The Mechanism of Oxidation of Hydroxamic Acids by Alkaline Ferricyanide

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The oxidation of hydroxamic acids, RCO·NH·OH (I) *via* their radical-anions $RCO-\dot{N}-\bar{O}$ (II) gives *NO*-diacylhydroxylamines RCO·NH·O·COR (III) in high yields. These compounds are active acylating agents. It is suggested that an unstable dimer of (II) gives (III) by intramolecular rearrangement.

N-Alkylhydroxamic acids oxidise similarly to give *N*-alkyl-*NO*-diacylhydroxylamines together with oximes or their oxidation products.

Both kinetic and e.s.r. studies have been used to elucidate the reaction mechanism.

THE discovery that many oxidants, *e.g.* periodic acid[1], bromine[2], iodine[3], mercuric oxide[3], t-butylhypochlorite[2], *N*-bromosuccinimide[1], and potassium ferricyanide[1] can convert hydroxamic acids RCO·NH·OH (I) into an active acylating agent has aroused particular interest since Boyland and Nery[3] have suggested that this type of acylation may be of biological significance in

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[1] B. Sklarz and A. F. Al-Sayyab, *J. Chem. Soc.*, **1964**, 1318.
[2] J. E. Rowe and A. D. Ward, *Austral J. Chem.*, **1968**, **21**, 2761.
[3] E. Boyland and R. Nery, *J. Chem. Soc.* (*C*), **1966**, 354.

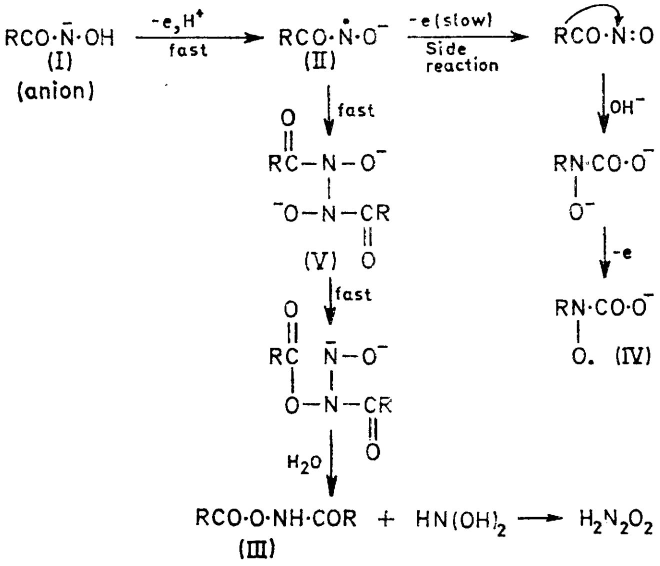
connection with the carcinogenicity of urethane[4] and possibly of many aromatic amines which can be oxidised *in vivo* to organic hydroxylamines.[5] In 1967 the e.s.r. study of Minor, Waters, and Ramsbottom[6] showed that the rapid oxidations of alkaline solutions of (I) by potassium ferricyanide gave transient free radicals, $RCO{\cdot}\dot{N}{\cdot}O^-$, (II) and on this basis Boyland and Nery suggested that the acylation might be a reaction involving $RCO{\cdot}$ free radicals.[3] E.s.r. spectra of secondary radicals, thought to be produced by further oxidation of (II), were also described.

We have now made a detailed study of the mechanism and products of this oxidation: our results, described below, lead us to the conclusion that the acylations themselves do not involve acyl radicals but are secondary reactions of *NO*-diacylhydroxylamines, $RCO{\cdot}NH{\cdot}O{\cdot}CO{\cdot}R$ (III), which can be isolated in yields of up to 95% by working up immediately the various reaction products. The dibenzyl compound (III; $R = PhCH_2$), like other *O*-acylhydroxylamines,[7] when kept in solution does acylate benzylamine in the prolonged reaction time that previous workers (notably Sklarz and Al-Sayyab,[1] who first discovered the acylation) had allowed before working up their products. Oxidation of (I; $R = PhCH_2$) in the presence of benzylamine did yield some *N*-benzylphenylacetamide, $PhCH_2{\cdot}NH{\cdot}CO{\cdot}CH_2Ph$. An oxidation, with excess of oxidant, in the presence of cyclohexylamine gave some *N*-cyclohexylphenylacetamide, but the acylating intermediate in this process would appear to be the unstable *NN*-di(phenylacetyl)hydroxylamine, $(PhCH_2CO)_2NOH$, which was isolated from a one-equivalent oxidation. This compound hydrolysed easily in water and readily rearranged thermally to (III; $R = PhCH_2$).

However, oxidation of (I; $R = PhCH_2$) in the presence of sodium azide or diethylamine did not yield any $PhCH_2{\cdot}CON_3$ or $PhCH_2{\cdot}CO{\cdot}NEt_2$, as would have been expected if free $(PhCH_2{\cdot}CO)^+$ cations had been produced during the reaction. Sklarz and Al-Sayyab[1] also found that no acylation of diethylamine occurred in their reaction, and we have shown that there was no appreciable reaction between diethylamine and (III; $R = PhCH_2$) when equivalent amounts of each were kept in solution overnight.

The stoicheiometry of the reaction has been determined by monitoring the disappearance of the ferricyanide anion spectroscopically at 415 nm. In 10^{-3}M-solutions at pH 9·5 or over, one equivalent of oxidant was consumed in less than 4 min., with up to 0·4 equivalents more being slowly reduced during the next hour. The addition of sodium ethylenediamine tetra-acetate (EDTA) to remove trace metals caused the consumption to be limited to the fast reaction, *i.e.* a one-equivalent oxidation.

From this and our product studies, we propose reaction Scheme 1, in which (III) is formed by an intramolecular rearrangement of (V), the dimer of the radical (II). The slow secondary oxidation, mentioned above, we assign to oxidation of the hyponitrite formed in the reaction.



SCHEME 1

Scheme 1 is in accord with the mechanism put forward by Cooley, Mosher, and Khan[8] for the oxidation of *O*-alkylhydroxamic acids from which the radical dimers can be isolated. However these dimers cannot undergo acyl-group transfer and decompose with loss of nitrogen.

By an e.s.r. study by the flow method, we found that in alkaline solution periodate, bromine, and iodine also yielded transient radicals (II) from (I; $R = PhCH_2$) and have confirmed, with a more sensitive instrument, that the ferricyanide oxidation of (I; $R = PhCH_2$) does give $PhCH_2{\cdot}N(O{\cdot}){\cdot}COO^-$ (IV) as a secondary radical. The signal of (IV) appears in ferricyanide oxidations of (I; $R = PhCH_2$) in aqueous sodium hydroxide immediately the flow is stopped, reaches a maximum in about 12 sec., and then decays completely over a further 2·5 min. whereas the signal of (II; $R = PhCH_2$) vanishes immediately the flow is stopped. Consequently the half-life of (II) must be of the order of 10^{-2} sec. and the much more stable radical (IV) could result from quite a minor side-reaction such as oxidation of (II) through $PhCH_2{\cdot}CO{\cdot}NO$ as tentatively suggested by Minor, Waters, and Ramsbottom.[6]

Radical (IV) was also produced by the one-electron oxidation of *N*-benzylhydroxylamine dissolved in aqueous sodium carbonate, in which the corresponding *N*-hydroxycarbamate is present:[9]

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[4] E. Boyland and R. Nery, *Biochem. J.*, 1965, **94**, 198.

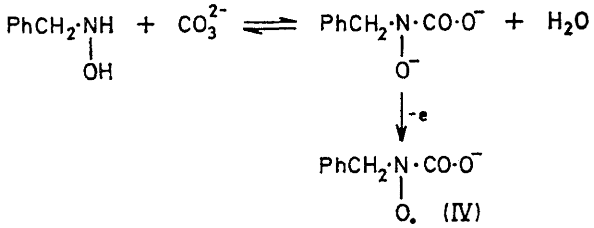
[5] J. W. Cramer, J. A. Miller, and E. C. Miller, *J. Biol. Chem.*, 1960, **235**, 885.

[6] D. F. Minor, W. A. Waters, and J. V. Ramsbottom, *J. Chem. Soc.* (*B*), 1967, 180.

[7] B. O. Handford, J. H. Jones, G. T. Young, and T. F. N. Johnson, *J. Chem. Soc.*, 1965, 6814.

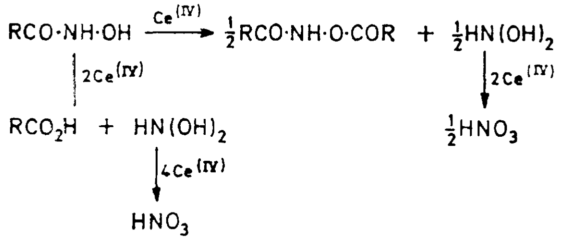
[8] J. H. Cooley, M. W. Mosher, and H. A. Khan, *J. Amer. Chem. Soc.*, 1968, **90**, 1867.

[9] See C. J. W. Gutch and W. A. Waters, *J. Chem. Soc.*, 1965, 751.



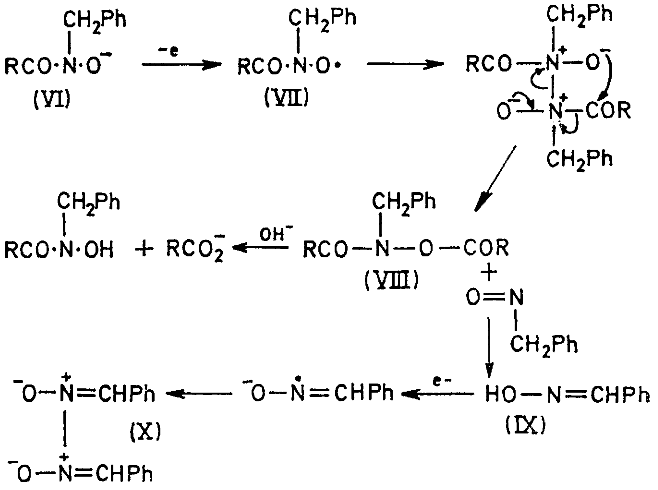
The oxidation of *N*-benzylhydroxylamine in aqueous sodium carbonate gave benzaldehyde azine *NN'*-dioxide in high yield (see Experimental section). This decomposes both thermally[10] and by hydrolysis to give benzaldehyde, and traces of the latter were always a minor product of oxidation of (I; R = $PhCH_2$). This is assumed to be formed *via* the side reaction shown in Scheme 1.

We have also examined the oxidation of (I; R = $PhCH_2$) in neutral ethanol with aqueous ceric ammonium nitrate. Compound (III) is again formed, but in lower yield (51%) and there is more hydrolysis to the carboxylic acid (13%) and its ethyl ester (28%). Experimentally, up to 4·7 equivalents of Ce^{IV} can be consumed per mole of (I): if the postulated intermediate $HN(OH)_2$ were oxidised to nitrate then, by calculation from the observed yields of products, 4·7 equivalents would be required. This is a rational suggestion because aqueous ceric ammonium nitrate rapidly oxidises nitrite to nitrate.



Scheme 2

Further support for the mechanism of Scheme 1 is provided by our study of the oxidation of *N*-benzylphenylacethydroxamic acid, $PhCH_2{\cdot}CO{\cdot}N(CH_2Ph){\cdot}OH$ (VI), by aqueous ferricyanide in alkali at pH 10—11. One equivalent of ferricyanide is consumed rapidly and then a further two more slowly. The transient radical $PhCH_2{\cdot}CO{\cdot}N(CH_2Ph){\cdot}O{\cdot}$ (VII) was formed initially and again the *NO*-diacylhydroxylamine $PhCH_2{\cdot}CO{\cdot}N{\cdot}(CH_2Ph){\cdot}O{\cdot}CO{\cdot}CH_2Ph$ (VIII) was a major product that could be isolated; it was shown to be an active acylating agent. Together with (VIII), α-benzaldehyde oxime (IX) is liberated at an early stage in the oxidation (by use of only 1 equiv. of ferricyanide) and this oxidises further to give eventually benzaldehyde azine *NN'*-dioxide (X). *N*-Benzylbenzohydroxamic acid oxidised in a similar way. Scheme 3 summarises the course of this oxidation; it would also explain satisfactorily the course of the oxidations of *N*-arylhydroxamic acids recently described by Forrester, Ogilvy, and Thomson.[11]



Scheme 3

EXPERIMENTAL

Oxidations of Hydroxamic Acids (I).—The acids (for preparation see ref. 6) (0·0025 mole) dissolved in M-sodium carbonate (50 ml.) were oxidised at room temperature with saturated aqueous potassium ferricyanide under the following conditions: (*a*) the oxidant (1·0 equiv.) was added dropwise during 10 min. to a rapidly stirred solution of the acid; (*b*) as in (*a*) but with 2·0 equiv. of oxidant; (*c*) the dissolved acid was added dropwise to 2·0 equiv. of oxidant; (*d*) as in (*a*) but after addition of 0·025 mole of disodium salt of EDTA; (*e*) as in (*b*) but after addition of 0·025 mole of sodium azide. As soon as a test (KI–starch) showed complete consumption of the ferricyanide the precipitated compound (III) was collected. Neutralisation (pH 7) and ether extraction gave a little more (III), following which acidification to pH 3 liberated the corresponding acid RCO_2H. A trace of benzaldehyde, detected by smell and characterised as its 2,4-dinitrophenylhydrazone, was produced in all oxidations of (I; R = $PhCH_2$) but no corresponding products could be isolated from the other acids.

		Isolated yields (%)		
(I; RCO·NH·OH)	Conditions	(III)	RCO_2H	Total RCO *
$PhCH_2$	(*a*)	85	20	105
	(*b*)	75	25	100
	(*c*)	68	32	100
	(*d*)	75		
	(*e*)	71	22	93
Ph	(*a*)	86	7	93
	(*c*)	66	34	100
$PhCH_2{\cdot}CH_2$	(*a*)	76		76
Ph_2CH	(*c*)	96		96

* Based on yields of (III) and RCO_2H.

An oxidation by procedure (*a*) of (I; R = $PhCH_2$) with the addition of 1·0 equiv. of cyclohexylamine gave 49% of the very unstable *NN*-diphenylacetylhydroxylamine which precipitated from the alkaline solution, together with only 15% of (III) and 30% of phenylacetic acid. When 2 equiv. of ferricyanide were used [procedure (*c*)] some 35% of

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[10] L. Horner, L. Hockenberger, and W. Kirmse, *Chem. Ber.*, 1961, **94**, 290.

[11] A. R. Forrester, M. M. Ogilvy, and R. H. Thomson, *J. Chem. Soc.* (*C*), 1970, 1081.

$PhCH_2\cdot CO\cdot NH\cdot C_6H_{11}$ was formed together with *NN*-diacylhydroxylamine, a little (III) (9%), and slightly more phenylacetic acid (34%).

Oxidation of (I; R = $PhCH_2$) [by procedure (*c*)] with the addition of 1·0 equiv. of benzylamine to the hydroxamate solution resulted in a 56% yield of *N*-benzylphenylacetamide, m.p. 119—120° (lit.,[1] 121°).

Products.—(i) NO-*Dibenzoylhydroxylamine* (III; R = Ph) had m.p. 168° (lit.,[3] 168°). The mass spectrum at 155° inlet showed major peaks at *m/e* = 122 and 119 corresponding to fragmentation to $PhCO_2H^+$ and $PhNCO^+$ (*cf*. Walling and Naglieri[12]). (ii) NO-*Di(phenylacetyl)hydroxylamine* (III; R = $PhCH_2$) had m.p. 166° (lit.,[13] 165°): τ (Me_2SO) 2·6—2·8 (10H two partly superimposed singlets), 6·15 (2H, s, $PhCH_2CO$), 6·4 (2H, s, $PhCH_2\cdot CO$); 1H variable at -ve τ (N*H*). The mass spectrum at 170° showed a weak molecular ion at *m/e* = 269 and strong fragment ions at *m/e* 136 and 133 ($PhCH_2CO_2H^+$ and $PhCH_2NCO^+$). (iii) NO-*Di*(β-*phenylpropionyl*)*hydroxylamine* (III; R = $PhCH_2\cdot CH_2$) had m.p. 72—73° (Found: C, 72·5; H, 6·1; N, 4·9. $C_{18}H_{19}NO_3$ requires C, 72·7; H, 6·4; N, 4·7%; *M*, 297): $\nu_{max.}$ 1672 and 1808 cm.$^{-1}$; τ ($F_3C\cdot CO_2H$) 2·75 (10H, s, C_6H_5), 6·6—7·4 (8H, complex, $PhCH_2\cdot CH_2\cdot CO$). The molecular ion at *m/e* 297 was weak, but there were major peaks at 150 and 147 ($PhCH_2\cdot CH_2CO_2H^+$ and $PhCH_2\cdot CH_2NCO^+$) and corresponding metastable peaks at 75·8 and 72·7. (iv) NO-*Bis(diphenylacetyl)hydroxylamine* (III; R = Ph_2CH) had m.p. 152—154°; $\nu_{max.}$ 1669 and 1763 cm.$^{-1}$; fragmentation to $Ph_2CHCO_2H^+$ and $Ph_2CH\cdot NCO^+$ (*m/e* 212 and 209) (Found: C, 79·6; H, 5·5; N, 3·3. $C_{28}H_{23}NO_3$ requires C, 79·8; H, 5·5; N, 3·3%). (v) NN-*Di(phenylacetyl)hydroxylamine* had m.p. 138—139° and was stable only in the solid form (Found: C, 71·0; N, 6·0; N, 5·2. $C_{16}H_{15}NO_3$ requires C, 71·3; H, 5·6; N, 5·2%). In benzene or chloroform its carbonyl absorption at 1720 cm.$^{-1}$ soon disappeared, being replaced by absorption due to (III; R = $PhCH_2$). In warm ethanol or water rapid hydrolysis to (I; R = $PhCH_2$) occurred.

Acylation of Benzylamine with NO-*Di(phenylacetyl)hydroxylamine* (*cf*. ref. 1).—Compound (III; R = $PhCH_2$) (1·35 g.) and benzylamine (0·54 g.) dissolved in dimethylformamide (50 ml.) and water (50 ml.) were stored overnight. The solution was then added to 2N-sodium carbonate (100 ml.) and extracted with ether to give *N*-benzylphenylacetamide (6·55 g., 50%), m.p. 118—119° (lit.,[1] 121°), identified by comparison of its i.r. spectrum with that of authentic material. The remaining alkaline solution contained both phenylacetic acid and (I; R = $PhCH_2$).

Oxidations of Phenylacethydroxamic Acid with Ceric Ammonium Nitrate.—(*a*) A solution of (I; R = $PhCH_2$) in ethanol was added dropwise to aqueous ceric ammonium nitrate. Reaction was complete after addition of 0·3 equivalents of (I) and the isolated products were (III) (35%), $PhCH_2CO_2Et$ (25%), and $PhCH_2\cdot CO_2H$ (30%).

(*b*) When the ceric salt was added to (I) in ethanol 4·7 equivalents of Ce^{IV} were consumed to give (III) (50%), $PhCH_2\cdot CO_2Et$ (28%), and $PhCH_2\cdot CO_2H$ (13%).

N-*Benzylphenylacethydroxamic Acid* (VI).—*N*-Benzylisobenzaldoxime[14] was found to give more tribenzylamine, m.p. 92°,[15] than monobenzylhydroxylamine when hydrolysed with hot concentrated hydrochloric acid,[14] but the latter was obtained in over 60% yield by shaking the oxime with equal volumes of 2M-hydrochloric acid and ether for 2 hr. at room temperature, and then separating the aqueous layer (Na_2CO_3), basifying it and then extracting it with ether; m.p. 57° (lit.,[13] 57°). Benzylhydroxylamine (0·05 mole) and triethylamine (0·05 mole) in dry chloroform (20 ml.) were treated dropwise at 0° with an equivalent of phenylacetyl chloride in chloroform (20 ml.) and the mixture was stored overnight. After removal of solvent the residue was extracted with boiling ether and the extract was crystallised from benzene, m.p. 98—99°, yield 70% (Found: C, 74·7; H, 6·0; N, 5·9%; M^+, *m/e* 241. $C_{15}H_{15}NO_2$ requires C, 74·7; H, 6·3; N, 5·8%; *M*, 241), $\nu_{max.}$ 1620 cm.$^{-1}$, τ (CF_3CO_2H) 2·7 (10H, s, 2 × C_6H_5), 4·95 (2H, s, $PhCH_2\cdot CO$), 5·85 (2H, s, $PhCH_2N$). This compound gave a colour with ferric chloride. Evidently the compound, m.p. 112—115°, described by Minor, Waters, and Ramsbottom[6] cannot have the structure then assigned to it.

N-Benzylbenzohydroxamic acid, prepared similarly, had m.p. 108° (lit.,[16] 106—107°) (Found: C, 74·0; H, 5·8; N, 6·2%; M^+, *m/e* 227. Calc. for $C_{14}H_{13}NO_2$: C, 74·1; H, 5·8; N, 6·2%; *M*, 227).

Oxidation of N-*Benzylphenylacethydroxamic Acid* (VI).—Spectrometric monitoring of the oxidation of (VI) with aqueous ferricyanide at pH 10—11 showed that 1 equiv. of oxidant was consumed in *ca*. 40 sec. and then, more slowly, a further 2 equiv. The e.s.r. spectrum of the transient benzyl phenylacetyl nitroxide radical, $PhCH_2\cdot CO\cdot N(CH_2Ph)O\cdot$, obtained by rapid flow (240 ml./min.) oxidation in 0·1M-sodium hydroxide showed a_N = 0·79; a_{NCH} 0·56 mT: *g*, 2·0061. Correspondingly the benzyl benzoyl nitroxide radical showed a_N 0·78; a_{NCH_2} 0·58 mT; *g*, 2·0060 and the methyl phenylacetylnitroxide radical a_N 0·76; a_{NCH} 0·87 mT; *g*, 2·0065. As would be expected the a_N hyperfine splitting constants are slightly higher than those reported[11,17] for acyl aryl nitroxides, but accord with those found for acyl methyl nitroxides.[6] Since the acid (VI) is sparingly soluble in aqueous sodium carbonate, product studies were made of oxidations in N-sodium hydroxide. After using 1 equiv. of ferricyanide in 2M-solutions and working up the reaction products immediately, 43% of (VI) was recovered unchanged and the reaction products were *N*-benzyl-*ON*-di(phenylacetyl)hydroxylamine (VIII) (15%), α-benzaldehyde oxime (IX) (35%) and phenylacetic acid, 30%, together with a little benzaldehyde. After using 2 equiv. of ferricyanide an 83% yield of (VIII) resulted together with 22% of phenylacetic acid and 12% of benzaldazine *NN'*-dioxide (X). Prolonged oxidation with 3 equiv. of ferricyanide gave phenylacetic acid and (X).

N-*Benzyl*-ON-*diphenylacetylhydroxylamine* (VIII) was only obtained as an oil. It had $\nu_{max.}$ 1670 and 1780 cm.$^{-1}$; τ ($CDCl_3$) 2·6—3·0 (complex, 3 × C_6H_5), 5·18 (s, $PhCH_2\cdot N$), 6·42, 6·46 (2 s, 2 × $PhCH_2\cdot CO$). Slow hydrolysis to (VI) occurred in water, as shown by the appearance of a ferric chloride colour.

Compound (X) which decomposes in light to benzaldehyde and nitrogen[10] was also the product of oxidation of both α- and β-benzaldehyde oximes with potassium ferricyanide

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[12] C. Walling and A. N. Naglieri, *J. Amer. Chem. Soc.*, 1960, **82**, 1820.

[13] I. de Paolini, *Gazzetta*, 1932, **62**, 1053.

[14] L. W. Jones and M. C. Sneed, *J. Amer. Chem. Soc.*, 1917, **39**, 674.

[15] Heilbron and Bunbury, 'Dictionary of Organic Compounds,' Eyre and Spottiswoode, London, 1965.

[16] O. Exner, *Coll. Czech. Chem. Comm.*, 1956, **21**, 1500.

[17] H. G. Aurich and F. Baer, *Tetrahedron Letters*, 1965, **43**, 3879.

in aqueous sodium carbonate at room temperature. It had m.p. 108—109° (lit.,[17] 107·5—108·5°), gave a molecular ion at *m/e* 240 and a strong nitrone absorption band at 1585 cm.$^{-1}$ but no carbonyl absorption.

Oxidation of Hydroxylamines RNHOH.—Oxidation of *N*-benzylhydroxylamine in M-sodium carbonate with 10^{-2}M-ferricyanide using a flow rate of 100 ml./min. gave a sharply defined spectrum of the secondary radical of (I; R = $PhCH_2$) showing a_N 1·11; $a_{C-H(2)}$ 0·72 mT; *g* 2·0059 corresponding to $PhCH_2 \cdot N(O\cdot) \cdot CO{-}O^-$; ceric sulphate oxidation in 2N-acid gave the spectrum of $PhCH_2 \cdot NH \cdot O\cdot$ described previously.[6] The corresponding spectrum from benzhydryl hydroxylamine showed $a_N = a_{NH}$ = 1·39; $a_{C-H(1)}$ 1·54 mT; *g* 2·0055, but the spectrum of this hydroxylamine in alkaline carbonate was evidently that of a mixture of radicals.

Oxidation of benzylhydroxylamine in M-sodium carbonate with 3 equiv. of ferricyanide gave 84% of benzaldehyde azine *NN'*- dioxide (X) together with a little *trans* α-azoxytoluene $PhCH_2 \cdot N{:}NO \cdot CH_2Ph$, detected by t.l.c. This was also isolated from the autoxidation of benzylhydroxylamine, and had m.p. 205—209°, *m/e* 206, lit.,[18] m.p. 209°.

Similar oxidation of benzhydrylhydroxylamine gave benzophenone oxime in 81% yield.

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[18] B. M. Lynch and K. H. Pausacker, *J. Chem. Soc.*, 1954, 3340.