

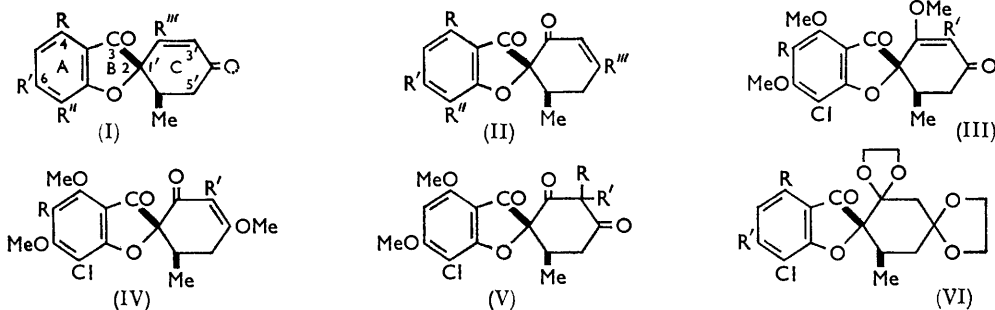
22. Griseofulvin Analogues. Part IX.* Proton Magnetic Resonance Studies.

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The proton magnetic resonance spectra of deuteriochloroform solutions of griseofulvin and of 43 representative analogues containing ethoxyl, isopropoxyl, acetoxyl, bisethylene ketal, methylthio-, methylamino-, pyrrolidin-1-yl, and halogen substituents and of a heavy-water solution of sodium griseofulvate have been recorded. The spectra are discussed and structural assignments made.

WE have examined, and have attempted to provide an interpretation of, the proton magnetic resonance (p.m.r.) spectra of griseofulvin and of 44 selected analogues and derivatives, whose infrared spectra we have discussed previously.¹ Our p.m.r. observations are complementary to, and extend, those contained in three accounts of the p.m.r. spectra of griseofulvin and of certain analogues encountered during the synthesis of griseofulvin, which appeared while this paper was being prepared.²

The proton magnetic resonance spectra of the griseofulvin analogues proved to be of considerable interest and provided structural information that supplemented in a remarkable way what had been obtained from infrared measurements. We were able to assign each spectral line or group of lines to a particular proton or group of protons and, conversely, to correlate each proton with a particular line or group of lines. Changes in chemical shift could be associated with differences in the substitution pattern and in the stereochemical configuration of the molecules (cf. Arison *et al.*^{2c}).



Discussion.—Our structural assignments for the various spectral lines, except for those between 6·5 and 7·9 τ (see below), are summarised in Tables 1—3.

Alkoxy, alkylthio, and hydroxyl protons. The methoxyl resonances are readily identified as sharp three-proton singlets between 5·80 and 6·37 τ (Table 1). In griseofulvin (I; R = R' = R''' = MeO, R'' = Cl), the 4- and 6-methoxyl groups appear at 6·00 and 5·94 τ and in isogriseofulvin (II; R = R' = R''' = MeO, R'' = Cl) at 6·09 and 5·97 τ , respectively. The 6-methoxyl absorbs at a slightly lower field strength than the 4-methoxyl group, since the former is adjacent to a chlorine atom, which induces a downward chemical shift.

This assignment, which differs from that of Arison *et al.*,^{2c} is supported by our measurements on the 6-acetoxy-4-methoxy- and the 4-acetoxy-6-methoxy-analogues, the 6-ethoxy-4-methoxy- and the 4-ethoxy-6-methoxy-analogues, the 6-isopropoxy-4-methoxy- and

* Part VIII, Page and Staniforth, *J.*, 1963, 1814.

¹ Page and Staniforth, *J.*, (a) 1962, 1292; (b) 1963, 1814.

² (a) Gerecke, Kyburz, von Planta, and Brossi, *Helv. Chim. Acta*, 1962, **45**, 2241; (b) Taub, Kuo, Slates, and Wendler, *Tetrahedron*, 1963, **19**, 1; (c) Arison, Wendler, Taub, Hoffsommer, Kuo, Slates, and Trenner, *J. Amer. Chem. Soc.*, 1963, **85**, 627.

the 4-isopropoxy-6-methoxy-analogues, and the 4-methoxy-6-pyrrolidin-1-yl and the 6-methoxy-4-pyrrolidin-1-yl analogues of griseofulvin. The 4-methoxy protons of the 6-acetoxy-4-methoxy-analogue (I; $R = R''' = \text{MeO}$, $R' = \text{AcO}$, $R'' = \text{Cl}$) absorb at a slightly higher field (6.06 τ) than the 6-methoxy protons of the 4-acetoxy-6-methoxy-analogue (I; $R = \text{AcO}$, $R' = R''' = \text{MeO}$, $R'' = \text{Cl}$) (5.98 τ); the methyl protons of the corresponding 6- and 4-acetoxy-groups absorb at 7.60 and 7.63 τ , respectively.

Elimination of the 7-chlorine atom, as in dechlorogriseofulvin (I; $R = R' = R''' = \text{MeO}$, $R'' = \text{H}$), causes the 4- and 6-methoxyl protons to absorb at the same frequency, namely, 6.08 τ . Introduction into the griseofulvin molecule of a 3'-chlorine atom has little effect, but a 5-chlorine atom displaces the 4- and 6-methoxyl lines to 5.92 and 5.83 τ , respectively (Table 2).

The 4- and 6-methoxyl protons of isogriseofulvin absorb at a slightly higher field strength than do those of griseofulvin, indicating that a 2'-methoxyl group has a smaller shielding effect on the 4- and the 6-methoxyl resonance than has a 2'-ketone group. A similar effect is observed when the ring-c methoxyl group is replaced by a hydrogen or halogen atom, or by ethoxyl, methylamino-, or pyrrolidin-1-yl, groups. If the 4- and 6-methoxyl are replaced by ethoxyl groups, both the triplets associated with the methyl and the quadruplets associated with the methylene protons of the ethoxyl group are displaced upfield in going from the 4'-oxo-2'-enol ether (I; $R = R' = R''' = \text{EtO}$, $R'' = \text{Cl}$) to the 2'-oxo-4'-enol ether (II; $R = R' = R''' = \text{EtO}$, $R'' = \text{Cl}$). This displacement provides an additional test for distinguishing between analogues of griseofulvin and of isogriseofulvin (cf. Page and Staniforth^{1a}).

The proton resonance frequencies for both 2'- and 4'-methoxyl groups are, as might be expected, higher than those for the corresponding 4- and 6-methoxyl group (Table 1). The frequency for a 2'-methoxyl group (6.33—6.37 τ) is, however, also higher than that for the corresponding 4'-methoxyl (6.14—6.20 τ). This frequency difference may be attributed to the spiro-configuration of the molecule. A model shows that the 2'-methoxyl group is brought into the shielding zone of the aromatic A-ring, but hardly approaches the deshielding zone of the 3-ketone; the nett result is therefore shielding and higher τ values for the 2'-methoxyl group. The protons associated with 2'-ethoxyl, 2'-methylamino-, and 2'-pyrrolidin-1-yl groups, also, appear at higher field strengths than do those for the corresponding 4'-substituents. Introduction of a 3'-halogen atom displaces the 2'- and 6'-methoxyl lines downfield to 5.94—5.99 and 5.91—5.97 τ , respectively (Table 2).

The proton resonances for 4- and 6-methylthio-groups parallel those for 4- and 6-methoxyl groups and appear at 7.43 and 7.38 τ , respectively, in the 4'-oxo-2'-enol ether (I; $R = R' = \text{MeS}$, $R'' = \text{Cl}$, $R''' = \text{MeO}$) and at 7.48 and 7.41 τ , respectively, in the 2'-oxo-4'-enol ether (II; $R = R' = \text{MeS}$, $R'' = \text{Cl}$, $R''' = \text{MeO}$). Similarly, the methylthio-proton resonance line for the 2'-methylthio-analogue of griseofulvin (I; $R = R' = \text{MeO}$, $R'' = \text{Cl}$, $R''' = \text{MeS}$) appears at a higher field, namely, 7.75 τ .

The ethylene protons of the 2',4'-bisethylene ketals (VI) form two simple A_2B_2 systems, which give two four-proton multipliers between 5.53 and 6.36 τ (Table 3).

The hydroxyl group in the 6-hydroxy-4-methyl-analogue of griseofulvin absorbed at a low field strength, namely, 1.82 τ (Table 1). The N-H protons in the 2'-methylamino- and 4'-methylamino-analogues gave multiplets at 5.28 and 4.90 τ , respectively, which disappeared when the deuteriochloroform solutions were shaken with heavy water; the methyl doublets at 7.28 and 7.17 τ , respectively, collapsed to give singlets.

Methyl groups. The 6'-methyl groups of griseofulvin and isogriseofulvin couple with the 6'-proton to give characteristic doublets ($J = 6$ c./sec.) centred at 9.04 and 8.98 τ , respectively (Table 1). This frequency displacement between the 4'-oxo-2'- and the 2'-oxo-4'-enol ether system also occurs when the ring-c methoxyl is replaced by a hydrogen or a chlorine atom or by an ethoxyl, methylamino-, or pyrrolidin-1-yl group (Table 1) and when methyl and halogen substituents are introduced at carbon-3' (Table 2). The effect is probably associated with the bond angle change at spiro-carbon-2 in going from the

TABLE I.
Proton-resonance lines (τ values) for griseofulvin and isogriseofulvin analogues (J values in c./sec. in parentheses).

No.	Compound						Ref.	6-Subst.	2'-Subst.	3'-H	4'-Subst.	6'-Me
	R	R'	R''	R'''	5-H	6-Subst.						
1	MeO	MeO	Cl	H	3.83	5.95	3.42(d,1.0)	3.84(d,1.0)	—	3.84(d,1.0)	—	9.08(d,6)
2	MeO	MeO	Cl	H	3.90	5.97	6.07	3.92(d,1.0)	—	3.92(d,1.0)	2.85(d,1.0)	8.98(d,6)
3	Me	HO	Cl	MeO	3.80(d,0.8)	1.82	7.63(d,0.8)	4.44(d,1)	—	4.44(d,1)	6.14	9.01(d,6)
4	I	MeO	Cl	MeO	3.46(d,0.8)	5.94	7.39(d,0.8)	4.42	6.33	4.42	—	9.08(d,6)
5	II	Me	Cl	MeO	3.52(d,0.8)	5.98	7.47(d,0.8)	4.52(d,1)	—	4.52(d,1)	6.18	9.00(d,6)
6	I	MeO	H	MeO	3.76(d,2)*	6.08	6.08	4.46	6.35	4.46	—	9.05(d,6)
7	I	MeO	Cl	MeO	3.82	5.94	6.00	4.45	6.36	4.45	—	9.04(d,6)
8	II	MeO	Cl	MeO	3.90	5.97	6.09	4.58(d,0.5)	—	4.58(d,0.5)	6.20	8.98(d,6)
9	I	EtO	Cl	MeO	3.86	5.98	5.79(q,7)	4.47	6.37	4.47	—	9.05(d,6)
10	I	MeO	Cl	MeO	3.87	5.74(q,7)	6.03	4.47	6.37	4.47	—	9.05(d,6)
11	I	PrHO	Cl	MeO	3.83	8.47(t,7)	5.25(m,6)	4.47	6.36	4.47	—	9.04(d,6)
12	I	MeO	Cl	MeO	3.87	8.57(d,6)	6.04	4.46	6.37	4.46	—	9.04(d,6)
13	I	Pyrrol- idin-1-yl	Cl	MeO	4.28	6.43(t,6)	8.01(m)	4.46	6.37	4.46	—	9.04(d,6)
14	I	MeO	Cl	MeO	4.20	8.01(m)	6.09	4.47	6.37	4.47	—	9.04(d,6)
15	I	AcO	Cl	MeO	3.54	5.98	7.63	4.46	6.36	4.46	—	9.06(d,6)
16	I	MeO	Cl	MeO	3.60	7.60	6.06	4.42	6.36	4.42	—	9.04(d,6)
17	II	MeO	Cl	MeO	3.65	7.60	6.10	4.55(d,1)	—	4.55(d,1)	6.20	8.96(d,6)
18	I	MeO	Cl	EtO	3.85	5.94	6.00	4.49	6.12(q,7)	4.49	—	9.05(d,6)
19	II	MeO	Cl	EtO	3.91	5.98	6.08	4.58(d,0.5)	8.80(t,7)	4.58(d,0.5)	6.00(q,7)	9.00(d,6)
20	I	MeO	Cl	EtO	3.86	5.73(q,7)	6.02	4.49	—	4.49	8.60(t,7)	9.05(d,6)
21	I	EtO	Cl	EtO	3.89	8.44(t,7)	5.81(q,7)	4.51	—	4.51	—	9.05(d,6)
22	II	EtO	Cl	EtO	3.96	8.49(t,7)	8.47(t,7)	4.59(d,1)	6.15(q,7)	4.59(d,1)	5.91(q,7)	8.99(d,6)
23	I	MeO	Cl	MeS	3.82	8.60(t,7)	6.01(q,7)	—	—	—	8.56(t,7)	9.05(d,6)
24	I	MeO	Cl	Cl	3.82	5.93	5.98	4.08	7.75	4.08	—	9.03(d,6)
25	II	MeO	Cl	Cl	3.89	5.93	5.99	3.58	—	3.58	—	8.98(d,6.5)
26	I	MeO	Cl	MeNH	3.80	5.98	6.06	4.74	5.28(m)	4.74	—	9.10(d,6)
27	II	MeO	Cl	MeNH	3.92	5.92	5.98	4.83(d,0.5)	7.28(d,5)	4.83(d,0.5)	4.90(m)	9.03(d,6.5)
28	I	MeO	Cl	Pyrrol- idin-1-yl	3.80	5.92	5.97	4.74	6.85(m)	4.74	7.17(d,5)	9.14(d,6)
									8.18(m)			

TABLE 1. (Continued.)

No.	Compound				5-H	6-Subst.	2'-Subst.	3'-H	4'-Subst.	6'-Me
	R	R'	R''	R'''						
29	MeO	MeO	Cl	Pyrrol- idin-1-yl	3.92	5.95	—	4.90(d,0.5)	6.60(m) 8.00(m)	9.01(d,6)
30	MeS	MeS	Cl	MeO	3.42	7.38	6.35	4.43	—	9.07(d,6)
31	MeS	MeS	Cl	MeO	3.46	7.41	—	4.54(d,1)	6.19	9.00(d,6)

Unless otherwise indicated, values refer to singlet absorptions. For multiplets, d = doublet, t = triplet, q = quadruplet, m = multiplet.

* 7-Hydrogen.

Refs.: *a*, Mulholland, *J.*, 1952, 3994. *b*, Goodall, Gregory, and Walker, *J.*, 1963, 1610. *c*, MacMillan, *J.*, 1953, 1697. *d*, Grove, MacMillan, Mulholland, and Rogers, *J.*, 1952, 3949. *e*, Arkley, Attenburrow, Gregory, and Walker, *J.*, 1962, 1260. *f*, Grove, MacMillan, Mulholland, and Rogers, *J.*, 1952, 3977. *g*, Duncanson, Grove, MacMillan, and Mulholland, *J.*, 1957, 3555. *h*, MacMillan, *J.*, 1959, 1823. *i*, Stephenson, Walker, Warburton, and Webb, *J.*, 1962, 1282.

TABLE 2.

Proton-resonance lines (τ values) for 5- and 3'-substituted griseofulvin and isogriseofulvin (J values in c./sec. in parentheses).

No.	Compound				5-Subst.	6-Subst.	2'-Subst.	3'-Subst.	4'-Subst.	6'-Me
	R	R'	R''	R'''						
32	H	Me	—	—	3.82	5.93	6.25	8.16	—	9.06(d,6)
33	H	Me	—	—	3.90	5.96	—	8.30	6.11	8.96(d,6)
34	Cl	H	—	—	—	5.92	6.33	4.42	—	9.03(d,6)
35	Cl	H	—	—	—	5.92	—	4.54(d,1)	6.20	9.00(d,6)
36	H	Cl	—	—	3.80	5.93	5.99	—	—	9.03(d,6)
37	H	Cl	—	—	3.89	5.94	—	—	5.94	8.94(d,6)
38	H	Br	—	—	3.92	5.97	—	—	5.97	8.95(d,6)
39	Cl	Cl	—	—	—	5.80	5.94	—	—	9.03(d,6)
40	Cl	Cl	—	—	—	5.88	—	—	5.91	8.93(d,6)
41	Pr ⁿ	H	—	—	9.03(t,7) 8.53(m) 7.38(t,7)	5.89	6.32	4.40	—	9.05(d,6)

Refs.: *e*, See Table 1. *j*, Gregory, Holton, Robinson, and Walker, *J.*, 1962, 1269. *k*, Walker, Warburton, and Webb, *J.*, 1962, 1277.

TABLE 3.

Proton-resonance lines (τ values) for griseofulvic acid derivatives (J values in c./sec. in parentheses).

No.	Compound				5-H	6-MeO	2'-Subst.	3'-Subst.	4'-Subst.	6'-Me
	R	R'	R''	R'''						
42	V	Na	H*	—	3.69	5.96	—	4.80	—	9.17(d,5.5)
43	V	Me	Me	—	3.86	5.95	—	8.62	—	8.95(d,6)
44	VI	MeO	MeO	—	3.91	5.97	5.53—6.36(m)	7.53(d,11)	5.53—6.36(m)	9.20(d,6)
45	VI	MeS	MeS	—	3.50	7.42	5.53—6.36(m)	8.16(d,11) 7.53(d,11) 8.15(d,11)	5.53—6.36(m)	9.23(d,6)

* Sodium salt of enolic form, examined in D₂O solution (external standard, SiMe₄ in CCl₄).

Refs.: *e*, *i*, See Table 1. *j*, See Table 2.

griseofulvin to the isogriseofulvin form; this change causes the 6'-methyl group of isogriseofulvin to be brought nearer to the 1-oxygen atom.

3'-Methyl substituents in griseofulvin and isogriseofulvin (Table 2) give sharp singlets at 8.16 and 8.30 τ , respectively. In griseofulvin (I; R = Me, R' = R'' = MeO, R'' = Cl) and isogriseofulvin analogues (II; R = Me, R' = R'' = MeO, R'' = Cl), 4-methyl groups couple with the proton on carbon-5 and yield broad singlets ($J = 0.8$ c./sec.) at 7.39 and 7.47 τ , respectively.

Aromatic protons. The aromatic proton at carbon-5 appears as a singlet at 3.82 and 3.90 τ in the p.m.r. spectra of griseofulvin and isogriseofulvin, respectively; this displacement is a further characteristic of the change from a 4'-oxo-2'- to a 2'-oxo-4'-enol ether (see Table 1). It is noteworthy that a change from a "normal" to an "iso" substitution pattern in ring C has a greater effect on the p.m.r. absorption than on the infrared absorption of ring-A.^{1a}

Introduction of a 5-chlorine atom or a 5-n-propyl group causes the aromatic proton line to disappear (Table 2). Replacement of the 7-chlorine atom by hydrogen, as in dechlorogriseofulvin (I; R = R' = R'' = MeO, R'' = H), leads to the appearance of a single-proton doublet centred at 3.76 τ ($J = 2$ c./sec.), which is coupled with a doublet centred at 3.95 τ and associated with the 5-proton.²

6-Demethylation of the 4-methyl-6-methoxy-analogue of isogriseofulvin (cf. the 6-hydroxy-4'-isopropoxy-analogue of isogriseofulvin^{2c}) displaces upfield lines associated with the 4-methyl protons and the proton on carbon-5 (Table 1).

Olefinic protons. The olefinic protons at carbon-3' in griseofulvin and isogriseofulvin give, respectively, a sharp singlet at 4.45 and a narrow doublet centred at 4.58 τ ($J = 1.0-0.5$ c./sec.); this behaviour is another characteristic feature of the change from the 4'-oxo-2'- to the 2'-oxo-4'-enol ether series (Table 1). The slight splitting of the 3'-proton peak for isogriseofulvin is probably caused by long-range interaction with the 5' α -proton.

Protons on carbon-5' and -6'. The protons on carbon-5' and -6' in griseofulvin analogues form a typical ABC system and give rise to a complex pattern of bands between 6.5 and 7.9 τ . Because of their complex nature, these bands have not been listed in the Tables. Two characteristic "doublets" centred at about 6.97 and 7.62 τ ($J = 11$ c./sec.) appear in the spectra of most griseofulvin analogues; isogriseofulvin analogues show similar "doublets" centred at about 6.82 and 7.56 τ ($J = 13$ c./sec.). Preliminary attempts to resolve the peaks by spin-decoupling methods have been unsuccessful.

The relatively broad band pattern between 6.5 and 7.9 τ shown by griseofulvin and isogriseofulvin analogues suggests that in these compounds there is a relatively large chemical shift between the 5' α - and 5' β -protons. This behaviour contrasts with that of epigriseofulvin in which a narrow band pattern suggests a small chemical shift between the 5' α - and 5' β -protons. Arison *et al.*^{2c} have used this difference in chemical shift to deduce the stereochemical configuration of the 6'-methyl group of griseofulvin and epigriseofulvin.

Experimental.—The spectra were measured at 38° with a Varian Associates A-60 p.m.r. spectrometer at a sweep rate of 1 c./sec./sec. and were calibrated against tetramethylsilane (SiMe₄ = 10 τ) used as an internal standard. The compounds were in 5% w/v solution in deuteriochloroform, except sodium griseofulvate, which was in heavy water. H-O and H-N signals were recognised through their disappearance on deuteration.

The compounds were prepared in these laboratories and had the physical properties described in the references listed in the Tables.