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REVIEW ARTICLE NUMBER 5

NATURALLY OCCURRING FLAVANS UNSUBSTITUTED IN THE HETEROCYCLIC RING

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Key Word Index—Hetero ring-unsubstituted flavans, flavan-*O*-glycosides, distribution, isolation, structure elucidation, biological properties

Abstract—The naturally occurring free and glycosylated flavans are reviewed The distribution, methods of isolation and structure elucidation of flavans, by comprehensive spectral analyses, are presented Biological properties of flavans are appraised

INTRODUCTION†

The term flavan is applied, collectively, to a large group of naturally occurring compounds possessing a 3,4-dihydro-2-phenyl-2H-1-benzopyran (2-phenylchroman) nucleus Flavans substituted in the heterocyclic ring (3 and 4-positions, e g catechins) are frequently encountered in nature, but the unsubstituted flavans have rarely been found due, presumably, to their instability in solution leading to polymeric products The scientific literature on this latter class of compounds is scattered There has been so far no comprehensive report on the phytochemistry and biological properties of this class of compounds The present review records the distribution, spectral and biological properties of this class of flavan

DISTRIBUTION

Flavans consisting of a C_6–C_3–C_6 carbon skeleton having substitution in A and B rings but devoid of any substitution in the heterocyclic ring have been found to co-occur with chalcones [1–4], flavanones [1, 4], flavan-3,4-diols, flavonols [4], 1,3-diphenylpropanes [3, 5] and xanthones [6] The flavans isolated and identified so far, from plant sources, are listed in Table 1

The first two flavans reported from natural sources were 5,7,4′-trimethoxyflavan (**1**) [1] and 4 -methoxyflavan (**2**) [7] 5,7,4′-Trimethoxyflavan (**1**) was isolated from fully methylated resin of *Xanthorrhoea preissii* [1] (it was present in the original resin as a phenolic precursor of an unknown degree of methylation) To date, eighteen free flavans (aglycones) with varying degrees of oxygenation (mono to penta) in the terminal rings (A, B), and of varying patterns of oxygenation, are known in the literature (Table 1) There are only four reports of flavan-*O*-glycosides in nature and the sugar moieties (glucose and xylose) being distributed in ring A at position 7 or 5 [6, 8, 9] or in ring B [4] These simple flavans and flavan-*O*-glycosides are so far restricted to seven plant families, viz Ericaceae [9], Gentianaceae [6], Leguminosae [4, 10], Liliaceae [1, 7, 11], Myristicaceae [3, 5, 12], Santalaceae [8] and Amaryllidaceae [Ghosal, S, Srivastava, R S and Singh, S K, unpublished work] Recently, Gomez *et al* [10] reported for the first time flavans prenylated at position 8, from *Tephrosia madrensis* (Leguminosae) Additionally, three biflavans (**43, 44, 45**) were encountered in the Liliaceae and Palmae [13–15]

ISOLATION AND STRUCTURAL ELUCIDATION

Extraction and separation

Extraction of naturally occurring flavans (free and glycosylated) is usually carried out on dried plant materials The classical method of continuous extraction using increasingly polar solvents (light petrol, chloroform, methanol) has proved to be efficient [4, 6] Strongly polar solvents (acetone, ethanol, methanol) are also directly employed for extraction of dried and milled plant materials followed by solvent-gradient fractionation of the polar extractives Flavans of varying polarities are often separated by column chromatography on silica gel using solvents of graded polarity (hexane, benzene, ethyl acetate and different proportions of mixtures thereof) as eluants Preparative TLC on silica gel (using EtOAc–MeOH–H_2O, 18 1 1, MeOH–$CHCl_3$–HOAc, 8 1 1) is employed in instances of molecules difficult to separate [16] In a number of cases the isolation is accomplished after derivatization, because of instability of the parent compounds, into permethyl ethers or acetates followed by column chromatography [1, 2] Strongly polar compounds are occasionally obtained after passing the methanol solutions over Sephadex LH20 using methanol as eluant [16] Strongly polar flavans are also separated by

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†The information covers the literature up to January 1984

Table 1 Distribution of flavans in higher plants

Flavan	Mp °C	$[\alpha]_D$	Source
5,7,4'-Tri-OMe (1)	109–111	—	*Xanthorrhoea preissii* Endl [1]
4'-OMe- (2)	83–84	—	[Canadian beaver (*Caster fiber*)] [7]
(−)-4'-OH-7-OMe- (3)	148 5–149 5	−15 6° (EtOH)	*Stypandra grandis* C T White [11]
(−)-4'-OH-7-OMe-8-Me- (4)	126–127	−22 4° ($CHCl_3$)	*Dianella revoluta* R Br [11]
(2*R*)-5-OH-7-*O*-Glc-Koaburarin (5)	221–222	−64 2° (EtOH)	*Enkianthus nudipes* Lour [9]
(2*S*)-7-OH-5-OMe-6-Me- (9)	122–124	−9 25° ($CHCl_3$)	*Dracaena draco* L [2] (Dragon's blood)
(2*S*)-7-OH-5-OMe- (11)	85–87	−6 35° ($CHCl_3$)	*Dracaena draco* L [2] (Dragon's blood)
(+)-3',4'-Di-OH-5,7-di-OMe (13)	98–100	—	*Iryanthera coriacea* Ducke [12]
(±)-2'-OH-7-OMe-4',5'-(O CH_2 O)- (15)	164–166	—	*Iryanthera juruensis* Warb [12]
7,4'-Di-OH-5-*O*-β-D-Xyl- (17)	243	−31 8° (EtOH)	*Buckleya lanceolata* Miq [8]
7,5'-Di-OH-4'-OMe-3'-*O*-β-D-Glc-Auriculoside (21)	140	−77 0° (MeOH)	*Acacia auriculiformis* A Cunn [4]
(±)-7,4'-Di-OH-3'-OMe- (26)	157–159	—	*Iryanthera elliptica* Ducke [5]
7-OH- (28)	—	—	*Narcissus pseudonarcissus** L [16]
7,4'-Di-OH- (29)	197–198	—	*Narcissus pseudonarcissus** L [16]
(−)-7,4'-Di-OH-8-Me- (30)	132–135	−36 4° ($CHCl_3$)	*Narcissus pseudonarcissus** L [16]
7,2'-Di-OH-6,8-di-Me-4',5'-(O CH_2 O)- (31)	168–170	—	*Iryanthera laevis* Markgr [3]
7,2'-Di-OH-5,8-di-Me-4',5'-(O CH_2 O)- (33)	174–176	—	*Iryanthera laevis* Markgr [3]
5,2'-Di-OH-7-OMe-6,8-di-Me-4',5'-(O CH_2 O)- (35)	164–166	—	*Iryanthera laevis* Markgr [3]
2'-OH-7-OMe-4',5'-(O CH_2 O)- (15)	164–166	—	*Iryanthera laevis* Markgr [3]
7-OH-4'-OMe-Broussin (37)	120–122	−17 4° ($CHCl_3$)	*Broussonetia papyrifera* Vent† [22] (paper mulberry)
5,7-Di-OMe-8-pren- (38)	76–77	−79 5° ($CHCl_3$)	*Tephrosia madrensis* Seem [10]
7-OH-3',4'-di-OMe-5'-*O*-β-D-Glc-Diffutin (40)	144–145	−46 3° (MeOH)	*Canscora diffusa* R Br [6]
2'-OH-7-OMe-4',5'-(O CH_2 O)- (15)	140	—	*Zephyranthes flava* Roem et Schult‡
7,4'-Di-OH-3'-OMe- (26)	145	—	*Zephyranthes flava* Roem et Schult‡
7-OH-3',4'-(O CH_2 O)- (42)	109	—	*Zephyranthes flava* Roem et Schult‡
Xanthorrhone (43)	193–196	—	*Xanthorrhoea* (*preissii* ?) [14]
14-Hydroxyxanthorrhone (44)	190–193	—	*Xanthorrhoea* (*preissii* ?) [14]
Biflavan 12 (45)	—	—	*Daemonorops draco* Blume [15]

*Not native, produced only when bulbs were inoculated with conidia of *Botrytis cinerea*

†Not native, produced when wounded

‡Ghosal *et al*, unpublished work

Glc, glucose, Xyl, xylose, pren, prenyl

HPLC on ODS Hypersil (5 μM) column using isocratic elution with 35% methanol in 5% formic acid [16] as well as on reversed phase (C_8 or C_{18} bondapak) columns using methanol–water (4 1, 7 3) as solvent [6]

The thin layer chromatoplates are viewed in UV light (λ254 nm), and after keeping the chromatoplates in I_2 vapour when reddish-purple colours are developed [6, 11] This forms a basis for their detection even when admixed with a number of unrelated phenolic constituents Spraying plates with dilute methanolic sulphuric acid (2%) produces reddish-brown colorations [6] Flavan glycosides having free phenolic groups respond to ferric reagent and Feigels test [4] A yellow coloration is developed when chromatograms are sprayed with diazotized *p*-nitroaniline [16] The purity of the isolated flavans can also be tested by analytical HPLC [6]

The hydrolysis of glycosides is carried out by warming methanol solutions with dil hydrochloric acid according to procedures followed for flavonoids [4, 6, 17] Enzymatic hydrolysis (with emulsin) is also of interest in case of glucosides [4, 6, 9] Aglucones are separated by column chromatography or preparative TLC on silica gel (C_6H_6–EtOAc, 3 1, or C_6H_6–$CHCl_3$, 1 1) [4, 6] The glycone moieties are identified as their alditol acetates by GC (5%, Silar 10C on Gas-chrom Q, temp 210°, flow rate 40 ml/min, N_2) [6]

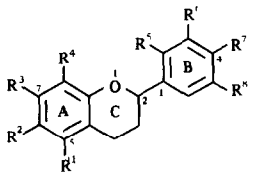
Spectral properties of flavans

Comprehensive spectral analyses (UV [1–16, 22], IR [3, 5, 6, 8–12, 22], ^1H NMR (1–16, 22], and ^{13}C NMR [4, 6, 18]) of flavans are reported These data together

with chemical interconversions [2, 6], and synthesis [11, 16] have led to establishment of their structures (Scheme 1)

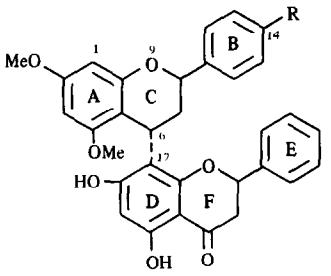
UV spectra of flavans

Flavans exhibit UV maxima of a simple benzenoid chromophore having an unconjugated aromatic system There are two major adsorption maxima in the regions 220–225 and 275–290 nm, with additional maxima (or shoulders) in the higher wavelength region in cases where a number of functional groups like hydroxyl/methoxyl, methylenedioxy and methyl, are attached to the terminal ring(s) The lower wavelength maximum is of higher intensity in most flavans Glycosylation of the 7-hydroxyl group results in an additional maximum at 251 nm [9],



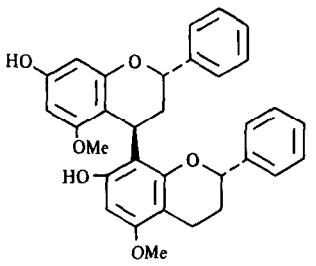
Flavan	Substitution pattern							
	R^1	R^2	R^3	R^4	R^5	R'	R^7	R^8
1	OMe	H	OMe	H	H	H	OMe	H
2	H	H	H	H	H	H	OMe	H
3	H	H	OMe	H	H	H	OH	H
4	H	H	OMe	Me	H	H	OH	H
5	OH	H	OGlc	H	H	H	H	H
6	OH	H	OH	H	H	H	H	H
7	OAc	H	OAc	H	H	H	H	H
8	OMe	H	OMe	H	H	H	H	H
9	OMe	Me	OH	H	H	H	H	H
10	OMe	Me	OAc	H	H	H	H	H
11	OMe	H	OH	H	H	H	H	H
12	OMe	H	OAc	H	H	H	H	H
13	OMe	H	OMe	H	H	OH	OH	H
14	OMe	H	OMe	H	H	OAc	OAc	H
15	H	H	OMe	H	OH	H	$-OCH_2O-$	
16	H	H	OMe	H	OAc	H	$-OCH_2O-$	
17	OXyl	H	OH	H	H	H	OH	H
18	OXyl	H	OMe	H	H	H	OMe	H
19	OXyl(OAc)$_3$	H	OAc	H	H	H	OAc	H
20	OXyl(OAc)$_3$	H	OMe	H	H	H	OMe	H
21	H	H	OMe	H	H	OGlc	OMe	OH
22	H	H	OAc	H	H	OGlc(OAc)$_4$	OMe	OAc
23	H	H	OMe	H	H	OGlc	OMe	OMe
24	H	H	OH	H	H	OH	OMe	OH
25	H	H	OAc	H	H	OAc	OMe	OAc
26	H	H	OH	H	H	OMe	OH	H
27	H	H	OAc	H	H	OMe	OAc	H
28	H	H	OH	H	H	H	H	H
29	H	H	OH	H	H	H	OH	H
30	H	H	OH	Me	H	H	OH	H
31	H	Me	OH	Me	OH	H	$-OCH_2O-$	
32	H	Me	OAc	Me	OAc	H	$-OCH_2O-$	
33	Me	H	OH	Me	OH	H	$-OCH_2O-$	
34	Me	H	OAc	Me	OAc	H	$-OCH_2O-$	
35	OH	Me	OMe	Me	OH	H	$-OCH_2O-$	
36	OAc	Me	OMe	Me	OAc	H	$-OCH_2O-$	
37	H	H	OH	H	H	H	OMe	H
38	OMe	H	OMe	pren	H	H	H	H
39	OH	H	OMe	pren	H	H	H	H
40	OGlc	H	OH	H	H	OMe	OMe	H
41	OGlc(OAc)$_4$	H	OAc	H	H	OMe	OMe	H
42	H	H	OH	H	H	$-OCH_2O-$		H

Glc = glucose, Xyl = xylose, pren = prenyl



43 Xanthorrhone , R=H

44 14 - Hydroxyxanthorhone , R= OH



45 Biflavan 12

Scheme 1 Naturally occurring biflavans and their derivatives

while prenylation at C-8 produces an additional maximum at 263 nm [10] Flavans with 3',4'-methylenedioxy and additional C_2 hydroxyl substituents, in ring B, exhibit maxima in the region 300 nm or above in addition to the maximum at 280–290 nm [3, 12] Thus, flavans highly substituted in A and B rings tend to absorb at wavelengths greater than usual [3, 12] Acetylation of the hydroxyflavans produces hypsochromic shifts and hypochromic effects in both maxima

The addition of the usual shift reagents [17] to an alcoholic solution of flavans induces considerable shifts in the UV maxima of flavans 7-Hydroxylated flavans exhibit bathochromic shifts of both the maxima with sodium methoxide [6] or sodium hydroxide [3, 5] The presence of *ortho*-dihydroxyl groups in ring B produces the expected bathochromic shifts on addition of a mixture of sodium acetate and boric acid [12], this property being similar to that exhibited by other flavonoids [17]

The biflavans, e g xanthorrhone (**43**) and 14-hydroxyxanthorrhone (**44**), exhibit two maxima (295 and 336 nm) in their UV spectrum The lower wavelength maximum is shifted hypsochromically to 245 on addition of potassium hydroxide Aluminium chloride, on the other hand, shifts the lower wavelength maximum hypsochromically and the upper wavelength maximum bathochromically with the appearance of a new maximum around 315 nm [14]

IR spectra of flavans

The IR spectra of flavans are not used for diagnostic purposes However, the IR data are largely used to detect the presence of oxygen functions (hydroxyl/methoxyl/methylenedioxy/acetoxyl/glucosyloxy) in rings A and B [3, 6, 8, 9] The IR data also form useful adjuncts to structural elucidation and are often quoted in support of identities of newly isolated flavans with known standards [3, 5, 6, 8–12, 22]

^{1}H NMR spectra of flavans

The proton magnetic resonance spectral analyses of flavans are carried out in deuterochloroform, carbon tetrachloride, deuterodimethylsulphoxide, acetone-d_6, pyridine-d_5, methanol-d_4 and mixtures thereof but the bulk of the published data refers to deuterochloroform, deuterodimethylsulphoxide and acetone-d_6 as solvents

The chemical shifts of the protons of free flavans indicate the aromatic (rings A and B) protons (in the range δ 5 96–6 98 and 6 31–7 53, respectively) and aliphatic (ring C) protons (in the range δ 1 8–5 1) suggesting the presence of two aromatic rings as shown in the partial structure Ar CH(–O)–CH_2CH_2 Ar' The effects of various solvents on chemical shifts of rings A, B and C protons provide important information [2, 4, 6, 9, 16] The influence of neighbouring hydroxyl, alkoxyl, alkyl and acetoxyl substituents on the chemical shift of ring protons is also significant (*vide infra*) It appears that a slight but consistent paramagnetic (downfield) shift of the proton signal occurs upon methylation of *ortho* (0 05–0 20 ppm in deuterochloroform, 0 04–0 09 ppm in deuterodimethyl sulphoxide) and *para* (0 02–0 04 ppm) hydroxyl groups in flavans Acetylation of hydroxyl groups produces deshielding effects on signals of the *ortho*/*meta*/*para* protons of the corresponding flavan ring which is similar in nature and magnitude to those noted for other flavonoids [17]

The five aliphatic protons (ring C) are also completely analysable The one proton quartet signal (in the range δ 4 8–5 1) corresponds to X of an ABX and represents the benzylic axial oxymethine H which is vicinal to two other hydrogens (OC<u>H</u>CH_2) The latter two protons and an additional pair of hydrogens (associated with C-4) exhibit two two-proton multiplets (in the range δ 1 8–2 3 and 2 6–3 0) ascribable to 2H-C-3 and 2H-C-4, respectively

An important application of ^{1}H NMR spectra of flavans constitutes the stereochemical assignment at C-2 Thus, the coupling patterns and constants of H-C-2 and 2H-C-3 [4, 6, 9] of free flavans and flavan aglucones suggest that the aryl ring in naturally occurring flavans is α and equatorial

The above method of analysis is also applicable to glycosyloxy flavans In glycosyloxy flavans, the signal of the anomeric proton of the sugar moiety and the methine proton of the benzyl ether system overlap [4, 6, 8] Acetylation of hydroxyl groups (in the ring and glycosyl moiety) produces considerable downfield shift of the neighbouring ring protons [2, 4, 6, 9, 12] Appreciable influence of different solvents on chemical shifts of glycosyloxy flavans is observed [6]

Some specific examples of application of ^{1}H NMR spectra to structure elucidation of flavans are given below

Ring-A protons

7-Oxygenated flavans (**3, 15, 16, 21–29**) [4, 5, 11–13, 16] The H-5 and H-6 protons appear as two *ortho* coupled doublets (J = *ca* 8 5 Hz) in the range δ 6 7–6 98 and 6 20–6 52, respectively The H-6 further *meta* couples (J = *ca* 2 5 Hz) with H-8 proton, the latter appears in the range δ 6 13–6 47, as a singlet or a doublet (J = 2 5 Hz) [4]

depending on the substituent/solvent Acetylation of the hydroxyl group at C-7 causes shift of the H-6 and H-8 signals downfield (by 0 07–0 28 ppm), the shift is maximum (*ca* 0 28 ppm) in case of the H-8 signal, which even reverses their positions relative to each other

5,7-*Dioxygenated flavans* (**1, 5, 11, 13, 17, 40**) [2, 6, 8, 9, 12] The *meta* related protons of the phloroglucinol type A-ring, exhibit signals as doublets (J = *ca* 2 5 Hz) in the range δ 5 96–6 51 (H-6) and 6 00–6 61 (H-8) The H-8 proton signal appears downfield as compared to H-6 in a few flavans Both these signals are shifted downfield on acetylation (by 0 1–0 2) The acetate shift has been used to locate the glucosyloxy signal in some flavans Thus, the comparative downfield chemical shift experienced by the H-6 (0 36 ppm) over the H-8 (0 27 ppm) signal, on acetylation, located the *O*-glucosyl substituent at C-5 in diffutin (**40**) [6]

Ring-B protons

In case of flavans having no oxygen substituent in the B-ring, all the protons appear as a symmetrical multiplet in the range δ 7 25–7 43 (i e somewhat downfield from the A-ring protons) (**5, 9, 11, 28, 38**) [2, 9, 10, 16]

4′-*Oxygenated flavans* (**1–4, 17, 29, 30**) [1, 7, 8, 11, 16] In the ^{1}H NMR spectra of the 4′-oxygenated flavans, the protons (H-2′, H-3′, H-5′, H-6′) exhibit an A_2B_2 quartet (J = 8 Hz) in the range δ 6 80–7 53 The H-3′ and H-5′ doublet occurs upfield (δ 6 80–7 5) compared to the H-2′, H-6′ doublet (δ 7 11–7 53) due to the shielding effect of the 4′-hydroxyl The downfield branch of the A_2B_2 quartet shows fine splitting (J = 0 5 Hz) which can be removed by irradiation at the frequency corresponding to the one proton resonance at δ 4 95 [11] This indicates allylic coupling between the benzenoid protons and the methine proton of a benzyl ether system In the spectrum of the flavan acetates the A_2B_2 pattern is displaced downfield (by about 0 06–0 21 ppm) which shows that the hydroxyl group is located in the *para* disubstituted benzene ring

3′,4′-*Dioxygenated flavans* (**13, 14, 26, 27, 40**) [5, 6, 12] The H-2′, H-5′ and H-6′ protons in a C-3′, C-4′ dihydroxy system appear as a broad singlet (δ 6 93) [5] or as a complex multiplet (δ 6 75–7 1) [6, 12] Acetylation of the hydroxyls produces a deshielding effect (by about 0 43 ppm) on these protons (3H multiplet)

3′,4′,5′-*Trioxygenated flavans* (**21–25**) [4] In auriculoside (**21**), the two *meta* coupled (J = 1 5 Hz) nonequivalent protons, H-2′ and H-6′, appear as doublets at δ 6 57 and 6 5, respectively The positions of the two signals are reversed [δ 6 67 (H-2′) and δ 7 01 (H-6′) in the corresponding acetate derivative (**22**)] In auriculin (the aglucone, **24**), both H-2′ and H-6′ are equivalent and thus appear as a two-proton singlet at δ 6 31

2′,4′,5′-*Trioxygenated flavans* (**15, 16, 31–36**) [3, 12] All the flavans reported in this group are 2′-hydroxy-4′,5′-methylenedioxy compounds The H-3′ and H-6′ protons appear as singlets in the range δ 6 40–6 78 Free rotation of the C-2 aryl group, containing a C-2′ hydroxyl function is not feasible because of its hydrogen bonding with the heterocyclic ring oxygen and thus, the H-6′ and H-3 (ring-C) protons interfere with each other in their ^{1}H NMR resonance [12]

There are only a few examples of the use of ^{1}H NMR analysis in the elucidation of the structure of biflavans The ^{1}H NMR spectra of xanthorrhone (**43**) and hydroxyxanthorrhone (**44**) differ from each other by the presence of two A_2B_2 doublets (δ 6 81, 7 21, J = 9 Hz) in the latter compound due to an extra hydroxyl group present in ring B at the *para* position (C-14) [14] The presence of only three aromatic protons in the upfield region suggests the linkage of the two units through the phloroglucinol ring of the flavanone moiety Double irradiation experiments and presence of seven aliphatic protons (in the range δ 2 12–5 35) support a –CH–CH_2–CH– system in the biflavan

^{13}C NMR spectra of flavans

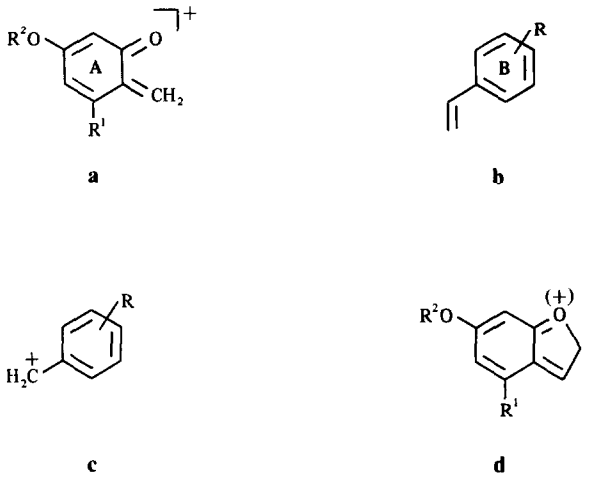
^{13}C NMR (proton-noise decoupled and single frequency off-resonance decoupled) spectra of only a few flavans have been recorded so far [4, 6, 18, Frahm, A W, pers comm] However, the assignments of the respective carbon signals enable the technique to be used in the determination of structure of a new flavan The solvents commonly used are, as in ^{1}H NMR, deuterochloroform ($CDCl_3$), deuteromethanol (CD_3OD) and hexadeuterodimethyl sulphoxide (DMSO-d_6) ^{13}C resonance for flavans occurs over a range of δ0–170 downfield from TMS The absence of carbonyl groups in a simple flavan is indicated by the absence of any ^{13}C signal beyond δ170 The ranges of chemical shifts for carbon types encountered in flavans are given in Table 2 The introduction of an alkoxy (hydroxyl/methoxyl/glucosyloxy) group causes upfield shift of the β-carbons as in the case of other flavonoids [19] Another interesting feature of the spectra of heavily substituted flavans (e g 5-glucosylated flavans [6]) is the pronounced upfield shift of the C-3 (*ca* 6 ppm) and C-4 (*ca* 2 ppm) signals, presumably due to a steric compression of these carbons by the glucosyloxy function [20] Concomitant upfield shifts take place in the signals of the glucosyl carbons

EIMS of flavans

Flavans, unless highly substituted, are not fragmented before providing the molecular ion peak as one of the most abundant peaks [12, 16] Distribution of the substituents (alkoxy/alkyl) in rings A and B is shown in the fragment ions formed by the retro-Diels–Alder reaction involving the heterocyclic ring [1, 4, 6, 7, 21] The initial products of the retro-Diels–Alder fragmentation are the ions **a** and **b** The ion **c** arises by a concomitant H capture thereby resulting in the formation of ion **d** from rings A

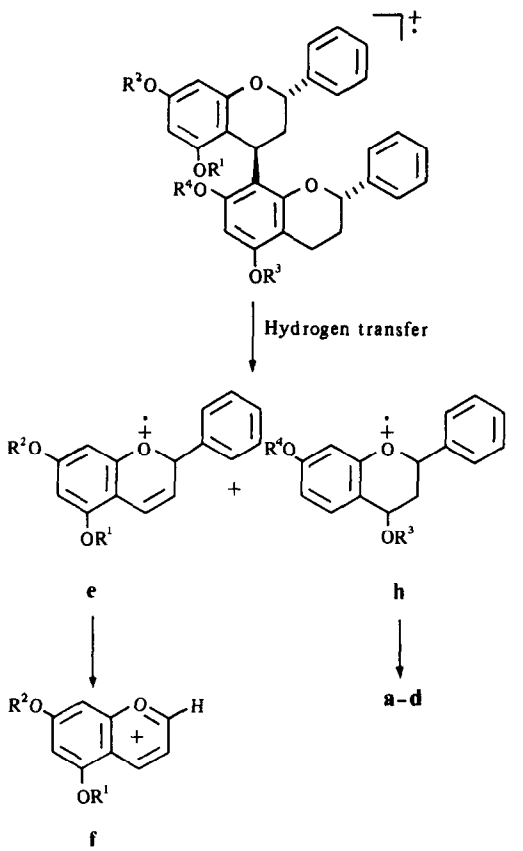
Table 2 ^{13}C chemical shift ranges for various carbon types encountered in flavans

Carbon type	Chemical shift range [δ-values (ppm) from TMS]
Aromatic	
(a) Oxygenated	155–159 (no *o*/*p*-oxygenation), 148–152 (with *o*/*p*-oxygenation),
(b) Non-oxygenated	127–131 (no *o*/*p*-oxygenation), 101–115 (with *o*/*p*-oxygenation)
Aliphatic	
(a) Oxygenated (sugars)	60–102
(b) Non-oxygenated	19–31
(c) Methylenedioxy	98–100
(d) Aromatic OMe	55–60



R = OH/OMe , R^1 = H/OH/OMe , R^2 = H/Me

Scheme 2 Principal initial ions produced in the mass spectral fragmentation of flavans



Scheme 3 Principal fragment ions produced in the mass spectra of biflavans

and C The appearance of ion **d** is strong evidence in favour of the flavan nucleus

The biflavans (e g **43** and **44**) also exhibit identifiable molecular ion peaks Accurate mass measurements (m/z 524 18 and m/z 540 17, respectively) have established the molecular formulae of **43** and **44** [14] The molecular ion of the biflavan initially undergo fragmentation of the two units to provide ions **e** and **h** by a hydrogen transfer The ions **e** and **h** experience further fragmentation to provide, respectively, ions **f** and **a–d**

BIOLOGICAL PROPERTIES

Naturally occurring flavans exhibit a number of important biological activities which, if exploited properly, may lead to valuable new drugs or agro-chemicals

Antimicrobial activity

Recent reports have shown antibacterial action of flavans and the defensive role played by these compounds against microorganisms [22] Broussin (**37**) was isolated only from wounded xylem tissue of paper mulberry (*Broussonetia papyrifera* Vent.) shoots It was not present in the healthy xylem tissue of *B papyrifera* Subsequently, the flavan was tested for its activity against *Bipolaris leersiae* and was found to significantly inhibit (in 10^{-4}–10^{-5} M concn) the growth of the microorganism Hence it was ascribed as a phytoalexin [22]

Flavans were also found to be active against *Botrytis cinerea* Pers ex Fr [16] Three flavans, 7-hydroxyflavan (**28**), 7,4′-dihydroxyflavan (**29**) and 7,4′-dihydroxy-8-methylflavan (**30**), were produced by *Narcissus pseudonarcissus* L when the scales of its bulbs were inoculated with suspensions of conidia of *B cinerea* The growth of the fungus was restricted within the lesions formed beneath the inoculum droplets (in *N pseudonarcissus*) The three flavans were absent in fresh or frozen and thawed healthy tissues of *N pseudonarcissus* In addition to showing fungitoxic activity on TLC plate bioassays, samples of **28–30** were also shown to be active against germinated spores of *B cinerea* in liquid culture ED_{50} values against the germ tube growth were 22, 65 and 32 μg/ml, for **28, 29** and **30**, respectively [16]

Flavans were also shown to produce bactericidal activity against gram-positive bacteria [23] and anti-viral activity [24]

Pharmacological activity

Auriculoside (**21**) was the first flavan glycoside to be investigated pharmacologically The initial results with auriculoside, as reported by Sahai *et al* [4], revealed only a minor CNS depressant activity However, subsequent studies with related glucosyloxy flavans showed pronounced adaptogenic (anti-stress/anti-anxiety) activity [6, 25] Thus, diffutin (**40**) isolated from *Canscora diffusa*, was initially found to produce a mild CNS depressant action (potentiation of barbiturate hypnosis and morphine analgesia in laboratory animals, in doses of 20–50 mg/kg ip) [6, 25] While elucidating the mechanism of this CNS depressant action of diffutin, it was found to be associated with anti-stress and anti-anxiety activities (collectively termed 'adaptogenic activity') in the battery of tests designed for such activities in laboratory animals [6, 25] Diffutin also exhibited a marked positive inotropic

effect in perfused frog heart in doses of 10–30 mcg (no arrhythmogenic property). It further potentiated the contractile responses of guinea-pig vas deferens to catechol amines (by a process other than the uptake inhibition of adrenaline) Diffutin was found to be non-toxic up to 500 mg/kg in dog In summary, these results provide a reasonable explanation for the therapeutic use of *Canscora diffusa* in some mental disorders, e g melancholia, in the Indian system of medicine The plant extract is used as a substitute of *C decussata* Schult, the latter contains xanthone glucosides as its active principle [26, 27]

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