N-Chlorosuccinimide-Promoted Oxidative Decarboxylation of α-Amino Acids in Aqueous Alkaline Medium

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Kinetics of oxidation of twelve α-amino acids (AA) by *N*-chlorosuccinimide (NClS) in aqueous alkaline media have been studied and compared with those of *N*-bromosuccinimide (NBS) oxidation. Analysis of the results shows that the observed rate of oxidation is first-order in [oxidant] and zero-order in [substrate]. The rate of oxidation increases with increase in $[OH^-]_{free}$ in [NClS], the exception being the amino acids having β-alkyl substituent such as valine, leucine etc. Perusal of the results shows that NClS/NBS reacts with α-amino acid anion to produce α-amino acyl hypohalite which then decomposes in the rate-determining step to give the products. The intermediate α-amino acyl hypohalite is identified by UV-visible absorption spectra. Glycine behaves differently from other amino acids in both oxidants. The proposed mechanism is consistent with the observed kinetics.

Amino acid metabolism is one of the well-documented process. However the mechanism of the chemical oxidative decarboxylation of α-amino acids is not well-understood and still an area of experimentation. As a part of our broad program on the mechanistic aspects of the oxidation of amino acids, we have studied the kinetics of twelve α-amino acids viz., glycine, alanine, α-amino butyric acid, norvaline, valine, norleucine, leucine, isoleucine, serine, threonine, phenylalaine, and glutamine with *N*-chlorosuccinimide in aqueous alkaline medium. The oxidation of α-amino acids by *N*-bromosuccinimide is also studied and the results are compared with the oxidation by *N*-chlorosuccinimide.

Results and Discussion

Oxidation by *N*-Chlorosuccinimide: The kinetics of oxidation of α-amino acids by *N*-chlorosuccinimide (NClS) are investigated at pseudo-first-order conditions by keeping an excess of the [amino acid] (i.e. more than 10 times) over [oxidant] and also with $[OH^-]_{total} \geqslant$[amino acid]. Here [oxidant] or [NClS]/[NBS] means the concentration of active (positive) chlorine or bromine. Plots of log[oxidant] vs. time are linear even up to 70% conversion of [oxidant]. Values of k_{obs}, the pseudo-first-order rate constants, are unaffected by the increase in $[NClS]_0$ in all the amino acids. This clearly shows that the rate is first-order with respect to [oxidant]. Glycine alone behaves differently and $[NClS]_0$ shows an inverse relation on k_{obs}.

Values of k_{obs} are unaffected by the change in [amino acid] at constant $[OH^-]_{free}$ where $[OH^-]_{free} = [OH^-]_T - [\text{amino acid}] - [\text{succinimide}]$. This clearly shows that the rate is zero-order with respect to [amino acid].

The rate constant remains constant with change in $[OH^-]_T$ when $[OH^-]_T \leqslant [\text{amino acid}] + [\text{succinimide}]$ and then increases linearly in glycine, alanine, α-amino butyric acid, norvaline, norleucine, serine, threonine, and glutamine (Fig. 1). The effect of $[OH^-]$ on k_{obs} was studied in the absence of succinimide also. The plots of k_{obs} vs. $[OH^-]_{free}$ are straight lines.

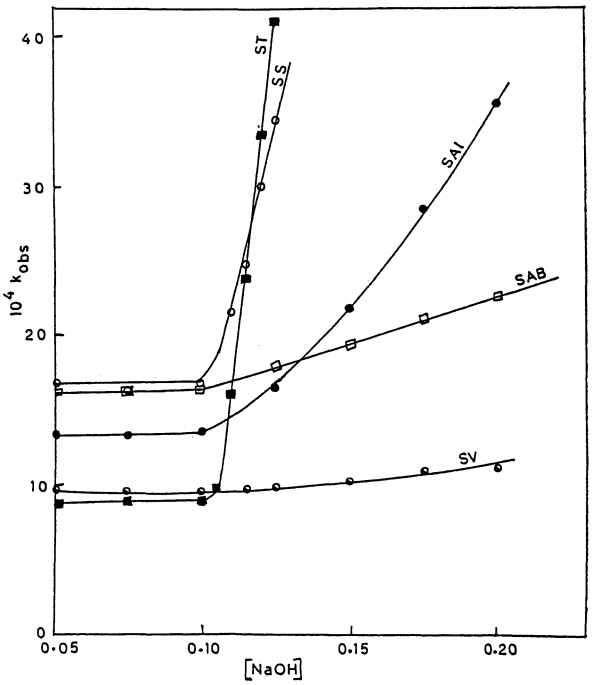


Fig. 1. Plot of k_{obs} vs. [NaOH] at 35 °C.
[Amino acid]=0.05 M; [Succinimide]=0.05 M, SAL-Alanine; $[NClS]=4.1\times10^{-3}$ M, SAB-Butryne; $[NClS]=3.6\times10^{-3}$ M, SV-Valine; $[NClS]=3.18\times10^{-3}$ M, SS-Serine; $[NClS]=3.85\times10^{-3}$ M, ST-Threonine; $[NClS]=3.34\times10^{-3}$ M.

The change in [succinimide] has no effect on k_{obs} in valine, leucine, isoleucine, and phenylalanine at constant $[OH^-]_T$. But in the other amino acids k_{obs} shows an inverse dependence on [succinimide] at constant $[OH^-]_T$. However at constant $[OH^-]_{free}$, k_{obs} is not at all affected by the change in [succinimide]. This clearly shows that the rate is zero-order with respect to [succinimide]. Formaldehyde, chloride ion, and ionic strength have no effect on k_{obs}.

Table 1. Rate Constants for the Oxidation of α-Amino Acids by NBS[a] at 35 °C

10^3 [NBS]	[Alanine]	$10^4 \times k_{obs}$
M	M	s^{-1}
1.98	0.05	54.9
3.88	0.05	54.9
3.88	0.075	57.0
3.88	0.05*	58.0

a) $[NaOH]_f$=0.00, [succinimide]=0.05 M; *[succinimide]=0.05, $[NaOH]_f$=0.0 M.

Oxidation by *N*-Bromosuccinimide (NBS). Under pseudo-first-order conditions plots of log[oxidant] vs. time are linear for two half lives indicating a first-order dependence on [oxidant]. Except glycine the rate is fast with half life approximately 3—4 minutes. However the results are reproducible within ±5% when the experiments are repeated with utmost care (Table 1). Moreover the extrapolations of log[oxidant] vs. time plots give the initial concentrations of the oxidant used with an error of ±5%. This indicates that there is no fast side reaction. The values of k_{obs} are unaffected by the increase in [oxidant].

The values of k_{obs} are unaffected with increase in [amino acid] at constant $[OH^-]_{free}$ indicating that the rate is zero-order with respect to [amino acid]. However in glycine alone, the rate of oxidation shows a linear dependence on [glycine] at constant $[OH^-]_f$ both in the presence and absence of succinimide. Hence plots k_{obs} vs. [glycine] at constant $[OH^-]_{free}$ both

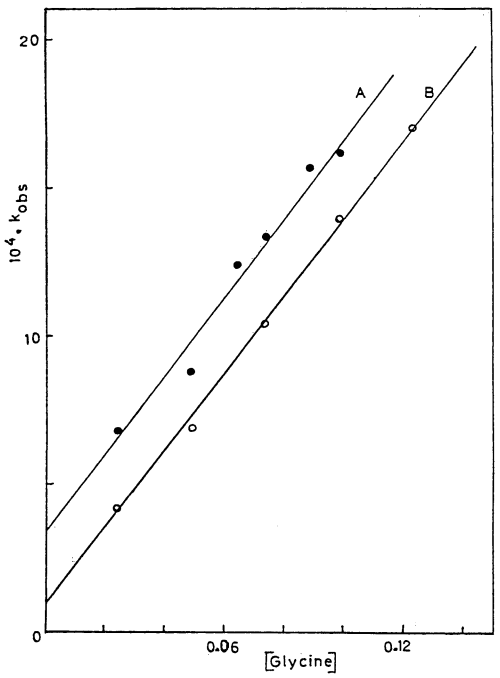


Fig. 2. Plot of k_{obs} vs. [Glycine] at 35 °C.
A. [Imide]=0.05 M; [NBS]=3.88×10⁻³ M; $[NaOH]_f$=0.0 M. B. [NBS]=3.88×10⁻³ M; $[NaOH]_f$=0.0 M; [Imide]=0.0 M.

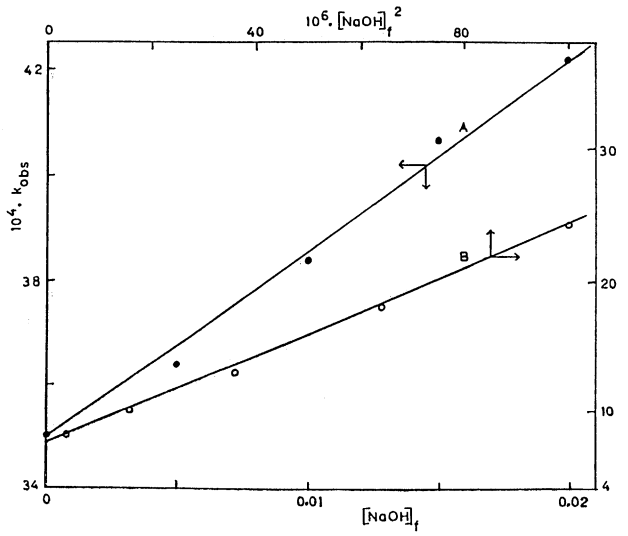


Fig. 3. A. Plot of k_{obs} vs. $[NaOH]_f$ at 35 °C. [Valine]=0.05 M; [NBS]=3.86×10⁻³ M; [Imide]=0.0 M. B. Plot of k_{obs} vs. $[NaOH]_f^2$ at 35 °C. [Glycine]=0.05 M; [NBS]=3.88×10⁻³ M; [Imide]=0.0 M

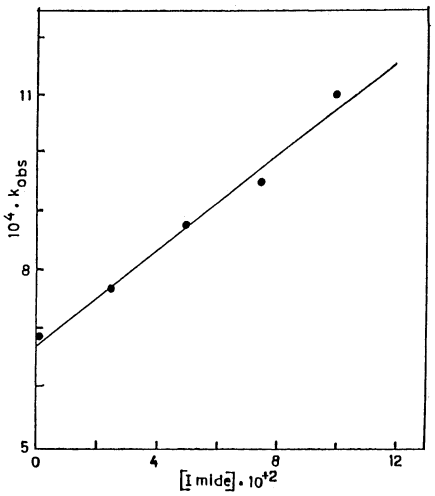


Fig. 4. Plot of k_{obs} vs. [Imide] at 35 °C. [Glycine]=0.05 M; $[NaOH]_f$=0.0 M; [NBS]=3.88×10⁻³ M.

in the presence and absence of succinimide give straight lines with identical slope (Fig. 2).

We could not study the effect of $[OH^-]_{free}$ on the amino acids with linear side chain since the rate is too fast to be measured. However the rate of oxidation of glycine and valine at various $[OH^-]_f$ can be followed. The plot of k_{obs} vs. $[OH^-]_f^2$ is a straight line with an intercept in glycine whereas k_{obs} vs. $[OH^-]_{free}$ is a straight line in valine (Fig. 3).

The plot of k_{obs} vs. [succinimide] at constant $[OH^-]_{free}$ is a straight line in glycine alone whereas in all other amino acids the rate is zero-order with respect to [succinimide] (Fig. 4).

Spectral Analysis: To a solution containing equimolar amount of amino acid and NaOH was added a solution of NClS/NBS and the UV-visible

Table 2. Absorption Maximum of the Intermediate

Amino acid	λ_{max} / nm	
	NClS	NBS
Glycine	283	306
Alanine	281	294
Valine	282	294
Serine	283	298

[Amino acid]=[NaOH]=0.1 M, [NClS]/[NBS]=0.015 M.

Table 3. Aldehyde Yield in the Oxidation of α-Amino Acids

Amino acid	Aldehyde yield/%	
	NClS	NBS
Glycine	0(4, 24)	0(4, 24)
Alanine	55(24, 48)	30(24), 13(48)
α-Amino butyric acid	60(24, 48)	35(24, 48)
Norvaline	65(24)	40(24)
Valine	52(24)	40(24)
Norleucine	75(24)	50(24)
Leucine	—	58(24)
Serine	100(24)	60(24)
Threonine	140(24)	70(24)
Glutamine	0(24)	—

Values given in parentheses are time in hrs after which the yield is measured. %yield is calculated based on [oxidant]. Very high yield in serine and threonine may be due to the formation of osazones by α-hydroxy aldehydes (Ref. E. Earl Royals, "Advanced Organic Chemistry," Prentice-Hall Inc., Englewood cliffs, N.J. (1958) p. 656). [AA]=[NaOH]=0.05 M ; [Oxidant]= ca. 0.03 M.

spectrum of the mixture was obtained. This mixture exhibits well defined absorption maximum (Table 2) and this absorption maximum is found to decrease with time. This clearly shows that the reaction involves the formation of an intermediate which then decays to products.

Product Analysis: The oxidation of amino acids by *N*-haloimides (NClS and NBS) in alkaline media results in the formation of the corresponding aldehyde, ammonia, and carbondioxide. The overall stoichiometry of the oxidation reaction may be represented as

$$1\ \text{NClS/NBS} + 1\ \text{Amino acid} \longrightarrow \text{Aldehyde} + CO_2 + NH_3 + Cl^-/Br^- + \text{succinimide.} \quad (1)$$

The analysis of the product (aldehyde) yields is given in Table 3. One of the interesting experimental observation is that aldehydes from serine and threonine are precipitated as 2,4-dinitrophenylhydrazone only after 3—4 h, whereas all other aldehydes are precipitated instantaneously. Analysis of the results given in Table 3 along with this observation clearly shows that either the self condensation of aldehyde molecules or condensation of aldehyde with imine may be responsible for the poor yield. Very high yield obtained in serine and threonine may be due to the formation of osazones.

Aliphatic amino acids exist as dipolar ionic form in water. The reaction involved in alkaline medium is

$$RCH(\overset{+}{N}H_3)COO^- + OH^- \longrightarrow RCH(NH_2)COO^- + H_2O \quad (2)$$

and with strong alkali such as NaOH the reaction is complete and quantitative.

Succinimide has no effect on k_{obs} in all the amino acids at constant $[OH^-]_{free}$, exception being glycine in NBS oxidation. The apparent effect of [succinimide] on k_{obs} at constant $[OH^-]_T$ (in NClS oxidation) is due to the change in $[OH^-]_{free}$ as shown by Eq. 3

$$\rangle N\text{-}H + OH^- \longrightarrow \rangle N^- + H_2O. \quad (3)$$

This reaction is possible because succinimide is a weak acid[1] with pK_a 9.6.

In alkaline medium *N*-halo compounds give hypohalite anion as,

$$\rangle N\text{-}X + 2OH^- \longrightarrow \rangle N^- + OX^- + H_2O. \quad (4)$$
(X=Br or Cl)

The reactive species of the oxidant could be either hypohalite anion or *N*-halosuccinimide itself. The formation of OCl^- from NClS and OBr^- from NBS may be ruled out on the ground that the intermediate should have a maximum at 292 nm[3] and 330 nm[3] respectively.

Let us consider the nature of the intermediate indicated by the absorption at ca. 280 nm in NClS-α-amino acid system so that we can have a clear insight into the mechanism. The work of Metcalf[4] has clearly established that *N*-chloro amine, one of the possible intermediates, shows an absorption spectrum with a λ_{max} at 253 nm. Anbar and Dostrovsky[3] have shown that the general appearance of the spectra of alkyl hypochlorites such as acetyl hypochlorite CH_3COOCl in CCl_4 show an absorption maximum at 265 nm. The absorption maximum of the intermediate from various amino acids (Table 2) occurs at the same wavelength irrespective of the structure of α-amino acids. Comparison of the spectrum of the intermediate with that of alkyl hypochlorites reported by Anbar and Dostrovsky[3] shows that the intermediate shown by the absorption at ca. 280 nm may be due to the formation of α-amino acyl hypochlorite $RCH(NH_2)COOCl$. Similarly the intermediates in NBS oxidation with λ_{max} ca. 295 nm is due to acyl hypobromites, since the general appearance of the spectra of hypobromites in CCl_4 shows a maximum at 280 nm with a weak shoulder at 340 nm.[3,4] This clearly shows that the intermediate formed on the oxidation of amino acids by *N*-halosuccinimide in alkaline medium is α-amino acyl hypohalite.

Amino acid concentration has no effect on the rate in all amino acids. This clearly shows that the decomposition of the intermediate, α-amino acyl

hypochlorite, which is formed rapidly and quantitatively, may be the rate-determining step. The α-hydrogen atom of the amino acid is acidic and it can be removed to give α-carbanion by the interaction with strong alkali.[5] The α-hydrogen atom can also be removed very easily as shown by the study of metal chelates of simple α-amino acids and peptides.[6] The catalytic effect of hydroxide ion on k_{obs} can be explained with the help of an interaction/complex formation between the hydroxide ion and α-hydrogen of the amino acid as shown in Scheme 1.

$$\rangle N\text{-}X + RCH(NH_2)COO^- \xrightarrow{fast} RCH(NH_2)COOX + \rangle N^- \quad (X=Cl \text{ or } Br) \tag{5}$$

$$RCH(NH_2)COOX + OH^- \underset{}{\overset{K_1}{\rightleftharpoons}} R\text{-}\underset{NH_2}{\overset{H\cdots{}^-OH}{C}}\text{-}COOX \tag{6}$$

$$RCH(NH_2)COOX \xrightarrow{k_1} \text{products} \tag{7}$$

$$R\text{-}\underset{NH_2}{\overset{H\cdots{}^-OH}{C}}\text{-}COOX \xrightarrow{k_2} \text{products} \tag{8}$$

Scheme 1.

According to the reaction Scheme 1, the rate equation can be written as,

$$k_{obs} = -\frac{d[RCH(NH_2)COOX]}{dt} \cdot \frac{1}{[RCH(NH_2)COOX]} = k_1 + K_1k_2[OH^-]_{free} \tag{9}$$

Equation 9 explains all the experimental observation. At $[OH^-]_{free}=0.0$ Eq. 9 reduces to $k_{obs}=k_1$. The values of k_1 and K_1k_2 are tabulated in Table 4. Perusal of the results (Table 4) shows that the value of k_1 calculated from the plot of k_{obs} vs. $[OH^-]_{free}$ agrees well with the one calculated at zero $[OH^-]_f$. The only exception is threonine wherin the plot k_{obs} vs. $[OH^-]_{free}$ gives a small negative intercept which can be considered as equal to zero. This may be due to the high reactivity of the hydroxide complex of threonine, as observed by the very high value of K_1k_2, the value of k_1 may be neglected at non zero $[OH^-]_{free}$.

Table 4. Rate Constants for the Oxidation of α-Amino Acids at 35 °C

Amino acid	NClS		NBS
	10^4k_1	$10^4K_1k_2$	10^4k_1
	s^{-1}	$M^{-1}s^{-1}$	s^{-1}
Glycine	2.2	—	0.6
Alanine	13.3(11.8)	190.0	55.0
α-Amino butyric acid	16.4(16.0)	72.0	65.0
Norvaline	14.7(14.6)	60.0	60.8
Norleucine	16.0(15.5)	44.0	64.9
Valine	9.6	ca. 0.0	35.0
Leucine	18.0	ca. 0.0	67.6
Isoleucine	11.6	ca. 0.0	45.7
Serine	15.6(8.4)	850	73.0
Threonine	8.4(0.0)	1450	34.5
Phenylalanine	16.3	ca. 0.0	Very fast
Glutamine	18.1	220	—

Values given in the parentheses are calculated using Eq. 9 i.e. plot of k_{obs} vs. $[NaOH]_f$.

Glycine behaves differently from other amino acids both in the oxidation by NClS and NBS. In NClS oxidation glycine differs from other amino acids in the respect that the rate of the oxidation at constant $[OH^-]_{free}$ decreases with increase in [oxidant]. This different behavior of glycine can be explained by the assumption that glycine hypochlorite dimerizes to give a nonreactive intermediate. Gopalakrishnan and Hogg[7] observed this type of dimerization in glycine alone in the oxidation of α-amino acids by *N*-bromosuccinimide in buffered media. For glycine alone, we can propose the following reaction scheme to explain the observed experimental facts.

$$CH_2(NH_2)COO^- + \rangle N\text{-}Cl \xrightarrow{fast} CH_2(NH_2)COOCl + \rangle N^- \tag{10}$$

$$CH_2(NH_2)COOCl + OH^- \underset{}{\overset{K_1}{\rightleftharpoons}} HO^-\cdots H\text{-}\underset{H}{C}\text{-}(NH_2)COOCl \tag{11}$$

$$2CH_2(NH_2)COOCl \underset{}{\overset{K_2}{\rightleftharpoons}} \text{Dimer} \tag{12}$$

$$CH_2(NH_2)COOCl \xrightarrow{k_1} \text{products} \tag{13}$$

$$HO^-\cdots H\text{-}\underset{H}{C}\text{-}(NH_2)COOCl \xrightarrow{k_2} \text{products} \tag{14}$$

Scheme 2.

This reaction Scheme 2 differs from the earlier scheme only in the reaction (12) namely the dimerization of glycine hypochlorite.

$$-\frac{d}{dt}[CH_2(NH_2)COOCl] = k_1[CH_2(NH_2)COOCl]_f + k_2[HO^-\cdots H\text{-}\underset{H}{C}\text{-}(NH_2)COOCl] \tag{15}$$

$$[CH_2(NH_2)COOCl]_T = [CH_2(NH_2)COOCl]_f + [\dot{C}H_2(NH_2)COOCl] + 2[\text{Dimerized product}] \tag{16}$$

We can write Eq. 16 as,

$$[CH_2(NH_2)COOCl]_T = [CH_2(NH_2)COOCl]_f + K_1[CH_2NH_2COOCl]_f[OH^-]_f + 2K_2[CH_2(NH_2)COOCl]_f^2 \tag{17}$$

Substituting the term $[CH_2NH_2COOCl]_f$ from Eq. 17 in 15 and rearranging, we get

$$k_{obs}=\frac{k_1+K_1k_2[OH^-]_f}{1+K_1[OH^-]_f+2K_2[CH_2NH_2COOCl]_f} \quad (18)$$

This equation can be approximated as

$$k_{obs}=\frac{k_1+K_1k_2[OH^-]_f}{1+K_1[OH^-]_f+2K_2[CH_2NH_2COOCl]_T} \quad (19)$$

Since $[CH_2(NH_2)COOCl]_T$ is formed quantitatively it can be replaced by $[NClS]_T$ as

$$k_{obs}=\frac{k_1+K_1k_2[OH^-]_f}{1+K_1[OH^-]_f+2K_2[NClS]_T} \quad (20)$$

Equation 20 explains the inverse dependence of k_{obs} on [oxidant]. At zero concentration of free hydroxide ion, Eq. 20 will be modified as

$$k_{obs}=\frac{k_1}{1+2K_2[NClS]_T} \quad (21)$$

Therefore a plot of k_{obs}^{-1} vs. $[NClS]_T$ should be a straight line with a slope of $2K_2/k_1$ and intercept of $1/k_1$. Such a plot is found to be a straight line (Fig. 5) with k_1 as 2.2×10^{-4} s^{-1} and K_2 as 101.3 M^{-1}. The effect of $[NClS]_T$ on k_{obs} is also studied at various $[OH^-]_f$, viz. 0.02, 0.03, 0.04, and 0.05 M. Similarly the effect of $[OH^-]_f$ on k_{obs} at constant $[NClS]_T$ is also studied. One of the interesting observation is that at $[OH^-]_f$=0.05 M, k_{obs} is independnt of $[NClS]_T$. Also the plot of k_{obs} vs. $[OH^-]_f$ at constant [NClS] gives a straight line with a small negative intercept which can be considered as equal to zero. These observation can be explained by the assumption that $k_1<K_1k_2[OH^-]_f$ and can be neglected. Therefore Eq. 21 becomes

$$k_{obs}=\frac{K_1k_2[OH^-]_f}{1+K_1[OH^-]_f+2K_2[NClS]_T} \quad (22)$$

According to Eq. 22, plot of k_{obs}^{-1} vs. $[NClS]_T$, at constant $[OH^-]_T$, should be a straight line (Fig. 5) with a slope of $2K_2/K_1k_2[OH^-]_f$ and an intercept of $1+K_1[OH^-]_f/K_1k_2[OH^-]_f$. The slope and intercept thus obtained at various $[OH^-]_f$ should be a function of $[OH^-]_f^{-1}$. The plot of (slope) vs. $[OH^-]_f^{-1}$ should be a straight line passing through the origin and the plot of (intercept) vs. $[OH]_f^{-1}$ should be a straight line with a positive intercept. Actually the plot of (intercept) vs. $[OH^-]_f^{-1}$ (Fig. 6) gives a straight line with positive intercept while the plot (slope) vs. $[OH^-]_f^{-1}$ is a straight line with negative intercept. This may probably due to the error involved in the slope obtained from the plots (Fig. 5) using Eq. 22. The values of k_2 and K_1 are calculated from the plot of (intercept) vs. $[OH^-]_f^{-1}$ as 45×10^{-4} s^{-1} and 58.0 M^{-1}. The value of K_2 is also calculated using the slope from the plots in Fig. 6 as 4×10^2 M^{-1} which is comparable in magnitude with the one calculated from the plot of k_{obs} vs. $[NClS]_T$ at zero $[OH^-]_f$. Equation 22 explains the observation that at 0.05 M of $[OH^-]_f$ k_{obs} is independent of [NClS]. This is because the denominator remains almost constant (changes from 4.26 to 5.34) for the variation of [NClS] from 1.8×10^{-3} M to 7.2×10^{-3} M. Similarly at constant [NClS], the change in the value of the denominator is not in proportion with that of $[OH^-]$ (for a five fold change of $[OH^-]_f$ i.e 0.01 to 0.05 M, the denominator changes from 2.31 to 4.62 at $[NClS]=3.6\times10^{-3}$) and fortuitously we get a straight line for k_{obs} vs. $[OH^-]_f$. In the above discussions we use the value of K_1 calculated at zero $[OH^-]_f$ since it is free from other equilibrium and

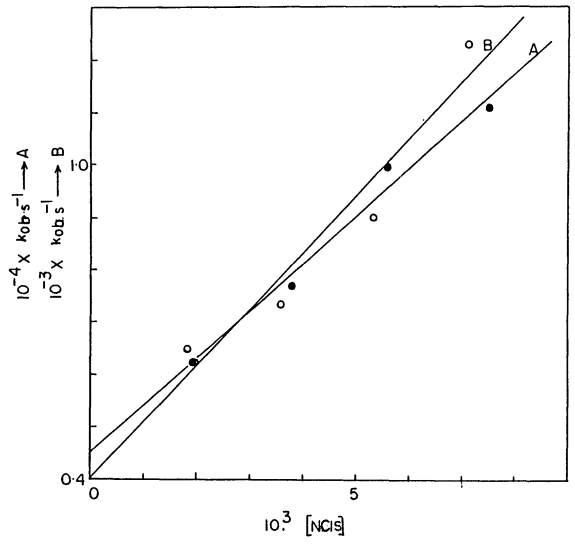


Fig. 5. Plot of k_{obs}^{-1} vs. [NClS] at 35 °C.
A. [Glycine]=0.05 M; [Succinimide]=0.05 M; [NaOH]$_f$=0.0 M. B. [Glycine]=0.05 M; [Succinimide]=0.05 M; [NaOH]$_f$=0.02 M.

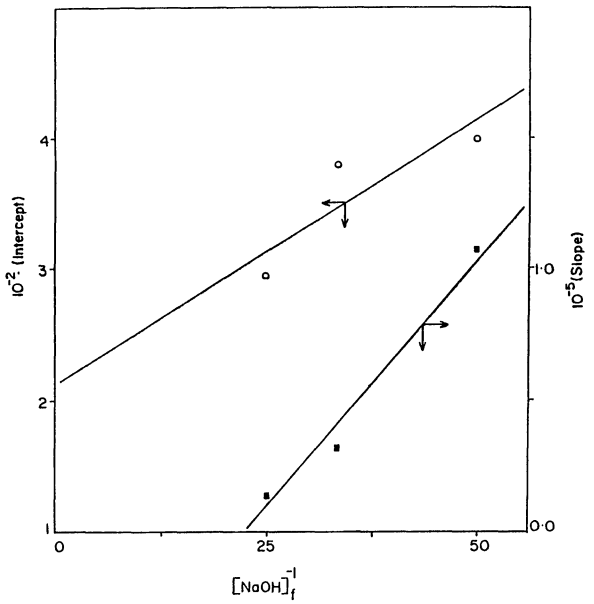
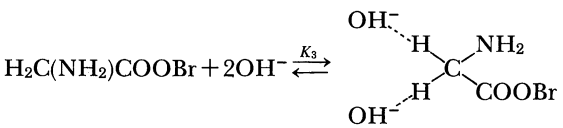


Fig. 6. Plot of intercept and slope (from Fig. 5) vs. $[OH^-]_f^{-1}$.

is considered more reliable.

The interesting observation in the NBS oxidation is that glycine shows a second order dependence on $[OH^-]_{free}$. Not only that the break down of glycyl hypobromite is also influenced by glycine anion (vide substrate effect) and succinimide anion. All other amino acids show zero-order dependence on each amino acid anion and succinimide anion. This clearly shows that the break down of glycyl hypobromite is base-catalysed. In order to examine the possibility of general base-catalysis, the kinetics of oxidation of glycine in the presence of sodium acetate was studied. Sodium acetate concentrations have no effect on k_{obs}. This is not surprising since glycine anion and succinimide anion are very strong bases than acetate anion (pK_a of glycine(classical), succinimide, and acetic acid are 9.78, 9.60, and 4.76 respectively). Thus the breakdown of glycyl hypobromite can be depicted, bearing the experimental observation as,



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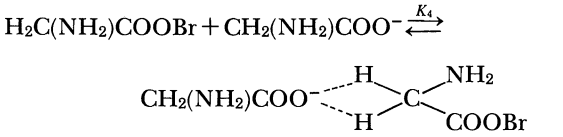

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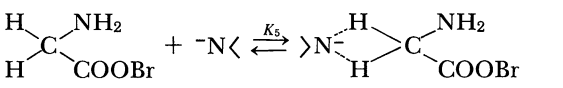

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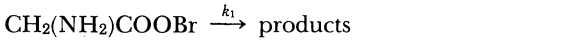

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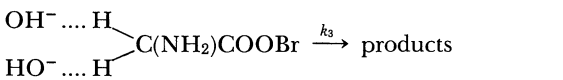
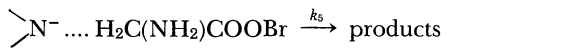


(27)

(28)

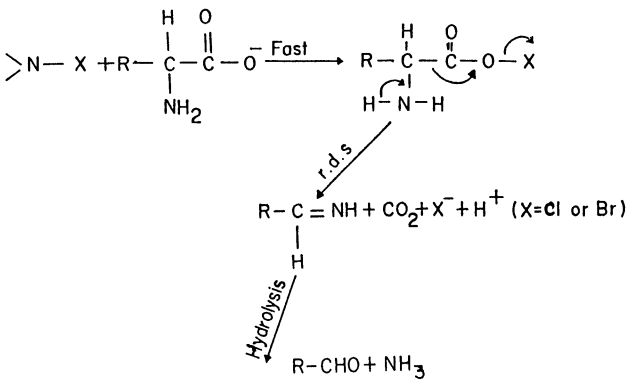


(29)

Scheme 3.

The reaction shown in Eq. 23 is actually not a trimolecular one but for simplicity the multiple step reaction is shown in one step. Similarly the interaction of the weak nucleophile with either of the hydrogens is simply depicted as in Eq. 24 and 25 and the structures in these equations do not mean the interaction of the nucleophile with the two hydrogen atoms simultaneously. We could not kinetically observe the dimerization of glycyl hypobromite eventhough this was observed at pH 4.00 by Gopalakrishnan and Hogg.[7] The rate equation for the above scheme can be written as

$$k_{obs} = k_1 + K_3 k_3 [OH^-]_f^2 + K_4 k_4 [\text{Glycine anion}] + K_5 k_5 [\text{succinimide anion}]. \quad (30)$$



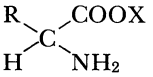
Scheme 4.

The equation explains the plots of k_{obs} vs. [glycine anion], k_{obs} vs. $[OH^-]_f^2$, and k_{obs} vs. [succinimide]. The values of K_4k_4 both in the presence and absence of succinimide anion are identical as $132\times10^{-4}\,M^{-1}\,s^{-1}$. The values of K_3k_3 and K_5k_5 are $16.1\,M^{-2}\,s^{-1}$ and $40.5\times10^{-4}\,M^{-1}\,s^{-1}$ respectively. The value of K_5k_5 obtained from the effect of glycine anion in the presence of succinimide at zero $[OH^-]_f$ is $50\times10^{-4}\,M^{-1}\,s^{-1}$. Similarly the values of k_1 from different plots are $8\times10^{-5}\,s^{-1}$, $2\times10^{-5}\,s^{-1}$, and $9\times10^{-5}\,s^{-1}$ (average $6\times10^{-5}\,s^{-1}$).

Based on the experimental observation we can propose a reaction scheme for the decomposition of the intermediate, α-amino acyl hypohalite (Scheme 4). Perusal of the results (values of k_1) shows that slight electron-releasing effect of alkyl group favours the reaction and this may be due to the stabilization of any partial positive charge that develops on the α-carbon in the transition state due to the possible nonconcertedness of bond making and bond-breaking i.e. the carbon-carbon bond cleavage to precede carbon-nitrogen bond formation to some extent.

Comparison of the results of the oxidation by *N*-chlorosuccinimide with that of *N*-bromosuccinimide is interesting. Glycine shows a first order dependence of $[OH^-]_f$ while valine is independent in NClS oxidation. In NBS oxidation glycine shows a second order dependence on $[OH^-]_f$ whereas valine shows a first order dependence. Not only that, the breakdown of glycyl hypobromite is catalysed by both the glycine anion and succinimide anion which is not observed in the breakdown of glycyl hypochlorite. This clearly shows that the formation of acyl hypobromite makes the α-hydrogens more reactive towards the nucleophile than in acyl hypochlorite. This is further confirmed by the correlation of k_1 with structure.

Perusal of the structure of the intermediate acyl hypohalites shows difference only in the alkyl substituent R at the α-carbon. Therefore



we can correlate k_1 with σ^*, the electron-donating power of the substituent and E_S, steric substituent

constant of the alkyl group using the Taft's equation[8]

$$\log k_1 = \log k_0 + \rho^* \sigma^* + \delta E_S \quad (31)$$

The results obtained can be expressed as

$$\log k_1 = -3.01 - 1.651\,\sigma^* + 0.16\,E_S \quad (32)$$
(Multiple correlation R=0.912)

$$\log k_1 = -2.61 - 3.99\,\sigma^* + 0.39\,E_S \quad (33)$$
(R=0.95)

for NClS and NBS oxidations respectively. In this analysis we include glycine, alanine, α-amino butyric acid, norvaline, valine, norleucine, leucine, and isoleucine only. This equation is valid only for a narrow range of σ^* i.e. from +0.49 to −0.19 only and too much importance can't be attached. Here we made the assumption that the polar and steric effects are additive and this assumption is proved successful in many free energy correlation.[8] But the assumption that the steric effects are additive does not seem to be justified and is not normally done.[8,9] However this correlation analysis substantiates the fact that in acyl hypobromite the inductive effects are more pronounced than in acyl hypochlorite and also the negative sign of the ρ^* term in Eqs. 32 and 33 indicates that the electron-releasing substituents accelarate the reaction.

The α-amino acids with electron-withdrawing substituents such as serine (R, CH_2OH σ^*=0.31), threonine (R, CH_3CHOH, σ^*=0.12), glutamine (R, CH_2-CH_2CONH_2, σ^*=0.19), and phenylalanine (R, C_6H_5-CH_2,σ^*= 0.27) show vertical deviation to Eqs. 32 and 33 and the mechanism may be different in these amino acids. The decomposition of phenylalanyl hypochlorite is not influenced by hydroxide ion (Table 4) and it is similar to valine, leucine etc. This suggest that phenyl group in the β-carbon inhibits the interaction of α-hydrogen with the hydroxide ion. The rate of decomposition of phenylalanyl hypobromite is too fast to be measured by the experimental methods employed here. These observation shows that the substituent $C_6H_5CH_2$ behaves like a strong electron donor but the positive σ^* value suggests that it should be an electron attractor. Similarly the substituents in serine and threonine i.e. CH_2OH- and CH_3CHOH- behave like electron donating groups similar to CH_3 etc. as evidenced by the large k_1 values whereas the hydroxide ion catalysed decomposition shows that these substituents behave similar to that of glycine (σ^*=+0.49). This abnormal reactivity observed in these amino acids namely phenylalanine, serine, and threonine may be due to alpha effect.[10]

Experimental

Amino acids were purchased from Loba-Indo Austranal Co (INDIA) and Sigma (USA). *N*-Chlorosuccinimide and *N*-bromosuccinimide both from Fluka (USA) were recrystallized from hot water and its purity was checked by iodometric determinations. Other chemicals used were of analytical grade. The reactions were carried out under pseudo-first-order conditions in a blackened reaction vessels. The amount of unreactive active chlorine/bromine was estimated by titrating against standard thiosulphate at suitable time intervals. The pseudo-first-order rate constants calculated from the plots of log [Vol. thio] vs. time were reproducible within ±5%.

Stoichiometry and Product Analysis: To a solution of amino acid with equimolar amount of sodium hydroxide NClS/NBS solution was added and diluted to a constant volume. The final concentrations of amino acid and NClS/NBS were always in the ratio of 0.05 M:0.03 M. After being kept for 24 hours aldehyde was estimated by 2,4-dinitrophenylhydrazone method.[11] The unreacted α-amino acid (in the form of anion) was estimated by titrating with standard HCl using Methyl Orange indicator. The accuracy of the titration was improved by the addition of 5 ml of formaldehyde (30% solution) to the titration mixture.[12]

Spectral Measurements: The concentration of amino acid (with equimolar NaOH) and NClS/NBS in the mixture was kept in the ratio of 0.1 M:0.015 M. The spectra were measured immediately on a Hitachi 557 spectrophotometer at room temperature using 1 cm cells.

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