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The Structure of a Citric Anhydride Derivative, $C_8H_6O_7$*†

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Abstract. $M_r = 214{\cdot}13$, orthorhombic, $P2_12_12_1$, $a = 9{\cdot}495\ (2)$, $b = 16{\cdot}286\ (3)$, $c = 5{\cdot}841\ (1)$ Å, $V = 903{\cdot}2\ (3)$ Å^3, $Z = 4$, $D_x = 1{\cdot}575$ g cm^{-3}, Cu $K\alpha$, $\lambda = 1{\cdot}5418$ Å, $\mu = 11{\cdot}4$ cm^{-1}, $F(000) = 440$, $T = 293$ K, $R = 0{\cdot}036$ ($wR = 0{\cdot}053$) for 999 reflections. The structure is of interest because the compound is a derivative of citric anhydride and consists of two fused five-membered rings, one of which is an anhydride and the other a γ-lactone, with one carbon of the fusion bearing an acetoxy group. The molecule has bond lengths and angles comparable with those of other anhydrides. The packing is determined by interaction between carbonyl and ether oxygen atoms.

Introduction. Citric anhydride, derived from the α-hydroxycarboxylic acid citric acid, has been invoked in the reaction catalyzed by the enzyme citrate synthase so that the reversibility of the conversion of low-energy citrate into high-energy acetyl-CoA may be accounted for (Spector, 1972; Eggerer & Remberger, 1963; Eggerer, Remberger & Grünewälder, 1964; Wunderwald & Eggerer, 1969; Buckel & Eggerer, 1969; Buckel, 1976); however, detailed high-resolution X-ray crystallographic studies of citrate bound to the enzyme citrate synthase (Remington, Wiegand & Huber, 1982) suggest that there is not room in the active site for such an anhydride to be formed.

The existence of citric anhydride as a well-defined molecule has been reported by Repta (Repta, Robinson & Higuchi, 1966; Repta, 1969; Repta & Higuchi, 1969), who prepared solid citric anhydride. His preparation has been repeated, and the infrared spectrum of the solid was consistent with citric anhydride (Smart, 1984) but we have not been able to obtain crystals of the product (only very small amounts of crystalline anhydrous citric acid when the crystallization medium is not sufficiently dry). A report that citric acid exists in aqueous solution in equilibrium with its anhydride is really an extrapolation from the existence of anhydrides of acids such as succinic acid (Higuchi, Miki, Shah & Herd, 1963; Higuchi, McRae & Shah, 1966; Higuchi, Eberson & McRae, 1967) and no firm evidence for the existence of citric anhydride in aqueous solutions is available.

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† (2*S*,3*S*)-Tetrahydro-3-acetoxy-5-oxo-2,3-furandicarboxylic anhydride.

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It seems that citric acid does not form an anhydride readily. This is not unexpected because there are conformational preferences in citric acid or the citrate ion that prevent two carboxylic acid groups from coming close enough to each other for dehydration to occur. There are, of course, two types of anhydrides that have the potential for formation. One involves the central carboxyl group (with a hydroxyl group in the α-position) and a terminal carboxyl group, and the other involves two terminal carboxyl groups; the different sizes of the rings so generated may favor one over the other. In general, in citrates and in other molecules containing a similar α-hydroxycarboxylic acid grouping, the hydroxyl group lies approximately in the plane of the carboxyl group (Jeffrey & Parry, 1952; Nordman, Weldon & Patterson, 1960; Glusker, van der Helm, Love, Dornberg, Minkin, Johnson & Patterson, 1965; Johnson, 1965; Glusker, Minkin & Patterson, 1969; Gabe, Glusker, Minkin & Patterson, 1967; Roelofsen & Kanters, 1972; Glusker & Carrell, 1973; Carrell & Glusker, 1973; Sheldrick, 1974; Mastropaolo, Powers, Potenza & Schugar, 1976; Strouse, Layten & Strouse, 1977; Gavrushenko, Carrell, Stallings & Glusker, 1977; Swanson, Ilsley & Stanislowski, 1983). This means that both the hydroxyl group and the central carboxyl group are perpendicular to the main backbone of the molecule, as shown in Fig. 1. This has been ascribed (Newton & Jeffrey, 1977) to hydrogen-bond formation; in the case of metal salts it could be due to metal coordination. However, this observed coplanarity can no longer be maintained when an internal anhydride is formed and the two carboxyl groups need to approach each other in order for a chemical reaction to occur. This may, in part, explain the difficulty in preparing an anhydride.

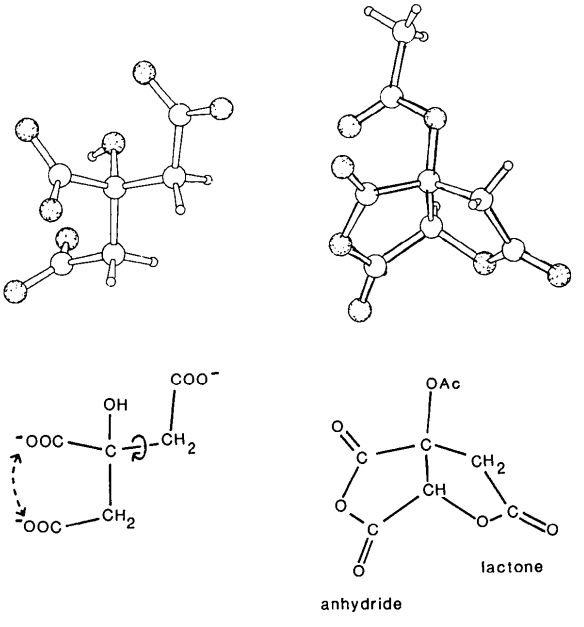
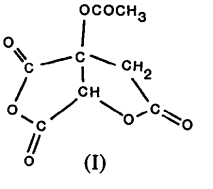


Fig. 1. The formation of citric anhydride (left) and the relationship of citric anhydride to the compound studied (right).

In this paper we describe the structure of a compound related to citric anhydride, and examine some of the reasons for constraints described above in the citric acid molecule or the citrate ion.

Experimental. (2*S*,3*S*)-Tetrahydro-3-acetoxy-5-oxo-2,3-furandicarboxylic anhydride, (I), prepared by one of us (RG) and crystallized as prisms. Crystal, approximate size 0·2 × 0·4 × 0·6 mm, sealed in Lindemann tube in dry argon. Four-circle automated diffractometer, θ–2θ scan, max. $2\theta = 138°$. 14 reflections (θ 15–29°) for measuring lattice parameters. 1005 reflections measured, 999 with $I > \sigma(I)$, $\sigma(I)$ derived from counting statistics. $\sigma(F) = (F/2)[\sigma^2(I)/(I)^2 + \delta^2]^{1/2}$, δ is measured instrumental uncertainty 0·017. Three standard reflections monitored at intervals during data collection showed no decay of intensities. No absorption correction. Data corrected for geometrical factors and put on an absolute scale with a Wilson plot. h 0–11, k 0–19, l 0–7. Structure solved with *MULTAN* (Germain, Main & Woolfson, 1971) on 240 E values ≥1·20. Refinement by full-matrix least-squares methods (Gantzel, Sparks, Long & Trueblood, 1969; Carrell, 1975), anisotropic temperature factors for C and O, isotropic for H which were located from a difference map after a few cycles of refinement of heavier atoms. $\sum w(||F_o|-|F_c||)^2$ minimized, $w = 1/\sigma^2(F)$. Atomic scattering factors from Cromer & Mann (1968) and Stewart, Davidson & Simpson (1965). Final $R = 0{\cdot}036$, $wR = 0{\cdot}053$. Final difference Fourier map contained no peak greater than 0·13 e Å^{-3}; max. $\Delta/\sigma = 0{\cdot}22$ for C and O, 0·05 for H. Atomic coordinates are listed in Table 1,* and some important torsion angles in Table 2.



(I)

Discussion. The structure of the molecule studied is shown in Fig. 2, which also shows bond lengths and angles. The molecule is both an anhydride and a γ-lactone and may be considered to be derived from a hydroxycitric acid. It provides partial structural proof of the existence of citric anhydride.

* Lists of structure factors, anisotropic thermal parameters and a view of the crystal packing have been deposited with the British Library Lending Division as Supplementary Publication No. SUP 39660 (10 pp.). Copies may be obtained through The Executive Secretary, International Union of Crystallography, 5 Abbey Square, Chester CH1 2HU, England.

Table 1. *Atomic parameters for (2S,3S)-tetrahydro-3-acetoxy-5-oxo-2,3-furandicarboxylic anhydride*

	x	*y*	*z*	⟨*B*⟩
O(1)	0·8319 (2)	0·5084 (1)	0·0241 (4)	6·39 (8)
O(2)	0·6388 (2)	0·5822 (1)	0·0706 (3)	4·28 (5)
O(3)	0·7685 (2)	0·7706 (1)	0·2129 (3)	3·57 (5)
O(4)	0·5548 (2)	0·6612 (1)	0·5877 (3)	4·25 (5)
O(5)	0·3895 (2)	0·5983 (1)	0·3837 (4)	5·75 (7)
O(6)	0·7454 (2)	0·7302 (1)	0·7056 (3)	5·86 (7)
O(7)	0·5653 (2)	0·8213 (1)	0·3350 (4)	5·96 (8)
C(1)	0·7764 (3)	0·5663 (1)	0·1082 (4)	4·21 (7)
C(2)	0·8414 (2)	0·6314 (1)	0·2562 (4)	4·28 (7)
C(3)	0·7250 (2)	0·6932 (1)	0·3006 (3)	3·10 (5)
C(4)	0·5954 (2)	0·6553 (1)	0·1876 (3)	3·46 (6)
C(5)	0·4992 (2)	0·6333 (1)	0·3867 (4)	3·92 (7)
C(6)	0·6823 (3)	0·6992 (1)	0·5532 (3)	3·73 (7)
C(7)	0·6732 (3)	0·8321 (1)	0·2352 (4)	4·22 (8)
C(8)	0·7229 (4)	0·9092 (1)	0·1260 (6)	5·85 (11)
H(1C2)	0·922 (5)	0·656 (3)	0·183 (9)	12·3 (4)
H(2C2)	0·892 (3)	0·616 (2)	0·380 (5)	4·5 (6)
H(4)	0·549 (3)	0·691 (2)	0·065 (6)	5·9 (7)
H(1C8)	0·811 (3)	0·920 (2)	0·153 (6)	5·5 (7)
H(2C8)	0·696 (4)	0·903 (2)	0·007 (9)	8·7 (9)
H(3C8)	0·683 (5)	0·954 (3)	0·19 (1)	10·2 (3)

Note. Positional parameters, *x*, *y*, *z*, are expressed as fractions of cell edges. Average equivalent isotropic temperature factors are in the form $\exp(-B\sin^2\theta/\lambda^2)$ with *B* values (Å²) calculated from $\langle B\rangle = \sum_{i=1,3} B_{ii}/3$. Atoms were refined anisotropically except for the hydrogen atoms. The numbers in parentheses after each parameter represent the e.s.d. with respect to the last digits quoted.

Table 2. *Torsion angles (e.s.d. 0·2–0·3°)*

(*a*) Anhydride ring		(*c*) Other	
C(5)–O(4)–C(6)–C(3)	−3·4	C(1)–O(2)–C(4)–C(5)	−109·2
O(4)–C(6)–C(3)–C(4)	6·6	C(4)–C(3)–O(3)–C(7)	61·8
C(6)–C(3)–C(4)–C(5)	−6·9	C(6)–C(3)–O(3)–C(7)	−55·9
C(3)–C(4)–C(5)–O(4)	5·5	C(1)–C(2)–C(3)–C(6)	115·8
C(4)–C(5)–O(4)–C(6)	−1·4	O(3)–C(3)–C(4)–O(2)	113·8
		O(3)–C(3)–C(6)–O(4)	131·7
(*b*) Lactone ring		O(3)–C(3)–C(6)–O(6)	−49·1
C(1)–C(2)–C(3)–C(4)	4·2	O(2)–C(4)–C(5)–O(5)	−59·8
C(2)–C(3)–C(4)–O(2)	−5·1	O(2)–C(4)–C(5)–O(4)	121·2
C(3)–C(4)–O(2)–C(1)	4·2		
C(4)–O(2)–C(1)–C(2)	−1·4		
O(2)–C(1)–C(2)–C(3)	−2·0		

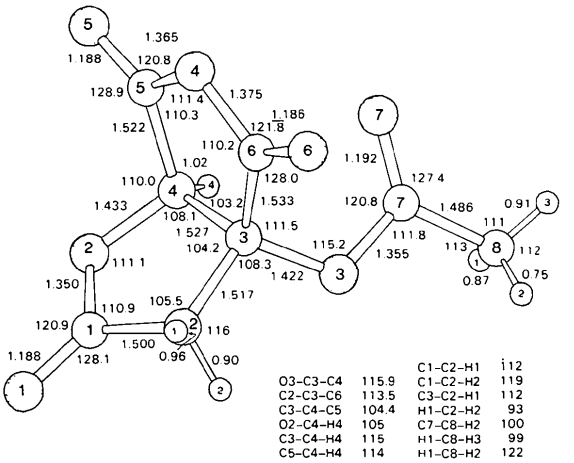


Fig. 2. A view of the molecule with distances and angles. E.s.d.'s are 0·002–0·003 Å for distances (0·03–0·06 Å if hydrogen is involved) and 0·1–0·2° for angles (2–5° if hydrogen is involved).

A review of the Cambridge Crystallographic Data File (Allen, Bellard, Brice, Cartwright, Doubleday, Higgs, Hummelink, Hummelink-Peters, Kennard, Motherwell, Rodgers & Watson, 1979; Murray-Rust, Stallings, Monti, Preston & Glusker, 1983) showed that of 37 anhydrides that have been reported in the literature only four have substituents other than carbon or hydrogen on C(3) and C(4), and no generalizations could be made from these. However, in 31 anhydrides it was shown that a change in the substituents on C(3) and C(4) from carbon to hydrogen resulted in a lengthening of the carbonyl group C=O bond and a shortening of C–C bond lengths, possibly because a hydrogen atom on C(3) or C(4) becomes labile as a result of the proximity of the carbonyl group. Data are given in Table 3 and show that in the structure here [with oxygen atoms attached to C(3) and C(4)] there is a lengthening of C(4)–C(5) and C(3)–C(6) and a shortening of C(5)–O(4) and C(6)–O(4).

The molecule shows no great evidence of strain when compared with other anhydrides such as a succinic anhydride derivative (Malmros & Wagner, 1978) or with a lactone of hydroxycitric acid (Glusker, Minkin & Casciato, 1971). The central carboxyl group [C(6), O(6), O(4)] cannot lie in the plane of the acetylated hydroxyl group [O(3), C(7), O(7), C(8)], as it would if it were a free citrate ion, because the central carboxyl group is part of the anhydride ring system and is coplanar with a terminal carboxyl group that forms the other half of the anhydride. Lactones of hydroxycitric acid are extremely stable and can be crystallized readily.

There are no hydrogen atoms attached to oxygen atoms in this molecule so that there is no potential for hydrogen-bond formation. The packing of molecules in the crystal structure is therefore determined by the packing of oxygen atoms, mainly carbonyl groups with their inherent dipoles interacting with ether oxygen atoms. The shortest packing distances involve the lactone carbonyl group O(1) interacting with furan (lactone and anhydride) oxygen atoms, O(2) and O(4), of another molecule at distances of 2·737 (2) and 2·981 (2) Å, respectively, and the anhydride carbonyl group O(6) interacting with O(3) (attached to the acetyl group) of another molecule at 2·652 (2) Å. The short O(1)···O(4) distance of 2·98 Å may result from the interaction of negatively charged carbonyl O(1) with a slightly positively charged O(4).

The hydrolysis of citryl-CoA *via* citric anhydride has been illustrated by Spector (1972) as

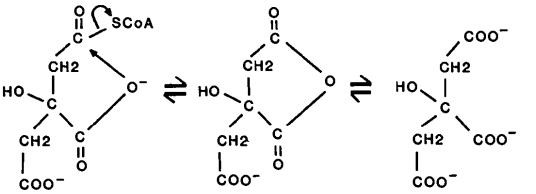


Table 3. *Bond lengths (in Å) in anhydrides*

Substituents at C(3) and C(4)	Lengths C(5)–O(5) and C(6)–O(6)	Lengths C(5)–O(4) and C(6)–O(4)	Lengths C(4)–C(5) and C(3)–C(6)	Length C(3)–C(4)
C, C, C, C*	1·183, 1·178	1·387, 1·384	1·511, 1·511	1·547
C, H, C, H†	1·189, 1·188	1·387, 1·389	1·497, 1·503	1·534
H, H, H, H‡	1·192, 1·189	1·384, 1·372	1·466, 1·484	1·512
This work				
C, O, H, O	1·188, 1·186	1·365, 1·375	1·522, 1·533	1·527

* 14 entries.
† 16 entries.
‡ 1 entry.

Our results would indicate that C–O cleavage in the anhydride on hydrolysis is more likely for the C(6)–O(4) than the C(5)–O(4) bond (*i.e.* the one nearest the substituted hydroxyl group) since the former bond is slightly longer and more twisted than the latter. Since carbonyl groups are polarized, with the negative end of the dipole moment on the oxygen atom, it would be expected that hydrolysis would result from nucleophilic substitution on the carbon atom (Berliner & Altschul, 1952). Another electron-withdrawing group would enhance this reactivity. Citric anhydride, with a hydrogen atom in place of the acetyl group (to give a hydroxyl group), would be expected to be much more reactive and this may be the reason that it is so hard to prepare.

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