P A R T - II

STRUCTURE OF GUGGULLIGNAN - 1

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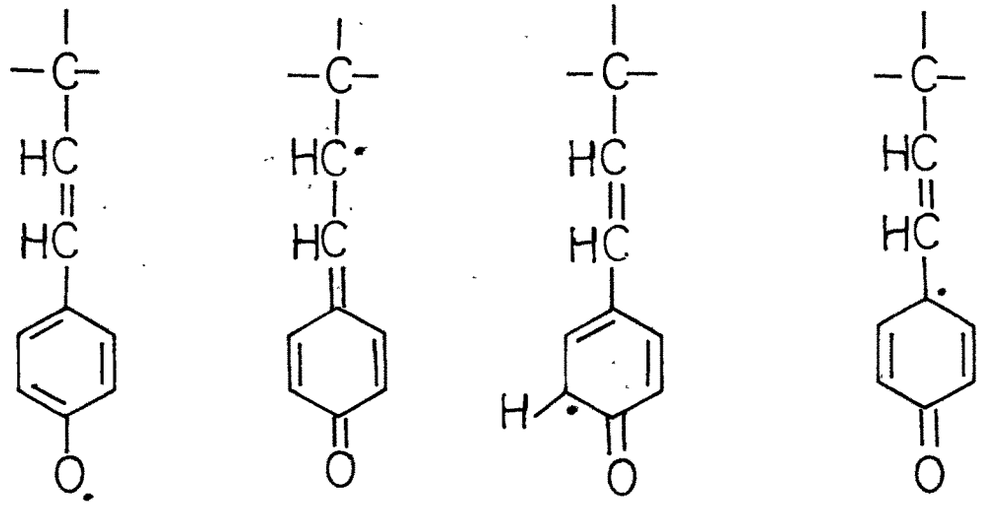
Abstract

The structure of guggullignan-1 was revised based on the high-field ^{1}HNMR (500 MHz) spectrum.

INTRODUCTION

Formation and classification of lignans

Lignans are a group of compounds composed of two phenylpropane (C_6-C_3) units linking at side chains. They are formed by oxidative dimerisation of monomeric C_6-C_3 phenols.[1] Pummerer suggested a radical theory for the oxidative coupling of phenols.[2] Differently substituted 1-(p-hydroxy phenyl)-1-propenes in their various oxidation states may yield a radical for which the mesomeric forms a-d may be considered. Their dimerization or combination, and subsequent re-aromatization will then lead to the different types of lignans.



a b c d

The most abundent lignans occurring in nature, originate in a combination of two b radicals.

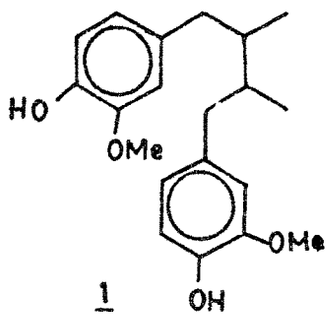
Broadly, lignans can be classified,[3] based on their skeletal structure, into 5 groups *viz*;

1) 1,4-Diaryl butane derivatives,
   e.g., (a) Dihydroguaiaretic acid (1)
   [1,4-Bis-(4-hydroxy-3-methoxyphenyl)-2,3-dimethyl butane],
   (b) Guaiaretic acid (2)
   [1,4-Bis-(4-hydroxy-3-methoxyphenyl)-2,3-dimethyl-1-butene].

2) 2,3-Dibenzylbutyrolactone derivatives.
   e.g., (a) Hinokimin (3)
   [2,3-Bis-(3,4-methylenedioxybenzyl)-butyrolactone],
   (b) Matairesinol (4)
   [2,3,-Bis-(4-hydroxy-3-methoxybenzyl)-butyrolactone].

3) Tetrahydrofuran derivatives.
   e.g., (a) Olivil (5)
   [2,5-Bis-(4-hydroxy-3-methoxyphenyl)-3,4-bis-(hydroxy methyl) tetrahydrofuran],
   (b) Lariciresinol (6)
   [4-(4-hydroxy-3-methoxybenzyl)-2-(4-hydroxy-3-methoxy-phenyl)-3-hydroxy methyl tetrahydrofuran].

4) Tetrahydrofurofuran derivatives
   e.g., (a) Pinoresinol (7).
   [2,6-Bis-(4-hydroxy-3-methoxyphenyl)-3,7-dioxabicyclo (3.3.0) octane],



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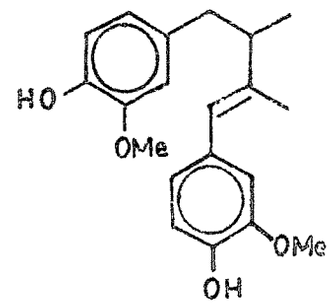

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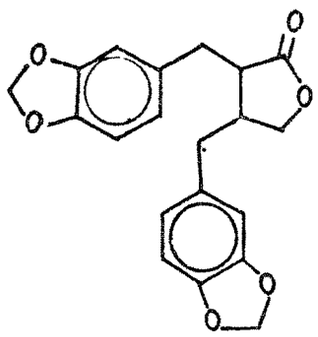

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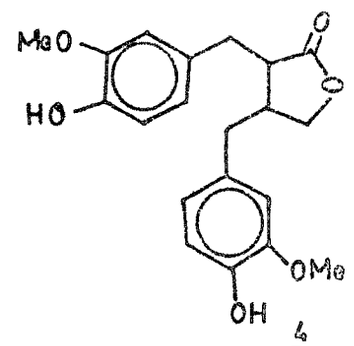

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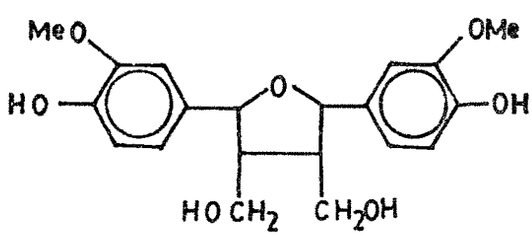

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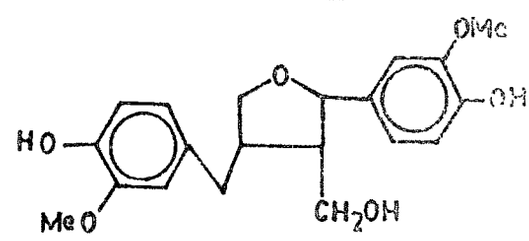

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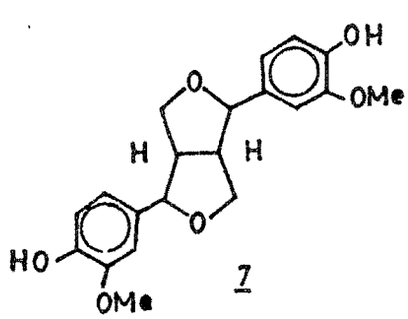

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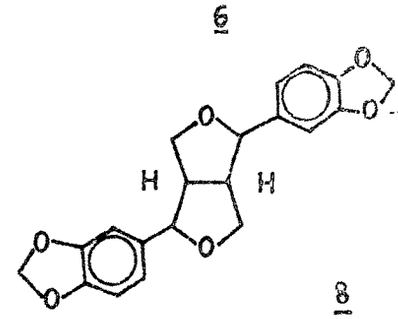

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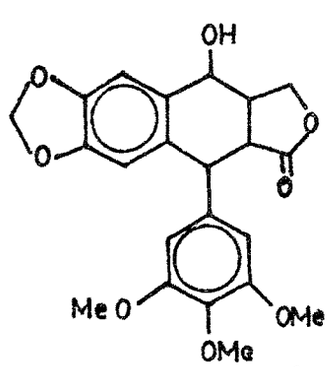

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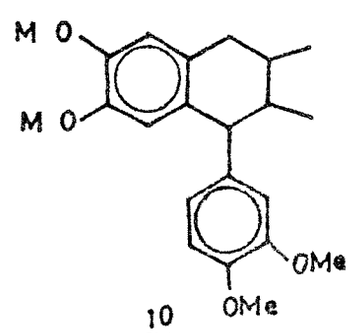

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Fig. 1

(b) Sesamin (8)

[2,6-Bis-(3,4-methylenedioxyphenyl)-3,7-dioxabicyclo-(3.3.0)-octane].

5) 4-Aryl tetrahydronaphthalene derivatives.

e.g., Podophyllotoxin (9)

[1-Hydroxy-2-hydroxymethyl-6,7-methylenedioxy-4-(3,4,5-trimethoxyphenyl)-1,2,3,4-tetrahydronaphthalene-3-carboxylic acid lactone],

(b) Galcatin (10).

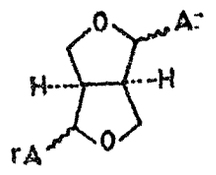
[2,3-Dimethyl-6,7-methylenedioxy-4-(3,4-dimethoxyphenyl)-1,2,3,4-tetrahydronaphthalene].

Apart from the above type of lignans, there are neo-lignans and cyclolignans which are formed by the dimerization of allyl phenols and phenylpropanes.

Guggullignans belong to tetrahydrofurofuran derivatives group and hence this group will be briefly described here.

Tetrahydrofurofuran derivatives
(2,6-Diaryl-3,7-dioxabicyclo [3.3.0] octanes)

The 2,6-diaryl-3,7-dioxabicyclo [3.3.0] octanes are one of the largest group of lignans and are widely distributed in



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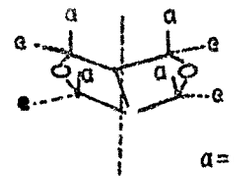

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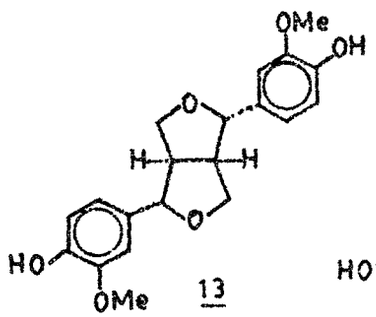
a = axial
e = equtorial



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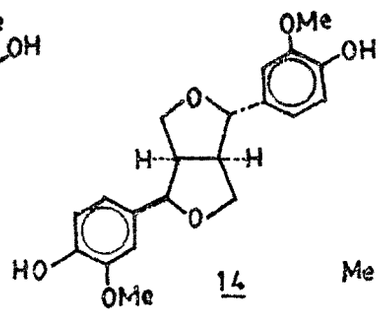

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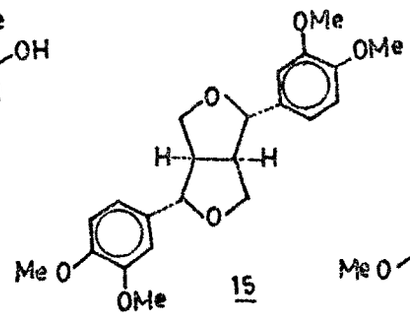

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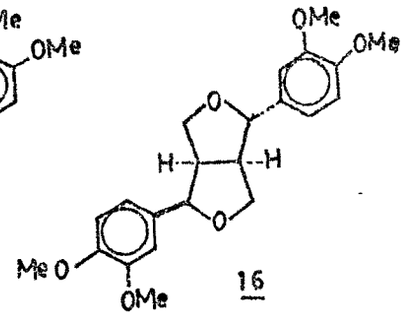

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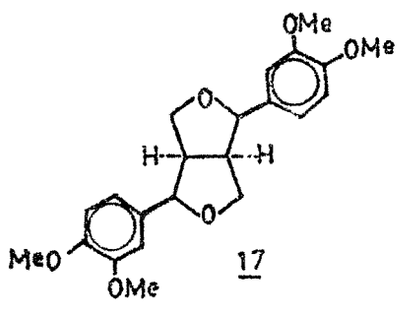

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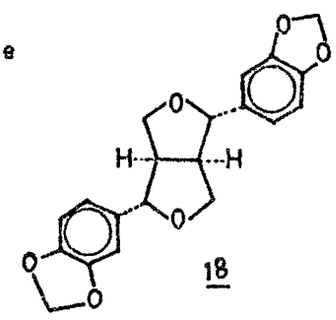

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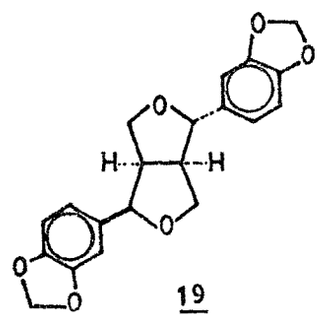

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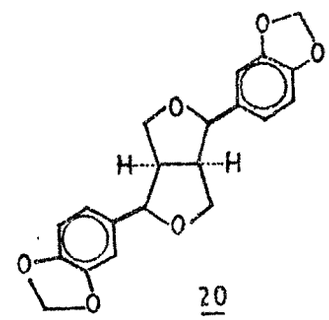

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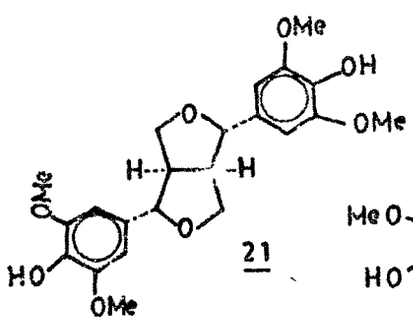

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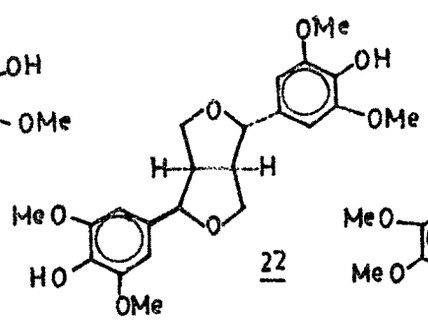

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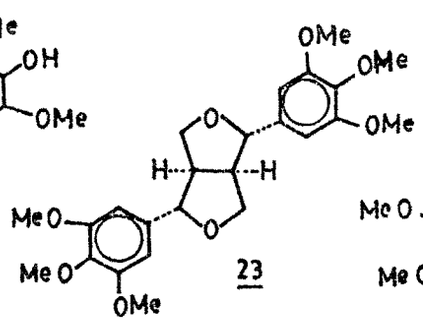

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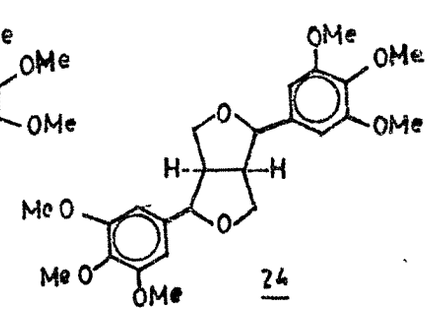

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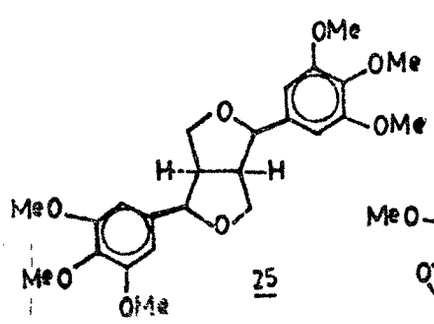

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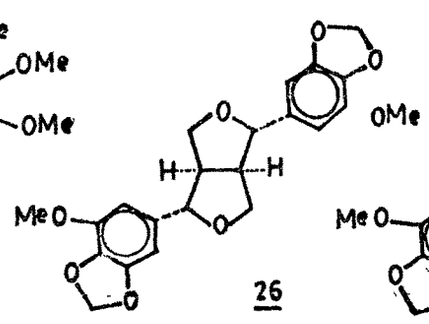

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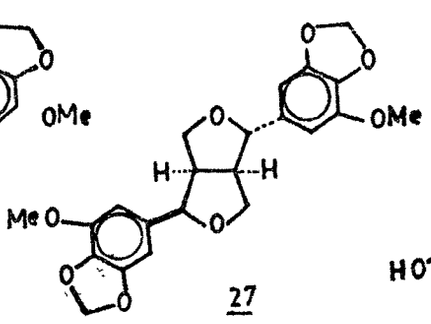

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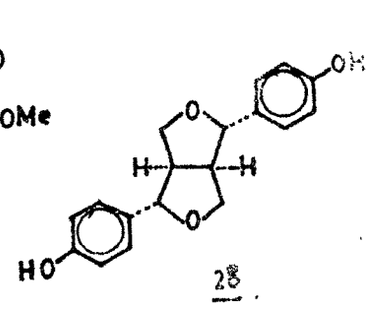

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Fig. 2

nature. They are present in the roots, heartwood, foliage, fruits, c resinous exudates of plants.[3] These lignans contain the basic 3,7-dioxabicyclo [3.3.0] octane skeleton (11) and have aryl (or occationally aryloxy) group attached to carbon atoms 2 and 6. They may also have hydroxy or acetoxy groups attached to one or more of the non-aromatic carbon atoms. The two heterocyclic rings would be expected to be cis-fused since otherwise the bicyclic system would be highly strained. This stereochemical assignment has been proved by degradation exoeriments.[4] Nitric acid oxidation of these lignans established[5] that the aryl groups are attached to carbon atoms 2 and 6. Substituents attached to carbon atoms 2,4,6 and 8 can occuoy an axial oosition (aoproximately oarallel to the axis of the molecule) or an equitorial position (approximately perpendicular to the axis of the molecule) (12, Fig. 2). For symmetrically substituted compounds, three stereochemically distinct series are, therefore possible in which the aryl groups are diequitorial, equitorial-axial and diaxial. The known naturally occurring 2,6-diaryl-3,7-dioxabicyclo [3.3.0] octanes are listed in Table 1.

Spectroscopy in the structure elucidation of 2,6-diaryl-3,7-dioxabicyclo [3.3.0] octanes.

PMR: PMR spectrum of these lignans is quite characteristic and is very heloful in determining the different substituents and substitution pattern on the aromatic ring. The most imoortant

Table 1. Naturally occurring 2,6-diaryl-3,7-dioxabicyclo-3.3.0 octanes. (symmetrically substituted)

	Name	Structure	m.p.	$[\alpha]_D$*	Ref.
1	Pinoresin	13	120-1°	82.4°	6
2	Epipinoresin	14	137-8°	130.4°	6
3	Eudesmin	15	107°	64.4°	7
4	Epieudesmin	16	125-6°	119°	8
5	Diaeudesmin	17	157-8°	+316°	9
6	Sesamin	18	123°	71°	8
7	Episesamin	19	121-5°	124°	8
8	Diasesamin	20	168-71°	+385°	8
9	Syringaresinol	21	172-7°	62.2	10
10	Episyringaresinol	22	210-11°	172°	10
11	Yangambin	23	121-3°	+46.6°	10
12	Epiyangambin	24	118-20°	+ 119°	10
13	Diayangambin	25	151-2°	+ 289°	11
14	Excessin	26	122-3°	+ 44	11
15	Epiexcessin	27	164.5-5.5°	+ 131°	12
16	Eigballinol	28	264-6°	-7.1°	12

* The sign of the specific rotation is indicated in cases where only one enantiomer has been isolated.

feature of nmr is that it easily differentiates diequitorial, diaxial and equitorial-axial lignans.

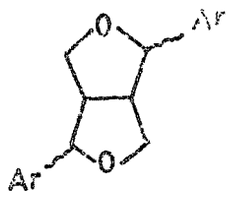
Pelter et al.[8,13] proposed a criterion for establishing the stereochemistry of these compounds, based on the direct field effect of an axial aryl group on the methylene protons of the other heterocyclic ring. The axial aryl group at C-2 for example, is held very close to the axial hydrogen atom at C-8. The axial hydrogen atom at C-8 lies within the shielding cone of the benzene ring and is therefore moved upfield. If on the other hand, there were an equitorial aryl group at C-2, then both the methylene protons at C-8 lie within the deshielding cone of the benzene ring and will be moved downfield. In the diequitorial compounds the methylene protons always resonate between 3.75 and 4.70 ppm, whereas in the diaxial compounds they resonate between 3.25 and 4.00 ppm. The equitorial axial lignans show one signal in each of these regions of the spectrum.

The episeries (equitorial-axial lignans) is anomalous in showing one of the benzylic protons at high field and this feature is shared by all the known epi-compounds. Pelter et al.[14] suggested that this is an important and diagnostic characteristic feature of the episeries and like the C-8 axial proton, it is a result of direct anisotropic field effect of an axial aryl group. This analysis was supported by the fact that the

episeries is the only one in which a benzylic proton is held close to an axial aryl group of the opposite ring. In the axial series, there are no such axial benzylic protons to be influenced, while in the diequitorial series both the aryl groups are equitorial. This indicates that, in the eoiseries the highfield proton is the axial proton and this easily solves the problem of assigning the stereochemistry at C-2 and C-6. This method was tested on the complete eudesmin series[13,14] (15, 16, 17), sesamin series[13] (18, 19, 20) and the gmelinols.[13] These criteria have also been used to assign the configuration of many other compounds including excelsin (26), epiexelsin (27),[11] yangambin (23) and diayangambin (25).[10]

Mass spectra: The mass spectra of 2,6-diaryl-3,7-dioxabicyclo-[3.3.0]octanes were first studied by Pelter[15] and Dulfield,[16] who defined the basic fragmentation pathways, which have been shown to be general and used extensively to assign structures in this field.[1]

For the lignans of type 11 the molecular ion is always intense and the most abundent fragments ions are 29-39 (Fig. 3). A study[1] of the metastable peaks showed clearly that many of the most abundent ions arise by more than one fragmentation



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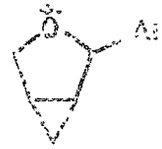

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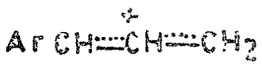

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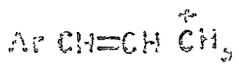

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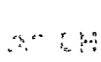

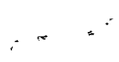

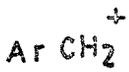

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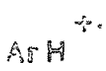

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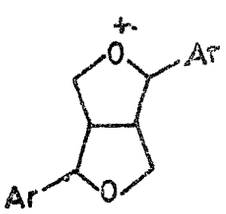

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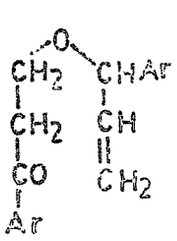

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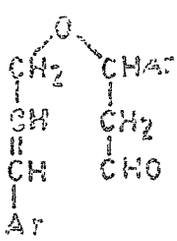

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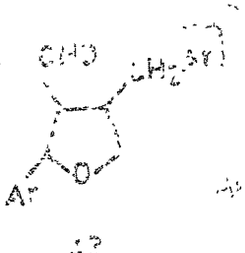

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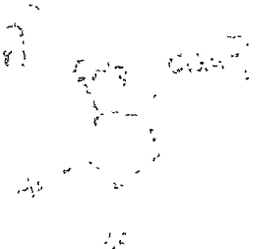

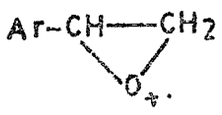

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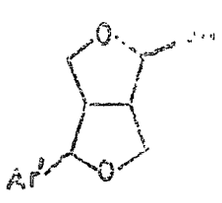


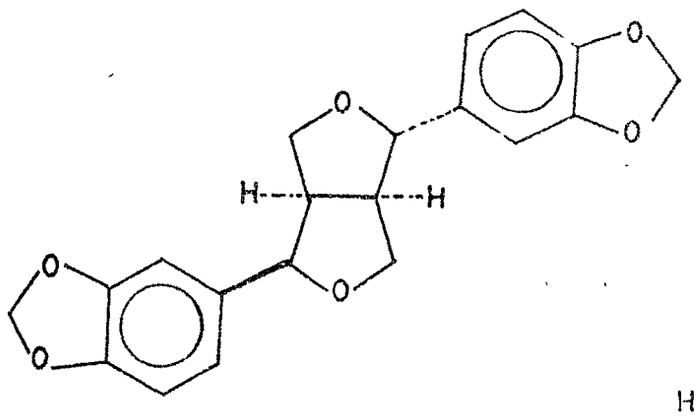
46

Fig. 3

pathways. Pelter[15] attempted to correlate the mass spectra of these compounds with those of simple tetrahydrofurano-lignans and put forward the hypothesis that the molecular ion can exist in various forms (40-44, Fig. 3). For a diagnostic view point, the abundant ion 33 is probably of the most significant. It corresponds to a "vertical cleavage of the molecule and shows the presence of the moiety Ar-CH-CH-CH_2-. Another peak which is seen weakly in this series, but strong in others is due to the ion 45 which is also a precursor of 37 and corresponds to a "horizontal" cleavage. If the two fragmentations are taken together, much structural information may be obtained. Although mass spectrometry is much useful for assigning the gross structures of these compounds, in no case has it been shown to distinguish reliably between stereo-isomers.[17] Ofcourse, for unsymmetrically substituted lignans (46); the mass spectra help to define the aryl groups that are present as well as the nature of the nucleus to which they are attached.[1]

GUGGULLIGNAN-1

Sukh Dev and co-workers[18,19] have isolated four lignans from C. mukul, viz. (+)-sesamin (19), oluviatilol (47), guggullignan-1 (50) and guggullignan-2 (49) (Fig. 4). Guggullignan-1 was tentatively assigned[19] the structure 48



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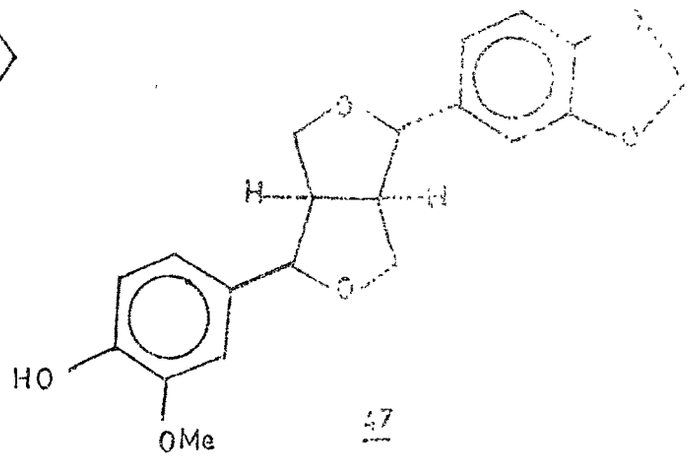

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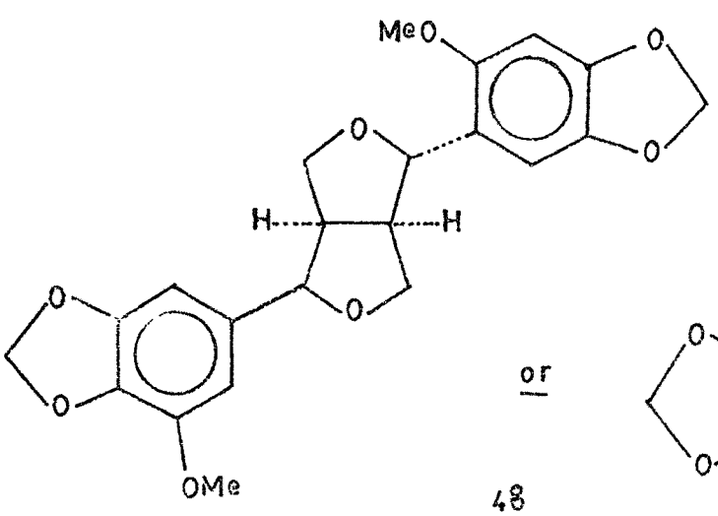
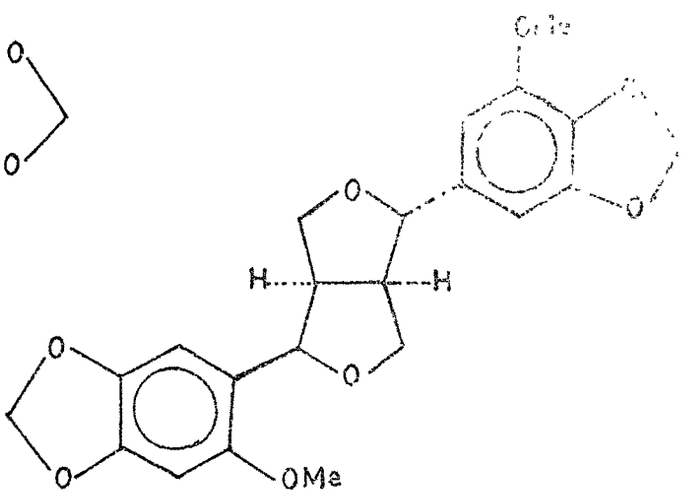


or



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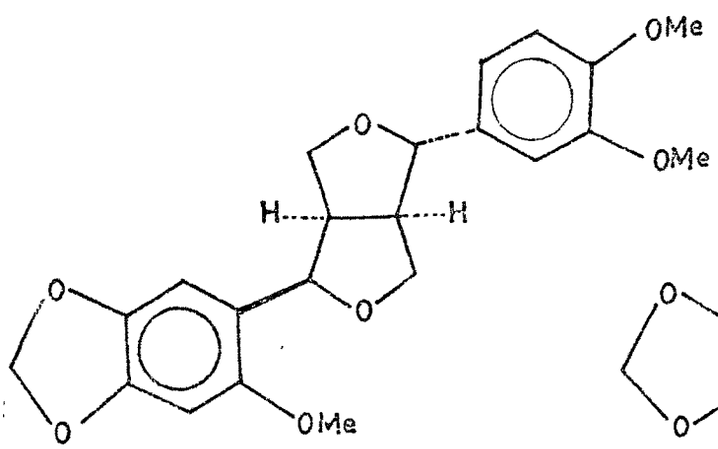

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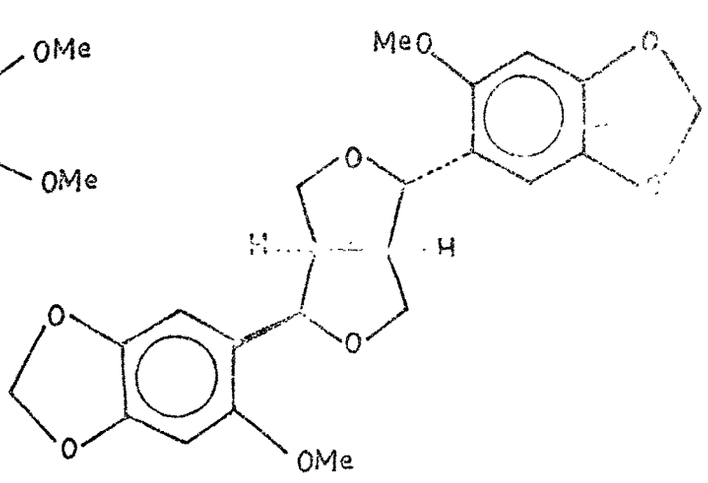


50

Fig. 4

based on the spectral data. Now the structure of guggullignan-1 has been revised as 50 based on high resolution PMR.

Previous structural assignment

Previously [19] structure 48 was assigned to guggullignan-1 based on the following spectral characteristics.

Guggullignan-1 analysed for $C_{22}H_{22}O_8$ (M^+, m/e 414). The PMR (90 MHz) spectrum (Fig. 8) indicated this compound to be a lignan of the fused bis-tetrahydrofuran series, and aliphatic protons region was suggestive of an epi-eudesmin (16) type structure (Table 2). Irradiation of the methine signal at 2.91 ppm caused the doublet at 4.41 ppm to collapse to a singlet, while irradiation at 3.33 ppm caused the doublet at 4.82 ppm to collapse to a singlet. This indicated that methine hydrogen at C-1 and C-5 in guggullignan-1 are non-equivalent. Of the four methylene protons, one proton appeared as a multiplet between 4.05-4.21, one between 3.22-3.45 ppm and two protons resonated as a multiplet between 3.76-3.87 ppm. These chemical shifts are in consistent with those of epi-eudesmin and other epi-compounds. Out of two benzylic protons, one appeared in the down-field (4.82 ppm, J = 5 Hz) and the other in the up-field (4.41 ppm, J = 7Hz). This is a characteristic feature of the episeries. Furthermore, the pmr spectrum indicated that guggullignan-1 has two methoxy groups (6H, s, 3.92 ppm), four methylene dioxy protons (4H, s, 5.97 ppm). Four aromatic protons

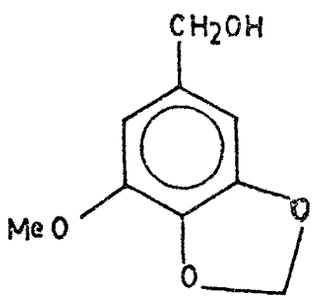
Table II - PMR SPECTRA OF EPIEUDESMIN AND GUGGULLIGNAN-I(50)

Protons	Epieudesmin (16)	Guggullignan (50)
1H	3.35, m	3.3, m
2H	4.85, d, J= 5.5Hz	4.82, d, J = 5Hz
4 H	4.1- 4.4, m	4.05-4.21, m
4 H	3.7-3.9, m	3.76-3.87 m
5H	2.9, m	2.91, m
6H	4.45, d, J = 7Hz	4.41, d, J = 7Hz
8 H	3.70-3.9, m	3.76-3.87, m
8 H	3.25-3.45, m	3.22-3.45, m
OCH_3	3.86, 3.90, s	3.92, s (6H)
Aromatic	6.9-7.1, m,	6.53, 6.58, 6.56, s, (4H)
O_2CH_2	-	5.97,s(4H).

appeared as two one-proton singlet at 6.53 ppm and 6.58 ppm and one two-proton singlet at 6.56 ppm. Mass spectrum revealed both vertical and horizontal cleavage, a characteristic feature of lignans. Further, it indicated that each aromatic ring has one methylene dioxy group and one methoxy group.

Substitution pattern on the aromatic ring was determined by comparative study of the aromatic region in the nmr spectra of epi-exelsin (27),[11] sesangolin (51)[20], 3,4-methylenedioxy-5-methoxy benzylalcohol (52), 3,4-dimethoxybenzyl-alcohol (53), 4-hydroxy-3-methoxybenzyl-alcohol (54), and 2-methoxy-3,4-methylenedioxy benzylalcohol (55, Fig. 5). The four aromatic protons of epi-excelsin (27) appear as a singlet at 6.5 ppm, as do the aromatic protons of 52 which resonate as a singlet at 6.5 ppm. The two para-protons of sesangolin[20] appear as singlets at 6.43 ppm and 6.84 ppm. The protons of the unsubstituted methylenedioxyphenyl ring in these lignans resonate as a multiplet between 6.7-6.76 ppm, as do the aromatic protons of 53 and 54 which resonate as a multiplet between 6.81-6.9 and 6.8-6.98 ppm respectively. In case of 55 the aromatic protons resonate as an AB quartet centered at 6.6 ppm.

Guggullignan-1 revealed two one-proton singlets at 6.53 ppm and 6.58 ppm, which indicated that the two aromatic



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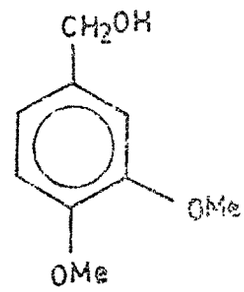

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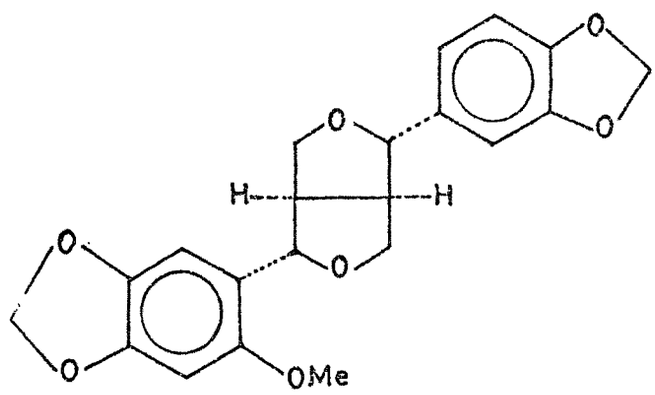

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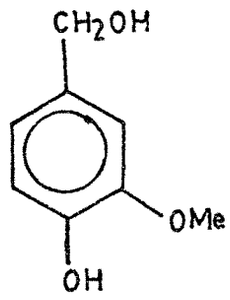

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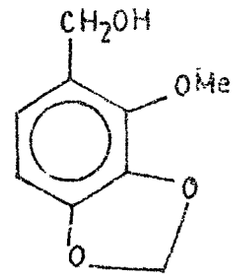


55

Fig. 5

protons are para with respect to each other, and a two proton singlet at 6.56 ppm which was assigned to two meta protons of the other aromatic ring. From this, it was tentatively concluded that, out of four aromatic protons, two were para with respect to each other (resonating at 6.53 and 6.58 ppm), and the other two were meta with respect to each other, resonating between the para-protons (6.56 ppm), thereby giving an appearance of an ill-defined multiplet (Fig. 8). Therefore guggullignan-1 was tentatively assigned the structure 48, leaving the stereochemistry at C-2 and C-6 unclarified.

PRESENT WORK

The structure of guggullignan-1 was revised as 50 based on the high-field pmr (500 MHz).

In guggullignan-1, the aromatic protons appear as an illdefined multiplet centered at 6.56 ppm, which has been defined[19] as a two one-proton singlets at 6.53 ppm and 6.58 ppm (para protons) and a two-proton singlet at 6.56 ppm (metaprotons). Based on this interpretation, structure 48 was assigned. Since pmr spectrum of guggulignan-1 was recorded using a 90 MHz pmr spectrometer, the signals for aromatic protons were not resolved because of their narrow range of chemical shifts.

Hence the interpretation of the illdefined multiplet[19] in guggullignan-1 was less secured and a reclarification was needed.

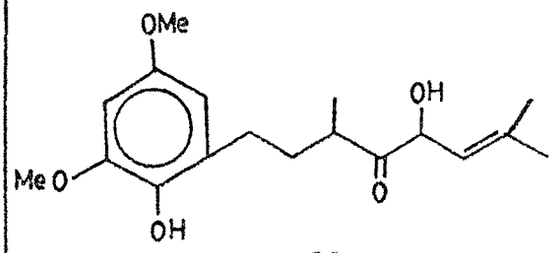
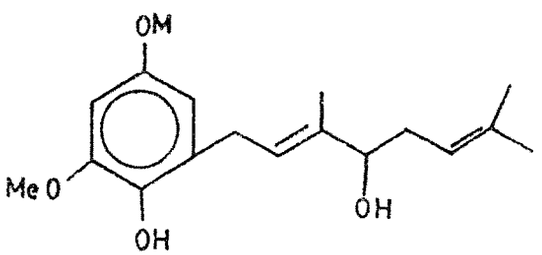
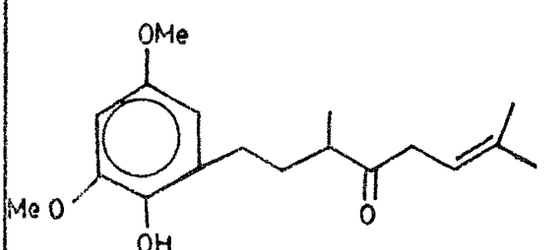
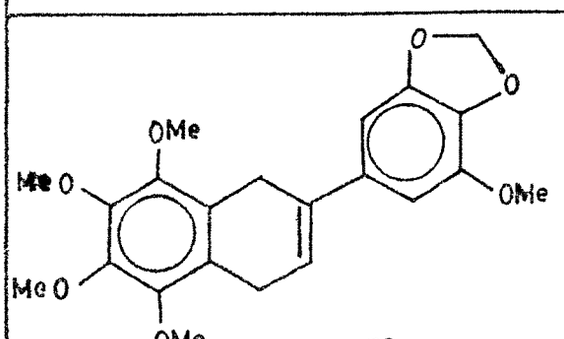
The meta-protons resonate as two doublets with coupling constants 2Hz, if they are in a different chemical environment. However, para-protons resonate as a two-singlet and ortho-protons resonate as AB quartet.[21] This behaviour of meta- and para-protons (which seems to be present in guggullignan-1) was examined in the high-field nmr of a number of natural products, having similar aromatic ring substituents, as that of guggullignan-1 (Fig. 6 and 7). The meta-protons of compound 56 (Fig. 6) resonate as two doublets at 6.52 ppm and 6.56 ppm (J = 2.9 Hz), meta-protons of the similar compounds 57 and 58 resonate as two doublets at 6.53 ppm, 6.56 ppm (J = 2.9 Hz) and 6.52 ppm, 6.56 ppm (J = 2.9 Hz) respectively. These protons in compound 59 resonate at 7.14 ppm and 6.65 ppm as doublets (J = 1.5 Hz). The meta-protons of 60 appear as two doublets at 6.56 ppm and 6.65 ppm (J = 1.5 Hz). Compound 61 showed the similar spectral behaviour. The meta-protons of 62 and 63 resonate as doublets at 6.32 ppm, 6.39 ppm (J = 1.2 Hz) at 6.65 ppm, 7.17 ppm (J = 2.5 Hz) respectively.

The para-protons of the unsymmetrically substituted ring resonate as singlets. This feature was examined on many

natural products in high resolution pmr (Fig. 7). In all the cases para-protons appeared as singlets which was quite evident from Fig. 7.

From the above studies, it can be concluded that the aromatic meta-protons resonate as doublets (J= 2.5 Hz), if they are in a different chemical environment, and the para-protons resonate as singlets.

In the 500 MHz pmr of guggullignan-1, the four aromatic protons resonate as four sharp singlets at 6.579 ppm, 6.556 ppm, 6.540 ppm and 6.506 ppm. This clearly indicates that in guggullignan-1, all the four protons are para-ones, but not two-para and two-meta, as assigned earlier.[19] Hence, the structure of guggullignan-1 is 50. If the earlier structure 48 was correct, then one would have expected two singlets and two doublets. The chemical shift assignment[19] for other aliphatic protons (Table 2) were confirmed by decoupling experiments (Fig. 10). Since guggullignan-1 is a symmetrical molecule, the stereochemical problem at C-2 and C-6 does not arise.

Compound	Magnetic field (MHz)	Chemical shift δ (ppm)
 56	360	2 d 6.52 6.56 J = 2.9 Hz
 57	360	2 d 6.53 6.56 J = 2.9 Hz
 58	360	2 d 6.52 6.56 J = 2.9
 59	270	2 d 7.14 6.65 J = 1.5 Hz

δ Values given are for meta-protons.

Fig. 6 (contd.)

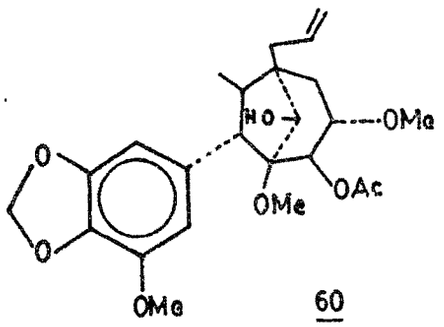
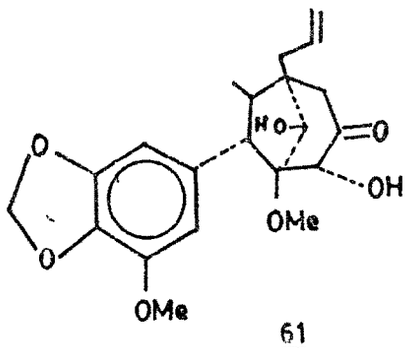
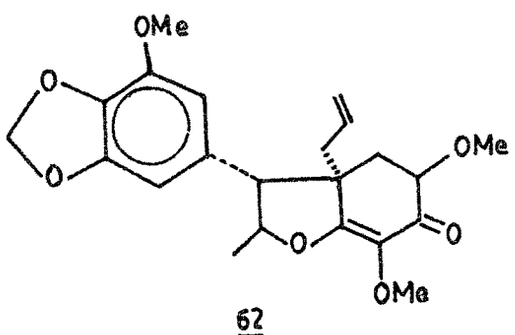
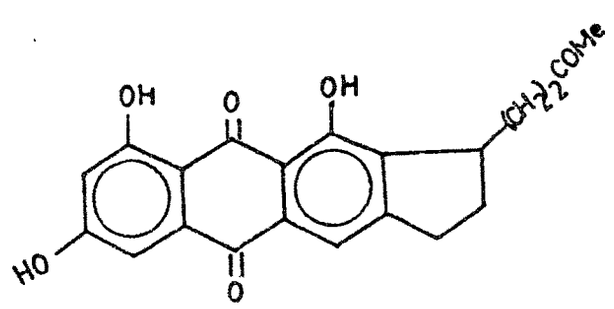
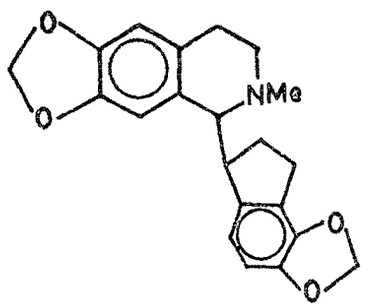
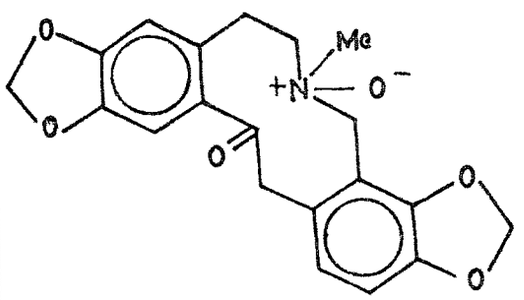
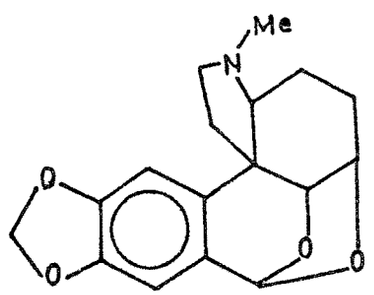
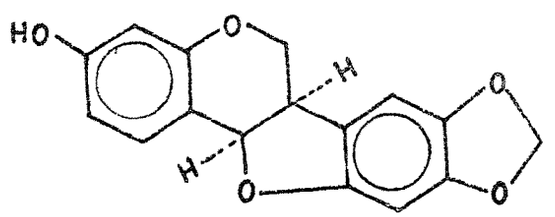
Compound	Magnetic field (MHz)	Chemical shift, δ (ppm)	Ref.
 60	270	2 d 6.56 6.65 J=1.5Hz	24
 61	100	2 d 6.55 6.73 J=1.2 Hz	
 62	270	2 d 6.32 6.39 J=1.5 Hz	25
 63	250	2 d 6.65 7.17 J= 1.5	26

Fig. 6

Compound	Magnetic field (MHz)	Chemical shift, δ (ppm)	Ref.
 64	200	2 s 6.7 6.36	
 65	200	2s 7.13 6.77	
 66	300	2s 6.8 6.55	
 67	250	2s 6.89 6.40	

δ Values given are for para-protons

Fig. 7 (contd.)

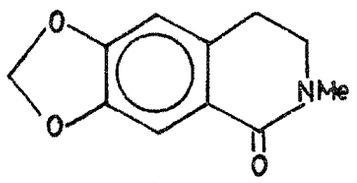
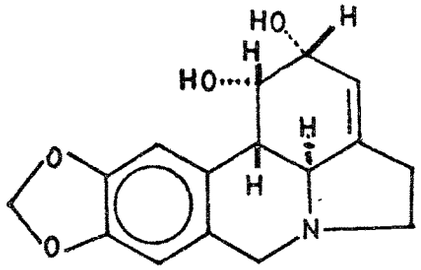
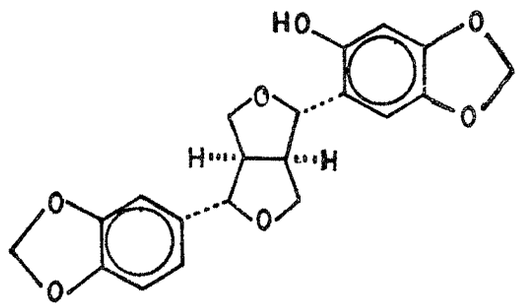
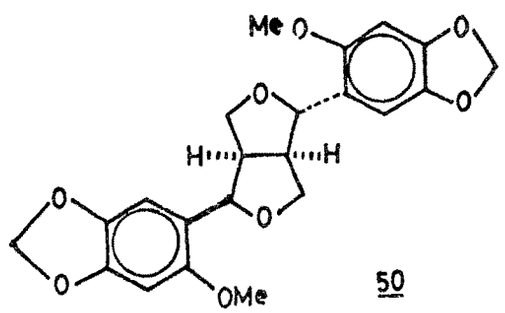
Compound	Magnetic field (MHz)	Chemical shift, δ (ppm)	Ref.
 68	200	2s, 6.61 7.54	31
 69	270	2s, 6.80 6.98	32
 70	90	2s, 6.80 6.42	33
 50	500	4s, 6.579 6.556 6.540 6.506	

Fig. 7

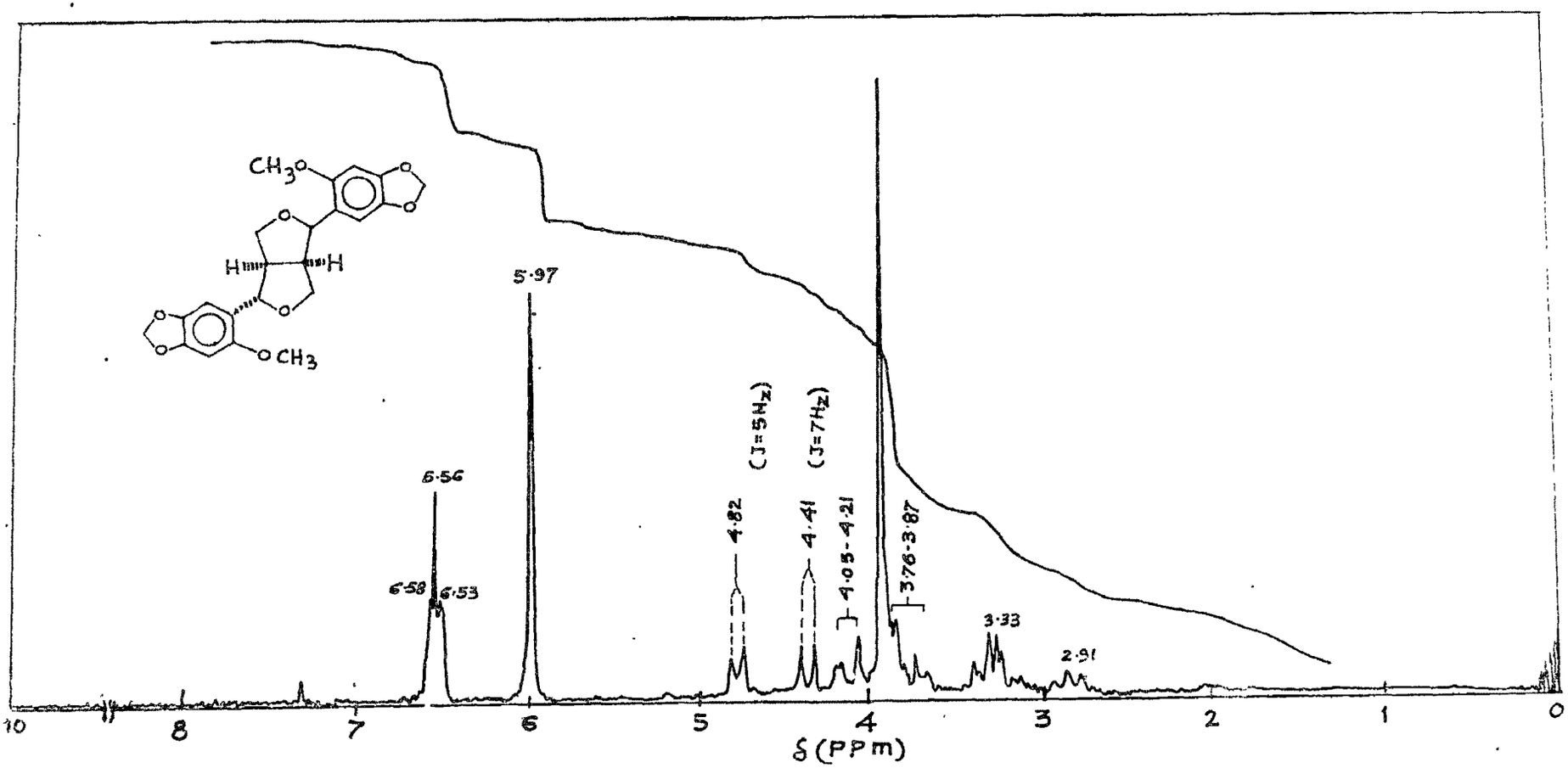


Fig. 8 - PMR (90 MHz) Spectrum of guggulilignan-1

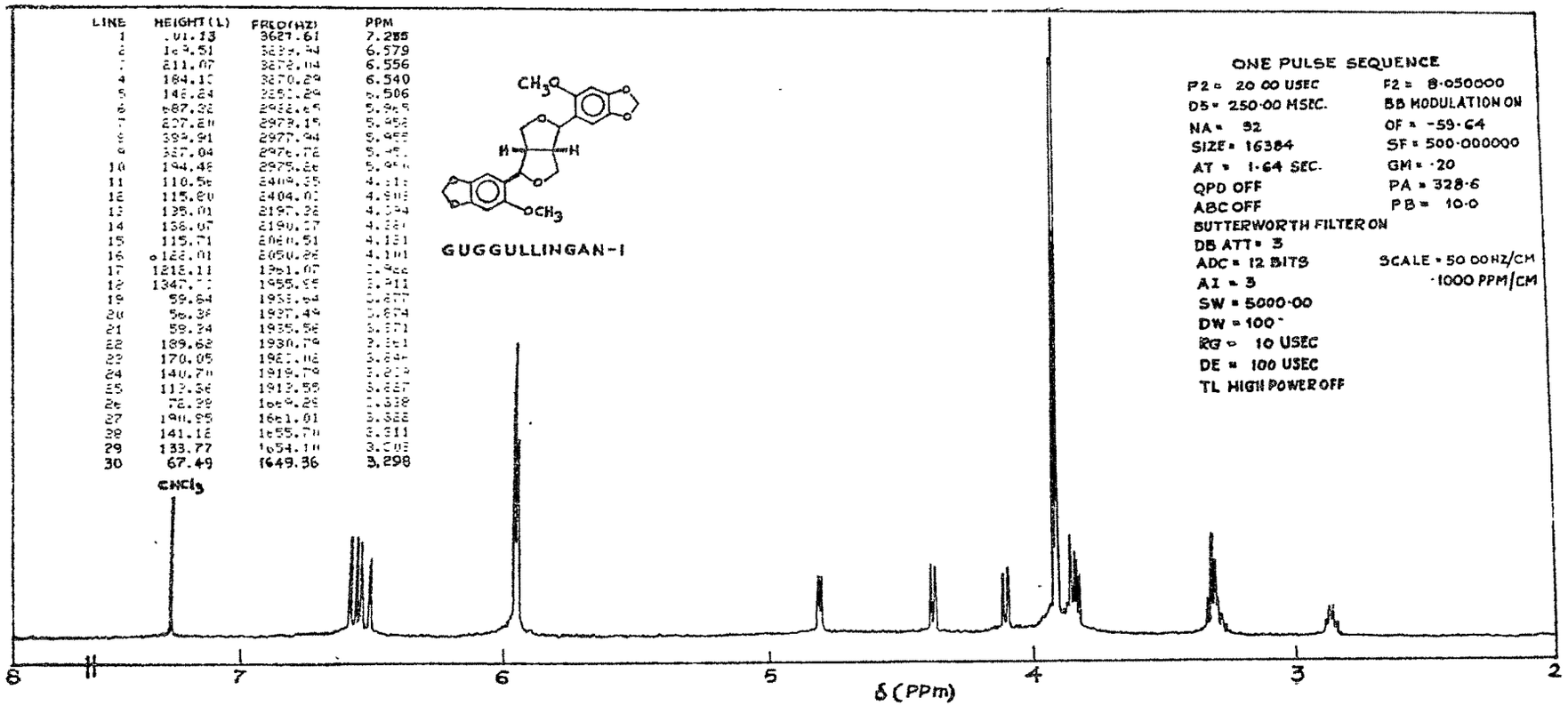


Fig. 9 - PMR (500 MHz) spectrum of guggulilignan-1

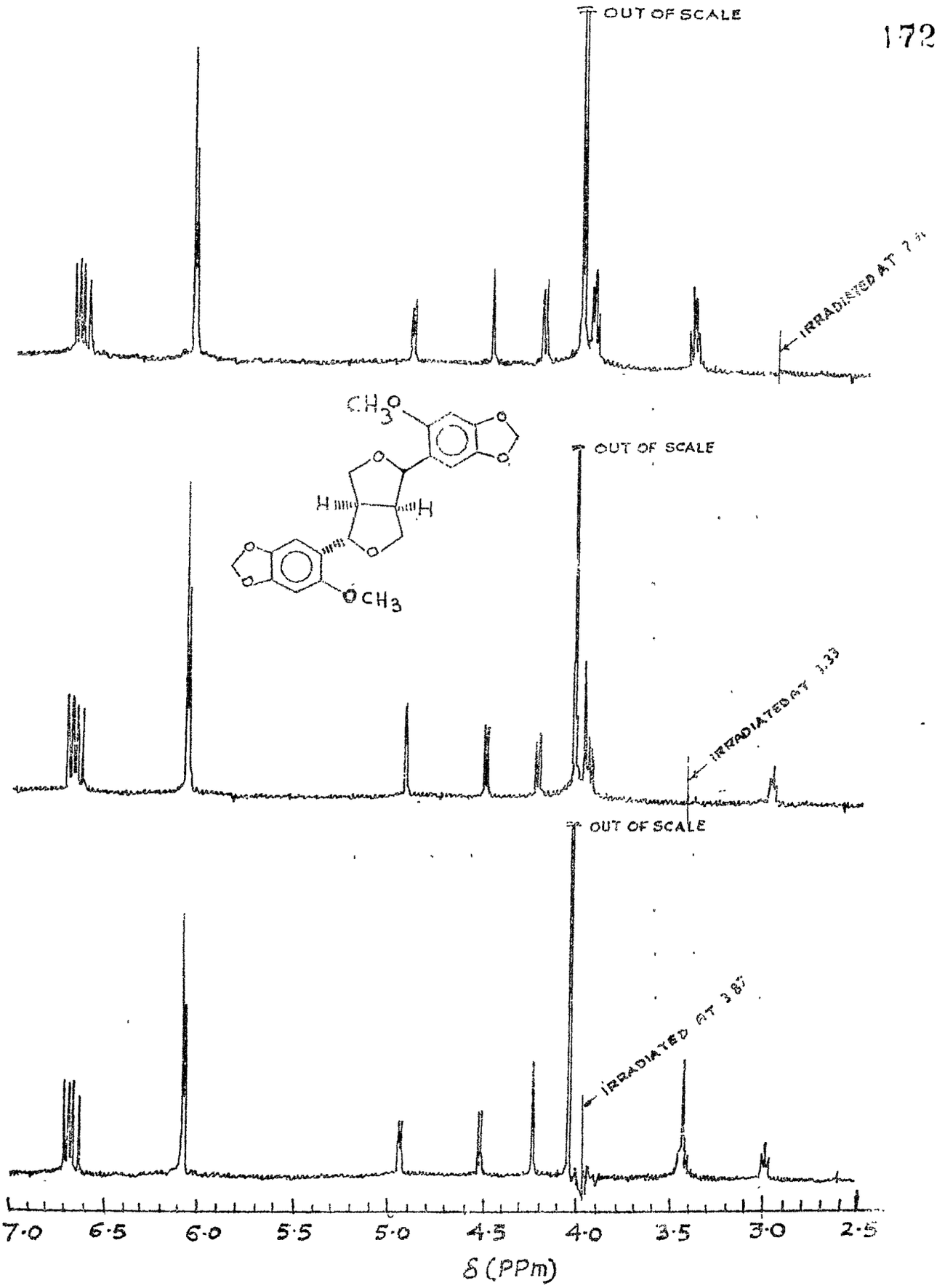


Fig. 10 - Decoupled PMR (500 MHz) spectra of guggullignan-1

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