933. *Electron Spin Resonance Spectra of the Carboxyhydroxymethyl Radical trapped after γ-Irradiation of Glycollic Acid.*

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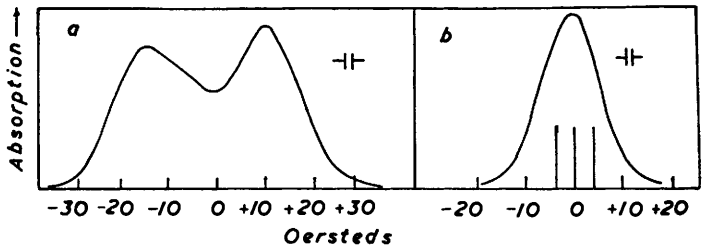
Electron resonance spectroscopy is used to identify the radical obtained on the irradiation of crystalline glycollic acid with gamma-rays as carboxyhydroxymethyl, HO·CH·CO_2H.

HIGH-ENERGY radiation can break many molecules into free-radical fragments whose chemical nature has mostly been inferred from the isolation of stable end-products. If, however, the radicals can be trapped in a solid they themselves can be investigated by electron resonance spectroscopy and identified. *X*-Irradiated glycollic acid has previously [1] been thus investigated, but the conclusions were only tentative and are not in agreement with the present extended experimental findings.

Spectra.—γ-Irradiated polycrystalline glycollic acid showed two lines, 25 oersteds apart, of line-width $\Delta H_{m.s.}$ ~9 oersteds at room temperature. The central minimum occurred at a field appropriate to $g \sim 2$. Though of nearly equal intensity, the high-field component was slightly narrower and with a greater peak intensity as shown in the Figure, *a*. Some

Electron resonance spectra at 9200 *Mc./sec.,* 3300 *oersteds, centred on* $g \sim 2$ *for* (*a*) *γ-irradiated glycollic acid and* (*b*) *γ-irradiated* [αα-$^2H_2$]*glycollic acid. The vertical lines show expected splitting by the deuterium nuclear moment.*



spectra showed signs of further structure with an additional, unresolved, weak feature just to the low-field side of the centre. This weak extra peak appeared to decay faster than the remainder of the spectrum and was absent from the other spectra described below; it is not considered significant. The signal was unchanged on opening the evacuated sample to air and was unchanged in intensity during three days but decayed to about 1/10th of the original intensity in six months. The spectrum was unchanged at 90° K but no irradiations at this temperature were undertaken. Calcium glycollate gave a very similar spectrum after irradiation though the lines were slightly closer (separation, 22 oersteds) and broader ($\Delta H_{m.s.}$ ~11 oersteds). [*carboxy*-$^2H_1$: *hydroxy*-$^2H_1$]Glycollic

[1] Gordy, Ard, and Shields, *Proc. Nat. Acad. Sci. U.S.A.*, 1955, **41**, 996.

acid, $DO{\cdot}CH_2{\cdot}CO_2D$, treated in the same way, gave the same spectrum as the protio-compound. In contrast, [$\alpha\alpha$-$^2H_2$]glycollic acid, $HO{\cdot}CD_2{\cdot}CO_2H$, gave a single-line spectrum (Figure, *b*) which was rather broad ($\Delta H_{m.s.}$ ~12 oersteds).

Discussion.—The two-peaked pattern of the Figure, *a*, shows [2] that there is one, and only one, hydrogen atom of spin $\frac{1}{2}$ which couples strongly to the free electron. The persistence of this pattern in the calcium salt shows that it is not the acidic hydrogen atom. The hydroxylic hydrogen atom is eliminated because the *OO'*-dideutero-compound retains the pattern. The conclusion is inescapable that the hydrogen atom which couples was initially one of those on the CH_2 group, and this is confirmed by the disappearance of the two peaks when these atoms were replaced by deuterium. The spectrum predicted for the radical containing the C–D group would be three lines of equal intensity separated by only 6 oersteds. This is less than the line-width of each, ~9 oersteds, and a numerical synthesis of a derivative curve of this nature shows that the overlapping of lines is so strong that only one peak would be apparent if the individual lines had a width of ~9 oersteds and either Gaussian or Lorentzian shape. All the spectra show slight asymmetry with a sharper feature on the high-field side and this is tentatively ascribed to an anisotropic *g*-factor which can distort the line shape in polycrystalline materials.

These considerations show that one, and only one, of the central hydrogen atoms is lost in forming the stable radical. In the absence of measurements with ^{17}O- and ^{13}C-containing materials it cannot be proved that the remainder of the skeleton is unchanged, but there is no reason to suspect that it is not intact. The evidence thus strongly favours the structure $HO{\cdot}\dot{C}H{\cdot}CO_2H$ for the trapped radical. Apart from the very small inflection mentioned above, there is no indication of a second radical. One possible step is $\gamma + HO{\cdot}CH_2{\cdot}CO_2H \longrightarrow HO{\cdot}\dot{C}H{\cdot}CO_2H + H$, though this may occur in two separate stages, loss of an electron followed by a proton. If this were followed by $H + HO{\cdot}CH_2{\cdot}CO_2H \longrightarrow HO{\cdot}\dot{C}H{\cdot}CO_2H + H_2$ or $H + H \longrightarrow H_2$, no different radicals would be formed.

Experimental

The electron resonance spectra were measured at 9200 Mc./sec., 3300 oersteds, with 490 c./sec. modulation and with display of the derivative of the absorption on a pen recorder by use of the spectrometer described elsewhere.[3]

The compounds (50—150 mg.) were irradiated by a 200 c ^{60}Co γ-radiation source for 48 hr. in a sealed tube under high vacuum. The dose-rate was *ca.* $1{\cdot}5 \times 10^{18}$ ev g.$^{-1}$ min.$^{-1}$ and the radical yield *ca.* 2×10^{18} radicals/g.

Calcium Glycollate.—Glycollic acid (2 g.) in water (20 ml.) was neutralised with excess of calcium carbonate. The suspension was boiled, filtered, and allowed to cool. The crystalline salt was filtered off and recrystallised twice from hot water.

[carboxy-$^2H_1$: hydroxy-$^2H_1$]*Glycollic Acid.*—Deuterium oxide (10 ml., 99·78% D_2O) was distilled *in vacuo* on to glycollic acid (1 g.). The solution was kept for 1 hr., and then the solvent was removed by freeze-drying. This process was repeated with two further portions (10 ml.) of deuterium oxide, and the product was rapidly transferred to the irradiation tube in a dry-handling box. An infrared spectrum showed the material to be at least 90% exchanged and we believe that the 10% of OH groups found were reintroduced in preparing the sample for infrared spectroscopy.

[$\alpha\alpha$-$^2H_1$]*Glycollic Acid.*—" AnalaR " oxalic acid (10 g.) was dehydrated at 100° over phosphoric oxide *in vacuo*. The anhydrous oxalic acid was converted into the dideutero-form by freeze-drying it thrice from solution in deuterium oxide (10 ml., 99·78% D_2O) in a vacuum system. " AnalaR " sulphuric acid (1 ml.) was similarly converted into deuterium sulphate.

The anhydrous [$^2H_2$]oxalic acid and the deuterium sulphate were dissolved in deuterium oxide (10 ml., 99·78% D_2O) and transferred to a small electrophoresis cell in a dry-handling box. The solution was electrolysed at 60° with lead electrodes (12 v, 1 a, cathode area 25 cm.^{2}) by Ershov and Pyatnitskaya's method.[4]

[2] Wertz, *Chem. Rev.*, 1955, **55**, 829.
[3] Abraham, Ovenall, and Whiffen, *Trans. Faraday Soc.*, 1958, **54**, 1128.
[4] Ershov and Pyatnitskaya, *J. Chem. Ind.* (*U.S.S.R.*), 1941, **18**, No. 12, 13; *Chem. Abs.*, 1944, **38**, 3912.

The electrolysis was continued until, on cooling to room temperature, no oxalic acid crystallised. Further precautions to exclude moisture were unnecessary after this stage. The solution was adjusted to pH 7·5 with calcium hydroxide, stirred for 30 min., and neutralised with excess of carbon dioxide. The solution was boiled, filtered, and passed down a column of Zeo-Karb 225 (H^+ form). The acid eluate was then neutralised with excess of calcium carbonate, boiled, filtered, and concentrated. On cooling, the solution deposited crystals of crude calcium [$\alpha\alpha$-$^2H_2$]glycollate. This salt was recrystallised twice from hot water, and the free acid obtained by passing a solution of the salt down a column of Zeo-Karb 225 (H^+ form). The eluate was freeze-dried and sublimed twice *in vacuo* at 35° to yield the dideutero-acid (0·068 g.), m. p. 76°. The mixed m. p. with authentic glycollic acid was 77° (authentic glycollic acid has m. p. 79°). Its infrared spectrum was consistent with this structure with C–D stretching frequencies of the CD_2 group at 2169 and 2108 cm.$^{-1}$ and showed glycollic acid to be completely absent. Interpolation between the infrared spectra of glycollic acid (very strong band at 1090 cm.$^{-1}$) and [$\alpha\alpha$-$^2H_2$]glycollic acid (corresponding band at 1135 cm.$^{-1}$) suggests that [α-$^2H_1$]glycollic acid, $CHD(OH)\cdot CO_2H$, would probably absorb very strongly near 1112 cm.$^{-1}$; no such band was found in the product and its spectrum indicated it to be over 98% of the desired isotopic species.

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