Structure of K-76, a Complement Inhibitor produced by *Stachybotrys complementi nov. sp.* K-76

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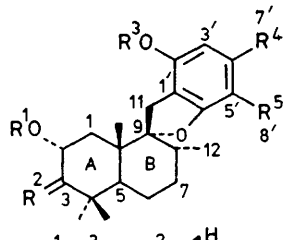
Summary The structure of 'K-76' (**1**), a complement inhibitor containing a spirobenzofuran unit, obtained from *Stachybotrys complementi nov. sp.* K-76, is reported.

THERE are several reports that the complement system participates in rheumatoid arthritis,[1] glomerulonephritis,[2] and other immune-complex diseases.[3] Therefore, we examined the abilities of various microbial products to inhibit this system, and found that the culture filtrate of *Stachybotrys complementi nov. sp.* K-76 strongly inhibited the complement system. We isolated the active compound (**1**) and named it 'K-76'.[4]



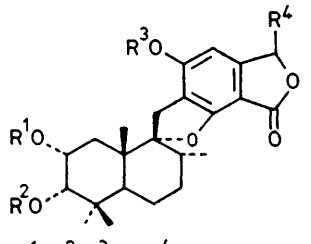
(1) $R^1=R^3=H$, $R^2=$  , $R^4=R^5=CHO$

(2) $R^1=R^3=H$, $R^2=$  , $R^4=R^5=CH_2OH$

(3) $R^1=R^3=Ac$, $R^2=$  , $R^4=R^5=CH_2OAc$

(4) $R^1=R^3=Ac$, $R^2=O$, $R^4=R^5=CH_2OAc$

(5) $R^1=H$, $R^2=$  , $R^3=SO_2C_6H_4Br$, $R^4=R^5=CH_2OH$



(7) $R^1=R^2=R^3=H$, $R^4=OH$

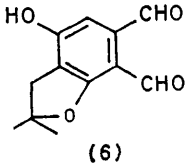
(8) $R^1=R^2=R^3=R^4=H$

(9) $R^1=R^2=R^4=H$, $R^3=Me$

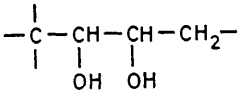
(10) $R^1=R^2=$  , $R^3=R^4=H$

(11) $R^1=COC_6H_4OMe$, $R^2=R^4=H$, $R^3=Me$

(12) $R^1=R^2=COC_6H_4OMe$, $R^3=Me$, $R^4=H$



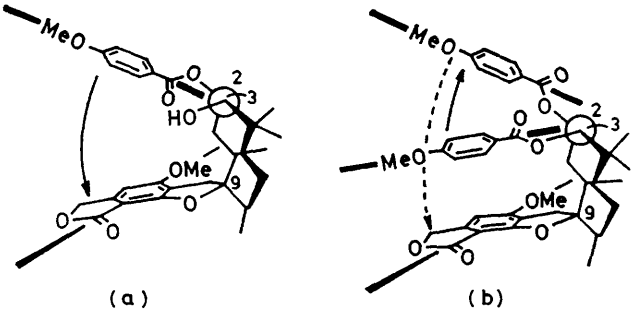
(6)



(13)

'K-76', $C_{23}H_{30}O_6$, m.p. 176 °C (decomp.), showed $[\alpha]_D^{20}$ −48° (MeOH); λ_{max} (EtOH) 246 (ε 16,500), 307 (6700), and 359 (5400) nm; λ_{max} (KBr) 1660, 1585, 1385, and 880 cm^{-1}; ^1H n.m.r. (C_6D_5N) δ 0·82 (3H, d, *J* 5 Hz, 12-H), 0·93, 1·04, and 1·27 (each 3H, s), 3·72 (1H, d, *J* 2 Hz, 3-H), 4·30 (1H, m, $W_{1/2}$ 20·4 Hz, 2-H), 7·41 (1H, s, 3'-H), and 10·75, and 11·02 (each 1H, s, -C*H*O). These data suggest that (**1**) is a 'triprenyl phenol'[5] containing two aromatic aldehyde groups. Mild oxidation of (**1**) with silver oxide afforded the lactol (**7**) [δ(CD_3OD) 6·45 and 6·53, 1H, 7'-H], which was reduced with $NaBH_4$ and then treated with acid to give the lactone (**8**), ν_{max} (CCl_4) 1755 cm^{-1}, δ[$(CD_3)_2CO$] 5·12, 2H, s, 7'-H. Compound (**8**) was treated with diazomethane to yield the monomethyl ether (**9**) [δ(CD_3OD) 3·83, 3H, s]. The above evidence, the u.v. spectrum of (**8**) [λ_{max} (EtOH) 268 nm],[6] the ^{13}C n.m.r. spectrum of the reduction product (**2**) δ[$(CD_3)_2SO$] 110·3(C-1'), 152(C-2'), 106·7(C-3'), 141·8-(C-4'), 110·8(C-5'), 160·3(C-6'), and 95·0(C-9) p.p.m. and the ^1H n.m.r. spectrum [δ(C_6D_5N) 2·82 and 3·20 (each 1H, d, *J* 16·6 Hz, 11-H)] of (**1**) show that (**1**) has the partial structure (**6**). The lactone (**8**) was treated with 2,2-dimethoxypropane and *p*-$MeC_6H_4SO_3H$ in acetone affording the acetonide (**10**). Acetylation of (**2**) with acetic anhydride and pyridine gave the tetra-acetate (**3**) as the major product, which was oxidized by Jones reagent to yield the monoketone (**4**) [ν_{max} (KBr) 1730 and 1750 cm^{-1}]. These data indicate that K-76 has the partial structure (**13**).

K-76 has three tertiary methyl groups, one secondary methyl group and the partial structure (**13**) in its sesquiterpene unit which suggests that the sesquiterpene unit is a drimane type skeleton. There are two hydroxy groups at C-2 and C-1 or C-3. Solvent shifts [δ($CDCl_3$)−δ(C_6D_6)][7] of the tertiary methyl groups of (**4**) (0, +0·48, +0·24 p.p.m.) indicated that one hydroxy group is attached at C-3. The signal of the C-7' methylene of (**3**) was a singlet at δ 5·18 and that of C-8' was an AB quartet at δ 4·84 and 5·62 (*J* 12·5 Hz), although that of (**4**) was a singlet at δ 5·17. This indicates that the C-3 hydroxy group of (**3**) was hydrogen bonded to the C-8' acetoxy group. In confirmation of this relationship, the A,B ring juncture is *trans* and the C–O bond at C-9 is axial. Thus we conclude that K-76 has structure (**1**).



(a) (b)

FIGURE. Chirality models of the monobenzoate (**11**) (a) and the dibenzoate (**12**) (b).

This structure was confirmed by *X*-ray analysis of the *p*-bromobenzenesulphonate (**5**):† $C_{29}H_{37}BrSO_8$, m.p. 204—207 °C, monoclinic, space group $P2_1$, $a = 15{\cdot}150$, $b = 10{\cdot}300$, $c = 10{\cdot}867$ Å, $\beta = 103{\cdot}15°$, $Z = 2$. 2222 independent reflections were collected on a Hilger-Watts four circle diffractometer. The structure was solved by the direct method using the MULTAN program, and refined to an *R* value of 0·106 by the block-diagonal least-squares technique with anisotropic thermal parameters. The molecule has structure (**5**).

The absolute configuration was established by the dibenzoate chirality method,[8] because we could not find a suitable crystal to determine the absolute structure by the *X*-ray method. The c.d. spectra of the monobenzoate (**11**) {$[\theta]_{271\text{ nm}}$ −63,430, $[\theta]_{251\text{ nm}}$ +35,240 and the dibenzoate (**12**) $[\theta]_{261\text{ nm}} = +144{,}470$, $[\theta]_{242\text{ nm}} = -24{,}240$} indicated that the absolute configuration of K-76 is as shown in (**1**) (Figure).

K-76 (**1**) and the sodium salt of the oxidation product (**7**) strongly inhibited the complement system *in vitro*[9] and preliminary experiments indicate that these substances improve the symptoms of experimental glomerulonephritis.

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† The atomic co-ordinates for this work are available on request from the Director of the Cambridge Crystallographic Data Centre, University Chemical Laboratory, Lensfield Road, Cambridge CB2 1EW. Any request should be accompanied by the full literature citation for this communication.

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