Stimulation of *Striga hermonthica* Seed Germination by 11β,13-Dihydroparthenolide

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Germination tests of *Striga hermonthica* seeds were performed using 11β,13-dihydroparthenolide (DHP), a sesquiterpene lactone from *Ambrosia artemisifolia* (common ragweed). In each test series aqueous solutions with acetone or methanol were used as negative controls. Test solutions of the strigol analogue 2-methyl-4-((2-oxo-2,3,3a,8b-tetrahydro-4*H*-indeno[1,2-*b*]furan-3-ylidene)methoxy)-but-2-en-4-olide (GR 24) in methanol as carrier system were used as positive controls. A decrease in concentration of DHP from 10^{-3} to 10^{-20} M resulted in several unusual "minima" and "maxima" of stimulatory activities, suggesting that the mode of action of DHP could be hormonal. The crystal structure of DHP was determined by X-ray crystallography, and the use of molecular modeling in predicting the biologically active conformations is proposed.

Keywords: *Striga hermonthica; Scrophulariaceae; 11β,13-dihydroparthenolide; seed germination; X-ray crystallography; molecular mechanics*

INTRODUCTION

Chlorophyllous obligate root parasites of the genus *Striga* (Scrophulariaceae) are a major constraint to crop production in West and East Africa and parts of Asia (Sauerborn, 1991). In particular, *Striga hermonthica* (Del.) Benth., which causes losses of yield up to 70% in major food crops of the Gramineae including sorghum (*Sorghum bicolor* (L.) Moench), millet (*Pennisetum americanum* (L.) K. Schum.), rice *(Oryza sativa* L.), and maize (*Zea mays* L.), is the species of greatest economic importance in Africa (Olivier *et al*., 1991).

Striga hermonthica has a high level of genetic diversity (Bharathalakshmi *et al*., 1990), even in comparison with other species that share its life-history features. Approximately 37 000 seeds are produced annually per *S. hermonthica* plant (Laing, 1984). High fecundity together with high viability results in a large seedbank which can rapidly build up and survive in the soil for many years. The seeds will not germinate unless they are exposed to an exogenous chemical stimulant (at suitable temperature and moisture) usually released from the roots of plants (Rugutt, 1996). After germination, endosperm nutrients can sustain the seedlings for 3−7 days. If the seedling does not attach to a host and successfully establish a parasitic link within this period, it dies (Worsham, 1987).

In the United States, *Striga* spp. are effectively eradicated using ethylene, which is injected into the soil (Eplee, 1984). This method is of limited application in developing countries because ethylene is a gas under relatively high pressure. In addition, its use for large-scale control requires transport and application equipment that either are not available or are not economically effective for the resource-poor farmers (Sand and Manley, 1990). In Africa, control methods for *S. hermonthica* include cultural procedures, hand-weeding, nonhost crop rotation, the use of nitrogen fertilizers, resistant crop cultivars, and germination stimulants (Musselman, 1987). Nonhost crop rotation is currently the most effective method and involves the use of crops whose exudates induce "suicidal" germination of *Striga* seeds but are themselves not parasitized. However, successful implementation of this effective method has been limited because little is known about the relative efficacy of nonhost crops or the crop-specific chemical compounds responsible for germination (Ariga and Berner, 1993; Berner *et al*., 1995).

Several years ago, the sesquiterpene strigol (**1**) was isolated (Cook *et al.*, 1966) from root exudates of cotton (*Gossypium hirsutum* L.), a nonhost plant, and found to be a highly potent germination stimulus for *Striga* spp. seeds (Cook *et al*., 1972, and references therein). Subsequently, a number of synthetic analogues have been synthesized (Johnson *et al*., 1981) and tested as *Striga* spp. germination stimulants (Pepperman *et al*., 1982). However, the difficult and expensive syntheses (MacAlpine *et al*., 1976) as well as the requirement of many different types of chemical equipment make the use of strigol (**1**) and its analogues prohibitively expensive for control of *Striga* spp. in small-scale farming (Vail *et al*., 1990).

Recently, Fischer *et al*. (1990, 1991) reported a number of strigol-like compounds, all sesquiterpene lactones (SLs) that stimulate germination of *Striga asiatica* seeds. Dihydroparthenolide (DHP, **2**), a sesquiterpene lactone found in *Ambrosia artemisifolia* (a nonhost plant), induced 70% germination at 10^{-9} M. This level of stimulation is close to that of strigol (**1**). Encouraged by this result, and as part of an ongoing program concerned with investigation of natural products as lead compounds for new biologically active compounds, we have screened various concentrations of DHP (**2**) on germination of temperature- and moisture-conditioned *S. hermonthica* seeds.

MATERIALS AND METHODS

General. All bioassays were done at the International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria.

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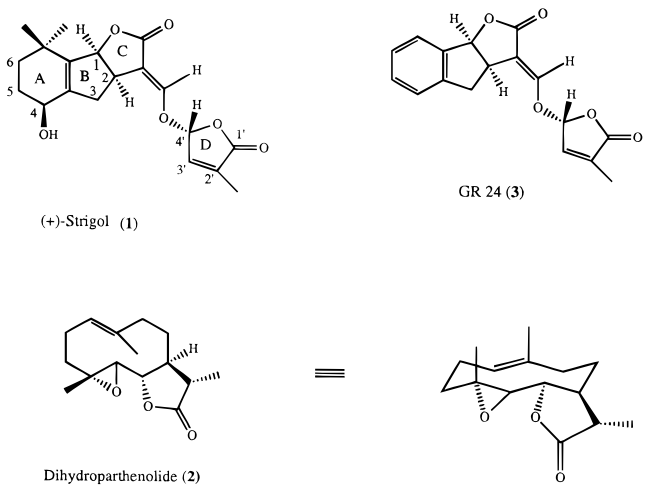


Seeds of *S. hermonthica* were collected in 1993 by D. K. Berner from sorghum plants in Bida, Nigeria. Viability of the seeds was estimated using tetrazolium red (2,3,5-triphenyltetrazolium chloride) test (Hsiao *et al.*, 1979). Approximately 400 seeds were placed on a clean sheet of filter paper in a Petri dish. The filter paper was then saturated with 1.0% tetrazolium red solution (pH 7). The Petri dish was sealed and incubated at 35 °C for 5 days. Embryos of viable seeds stained pink when observed under a microscope. Very pale pink and unstained embryos indicated nonviable seeds. Viability experiments were repeated eight times, and the mean percentage viability was determined to be 87%.

Seed Conditioning. For each experiment, about 1440 *S. hermonthica* seeds were surface sterilized by completely immersing in 3.6 mL of 1% (w/v) sodium hypochlorite (NaOCl) to which three drops of the detergent Tween 80 [polyoxyethylene(20) sorbitan monooleate] were added. After shaking, the mixture was allowed to stand for 4 min. Floating seeds were decanted and discarded. The remaining seeds were transferred to a Buchner funnel lined with Whatman no. 1 filter paper and rinsed several times with sterile deionized water until the chlorine smell disappeared. The filter paper containing the surface sterilized seeds was removed and air-dried. The seeds were then conditioned on 100 glass fiber disks (5.0 mm diameter) arranged on top of two sterile Whatman no. 1 filter papers in 9.5 cm diameter Petri dishes. Then 25−40 seeds were carefully sprinkled on each glass fiber disk. The Petri dishes were sealed with parafilm and incubated at 28 °C in the dark for 10 days. Throughout the conditioning period the filter papers were saturated with sterile deionized water.

Measurement of Germination. Pure samples (>98%) of DHP (**2**) and GR 24 (**3**) were provided by N. H. Fischer and D. K. Berner, respectively. Because of the low water solubility of both compounds, aqueous stock solutions were prepared using acetone and methanol as carriers. Two sets of stock solutions of 10^{-3} M DHP (**2**) were prepared by dissolving 2.5 mg in 0.1 mL of of acetone or methanol and diluted with 9.9 mL of sterile deionized water to obtain the desired concentration. The 10^{-3} M GR 24 (**3**) was similarly prepared by dissolving 2.98 mg in 0.1 mL of methanol before diluting with 9.9 mL of sterile deionized water. The stock solutions were refrigerated at about 4 °C. For each experiment, fresh solutions of lower concentrations were prepared by serial dilution of stock solutions and evaluated as germination stimulants for conditioned *S. hermonthica* seeds. In all experiments, conditioned seeds treated with sterile deionized water containing 0.1% (v/v) acetone or methanol served as negative controls. For each test, 12 glass fiber disks containing (approximately 360) conditioned seeds were arranged in a 9.0 cm diameter sterile Petri dish lined with two (Whatman no. 1) filter papers which had been moistened with 2.0 mL of sterile deionized water. Using a micropipet, aliquots of 600 μL of test solutions were uniformly applied to each Petri dish containing disks of conditioned seeds. The Petri dishes were sealed with parafilm and incubated at 28 °C for 24 h. Germinated and nongerminated seeds were counted under a binocular dissecting microscope at 20× magnification. Seeds were considered to have germinated if the radicle had protruded from the seed coat. Because of the difficulties in reproducing *Striga* bioassays (Rugutt, 1996), it was necessary to replicate each test in three separate Petri dishes per experiment and repeat the experiments 12 times over a period of 3 weeks. The germination data were transformed to arc sine (Gomez and Gomez, 1984) and processed by analysis of variance (SAS, 1989).

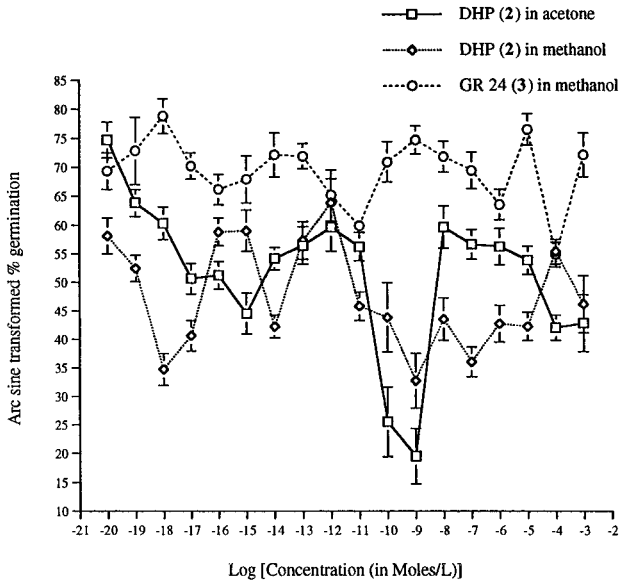


Figure 1. Effect of dihydroparthenolide (DHP, **2**) on gemination of conditioned *Striga hermonthica* seeds. Data are means ± SE of 12 different experiments performed in triplicate (P = 0.05). Control is GR 24 (**3**) in methanol. Small vertical lines: standard error of mean value.

X-ray Experimental. X-ray data for DHP (**2**) (atomic coordinates, thermal parameters, bond distances and angles, and observed and calculated structure parameters have been deposited with the Cambridge Crystallographic Data Center and can be obtained upon request from Dr. Olga Kennard, University Chemical Laboratory, 12 Union Road, Cambridge CB2 IEZ, UK): A colorless needle fragment of dimensions 0.55 × 0.40 × 0.40 mm was used for data collection on an Enraf-Nonius CAD4 diffractometer equipped with Cu Kα radiation (λ = 1.541 84 Å), and a graphite monochromator. Crystal data: $C_{15}H_{22}O_3$, M_r = 250.3, orthorhombic space group $P2_12_12_1$, a = 7.8964(5) Å, b = 9.0571(4) Å, c = 20.068(1) Å, V = 1435.2-(2) Å^3, Z = 4, d_c = 1.159 g cm^{-3}, T = 26°. Intensity data were measured by $\omega-2\theta$ scans of variable rate. Five octants of data were collected within the limits $2 < \theta < 75°$, and agreement between redundant data was R_{int} = 0.029. Data reduction included corrections for background, Lorentz, polarization, and absorption effects. Absorption corrections (μ = 6.0 cm^{-1}) were based on ψ scans, with minimum relative transmission coefficient 95.1%. Of 2949 unique data, 2870 had $I > \sigma(I)$ and were used in the refinement. The structure was solved by direct methods using SIRPOW.92 (Altomare *et al.*, 1994) and refined by full-matrix least squares, treating nonhydrogen atoms anisotropically, using the Enraf-Nonius MolEN programs (Fair, 1990). Hydrogen atoms were located using difference maps and refined isotropically, except for those of the methyl groups, which were placed in calculated positions. Convergence was achieved with R = 0.0368, R_w = 0.0460, and GOF = 2.210. The absolute configuration was determined by refinement of the mirror-image structure under identical circumstances, yielding R = 0.0370, R_w = 0.0463, and GOF = 2.221. The crystal structure is illustrated in Figure 2, and its coordinates are collected in Table 2.

Molecular Modeling. Due to our interest in the unique structural features responsible for *S. hermonthica* seed germination stimulatory activities in DHP (**2**) relative to both strigol (**1**) and GR 24 (**3**), MM calcuations were performed using a PCMODEL molecular mechanics (MM) program

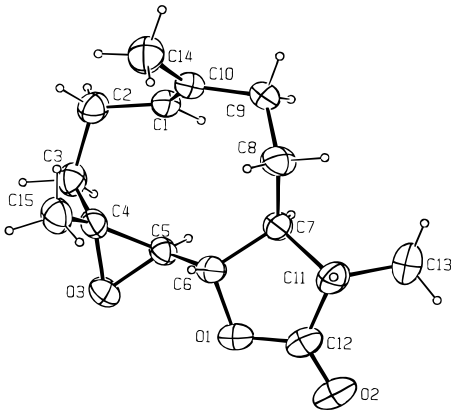
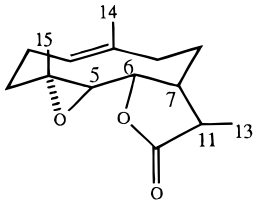


Figure 2. ORTEP drawing of 11β,13-dihydroparthenolide (DHP, **1**).

Table 1. Calculated Conformational Energies (kcal/mol), van der Waals Volume (VDW), and Dipole Moment (DP) of Conformations (2−2b) of 11β,13-Dihydroparthenolide (DHP) Molecule



conformation	Me-14	H-11	energy	VDW	DP
2	β	β	25.40	4.78	5.41
2a	β	α	26.88	5.12	5.41
2b	α	β	104.04	1.84	2.81

Table 2. Positional Parameters and Their Estimated SDS for Dihydroparthenolide (2)

atom	x	y	z	B_{eq} (Å^2)
O1	0.5422(1)	0.85320(9)	0.33911(5)	5.48(2)
O2	0.6955(2)	0.9908(1)	0.27040(6)	7.88(3)
O3	0.3633(1)	0.77499(9)	0.46094(5)	5.83(2)
C1	0.6443(1)	0.4084(1)	0.48361(6)	4.61(2)
C2	0.5128(2)	0.4136(2)	0.53754(7)	5.49(3)
C3	0.4253(2)	0.5661(2)	0.53881(6)	5.43(2)
C4	0.3752(1)	0.6160(1)	0.46987(7)	4.70(2)
C5	0.5051(1)	0.6971(1)	0.43259(6)	4.25(2)
C6	0.5275(1)	0.6976(1)	0.35864(6)	4.24(2)
C7	0.6939(2)	0.6272(1)	0.33493(5)	4.34(2)
C8	0.6927(2)	0.4612(1)	0.31830(6)	5.52(3)
C9	0.7624(2)	0.3587(1)	0.37245(7)	5.22(2)
C10	0.6378(2)	0.3314(1)	0.42745(6)	4.59(2)
C11	0.7363(2)	0.7233(2)	0.27464(7)	5.97(3)
C12	0.6619(2)	0.8706(2)	0.29263(7)	5.70(3)
C13	0.9215(3)	0.7326(3)	0.2557(1)	10.61(5)
C14	0.5067(2)	0.2164(2)	0.41198(9)	6.74(3)
C15	0.2339(2)	0.5327(2)	0.43682(9)	6.13(3)

developed by Burkert and Allinger (1982). This program uses the MMX force field. For DHP (**2**) and strigol (**1**), energy minimization calculations were carried out using the coordinates of X-ray crystallographic analyses by us (Table 2) and Coggon *et al*. (1973), respectively. In the case of the synthetic strigol analogue GR 24 (**3**), the coordinates of strigol (**1**) were used in MM calculations except in the A ring.

RESULTS

The curves of means of arc sine transformed percentage germination of *S. hermonthica* seeds plotted against the log [concentration] of DHP (**2**) and GR 24 (**3**) are shown in Figure 1. In all experiments, the negative controls did not show observable germination under the microscope. All concentrations of DHP (**2**) and GR 24 (**3**) showed some activity in stimulating germination of conditioned seeds. GR 24 (**3**) in methanol as a carrier system exhibited high stimulatory effects (54−79%) across all concentrations. Solutions of DHP (**2**) prepared in acetone not only exhibited a broader range of stimulation (19−75%) than those in methanol (32−64%) but also produced the highest stimulation of germination (75%) at 10^{-20} M. Solutions of DHP (**2**) in methanol showed two sets of "plateaus" of 42% and 59% germination between the 10^{-5}−10^{-6} M and 10^{-15}−10^{-16} M concentration range, respectively. Unlike DHP (**2**) in acetone, GR 24 (**3**) elicited a single "plateau" of 72% germination between 10^{-13} and 10^{-14} M. It is of interest to note that while 10^{-9} M solutions of DHP (**2**) in acetone and methanol showed the lowest germination stimulatory activities of 19 and 32%, respectively, a 10^{-9} M solution of GR 24 (**3**) elicited a high germination rate of 71%.

While NMR studies have shown that strigol (**1**), DHP (**2**), and GR 24 (**3**) are the stable conformations at room temperature (Cook *et al*., 1972; Johnson *et al*., 1981; Fischer *et al*., 1989), we cannot be certain that they are the only forms populated since the NMR method is unable to distinguish between a single "fixed" conformation or a mixture of conformations in rapid equilibrium resulting in an averaged symmetry equivalent to that of a single conformation. To solve this enigma, the utilization of MM calculations on the conformational analysis of germination stimulants is inevitable (Osawa *et al*., 1989; Shimazaki *et al*., 1991, and references therein). The conformational energy (CE), van der Waals volumes (VDW), and dipole moments (DP) of strigol (**1**) and GR 24 (**3**), were determined to be 43.71 kcal/mol, 2.35, 6.38, and 45.37 kcal/mol, 1.92 and 5.60, respectively. Analysis of the conformers of DHP (**2**) indicated that there are at least two conformers, **2** itself and **2a** (Table 1), which are candidates for the biologically active structure (within the context of our "stimulant−receptor interaction model" proposed below). The energy of conformer **2a** with H-11 in the α-orientation was within 1.48 kcal/mol of the energy of conformer **2**. The interconversion between the two conformers (**2** and **2a**) may thus be described as a large-amplitude torsional motion. However, the energy barrier for the interconversion of conformer **2** to **2b** (Me-14 in α-orientation) was calculated to be large (104.04 kcal/mol). The VDW of strigol (**1**) and GR 24 (**3**) are comparable but differ from that of DHP (**2**).

DISCUSSION

The theme of the song of those who sing the praises of classical *Striga* spp. germination stimulants is entitled "Potent if strigol-like". The lyrics create testing of a wide array of compounds for stimulatory activities. In the present communication, we have shown, for the first time, that DHP (**2**) is a highly active *S. hermonthica* seed germination stimulant (Figure 1). In accordance with previous results on *S. asiatica* by Fischer *et al*. (1990), we also observed several "minima" and "maxima" of stimulation of germination across a broad concentration range. An interesting and potentially important observation in the present study was the stimulation of germination of *S. hermonthica* seeds by DHP (**2**) at concentrations lower than 10^{-15} M. This activity is considerably higher than the activities of other natural compounds reported to affect seed germination (Duke, 1986) and suggests that DHP (**2**) may be representative

of a new class of plant growth hormones (Kleczkowski, 1995). The fact that 10^{-9} M solutions of DHP (**2**) induced the lowest stimulation may be interpreted as a lack of intrinsic hormone-like activity and that the ability to stimulate germination probably failed at this concentration. Indeed, additional experiments will be necessary to establish the actual factors responsible for this puzzling behavior.

Structural Differences. GR 24 (**3**) possesses an aromatic A ring and contains all of the features of strigol (**1**) with the exception of both of the *gem*-dimethyl groups and the introduction of formal double bonds in the 4,5- and 6,7-positions. In addition, GR 24 (**3**) does not have a hydroxyl group at the 4-position as does strigol (**1**). Other structural similarities of **1** and **3** that merit comment include the butenolide ring structure (D ring of strigol (**1**)) joined to another methyleneoxy bridge.

On the other hand, the structure of DHP (**2**), *per se*, offers several rationale for its stimulatory properties. Firstly, it has C-14 and C-15 lying *syn* on the β-face of the medium ring. Secondly, it does not contain the common alkylating α-methylene γ-lactone moiety. Thirdly, it has a germacranolide skeleton with a 4,5-epoxide of a 1(10), 4-cyclodecadiene system. Although the epoxide function is not directly accessible due to steric hindrance of the associated medium ring structure, it provides increased reactivity, as well as regio- and stereospecificity of subsequent intramolecular cyclizations (Fischer, 1991). The transannular cyclization generate a cationic center at C-10 which can be a receptor for biological nucleophiles, in particular, thiol-containing amino acids (Hall *et al*., 1977). It should be noted that DHP (**2**) is stable at room temperature and its lactone ring is similar to the butenolide ("D") ring of strigol (**1**), except that it lacks the C-2′═C-3′ double bond. Strigol (**1**) and its analogues undergo alkaline hydrolysis or degradation during storage, leading to variable results in *Striga* bioassays (Babiker *et al*., 1987, 1992). The absence of butenolide and C-13 exocyclic methylene moieties in DHP (**2**) makes it an attractive nontoxic candidate in *Striga* spp. control.

Stimulant−Receptor Interaction Model. The presence of receptor sites in *Striga* spp. seeds in which the reactive sites of germination stimulants attach and induce the production of ethylene has been proposed (Rugutt, 1996). Plausible features of receptor models may include (1) the use of the entire strigol (**1**) molecule in the construction of the models instead of parts of the molecule (2) the receptor has a specific and probably flexible van der Waals volume (VDW) (3) the use of the complete structure of different conformers in the calculations of conformational energies, and (4) addition of flexibility to the model with respect to the required location of the actiphore of the germination stimulants. The receptor sites may be complementary to a wide variety of functional groups including the double bond, lactone moiety, hydroxyl, and/or methyl groups.

The high activity of strigol (**1**) (Kendall *et al*., 1979) compared to its synthetic analogue GR 24 (**3**) (Babiker *et al*., 1987) indicate that not only the *gem*-dimethyl groups but also the hydroxyl group (OH-4) may efficiently interact with a hydrophobic region of the assumed receptor cavity, a region which is complementary to van der Waals surface of the substrate molecule (Liljefors *et al*., 1985). The shape of the part of the receptor cavity interacting with the A, B, C, and D rings of strigol (**1**) may be a deep "groove" or "pocket", at least fully circumscribing the actiphore (rings C and D). Thus, the ability of a compound to stimulate germination of *Striga* spp. seeds can be predicted on the basis of its ability to mimic strigol (**1**) with respect to the spatial locations of the crucial molecular parts (Hauck *et al*., 1992). The different stimulatory activities of DHP (**2**) and GR 24 (**3**) may be, according to our stimulant−receptor interaction model, due to the differences in binding capabilities to the receptor, or to form an "activated complex" with the receptor. This is reflected in the differences in the conformational energies (Table 1).

It is hoped (we are real optimists) that germination data obtained in our study can be extrapolated to screenhouse and, eventually, field experimentation. The results obtained by MM calculations should be of great value in establishing the groundwork in explaining structure−activity relationships of agricultural compounds that exhibit *Striga* stimulatory activities.

ABBREVIATIONS USED

Spp., species; MM, molecular mechanics; VDW, van der Waals volume; DP, dipole moment; CE, conformational energy; GR 24, 2-methyl-4-((2-oxo-2,3,3a,8b-tetrahydro-4*H*-indeno[1,2-*b*]furan-3-ylidene)methoxy)but-2-en-4-olide; DHP, 11βH,13-dihydroparthenolide; SLs, sesquiterpene lactones; Me, methyl.

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LITERATURE CITED

Altomare, A.; Cascarano, G.; Giacovazzo, C.; Guagliardi, A.; Burla, M. C.; Polidori, G.; Camalli, M. SIRPOW.92 - a program for automatic solution of crystal structures by direct methods optimized for powder data. *J. Appl. Crystallogr.* **1994**, *27*, 435−436.

Ariga, E. S.; Berner, D. K. Response of *Striga hermonthica* seeds to different germination stimulants and stimulant concentrations. *Phytopathology* **1993**, *83*, 1401.

Babiker, A. G. T.; Hamdoun, A. M.; Rudwan, A.; Mansi, N. G.; Faki, H. H. Influence of soil moisture on activity and persistence of the strigol analogue GR 24. *Weed Res.* **1987**, *27*, 173−178.

Babiker, A. G. T.; Parker, C.; Suttle, J. C. Induction of *Striga* seed germination by thidiazuron. *Weed Res.* **1992**, *32*, 243−248.

Berner, D. K.; Kling, J. G.; Singh, B. B. *Striga* Research and Control. A perspective from Africa. *Plant Dis.* **1995**, *79*, 652−660.

Bharathalakshmi; Werth, C. R.; Musselman, L. J. A study of genetic diversity among host-specific populations of the witchweed *Striga hermonthica* (Scrophulariaceae) in Africa. *Plant Syst. Evol.* **1990**, *172*, 1−12.

Burkert, U.; Allinger, N. L. *Molecular Mechanics*; ACS Monograph 177; American Chemical Society: Washington, DC, 1982; pp 1−314.

Coggon, P.; Luhan, P. A.; McPhail, A. T. Crystal and molecular structure of the germination stimulant strigol by X-ray analysis. *J. Chem. Soc., Perkin Trans. 2* **1973**, 465−469.

Cook, C. E.; Whichard, L. P.; Turner, B.; Wall, M. E.; Egley, G. H. Germination of witchweed (*Striga lutea* Lour.): Isolation and properties of a potent stimulant. *Science* **1966**, *154*, 1189−1190.

Cook, C. E.; Whichard, L. P.; Wall, M. E.; Egley, G. H.; Coggon, P.; Luhan, P. A.; McPhail, A. T. Germination stimulants. II. The structure of strigol—A potent seed germination

stimulant for witchweed (*Striga lutea* Lour.). *J. Am. Chem. Soc.* **1972**, *94*, 6198−6199.
Duke, S. O. Naturally-occurring chemical compounds as herbicides. *Rev. Weed Sci.* **1986**, *2*, 15−44.
Eplee, R. E. Chemical Control of *Striga*. In *Striga: Biology and Control;* Ayensu, E. S., Doggett, H., Keynes, R. D., Marton-Lefevre, J., Mussleman, L. J., Parker, C., Pickering, A., Eds.; ICSU Press: Paris, 1984; pp 113−123.
Fair, C. K. MolEN. *An Interactive System for Crystal Structure Analysis;* Enraf-Nonius: Delft, The Netherlands, 1990.
Fischer, N. H. Sesquiterpene lactones: Biogenesis and biomimetic transformations. In *Biochemistry of the Mevalonic acid pathway to terpenoids.* Towers, G. H. N., Stafford, H. A., Eds.; Plenum Press: New York, 1991; Chapter 4, pp 161−201.
Fischer, N. H.; Weidenhamer, J. D.; Bradow, J. M. Dihydroparthenolide and other sesquiterpene lactones stimulate witchweed germination. *Phytochemistry* **1989**, *28*, 2315−2317.
Fischer, N. H.; Weidenhamer, J. D.; Riopel, J. L.; Quijano, L.; Menelaou, M. A. Stimulation of witchweed germination by sesquiterpene lactones: Structure-activity study. *Phytochemistry* **1990**, *29*, 2479−2483.
Gomez, K. A.; Gomez, A. A. Arc sine Transformations. In *Statistical Procedures for Agricultural Research*, 2nd ed.; Gomez, K. A., Gomez, A. A., Eds.; An International Rice Research Institute Book; Wiley: New York, 1984; pp 306−308.
Hall, I. H.; Lee, K. H.; Mar, E. C.; Starnes, C. O. Antitumor agents. 21. A proposed mechanism for inhibition of cancer growth by tenulin and helenalin and related cyclopentenones. *J. Med. Chem.* **1977**, *20*, 333−337.
Hauck, C.; Müller, S.; Schildknecht, H. A germination stimulant for parasitic plants from *Sorghum bicolor*, a genuine host plant. *J. Plant Physiol.* **1992**, *139*, 474−478.
Hsiao, A. I. The effect of sodium hypochlorite and gibberillic acid on seed dormancy and germination of wild oats (*Avena futua*). *Can. J. Bot.* **1979**, *57*, 1729−1734.
Johnson, A. W.; Gowda, G.; Hassanali, A.; Knox, J.; Monaco, S.; Rozaviz, Z. The preparation of synthetic analogues of strigol. *J. Chem. Soc., Perkin Trans. 1* **1981**, 1731−1743.
Kendall, P. M.; Johnson, J. V.; Cook, C. E. Synthetic route to an aromatic analogue of strigol. *J. Org. Chem.* **1979**, *44*, 1421−1424.
Kleczkowski, K. Phytohormone Conjugates: Nature and Function. *Crit. Rev. Plant Sci.* **1995**, *14*, 283−298.
Laing, E. *Striga hermonthica:* its adaptations and distribution in Ghana. In *Striga Biology and Control*; Ayensu, E. S., Doggett, H., Keynes, R. D., Eds.; ICSU Press: Paris, and the International Development Center, Ottawa, 1984; pp 63−70.
Liang, W.-H.; Wen, J.-Q.; Liang, H.-G. Stimulation of ethylene production in aged potato tuber slices by salicyclic acid. *Phytochemistry* **1997**, *44*, 221−223.
Liljefors, T.; Thelin, B.; van der Pers, J. N. C.; Lofstedt, C. Chain-elongated analogs of a pheromone component of the turnip moth, *Agrotis segetum*. A structure-activity study using molecular mechanics. *J. Chem. Soc., Perkins Trans. 2* **1985**, 1957−1962.
MacAlpine, G. A.; Ralphael, R. A.; Shaw, A.; Taylor, A. W.; Wild, H. J. Synthesis of the germination stimulant (±)-strigol. *J. Chem. Soc., Perkins Trans. 1* **1976**, 410−416.
Musselman, L. J. Taxonomy of witchweeds. In *Parasitic Weeds in Agriculture Vol. 1 Striga*; Musselman, L. J., Ed.; CRC Press: Boca Raton, FL, 1987; pp 3−12.
Olivier, A.; Ramaiah, K. V.; Leroux, G. D. Assessment of sorghum (*Sorghum bicolor*) resistance to *Striga hermonthica*. In *Proceedings of the Fifth International Symposium on Parasitic Weeds*; Ransom, J. K., Musselman, L. J., Worsham, A. D., Parker, C., Eds.; CIMMYT: Nairobi, 1991; pp 114−121.
Osawa, E.; Goto, H.; Oishi, T.; Ohtsuka, Y.; Chuman, T. Application of molecular mechanics to natural products chemistry. *Pure Appl. Chem.* **1989**, *61*, 597−600.
Pepperman, A. B.; Connick, W. J.; Vail, S. L.; Worsham, A. D.; Pavlista, A. D.; Moreland, D. E. Evaluation of precursors and analogues of strigol as witchweed (*Striga asiatica*) seed germination stimulants. *Weed Sci.* **1982**, *30*, 561−566.
Rugutt, J. K. Control of African *Striga* species by natural products from native plants. Ph.D. Thesis, Louisiana State University, 1996, pp 1−228.
Sand, P. F.; Manley, J. D. The witchweed eradication program survey, regulatory and control. In *Witchweed Research and Control in the United States*; Sand, P. F., Westerbrooks, R. G., Eds.; Champaign, IL, 1990; pp 141−150.
SAS Institute Inc. *SAS/STAT User's Guide*, Version 6, 4th ed.; SAS: Cary, NC, 1989; Vol. 1, p 943; Vol. 2, p 846.
Sauerborn, J. The economic importance of the phytoparasite *Orobanche* and *Striga*. In *Proceedings of the Fifth International Symposium of Parasitic Weeds*; Ransom, J. K., Musselman, L, J., Worsham, A. D., Parker, C., Eds.; CIMMYT: Nairobi, 1991; pp 137−144.
Shimazaki, K.; Mori, M.; Okada, K.; Chunman, T.; Goto, H.; Osawa, E.; Sakakibara, K.; Hirota, M. Conformational analysis of periplanone analogues by molecular mechanics calculations. *J. Chem. Ecol.* **1991**, *17*, 779−788.
Vail, S. L.; Dailey, O. D.; Blanchard, E. J.; Pepperman, A. B.; Riopel, J. L. Terpenoid precursors of strigol as seed germination stimulants of Broomrape (*Orobanche ramosa*) and witchweed (*Striga asiatica*). *J. Plant Growth Regul.* **1990**, *9*, 77−83.
Worsham, A. D. Germination of witchweed seeds. In *Parasitic Weeds in Agriculture. Volume 1: Striga*; Musselman, L. J., Ed.; CRC Press: Boca Raton, FL, 1987; pp 45−61.

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