

the previous H_2O layer. The aqueous solution was washed with 20 ml of Et_2O , and the pH adjusted to 5 with 5% HCl . After saturating with NaCl , the aqueous phase was extracted twice with 50 ml of ethyl acetate. The combined ethyl acetate extracts were dried (Na_2SO_4) and evaporated to give an oil which solidified upon stirring with 40 ml of H_2O .

Recrystallization of the solid from aqueous ethanol gave 2.2 g (55%) of the product 2 as a white crystalline powder (m.p. 124–125 $^\circ\text{C}$). Elemental analysis, NMR and IR data confirmed that the product was the hydroxamic acid 2.

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- 2 M.D. Corbett and B.R. Chipko, *Biochem. J.* 165, 263 (1977).
- 3 M.D. Corbett, B.R. Chipko and J.H. Paul, *J. env. Path. Toxic.* in press.
- 4 J.H. Weisburger and E.K. Weisburger, *Pharmac. Rev.* 25, 1 (1973).
- 5 M.D. Corbett and D. Baden, unpublished results.
- 6 E.E. Smissman and M.D. Corbett, *J. org. Chem.* 37, 1847 (1972).
- 7 Y. Kasai, T. Tanimura and Z. Tamura, *Analyt. Chem.* 47, 34 (1975).

Degradation of 2,3,7,8-tetrachlorodibenzo-p-dioxin in organic solvents by gamma ray irradiation

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Summary. The effect of gamma ray irradiation on 2,3,7,8 tetrachlorodibenzo-p-dioxin (TCDD) dissolved in organic solvents is described; TCDD is degraded with the loss of 1 or more chlorine atoms.

2,3,7,8-Tetrachlorodibenzo-p-dioxin (TCDD) is recognized as one of the most potent low mol. wt toxins known. TCDD has a number of toxic effects¹⁻⁶, such as teratogenicity, mutagenicity, immunosuppression and carcinogenicity; furthermore, it is a chemically and metabolically inert compound with a very long half-life both in the environment⁷⁻¹⁰ and in living systems¹¹⁻¹⁹. Therefore the possibility of TCDD accumulation in the food chain represents a serious health hazard.

The most serious case of environmental contamination by TCDD occurred about 2 years ago near the Italian town of Seveso, when a large, densely populated area was contaminated as a consequence of an explosion in a reactor producing 2,4,5-trichlorophenol²⁰. Since the population is still living in the contaminated areas, the problem of decontamination must be solved urgently, in order to reduce the possibility of adverse biological effects due to chronic exposure.

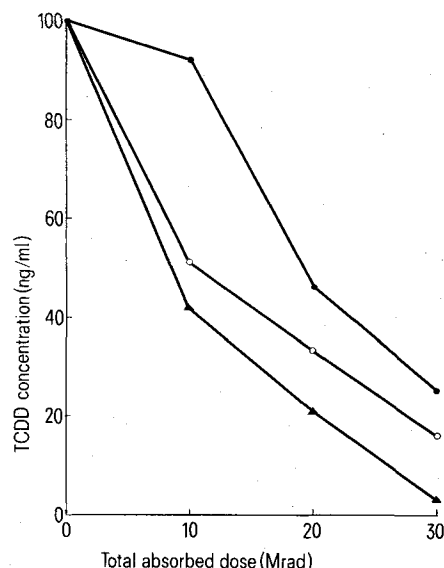
So far the only known methods of degrading the TCDD molecule are photoreduction by UV rays in organic solvents^{21,22} and incineration at a temperature above 750 $^\circ\text{C}$ ²³. These methods are now under investigation with a view to their practical application to the situation in Seveso.

As an attempt to contribute to solving this problem, we report the outcome of preliminary experiments on the effects of gamma rays on TCDD dissolved in organic solvents.

All irradiations were accomplished with an experimental irradiator (BC-27 model) containing a 10,000-Ci ^{60}Co source. The dose rate was 10^6 rad/h. The changes caused by gamma ray irradiation were studied varying the total dose of radiation absorbed by solutions of TCDD in dioxane, acetone or ethanol at a concentration of 100 ng/ml. Ali-

quots of 0.5 ml were put into glass screw-capped vials which were then submitted to irradiation at room temperature.

The irradiated solutions were directly analyzed for TCDD content and compared with control samples prepared and



Effect of gamma ray irradiation on TCDD concentration. TCDD was dissolved in ethanol (▲), acetone (○) and dioxane (●) at a concentration of 100 ng/ml. Solutions were irradiated at a dose rate of 10^6 rad/h and their TCDD concentration was measured after 10, 20 and 30 h.

handled in the same manner. The samples were also analyzed for the presence of trichlorodibenzo-p-dioxins and dichlorodibenzo-p-dioxins possibly formed as a consequence of the irradiation. Analyses were carried out by mass fragmentography using an LKB 2091-B gas chromatograph-mass spectrometer operating in the EI mode, equipped with an LKB 2130 computer system for data acquisition and calculation.

The results summarized in the figure indicate that TCDD disappearance is directly related to the total dose of radiation absorbed. Although these data are merely preliminary, it is also clear that the type of solvent used is important for the efficiency of the degradation process.

Since it is known that substances chemically related to TCDD retain a high degree of toxicity, an analysis was carried out in order to identify compounds other than TCDD, present in irradiated solutions. In all these samples, it was possible to detect trichloro- and dichloro-dibenzo-p-dioxins, suggesting that the degradation process occurs, at least in part, with the loss of chlorine atoms from the TCDD molecule. Further studies are in progress to verify the possible application of this method to degradation of TCDD in contaminated soil samples.

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- 1 R.D. Kimbrough, Crit. Rev. Toxic. 2, 445 (1974).
- 2 J.A. Moore, Ecol. Bull. (Stockholm), in press.
- 3 M.W. Harris, J.A. Moore, J.G. Vos and B.N. Gupta, Envir. Hlth Perspect. 5, 101 (1973).
- 4 B.A. Schwetz, J.M. Norris, G.L. Sparschu, V.K. Rowe, P.J. Gehring, J.L. Emerson and C.G. Gerbig, Envir. Hlth Perspect. 5, 87 (1973).
- 5 D. Neubert, P. Zens, A. Rothenwallner and H.-J. Merker, Envir. Hlth Perspect. 5, 67 (1973).
- 6 World Health Organization, IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Man, vol. 15, p. 41. IARC, Lyon 1977.
- 7 C.S. Helling, A.R. Isensee, E.A. Woolson, P.D.J. Ensor, G.E. Jones, J.R. Plimmer and P.C. Kearney, J. envir. Qual. 2, 171 (1973).
- 8 P.C. Kearney, E.A. Woolson and C.P. Ellington, Jr, Envir. Sci. Technol. 6, 1017 (1972).
- 9 A.L. Young, C.E. Thaiken, E.L. Arnold, J.M. Cupello and L.G. Cockerham, in: Fate of 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) in the Environment: Summary and Decontamination Recommendations, United States Air Force Academy, Colorado, 1976.
- 10 B. Commoner and R.E. Scott, in: Accidental Contamination of Soil with Dioxin in Missouri: Effects of Countermeasures, Center for the Biology of Natural Systems, Washington University, Saint Louis, Missouri, 1976.
- 11 F. Matsumura and H.J. Benezet, Envir. Hlth Perspect. 5, 253 (1973).
- 12 A.R. Isensee and G.E. Jones, J. agric. Fd. Chem. 19, 1210 (1971).
- 13 A.R. Isensee, Ecol. Bull. (Stockholm), in press.
- 14 A.R. Isensee and G.E. Jones, Envir. Sci. Technol. 9, 668 (1975).
- 15 J.P. Van Miller, R.J. Marlar and J.R. Allen, Fd. Cosmet. Toxic. 14, 31 (1976).
- 16 W.N. Piper, J.Q. Rose and P.J. Gehring, Adv. Chem. Ser. 120, 85 (1973).
- 17 J.Q. Rose, J.C. Ramsey, T.H. Wentzler, R.A. Hummel and P.J. Gehring, Fd. Cosmet. Toxic. 36, 209 (1976).
- 18 G.F. Fries and G.S. Marrow, J. agric. Fd. Chem. 23, 265 (1975).
- 19 J.H. Vinopal and J.E. Casida, Archs envir. Contam. Toxic. 1, 122 (1973).
- 20 S. Garattini, Biomedicine 26, 28 (1977).
- 21 J.R. Plimmer, U.I. Klingebiel, D.G. Crosby and A.S. Wong, Adv. Chem. Ser. 120, 44 (1973).
- 22 D.G. Crosby, A.S. Wong, J.R. Plimmer and E.A. Woolson, Science 173, 748 (1971).
- 23 R.H. Stehl, R.R. Papenfuss, R.A. Bredeweg, and R.W. Roberts, Adv. Chem. Ser. 120, 119 (1973).

(±)9,10-dihydroxy- $\Delta^{6a(10a)}$ -tetrahydrocannabinol and (±)8,9-dihydroxy- $\Delta^{6a(10a)}$ -tetrahydrocannabinol: 2 new cannabinoids from *Cannabis sativa* L.¹

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Summary. The structures of 2 new polyhydroxylated cannabinoids, (±)9,10-dihydroxy- $\Delta^{6a(10a)}$ -tetrahydrocannabinol and (±)8,9-dihydroxy- $\Delta^{6a(10a)}$ -tetrahydrocannabinol, obtained from a hexane extract of an Indian *Cannabis* variant were determined by spectral means and correlation with cannabinol.

To date more than 50 cannabinoids, among which the psychotomimetic Δ^9 -tetrahydrocannabinol have been isolated and/or detected in *Cannabis*³. In a previous communication from our laboratory⁴ we have reported the isolation and structure elucidation of 2 new cannabinoids, namely (+)-cannabitriol and (-)-10-ethoxy-9-hydroxy- $\Delta^{6a(10a)}$ -tetrahydrocannabinol from a *Cannabis* extract. The presence of these hydroxylated cannabinoids and reports on polyhydroxylated cannabinoids as minor constituents of *Cannabis*⁵⁻⁸ have stimulated our interest in the isolation and characterization of minor components of *Cannabis*.

Material and methods. A polar fraction obtained by silica gel column chromatography of the hexane extract of an Indian variant of *Cannabis sativa* L. showed the presence of 2 phenols; **1a** and **2a**, with R_f 0.58 and 0.51 (EtOAc-CHCl₃; 1:1). These compounds were only observed in the GC as their TMS derivatives, **1b** and **2b** (RRT 0.28 and 0.35, respectively)⁹. Separation of the 2 compounds was

achieved by repeated chromatography. For details on instrumentation as well as preparation of different derivatives see Elsohly et al.⁴.

Results and discussion. Compound **1a**, R_f 0.58, C₂₁H₃₀O₄ (HRP-MS), was obtained as a yellow optically inactive oil. The IR-spectrum showed bands at ν_{\max} (CHCl₃) 3500–3150 (br.) (OH) and 1620 cm⁻¹ (C=C, Ar) and no carbonyl absorption. The UV-spectrum was characteristic for a styrene chromophore¹⁰, λ_{\max} MeOH 302 (sh, log ϵ 3.76) nm, 276 (3.88) and 228 (2.92).

The ¹H-NMR-spectrum was indicative for the olivetol moiety of a cannabinoid. The 2 aromatic protons were found at δ 6.40 (s, 1 H) and δ 6.44 (s, 1 H) and the terminal CH₃ group of the pentyl side chain at δ 0.97 (s, 3 H) and the benzylic protons at δ 2.28 (broad, m). In addition resonances were found at δ 1.52 (s, 3 H), 1.47 (s, 3 H) and 1.28 (s, 3 H) assigned to methyl groups on