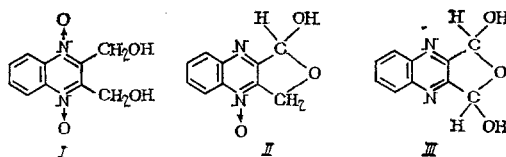


INVESTIGATION OF THE KINETICS OF THE REDOX REACTIONS OF DIOXIDINE

K. P. Tetenchuk, G. G. Dvoryantseva,
I. S. Musatova, and A. S. Elina

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Dioxidine [2,3-bis(hydroxymethyl)quinoxaline 1,4-dioxide] (I) is an effective medicinal preparation that is used in the treatment of acute bacterial infections [1, 2]. It is known that the N-oxides of α -hydroxyalkyl derivatives of azines display a capacity for undergoing various types of redox reactions in both alkaline and acidic media [3-7]. It has been previously shown that I in the presence of alkaline reagents undergoes redox transformations that lead to the formation of 2-(hydroxymethyl)-3-formylquinoxaline 1-oxide, which is isolated in the form of cyclic hemiacetal II, or, in the case of excess alkaline reagent, to the formation of 2,3-diformylquinoxaline cyclic hemiacetal III [4].



The kinetics and mechanisms of these reactions and the ability of I to undergo redox transformations in acidic media have not been investigated. In addition, these data are of considerable interest in the study of a number of problems involved in the technology of the manufacture of dioxidine (I) and its metabolism.

In the present research we investigated the kinetics of the redox reactions of I in aqueous and alcohol (CH_3OH , $\text{C}_2\text{H}_5\text{OH}$) solutions in the presence of alkaline (NaOH) and acidic (HCl) reagents. The reaction was studied in alkaline media at I concentrations from 10^{-3} to 0.5 M and at various alkali concentrations (from $2 \cdot 10^{-3}$ to 0.5 M). The data on the kinetics of the redox reaction in an acidic medium presented below were obtained for 10^{-3} M solutions of I in a 0.3 M alcohol (CH_3OH) solution of hydrochloric acid. All of the measurements were made at 25°C.

We used UV spectrophotometry as the principal method of investigation. It is known that the N-oxidation of quinoxaline derivatives has a characteristic effect on the positions and intensities of the π - π^* transitions observed above 210 nm [8]. The possibility of the application of UV spectroscopy for the study of the redox reactions of I was determined by the fact that the changes in the UV spectra observed on passing from alkyl and hydroxyalkyl derivatives of quinoxaline to their N-oxides and N,N'-dioxides are similar to the changes that occur in the N-oxidation and N,N'-dioxidation of unsubstituted quinoxaline (Table 1 and Fig. 1a).

The investigations showed that the redox transformations of I in alcoholic alkaline media proceed in two consecutive kinetically controlled steps: The first step involves the formation of N-oxide II, and the second step involves the conversion of II to dialdehyde hemiacetal III. Thus in the measurement of the UV spectra of 10^{-3} M solutions of I in alcohol in the presence of alkali at a molar ratio of I and NaOH of 1:2 we observed that a successive decrease in the intensities of the bands at 265 and 380 nm, which are characteristic for I, and a parallel increase in the intensities of the bands at 322 and 340 nm, which are characteristic for II, occur during a period of 3 h from the start of the reaction. The presence of a distinct isobestic point at 348 nm constitutes evidence that during this period of time the process is restricted only by the first step of the redox reaction (Fig. 1b). The subsequent changes in the UV spectra are accompanied by a further decrease in the intensities of the bands related to I, a simultaneous decrease in the intensity of the band at 340 nm, which is characteristic for II, and an increase in the intensity of the absorption at 320 nm. A small hypsochromic shift (from 243 nm to 238 nm) and a decrease in the intensity of the short-wave

S. Ordzhonikidze All-Union Scientific-Research Institute of Pharmaceutical Chemistry, Moscow. Scientific-Research Institute of Pharmaceutical Chemistry, Novokuznetsk. Translated from *Khimiko-farmatsevticheskii Zhurnal*, Vol. 18, No. 5, pp. 598-604, May, 1984. Original article submitted June 24, 1983.

TABLE 1. UV Spectra of Quinoxaline Derivatives in 96% Ethanol

Compound	λ_{\max} , nm (lg ϵ)	
Quinoxaline	318 (3,60)	235 (4,30)
2,3-Dimethylquinoxaline	318 (3,86)	235 (4,37)
2,3-Bis(hydroxymethyl)quinoxaline	318-300 (3,88)	235 (4,49)
2,3-Diformylquinoxaline hemiacetal	322 (3,90)	238 (4,45)
Quinoxaline 1-oxide	341 (3,84) 324 (3,93)	238 (4,58)
2-(Hydroxymethyl)-3-formylquinoxaline hemiacetal 1-oxide	340 (3,93) 322 (4,00)	243 (4,60)
Quinoxaline 1,4-dioxide	385-378 (3,69)	300-285 (3,40) 256 (4,23) 240-232 (4,32)
2,3-Dimethylquinoxaline 1,4-dioxide	376 (4,13)	290 (3,25) 260 (4,56) 235 (4,30)
2,3-Bis(hydroxymethyl)quinoxaline 1,4-dioxide (dioxidine)	380 (4,09)	300-290 (3,44) 265 (4,45) 235 (4,35)

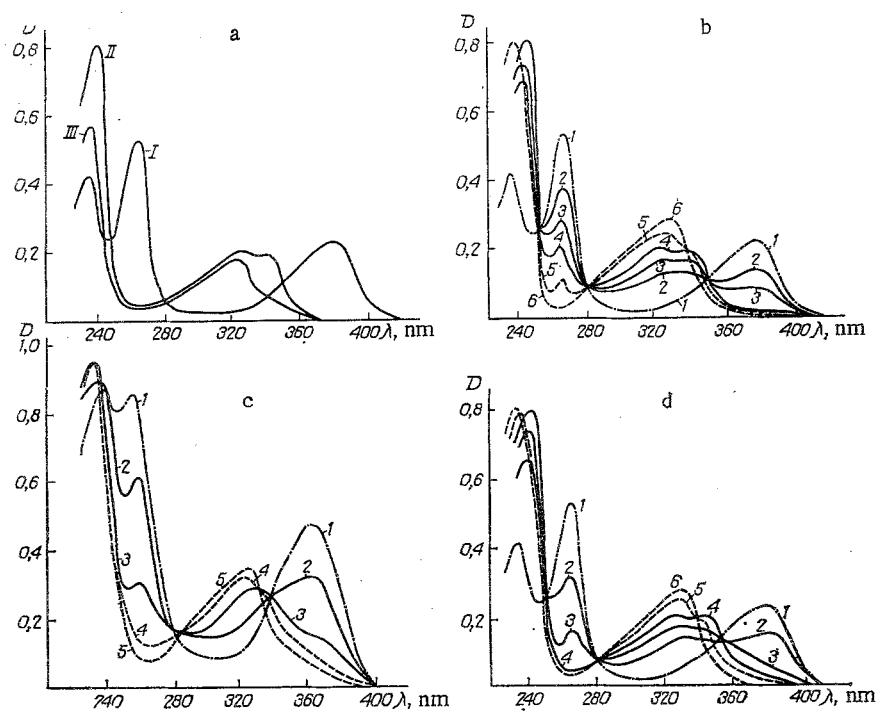


Fig. 1. UV spectra of dioxidine in aqueous and alcohol media in the presence of alkaline and acidic agents: a) I-III in 96% ethanol; b) I in a $2 \cdot 10^{-3}$ M alcohol solution of NaOH, $C_0 = 10^{-3}$ M: 1) starting solution, $t = 2$; 2) $t = 1$ h; 3) $t = 2$ h; 4) $t = 3$ h; 5) $t = 6$ h; 6) $t = 9$ h; c) I in a 0.5 M aqueous solution of NaOH, $C_0 = 10^{-3}$ M: 1) $t = 0$; 2) $t = 1$ h; 3) $t = 3$ h; 4) $t = 6$ h; 5) $t = 8$ h; d) I in a 0.3 M alcohol solution of HCl, $C_0 = 10^{-3}$ M: 1) $t = 0$; 2) $t = 15$ min; 3) $t = 30$ min; 4) $t = 1$ h; 5) $t = 56$ h; 6) $t = 64$ h.

absorption band are simultaneously observed. A comparison with the spectra of I-III (see Fig. 1a) makes it possible to conclude that the indicated changes in the absorption curves during the period from 3 h to 9 h from the start of the reaction are due to the successive occurrence of the first and second steps of the redox reaction. The appearance of a second isobestic point at 337 nm correspond to a second step, i.e., to conversion of N-oxide II to dialdehyde hemiacetal III. The spectra measured 9 h after the start of the reaction correspond to complete conversion of I to final product. The kinetic curves of the redox

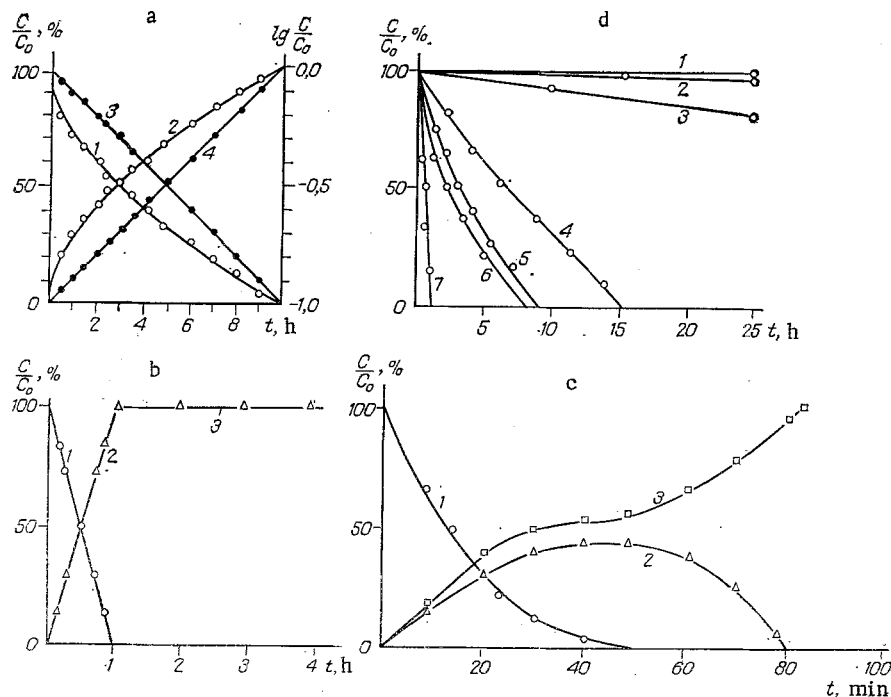


Fig. 2. Kinetic curves of the redox reaction of dioxidine: a): 1) consumption of I; 2) accumulation of II and III, $C_0(I) = 10^{-3}$ M; b): 1) consumption of I; 2) accumulation; 3) consumption of II in an 0.3 M alcohol solution of HCl, $C_0 = 10^{-3}$ M; c): 1) consumption of I; 2) accumulation and consumption of II; 3) accumulation of II in a 0.5 M alcohol solution of NaOH, $C_0 = 0.5$ M; d) kinetic curves of the consumption of I at various alkali and acid concentrations in aqueous and alcohol solutions: 1) 12 M HCl/H₂O, $C_0 = 10^{-3}$ M; 2) 0.03 M NaOH/CH₃OH, $C_0 = 1$ M; 3) $2 \cdot 10^{-3}$ M NaOH/H₂O, $C_0 = 10^{-3}$ M; 4) 0.1 M NaOH/H₂O, $C_0 = 10^{-3}$ M; 5) $2 \cdot 10^{-3}$ M NaOH/CH₃OH, $C_0 = 10^{-3}$ M; 6) 0.5 M NaOH/H₂O, $C_0 = 10^{-3}$ M; 7) 0.3 M HCl/CH₃OH, $C_0 = 10^{-3}$ M.

transformation of I (curve 1) and the accumulation of the reaction products (curve 2) are presented in Fig. 2a. The two curves intersect at one point corresponding to reaction time $t = 3$ h. The branch of curve 2 corresponding to the time interval from $t = 0$ to $t = 3$ h consequently characterizes an increase in the concentration of only N-oxide II. The 50% decrease in the concentration of I corresponds to the first step in the reaction under the conditions indicated above. An analysis of the kinetic curves by the usual method [9] showed that both the overall redox reaction $I \rightarrow III$ and the first step in this process $I \rightarrow II$ obey a first-order kinetic equation. The time of half conversion of I calculated from the first-order equation ($\tau_{1/2} = 3$ h) corresponds precisely with the time interval determined by the point of intersection of kinetic curves 1 and 2 presented in Fig. 2a. The rate constant of the first step $I \rightarrow II$ ($K_1 = 6.7 \cdot 10^{-5} \text{ sec}^{-1}$) proved to be close to the rate constant of the overall reaction $I \rightarrow III$ (Table 2).

A similar analysis of the kinetic curves measured in alcohol and aqueous solutions of I with various concentrations of alkali and I showed that the sequence of two steps in the redox reaction $I \rightarrow II \rightarrow III$ is retained under all of the examined conditions and satisfactory conformity of the reaction rates with a first-order kinetic equation is observed. The kinetic curves (see Fig. 2) and the data on the rate constants and half-conversion times of I (see Table 2) make it possible to examine the effect of the medium and the concentration of the alkaline reagent on the kinetics of the process.

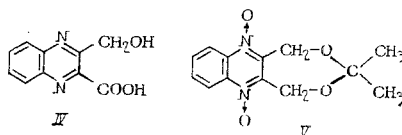
At a fixed concentration of I and the alkali (10^{-3} M solutions of I, molar ratio of I and NaOH of 1:2) the transition from an alcohol to an aqueous medium leads to marked retardation of the reaction: the rate constant decreases by a factor of ~ 1.5 . Under the conditions of a slow $I \rightarrow III$ reaction each step of the process cannot be accurately isolated; however, a qualitative examination of the UV spectra makes it possible to conclude that the rate constants of the first and second steps decrease symbatically. An increase in the alkali con-

TABLE 2. Rate Constants (K) and Half-Conversion Times ($\tau_{1/2}$) of Dioxidine (I) in Redox Reaction I \rightarrow III in Alkaline Media ($t = 25^\circ\text{C}$)

C_0 (I), M ^a	C_0 (I)/C (NaOH) ^b	Medium	K, sec ⁻¹	$\tau_{1/2}$, h
10^{-5}	1:2	CH ₃ OH	$6,8 \cdot 10^{-5}$	2,83
10^{-3}	1:2	H ₂ O	$1,8 \cdot 10^{-6}$	106,94
10^{-3}	1:100	H ₂ O	$2,7 \cdot 10^{-5}$	7,13
10^{-3}	1:500	H ₂ O	$9,0 \cdot 10^{-5}$	2,14
1,0	1:0,03	CH ₃ OH	$5,5 \cdot 10^{-7}$	350,00
0,5	1:1	CH ₃ OH	$1,5 \cdot 10^{-3}$	0,13

^aInitial concentration of I. ^bMolar ratios of I and NaOH.

centration in aqueous solution leads to an increase in the rate of redox transformation of I; however, the rate constants of the reaction become comparable to the values found for the alcohol medium (see Fig. 1c and Fig. 2d) only in the case of large molar excesses of alkali (1:100, 1:500). As the concentrations of I (up to 0.5 M) and alkali (up to 1.0 M) in water are increased simultaneously, the reaction rate increases to such an extent that it cannot be monitored by means of the methods of investigation used in this research. Under these conditions, in addition to II and III, we isolated 2-(hydroxymethyl)-3-carboxyquinoxaline (IV), which is evidently formed as a consequence of further transformations of dialdehyde hemiacetal III via the Cannizzaro reaction. At the same time, a decrease in the rate of reaction I \rightarrow III by two orders of magnitude is observed when the concentration of I is increased from 10^{-3} to 1 M in an alcohol medium but in the presence of catalytic amounts of alkali.



An independent confirmation of the sequence of the steps of the redox reaction I \rightarrow II \rightarrow III was obtained by means of PMR spectroscopy. A comparison of the spectra of I-III measured in methanol (Fig. 3, spectra a-c) clearly illustrates the characteristic (for these compounds) differences in the positions and relative intensities of the signals of the methyldyne, methylene, and aromatic protons.

In addition to signals of the starting substance, signals of N-oxide II and dialdehyde hemiacetal III of comparable intensity are present in the spectrum of an alcohol solution measured 10 min after preparation in the case of an equimolar ratio of I and alkali (0.5 M) (Fig. 3d). The changes in the spectra with time are accompanied by a successive decrease in the intensities of the signals of the starting substance until they disappear completely 50 min after the start of the reaction and by an increase in the intensities of the signals of II and III. The further changes in the spectra correspond only to the second step of the reaction II \rightarrow III up to the point of complete conversion of N-oxide II to dialdehyde hemiacetal III. The results obtained are illustrated by the kinetic curves measured from the intensities of the corresponding signals in the PMR spectra (see Fig. 2c). In the case of close molar ratios of I and NaOH in a methanol medium an increase in the concentration of I from 10^{-3} to 0.5 M leads to an increase in the reaction rate by two orders of magnitude (see Fig. 2). It is important to note that signals of a dialdehyde hemihydrate of comparable intensity are observed in the PMR spectrum of a solution of N-oxide II as the starting substance measured under the same conditions 10 min after dissolving of II (Fig. 3g). The set of data obtained constitute evidence that in alkaline, alcohol, and aqueous media the rate constants of the two consecutive steps of the reaction are comparable in magnitude ($K_1 \approx K_2$) and increase symbatically with respect to increases in the concentration of starting I and alkali.

The investigations carried out by UV spectrophotometry showed that the same redox reaction is observed in alcohol solutions of hydrochloric acid. However, in the case of comparable molar concentrations of alkali or acid in an alcohol medium (0.2-0.3 M) the reaction in acidic solutions proceeds considerably more slowly. At room temperature the redox transformation of I proceeds at a kinetically controlled rate at an I concentration of 10^{-3} M in an 0.3 M alcohol solution of HCl (Fig. 1d). Two successive steps with the formation of N-oxide

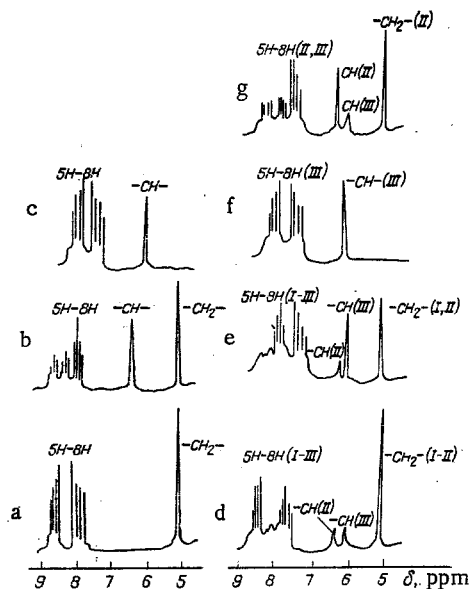


Fig. 3. PMR spectra: I (a), II (b), and III (c) in CD_3OD ; I in 0.5 M $\text{NaOH}/\text{CD}_3\text{OD}$, $C_0 = 0.5$ M when $t = 10$ min (d), $t = 40$ min (e), and $t = 90$ min (f); II in 0.5 M $\text{NaOH}/\text{CD}_3\text{OD}$, $C_0 = 0.5$ M when $t = 10$ min (g).

II as an intermediate are also observed under these conditions. However, the ratio of the rates of the first and second steps of the reaction changes markedly in an acidic medium. Under the indicated conditions the rate constant of the second step ($K_2 = 3.6 \cdot 10^{-6} \text{ sec}^{-1}$) is almost two orders of magnitude lower than the rate constant of the first step ($K_1 = 6.0 \cdot 10^{-4} \text{ sec}^{-1}$). It follows from the kinetic curves (see Fig. 2b) that in an acidic medium at low I concentrations (10^{-3} M) the two steps of the process are separated completely, and the formation of III is observed after complete conversion of I to II. Replacement of alcohol by water in an acidic medium is accompanied by a considerably more pronounced decrease in the rate of the redox reaction and primarily the rate of its first step as compared with the similar replacement of the solvent in the presence of alkali (see Fig. 2d). Even in concentrated aqueous solutions of HCl (up to 12 M) at room temperature the UV spectra of 10^{-3} M solutions of I remain virtually unchanged for at least 24 h. This result is confirmed by monitoring of the composition of the reaction mixture by means of thin-layer chromatography (TLC).

The marked difference in the rate constants of the first and second steps of the reaction enabled us to preparatively isolate both products of the redox transformation of I in an acidic medium and to confirm their structures by means of elementary analysis and physico-chemical methods of investigation. The isolation of N-oxide II in 76% yield is achieved by heating I in concentrated HCl at 80–85°C. Under these conditions final product III can be isolated in only ~2% yield. When we treated the reaction mixture with acetone, in addition to II and III, we isolated a compound, the structure of which, according to the results of elementary analysis and data from the IR, PMR, and mass spectra, corresponds to the O-isopropylidene derivative (V) of 2,3-bis(hydroxymethyl)quinoxaline 1,4-dioxide. When V is heated in 96% ethanol, it is converted quantitatively to I.

Thus, we obtained the following principal results on the basis of our investigation. Two successive steps in the redox transformation of dioxidine to dialdehyde hemiacetal III with the formation of N-oxide II as an intermediate are observed in aqueous and alcohol solutions in the presence of alkali or acid. In alkaline media the two steps proceed at comparable rates, whereas in an alcohol solution of acid the rate constant of the second step is two orders of magnitude lower than the rate constant of the first step of the reaction. At a fixed concentration of dioxidine in the presence of both alkali and acid the reaction rate decreases sharply on passing from alcohol to aqueous solutions. In alkaline media the reaction rate increases as the concentrations of the starting substance and (or) alkali increase.

EXPERIMENTAL

All of the experiments were carried out under conditions that provided for protection of the investigated solutions from the action of light. The UV spectra were recorded with a Specord UV-vis spectrophotometer (East Germany). The PMR spectra of solutions in d_6 -DMSO were obtained with a Tesla BS-497 spectrometer (Czechoslovakian SSR) (100 MHz) with tetramethylsilane as the internal standard. The IR spectra of KBr pellets of the compounds were recorded with a UR-20 spectrometer (East Germany). The mass spectra were obtained with a

Varian MAT-112 chromatographic mass spectrometer (West Germany) at an ionizing-electron energy of 70 eV and an ionization-chamber temperature of 180°C with direct introduction of the samples into the ion source. Chromatography was carried out on Silufol UV-254 plates in a chloroform-heptane-ethanol system (50:45:15) with development in UV light.

Redox Reactions of Dioxidine (I) in an Acidic Medium. A) A mixture of 1 g of I and 3 ml of hydrochloric acid (sp. gr. 1.19) was heated at 80–85°C for 1 h, after which it was cooled and treated with 10 ml of acetone. After 2 h, workup gave 0.1 g (8.5%) of V with mp 199–200°C (dec.). Found, %: C 59.47; H 5.46; N 10.46. $C_{13}H_{14}N_2O_4$. Calculated, %: C 59.53; H 5.46; N 10.67. Mass spectrum, m/e: 262.2 (M^+). PMR spectrum, δ , ppm: 1.42 s (CH_3), 5.06 s (CH_2), and 7.86–8.38 m (5H–8H).

Workup of the filtrate after 3 h gave 0.76 g (82.6%) of II, which was identical to a genuine sample of II [4] with respect to its melting point and IR and PMR spectra. See Table 1 and Fig. 1a for the UV spectrum.

B) A 3-ml sample of hydrochloric acid (sp. gr. 1.19) was added to 1 g of I, and the mixture was heated at 80–85°C for 1 h, after which it was cooled to 20–25°C. After 2 h, workup gave 0.44 g of a crystalline substance, which was subjected to sublimation at 115–120°C (2 mm) to give 0.02 g of 2,3-diformylquinoxaline with mp 175–176°C (dec.) (mp 177–178°C [10]). The substance obtained when this product was dissolved in aqueous ethanol was identical to dialdehyde hemiacetal III, according to its R_f value and UV spectrum.

2-(Hydroxymethyl)-3-carboxyquinoxaline (IV). A mixture of 0.5 g of I and 5 ml of a 1 N aqueous solution of sodium hydroxide was maintained at 20–25°C for 24 h, after which the mixture was acidified to pH 1.0 with a 1-N solution of hydrochloric acid to give 0.44 g (97%) of IV with mp 114°C (dec., from alcohol). Found, %: C 58.62; H 4.01; N 13.67. $C_{10}H_8N_2O_3$. Calculated, %: C 58.82; H 3.96; N 13.71. PMR spectrum, δ , ppm: 5.00 s (CH_2), 6.65 broad s (OH) (vanishes when D_2O is added), and 7.90–8.12 m (5H–8H). IR spectrum, ν_{max} , cm^{-1} : 1750 (C=O); 3440, 3110 (OH_{assoc}).

LITERATURE CITED

1. A. S. Elina and E. N. Radeiskaya, in: Collection of Research from the All-Union Scientific-Research Institute of Pharmaceutical Chemistry [in Russian], Vol. 2, Moscow (1971), pp. 197–209.
2. E. N. Padeiskaya, in: New Chemotherapeutic Preparations for the Treatment of Patients Afflicted with Infectious Diseases [in Russian], Moscow (1976), pp. 89–103.
3. W. S. Chilton and A. K. Butler, *J. Org. Chem.*, **32**, 1270–1272 (1967).
4. A. S. Elina, L. G. Tsyrul'nikova, and G. P. Syrova, *Khim. Geterotsykl. Soedin.*, No. 1, 149–153 (1969).
5. C. W. Muth, J. C. Patton, B. Bhattacharya, et al., *J. Heterocycl. Chem.*, **9**, 1299–1304 (1972).
6. J. P. Dirlam and I. W. McFarland, *J. Org. Chem.*, **42**, 1300–1304 (1977).
7. A. S. Elina, I. S. Musatova, R. M. Timkova, et al., *Khim. Geterotsykl. Soedin.*, No. 8, 1106–1108 (1980).
8. M. M. Kaganskii, G. G. Dvoryantseva, I. V. Sokolova, et al., *ibid.*, No. 1, 118–124 (1975).
9. L. Hammett, *Fundamentals of Physical Organic Chemistry* [Russian translation], Moscow (1972), pp. 71–126.
10. E. J. Moriconi, A. J. Fritsch, et al., *J. Org. Chem.*, **30**, 1542 (1965).