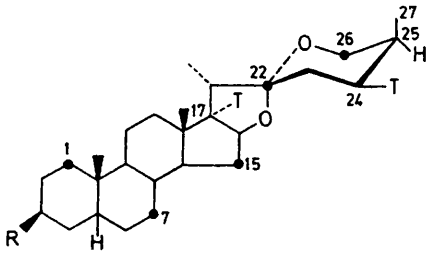
Biogenesis of Spirostan Sapogenins: Origin of the 26-Carbon in Sarsasapogenin

By ANTONIO G. GONZÁLEZ,* COSME G. FRANCISCO, RAIMUNDO FREIRE, ROSENDO HERNÁNDEZ, JOSÉ A. SALAZAR, and ERNESTO SUÁREZ

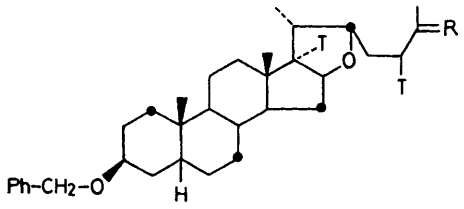
(*Department of Organic and Biochemistry, University of La Laguna, Instituto de Investigaciones Químicas, C.S.I.C., Tenerife, Spain*)

Summary It is shown that the 26-carbon in sarsasapogenin biosynthesized in *Agave attenuata* Solm. is derived from C-2 of mevalonic acid.

IT has been suggested that the determinant step in the formation of the side chain of 25*R*- ('iso') and 25*S*- ('neo') spirostan sapogenins is the biogenetic oxidation of the *cis*



(1) R = OH
(2) R = O – CH_2– Ph



(3) R = H, $\dot{C}H_2OH$
(4) R = H, $\dot{C}H_2OTs$
(5) R = H, $\dot{C}H_2I$
(6) R = $=\dot{C}H_2$
(7) R = H, ~OH
• = 2 –^{14}C of MVA

or *trans* methyl group of the isopropylidene function in the $\Delta^{2,4}$-precursor.[1] On the other hand, it has been shown that in a *Dioscorea*[2] there is no *in vivo* conversion of diosgenin (25*R*) into yamogenin (25*S*) and that in a *Digitalis*[3] the C-27 of tigogenin (25*R*) comes from C-2 of MVA. We show here that the 26-carbon in sarsasapogenin (**1**), a 25*S*-spirostan sapogenin, is derived from C-2 of MVA.

[2-^{14}C,(4*R*)-4-^{3}H]MVA was administered to a culture of *Agave attenuata* Solm. After 3 days, extraction of the sapogenins gave sarsasapogenin (**1**; incorporation 0·9%) labelled with ^{3}H in positions 17 and 24, and with ^{14}C in 1, 7, 15, 22, and 26 (or 27), the atomic ratio ^{3}H:^{14}C being 2·02:5. It was diluted with carrier material and after formation of the corresponding benzyl ether (**2**), it was treated with $LiAlH_4$–$AlCl_3$ to give the dihydro-derivative (**3**). This was tosylated and the resulting compound (**4**) transformed into the iodide (**5**). Elimination of the iodine with aqueous KOH at reflux yielded the methylene compound (**6**) (121 d.p.m. of ^{14}C/mg) whose ^{3}H:^{14}C ratio, 1·98:5, was unchanged. Treatment of compound (**6**) with ozone followed by reduction with $LiAlH_4$ gave the alcohol (**7**) [99 d.p.m. of ^{14}C/mg; m.p. 148—151° (n-hexane), ν_{max} ($CHCl_3$) 3380 cm^{-1}, τ ($CDCl_3$) 8·79 (d, J 6·5 Hz, 25-Me)], which after recrystallization to constant specific radioactivity exhibited a ^{3}H:^{14}C ratio of 2:4·04. This confirms that in sarsasapogenin (**1**) the C-26 was labelled. Hence, we may deduce that in the formation of 25*S*- and 25*R*-spirostan sapogenins the oxidation of the isopropylidene function of the Δ^{24}-precursor takes place at a different methyl group. Moreover, our result excludes the possibility that the differentiation between 25*R*- and 25*S*-spirostan sapogenins occurs by a different attack of the incoming hydrogen at C-25 during the biological hydrogenation of the Δ^{24}-precursor.[4]

One of us (R.H.) thanks the C.S.I.C. for a postdoctoral fellowship. This work was performed within the Programme of Chemistry 1971 conceded by the Foundation Juan March.

(*Received, 5th August* 1974; *Com.* 1000.)

[1] K. Takeda, *Progr. Phytochemistry*, 1972, **3**, 318.
[2] R. D. Bennett, E. Heftmann, and R. A. Joly, *Phytochemistry*, 1970, **9**, 349.
[3] R. Joly and Ch. Tamm, *Tetrahedron Letters*, 1967, 3535.
[4] L. Canonica, F. Ronchetti, and G. Russo, *J.C.S. Chem. Comm.*, 1972, 1309.