Stereochemistry of Culmorin Biosynthesis

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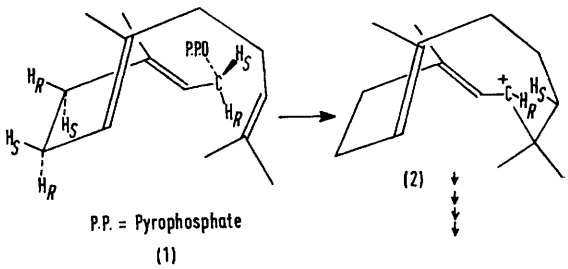
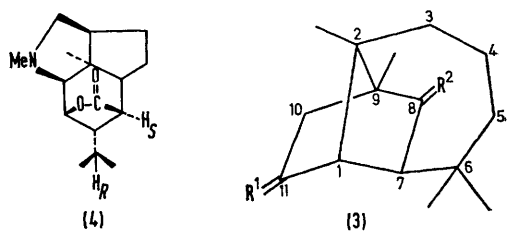
Summary The labelling pattern in the sesquiterpenoid culmorin biosynthesised from [2(*S*)-2-^{3}H,2-^{14}C]- and [5(*R*)-5-^{3}H,2-^{14}C]-mevalonate shows that the *endo*-C-10 proton originates from the 2(*S*) position whilst the C-11 hydroxy-group has displaced a *pro*-5(*R*) proton; in contrast to dendrobine an eleven-membered ring is implicated, and a *pro*-5(*S*)-mevalonoid proton migrates at an early stage in the biosynthesis.

SOME recent reports[1,2] on the biosynthesis of those sesquiterpenoids in which the primary cyclization of farnesyl pyrophosphate is between the terminal pyrophosphate and the distal double bond, have described a hydrogen migration. The biosynthetic function of these rearrangements is to transfer a carbonium ion from the C-10 or C-11 (the distal end) of farnesyl pyrophosphate back to C-1 to initiate further isomerization and secondary cyclizations of the 10- or 11-membered ring. We have studied the stereochemistry of *pro*-2(*S*)- and *pro*-5(*R*)-mevalonoid labelling of culmorin (**3**; $R^1 = R^2 = \beta$-H, α-OH)[3] in this context.

The two [2-^{3}H]mevalonoid hydrogen labels which were located[1] at C-10 could be displaced by enolization. The stereochemistry of labelling at this centre was determined by utilizing the differing rates of exchange of the *exo*- and *endo*-C-10 protons.[4] [2(*S*)-2-^{3}H,2-^{14}C]Mevalonic acid was fed to *Fusarium culmorum*. The culmorin was isolated and oxidized to the diketone (**3**; $R^1 = R^2 = O$). The results are summarised in the Table. After treatment of the diketone (**3**) with NaOD–D_2O for 1·5 h, the product contained 69·3% [$^{2}H_1$] and 30·7% [$^{2}H_2$] species. The *exo*-C-10 proton resonance (δ 2·28, J 18 Hz) had disappeared whilst the *endo*-C-10 proton resonance (δ 1·94, J 18 Hz) had collapsed. However, the product retained the [2(*S*)-2-^{3}H] label. Prolonged treatment (16 h) led to 29·0% [$^{2}H_2$] and 71·0% [$^{2}H_3$] species and to losses of the 2(*S*)-label [^{3}H:^{14}C ratio 2·81:1]. Thus the *endo*-C-10 proton of culmorin originates from the 2(*S*)-position of mevalonate.





Culmorin was shown to incorporate five of the possible six [5-^{3}H]mevalonoid hydrogen labels, *i.e.*, one more than would be expected in the absence of a hydride shift.[1] [5(*R*)-5-^{3}H,2-^{14}C]Mevalonic acid was fed to *F. culmorum*.

TABLE. The incorporation of [2(*S*)-2-^{3}H,2-^{14}C]- and [5(*R*)-5-^{3}H,2-^{14}C]mevalonates into culmorin.

	[2(*S*)-2-^{3}H,2-^{14}C]	[5(*R*)-5-^{3}H,2-^{14}C]
Mevalonate ^{3}H : ^{14}C ..	4·38 : 1	6·56 : 1
Atom ratio	3 : 3	3 : 3
μCi ^{14}C fed	60	108
Culmorin ^{3}H : ^{14}C ..	3·70 : 1	4·63 : 1
Atom ratio	2·53 : 3	2·1 : 3
% Incorporation ..	4·3	0·34
Culmorin diketone ^{3}H : ^{14}C	3·68 : 1	4·52 : 1
Atom ratio	2·98 : 3[a]	2·0 : 3
^{3}H : ^{14}C after exchange ..	3·59 : 1	2·41 : 1
Atom ratio	2·91 : 3	1·1 : 3
Degree of deuteriation ..	69·3% [$^2H_1$]	3% [$^2H_2$]
	30·7% [$^2H_2$]	97% [$^2H_3$]

[a] Atom ratio based on culmorin = 3 : 3 to allow for losses due prenyl isomerase.

The culmorin retained only two labels and there was no change in the ^{3}H : ^{14}C ratio on oxidation to the diketone. Thus the C-11 hydroxy-group has displaced a *pro*-5(*R*)-mevalonoid hydrogen atom. The stereochemical relationship between the *pro*-2(*S*) and *pro*-5(*R*) labels is that which would be expected from the folding of farnesyl pyrophosphate (**1**). When the diketone was treated with 1·5N NaOD–D_2O for 3 days, the product contained 3% [$^2H_2$] and 97% [$^2H_3$] whilst both the C-10 and the C-7 proton resonances had disappeared. In the exchange reaction there was a loss of a [5(*R*)-5-^{3}H]mevalonoid label from C-7. Thus in culmorin this centre, which originates from C-1 of farnesyl pyrophosphate, bears a 5(*R*)-label and a [5(*S*)-5-^{3}H]mevalonoid hydrogen {≡[1(*S*)-1-^{3}H]farnesyl} has migrated, probably to C-5.[1] In the biosynthesis of dendrobine (**4**)[2] the *pro*-1(*R*)-hydrogen of farnesol is transferred to C-8. This apparent dichotomy in the hydride shifts [10-membered ring: 1(*R*)-hydrogen migrates; 11-membered ring: 1(*S*)-hydrogen migrates]† may be rationalized if we assume that the initial attack of C-1 of farnesyl pyrophosphate (**1**) on the distal double bond leads to a cyclopropyl ion. Collapse of this to an 11-membered ring (*i.e.*, the formation of a C-1–C-11 bond) (**2**) involves the transfer of the appropriately oriented *pro*-1(*S*) hydrogen atom whereas collapse to a 10-membered ring (C-1–C-10 bond formation) involves the migration of the *pro*-1(*R*)-hydrogen atom. The carbonium ion now resides on C-1 of the farnesyl system from whence it may be involved in the further isomerisation and cyclisation of the carbon skeleton.

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† Professor D. Arigoni, E. T. H., Zurich, has made the same observation in sativene and longifolene biosynthesis [F. Dorn, P. Bernasconi, and, and D. Arigoni, *Chimia* (*Switz.*), 1975, **29**, 25].

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