Brief Communications

Chemistry of naphthazarin derivatives
10.* First direct observation of prototropic tautomerism of 1´-hydroxyalkylnaphthazarins by IR spectroscopy

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It is found by IR spectroscopy that 1´-hydroxyalkylnaphthazarins in aprotic organic solvents exist as a mixture of the 1,4- and 1,5-naphthoquinonoid tautomers.

Key words: 1´-hydroxyalkylnaphthazarins, (2-alkyl-1´,5,8-trihydroxy-1,4-naphthoquinones); shikalkin, shikonin, alkannin; prototropic tautomerism; IR spectroscopy of 1´-hydroxyalkylnaphthazarins in solutions.

We have previously[1,2] reported the use of IR spectroscopy to study the prototropic tautomerism of mono- and dihydroxy derivatives of naphthazarin (5,8-dihydroxy-1,4-naphthoquinone) (**1**). Based on analysis of the frequencies of the stretching vibration of the β-hydroxy groups at C(2) and the intensity of the corresponding absorption bands, we have shown that in aprotic media these compounds exist as a mixture of the tautomeric 1,4-naphthoquinonoid forms of the types **1(A)** and **1(B)** (Scheme 1).

The problem on the tautomerism of 1´-hydroxyalkylnaphthazarins **2a,b** remains unclear, although there were several attempts to elucidate whether shikalkin (**2b**), *viz.*, one of the representatives of this class of compounds, is prone to similar mutual transformations in solutions or not. Only NMR (^{1}H and ^{13}C) spectroscopy was used[3—5] to solve this problem. However, the high rate of tautomeric transitions in naphthazarins (in the NMR time scale) did not allow one to determine the tautomeric composition of shikalkin in solutions. Based on the NMR study, the researchers have drawn different conclusions about the forms of this compound in a solution: only forms **C** and **D** [3]; forms **A**, **B**, **C**, and **D** [4]; and predominantly form **A** [5] (Scheme 2).

We applied IR spectroscopy to study the tautomeric equilibrium in solutions of 1´-hydroxyalkylnaphthazarins. In the framework of this method, we used the possibility of the hydroxy group at C(1´) to form hydrogen bonds different in strength, depending on the localization of the

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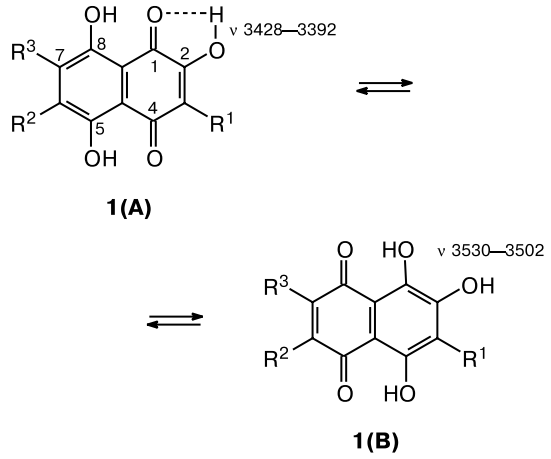
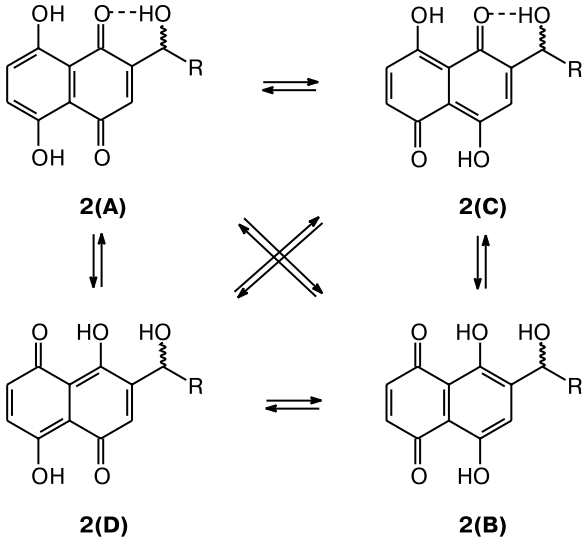
Scheme 1



R^1, R^2, R^3 = H, Alk, OMe

Scheme 2



R = Me (**a**), $CH_2CH{=}CMe_2$ (**b**)

1′-hydroxyalkyl substituent (in the quinonoid or benzenoid ring). Indeed, in the IR spectra of 1′-hydroxyalkylnaphthazarins, the frequencies of the stretching vibration of the hydroxy group (ν(OH)) at C(1′) differ if the hydroxyalkyl substituent is localized in different rings. For example, a solution of compound **2a** in hexane is

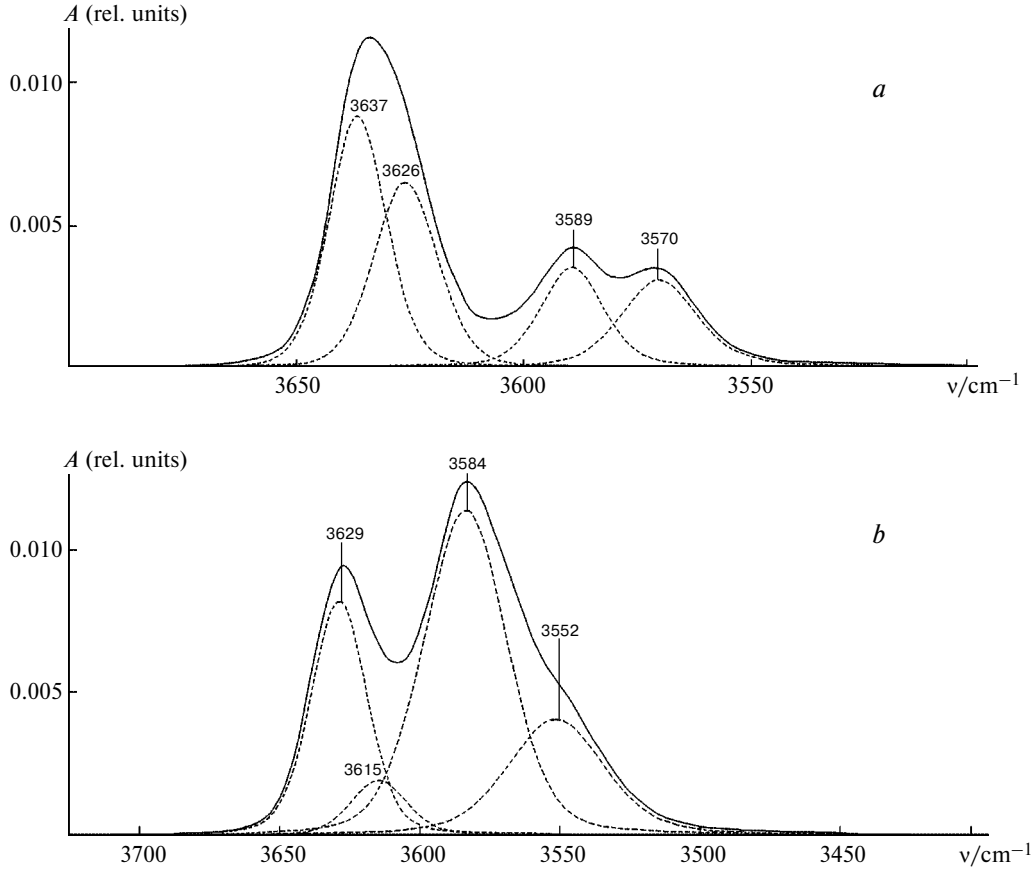


Fig. 1. IR spectra of compounds **2a** (*a*) and **2b** (*b*) in hexane (the region of ν(OH) frequencies) (dotted lines show the deconvolution of the ν(OH) absorption band contour).

characterized by the complicated IR spectrum in the absorption region of these hydroxy groups (Fig. 1, *a*). In addition, the region considered contains three well resolved ν(OH) absorption bands at 3635, 3589, and 3570 cm^{-1} instead of the expected two bands. The pronounced asymmetry of the low-frequency shoulder is observed for the high-frequency band.*

The mathematical deconvolution of the spectral contour by the Levenberg—Marquardt algorithm[6] gave four components (see Fig. 1, *a*). A similar result was obtained by analysis of the contours of the ν(OH) absorption bands of compound **2a** in CCl_4 and $CDCl_3$. The trivial case where a solution contains associates with different compositions can be excluded on the basis of the experiments on dilution. Therefore, the data obtained suggest that compound **2a** in hexane, CCl_4, and $CDCl_3$ exists as a mixture of four tautomers, *i.e.*, 1,5-naphthoquinonoid forms **2a(C)** and **2a(D)** exist in a solution in addition to 1,4-naphthoquinonoid forms **2a(A)** and **2a(B)** (see Scheme 2). We assigned two lower-frequency components of the ν(OH) band contour at 3589 and 3570 cm^{-1} (see Fig. 1) to tautomers **2a(A)**, **2a(C)** or **2a(C)**, **2a(A)**, respectively, because their 1′-hydroxy group is bound to the carbonyl group at the C(1) atom by the intramolecular hydrogen bond. The ratio of the sum of areas of two higher-frequency bands (S_b), which are assigned to tautomers **2a(B)** and **2a(D)**, to the total area of all bands (S) is equal to 0.66.

The high-frequency region of the IR spectrum of shikalkin (**2b**), which is a racemic mixture of epimers of shikonin (1′*R*) and alkannin (1′*S*),[7] in hexane exhibits two resolved ν(OH) absorption bands of the 1′-hydroxy group at 3629 and 3584 cm^{-1} with a shoulder at ~3550 cm^{-1} (Fig. 1, *b*).

The mathematical deconvolution of the spectral contour of the ν(OH) absorption band of shikalkin gave four components (see Fig. 1, *b*), which also indicate the presence of all four tautomers of compound **2b** (**A—D**) in a solution. The S_b/S ratio is 0.30 and remains unchanged for a solution of shikalkin in $CDCl_3$. A comparison of the S_b/S values obtained for compounds **2a** and **2b** shows that in a solution of **2a** the overall concentration of tautomers **2a(A)** + **2a(C)** is approximately halved compared to the corresponding overall concentration of tautomers **2b(A)** + **2b(C)**. Thus, the structure of the 1′-hydroxyalkyl substituent in compounds **2a** and **2b** has a strong effect on the tautomeric equilibrium. Perhaps, this is related to the presence (absence) of the double bond in their side chains. A more detailed assignment of the observed components of the ν(OH) band to particular types of tautomers and the determination of their quantitative composition are beyond the present study.

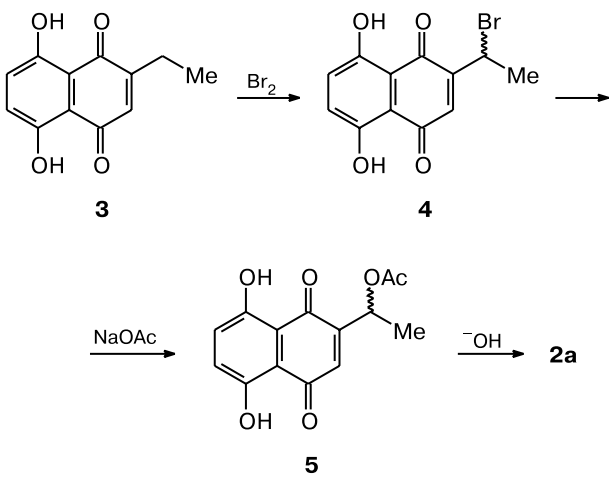
* The stretching vibrations of α-O—H in the compounds under study appear as a broad diffuse band at 3400—2200 cm^{-1}.

It should be noted that the observed tautomeric equilibrium does not contradict the modern quantum-chemical calculations. For example, the *ab initio* DTF calculations using the B3LYP functional in the extended 6-311G** basis set [8] show that the energy barrier for a process of the **2(A)**→**2(C)** type is lower than 5 kcal mol^{-1} in the case of naphthazarin, which allows the 1,5-naphthoquinonoid forms of the types **2(C)** and **2(D)** to exist in solutions at room temperature.

Experimental

Natural shikalkin[9] (**2b**)* was used in the present work. Compound **2a** was synthesized from ethylnaphthazarin (**3**)[1] through intermediates **4** and **5** (Scheme 3).

Scheme 3



IR spectra of compounds **2a,b** were recorded with a Bruker Vector 22 FT IR spectrometer with a resolution of 2.0 cm^{-1} in solutions of $CDCl_3$, CCl_4, and hexane using cells with CaF_2 windows and a layer thickness of 0.40—2.50 mm. The concentration of solutions of the compounds under study was 2—5 mmol L^{-1}. ^1H NMR spectra were recorded with a Bruker AC-250 spectrometer in $CDCl_3$ using Me_4Si as the internal standard. Mass spectra (EI) were obtained with an LKB-9000S instrument with direct injection at an energy of ionizing electrons of 70 eV. Silica gel L 80/160 μm (H^+) was used for column chromatography.[10] The reaction course was monitored using TLC (Merk 60F-254 plates, a hexane—acetone (3 : 1) mixture). Elemental analysis was carried out with a Flash EA1112 C,H,N-analyzer (Institute of Chemistry and Applied Ecology, Far-Eastern State University, Vladivostok).

2-(1-Bromoethyl)-5,8-dihydroxy-1,4-naphthoquinone (4). A solution of Br_2 (60 μL) in CCl_4 (10 mL) was added dropwise to a solution of ethylnaphthazarin (**3**) (218 mg, 1.0 mmol) in

* A shikalkin sample was kindly presented by S. A. Fedoreev and N. P. Mishchenko (Laboratory of Chemistry of Natural Compounds, Pacific Institute of Bioorganic Chemistry, FEB of the RAS).

CCl_4 (150 mL) with stirring at ~20 °C. After the end of the reaction, the mixture was concentrated. Product **4** was isolated in 86% yield (278 mg) by column chromatography (benzene), m.p. 125—127 °C. ^{1}H NMR, δ: 1.99 (d, 3 H, Me, J = 6.8 Hz); 5.47 (dq, 1 H, H(1′), J_1 = 6.8 Hz, J_2 = 0.7 Hz); 7.20 (s, 2 H, H(6), H(7)); 7.28 (d, 1 H, H(3), J = 0.7 Hz); 12.40 and 12.64 (both s, 1 H each, α-OH). MS, m/z (I_{rel} (%)): 296/298 $[M]^+$ (19), 217/219 $[M - Br]^+$ (100). Found (%): C, 48.33; H, 3.15. $C_{12}H_9O_4Br$. Calculated (%): C, 48.51; H, 3.05.

2-(1-Acetoxyethyl)-5,8-dihydroxy-1,4-naphthoquinone (5). A mixture of bromoethylnaphthazarin **4** (238 mg, 0.8 mmol) and NaOAc (330 mg, 4.0 mmol) in a HOAc—$CHCl_3$ (3 : 1, 12 mL) solution was refluxed for 2 h. The reaction mixture was concentrated, and a residue was diluted with H_2O (50 mL) and extracted with EtOAc (3×100 mL). The extract was dried over anhydrous Na_2SO_4 and concentrated. The residue was chromatographed (benzene—TFA, 100 : 1), and product **5** was obtained in 50% yield (110 mg), m.p. 155—157 °C. ^{1}H NMR, δ: 1.54 (d, 3 H, Me, J = 6.6 Hz); 2.15 (s, 3 H, OMe); 6.07 (dq, 1 H, H(1′), J_1 = 1.2 Hz, J_2 = 6.6 Hz); 7.05 (d, 1 H, H(3), J = 1.2 Hz); 7.06 and 7.18 (both s, 1 H each, H(6), H(7)); 12.47 and 12.56 (both s, 1 H each, α-OH). MS, m/z (I_{rel} (%)): 276 $[M]^+$ (12), 256 (21), 240 (16), 234 (15), 219 (18), 218 (30), 216 (46), 190 (17). Found (%): C, 60.55; H, 4.44. $C_{14}H_{12}O_6$. Calculated (%): C, 60.87; H, 4.38.

5,8-Dihydroxy-2-(1-hydroxyethyl)-1,4-naphthoquinone (2a). A suspension of acetoxyethylnaphthazarin **5** (110 mg, 0.4 mmol) in a TFA—H_2O (2 : 1, 5 mL) mixture was refluxed for 30 min. The reaction mixture was concentrated, and a residue was dissolved in H_2O (50 mL) and extracted with EtOAc (3×20 mL). The extract was dried over anhydrous Na_2SO_4 and concentrated. The residue was chromatographed (benzene—acetone, 50 : 1), and product **2a** was obtained in 58% yield (50 mg), m.p. 118—122 °C. IR ($CDCl_3$), ν/cm^{-1}: 3613, ~3560 (C(1′)—OH), 3050 (α-OH), 1657, 1610 (C=O), 1659 (C=C). ^{1}H NMR, δ: 1.55 (d, 3 H, Me, J = 7.4 Hz); 5.08 (dq, 1 H, H(1′), J_1 = 7.4 Hz, J_2 = 1.0 Hz); 7.16 (d, 1 H, H(3), J = 1.0 Hz); 7.20 (s, 1 H, H(6)); 7.12 (s, 1 H, H(7)); 12.47 and 12.59 (both s, 1 H each, α-OH). MS, m/z (I_{rel} (%)): 234 $[M]^+$ (100), 219 $[M - Me]^+$ (22), 216 $[M - H_2O]^+$ (76). Found (%): C, 61.40; H, 4.37. $C_{12}H_{10}O_5$. Calculated (%): C, 61.54; H, 4.30.

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