pH values above ~6 to give the limiting yields: $G(N_2) \simeq 2.7$, $G(NH_3) \simeq 2.7$. This effect of pH is interpreted by Lefort and Tarrago as evidence for the equilibrium

$$NH_3^+ + H_2O \rightleftarrows NH_2 + H_3O^+$$

That is, decomposition of hydroxylamine is not observed at low pH because of a reconstitution reaction between $NH_3^+$ and the product of reaction (3.1) i.e.

$$NH_2OH \rightarrow NHOH + H^+$$

$$NH_3^+ + NHOH + H_2O \rightarrow NH_3OH^+ + NH_2OH$$

Lefort and Tarrago proposed that $NH_3^+$ does not react with $NH_3OH^+$ because of charge repulsion. However, as the pH is increased, $NH_3^+$ dissociates to $NH_2$ in which form it is assumed to react preferentially with $NH_3OH^+$ and/or $NH_2OH$

$$NH_2 + NH_3^+OH \rightarrow NH_3 + NH_2O + H^+$$

to give nitrogen through the step,

$$2NH_2O \rightarrow N_2 + 2H_2O$$

(3.3) $NH_3OH^+ + e^-_{aq} \rightarrow NH_2 + H_2O$

The data of Lefort and Tarrago may also be explained in terms of electron capture by $NH_3OH^+$ as indicated by equation (3.3). On this assumption the observed pH effect would arise as a result of a competition between $NH_3OH^+$ and $H_3O^+$ for $e^-_{aq}$. Subsequent steps leading to observed products would be essentially the same as those given above. However, the reconsitution reaction in acid solution would in this case correspond to

$$H + NH_2OH^+ \rightarrow NH_3OH^+$$

or the equivalent.

(4.) <u>Amino Acids</u>

(4.1) $NH_3^+CH(R)COO^- + OH \rightarrow NH_3^+\dot{C}(R)COO^- + H_2O$

The radiation-induced reactions of glycine and alanine in neutral solution containing oxygen lead to the formation of ammonia, α-ketoacid, aldehyde, and hydrogen peroxide.[13,14] Both the qualitative and quantitative aspects of the degradation are consistent with a mechanism[15] involving hydrogen abstraction by OH at the α-carbon position via reaction (4.1) and the steps

$$NH_3^+\dot{C}(R)COO^- + O_2 \rightarrow NH_3 + RCOCOOH + HO_2$$

$$\rightarrow NH_3 + RCHO + CO_2 + HO_2$$

$$e^-(H) + O_2 \rightarrow O_2^-(HO_2)$$

$$O_2^- + H^+ \rightleftarrows HO_2$$

$$2HO_2 \rightarrow H_2O_2 + O_2$$

The reaction between $NH_3^+\dot{C}(R)COO^-$ and $O_2$ as written above represents only the over-all stoichiometry. A detailed consideration of the nature of the intermediate involved in cleavage of the N-C bond is given elsewhere;[16] the main point here is that all OH radicals appear to react preferentially at the $\alpha$-carbon position of the simple amino acids in the zwitterion form. Maxwell et al.[13] report for 1M glycine under $\gamma$-rays the values $G(NH_3) \simeq 4.3$, $G(\Sigma C{=}O) \simeq 4.5$ $G(H_2O_2) \simeq 3.6$. Similar results[14] are obtained with alanine at concentrations above $\sim 0.1$M. Other amino acids give lower ammonia yields as a result of OH attack at side-chain loci.[16] Product yields from solutions of the zwitterions of glycine and alanine are markedly dependent on solute concentration and do not begin to level off at concentrations much below 0.5M and 0.1M respectively.* This apparent low reactivity has been shown by Rabani and Stein[17] to be a property of the zwitterion forms. They found

*At these concentrations of dipolar ions the dielectric constant of the solution is considerably increased over that for pure water. The molar dielectric increment for glycine is ~25 and it would seem that the modes of water decomposition might be appreciably modified by dipole solutes particularly in view of the marked influence of both dielectric constant and relaxation time on primary processes in a condensed phase.

that the velocity of the reaction of OH with the zwitterions of glycine and alanine are respectively ~500 and ~30 times slower than the velocity of the corresponding reactions with the negative ion forms. Actually, as indicated by equations (4.2), (4.3) below, reaction of the amino acid negative-ion involves two loci—the N-H bond of the uncharged amino group as well as the C-H bond at the α-carbon position.

$$(4.2)\quad NH_2CH(R)COO^- + OH \rightarrow NH_2\dot{C}(R)COO^- + H_2O$$

$$(4.3)\quad NH_2CH(R)COO^- + OH \rightarrow \dot{N}HCH(R)COO^- + H_2O.$$

Jayson et al.[18] found in their radiolysis studies of alkaline solutions of the α-amino acids that hydroxylamine as well as ammonia is produced in the presence of oxygen. Formation of hydroxylamine is associated with OH attack at the uncharged amino group via reaction (4.3) followed for example by

$$\dot{N}HCH(R)COO^- + O_2 \rightarrow O_2NHCH(R)COO^-$$

$$O_2NHCH(R)COO^- + HO_2 \rightarrow NH_2OH + RCOCOO^- + O_2$$

Although this effect of pH on the reactivity of the nitrogen locus is analogous to that involved in the radiolysis of aqueous ammonia, it should be noted that the reaction of OH with glycine and alanine even at high pH values still occurs predominantly at the α-carbon atom.

$$(4.4)\quad NH_3^+CH(R)COO^- + e^-_{aq} \rightarrow H + NH_2CH(R)COO^-$$

$$(4.5)\quad \rightarrow H_2 + NH_2\dot{C}(R)COO^-$$

$$(4.6)\quad \rightarrow NH_3 + \dot{C}H(R)COO^-$$

Several years ago Maxwell and co-authors[19] first established the principal stoichiometric relationships involved in the over-all radiation chemistry of oxygen-free solutions of glycine under $\gamma$-rays. Major products from 1.0M solutions at pH 6.5 include hydrogen, ammonia, acetic acid, glyoxylic acid, formaldehyde and carbon dioxide. Shortly thereafter it was shown[20] that these chemical changes could be interpreted both qualitatively from known values of the radical yields in water radiolysis and the reaction scheme:

$$H + NH_3^+CH(R)COO^- \rightarrow NH_4^+ + \dot{C}H(R)COO^-$$

$$\rightarrow H_2 + NH_3^+\dot{C}(R)COO^-$$

$$OH + NH_3^+CH(R)COO^- \rightarrow H_2O + NH_3^+\dot{C}(R)COO^-$$

$$\dot{C}H(R)COO^- + NH_3^+CH(R)COO^- \rightarrow RCH_2COO^- + NH_3^+\dot{C}(R)COO^-$$

$$2\,NH_3^+\dot{C}(R)COO^- \rightarrow NH_2^+{=}C(R)COO^- + NH_3^+CH(R)COO^-$$

$$H_2O_2 + NH_3^+\dot{C}(R)COO^- \rightarrow NH_2^+{=}C(R)COO^- + H_2O + OH$$

$$H_2O + NH_2^+{=}C(R)COO^- \rightarrow NH_3 + RCOCOOH$$

$$\rightarrow NH_3 + RCHO + CO_2$$

Independent evidence for the proposed intermediates $\dot{C}H(R)COOH$ and $NH_2\dot{C}(R)COOH$, is to be found in the observation[20] that succinic acid, aspartic acid, and diaminosuccinic acid are produced in the radiolysis of oxygen-free solutions of glycine, albeit in relatively low yield.

The mechanism given above is also applicable with minor adjustments in the two branching ratios to corresponding data[21] obtained in the $\gamma$-radiolysis of aqueous solutions of alanine.

It is obvious, however, that this reaction scheme must be revised to take into account the fact that the reducing species initially formed in water radiolysis is $e^-_{aq}$ rather than H. The question here of course is: what are the reactions of $e^-_{aq}$ in the essentially neutral solutions of the zwitterion that are stoichiometrically equivalent to the H atom reactions formulated in the original mechanism?

One possibility is that the $NH_3^+$ group of the amino acid simply reacts as an acid with $e^-_{aq}$ to yield H much in the same way that $NH_4^+$ has been shown to react. The mechanism then would stand as written except for the prior addition of reaction (4.4). Dr. B. M. Weeks and Mrs. Sibyl Cole of this laboratory have investigated this possibility in the following way. To a series of 1.0M alanine solutions containing $NH_2CH(CH_3)COOH$ was added sodium formate in increasing amounts over the concentration range $5\times10^{-3}$M to 1.5M. These solutions at a pH of ~6.5 were evacuated and irradiated with $Co^{60}$ γ-rays for a dose of ~$1.5\times10^{19}$ ev/ml. Now, if the zwitterion of alanine simply converts $e^-_{aq}$ to H which then reacts with $NH_3^+(CH(CH_3)COO^-$ we would expect the yield of propionic acid to decrease with increasing formate concentration because a competing path for H removal has been introduced viz

$$H + HCOOH \rightarrow H_2 + COOH$$

We have found, however, that the value G(propionic) ≃ 1.0 is essentially independent of formate concentration although the value of $G(NH_3)$ decreases rapidly with increasing formate and reaches a minimal value at formate concentration of ~0.1M. These results would suggest that $e^-_{aq}$ reacts directly with $NH_3^+CH(CH_3)COO_-$ via reaction (4.6) to yield propionic acid without the

intermediate formation of a freely diffusing H atom.*

Additional evidence in support of this conclusion has been obtained by Dr. Weeks from studies of the interrelated effects of alanine concentration and pH on propionic acid yields. For example in 0.1M alanine at pH 0.5 the yield of propionic acid corresponds to $G \simeq 0.05$. The yield of propionic acid increases with pH and reaches a maximum of $G \simeq 1.0$ at pH 3.5. From the initial slope of the pH-yield curve it is estimated that the rate of reaction of alanine [in this case as the cation $NH_3^+CH(CH_3)COOH$] with $e^-_{aq}$ is approximately $10^{-2}$ that of the reaction of $e^-_{aq}$ with $H_3O^+$.** However, it appears that the yield of propionic acid is not determined exclusively by a simple competition between the amino acid and the hydronium ion for $e^-_{aq}$. This is evidenced by the fact the corresponding pH-yield plot for a 1.0M alanine solution over the same pH range does not coincide exactly with the pH-yield plot for 0.1M alanine when displaced one pH unit. Alanine in 1.0M solutions at low pH seems to be proportionately more effective than the proton in scavenging the decomposition products from water.

(5.) Primary Amines

(5.1) $NH_3^+CH_2R + OH \rightarrow NH_3^+\dot{C}HR + H_2O$

(5.2) $NH_2CH_2R + OH \rightarrow NH_2\dot{C}HR + H_2O$

(5.3) $\rightarrow \dot{N}HCH_2R + H_2O$

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*These results suggest that in the radiolysis of amino acids in the solid state electron capture may occur via $NH_3^+CH(R)COO^- + e^- \rightarrow NH_3 + \dot{C}H(R)COO^-$.

**It is assumed here that the slowness of the reaction,[22] $H + H^+ \rightarrow H_2^+$ precludes any major contribution of $H_2^+$ as a reactive intermediate in this system.

Although the primary aliphatic amines have been studied much less extensively than the amino acids there seems to be little question but that the radiation-chemical properties of the two classes of compounds are quite analogous—except, of course, for the unique property of low reactivity exhibited by the amino acid zwitterions. Jayson et al.[18] have studied the X-ray-induced formation of acetaldehyde from ethylamine in oxygenated solution over a wide range of pH. At pH values below 7 the removal of OH appears to occur via reaction (5.1) to give acetaldehyde and ammonia according to

$$NH_3^+\dot{C}HR + O_2 + H_2O \rightarrow NH_4^+ + RCHO + HO_2$$

As the pH is increased above 7, hydroxylamine appears as a product in addition to ammonia and the yield of the former reaches a limiting value of $G \simeq 0.6$ at pH 10 under which condition $G(CH_3CHO) \simeq 2.2$. Since the basic dissociation constant of ethylamine is $5.6 \times 10^{-4}$ it is assumed that reactions (5.2) and (5.3) occur in parallel at high pH to give ammonia, hydroxylamine and acetaldehyde through intermediates analogous to those considered in greater detail in Section (4).

$$(5.4) \quad NH_3^+CH_2R + e^-_{aq} \rightarrow NH_3 + \dot{C}H_2R$$

Although reactions of type (5.4) are apparently of importance in the radiolysis of oxygen-free solution of the amino acids, there is no direct experimental evidence either for or against the occurrence of this reaction in solution of the primary amines. In fact, it would appear from the literature that radiation-chemical studies of aliphatic amines in oxygen-free solution have not been undertaken.

There are however certain unexplained aspects of the data obtained by Jayson et al.[18] in their studies of oxygenated solutions of ethylamine

that appear now to be related to reaction (5.4). Before considering these we first consider briefly some general features of the radiation-induced oxidation of organic compounds in aqueous solution. Now, the oxidation of quite a large number of compounds can be represented in terms of the generalized scheme.[23]

$$RH + OH \rightarrow R^{\cdot} + H_2O$$

$$R^{\cdot} + O_2 \rightarrow R'H + HO_2$$

$$e^-_{aq} + O_2 \rightarrow O_2^-$$

$$e^-_{aq} + H_3\overset{+}{O} \rightarrow H + H_2O$$

$$H + O_2 \rightarrow HO_2$$

$$HO_2 \rightleftarrows H^+ + O_2^-$$

$$2HO_2 \rightarrow H_2O_2 + O_2$$

On the basis of this scheme, product yields would be independent of pH with $G(R'H) \simeq G_{OH}$, $G(H_2O_2) \simeq G_H + G_{OH} + G_{H_2O_2}$. The oxidation of formic acid[24] to carbon dioxide and hydrogen peroxide and of ethanol[25] to acetaldehyde and hydrogen peroxide conform to this generalized mechanism over the pH range 1 to 13.*

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*Somewhat higher product yields are observed at a low pH. These effects are attributed to an influence of pH on the primary yield of decomposition products from water.[26]

We have seen also that the radiation-induced deamination of glycine and alanine in oxygenated solution at pH 7 is in accord with this reaction scheme although the effects of pH in the radiolysis of amino acid solutions have not as yet been examined in any detail. However, Jayson et al.[18] in their early studies of the effects of X-rays on ethylamine in oxygenated solution report yields of both hydrogen peroxide and acetaldehyde over the pH range 1 to 12. The interesting thing here is that they report $G(CH_3CHO)$ to be essentially constant over the pH range 1 to 7 whereas $G(H_2O_2)$ drops rapidly from a value of ~4.2 at pH 1 to ~1.1 at pH 7. Since the ethylamine is essentially all in the salt form, $NH_3^+CH_2R$, over this pH range, the effect of pH on $G(H_2O_2)$ cannot be attributed to reactions involving OH—particularly in view of the constancy of $G(CH_3CHO)$. But, if the cation of the amine scavenges $e^-_{aq}$ via reaction (5.4) in competition with $O_2$ and $H_3O^+$ it follows that the course of the reaction will change as the pH is increased. The decrease of $G(H_2O_2)$ then would follow as a consequence of reaction (5.4) if as seems likely the subsequent reaction of $\dot{C}H_2R$ with oxygen gives an organic peroxide instead of hydrogen peroxide. Miss Winifred Bennett of this laboratory has reinvestigated the radiation chemistry of oxygenated ethylamine solutions from this standpoint and in fact finds that the yield of <u>total</u> peroxide (hydrogen peroxide plus organic peroxide) at pH 6.5 corresponds to $G \simeq 3.3$ for $Co^{60}$ $\gamma$-rays. From the slope of the pH-yield plot for hydrogen peroxide production it is estimated on the basis of the above argument that the rate of the reaction of $NH_3^+CH_2CH_3$ with $e^-_{aq}$ is $\sim 10^{-2}$ that of the reaction of $e^-_{aq}$ with $H_3O^+$. It is noted that a value of $10^{-2}$ for this ratio was also estimated for alanine in the cation form (Section 4).

(6.) Secondary Amines

(6.1) $RNH_2^+CH_2R + OH \rightarrow RNH_2^+\dot{C}HR + H_2O$

(6.2) $RNHCH_2R + OH \rightarrow RNH\dot{C}HR + H_2O$

(6.3) $\rightarrow R\dot{N}CH_2R + H_2O$

A principal radiation-induced reaction of secondary amines in oxygenated solution leads to formation of primary amine and aldehyde.[18,27] The degradation processes are related to those observed with the amino acids. Primary OH attack occurs predominantly at the carbon position $\alpha$ to nitrogen in the simpler amines. Diethylamine, for example, yields ethylamine and acetaldehyde throughout the entire pH range. Degradation apparently involves reaction (6.1) followed by

$$RNH_2^+\dot{C}HR + O_2 + H_2O \rightarrow RNH_3^+ + RCHO + HO_2$$

In strongly alkaline solution, the reactions of OH involve both the C-H and N-H bonds as indicated in reactions (6.2), (6.3). With diethylamine at pH values above 10, reaction (6.3) accounts for the removal of approximately ten percent of the OH radicals.[18] With the unsymmetrical amines, OH attack may occur at either of the two $\alpha$-carbon positions. Sarcosine for example gives formaldehyde as the principal carbonyl product together with smaller amounts of glyoxylic acid.[16]

Oxygen-free solutions of the simpler secondary amines have not been studied in any detail. Information on the effects of various types of substitution on the possible reactions of $e^-_{aq}$ with $RNH_2^+CH_2R$ would be of interest.

(7.) Peptides

(7.1) $RCONHCHR_2 + OH \rightarrow RCONH\dot{C}R_2 + H_2O$

(7.2) $RCONHCHR_2 + H \rightarrow RCONH\dot{C}R_2 + H_2$

Peptides show the characteristic properties of secondary amines toward the indirect action of ionizing radiation in oxygenated solution.[16,27] Simple peptides such as acetylglycine, acetylalanine, etc. undergo radiation-induced cleavage of the N-C bond to yield amide and carbonyl products. The degradation involves an initial OH attack via reaction (7.1) and the subsequent step

$$RCONH\dot{C}R_2 + O_2 + H_2O \rightarrow RCONH_2 + R_2CO + HO_2$$

In certain cases there is evidence for the formation of the dehydropeptide in the parallel reaction [16,28]

$$RCONH\dot{C}R_2 + O_2 \rightarrow RCON{=}CR_2 + HO_2$$

The importance of reaction (7.1) in the over-all radiation chemistry of a particular peptide will of course, depend on the nature of the substituents R. In the case of acetylglycine G(acetamide) $\simeq$ 3.0, whereas for a number of aqueous protein solutions G(amide) $\simeq$ 1.0.[16]

In oxygen-free solutions of the simpler peptides the evidence is that H also yields $RCONH\dot{C}R_2$ as indicated in reaction (7.2). Removal of the $RCONH\dot{C}R_2$ species in solutions of acetylglycine has been shown[28] to occur largely through dimerization to give 1,2 diamine derivatives

$$2RCONH\dot{C}R_2 \rightarrow \begin{array}{c} RCONHCR_2 \\ | \\ RCONHCR_2 \end{array}$$

and to a lesser extent through disproportionation

$$2RCONH\dot{C}R_2 \rightarrow RCONH{=}CR_2 + RCONHCHR_2$$

and through

$$RCONH\dot{C}R_2 + H_2O_2 \rightarrow RCONHC(OH)R_2 + OH$$

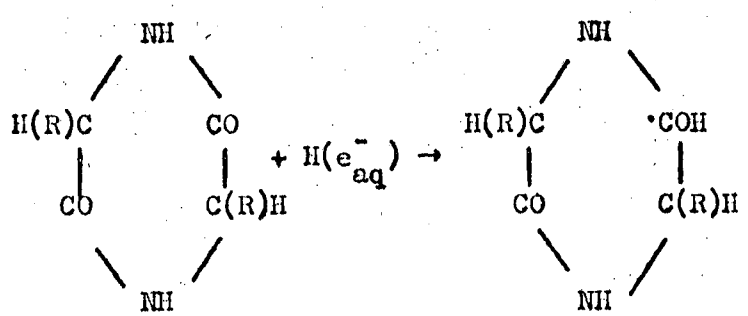
Products of the latter two reactions i.e. the dehydropeptide $RCON{=}CR_2$ and the dehydropeptide hydrate $RCONHC(OH)R_2$ yield ammonia and the corresponding carbonyl product on mild hydrolysis. Oxygen-free solutions of acetyl glycine (0.1M, pH 3) after γ-irradiation give on hydrolysis[28] G(ammonia) ≃ 0.90, G(glyoxylic acid) ≃ 0.50, G(formaldehyde) ≃ 0.20, G(diaminosuccinic acid) ≃ 1.7. The fact that aspartic acid and acetic acid are also detected in low yield with respective G values of ~0.13 and ~0.02 suggests that reductive cleavage to give the $\dot{C}H_2COOH$ radical occurs to a small extent in 0.1M acetylglycine solutions at pH 3. Studies of the effects of pH and solute concentration on the yields of these products have been undertaken to determine the contributions of reactions of the type

$$RCONHCHR_2 + H \rightarrow RCONH_2 + \dot{C}HR_2$$

$$RCONHCNR_2 + e^-_{aq} + H_2O \rightarrow RCONH_2 + \dot{C}HR_2 + OH^-$$

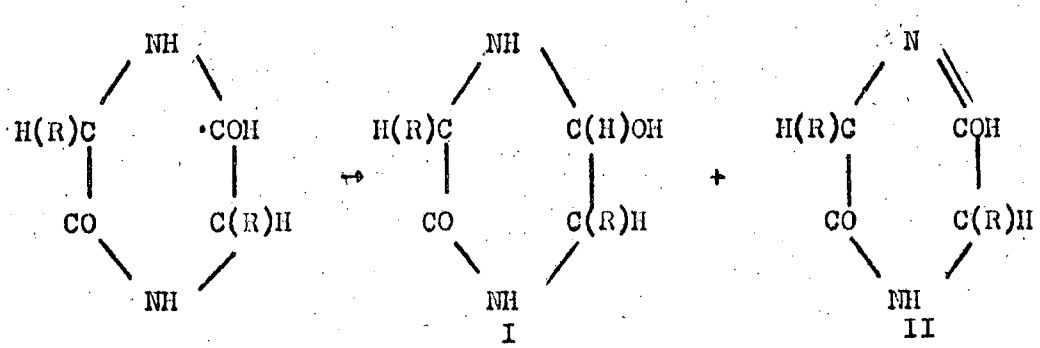
(8.) Cyclic Dipeptides

(8.1)

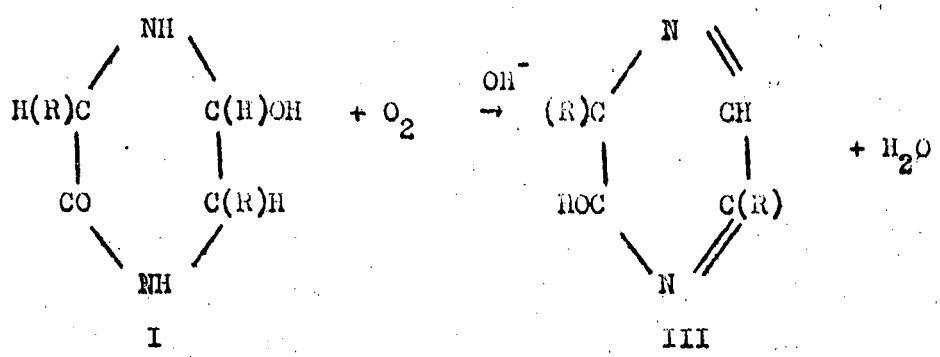


Evidence for a radiation-induced reduction of the peptide carbonyl group has been obtained in recent studies of the cyclic dipeptides in oxygen-free solution.[29] If an oxygen-free solution of alanine anhydride (0.1M, pH 7) is irradiated with γ-rays and then examined spectrophotometrically, it is found that the absorption spectrum shows negligible absorption above 2600°A as long as the solution is not exposed to the atmosphere. However, if oxygen is introduced, an absorption maximum in the longer wave-length region slowly appears. In neutral and basic solution the absorption maximum of the product from alanine anhydride is at 320 mμ; in acid solution there is a reversible shift to 340 mμ. Solutions of glycine anhydride exhibit similar effects except that the wave-length of maximum absorption is independent of pH.

These phenomena have been shown[29] to involve the reduction of the diketopiperazine to the corresponding 1,2-dihydropyrazine which is oxidized in air to give the pyrazine nucleus. The proposed mechanism includes the reduction step (8.1) followed by disproportion ation




where II is simply the enol form of the dipeptide. The subsequent reaction of I with oxygen is accompanied by isomerization i.e.




The yield of 2-hydroxy-3:6-dimethylpyrazine (III) in 0.1M alanine anhydride at ph 7 corresponds to G $\simeq$ 0.5 with $Co^{60}$ $\gamma$-rays. If the indicated mechanism for pyrazine formation is correct, the initial yield of reaction (8.1) must be in the order of G~ 1. Since the highest estimate for the primary H yield in neutral solution is ~0.45 it would appear that $e^-_{aq}$ rather than H is involved in the initial reduction reaction (8.1).

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FOOTNOTE AND REFERENCES

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1. E. Hayon and J. Weiss, Proceedings of the Second United Nations International Conference on the Peaceful Uses of Atomic Energy, 29. Chemical Effects of Radiation (United Nations, Geneva, Switzerland, 1958).

2. N. F. Barr and A. O. Allen, J. Phys. Chem., 63, 928 (1959).

3. G. Czapski and H. A. Schwarz, J. Phys. Chem., 66, 471 (1962).

4. T. Rigg, G. Scholes and J. Weiss, J. Chem. Soc., 3034 (1952).

5. V. L. Tal'roze and E. L. Frankevich, Proceedings of the First All-Union Conference on Radiation Chemistry. Moscow, 1957.

6. T. L. Cottrell, The Strengths of Chemical Bonds. (Butterworths Publications, Ltd., London, 1954).

7. D. Cleaver, E. Collinson and F. S. Dainton, Trans. Faraday Soc., 56, 1640 (1960).

8. J. Jortner, M. Ottolenghi, J. Rabani and G. Stein, J. Chem. Phys., 37, 2488 (1962).

9. H. A. Dewhurst and M. Burton, J. Am. Chem. Soc., 77, 5781 (1955).

10. A. O. Allen, Proceedings of the First United Nations International Conference on The Peaceful Uses of Atomic Energy 7, Nuclear Chemistry and the Effects of Radiation. (United Nations, Geneva Switzerland, 1955).

11. M. C. Sneed and R. C. Brasted, Comprehensive Inorganic Chemistry, V, (D. Van Nostrand Inc., New York).

12. M. Lefort and X. Tarrago, J. Inor. Nucl. Chem., 16, 169 (1961).

13. C. R. Maxwell, D. C. Peterson, and W. C. White, Radiation Research 2, 431 (1955).

FOOTNOTE AND REFERENCES (Continued)

14. G. Stein and J. Weiss, J. Chem. Soc., 3256 (1949).

15. W. M. Garrison and B. M. Weeks, J. Chem. Phys., 25, 685 (1956).

16. W. M. Garrison, M. E. Jayko and W. Bennett, Radiation Research 16, 483 (1962).

17. J. Rabani and G. Stein, Trans. Faraday Soc., 58, 2150 (1962).

18. G. G. Jayson, G. Scholes and J. Weiss, J. Chem. Soc., 2594 (1955).

19. C. R. Maxwell, D. C. Peterson, and N. E. Sharpless, Radiation Research 1, 530 (1954).

20. B. M. Weeks and W. M. Garrison, Radiation Research 9, 291 (1958).

21. N. E. Sharpless, A. E. Blair and C. R. Maxwell Radiation Research 417 ( ).

22. K. C. Kurien and M. Burton, Summary of Proceedings of the Fourth Informal Conference on the Radiation Chemistry of Water, (University of Notre Dame, March, 1961).

23. W. M. Garrison, Proceedings of the 6th International Congress on Nuclear Energy, (Rome, Italy, June 1959 p 67.).

24. E. J. Hart, J. Am. Chem. Soc., 76, 4198 (1954).

25. G. G. Johnson, G. Scholes and J. Weiss, J. Chem. Soc., 1358 (1957).

26. A. O. Allen, The Radiation Chemistry of Water and Aqueous Solutions (D. Van Nostrand Inc. New York, 1961).

27. M. E. Jayko and W. M. Garrison, J. Chem. Phys., 25, 1084 (1956).

28. W. M. Garrison and B. M. Weeks, Radiation Research, 17, 341 (1962).

29. M. Kland-English and W. M. Garrison, Nature 189, 302 (1961).

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