Synthesis of Thiogibberellins: *X*-Ray Molecular and Crystal Structure of 19,10-Thio-3-epigibberellin A_1

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The synthesis of thiogibberellins required for studies on structure–biological activity relationships is described. The 7-thiogibberellin analogues (**10**), (**11**), and (**13**) were prepared *via* sulphydrolysis of the corresponding symmetric anhydrides (**4**)—(**6**). Photolysis of (**10**) and (**13**) led, under intramolecular functionalization, to the 7,15α-thiolactones (**15**) and (**17**), respectively. For synthesis of 19,10-thiogibberellins, the $\Delta^{1(10)}$-thiocarboxylic intermediate (**22**) was prepared in 4 steps from GA_3 methyl ester (**18**). The regio- and stereo-selective γ-thiolactone ring closure in (**22**) was achieved by intramolecular photoaddition or upon treatment with hydrogen sulphide–pyridine to give (**23**). Deacetylation of (**23**) with sodium methoxide led *via* simultaneous retro-aldol reaction to the 3α-hydroxylated thiolactone methyl ester (**25**), which was demethylated to 19,10-thio-3-epi-GA_1 (**26**). The structure of the methyl ester (**25**) was independently confirmed by *X*-ray analysis.

Although a multitude of structurally modified gibberellins have been prepared[1-3] the synthesis of thio analogues of this phytohormone group has not been described hitherto. In extending our studies on structure and biological activity this paper addresses the syntheses of such thiogibberellins. As reported already,[2,4,5] preliminary reaction sequences were developed which led from GA_3 (**1**) and GA_1 (**3**), respectively, to the 7-thio analogues (**10**) and (**13**) as well as to the 19,10-thiolactone gibberellin derivatives (**23**), (**25**), and (**26**), leaving intact other structural requirements essential for phytohormone activity.[6,7] In the course of this work new photochemical ring-closure reactions leading from the thiocarboxylic acids (**10**) and (**13**), respectively, to 7,15α-thiolactones (**15**) and (**17**) and from the thiocarboxylic intermediate (**22**) to the 19,10-thiolactone structure (**23**) were discovered.

In this paper we summarize and supplement our investigations of the synthesis of thiogibberellins including the *X*-ray analysis of the prepared 19,10-thio-3-epi-GA_1 methyl ester (**25**).

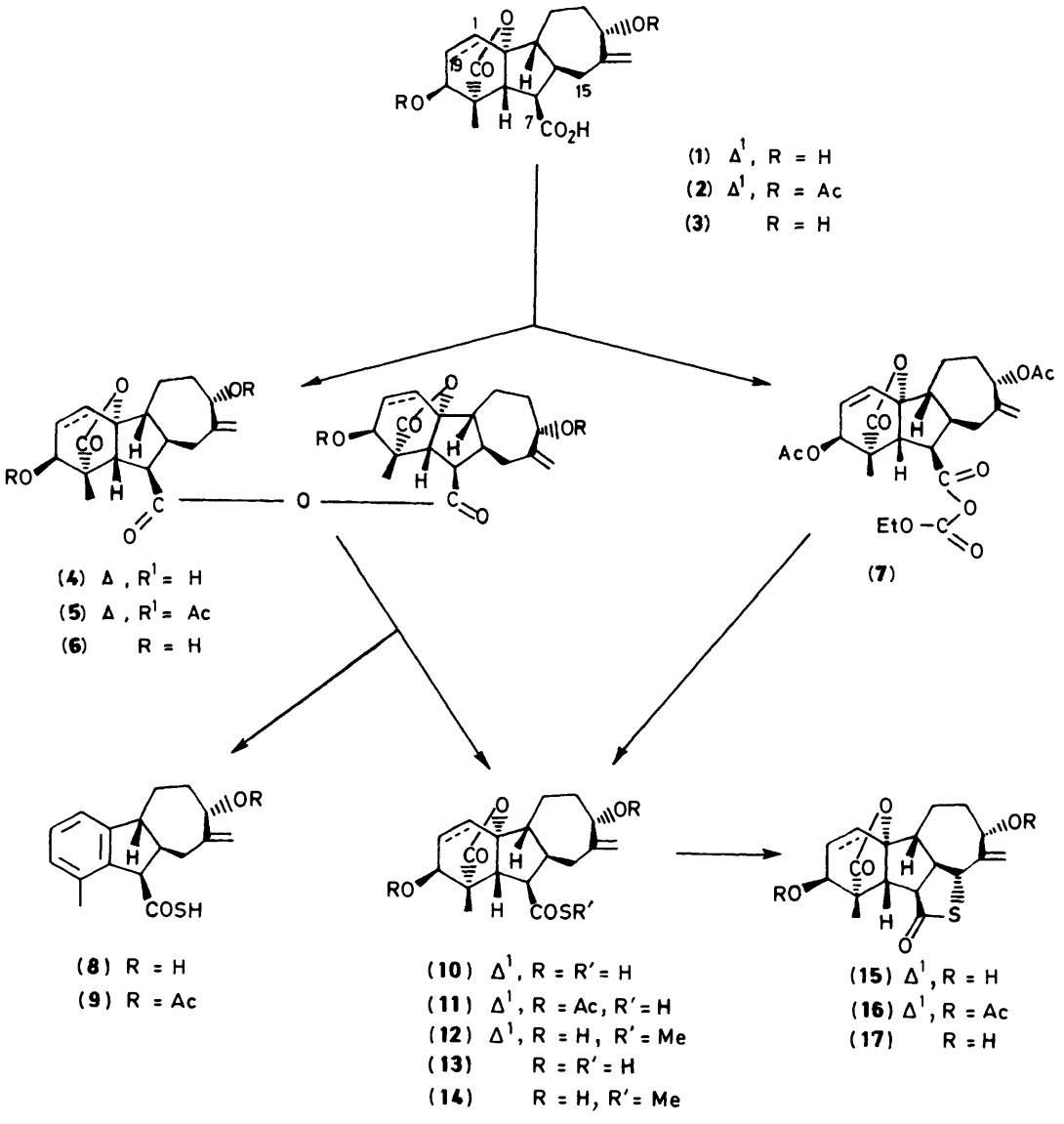
Results and Discussion

For synthesis of the 7-thio analogues, GA_3 (**1**), its 3,13-diacetate (**2**), and GA_1 (**3**) were transformed with dicyclohexylcarbodiimide (DCC) as previously described to the symmetric anhydrides (**4**),[8] (**5**),[9] and (**6**).[10] Upon sulphydrolysis of these anhydrides in dry pyridine the 7-thiogibberellins (**10**), (**11**), and (**13**) besides the corresponding starting compounds (**1**), (**2**), and (**3**), respectively, were obtained. In the case of the sulphydrolysis of anhydride (**4**) the 7-thioepiallogibberic acid (**8**) was also isolated as a minor product and was characterized as its 13-acetate (**9**). Compound (**11**) was prepared also from (**2**) *via* sulphydrolysis of the mixed anhydride (**7**) as well as *via* prolonged acetylation (acetic anhydride–pyridine, 7 days) of 7-thio-GA_3 (**10**) (Scheme 1).

The crystalline 7-thiogibberellins showed correct elemental compositions in the high-resolution mass spectra and exhibited a characteristic positive Cotton effect around 275 nm due to a (u.v.-inactive) $n \longrightarrow \pi^*$-transition of the thiocarboxy chromophore.[11] The ^{1}H n.m.r. spectra were similar to those of the parent sulphur-free gibberellins. In the case of 7-thio-GA_3 (**10**) the AB-spin system of the 5- and 6-proton could be separated only by using higher resolution (200 MHz), and then showed doublets at δ 3.35 and 3.33 (*J* 10 Hz). In the cation mass spectra a preferred loss of two hydrogens from the molecular is remarkable. Thus, in 7-thio-GA_1 (**13**) the $M^+ - 2$ peak is more than twice as intense (76%) as the molecular ion (33%), and in the mass spectrum of 7-thio-GA_3 diacetate (**11**) the $M^+ - 2$ peak appears as the peak of highest mass number. This loss of two hydrogens seems to be typical of the 7-thiocarboxy function and finds its parallel in the observed photochemical dehydrogenation of compounds of this structural type to 7,15α-thiolactones as described below. The corresponding methyl esters (**12**) and (**14**), prepared from acids (**10**) and (**13**) with ethereal diazomethane in ethyl acetate, showed normal molecular ions in their mass spectra.

Upon irradiation (at 254 nm) of 7-thio-GA_3 (**10**) in ethyl acetate under argon, the 7,15α-thiolactone (**15**) was formed in 34% yield and showed a positive Cotton effect at 280 nm with *a* +148. In the mass spectrum of (**15**) the molecular ion appears as the base peak, and upon deuterium exchange only two active hydrogens were indicated. Structure (**15**) is also in good accord with the ^{1}H n.m.r. spectrum, which shows a new one-proton doublet at δ 4.55 for the 15β-H as well as the distinct doublet character of the 17-H_2 signals at 4.81 and 5.23 (each 2 Hz), indicating one remained allylic coupling. With regard to the configuration at C-15, similar to that found for the corresponding gibberellin 7,15-lactones,[12] only a 15α-connection is possible because of an extreme distortion of the thiolactone ring in the case of 15β-annelation (Dreiding models). Long-termed acetylation of (**15**) gave the 3,13-diacetate (**16**). Similarly, 7-thio-GA_1 (**13**) was transformed photochemically into the corresponding thiolactone (**17**) in 38% yield. The observed regio-and stereo-selective ring closure of (**10**) and (**13**) to the 7,15α-thiolactones (**15**) and (**17**), respectively, represents to our knowledge the first case of such an intramolecular functionalization observed in the photochemistry of thiocarboxylic acids.

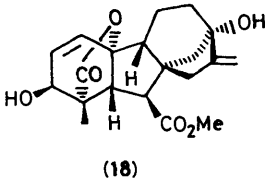
Our strategy for the synthesis of 19,10-thiogibberellin compounds involved the use of the lactone-opened $\Delta^{1(10)}$-19-oic



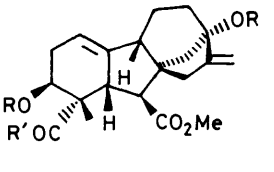
Scheme 1.

acid (**20**) as key intermediate for its transformation into the corresponding 19-thio analogue (**22**), followed by intramolecular thiolactone ring closure. Compound (**20**) was prepared as described earlier by catalytic hydrogenation of GA_3 methyl ester (**18**)[13] to the carboxylic acid (**19**) followed by prolonged acetylation.[14] Attempts to prepare the thioacid (**22**) from (**20**) *via* sulphydrolysis of its 19,19-anhydride or the *S*-phenyl ester[15] failed because of an unexpected stability of both derivatives. However, sulphydrolysis of the crude chloride (**21**) in pyridine gave the desired 19-thio compound (**22**) in 47% yield (based on consumed starting material) with the expected spectroscopic properties. Upon irradiation of (**22**) in absolute tetrahydrofuran (THF) (254 nm, argon) regio- and stereoselective intramolecular photoaddition of the thiocarboxy function to the $\Delta^{1(10)}$ double bond took place leading, in 72% yield, to the desired γ-thiolactone (**23**).

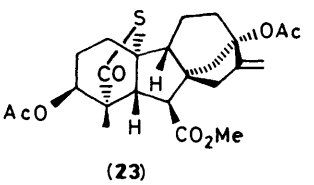
In the ^{1}H n.m.r. spectrum of (**23**) the doublet character of the 5-proton signal as well as the absence of a –S–C(–)–H resonance confirmed the original 19,10-thiolactone ring, and ruling out an alternative 19,1-thiolactone structure. For the photochemical transformation of (**22**) into (**23**) a radical addition of the thiocarboxylic function to the $\Delta^{1(10)}$ double bond *via* the intermediates (**a**) and (**b**) is proposed. Thus, contrary to the



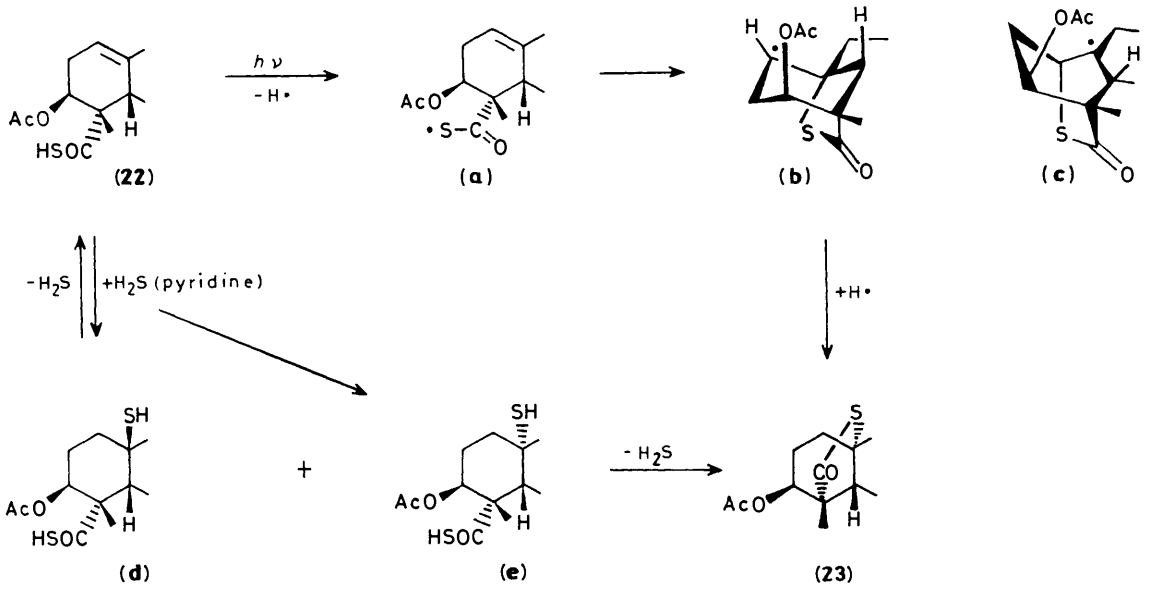
(**18**)



(**19**) R = H, R' = OH
(**20**) R = Ac, R' = OH
(**21**) R = Ac, R' = Cl
(**22**) R = Ac, R' = SH



(**23**)



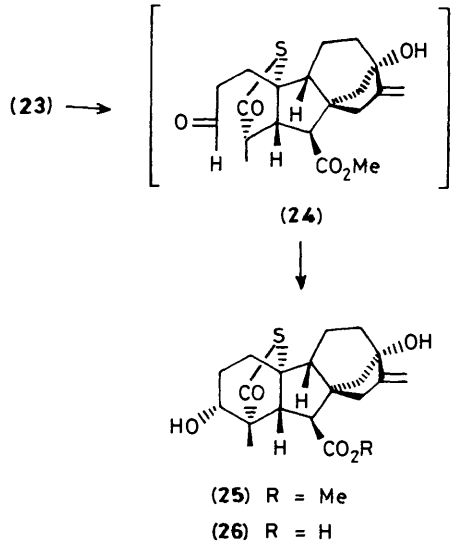
Scheme 2. Proposed mechanism for the radical and non-radical thiolactonization of compound (**22**) to the 19,10-thiogibberellin (**23**)

known[16] antiMarkovnikov radical photoaddition of simple thiocarboxylic acids to cyclic olefins, in the intramolecular thiolactonization to (**23**) exclusive Markovnikov addition is observed. This should be due to the higher stability of the intermediate *C*-radical (**b**) (ring A in a nearly chair conformation) in comparison with a corresponding anti-Markovnikov *C*-radical (**c**) which is highly strained because of the trigonality of C-10 (Dreiding models) (Scheme 2).

Furthermore it was found that the thiocarboxylic compound (**22**) could be also cyclized to (**23**) upon treatment with hydrogen sulphide in the presence of pyridine. For this pyridine-catalysed ring closure an addition of hydrogen sulphide to the $\Delta^{1(10)}$ double bond of (**22**) to give the 10-epimeric mercapto intermediates (**d**) and (**e**) can be assumed, followed by thiolactonization under hydrogen sulphide elimination from the 10α-compound (**e**). This led to an effective second method for preparation of thiolactone (**23**). Thus, prolonged treatment (7 h) of the crude chloride (**21**) with hydrogen sulphide in pyridine gave directly [89%, based on consumed acid (**20**)] the desired thiolactone (**23**) without isolation of the intermediate thiocarboxylic acid (**22**). In agreement with these results, in the preparation of the thiocarboxylic acid (**22**) *via* the chloride (**21**) the thiolactone (**23**) was found as a minor product.

Treatment of compound (**23**) with 2 mol equiv. of sodium methoxide in methanol smoothly yielded the dihydroxy thiolactone (**25**) with α-configuration of the 3-hydroxy group. Thus, in addition to deacetoxylation, inversion at C-3 has taken place due to a smooth alkali-catalysed retro-aldol reaction *via* the corresponding 3,4-seco-3-aldehyde (**24**), a reaction well known for ordinary lactones of the GA_1 series.[17,18] The inversion at position 3 was indicated in the ^1H n.m.r. spectrum of (**25**) by the high-field-shifted 5-and 6-H doublets at δ 2.99 and 2.54 as similarly observed for 3-epi-GA_1 compared with GA_1 (**3**).[18] Compound (**25**) was demethylated with lithium n-propanethiolate in hexamethylphosphoric triamide (HMPA) to give a 79% yield of 19,10-thio-3-epi-GA_1 (**26**) (Scheme 3).

For an independent structural proof of the synthesized 19,10-thiolactones an *X*-ray analysis of the methyl ester (**25**) has been performed, which confirmed unequivocally the presence of a γ-thiolactone ring as well as the equatorial 3α-hydroxy group in compounds (**25**) and (**26**). In our preliminary accounts[2,5] the



Scheme 3.

configuration at C-3 for compounds (**25**) and (**26**) was erroneously reported as 3β and must be corrected.

For data collection a single crystal of compound (**25**), $C_{20}H_{26}O_5S$, crystallized from acetone–hexane was used.

Crystal data.—Monoclinic, $a = 25.543(8)$, $b = 6.397(4)$, $c = 11.456(5)$ Å, $\beta = 95.19(3)°$, $Z = 4$, $D_x = 1.346$ g cm^{-3}, space group $C2$, m.p. 233 °C (decomp.). The intensities of 1 571 independent reflections were measured on a Hilger–Watts four-circle diffractometer within a 2θ sphere of 48°, using graphite-monochromated Mo-K_α radiation and an ω/2θ scan mode. The intensity data were corrected for Lorentz and polarization effects. No absorption or extinction corrections were applied.

Attempts to solve the structure by direct methods failed. Only the sulphur atom could be located in the *E*-map and this position has been corroborated by the Patterson function. It was expected that the molecular structure of the title compound

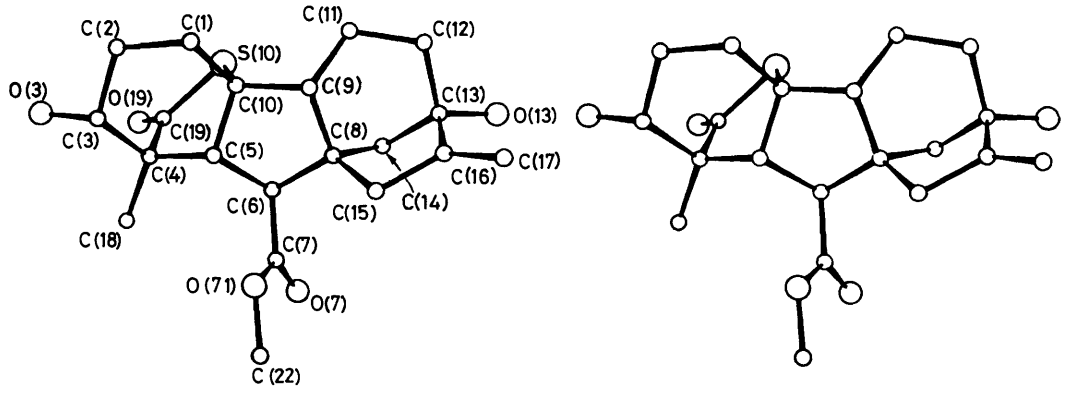


Figure 1. Stereoview of molecule (**25**)

would be similar to that of 1β-azidogibberellin A_1[19] with a sulphur atom replacing the oxygen atom O(10) in the lactone ring. The model was rotated around the sulphur atom and the residual was calculated by the computer program POSIT[20] in steps of 15° for three angles using 68 strong reflections with low indices.

The orientation of the molecule with the lowest *R*-value proved to be correct. A few cycles of full-matrix least-squares refinement of the non-hydrogen atoms with isotropic temperature factors by the program SHELX 76[21] reduced *R* to 0.17 for all reflections. A difference Fourier map revealed the positions of H(O3) and H(O13) while the rest of the hydrogen atoms were generated geometrically. The anisotropic refinement of the non-hydrogen atoms led to the final value of *R* 0.0756 for 1 271 reflections with $I > 3\sigma(I)$. The final atomic positional parameters are listed in the Table. In the last least-squares cycle the maximum LS shift/error was 0.43. Figure 1 shows a stereoview of the molecule. Bond distances, bond angles, and endocyclic torsion angles are given in Figure 2. The estimated standard deviations vary 0.008—0.012 Å for the bond distances, from 0.6—0.8° for the bond angles, and from 0.7—0.9° for the torsion angles.*

Ring A in the title compound adopts a chair conformation puckered in the fusion area C(4), C(5), C(10) and flattened in the area C(1), C(2), C(3) but less distorted than in the structure of 1β-azidogibberellin A_1.[19] The maximum value of the asymmetry parameter[22] is $\Delta C_S^{C(1)}$ 15.3°. Ring E has a nearly ideal C(5)-envelope conformation, as shown by the torsion angle C(10)–S(10)–C(19)–C(4) 0.1°. Compared with 1β-azidogibberellin A_1 the strain in this part of the molecule is relieved by the introduction of the sulphur atom. This can also be seen from the root-mean-square values of the deviations from the ideal tetrahedral angle of α 109.47° defined by $\delta(i) = \sum_{j=1-6}^{6}(\alpha_{ij} - \alpha)^2/6$ where α_{ij} is the bond angle *j* at the atom *i*. The values $\delta(i)$ for C(4) and C(10), indicating the extent of distortion, are 3.7° and 4.3°, and are smaller than the corresponding values of 5.5° and 4.9°, respectively, in 1β-azidogibberellin A_1. Ring B approximates to a 5β-envelope conformation distorted towards a 5β,10α-half-chair. Ring C has a boat conformation while the ring conformation of D is half-chair, as can be seen from Figure 2. The bond distance C(9)–C(10) 1.540(11) Å corresponds to the expected value, while in gibberellins with a γ-lactone ring this bond length is significantly shortened. The distances S(10)–C(10) 1.860(9) Å and S(10)–C(19) 1.799(8) Å are somewhat longer than the expected values for the bond lengths from sulphur to sp^3- and sp^2- hybridized C atoms.

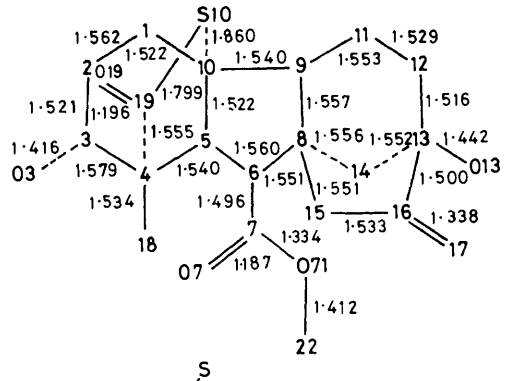
Table. Fractional atomic co-ordinates for compound (**25**), with e.s.d.s in parentheses

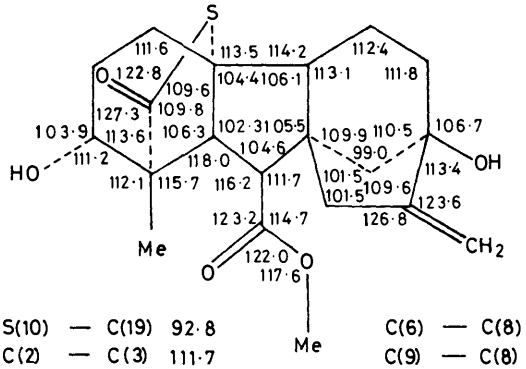
Atom	*x*	*y*	*z*
C(1)	0.269 5(3)	0.361 1(16)	0.283 9(7)
C(2)	0.268 6(3)	0.286 0(17)	0.413 5(7)
C(3)	0.320 9(4)	0.325 7(16)	0.484 5(6)
C(4)	0.370 1(3)	0.251 4(16)	0.421 4(6)
C(5)	0.367 2(3)	0.371 5(16)	0.304 5(7)
C(6)	0.411 8(3)	0.334 8(14)	0.222 4(6)
C(7)	0.461 4(3)	0.456 4(15)	0.250 5(6)
C(8)	0.386 0(3)	0.385 2(16)	0.097 7(7)
C(9)	0.325 7(3)	0.367 0(17)	0.106 2(7)
C(10)	0.319 0(3)	0.288 8(15)	0.231 1(6)
C(11)	0.297 5(3)	0.230 3(17)	0.007 6(6)
C(12)	0.314 5(3)	0.284 6(17)	−0.113 4(7)
C(13)	0.372 3(4)	0.341 5(17)	−0.107 7(7)
C(14)	0.403 0(3)	0.237 3(14)	0.000 2(6)
C(15)	0.397 8(3)	0.603 0(16)	0.046 6(7)
C(16)	0.381 3(3)	0.569 9(15)	−0.084 0(8)
C(17)	0.377 7(5)	0.716 2(17)	−0.167 8(8)
C(18)	0.421 6(4)	0.274 8(19)	0.500 3(7)
C(19)	0.361 5(3)	0.017 9(19)	0.386 9(7)
C(22)	0.499 5(4)	0.772 8(23)	0.318 8(9)
O(3)	0.316 7(3)	0.213 2(13)	0.589 7(5)
O(7)	0.503 4(2)	0.395 3(14)	0.229 4(6)
O(71)	0.454 6(2)	0.645 3(12)	0.296 2(5)
O(13)	0.391 1(2)	0.272 4(11)	−0.216 2(4)
O(19)	0.375 5(3)	−0.132 4(12)	0.443 6(6)
S(10)	0.324 0(1)	0.000 0(0)	0.246 6(2)

Each molecule is linked by hydrogen bonds to adjacent molecules related by a rotation axis O(13) ... O(7^{i}) 2.825(9) Å [symmetry code (i) $1 - x, y, -z$] and by translation in the *c*-direction O(3) ... O(13^{ii}) 2.817(9) Å with (ii) $x, y, z + 1$. The resulting molecular packing is given in Figure 3.

With regard to the biological activity of the thiogibberellins prepared in this study, the 7-thio analogue (**10**) and (**13**) were shown to undergo easy nucleophilic substitution of the SH group to afford the parent phytohormone even under the applied conditions of the bioassay. Therefore, the resulting bioactivities of compounds (**10**) and (**13**) have no significance. However, the stable 19,10-thio-3-epi-GA_1 (**26**), tested *e.g.* in the dwarf maize bioassay, showed an activity higher than that for GA_1. Detailed biological results will be reported elsewhere.

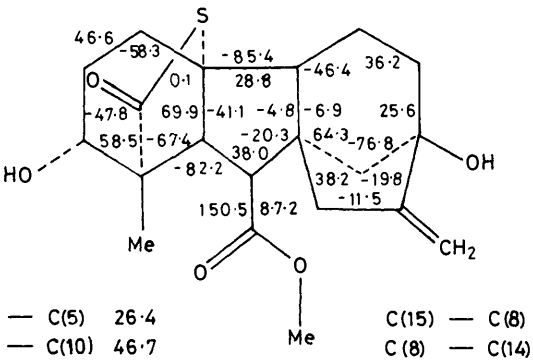
* *Supplementary data.* (see section 5.6.3 of Instructions for Authors, in the January issue). Anisotropic thermal parameters, hydrogen-atom parameters, and a full list of torsion angles have been deposited at the Cambridge Crystallographic Data Centre.





C(10) — S(10) — C(19) 92·8
C(1) — C(2) — C(3) 111·7
C(3) — C(4) — C(5) 105·7
C(3) — C(4) — C(19) 107·8
C(5) — C(4) — C(19) 105·3
C(18) — C(4) — C(19) 109·7
C(6) — C(8) — C(14) 114·3

C(6) — C(8) — C(15) 117·0
C(9) — C(8) — C(15) 108·5
S(10) — C(10) — C(1) 108·5
C(1) — C(10) — C(9) 115·9
O(13) — C(13) — C(14) 111·8
C(12) — C(13) — C(16) 111·9
C(14) — C(13) — C(16) 102·5
S(10) — C(10) — C(5) 104·3



C(19) — S(10) — C(10) — C(5) 26·4
C(19) — C(4) — C(5) — C(10) 46·7
S(10) — C(10) — C(5) — C(4) −46·1
S(10) — C(19) — C(4) — C(5) −26·1

C(15) — C(8) — C(14) — C(13) −50·5
C(8) — C(14) — C(13) — C(16) 42·7

Figure 2. Bond lengths (Å), bond angles (°), and selected torsion angles (°) for compound (**25**)

Experimental

M.p.s were determined on a Boetius hot-stage microscope and are corrected. Specific rotations were determined in EtOH if not otherwise stated. I.r. spectra were recorded on a Zeiss instrument Specord 75 IR for Nujol mulls. U.v. spectra were obtained on a Zeiss Specord UV-VIS instrument, and o.r.d. measurements were made on a Jasco spectropolarimeter ORD-UV5 for MeOH solutions. Routine mass spectra were obtained with the mass spectrometer of the 'Research Institute Manfred von Ardenne,' Dresden. High-resolution mass spectra were obtained with an AE MS 902S spectrometer. ^{1}H N.m.r. spectra were recorded on a Varian HA-100 (100 MHz) or Bruker WP-200 (200 MHz) instrument for [$^2H_6$]acetone solutions with tetramethylsilane as the internal standard. The photochemical reactions were performed in a Reading photoreactor (254 nm) with a quartz vessel at 15–20 °C under argon. Column chromatography was performed on Woelm silica gel for partition, and t.l.c. on Merck Kieselgel G-coated plates (0.5 nm thickness; 30 min activation at 120 °C before use; detection with 85% H_2SO_4 and 15 min heating at 110 °C).

General Procedure for the Synthesis of 7-Thiogibberellins from Symmetric Anhydrides.—Dry hydrogen sulphide was bubbled for 4 h through a solution of the gibberellin anhydride (2 g) in absolute pyridine (50 ml) at room temperature. The mixture was evaporated to dryness under reduced pressure and the residue was chromatographed on silica gel (100 g) with mixtures of chloroform–ethyl acetate as eluant (t.l.c. monitoring). Combined fractions were evaporated to dryness under reduced pressure. In this way the following compounds were prepared.

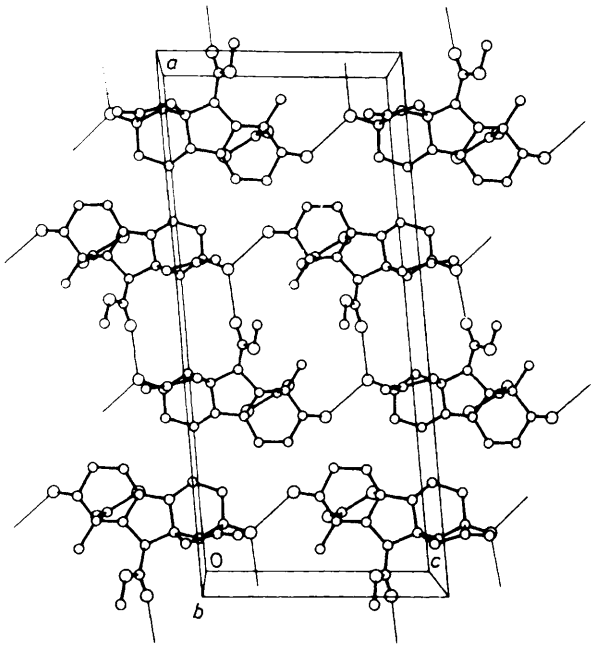


Figure 3. Molecular packing viewed along *b*. The hydrogen bonds are indicated by thin lines

(i) 7-Thioepiallogibberic *S*-acid (**8**) (55 mg) was obtained from GA_3 anhydride (**4**)[8] (2 g) as a minor product upon elution of the column with chloroform–ethyl acetate 7:3 v/v and crystallized from acetone–n-hexane, m.p. 116—118 °C; $[\alpha]_D^{24}$ +36.2° (*c* 0.353); R_F 0.58 (chloroform–methanol 9:1); $\nu_{max.}$ 890, 1 599, 1 692, 1 710, and 3 400 cm^{-1}; $\lambda_{max.}$ 238 (ε 8 240) and 280 nm (840); o.r.d. $[M]_{306}$ +2 840°, $[M]_{282}$ −1 015°, $[M]_{266}$ −3 550° (*a* +64); δ 2.26 (s, 18-H_3), 3.44 (m, 9-H), 3.98 (s, 6-H), 4.92 and 5.11 (17-H_2), and 7.1 (m, 1-, 2-, and 3-H); *m/z* 300 (M^+, 6%), 282 (19), 270 (48), 266 (77), 255 (29), 238 (95), 239 (100), and 221 (91).

(ii) 7-*Thiogibberellin* A_3 (S-*acid form*) (**10**) (614 mg) was obtained, from GA_3 anhydride (**4**) (2 g), upon elution with chloroform–ethyl acetate 4:6, and was crystallized from ethyl acetate–n-hexane, m.p. 201—206 °C (decomp.); $[\alpha]_D^{25}$ +54.5° (*c* 0.318); R_F 0.41 (chloroform–methanol 9:1) (Found: M^+, 362.2155. $C_{19}H_{22}O_5S$ requires *M*, 362.2093); $\nu_{max.}$ (KBr) 893, 1 655, 1 720, 1 760, 3 068, and 3 440 cm^{-1}; $\lambda_{max.}$ 242 nm (2 345); o.r.d. $[M]_{298}$ +5 320° and $[M]_{260}$ −10 050° (*a* +153); δ 1.25 (s, 18-H_3), 3.33 (d, *J* 10 Hz, 6-H), 3.35 (d, *J* 10 Hz, 5-H), 4.02 (d, *J* 3 Hz, 3-H), 4.91 and 5.23 (m, 17-H_2), 5.90 (dd, *J* 9, *J'* 3 Hz, 2-H), 6.40 (d, *J* 9 Hz, 1-H); *m/z* 362 (M^+, 13%), 360 (M^+ − 2, 12), 344 (20), 328 (32), 317 (42), 300 (62), 284 (100), 256 (55), and 239 (90). Further elution of the column with chloroform–ethyl acetate 1:9 gave GA_3 (**1**) (1 g).

(iii) 3,13-*Di*-O-*acetyl*-7-*thiogibberellin* A_3 (S-*acid form*) (**11**) (312 mg) was obtained from 3,3', 13,13'-tetra-acetylgibberellin A_3 anhydride (**5**)[9] (2 g), upon elution with n-hexane–chloroform 3:7, rechromatography, and crystallization from acetone–n-hexane, m.p. 223—226 °C (decomp.); $[\alpha]_D^{25}$ +183.0° (*c* 0.380); R_F 0.69 (chloroform–ethyl acetate 9:1) (Found: M^+ − 2, 444.1232. $C_{23}H_{24}O_7S$ requires *m/z*, 444.1243); $\nu_{max.}$ 898, 1 235, 1 708, 1 730, 1 740, and 1 778 cm^{-1}; $\lambda_{max.}$ 238 nm (2 630); o.r.d. $[M]_{298}$ +8 850° and $[M]_{264}$ −2 540 (*a* +114); δ 1.17 (18-H_3), 2.01 and 2.062 × OAc), 3.11 (d, *J* 10 Hz, 6-H), 3.37 (d, *J* 10 Hz, 5-H), 4.96 and 5.15 (17-H_2), 5.28 (d, *J* 3 Hz, 3-H), 5.80 (dd, *J* 9, *J'* 3 Hz, 2-H), and 6.30 (d, *J* 9 Hz, 1-H); *m/z* 444 (M^+ − 2, 4%), 428 (4), 412 (4), 402 (9), 384 (9), 370 (9), 352 (32), 342 (13), 326 (16), 308 (13), 298 (10), 281 (100), 266 (46), 238 (66), and 221 (99). Further elution of the column with n-hexane–chloroform 3:7 gave 3,13-di-*O*-acetyl-GA, (**2**) (1.3 g).

(iv) 7-*Thiogibberellin* A_1 (S-*acid form*) (**13**) (632 mg) was obtained from GA_1 anhydride (**6**)[10] (2 g), upon elution with chloroform–ethyl acetate 3:7, and was crystallized from ethyl acetate–n-hexane, m.p. 258—261 °C (decomp.); $[\alpha]_D^{26}$ +41.1° (*c* 0.294); R_F 0.41 (chloroform–methanol 9:1) (Found: M^+ −2, 362.2119. $C_{19}H_{22}O_5S$ requires *m/z*, 362.2093); $\nu_{max.}$ (KBr) 905, 1 720, 1 760, 3 070, and 3 440 cm^{-1}; $\lambda_{max.}$ 242 nm (2 100); o.r.d. $[M]_{300}$ +6 110° and $[M]_{260}$ −15 950° (*a* +220); δ 1.09 (s, 18-H_3), 3.09 (d, *J* 10 Hz, 6-H), 3.25 (d, *J* 10 Hz, 5-H), 3.68 (d, *J* 3 Hz, 3-H), and 4.83 and 5.14 (m, 17-H_2); *m/z* 364 (M^+, 33%), 362 (M^+ − 2, 76), 346 (52), 330 (85), 318 (32), 302 (63), 284 (58), 258 (100), and 240 (69). Further elution of the column with chloroform–ethyl acetate 3:7 gave GA_1 (**3**) (1.25 g).

3,13-*Di*-O-*acetyl*-7-*thiogibberellin* A_3 (S-*Acid Form*) (**11**).—(a) Via *sulphydrolysis of the mixed anhydride* (7). To a solution of diacetate (**2**) (861 mg) in absolute tetrahydrofuran (THF) (3 ml) and triethylamine (0.28 ml) at −5 °C was added a solution of ethylchloroformate (217 mg) in absolute THF (0.5 ml). The mixture was kept for 1 h at the same temperature, and the precipitate was then filtered off. To the filtrate of the mixed anhydride (**7**) was added a solution of triethylamine (0.28 ml) in absolute THF (10 ml), and dry hydrogen sulphide was bubbled for 2 h into the mixture at −20 °C. After evaporation to dryness under reduced pressure, the residue was chromatographed on silica gel (50 g). The fractions eluted with light petroleum–chloroform 3:7 v/v gave the title compound (**11**) (266 mg). Further elution with chloroform yielded starting compound (**2**) (365 mg).

(b) Via *acetylation of the* 7-*thiogibberellin* A_3 (**10**). A solution of compound (**10**) (86 mg) in a mixture of dry pyridine (1 ml) and acetic anhydride (1 ml) was left at room temperature for 7 days. Removal of the solvent under reduced pressure gave a gum which was chromatographed on silica gel to give, upon elution with n-hexane–chloroform 7:3 the title compound (**11**) (62 mg).

7-*Thiogibberellin* A_3 (S-*Acid Form*) *Methyl Ester* (**12**).—A solution of the thio-GA_3 (**10**) (71 mg) in ethyl acetate (5 ml) was treated with an excess of ethereal diazomethane for 0.2 h at room temperature. Removal of the solvent under reduced pressure gave a gum, which was chromatographed on silica gel. Upon elution with n-hexane–chloroform 4:6, the *S*-methyl ester (**12**) (39 mg) was obtained and was crystallized from acetone–n-hexane, m.p. 128—132 °C; $[\alpha]_D^{21}$ +49.3° (*c* 0.306); R_F 0.52 (chloroform–methanol 9:1); $\nu_{max.}$ 894, 1 680, 1 765, and 3 300 cm^{-1}; $\lambda_{max.}$ 236 nm (3 840); o.r.d. $[M]_{290}$ +1 540° and $[M]_{244}$ −1 080° (*a* +26); δ 1.15 (s, 18-H_3), 2.35 (s, SMe), 2.95 (d, *J* 10 Hz, 6-H), 3.38 (d, *J* 10 Hz, 5-H), 4.06 (3-H), 4.85 and 5.20 (m, 17-H_2), 5.95 (dd, *J* 9, *J'* 3 Hz, 2-H), and 6.47 (d, *J* 9 Hz, 1-H); *m/z* 376 (M^+, 27%), 358 (33), 328 (70), 314 (72), 300 (60), 283 (64), 266 (76), 256 (60), and 239 (100). Upon further elution with chloroform–ethyl acetate 6:4, starting 7-thio-GA_3 (**10**) (16 mg) was obtained.

7-*Thiogibberellin* A_1 (S-*Acid Form*) *Methyl Ester* (**14**).—The thio-GA_1 (**13**) (50 mg) was treated with diazomethane and worked up as described for compound (**12**), to give amorphous *S*-methyl ester (**14**) (27 mg), $[\alpha]_D^{29}$ +37.4° (*c* 0.303); R_F 0.52; $\nu_{max.}$ 900, 1 685, 1 760, and 3 400 cm^{-1}; *m/z* 378 (M^+, 65%), 360 (16), 346 (27), 330 (97), 303 (100), 266 (35), and 241 (65). Upon further elution with chloroform–ethyl acetate 6:4, starting 7-thio-GA_1 (**13**) (26 mg) was obtained.

13-O-*Acetyl*-7-*thioepiallogibberic* S-*Acid* (**9**).—Compound (**8**) (30 mg) was acetylated for 7 days at room temperature and worked up as described for diacetate (**11**). Chromatography on

silica gel gave, upon elution with n-hexane–chloroform 4:6, the amorphous 13-acetate (**9**) (14 mg), $[\alpha]_D^{21}$ +95.7° (*c* 0.193 in chloroform); R_F 0.70 (chloroform); $\nu_{max.}$ 890, 1 240, 1 600, 1 710, and 1 740 cm^{-1}; *m/z* 342 (M^+, 19%), 325 (7), 308 (48), 299 (10), 281 (100), 248 (38), 238 (74), and 221 (97).

15α-*Mercapto*-7-*thiogibberellin* A_3 (S-*Acid Form*) 7,15α-*Thiolactone* (**15**).—A solution of the 7-thio-GA_3 (**10**) (136 mg) in ethyl acetate (100 ml) was irradiated for 2 h (t.l.c. monitoring). The solvent was evaporated off under reduced pressure and the obtained residue was chromatographed on silica gel (7 g). From the fractions eluted with chloroform–ethyl acetate 9:1, GA_3 *thiolactone* (**15**) (43 mg) was obtained, and was crystallized from acetone–n-hexane, m.p. 263 °C (decomp.); $[\alpha]_D^{24}$ −126.0° (*c* 0.298); R_F 0.43 (chloroform–methanol 9:1) (Found: M^+, 360.1027. $C_{19}H_{20}O_5S$ requires *M*, 360.1032); $\nu_{max.}$ 890, 1 680, 1 760, 3 430, and 3 480 cm^{-1}; $\lambda_{max.}$ 241 nm (3 150); o.r.d. $[M]_{300}$ +430° and $[M]_{250}$ −14 350° (*a* +148); δ 1.29 (s, 18-H_3), 2.97 (s, 5- and 6-H), 3.99 (d, *J* 3 Hz, 3-H), 4.55 (d, *J* 2 Hz, 15β-H), 4.81 and 5.23 (d, *J* each 2 Hz, 17-H_2), 5.80 (dd, *J* 9, *J'* 3 Hz, 2-H), and 6.27 (d, *J* 9 Hz, 1-H); *m/z* 360 (M^+, 100%), 253 (6), 237 (7).

3,13-*Di*-O-*acetyl*-15α-*mercapto*-7-*thiogibberellin* A_3 (S-*Acid Form*) 7,15α-*Thiolactone* (**16**).—A solution of thiolactone (**15**) (37 mg) was acetylated for 7 days at room temperature and worked up as described for diacetate (**11**). Chromatography on silica gel gave, upon elution with chloroform–ethyl acetate, the diacetate (**16**) (18 mg), which was crystallized from chloroform–n-hexane, m.p. 247 °C (decomp.); $[\alpha]_D^{24}$ −10° (*c* 0.286); R_F 0.66 (chloroform–ethyl acetate 9:1); $\nu_{max.}$ 890, 1 240, 1 700, 1 740, and 1 780 cm^{-1}; $\lambda_{max.}$ 240 nm (3 630); o.r.d. $[M]_{300}$ +2 520° and $[M]_{251}$ −18 700° (*a* +212); *m/z* 444 (M^+, 42%), 402 (68), 384 (52), 358 (31), 340 (45), 324 (36), 314 (36), 298 (65), 280 (100), 270 (67), 262 (51), 252 (77), 238 (99), and 220 (94).

15α-*Mercapto*-7-*thiogibberellin* A_1 (S-*Acid Form*) 7, 15α-*Thiolactone* (**17**).—The 7-thio-GA_1 (**13**) (147 mg) was irradiated in ethyl acetate and worked up as described for the preparation of compound (**15**). Upon silica gel chromatography, elution with chloroform gave the thiolactone (**17**) (55 mg), which was crystallized from acetone–n-hexane, m.p. 165 °C (decomp.); $[\alpha]_D^{26}$ +30.5° (*c* 0.109); R_F 0.43 (chloroform–methanol 9:1); $\nu_{max.}$ 900, 1 680, 1 760, and 3 400 cm^{-1}; $\lambda_{max.}$ 233 (2 820); o.r.d. $[M]_{290}$ +200° and $[M]_{250}$ −2 940° (*a* +31); *m/z* 362 (M^+, 100%), 346 (15), 334 (9), and 318 (7).

Thiocarboxylic Acid (**22**).—To a solution of the acid (**20**)[14] (1.1 g) in absolute benzene (7 ml) was added freshly distilled thionyl chloride (7 ml) and the mixture was left at room temperature for 1 h. Evaporation under reduced pressure gave a gummy, crude chloride (**21**), which was dissolved in absolute pyridine (60 ml). Through this solution was bubbled a stream of dry hydrogen sulphide for 2 h at room temperature. Evaporation to dryness under reduced pressure gave a residue, which was chromatographed on silica gel. Elution with n-hexane–chloroform 7:3 yielded the *thiocarboxylic acid* (**22**) (248 mg), which was rechromatographed, and was then crystallized from acetone–n-hexane, m.p. 198 °C (decomp.); $[\alpha]_D^{24}$ −9.8° (*c* 0.268); R_F 0.32 (chloroform–ethyl acetate 98:2) (Found: M^+, 462.1732. $C_{24}H_{30}O_7S$ requires *M*, 462.1712); $\nu_{max.}$ 890, 1 710, 1 720, and 1 735 cm^{-1}; $\lambda_{max.}$ 245 nm (2 020); o.r.d. $[M]_{310}$ +240° and $[M]_{262}$ −4 000 (*a* +42); δ 1.40 (s, 18-H_3), 1.96 and 2.06 (s, AcO), 2.75 (d, *J* 6 Hz, 6-H), 3.69 (s, OMe), 4.95 (3- and 17-H), 5.25 (17-H), and 5.48 (1-H); *m/z* 462 (M^+, 4%), 429 (14), 420 (14), 402 (15), 400 (17), 369 (13), 342 (57), 311 (18), 299 (39), 281 (100), 240 (50), 223 (99). Further elution of the column with n-hexane–chloroform 7:1 and rechromatography gave thiolactone (**23**) (61 mg). Elution with chloroform yielded starting carboxylic acid (**20**) (587 mg).

3,13-*Di*-O-*acetyl*-19,10-*thiogibberellin* A_1 *Methyl Ester* (**23**).—(a) Via *photolysis of compound* (**22**). The thiocarboxylic acid (**22**) (70 mg) was irradiated in absolute THF (20 ml) for 2 h (t.l.c. monitoring). The solvent was evaporated off under reduced pressure and the residue was chromatographed on silica gel (4 g). From the fractions eluted with n-hexane–chloroform 7:3 the *thio-*GA_1 *derivative* (**23**) (50 mg) was obtained, and was crystallized from acetone–n-hexane, m.p. 183 °C (decomp.); $[\alpha]_D^{25}$ +69.9° (*c* 0.344); R_F 0.53 (chloroform–ethyl acetate 98:2) (Found: M^+, 462.1742. $C_{24}H_{30}O_7S$ requires *M*, 462.1712); $\nu_{max.}$ 890, 1 235, 1 255, 1 702, 1 730, and 1 745 cm^{-1}; $\lambda_{max.}$ 242 nm (3 030); o.r.d. $[M]_{304}$ −1 440°, $[M]_{256}$ +22 280°, and $[M]_{230}$ +17 250° (*a* −237); δ 0.88 (s, 18-H_3), 1.97 and 2.09 (s, OAc), 2.99 (d, *J* 11 Hz, 6-H), 3.18 (d, *J* 11 Hz, 5-H), 3.70 (s, OMe), 4.76 (3-H), and 4.92 and 5.13 (m, 17-H_2); *m/z* 462 (M^+, 21%), 431 (20), 420 (43), 402 (45), 379 (41), 361 (20), 242 (85), 319 (21), 300 (45), 282 (100), 241 (68), and 223 (70).

(b) Via *hydrogen sulphide treatment of compound* (**22**) *in pyridine*. Dry H_2S was bubbled through a solution of thiocarboxylic acid (**22**) (20 mg) in absolute pyridine (30 ml) for 7 h at room temperature. Work-up and chromatography on silica gel (2 g) gave, upon elution with n-hexane–chloroform 7:3, the thio-GA_1 derivative (**23**) (10 mg).

(c) Via *prolonged hydrogen sulphide treatment of chloride* (**21**) *in pyridine*. Chloride (**21**) was prepared from acid (**20**) (30 mg) and thionyl chloride (0.32 ml) in absolute benzene (1 ml) as described for compound (**22**). The crude product was dissolved in absolute pyridine (10 ml), and dry hydrogen sulphide was bubbled through the solution for 7 h at room temperature. Evaporation, and chromatography on silica gel (3 g) gave, upon elution with n-hexane–chloroform 7:1, the thio-GA_1 derivative (**23**) (11 mg). Further elution of the column with the same eluant yielded starting acid (**20**) (18 mg).

19,10-*Thio*-3-*epigibberellin* A_1 *Methyl Ester* (**25**).—The diacetate (**23**) (39 mg) was dissolved in 0.2M-methanolic sodium methoxide (1.69 ml) and the solution was left for 4 h at room temperature. After addition of dil. acetic acid (0.5 ml) the solution was evaporated to dryness under reduced pressure and the residue was chromatographed on silica gel (3 g). Elution with chloroform gave the *thio-*3-*epi-*GA_1 *methyl ester* (**25**) (22 mg), which was crystallized from acetone–n-hexane, m.p. 233 °C (decomp.); $[\alpha]_D^{24}$ +16.0° (*c* 0.285); R_F 0.57 (chloroform–ethyl acetate 1:1) (Found: M^+, 378.1523. $C_{20}H_{26}O_5S$ requires *M*, 378.1501); $\nu_{max.}$ 1 690, 1 725, and 3 400 cm^{-1}; $\lambda_{max.}$ 243 nm (2 670); o.r.d. $[M]_{300}$ −2 230°, $[M]_{252}$ +8 810°, and $[M]_{226}$ −9 050° (*a* −110.5); δ 0.99 (s, 18-H_3), 2.54 (d, *J* 11 Hz, 6-H), 2.99 (d, *J* 11 Hz, 5-H), 3.58 (3β-H), 3.68 (s, OMe), and 481 and 5.16 (17-H_2); *m/z* 378 (M^+, 100%), 360 (2), 350 (16), 346 (11), 332 (3), 328 (3), 319 (11), 300 (11), 285 (8), 272 (8), 267 (5), 257 (11), 241 (13), and 239 (10).

19,10-*Thio*-3-*epigibberellin* A_1 (**26**).—The methyl ester (**25**) (38 mg) was shaked with 0.5M-lithium propanethiolate solution in hexamethylphosphoric triamide (1.05 ml)[23] under argon for 5 h at room temperature. Ice was added, and the solution was acidified with 2M-hydrochloric acid and extracted with ethyl acetate. The organic layer was dried with Na_2SO_4 and concentrated to dryness under reduced pressure. The residue was chromatographed on silica gel (3 g). Elution with chloroform–ethyl acetate 9:1 gave the *thio-*3-*epi-*GA_1 (**26**) (28 mg), which was crystallized from ethyl acetate–n-hexane, m.p. 165 °C (decomp.); $[\alpha]_D^{24}$ +18.4° (*c* 0.298); R_F 0.60 (chloroform–ethyl acetate–acetic acid 5:4:1) (Found: M^+, 364.1320. $C_{19}H_{24}O_5S$ requires *M*, 364.1345); $\nu_{max.}$ 1 690, 1 710, and 3 400

cm^{-1}; $\lambda_{max.}$ 242 nm (1 600); o.r.d. $[M]_{302}$ −3 000°, $[M]_{254}$ +8 800°, and $[M]_{228}$ −9 000° (a −118); δ 1.06 (s, 18-H_3), 2.52 (d, J 11 Hz, 6-H), 2.98 (d, J 11 Hz, 5-H), 3.59 (3β-H), and 4.80 and 5.16 (17-H_2).

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