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structural formula of tetracycline with groupings A, B, and C

Figure 1: structural formula of tetracycline with groupings A, B, and C

Abstract

Full Text

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STUDY OF THE DISSOCIATION OF TETRACYCLINES BY INFRARED SPECTROSCOPY

(Presented by Academician M. M. Shemyakin, February 20, 1964)

Tetracycline antibiotics: tetracycline (I), oxytetracycline (II), and chlortetracycline (III) possess three dissociation constants associated with the tricarbonylmethane (A), dimethylamino (B), and phenoldiketone (C) groupings:

$$\begin{aligned} R_1 = H; R_2 = H \text{ (I). } & \quad pK_{a_1} = 3.30; pK_{a_2} = 7.74; pK_{a_3} = 9.27, \\ R_1 = H; R_2 = OH \text{ (II). } & \quad pK_{a_1} = 3.27; pK_{a_2} = 7.32; pK_{a_3} = 9.11, \\ R_1 = Cl; R_2 = H \text{ (III). } & \quad pK_{a_1} = 3.30; pK_{a_2} = 7.68; pK_{a_3} = 9.69. \end{aligned}$$

Meanwhile, up to the present time there has been no complete clarity in assigning the indicated constants to definite functional groupings of tetracycline.

In the detailed work of Stephens and co-workers¹, on the basis of determining the pK_a values of a series of model compounds, as well as of certain tetracycline derivatives, pK_{a_1} , pK_{a_2} , and pK_{a_3} were assigned to the groupings A, B, and C, respectively. However, in a note to this work doubt was expressed as to the correctness of the assignment of pK_{a_1} and pK_{a_2} . The fact that the experimental results allow an ambiguous interpretation indicates the insufficient reliability of the method used by the authors.

More reliable results should be obtained by studying the dissociation of tetracyclines by means of IR spectroscopy. The point is that IR spectra characterize the local bonds of dissociating groupings, and the changes in structure and electronic configuration of the molecule associated with dissociation lead to quite definite changes in the vibrational spectra, readily verifiable on model compounds.

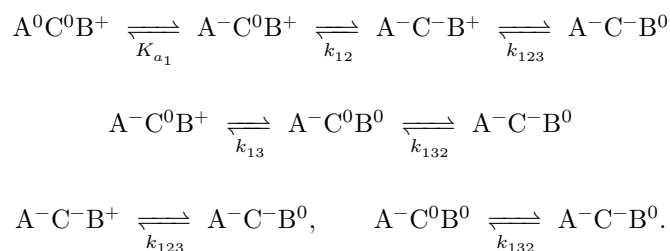
It should be noted that, because of interaction among the dissociating groups, the macroconstants obtained by potentiometric titration are not equal to the microconstants corresponding to individual acts of ionization². With the aid of IR spectroscopy we can assign to the dissociating groupings precisely those

structural formula of tetracycline derivative with substituents R_3 and R_4

Figure 2: structural formula of tetracycline derivative with substituents R_3 and R_4

microconstants that make the principal contribution to the corresponding macroconstants.

Anticipating somewhat, we shall depict the following scheme of tetracycline dissociation, constructed with allowance for the values of their pK_a^* .

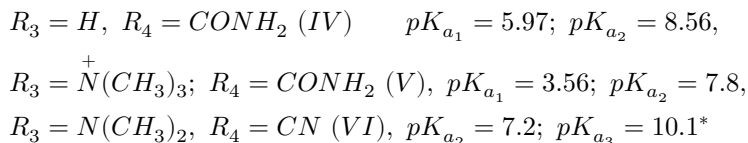


* The pK_a values of nirylytetracycline (VI) are given for a 50% solution of dimethylformamide in water; the pK_a values of the remaining compounds (I–V) are for aqueous solutions.

From this scheme it is evident that after ionization of the A^0 grouping, dissociation of both the C^0 and B^+ groupings is possible. From the data obtained by means of IR spectroscopy, it is apparent that the first dissociation path is predominantly realized. This also agrees with the calculations carried out in work ⁽³⁾, showing that the main contribution to the macroconstants K_{a_2} and K_{a_3} is made by the microconstants k_{12} and k_{123} .

Our work was complicated by the fact that the IR spectra of tetracyclines have not been sufficiently studied, although several investigations ^(4,5) have been devoted to this question. Therefore we also carried out work on assigning some bands in the IR spectra of tetracyclines—chiefly those which undergo changes upon dissociation.

For this purpose we used the following principal tetracycline derivatives:



In compounds IV–VI, which differ little in structure from the original tetracycline, the A and B groupings have been changed: in compound IV the dimethylamino group has been removed, while in V it has acquired a permanent charge;

structural formulas of auxiliary compounds VII, VIII, IX, and X

Figure 3: structural formulas of auxiliary compounds VII, VIII, IX, and X

in compound VI, owing to the influence of the $C \equiv N$ group, the hydroxyl at C_3 is dissociated already in the strongly acidic region.

The IR spectra of these compounds, as well as of the original tetracyclines, were recorded both in the solid state (in KBr pellets) and in aqueous solutions (in H_2O and D_2O) at pH values corresponding to the maximum content of each of the ionized forms of tetracycline, selected with allowance for the pK_a values of the latter. Thus, for example, for tetracycline these pH values were 1.5; 5; 8.7 and 11. In addition, the indicated compounds were studied in the pH range from 7 to 11 at intervals of 0.2-0.3 pH unit. Comparison of the IR spectra of compounds I-VI allowed us to reveal the spectral changes associated with dissociation of each of the three groupings, and thereby to assign pK_{a_1} to the tricarbonylmethane, pK_{a_2} to the phenoldiketone, and pK_{a_3} to the dimethylamino groupings, as well as to obtain additional data for assigning the IR spectra.

In addition to the principal compounds I-IV, we also investigated auxiliary compounds VII-X.

VII VIII IX X

Below we present the principal data on the IR spectra of tetracyclines in connection with their dissociation. In the region of pK_{a_1} for compounds I-V, but not VI, we observe the following spectral changes, indicating dissociation of the tricarbonylmethane grouping: the broad absorption band at 1550 cm^{-1} (vibrations of $C = O$ and $C = C$), characteristic of the enol ^(6,7), shifts by $30\text{-}40\text{ cm}^{-1}$ toward lower frequencies; the absorption band of $C = O$ of the amide group likewise decreases in frequency from 1670 cm^{-1} to $1640\text{-}1650\text{ cm}^{-1}$; the bands of the stretching vibrations of the enol in the region $2600\text{-}2800\text{ cm}^{-1}$ disappear.

Dissociation of the phenoldiketone grouping leads to a shift of the band at $1230\text{-}1240\text{ cm}^{-1}$, associated with vibrations of the phenolic hydroxyl, to $1260\text{-}1270\text{ cm}^{-1}$. At the same time, considerable changes occur in the region $820\text{-}870\text{ cm}^{-1}$, corresponding to deformation vibrations of CH of a polysubstituted aromatic ring, and a strong band appears at 1430 cm^{-1} . The latter is apparently associated with the $C_{11}\text{-}C_{12}$ grouping, which, after dissociation of the phenolic hydroxyl, is capable of forming a strong chelate ring, similar to what occurs in dimers of carboxylic acids, in whose spectra a strong band is also observed at 1400 cm^{-1} ⁽⁸⁾. This may be confirmed by the absence of the indicated band in isochlorotetracycline VII, in which the phenoldiketone grouping is destroyed.

Finally, as a result of dissociation of the dimethylammonium cation, characteristic changes are observed in the region of CH stretching vibrations. The uncharged dimethylamine group has two distinct bands at 2785 and 2825 cm^{-1} , which disappear upon formation of the dimethylammonium cation, as, for ex-

ample, in compounds VIII and X.

The study of the dissociation of tetracyclines by means of UV spectra, from the bands at 268 m μ and 355 m μ associated with groupings A and C, respectively (the assignment was made with the aid of model compounds VIII and IX), confirms our conclusions concerning the distribution of pK_a.

After the main results of our work had already been obtained and reported at the XV All-Union Conference on Spectroscopy⁽⁹⁾ in July 1963, an article by Leeson⁽³⁾ appeared, in which, on the basis of the results of potentiometric titration of tetracyclines, an assignment of pK_a was made that confirms our data.

In conclusion, we express our gratitude to M. N. Kolosov for providing samples and to E. I. Pokrovsky, N. A. Margolina, and E. F. Fedorova for discussion of the results and assistance in the work.

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CITED LITERATURE

- ¹ C. K. Stepheuz, K. Murai et al., *J. Am. Chem. Soc.*, **78**, 4155 (1956).
- ² J. T. Edsall, J. Wyman, *Biophysical Chemistry*, **1**, N. Y., 1958, p. 495.
- ³ L. J. Leeson, J. E. Kinger, A. Nach, *Tetrahedron Letters*, No. 18, 1155 (1963).
- ⁴ F. Meda, P. Scrocco, *Gazz. chim. Ital.*, **85**, 364 (1955).
- ⁵ J. R. Lacher, J. L. Bitner, J. L. Park, *J. Phys. Chem.*, **59**, 610 (1955).
- ⁶ C. Duvar, J. Leiomte, *Rev. chim. (Romin.)*, **7**, No. 1, 169 (1962).
- ⁷ O. Ya. Neiland, G. Ya. Vanag, *Usp. khim.*, **28**, 436 (1959).
- ⁸ D. Hadzi, N. Sheppard, *Proc. Roy. Soc. A*, **216**, 247 (1953).
- ⁹ B. G. Belen' kii, K. K. Kalininysh, E. I. Pokrovskii, *Infrared spectroscopy of aqueous solutions*, report at the XV All-Union Conference on Spectroscopy, Minsk, 1963.

Note: Figure translations are in progress. See original paper for figures.

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