SHORT PAPER

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5-Hydroxyorotic acid and orotic acid 5-sulfate†

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The title compounds were synthesised by application of the Elbs oxidation to orotic acid

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Two groups headed by Mingos[1] and by Perlepes[2] have summarised the importance of the coordination chemistry of orotic acid (uracil 6-carboxylic acid, **1**) and its derivatives. Interest in this area includes the role of metal ions in the biosynthesis of orotic acid (the precursor of all biologically important pyrimidines), metal complexes of orotic acid for therapeutic purposes, and in materials chemistry because of orotic acid's exceptionally interesting acceptor-donor properties. Complexes of 5-amino-orotic acid have been studied. We now report the synthesis of two new 5-substituted orotic acid derivatives, 5-hydroxyorotic acid, **3,** and orotic acid 5-sulfate, **2**. A paper on some gallium chemistry of 5-hydroxyorotic acid has been published.[3] X-ray crystallographic analysis showed that the anion $[Ga(5\text{-hydroxyorotate})_2(DMF)_2]^-$ self- assembles to form a two-dimensional anionic network and also confirms the structure of **3.**

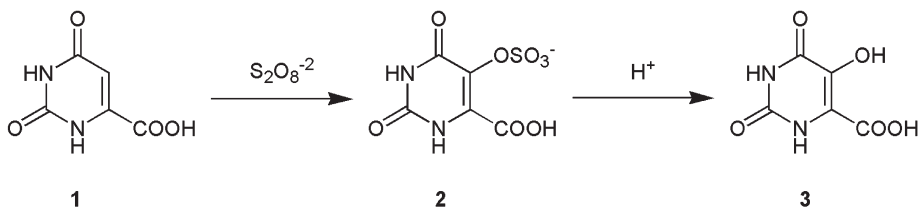
We have used the Elbs peroxydisulfate oxidation to carry out the transformation shown in Fig. 1. Hull[4] first showed the applicability of the Elbs oxidation to the pyrimidine series. The work through 1984 has been summarised.[5] Peroxydisulfate oxidations of pyrimidines not cited in ref. 5 are given in ref. 6.

Aside from the description of two new orotic acid derivatives, two points of synthetic interest emerged. When the reaction between orotic acid and peroxydisulfate ions was carried out under the usual conditions, that is in unstirred solution, the yield of sulfate ester was very poor. This was traced to the absence of oxygen; reasonable yields were found if the homogeneous reaction mixture was shaken continuously in air. Analysis of the products formed under anaerobic conditions showed the formation of urea in good yield. The rate of disappearance of peroxydisulfate under these conditions was about 10-times that in the presence of oxygen. Oxygen evidently inhibits a pathway leading to extensive oxidation of the pyrimidine ring. The effect of oxygen and the formation of urea recall the work of Sztumpf and Shugar[7] who described the inhibitory effect of oxygen on the uv-induced radical dimerisation of orotic acid and some analogs which they attributed to trapping of a triplet state (see also ref. 8). They also noted the instability in alkali of 5,6-dihydropyrimidines. Moschel and Behrman[9] found ring-cleavage and urea formation during the reaction of peroxydisulfate with thymine. The other feature of interest was the observation that 5-hydroxyorotic acid undergoes decarboxylation rather easily. When acid-catalysed hydrolysis of orotic acid 5-sulfate was carried out conventionally by refluxing in lM HCl, isobarbituric acid (5-hydroxyuracil) was the major product. Conversion of **2** into **3** was achieved by treatment of **2** with conc. sulfuric acid at room temperature followed by an ice quench. The relative ease of decarboxylation of 5-hydroxyorotic acid in contrast to orotic acid itself may have some importance in explaining the much debated catalytic proficiency of orotidine monophosphate decarboxylase.[10]

Experimental

Orotic acid 5-sulfate **2***:* Orotic acid monohydrate (Aldrich, 15.7 g, 0.09 mol), potassium peroxydisulfate (27g, 0.1 mol), and 85% KOH (26.5g, 0.4 mol) were dissolved in water (1l). The solution was divided into three equal portions and shaken at 35°C in three 2l Erlenmyer flasks for one week. The pH after that time was 10.5. Conc. HCl was added to the clear, colourless solution to bring the pH to 8.9 (pK_{a2} = 9.5). A crystalline precipitate of the potassium salt of orotic acid was removed by filtration after 12 h at 5°C, 5.8 g, 0.029 mol., 32%. The filtrate was then adjusted to pH 6 with additional conc. HCl. Fine needles of the dipotassium salt of the product **2** were collected after 12 h at 5°C (pK_{a3} = 8.7). The average yield for several preparations was 10g , 0.029 mol, 32% or, after correction for recovered orotic acid, 47%. These crystals darkened but did not melt when heated to 300°C. Anal.: Calcd. For $C_5H_2N_2O_8K_2S.H_2O$: C, 17.34; H, 1.16; N, 8.09; K, 22.58. Found: C, 17.09; H, 1.18; N, 8.26; K, 22.5. UV: λ_{max} 278 nm, ε 7300 $M^{-1}cm^{-1}$ (water). IR (Nujol): 3505, 3460, 1713, 1674, 1640, 1601, 1303, 1267, 1060, 845, 790, 755, 675 cm^{-1}. C, H, N analyses by Quantitative Technologies; potassium analysis by the tetraphenylboron method.

5-Hydroxyorotic acid **3***:* Orotic acid 5-sulfate (10 g, 0.029 mol), was finely powdered and then added slowly with stirring to conc. sulfuric acid (100 ml). The suspension was stirred briefly to dissolve the material. It was then allowed to stand for a few hours at RT. The clear solution was poured slowly onto 300 g of ice. A fine precipitate formed. This was filtered after standing overnight at RT. It was washed with cold water and then ethanol to yield 6 g, 0.029 mol, 100%, of the crude product. This material generally contained traces of orotic acid. Crystallisation was not successful in removing this. However, extraction of orotic acid under the following conditions yielded pure



1 **2** **3**

Figure 1

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† This is a Short Paper, there is therefore no corresponding material in *J Chem. Research (M).*

product: 10 g lots of the crude 5-hydroxyorotic acid were finely powdered and then stirred overnight (RT) with a 7:2:1 (v/v) mixture of isopropanol:water:conc. NH_4OH (500 ml). The undissolved material (5-hydroxyorotic acid) was filtered, washed with the same solvent mixture, and then ethanol. The process was repeated if tlc on silica (same solvent) showed the presence of orotic acid. This material was recrystallised by rapid dissolution in boiling 0.1 M HCl (4 g/l) and slow cooling. Anal.: Calcd. For $C_5H_4N_2O_5$. $2H_2O$: C, 28.9; H, 3.87; N, 13.5. Found: C, 29.1; H, 3.72; N, 13.5. UV: λ_{max} 308 nm, ε 7230 $M^{-1}cm^{-1}$ (0.05 M $NaHCO_3$). IR(Nujol): 3480, 3160, 1760, 1675, 1638, 1337, 1234, 965, 820, 760, 722 cm^{-1}. m.p. > 300°C.

5-Hydroxyorotic acid, ammonium salt: Alternatively, recrystallisation of crude 5-hydroxy orotic acid as the ammonium salt gives pure product. Crude 5-hydroxyorotic acid dihydrate (1g) was dissolved in boiling water (50 ml) containing 1 equiv. of conc. ammonia and filtered. The pH should be between 6 and 7. Fine needles formed upon cooling. These were washed with cold water and then acetone to yield the ammonium salt, 0. 8 g, 80%. Calcd. for $C_5H_7N_3O_5.H_2O$: C, 29.0; H, 4.38; N, 20.3. Found: C, 29.0; H, 4.16; N, 20.1. UV(water): λ_{max} 307 nm, ε 7300 $M^{-1}cm^{-1}$. IR(Nujol): 3550, 3370, 3190, 1716, 1667, 1635, 1310, 1250, 1044, 964, 819, 745, 720 cm^{-1}. m.p. >310°. R_f values, silica on Al: isopropanol: water: conc. ammonia- 7:2:1. Orotic acid 0.7, isobarbituric acid, 0.5, orotic acid 5-sulfate, 0.3, 5-hydroxyorotic acid, 0.0.

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