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NINE ISOFLAVONES FROM *TEPHROSIA MAXIMA*

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Key Word Index—*Tephrosia maxima*, Leguminosae, maxima isoflavones A, C, D, E, F and G, ^{1}H and ^{13}C NMR spectra

Abstract—The isolation and characterisation of nine closely related 3',4'-disubstituted isoflavones from the aerial parts and roots of *Tephrosia maxima* are reported Four of these compounds are new natural products, one of which contains a 7,8-methylenedioxy group, an uncommon feature in naturally occurring isoflavones ^{1}H and ^{13}C NMR data are given for all compounds

INTRODUCTION

As part of our continuing study of *Tephrosia* species [1, 2] we have undertaken a reexamination of the chemical constituents of the aerial parts and roots of *Tephrosia maxima* Pers We report here the isolation of four new isoflavones (**4**, **8**, **9** and **10**), besides the previously reported maxima isoflavones A (**1**) and C (**3**) [3–6]. Their structures have been established on the basis of spectroscopic and chemical evidence In addition we have isolated and characterised 7,6'-dimethoxy-3',4'-methylenedioxyisoflavone (**5**), 7,8,6'-trimethoxy-3',4'-methylenedioxyisoflavone (**6**), and 6-methoxy-7-hydroxy-3',4'-methylenedioxyisoflavone (7) [7–10]

RESULTS AND DISCUSSION

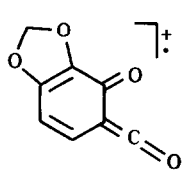
Chromatography of the petrol extract of the aerial parts of *T maxima* gave maxima isoflavones A and C, while chromatography of the chloroform extracts of the aerial parts and roots gave four new isoflavones which we designate as maxima isoflavones D, E, F and G Although the structures of maxima isoflavones A (**1**) and C (**3**) have been established on the basis of chemical evidence [4–6] no spectral data is available in the literature, nor authentic samples available for comparison Since the new isoflavones are closely related to them spectral evidence for the structures of maxima isoflavones A and C is briefly presented

The mp and UV spectrum of maxima isoflavone A (**1**) were in close accord with those reported in the literature [5] In the mass spectrum the molecular ion was observed at m/z 310 ($C_{17}H_{16}O_6$) and other significant fragments at m/z 164 and 146 could be rationalized in terms of the known fragmentation pattern of isoflavones [11] Together with the ions at m/z 165 and 135 in the mass spectrum of the corresponding deoxybenzoin (**11**), they established the presence of methylenedioxy substituents in both rings A and B

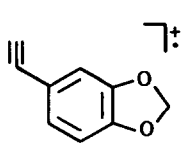
The fact that the 5 and 6 positions of **1** are unsubstituted was immediately evident from the ^{1}H NMR spectrum (Table 1) which shows an *ortho* coupled doublet (J = 9 Hz) at δ7 68 due to H-5 As shown in Table 2 the ^{13}C NMR spectrum also fully supports the proposed structure [12] The structure was further confirmed by double demethylenation followed by methylation to give 7,8,3',4'-tetramethoxyisoflavone (**2**) [5]

The structure proposed earlier for maxima isoflavone C (**3**) [6] is fully supported by its ^{1}H and ^{13}C NMR spectra (Tables 1 and 2), and by those of the corresponding deoxybenzoin (**12**) [13] (Tables 3 and 4) The mass spectrum of **3** showed a molecular ion at m/z 380 ($C_{22}H_{20}O_6$) in addition to a prominent fragment ion at m/z 312 corresponding to loss of C_5H_8 Another prominent ion at m/z 281, due to further loss of an OMe group is a characteristic feature observed for all compounds having a 6'-methoxyl group on ring B (*vide infra*)

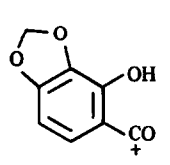
The first new compound, maxima isoflavone D (**4**), mp 223–224°, showed λ_{max} 262 nm and ν_{max} 1632 cm^{-1} typical of an isoflavone The ^{1}H NMR spectrum (Table 1) showed a singlet at δ8 24 confirming the isoflavone nucleus The sharp singlets at δ3 72 (6H) and δ6 19 (2H) indicated the presence of two methoxy and one methylenedioxy group Two doublets at δ7 64 and



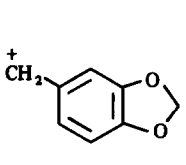
m/z 164



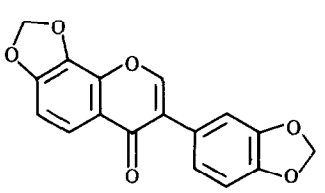
m/z 146



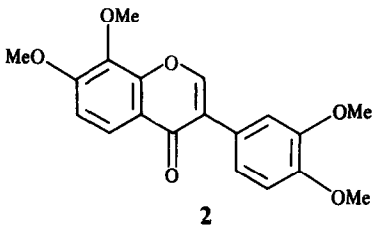
m/z 165



m/z 135



1



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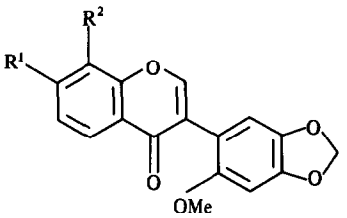
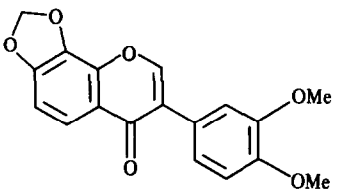
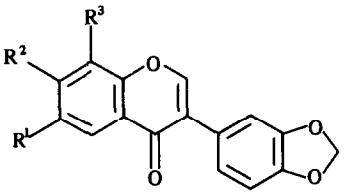


3 R^1=O—CH_2—CH=C(Me)(Me), R^2=H



4

5 R^1=OMe, R^2=H
6 R^1=R^2=OMe
9 R^1=OH, R^2=OMe
9a R^1=OAc, R^2=OMe
10 R^1=OH, R^2=H
10a R^1=OAc, R^2=H

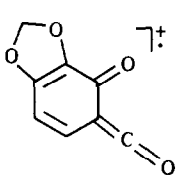


7 R=OMe, R^2=OH, R^3=H
8 R^1=H, R^2=OH, R^3=OMe
8a R^1=H, R^2=OAc, R^3=OMe
8b R^1=H, R^2=R^3=OMe

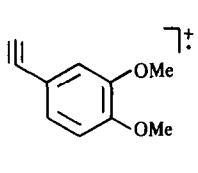
7 01 (J = 9 Hz) could be assigned to H-5 and H-6 respectively and the remaining three protons of the B ring were observed as a multiplet at δ6 8–7 2

That the methylenedioxy group and two methoxy groups were attached to carbons 7,8 and 3',4' in rings A and B was proven by demethylenation followed by remethylation to give 7,8,3',4'-tetramethoxyisoflavone (**2**) [5] In the mass spectrum of **4** the molecular ion was observed at m/z 326 ($C_{18}H_{14}O_6$) and the formation of significant fragment ions at m/z 162 and 164 was also consistent with the proposed structure Along with ions at m/z 151 and 165 in the mass spectrum of the corresponding deoxybenzoin (**14**), they confirm beyond doubt that the two methoxy groups are attached to ring B and the methylenedioxy group to ring A

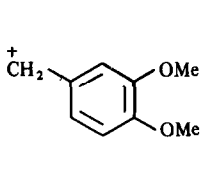
The ^{13}C NMR spectrum of **4** closely resembles that of maxima isoflavone A (**1**) (see Table 2), particularly with regard to the chemical shifts of the ring A carbon atoms The NMR and mass spectra of the product obtained by H_2O_2–KOH cleavage show it to be a *ca* 1 1 mixture of



m/z 164



m/z 162



m/z 151

Table 1 ^{1}H NMR spectra of isoflavones

Compound	Solvent	2′	5′	6′	2	5	6	8	OCH_2O	OMe	OH	OAc	O⌒
1	DMSO	6 9–7 1	*m*		8 36 *s*	7 68 *d*(9)	—	—	6 04 *s* 6 29 *s*	—	—	—	—
2	DMSO	7 0–7 2	*m*		8 48 *s*	7 88 *d*(8)	—	—	—	3 79 *s*(×2) 3 89 *s* 3 97 *s*	—	—	—
	$CDCl_3$	7 21 *d*(2)	7 06 *dd*(8,2)	6 92 *d*(8)	8 02 *s*	8 04 *s*	7 06 *d*(9)	—	—	3 92 *s* 3 93 *s* 4 01 *s*(×2)	—	—	—
3	DMSO	6 84 *s*	6 80 *s*	—	8 16 *s*	7 86 *d*(8)	7 02 *dd*(8,2)	7 09 *d*(2)	5 98 *s*	3 64 *s*	—	—	1 74 *s*, 4 65 *d*, 5 45 *m*
	$CDCl_3$	6 80*s*	6 58 *s*	—	7 85 *s*	8 15 *d*(9)	6 92 *dd*(9,2)	6 82 *d*(2)	5 90 *s*	3 68 *s*	—	—	1 76 *s*, 1 80 *s*, 4 58 *d*, 5 48 *t*
4	DMSO	6 8–7 2	*m*		8 24 *s*	7 64 *d*(9)	7 01 *d*(9)	—	6 19 *s*	3 72 *s*	—	—	—
	$CDCl_3$	7 18 *d*(2)	7 04 *dd*(8,2)	6 91 (8)	7 93 *s*	7 90 *d*(9)	6 98 *d*(9)	—	6 21 *s*	3 92 *s* 3 93 *s*	—	—	—
5	DMSO	6 86 *s*	6 82 *s*	—	8 12 *s*	7 98 *d*(9)	7 07 *dd*(9,2)	7 13 *d*(2)	6 00 *s*	3 66 *s* 3 91 *s*	—	—	—
	$CDCl_3$	6 83 *s*	6 62 *s*	—	7 88 *s*	8 18 *d*(9)	6 97 *dd*(9,2)	6 84 *d*(2)	5 95 *s*	3 73 *s* 3 91 *s*	—	—	—
6	DMSO	6 87 *s*	6 82 *s*	—	8 25 *s*	7 81 *d*(9)	7 27 *d*(9)	—	6 01 *s*	3 66 *s* 3 88 *s* 3 96 *s*	—	—	—
	$CDCl_3$	6 83 *s*	6 62 *s*	—	7 96 *s*	8 01 *d*(9)	7 04 *d*(9)	—	5 96 *s*	3 73 *s* 4 00 *s* 4 02 *s*	—	—	—
7	DMSO	6 8–7 2	*m*		8 30 *s*	7 44 *s*	—	6 94 *s*	6 04 *s*	3 88 *s*	—	—	—
8	DMSO		6 9–7 2 *m*		8 41 *s*	7 73 *d*(9)	7 02 *d*(9)	—	6 04 *s*	3 88 *s*	10 5 *br*	—	—
	$CDCl_3$	7 09 *d*(2)	6 99 *dd*(8,1)	6 85 *d*(8)	7 95 *s*	7 98 *d*(9)	7 05 *d*(9)	—	5 99 *s*	4 08 *s*	6 17 *s*	—	—
8a	$CDCl_3$		6 8–7 1 *m*		8 00 *s*	8 04 *d*(9)	7 19 (9)	—	5 96 *s*	3 98 *s*	—	2 35 *s*	—
8b	DMSO		6 9–7 2 *m*		8 44 *s*	7 86 *d*(9)	7 28 *d*(9)	—	6 04 *s*	3 88 *s* 3 96 *s*	—	—	—
9	DMSO	6 86 *s*	6 82 *s*	—	8 22 *s*	7 68 *d*(9)	7 00 *d*(9)	—	6 00 *s*	3 66 *s* 3 87 *s*	—	—	—
	$CDCl_3$	6 82 *s*	6 62 *s*	—	7 93 *s*	7 95 *d*(9)	7 03 *d*(9)	—	5 95 *s*	3 73 *s* 4 08 *s*	6 24 *s*	—	—
9a	$CDCl_3$	6 62 *s*	6 72 *s*	—	8 00 *s*	8 02 *d*(9)	7 10 *d*(9)	—	5 94 *s*	3 72 *s* 4 00 *s*	—	2 38 *s*	—
10	DMSO	6 79 *s*	6 74 *s*	—	8 04 *s*	7 86 *d*(9)	6 86 *dd*(9,2)	6 78 *m*	5 94 *s*	3 59 *s*	6 80 *s*	—	—
10a	$CDCl_3$	6 76 *s*	6 56 *s*	—	7 87 *s*	8 24 *d*(9)	7 08 *dd*(9,2)	7 22 *d*(2)	5 89 *s*	3 68 *s*	—	2 32 *s*	—

Table 2 ^{13}C NMR spectra of isoflavones

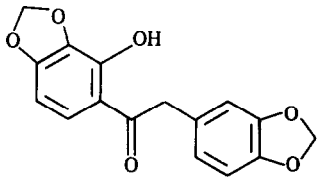
Compound	Solvent	1′	2′	3′	4′	5′	6′	2	3	4	4a	5	6	7	8	8a	OMe OAc	OCH_2O
1	DMSO	125 26	109 42	146 96	146 96	108 01	122 46	153 24	122 99	174 19	119 68	119 95	107 34	151 99	134 30	140 34		101 00
2	DMSO	124 18	112 71	148 26	148 61	111 57	121 25	153 63	123 01	174 92	118 36	120 85	110 95	156 09		149 79	55 49 (× 2)	103 64
																	56 44	
																	60 95	
3	DMSO	112 75	110 96	140 39	147 91	95 51	152 81	154 53	121 66	174 29	117 47	126 79	115 00	162 81	101 21	157 40	56 56	101 20
	$CDCl_3$	112 97	111 26	141 24	148 40	95 51	153 00	154 11	122 11	174 64	118 39	127 72	114 83	163 16	100 96	157 93	56 90	101 37
4	DMSO	124 08	112 87	148 26	148 70	111 56	121 33	153 15	123 09	174 29	119 76	119 96	107 32	151 94	134 29	140 40	55 51	103 62
5	DMSO	113 38	111 08	140 99	148 20	96 08	153 16	154 40	122 0		118 17	127 17	114 41	164 00	100 87	157 78	56 09	101 33
																	57 06	
6	DMSO	113 17	110 97	140 75	148 08	96 00	153 12	154 50	121 44	174 63	118 85	120 69	111 35	156 21	136 78	150 15	56 76	101 26
																	56 97	
																	61 02	
7	DMSO	125 91	109 37	146 93	146 93	108 03	122 28	152 95	122 63	174 16	116 13	104 70	146 80	151 70	102 72	152 83	55 79	100 97
8	DMSO	125 59	109 39	146 94	146 94	108 05	122 37	153 27	122 96	174 65	117 36	120 75	115 15	154 65	134 61	150 59	60 77	100 99
8a	$CDCl_3$	125 28	109 74	147 83	147 93	108 49	122 48	152 21	123 78	175 5	125 13	120 19	121 13	147 03	140 9	150 6	61 74	101 26
																	168 33	20 69
8b	DMSO	125 47	109 35	146 97	146 97	108 04	122 36	153 70	122 92	174 67	118 40	120 81	110 94	156 14	136 09	149 83	56 45	101 01
																	60 91	
9	DMSO	112 81	110 97	140 34	147 88	95 50	152 86	154 11	121 34	174 40	117 37	120 63	115 14	154 70	134 78	150 69	56 56	101 16
																	60 74	
9a	$CDCl_3$	112 27	111 10	141 31	148 68	95 53	153 05	154 15	122 49	175 53	123 86	119 99	121 13	150 74	141 31	146 89	56 89	101 46
																	61 73	
																	168 39	20 70
10	DMSO	112 88	110 98	140 33	147 83	95 50	152 80	154 23	121 46	174 29	116 57	127 15	115 05	162 48	102 13	157 46	56 53	101 12
10a	$CDCl_3$	112 22	110 89	140 87	148 31	95 28	152 75	154 29	122 01	174 98	121 96	127 15	119 41	156 40	111 00	154 85	56 63	101 25
																	168 25	20 96

Table 3 ^{1}H NMR spectra of deoxybenzoins

Compound	Solvent	2′	5′	6′	CH_2	6	5	3	OCH_2O	OMe	OH	O⌒		
11	$CDCl_3$		6 70 *m*		4 12 *s*	7 46 *d*(8)	6 44 *d*(8)	—	5 92 *s* 6 04 *s*	—	11 4 *s*		—	
12	$CDCl_3$	6 62 *s*	6 48 *s*	—	4 09 *s*	7 72 *d*(9)	6 38 *m*		5 83 *s*	3 70 *s*	12 60 *s*	5 40 *t*	4 48 *d*	1 72 *s* 1 77 *s*
13	$CDCl_3$	6 66 *s*	6 51 *s*	—	4 08 *s*	7 78 *d*(9)	6 38 *m*		5 85 *s*	3 70 *s* 3 78 *s*	12 62 *s*			
14	$CDCl_3$		6 79 *m*		4 18 *s*	7 50 *d*(8)	6 45 *d*(8)	—	6 04 *s*	3 88 *s* (× 2)	11 3 *s*			
15	$CDCl_3$	6 65 *s*	6 51 *s*	—	4 09 *s*	7 78 *d*(9)	6 30 *m*		5 85 *s*	3 70 *s*	12 45 *s*		—	

Table 4 ^{13}C NMR spectra of deoxybenzoins

Compound	Solvent	1′	2′	3′	4′	5′	6′	CH_2	CO	1	6	5	4	3	2	OMe	OCH_2O
11	$CDCl_2$	127 67	108 50	146 81	147 98	109 75	122 48	44 81	202 87	116 20	126 45	100 93	154 13	134 70	147 49	—	101 12
13	$CDCl_3$	113 38	110 48	141 18	147 44	94 92	152 19	38 88	202 54	115 06	131 96	100 96	166 04	107 56	165 56	55 52 56 48	102 66 101 22
14	$CDCl_3$	126 59	111 53	148 32	149 22	112 55	121 55	44 84	203 08	116 25	126 50	100 89	154 11	134 70	147 50	55 92	102 65
15	$CDCl_3$	113 52	110 51	141 19	147 50	94 99	152 23	38 96	203 03	114 85	132 75	103 50	165 19	108 03	163 10	56 51	101 23



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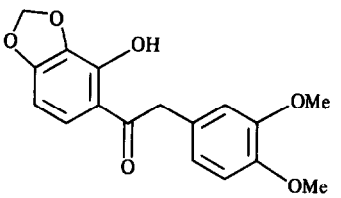

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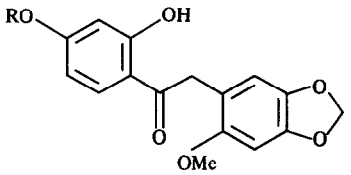


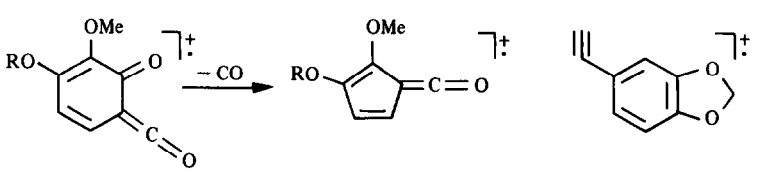
12 R = $CH_2CH{=}CMe_2$
13 R = Me
15 R = H

veratric and 2-hydroxypiperonylic acids, as would be expected The 7,8-methylenedioxy group is an unusual feature in the isoflavone series and maxima isoflavone D is only the second naturally occurring compound to possess this feature, the first being maxima isoflavone A [5]

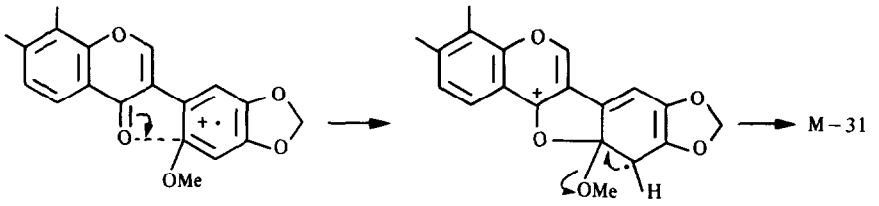
The second new compound, maxima isoflavone E (**8**), mp 268–270°, showed λ_{max} 256 nm and ν_{max} 1635 cm^{-1} indicative of an isoflavone The ^1H NMR spectrum showed singlets at δ8 41 (H-2), 3 88 (OMe) and 6 04 (OCH_2O) The two doublets at δ7 73 and 7 02 (J = 9 Hz) can be assigned to H-5 and H-6 and the multiplet at δ6 9–7 2 is attributed to the three protons of the B ring Maxima isoflavone E contains a hydroxyl group as evidenced by the formation of a monoacetate (**8a**), mp 165–167 5°, and a monomethyl ether (**8b**), mp 182–183° The methyl ether is isomeric with maxima isoflavone D (**4**) and is identical with purpuranin A [14] isolated from the pods of *Tephrosia purpurea* var *maxima*, although no direct comparison was possible

The mass spectrum of maxima isoflavone E showed a molecular ion at m/z 312 ($C_{17}H_{12}O_6$) The presence of ions at m/z 146, 166 and 138 for the parent compound and at m/z 146 and 152 for the methyl ether are again diagnostic of the substituents present in the two halves of the molecule The ^1H NMR spectrum is also indicative of substitution of C-7 and C-8 and the UV shifts ($\lambda_{max}^{MeOH+NaOAc}$ 269 nm) leave no doubt about the orientation of the hydroxyl and methoxyl groups [15] The ^{13}C NMR spectrum is consistent with the 7,8-substitution pattern and the shifts for ring A of the methyl ether (**8b**) compare closely with those of maxima isoflavone F (**9**) and 7,8,6′-trimethoxy-3′,4′-methylenedioxyisoflavone (**6**) (see Table 2) It is however clear that the presence of methoxy groups at C-7 and C-8 cause significant differences in the chemical shifts of C-6, C-7 and C-8 as compared with those when a methylenedioxy group is present, perhaps reflecting poorer overlap of the lone pairs on oxygen with the ring due to free rotation of the methoxyl groups

The third new compound, maxima isoflavone F (**9**), mp 257–259°, showed λ_{max} 254 nm and ν_{max} 1622 cm^{-1} characteristic of an isoflavone, and also showed a characteristic singlet at δ8 22 (H-2) in its ^1H NMR spectrum (Table 1) The sharp singlets at δ3 66, 3 87 and 6 00 indicated the presence of two methoxyl groups and one methylenedioxy group The two doublets at δ7 68 and 7 00 (J = 9 Hz) can be assigned to H-5 and H-6 and the singlets at δ6 82 and 6 86 are attributed to H-2′ and H-5′ respectively Maxima isoflavone F contains a hydroxyl group and readily formed a monoacetate (**9a**), mp 194–197°, and a monomethyl ether mp 214–216°, which was identical in all respects with 7,8,6′-trimethoxy-3′,4′-methylenedioxyisoflavone (**6**) [7, 8] It therefore only remained to determine the relative positions of the hydroxyl and methoxyl groups in ring A The UV shift with NaOAc to 266 nm clearly indicates that the hydroxyl is at C-7 A comparison of the ^1H and ^{13}C NMR spectra of maxima isoflavone F with the relevant parts of the



R = H m/z 166 R = H m/z 138 R = Me m/z 152 m/z 146



spectra of 7,8,6′-trimethoxy-3′,4′-methylenedioxy isoflavone (**6**) and maxima isoflavone E (**8**) convincingly supports the proposed structure (**9**) having a 7-hydroxyl and an 8-methoxyl group

The molecular ion of **9** was observed at *m/z* 342 ($C_{18}H_{14}O_7$) Other significant fragments ions were at *m/z* 311 [M − 31], 167, 176 and 137 All of the compounds having an extra methoxyl group at C-6′ on ring B show a prominent peak due to loss of methoxyl in their mass spectra and this can therefore be of great assistance in identifying this type of substitution pattern Such interactions have good analogies [11, 16]

The fourth new compound, maxima isoflavone G (**10**), mp 298–302°, was found to be a hydroxy isoflavone containing one methoxy and one methylenedioxy group That the hydroxyl was at C-7 was indicated by the UV shift with NaOAc (248 → 257 nm) The compound readily formed a monoacetate, mp 177–178°, and a monomethyl ether, mp 214–216° The methyl ether was identical in all respects with 7,6′-dimethoxy-3′,4′-methylenedioxy-isoflavone (**5**) [7, 9] Furthermore the parent isoflavone was identical in all respects with a sample obtained from maxima isoflavone C by acid treatment [6] Compound **10** shows a prominent peak in the mass spectrum at *m/z* 281 due to loss of methoxyl in addition to other important fragment ions are at *m/z* 137 and 176/175 The ^{13}C NMR shifts of the ring A carbon atoms are in complete accord with those reported for 7-hydroxyisoflavones [12] Alkaline cleavage of **10** yields the corresponding deoxybenzoin (**15**) [9, 13] whose ^1H and ^{13}C NMR spectra (Tables 3 and 4) fully support the proposed structure

The remaining compounds (**5**, **6** and **7**) were identified by their spectral characteristics (see Tables 1 and 2) Compounds **5** and **6** have been reported to be present in the trunk wood of *Pterodon appariciοi* (Leguminosae) [7] Compound **5** has been synthesised [9] while compound **6** has been prepared from 4-methoxypterocarpin [8] Compound **7** was reported from the wood of *Dalbergia riparia* (Leguminosae) [10]

The melting points of **5**, **6** and **7** isolated by us were slightly higher than those reported for these compounds in the literature and direct comparison was not possible However their characterisation was firmly established on the basis of their spectral data In the case of compound **5** it could be compared with the corresponding sample obtained from maxima isoflavone C (**3**) by selective dealkylation at the 7-position to give **10** followed by subsequent methylation to give **5** Furthermore alkaline cleavage gave the corresponding deoxybenzoin (**13**)

EXPERIMENTAL

Aerial parts and roots of *Tephrosia maxima* Pers were collected from plants growing around Visakhapatnam A voucher specimen has been deposited in the Botany Department of Andhra University

Dried and powdered aerial parts (2 3 kg) and roots (2 kg) were extracted separately with hot petrol followed by $CHCl_3$ and EtOH Examination of the EtOH extracts was deferred After removal of solvents the petrol and chloroform extracts left crude semi solid residues (39 g, 57 g) and (19 g, 24 g) respectively The crude residues were subjected to column chromatography on silica gel and eluted with benzene, $CHCl_3$ and MeOH, and mixtures thereof The fractions were further purified by prep TLC wherever necessary

Column chromatography of the petrol extract from the aerial parts afforded maxima isoflavones A (**1**) and C (**3**), while the $CHCl_3$ extract gave **1**, **3**, maxima isoflavone D (**4**), 7,6′-dimethoxy-3′,4′-methylenedioxyisoflavone (**5**), 7,8,6′-trimethoxy-3′,4′-methylenedioxyisoflavone (**6**), maxima isoflavone E (**8**) and 6-methoxy-7-hydroxy-3′,4′-methylenedioxyisoflavone (**7**) Column chromatography of the $CHCl_3$ extract of the roots gave maxima isoflavones F (**9**) and G (**10**), besides all the above mentioned isoflavones except **7** All the compounds gave positive Labat test for a methylenedioxy group

Maxima isoflavone A (**1**) The fraction eluted with C_6H_6–$CHCl_3$ (3 1) afforded **1** (3 2 g) as colourless needles from $CHCl_3$–MeOH, mp 228–230° (lit [3] mp 226–229°) UV λ_{max}^{MeOH} nm, 261, 295, IR ν_{max}^{KBr} cm^{-1} 3080, 2900, 1650, 1620, 1178, 1100, 1048, 815, 720 EIMS (probe) 70 eV *m/z* (rel int) 310 $[M]^+$ (91 5), 309 (17 7), 165 (10 5), 164 $[C_8H_4O_4]^+$ (100), 146 $[C_9H_6O_2]^+$ (54 6), 145 (25 6), 126 (11 5), 106 (28 0) (Found $[M]^+$ 310 0477 $C_{17}H_{10}O_6$ requires 310 0477)

Alkaline degradation of **1** Compound **1** (200 mg) was refluxed with 12% NaOH (12 ml) in EtOH (12 ml) for 20 min The reaction mixture was cooled, diluted with water (100 ml) and acidified with 20% H_3PO_4 The solid that separated after cooling was taken up in $CHCl_3$ and washed with 5% $NaHCO_3$ and water, dried and the solvent evaporated The residue was crystallized from $CHCl_3$–MeOH to give the deoxybenzoin **11** (75 mg), mp 132/143–144 5° (lit [4] 142–144°) UV λ_{max}^{MeOH} nm 217, 243, 295 IR ν_{max}^{KBr} cm^{-1} 3460, 2900, 1655, 1610, 1490, 1440, 1250, 1186, 934 EIMS (probe) 70 eV *m/z* (rel int) 300 $[M]^+$ (6 5), 165 $[C_8H_5O_4]^+$ (100), 135 $[C_8H_7O_2]^+$ (8 6) (Found $[M]^+$ 300 0634 $C_{16}H_{12}O_6$ requires 300 0634)

Oxidation of **1** *with alkaline* H_2O_2 Compound **1** (100 mg) was dissolved in 5% KOH in 80% EtOH (10 ml) by warming for 15 min To the soln 12% H_2O_2 was added drop by drop (total 4 ml) When the effervescence ceased, the reaction mixture was warmed for a few min, diluted with H_2O to 30 ml, acidified with dil HCl and extracted with Et_2O The Et_2O extract was divided into acidic and phenolic fractions by extracting with 5% $NaHCO_3$ and 5% NaOH The acidic fraction gave 20 mg piperonylic acid, mp 227–229° (Me_2CO–petrol) while the phenolic fraction afforded the deoxybenzoin (**11**) (30 mg)

7,8,3′4′-Tetrahydroxyisoflavone Compound **1** (200 mg) in dry C_6H_6 (20 ml) was treated with anhydrous $AlCl_3$ (2 g) and the mixture refluxed for 3 hr The reaction mixture was cooled and the supernatant decanted off and the solvent removed The two residues were treated with HCl (1 1) and the combined acid solutions extracted with Et_2O The yellow Et_2O soln was extracted with 1% NaOH and the alkali soluble part recovered immediately by acidification and extraction with Et_2O The

residue obtained after evaporation of the solvent was purified by column chromatography over silica gel The fraction eluted with $CHCl_3$–MeOH (96 4) afforded the tetrahydroxyisoflavone as light yellow prisms from MeOH, mp 320° (decomp) with darkening at *ca* 270° It gave a dark green colour with $FeCl_3$ and negative Labat test

7,8,3',4'-Tetramethoxyisoflavone (**2**) The hydroxy compound (25 mg) was methylated with Me_2SO_4 (0 4 ml) and anhydrous K_2CO_3 (1 g) in Me_2CO (10 ml) The product (**2**) crystallized from $CHCl_3$–MeOH as colourless needles (20 mg), mp 166–168° (lit [5] mp 168–170°) It gave no colour with $FeCl_3$ EIMS (probe) 70 eV *m/z* (rel int) 342 $[M]^+$ (100), 327 $[M-OMe]^+$ (17 3), 299 $[M-Me-CO]^+$ (10 7), 162 $[C_{10}H_{10}O_2]^+$ (5 7), 152 $[C_8H_8O_3]^+$ (9 1), 137 $[C_8H_8O_3-Me]^+$ (10 5) 119 (29 5) (Found $[M]^+$ 342 1103 $C_{19}H_{18}O_9$ requires 342 1103)

Maxima isoflavone C (**3**) The fraction eluted with C_6H_6–$CHCl_3$ (1 1) afforded **3** (2 7 g) as colourless large hexagonal prisms from MeOH, mp 145–147° (lit [6] mp 142–144°) UV λ_{max}^{MeOH} nm 244, 249, 267, 306, IR ν_{max}^{KBr} cm^{-1} 1645, 1612, 925 EIMS (probe) 70 eV *m/z* (rel int) 380 $[M]^+$ (26 0), 312 $[M-C_5H_8]^+$ (26 6), 281 $[M-C_5H_8-OMe]^+$ (35 0), 175 $[C_{10}H_7O_3]^+$ (7 6), 137 $[C_7H_5O_3]^+$ (12 1), 69 $[C_5H_9]^+$ (100) (Found M^+ 380 1260 $C_{22}H_{20}O_6$ requires 380 1260)

Alkaline degradation of **3** Compound **3** (100 mg) was refluxed with 12% NaOH in 50% EtOH (12 ml) for 30 min The product worked up as described under 'alkaline degradation of **1**' afforded the deoxybenzoin **12** as a crystalline solid mp 79° (lit [6] mp 79–81°, lit [13] mp 80°)

Maxima isoflavone D (**4**) The fraction eluted with $CHCl_3$–MeOH (99 5 0 5) crystallized from $CHCl_3$–MeOH to give compound **4** as colourless needles (2 9 g) mp 223–224° UV $\lambda_{max}^{CHCl_3}$ nm 262, 292, IR ν_{max}^{KBr} cm^{-1} 3040, 2920, 1632, 1602, 1090, 1052, 740 EIMS (probe) 70 eV *m/z* (rel int) 326 $[M]^+$ (100), 311 $[M-Me]^+$ (24 3), 283 $[M-Me-CO]^+$ (20 0), 240 (10 4), 164 $[C_8H_4O_4]^+$ (19 5), 162 $[C_{10}H_{10}O_2]^+$ (8 9), 147 (16 8), 134 (11 3), 119 (39 3), 106 (16 0) (Found $[M]^+$ 326 0790 $C_{18}H_{14}O_6$ requires 326 0790)

Alkaline degradation of **4** Compound **4** (200 mg) was hydrolysed with 12% NaOH in 50% EtOH (25 ml) and the product worked up as usual to give the deoxybenzoin **14** as colourless needles from $CHCl_3$–MeOH (85 mg), mp 122° It gave light green colour with $FeCl_3$ UV λ_{max}^{MeOH} nm 216, 240, 295, IR ν_{max}^{KBr} cm^{-1} 3456, 2920, 1650, 1610, 1090, 1020, 920 EIMS (probe) 70 eV *m/z* (rel int) 316 $[M]^+$ (11 1), 165 $[C_8H_5O_4]^+$ (100), 151 $[C_9H_{11}O_2]^+$ (13 4) (Found $[M]^+$ 316 0947 $C_{17}H_{16}O_6$ requires 316 0947)

Oxidation of **4** *with alkaline* H_2O_2 Compound **4** (150 mg) was subjected to H_2O_2–KOH cleavage as in the case of compound **1** The acid fraction (25 mg) obtained was shown to be an approx 1 1 mixture of veratric acid and 2-hydroxypiperonylic acid by NMR and MS

7,8,3',4'-Tetramethoxyisoflavone (**2**) *from* **4** Compound **4** (150 mg) was demethylated and demethylenated with anhydrous $AlCl_3$ in dry C_6H_6 and the phenol thus obtained was methylated with Me_2SO_4–K_2CO_3 in Me_2CO The product crystallized from $CHCl_3$–MeOH as colourless needles (75 mg), mp 166–168° It was found to be identical in all respects with compound **2** obtained from maxima isoflavone A (**1**)

7,6'-Dimethoxy-3'4'-methylenedioxyisoflavone (**5**) This compound was eluted with $CHCl_3$–MeOH (99 5 0 5) and purified by prep TLC It crystallized from $CHCl_3$–MeOH to give colourless needles (257 mg), mp 222–224° (lit [7] mp 210–212°, lit [9] mp 208–209°) UV $\lambda_{max}^{CHCl_3}$ nm 244, 250, 267, 307, IR ν_{max}^{KBr} cm^{-1} 1637, 1622, 1600, 1492, 1437, 1260, 1242, 1184, 933, 825 EIMS (probe) 70 eV *m/z* (rel int) 326 $[M]^+$ (80 9), 295 $[M-OMe]^+$ (100), 182 (10 3), 176 $[C_{10}H_8O_3]^+$ (17 5), 175 (18 9), 161 (21 1), 151 $[C_8H_7O_3]^+$ (45 7), 147 (13 0), 107 (10 8), 103 (12 0) (Found $[M]^+$ 326 0790 $C_{18}H_{14}O_6$ requires 326 0790)

Deoxybenzoin **13** *from* **5** Compound **5** when treated with 12% EtOH–KOH in the usual way gave **13** as colourless needles ($CHCl_3$–MeOH), mp 133–134° (lit [9] mp 131–132°) UV λ_{max}^{MeOH} nm 216, 232, 257, 308, IR ν_{max}^{KBr} cm^{-1} 3450, 2995, 1670, 1630, 928

7,8,6'-Trimethoxy-3',4'-methylenedioxyisoflavone (**6**) This compound was eluted with $CHCl_3$–MeOH (99 5 0 5) and purified by prep TLC Crystallization from $CHCl_3$–MeOH gave colourless prisms (144 mg), mp 215–216° (lit [7] mp 204–206°, lit [8] mp 204°) UV $\lambda_{max}^{CHCl_3}$ nm 248, 256, 308, IR ν_{max}^{KBr} cm^{-1} 1625, 1610, 1305, 1272, 1090, 970, 920 EIMS (probe) 70 eV *m/z* (rel int) 356 $[M]^+$ (100), 325 $[M-OMe]^+$ (85 4), 189 (12 6), 181 $[C_9H_9O_4]^+$ (41 5), 176 $[C_{10}H_8O_3]^+$ (24 0), 175 $[C_{10}H_7O_3]^+$ (28 7), 163 (18 6), 162 (10 5), 161 (21 7), 151 $[C_8H_7O_3]^+$ (4 9), 149 (11 2), 137 $[C_7H_5O_3]^+$ (14 8), 109 (10 3), 103 (11 7) (Found $[M]^+$ 356 0923 $C_{19}H_{16}O_7$, requires 356 0896)

6-Methoxy-7-hydroxy-3',4'-methylenedioxyisoflavone (**7**) The fraction eluted with $CHCl_3$–MeOH (98 2) was purified by prep TLC to give compound **7** as colourless needles (72 mg) from $CHCl_3$–MeOH, mp 278 5–281° (lit [10] mp 260–262°) UV λ_{max}^{MeOH} nm 224, 264, 298, 324, $\lambda_{max}^{MeOH+NaOAc}$ nm 225, 267, 294, 351, EIMS (probe) 70 eV *m/z* (rel int) 312 $[M]^+$ (100), 267 (12 9), 166 $[C_8H_6O_4]^+$ (31 5), 146 $[C_9H_6O_2]^+$ (40 0), 145 $[C_9H_5O_2]^+$ (20 0), 127 (13 5), 123 (16 5) (Found $[M]^+$ 312 0634 $C_{17}H_{12}O_6$ requires 312 0634)

Maxima isoflavone E (**8**) This compound was eluted with $CHCl_3$–MeOH (98 2) and purified by prep TLC Two crystallizations from EtOH afforded compound **8** as colourless needles (153 mg), mp 268–270°, UV λ_{max}^{MeOH} nm 218, 256, 298, $\lambda_{max}^{MeOH+NaOAc}$ nm 220, 269, 295, IR ν_{max}^{KBr} cm^{-1} 3190, 1635, 1595, 1070, 923, 902 EIMS (probe) 70 eV *m/z* (rel int) 312 $[M]^+$ (100), 166 $[C_8H_6O_4]^+$ (4 5), 146 $[C_9H_6O_2]^+$ (21 1), 141 (10 4), 138 $[C_7H_6O_3]^+$ (20 7) (Found $[M]^+$ 312 0634 $C_{17}H_{12}O_6$ requires 312 0634)

Maxima isoflavone E acetate (**8a**) A 20 mg sample of maxima isoflavone E (**8**) when acetylated with Ac_2O–C_5H_5N in the usual way gave the acetate (**8a**) (16 mg), mp 165–167 5° UV λ_{max}^{MeOH} nm 214, 260, 297 (unaffected by NaOAc), IR ν_{max}^{KBr} cm^{-1} 2920, 1750, 1643, 1600, 1495, 1225, 1032, 930, 875

Maxima isoflavone E methyl ether (**8b**) (= *purpuranin A*) Compound **8** (35 mg) was methylated with Me_2SO_4–K_2CO_3 in Me_2CO to give the methyl ether (**8b**) as colourless plates from MeOH (20 mg), mp 182–183° (lit [14] mp 189–191°) EIMS (probe) 70 eV *m/z* (rel int) 326 $[M]^+$ (100), 152 $[C_8H_8O_3]^+$ (10 7), 146 $[C_9H_6O_2]^+$ (10 0) (Found $[M]^+$ 326 0790 $C_{18}H_{14}O_6$ requires 326 0790)

Maxima isoflavone F (**9**) This compound was eluted with $CHCl_3$–MeOH (98 2) and crystallized from $CHCl_3$–MeOH (92 mg), mp 257–259° UV λ_{max}^{MeOH} nm 214, 254, 306, $\lambda_{max}^{MeOH+NaOAc}$ nm 223, 266, IR ν_{max}^{KBr} cm^{-1} 3150, 2920, 1622, 1425, 1035, 930, 910, 832, 792, 750 EIMS (probe) 70 eV (rel int) 342 $[M]^+$ (100), 311 $[M-OMe]^+$ (81 4), 299 (11 7), 297 (13 9), 296 $[M-OMe-Me]^+$ (31 9), 189 (12 2), 177 (10 4), 176 $[C_{10}H_8O_3]^+$ (25 3), 175 (25 3), 167 $[C_8H_7O_4]^+$ (28 1), 161 (31 6), 138 (10 4), 137 $[C_7H_5O_3]^+$ (7 9), 123 (25 9), 103 (14 3) (Found $[M]^+$ 342 0736 $C_{18}H_{14}O_7$ requires 342 0733)

Maxima isoflavone F acetate (**9a**) Compound **9** (20 mg) when acetylated with Ac_2O–C_5H_5N in the usual way afforded the acetate (**9a**) as colourless needles from $CHCl_3$–MeOH (15 mg), mp 194–197°, UV λ_{max}^{MeOH} nm 216, 252, 308 (unaffected by NaOAc), IR ν_{max}^{KBr} cm^{-1} 2910, 1770, 1635, 1470, 1320, 1055, 930, 875

Maxima isoflavone F methyl ether (**6**) Compound **9** was methylated with Me_2SO_4–K_2CO_3 in Me_2CO to give a methyl

ether (**6**), mp 214–216°, identical in all respects with the natural sample **6**

Maxima isoflavone G (**10**) This compound was eluted with $CHCl_3$–MeOH (96 4) It crystallized from MeOH as colourless needles (135 mg), mp 298–302° (lit [6] mp 298–300°) UV λ_{max}^{MeOH} nm 215, 248, 308, $\lambda_{max}^{MeOH+NaOAc}$ nm 213, 257, 316, IR ν_{max}^{KBr} cm^{-1} 3150, 2960, 1635, 1615, 1586, 1395, 1230, 1092, 950, 933, 832, 780 EIMS (probe) 70 eV *m/z* (rel int) 312 $[M]^+$ (100), 281 $[M-OMe]^+$ (75 0), 176 $[C_{10}H_8O_3]^+$ (10 0), 175 $[C_{10}H_7O_3]^+$ (13 0), 161 (10 1), 140 (20 4), 137 $[C_7H_5O_3]^+$ (23 4). (Found M^+ 312 0634 $C_{17}H_{12}O_6$ requires 312 0634) The natural sample (**10**) was identical in all respects with an authentic sample of 7-hydroxy-6′-methoxy-3′,4′-methylenedioxyisoflavone (6′-methoxy-pseudobaptigenin) [6] obtained from maxima isoflavone C by treatment with 2% EtOH–HCl at 75–80° for 2 hr

Maxima isoflavone G acetate (**10a**) Compound **10** (15 mg) was acetylated with Ac_2O–C_5H_5N in the usual way to give the acetate (**10a**), mp 177–178°, UV λ_{max}^{MeOH} nm 216, 262, 304, IR ν_{max}^{KBr} cm^{-1} 3050, 2900, 1750, 1638, 1610, 1480, 1420, 1160, 926, 902, 780

Maxima isoflavone G methylether (**5**) Compound **10** (20 mg) was methylated with Me_2SO_4–K_2CO_3 in Me_2CO to give the methyl ether (**5**), mp 214–216°, identical in all respects with the natural sample **5**

Deoxybenzoin **15** *from maxima isoflavone G* Compound **10** when treated with 12% EtOH–KOH afforded the deoxybenzoin (**15**) as colourless prisms from MeOH, mp 168–169° (lit [13] mp 167°, lit [9] mp 164–165°)

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REFERENCES

1 Venkato Rao, E and Ranga Raju, N (1979) *Phytochemistry* **18**, 1581
2 Pelter, A, Ward, R S Venkato Rao, E and Ranga Raju, N (1981) *J Chem Soc Perkin Trans* 1, 2491
3 Rangaswami, S and Rama Sastry, B V (1954) *Curr Sci (India)* **23**, 397
4 Rangaswami, S and Rama Sastry, B V (1956) *Proc Indian Acad Sci* **44a**, 279
5 Kukla, A S and Seshadri, T R (1962) *Tetrahedron* **18**, 1443
6 Rangaswami, S and Rama Sastry, B V (1959) *Arch Pharm* **292**, 170
7 Galina, E and Gottlieb, O R (1974) *Phytochemistry* **13**, 2593
8 Bouwer, D, Brink, C M, Engelbrecht, J P and Rall, G J H (1968) *J S Afr Chem Inst* **21**, 159
9 Suginone, H (1960) *Tetrahedron Letters* 16
10 Braz Filho, R, Leite De Almeida, M E and Gottlieb, O R (1973) *Phytochemistry* **12**, 1187
11 Mabry, T J and Markham, K R (1975) in *The Flavonoids* (Harborne, J B, Mabry, T J and Mabry, H, eds) p 97 Chapman & Hall, London
12 Pelter, A, Ward, R S and Bass, R J (1978) *J Chem Soc Perkin Trans* 1, 666
13 Anirudhan, C A and Whalley, W B (1963) *J Chem Soc* 6049
14 Subba Rao, N V (1956) *Curr Sci (India)* **25**, 396
15 Mabry, T J, Markham, K R and Thomas, M B (1970) *The Systematic Identification of Flavonoids*, p 165 Springer, New York
16 Pelter, A and Stainton, P (1967) *J Chem Soc* 1933