

In selecting the molecular formula for leptotene Takeda and Ohta² emphasized the carbon content. Their H-value (9.12 %) was very close to that required for C₄₀H₄₈ (9.16 %), although the C-value was about 1 % too low. For aryl carotenoids the hydrogen uptake may vary considerably with the hydrogenation conditions.

The name leptotene has priority over isorenieratene. However, the nomenclature of aromatic carotenoids will be discussed elsewhere.

Acknowledgements. This investigation was carried out at the initiative of Professor N.A. Sørensen and was made possible through the courtesy of Professor R. Kuhn, Max-Planck-Institut für Medizinische Forschung, Heidelberg, who kindly supplied the specimen of leptotene, and Professor B. C. L. Weedon, Queen Mary College, London, who provided a sample of synthetic isorenieratene.

A grant from *Norges Tekniske Høgskole* is gratefully acknowledged.

1. Grundmann, C. and Takeda, Y. *Naturwiss.* **25** (1937) 27.
2. Takeda, Y. and Ohta, T. *Z. Physiol. Chem.* **265** (1940) 233.
3. Takeda, Y. and Ohta, T. *Ibid.* **258** (1939) 6.
4. Takeda, Y. and Ohta, T. *J. Biochem. (Tokyo)* **36** (1944) 535.
5. Takeda, Y. and Ohta, T. *J. Pharm. Soc. Japan* **64** (1944) 67.
6. Goodwin, T. W. *The Comparative Biochemistry of the Carotenoids*, Chapman & Hall, London 1952, p. 120.
7. Ohta, T. *J. Pharm. Soc. Japan*, **71** (1951) 462.
8. Turian, G. *Helv. Chim. Acta* **36** (1953) 937.
9. Goodwin, T. W. and Jamikorn, M. *Biochem J.* **62** (1956) 269, 275.
10. Guadiano, A. *Rend. Ist. Super. Sanita* **22** (1959) 769; *Chem. Abstr.* **54** (1960) 13253c.
11. Tsumaki, T. and Yamaguchi, M. *J. Chem. Soc. Japan, Pure Chem. Sect.* **75** (1954) 297.
12. Yamaguchi, M. *Bull. Chem. Soc. Japan* **30** (1957) 111.
13. Yamaguchi, M. *Ibid.* **30** (1957) 979.
14. Yamaguchi, M. *Ibid.* **31** (1958) 51.
15. Yamaguchi, M. *Ibid.* **31** (1958) 739.
16. Yamaguchi, M. *Ibid.* **32** (1959) 1171.
17. Yamaguchi, M. *Ibid.* **33** (1960) 1560.
18. Liaaen Jensen, S. and Weedon, B. C. L. *Naturwiss.* (1964). *In press.*
19. Jensen, A. *Acta Chem. Scand.* **14** (1960) 2051.

20. Cooper, R. D. G., Davis, J. B. and Weedon, B. C. L. *J. Chem. Soc.* **1963** 5637.
21. Zechmeister, L. and Polgár, A. *J. Am. Chem. Soc.* **65** (1943) 1522.
22. Liaaen Jensen, S. *Kgl. Norske Videnskabs Selskabs Skrifter* **1962** No. 8.
23. Jensen, A. and Liaaen Jensen, S. *Acta Chem. Scand.* **13** (1959) 1863.
24. Jensen, A., Aasmundrud, O. and Eimhjellen, K. E. *Biochim. Biophys. Acta*. *In press.*

Received July 15, 1964.

Circular Dichroism and Chelation: Complexes of N,N'-Bis (2-butan-1-ol)-ethylenediamine

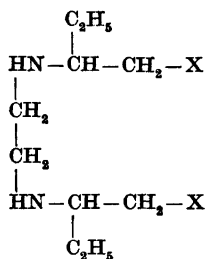
C. J. HAWKINS

Department I, Inorganic Chemistry, The H. C. Ørsted Institute, University of Copenhagen, Copenhagen, Denmark

Larsen and Olsen¹ have recently reported that optically active monodentate ligands co-ordinating through nitrogen (*e.g.* *d*-2-aminobutane) or oxygen (*e.g.* *d*-α-methylbutyrate) do not impose an observable optical activity onto the *d*-*d* transitions of metal ions. An optically active chelate, however, causes its complexes (*e.g.* mono (*l*-propylenediamine)copper(II)) to exhibit circular dichroism. A vicinal effect of the asymmetric carbon in the monodentate was not sufficient to cause a measurable activity but the puckering of the chelate ring lowered the effective symmetry and the Cotton effect was observed. Therefore, circular dichroism should prove to be a useful tool for determining whether an optically active ligand behaves as a chelate.

In this paper, the above observations will be applied to some metal complexes formed in solution by N,N'-bis(2-butan-1-ol)-ethylenediamine ('ethambutol'; structure below with X = OH). These complexes are important because they are thought to be responsible for the compound's considerable antituberculous activity.² From a comparison of the stability of these com-

plexes with the stability of complexes formed by a number of closely related compounds (*e.g.* *N,N'*-di-*sec*-butylethylenediamine), Hawkins and Perrin³ have concluded that ethambutol, both in its neutral and ionised forms, acts as a tetradentate ligand with copper(II) and nickel(II). The circular dichroism measurements presented in this paper have afforded further evidence to show that the hydroxyl groups must take part in the co-ordination.



The absorption spectra and circular dichroism are given in Fig. 1 for the 1:1 *d*-ethambutol-copper(II) complex in its 'neutral', 'half-ionised' and 'fully ionised' forms and in Fig. 2 for the corresponding nickel complexes.

The observed dichroism must arise from either the incorporation of the asymmetric centre into a chelate ring or, perhaps, a

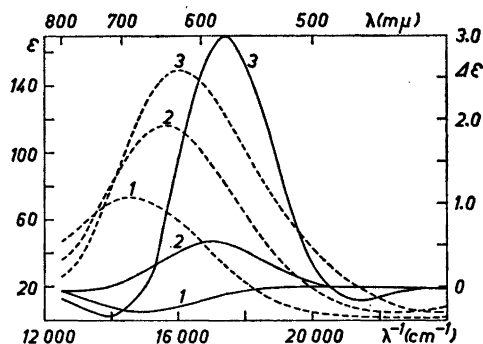


Fig. 1. Absorption (---) and circular dichroism (—) spectra for solutions of copper(II) nitrate (5×10^{-3} M) and *d*-ethambutol dihydrochloride (5×10^{-3} M) at 20° and $I = 0.1$ (NaNO_3). Concentrations of NaOH are: (1), 1.0×10^{-2} M; (2), 1.5×10^{-2} M; (3), 2.0×10^{-2} M. Species present are: (1), $\text{CuH}_2\text{L}^{2+}$; (2) CuHL^+ ; and (3), CuL^3 .

stereospecific effect of the unco-ordinated optically active butanol groups on the conformation of the ethylenediamine chelate ring. In order to estimate the importance of this latter proposal, the circular dichroism spectra of the corresponding copper(II) and nickel(II) complexes with *l*-*N,N'*-bis

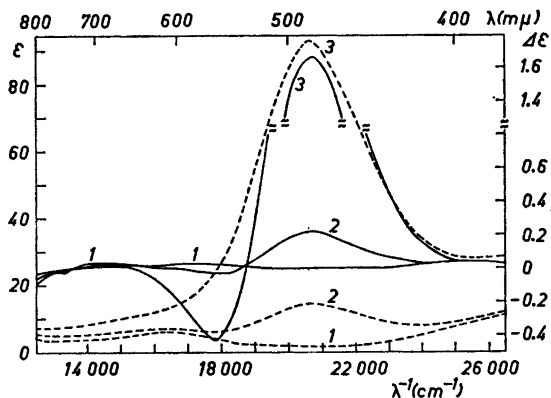


Fig. 2. Absorption (---) and circular dichroism (—) spectra for solutions of nickel(II) nitrate (5×10^{-3} M) and *d*-ethambutol dihydrochloride (5×10^{-3} M) at 20° and $I = 0.1$ (NaNO_3). Concentrations of NaOH are: (1), 1.0×10^{-2} M; (2), 1.5×10^{-2} M; (3), 2.0×10^{-2} M. Solutions (2) and (3) were clouded by slight precipitation. Species present are: (1), $\text{NiH}_2\text{L}^{2+}$; (2) and (3), uncertain composition.³

(1-(chloromethyl)propyl) ethylenediamine (structure above with X = Cl) were measured. In these complexes the optically active groups are unable to chelate.

The circular dichroism for the copper(II) complexes with this ligand* (Fig. 3) is

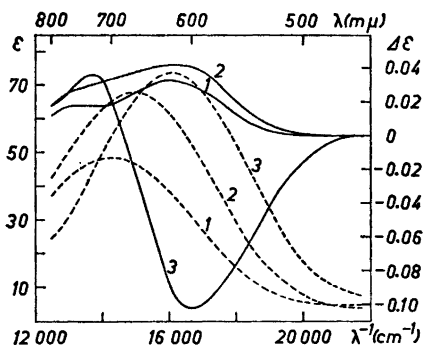


Fig. 3. Absorption (---) and circular dichroism (—) spectra for solutions of copper(II) nitrate (5×10^{-3} M) and *l*-N,N'-bis(1-(chloromethyl)propyl)ethylenediamine dihydrochloride (5×10^{-3} M) at 20° and $I = 0.1$ (NaNO_3). Concentrations of NaOH are: (1), 1.0×10^{-2} M; (2), 1.5×10^{-2} M; (3), 2.0×10^{-2} M. Species present probably have exclusive hydroxo bridging.

very much less than that for the corresponding ethambutol complexes and is of a different general shape. (Note the difference in the ordinate scale: 1 to 20). The nickel(II) complexes gave no observable dichroism and their absorption spectra did not show the same dramatic change as was found for the ethambutol complexes. Thus, the bulky unco-ordinated optically active groups on the nitrogens are able to impose a small amount of circular dichroism onto the *d-d* transitions of copper(II). However, this cannot account for the circular dichroism spectra of the ethambutol complexes which must, therefore, be due to the co-ordination of at least one, or more probably both, of the OH groups. Because the water molecules in the plane of the copper would be removed by the chelation

* By comparison with the complexes of N,N'-di-*sec*-butylethylenediamine,³ these complexes are probably polynuclear involving extensive hydroxo bridging.

of the butanol groups, these experiments also support the conclusion of Hawkins and Perrin³ that the protons must have been liberated from the ligand and not from co-ordinated water molecules to give the 'ionised' species.

Experimental. *d*-Ethambutol dihydrochloride and *l*-N,N'-bis(1-(chloromethyl)propyl)ethylenediamine dihydrochloride, supplied as research samples by Dr. A. Wander SA, Berne, and the Lederle Laboratories of the American Cyanamid Co., were dried at 100° and used without further purification. All other reagents were of AnalaR grade.

Solutions, prepared from standard solutions of metal nitrate and carbonate-free NaOH and weighed amounts of the ligand, were adjusted to an ionic strength of 0.1 with sodium nitrate. The investigations were carried out at 20° . From the stability constants and pK_a values determined by Hawkins and Perrin³ under identical conditions, it was concluded that, when 2, 3, and 4 equivalents of NaOH had been added, the copper(II) was almost completely in the forms, $\text{CuH}_2\text{L}^{2+}$, CuHL^+ and CuL , respectively. Unfortunately, similar data are not available for the nickel system.

Circular dichroism measurements were performed with a Roussel-Jouan Dichrograph possessing an extended wavelength scale (800–220 μ). The absorption spectra were measured with a Cary Model 14 recording spectrophotometer.

Acknowledgements. The author is most grateful to the Lederle Laboratories of the American Cyanamid Company and Dr. A. Wander SA, Berne, for research samples of ethambutol and related compounds, to Professor J. Bjerrum for working facilities in his Laboratory, and to members of the department for valuable discussions. The research was carried out while the author possessed an I.C.I.A.N.Z. Travelling Fellowship and a Rask-Ørsted Foundation grant for which he is very grateful.

1. Larsen, E. and Olsen, I. *Acta Chem. Scand.* **18** (1964) 1025.
2. Wilkinson, R. G., Cantrall, M. B. and Shepherd, R. G. *J. Med. Pharm. Chem.* **5** (1962) 835.
3. Hawkins, C. J. and Perrin, D. D. *Unpublished results.*

Received August 3, 1964.