

all lactones (Ruzic-Toros & Leban, 1978; Cheng, Koo, Mellor, Nyburg & Young, 1970; Svinning & Sorum, 1979; Berman, Carrell & Glusker, 1973). The cyclopentene ring makes an angle of 33.8 (4)° with the mean plane of the cyclohexane ring, and angles of 24.3 (5) and 92.7 (2)°, respectively, with the mean planes of the lactone rings I and II. The cyclohexane ring is fused to the lactone rings I and II making angles of 58.1 (2) and 58.9 (2)°, respectively.

Such bicyclic systems fused to each other impose considerable strain on the molecule (Sasaki & Hirata, 1973; Bucourt & Hainaut, 1965) and this contributes to the activity of the compound (Bowman & Rand, 1980). The stereo packing diagram down **b** is shown in Fig. 2. The molecules are stabilized by van der Waals forces.

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## Structure of Fluorescein Dipropionate Acetone Solvate

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**Abstract.** 3-Oxospiro[isobenzofuran-1(3H),9'-[9H]-xanthene]-3',6'-diyl dipropionate-acetone (2/1), C<sub>26</sub>H<sub>20</sub>O<sub>7.5</sub>, *M<sub>r</sub>* = 473.38, monoclinic, *P2<sub>1</sub>/c*, *a* = 14.917 (3), *b* = 11.964 (2), *c* = 13.633 (3) Å, β = 101.98 (5)°, *V* = 2380.0 Å<sup>3</sup>, *Z* = 4, *D<sub>x</sub>* = 1.321 g cm<sup>-3</sup>, λ(Cu Kα) = 1.5418 Å, μ = 7.93 cm<sup>-1</sup>, *T* = 298 K, *F*(000) = 992, *R* = 0.0556 for 2587 unique observed reflections. The title compound was prepared by esterification of fluorescein and on recrystallization from acetone it formed a crystalline inclusion compound with the solvent in the molar ratio 2:1.

**Introduction.** Fluorescein esters are highly sensitive agents for living biosubstances and much attention has been devoted to them recently. They can penetrate the cell membrane and can be hydrolyzed

- ### References
- BERMAN, H. M., CARRELL, H. L. & GLUSKER, J. P. (1973). *Acta Cryst.* **B29**, 1163–1165.  
 BIXON, M. & LIFSON, S. (1967). *Tetrahedron*, **23**, 769–784.  
 BOWMAN, W. C. & RAND, M. J. (1980). Editors. *Textbook of Pharmacology*, p. 38.7. Oxford: Blackwell Scientific Publications.  
 BUCOURT, R. & HAINAUT, D. (1965). *Bull. Soc. Chim. Fr.* pp. 1366–1378.  
 CHENG, P.-T., KOO, C. H., MELLOR, I. P., NYBURG, S. C. & YOUNG, J. M. (1970). *Acta Cryst.* **B26**, 1339–1348.  
 FRENZ, B. A. (1978). *The Enraf-Nonius CAD-4 SDP - A Real-Time System for Concurrent X-ray Data Collection and Crystal Structure Solution*. In *Computing in Crystallography*, edited by H. SCHENK, R. OLTJOF-HAZEKAMP, H. VAN KONINGSVELD & C. BASSI, pp. 64–71. Delft Univ. Press.  
 GANESH RAJ, S. P., JANARDHANAM, S. & RAJAGOPALAN, K. (1988). *Ind. J. Chem.* **27B**, 513–515.  
 MOTHERWELL, W. D. S. & CLEGG, W. (1978). *PLUTO*. Program for plotting molecular and crystal structures. Univ. of Cambridge, England.  
 RUZIC-TOROS, Z. & LEBAN, I. (1978). *Acta Cryst.* **B34**, 1226–1230.  
 SASAKI, K. & HIRATA, Y. (1973). *Acta Cryst.* **B29**, 547–553.  
 SVINNING, T. & SORUM, H. (1979). *Acta Cryst.* **B35**, 2967–2970.

by the lipolase therein (Rotman & Papermaster, 1966), producing the fluorescent substance fluorescein and giving bright yellow-green fluorescence under fluorescent microscopy. Because of the relationship between cell vitality and lipolase content, the living state of the cell may be determined by monitoring variations in fluorescent strength as the lipolase hydrolyzes fluorescein esters within the cell (Wang, Wang, Wang & Ni, 1984). In order to understand the mechanism of fluorescence in different esters, it was thought useful to study their molecular structures. In this paper we present the structure of fluorescein dipropionate.

**Experimental.** Fluorescein dipropionate (FDP), m.p. 439 K, was prepared from propionic acid anhydride and fluorescein; the results of elemental analysis

Table 1. Final positional and thermal parameters of non-H atoms with *e.s.d.*'s in parentheses

	x	y	z	B (Å <sup>2</sup> )
O(1A)	0.1099 (2)	0.6050 (3)	0.6015 (2)	4.81 (7)
O(1B)	0.7082 (2)	0.3617 (3)	0.6228 (2)	4.80 (7)
O(1)	0.3961 (2)	0.4368 (3)	0.6161 (2)	4.15 (6)
O(2)	0.3215 (2)	0.3152 (2)	0.3278 (2)	3.48 (6)
O(2B)	0.7025 (2)	0.2319 (3)	0.7385 (2)	6.49 (8)
O(2A)	0.1145 (2)	0.7405 (3)	0.4901 (2)	5.74 (8)
O(3)	0.2719 (2)	0.2122 (3)	0.1899 (2)	5.11 (7)
C(1)	0.3337 (3)	0.3031 (3)	0.4384 (2)	3.11 (8)
C(1B)	0.4334 (3)	0.3200 (3)	0.4848 (3)	3.14 (8)
C(1A)	0.2741 (3)	0.3889 (3)	0.4753 (3)	3.20 (8)
C(2B)	0.4586 (3)	0.3827 (3)	0.5722 (3)	3.34 (8)
C(2)	0.3005 (3)	0.1857 (3)	0.4487 (3)	3.12 (8)
C(2A)	0.3082 (3)	0.4471 (3)	0.5636 (3)	3.38 (8)
C(3A)	0.2535 (3)	0.5205 (4)	0.6026 (3)	3.89 (9)
C(3)	0.2931 (3)	0.1263 (4)	0.5342 (3)	4.03 (9)
C(3B)	0.5490 (3)	0.3947 (4)	0.6195 (3)	3.81 (9)
C(4B)	0.6152 (3)	0.3449 (4)	0.5789 (3)	3.92 (9)
C(4)	0.2598 (3)	0.0187 (4)	0.5212 (3)	5.0 (1)
C(4A)	0.1656 (3)	0.5362 (3)	0.5545 (3)	3.97 (9)
C(5A)	0.1284 (3)	0.4809 (4)	0.4652 (3)	4.3 (1)
C(5)	0.2337 (3)	-0.0298 (4)	0.4279 (4)	5.2 (1)
C(5B)	0.5933 (3)	0.2843 (4)	0.4906 (3)	4.4 (1)
C(6)	0.2403 (3)	0.0292 (4)	0.3428 (3)	4.6 (1)
C(6B)	0.5031 (3)	0.2717 (4)	0.4452 (3)	3.97 (9)
C(6A)	0.1839 (3)	0.4078 (4)	0.4280 (3)	3.96 (9)
C(7)	0.2740 (3)	0.1368 (3)	0.3553 (3)	3.46 (9)
C(7B)	0.7461 (3)	0.2989 (4)	0.7040 (3)	4.05 (9)
C(7A)	0.0910 (3)	0.7074 (4)	0.5634 (3)	4.2 (1)
C(8)	0.2876 (3)	0.2188 (4)	0.2803 (3)	3.79 (9)
C(8A)	0.0383 (3)	0.7739 (4)	0.6258 (4)	5.9 (1)
C(8B)	0.8439 (3)	0.3264 (3)	0.7193 (3)	4.9 (1)
C(9B)	0.8915 (4)	0.2566 (6)	0.8237 (4)	8.3 (2)
C(9A)	0.0160 (5)	0.8854 (6)	0.5977 (5)	11.9 (2)
O(C)*	0.5000	0.0970 (5)	0.250	17.0 (3)
C(1C)*	0.500	0.000	0.250	8.4 (3)
C(2C)*	0.5200 (5)	-0.0592 (6)	0.1640 (7)	12.5 (3)

\* Atoms of the acetone molecule.

agree with calculations. The dried fine powder of FDP was dissolved in acetone, and the solution was placed in a shady and cool place. After the solvent evaporated, small cubic crystals formed. A CAD-4K single-crystal diffractometer was used for this study. Crystal dimensions 0.20 × 0.20 × 0.35 mm,  $\theta$ - $2\theta$  scan, cell parameters from setting angles of 25 reflections ( $12 \leq \theta \leq 14^\circ$ );  $2\theta_{\max} = 120^\circ$ ,  $-15 \leq h \leq 15$ ,  $0 \leq k \leq 13$ ,  $0 \leq l \leq 16$ ; 15 standard reflections monitored, 1.5% variation. The total number of independent reflections was 3689, 2587 (70% of the total) of which had  $I > 3\sigma(I)$ . The space group was assumed to be  $P2_1/c$ . The structural solution was carried out on a PDD 12/14 computer. The data were corrected for  $L_p$ , time attenuation and empirical absorption (max. 0.98, min. 0.90), and weighted with Wilson statistics.  $B$  and  $K$  values were chosen so that the diffraction intensities were reduced to  $|F_o|$ 's on an absolute scale, and were used in the structure refinement.

*MULTAN82* (Main, Fiske, Hull, Lessinger, Germain, Declercq & Woolfson, 1982) was used to obtain the coordinates of 36 non-H atoms of the asymmetric unit by direct methods. The coordinates of the H atoms on the benzene ring and on the methylene group were determined by difference Fourier map, and those of the methyl-group H atoms were obtained by theoretical calculation. Finally, the

Table 2. Bond lengths (Å), bond angles (°) and torsion angles (°)

C(1A)—C(4A)	1.414 (4)	C(2)—C(7)	1.382 (4)
C(1A)—C(7A)	1.338 (8)	C(2A)—C(3A)	1.378 (5)
O(1B)—C(4B)	1.407 (4)	C(3A)—C(4A)	1.353 (5)
O(1B)—C(7B)	1.361 (4)	C(3)—C(4)	1.379 (6)
O(1)—C(2B)	1.373 (4)	C(3B)—C(4B)	1.366 (5)
O(1)—C(2A)	1.363 (4)	O(C)—C(1C)	1.160 (6)
O(2)—C(1)	1.488 (3)	C(4A)—C(5A)	1.395 (5)
O(2)—C(8)	1.367 (4)	C(5A)—C(6A)	1.371 (5)
O(2B)—C(7B)	1.188 (4)	C(5)—C(6)	1.379 (6)
O(2A)—C(7A)	1.193 (4)	C(5B)—C(6B)	1.368 (5)
O(3)—C(8)	1.209 (4)	C(6)—C(7)	1.379 (5)
C(1)—C(1B)	1.505 (5)	C(7)—C(8)	1.462 (5)
C(1)—C(1A)	1.510 (5)	C(7B)—C(8B)	1.475 (6)
C(1)—C(2)	1.505 (5)	C(7A)—C(8A)	1.501 (6)
C(1B)—C(2B)	1.393 (4)	C(8A)—C(9A)	1.410 (9)
C(1B)—C(6B)	1.392 (5)	C(8B)—C(9B)	1.481 (7)
C(1A)—C(2A)	1.392 (4)	C(4B)—C(5B)	1.384 (5)
C(1A)—C(6A)	1.385 (5)	C(4)—C(5)	1.379 (6)
C(2B)—C(3B)	1.375 (5)	C(1C)—C(2C)	1.453 (9)
C(2)—C(3)	1.389 (5)		
C(4A)—O(1A)—C(7A)	116.9 (3)	C(4B)—O(1B)—C(7B)	118.3 (3)
O(1A)—C(4A)—C(3A)	117.6 (3)	O(1B)—C(4B)—C(3B)	119.8 (3)
C(2A)—C(3A)—C(4A)	119.6 (4)	C(2B)—C(3B)—C(4B)	119.0 (3)
C(1A)—C(2A)—C(3A)	120.8 (3)	C(1B)—C(2B)—C(3B)	121.4 (3)
C(2A)—C(1A)—C(6A)	117.8 (3)	C(2B)—C(1B)—C(6B)	117.7 (3)
C(1A)—C(6A)—C(5A)	122.4 (4)	C(1B)—C(6B)—C(5B)	121.5 (4)
C(4A)—C(5A)—C(6A)	117.6 (4)	C(4B)—C(5B)—C(6B)	118.9 (4)
C(3A)—C(4A)—C(5A)	121.8 (4)	C(3B)—C(4B)—C(5B)	121.4 (4)
O(1A)—C(4A)—C(5A)	120.5 (4)	O(1B)—C(4B)—C(5B)	118.6 (3)
O(1A)—C(7A)—O(2A)	123.7 (4)	O(1B)—C(7B)—O(2B)	121.7 (4)
O(1A)—C(7A)—C(8A)	110.9 (4)	O(1B)—C(7B)—C(8B)	110.9 (4)
O(2A)—C(7A)—C(8A)	125.3 (4)	O(2B)—C(7B)—C(8B)	127.4 (4)
C(7A)—C(8A)—C(9A)	118.0 (5)	C(7B)—C(8B)—C(9B)	114.1 (5)
C(1)—C(1A)—C(2A)	119.5 (3)	C(1)—C(1B)—C(2B)	120.0 (3)
C(1)—C(1A)—C(6A)	122.6 (3)	C(1)—C(1B)—C(6B)	122.3 (3)
O(1)—C(2A)—C(1A)	123.4 (3)	O(1)—C(2B)—C(1B)	122.8 (3)
C(1A)—C(1)—C(1B)	111.6 (3)	C(2A)—O(1)—C(2B)	118.5 (3)
C(1A)—C(1)—O(2)	108.3 (3)	C(1B)—C(1)—O(2)	108.3 (3)
C(1A)—C(1)—C(2)	112.1 (3)	C(1B)—C(1)—C(2)	113.7 (3)
O(2)—C(1)—C(2)	102.2 (2)	O(2)—C(8)—C(7)	109.0 (3)
O(3)—C(8)—O(2)	120.8 (3)	O(3)—C(8)—C(7)	130.1 (4)
C(1)—C(2)—C(7)	110.3 (3)	C(2)—C(7)—C(8)	107.8 (3)
C(1)—C(2)—C(3)	129.8 (3)	C(6)—C(7)—C(8)	129.8 (3)
C(3)—C(2)—C(7)	119.9 (3)	C(2)—C(7)—C(6)	122.5 (3)
C(2)—C(3)—C(4)	117.3 (4)	C(3)—C(4)—C(5)	122.5 (4)
C(4)—C(5)—C(6)	120.3 (4)	C(5)—C(6)—C(7)	117.5 (4)
O(1)—C(2A)—C(3A)	115.8 (3)	O(1)—C(2B)—C(3B)	115.8 (3)
O(C)*—C(1C)*—C(2C)*	119.2 (4)	C(2C)*—C(1C)*—C(2C)*	121.6 (8)
C(1)—O(2)—C(8)	110.7 (3)		
C(7A)—O(1A)—C(4A)—C(3A)	105.5	C(7B)—O(1B)—C(4B)—C(3B)	81.6
C(7A)—O(1A)—C(4A)—C(5A)	-78.9	C(7B)—O(1B)—C(4B)—C(5B)	-103.0
C(4A)—O(1A)—C(7A)—O(2A)	3.9	C(4B)—O(1B)—C(7B)—O(2B)	-0.7
C(4A)—O(1A)—C(7A)—C(8A)	-175.2	C(4B)—O(1B)—C(7B)—C(8B)	179.1

\* Atoms of the acetone molecule.

coordinates were corrected by the block-diagonal least-squares method; 417 parameters refined; the refinement (on  $F$ ) finally converged to  $R = 0.0556$  and  $wR = 0.0533$ ;  $(\Delta/\sigma)_{\max} = 0.003$ ,  $(\Delta\rho)_{\max} = 0.20$ ,  $(\Delta\rho)_{\min} = -0.10 \text{ e } \text{Å}^{-3}$ .

**Discussion.** The coordinates of non-H atoms are listed in Table 1,\* and bond lengths, bond angles and torsion angles in Table 2. An *ORTEP* plot of the title compound with the atomic numbering scheme is

\* Lists of anisotropic thermal parameters and least-squares-planes data have been deposited with the British Library Document Supply Centre as Supplementary Publication No. SUP 52768 (3 pp.). Copies may be obtained through The Technical Editor, International Union of Crystallography, 5 Abbey Square, Chester CH1 2HU, England.

shown in Fig. 1, and the packing of the molecules in a unit cell is shown in Fig. 2.

The results of the structure determination show that the molecular skeleton is very close to that of dichlorofluoran [630-88-6] (Wang, Ren, Wang, He, Wang, Jin & Zhang, 1989). The mean C—C bond length in the benzene ring is 1.380 Å, characteristic of a typical conjugated system. O—C bond lengths show greater variation. Bond lengths O(2A)—C(7A), O(2B)—C(7B) and that of the carboxy carbonyl O(3)—C(8) are 1.209, 1.193 and 1.185 Å, respectively, characteristic of a C=O double bond, but those of the carboxy ester C—O, *i.e.* O(1A)—C(4A) and O(1B)—C(4B) are 1.414 and 1.407 Å, both characteristic of a C—O single bond. It is thus evident that O(1A) and O(1B) do not take part in conjugation with the benzene ring, and this is in agreement with the results of a  $^{13}C$  NMR study. The bond lengths between the carbonyl carbon and ester oxygen, O(1A)—C(7A) and O(1B)—C(7B), are 1.338 and 1.361 Å, respectively, and both lie between the bond lengths of single C—O and double C=O bonds; this is the result of the O—C(R)—O conjugation effect. The C(1)—O(2) bond length is 1.488 Å

which is about 0.06 Å longer than normal C—O bonds (1.43 Å), and this can be ascribed to steric hindrance and strain of the five-membered ring. The three C—C bond lengths of the asymmetric atom C(1) are 1.507 (2) Å, 0.03 Å shorter than the normal C—C single bond length (1.54 Å). Using  $E_a = k(\Delta d)^2$ , where  $E_a$  is the difference in bond energies,  $\Delta d$  is the difference in bond lengths in Å and  $k$  is a constant ( $1470 \text{ kJ mol}^{-1} \text{ \AA}^{-2}$ ), the increase in bond energy is  $1.3 \text{ kJ mol}^{-1}$ . All C—H bond lengths are within the reasonable range 0.83–1.09 Å.

The angle between the two benzene rings (planes 1 and 2) of the xanthene moiety is  $14.4^\circ$ , which is larger than that in the 3,6-dichlorofluoran molecule (Wang *et al.*, 1989). The five-membered lactone ring of the isobenzofuran moiety is also planar (plane 4), and the angle between this plane and the adjacent benzene ring (plane 3) is only  $0.8^\circ$ , so they may be considered as coplanar. The two rings of isobenzofuran are approximately perpendicular to the xanthene moiety; this is the inevitable outcome of the bonding of C(1) through  $sp^3$  hybridization. The skeleton of the whole molecule is T-shaped, but the plane angles are a little different from those of both 3,6-dichlorofluoran (Wang *et al.*, 1989) and 2,5,7-trichlorospiro[benzofuran-3(2H),9-[9H]-xanthene]-2-one (Kimura, 1985).

The rotation of the propionate C—O bond in the single crystal is frozen so that the whole molecule exists in a stable conformation. From the torsion angles in Table 2, the distances of C(7A) and O(2A) to plane 1 (+0.998 and 2.207 Å, respectively) and the distances of C(7B) and O(2B) to plane 2 (−1.081 and −2.119 Å, respectively), it can be seen (Fig. 1) that C(7A) and O(2B) protrude upwards above plane 1 and C(7B) and O(2B) protrude downwards below plane 2, and so the whole molecule is asymmetric.

Also, the results show that each unit cell contains two molecules of acetone. The carbonyl oxygen O(C) and carbonyl carbon C(1C) of the acetone molecule are situated on the secondary axis of the cell with the methyl carbon C(2C) of the acetone molecule on the ordinate position.

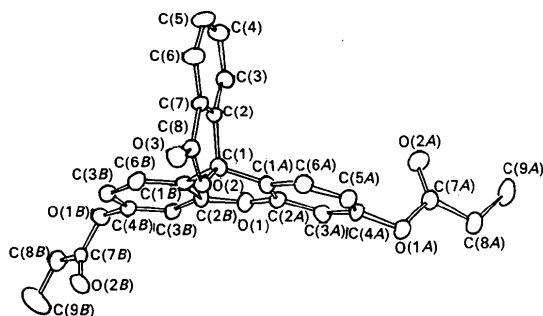


Fig. 1. ORTEP (Johnson, 1965) drawing of the structure.

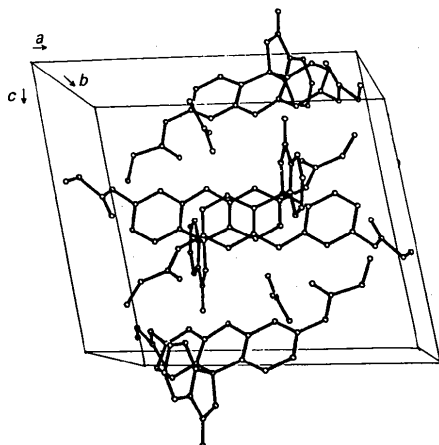


Fig. 2. The unit cell.

## References

- JOHNSON, C. K. (1965). ORTEP. Report ORNL-3794. Oak Ridge National Laboratory, Tennessee, USA.
- MAIN, P., FISKE, S. J., HULL, S. E., LESSINGER, L., GERMAIN, G., DECLERCQ, J.-P. & WOOLFSON, M. M. (1982) MULTAN82. *A System of Computer Programs for the Automatic Solution of Crystal Structures from X-ray Diffraction Data*. Univs. of York, England, and Louvain, Belgium.
- KIMURA, M. (1985). *Bull. Chem. Soc. Jpn.*, **58**, 909–910.
- ROTMAN, B. & PAPERMASTER, B. W. (1966). *Proc. Natl Acad. Sci. USA*, **55**(1), 134–141.
- WANG, L., REN, Y., WANG, Q., HE, F., WANG, Q., JIN, D. & ZHANG, P. (1989). *Chin. Sci. Bull.* **34**(21), 1789.
- WANG, Y., WANG, L., WANG, X. & NI, J. (1984). *J. Lanzhou Univ. Nat. Sci.* **20**(3), 132.