GC/MS Evaluation of Compounds in Dry and Conditioned *Striga* Species Seeds

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We have analyzed the dichloromethane (CH_2Cl_2) extracts of seeds of dry and conditioned *Striga hermonthica* (Del.) Benth., *S. aspera* (Willd.) Benth., and *S. gesnerioides* (Willd.) Vatke by means of gas chromatography (GC) coupled to mass spectrometry (MS). Sixteen compounds were identified on the basis of their mass spectra and their retention indices. All *Striga* spp. extracts contained tetradecanoic acid, *cis*,*cis*-9,12-octadecadienoic acid, *cis*-9-octadecenoic acid, and sitosterol. Also, 2,6-dimethoxy-*p*-benzoquinone (2,6-DMBQ) and several long chain aldehydes and *n*-hydrocarbons were detected in some of the extracts. The nature of the chemical changes induced by seed conditioning is discussed.

Keywords: *Striga aspera; S. hermonthica; S. gesnerioides; Scrophulariaceae; seed chemical constituents; CH_2Cl_2; GC/MS*

INTRODUCTION

Striga (Scrophulariaceae) is one of the most important genera of parasitic plants, infecting a range of major crops (e.g., sorghum, millet, maize, rice, sugar cane, and cowpea) in sub-Saharan Africa and Asia. Crop losses on cereals, attributable to *Striga hermonthica*, have been estimated to be as high as $7 billion per year in the savannas of West and Central Africa alone (M'boob, 1988).

Striga spp. are obligate root parasites wholly dependent upon a host from germination through flowering and reproduction. A *Striga* plant is capable of producing from 50 000 to 500 000 tiny seeds (each weighing about 7 μg) that may remain viable for 14 years in soil (Bebawi, 1984). Germination of *Striga* spp. seeds requires exposure to both appropriate environmental conditions and an exogenous germination stimulant, which in nature is provided by root exudates of various plants. After germination, endosperm nutrients can sustain the developing *Striga* seedlings for 3−7 days in the absence of a host (Worsham, 1987). If the seedling does not attach to a host and successfully establish a parasitic link within this period, the seedling dies. If a host root is in close proximity (2−3 mm) to the germinated *Striga* seedling, chemical signals are exchanged that direct the *Striga* radicle to the host root, initiate haustorium induction, and result in successful attachment and establishment of xylem-to-xylem connections between the parasite and host (Saunders, 1933). After successful attachment, developing *Striga* seedlings grow underground for 4−7 weeks prior to emergence. Numerous seedlings can usually be found attached to the same plant and often the same root. Most of the damage to the host plant occurs during this stage. Following emergence, *Striga* plants form chlorophyll and begin to photosynthesize but are still unable to survive in the absence of host attachment (Saunders, 1933). Symptoms of parasitism are often dramatic but nondescript, resembling drought stress, nutrient deficiency, and vascular disease. Severe plant stunting and zero economic yield often results (Berner *et al.*, 1995).

In the laboratory, *Striga* spp. seeds are sensitized to stimulant-induced germination by exposure to free water at a suitably warm temperature (around 25−30 °C), a process that has been termed "conditioning" (Vallance, 1951). In the absence of or insufficiency of conditioning, *Striga* spp. seeds respond either not at all or poorly to exogenous germination stimulants (Worsham, 1987). Several plausible explanations have been put forward to explain what happens during conditioning.

Brown and Edwards (1946) hypothesized that during conditioning *Striga* spp. seeds synthesize a substance that is the same as or similar to the stimulating substance released from plant roots. This hypothesis implies that during conditioning the amount of the substance formed in the seed itself gradually increases. The particularly interesting explanation by Schopfer (1943) states that the mere fact that *Striga* spp. seeds require exogenous germination stimulants suggests the loss of the capability of synthesizing a specific substance, which is essential for germination. Hsiao *et al.* (1979) suggested that some factor(s) beneficial to the conditioning process(es) are leached out of the *Striga* spp. seeds during conditioning. The possible germination promoters such as metabolic substrates or stimulants, which had accumulated or were synthesized during conditioning, are not leached out. More recently, Babiker *et al.* (1993) established that *Striga* spp. seeds have limited capacity to convert 1-aminocyclopropane-1-carboxylic acid (ACC), the immediate ethylene precursor, to ethylene. They suggested that the *de novo* synthesis and/or activation of proteins that act as targets for exogenous germination stimulants occurs during conditioning. Once these target proteins encounter the appropriate exogenous germination stimulant, endogenous ACC is

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S0021-8561(96)00097-0 CCC: $12.00

converted to ethylene and germination occurs. In the absence of an exogenous stimulant, protein synthesis is turned off and/or the proteins are inactivated and the seeds reenter a dormant stage. However, the existence of these proteins, whether inhibitors, stimulants, or target sites, has yet to be proven.

As there is no information pertaining to the chemical composition of dry and conditioned *Striga* spp. seeds, we attempted to establish the differences in dichloromethane (CH_2Cl_2) extracts by means of gas chromatographic/mass spectrometric (GC/MS) technique. Such information could help toward establishing *Striga* control programs.

MATERIALS AND METHODS

Seed Collection. Studies involving viable *Striga* spp. seeds were done at the International Institute of Tropical Agriculture, Ibadan, Nigeria. *Striga aspera* (Willd.) Benth. and *Striga gesnerioides* (Willd.) Vatke seeds were collected from sorghum (host crop) in Kano, Nigeria, during the 1992 and 1993 harvesting seasons, respectively. The seeds of *Striga hermonthica* (Del.) Benth. were collected from the same host crop in Abuja, Nigeria, in 1993.

Seed Surface Disinfestation (Sterilization) and Conditioning. For each experiment, *Striga* spp. seeds (2.0 g) were surface disinfested by soaking in 200 mL of 1% (w/v) sodium hypochlorite (NaOCl). Ten drops of the detergent Tween 80 [polyoxyethylene (20) sorbitan monooleate] were added. After being shaken, the mixture was allowed to stand for 4 min. Floating seeds were decanted and discarded. The remaining seeds were then transferred to a Buchner funnel lined with Whatman No. 1 filter paper and rinsed several times with sterile deionized water until the chlorine smell had disappeared. The filter paper containing the surface disinfested seeds was removed and air-dried. The dried seeds were then conditioned following a procedure already described (Vasudeva Rao, 1985). Two layers of Whatman No. 1 filter paper (9.0 cm diameter) were placed in a 9.5 cm diameter Petri dish and wetted with 3 mL of sterile deionized water. One hundred disks of 5.0 mm diameter were cut from Whatman GF/A glass microfiber filter paper and arranged on the wet filter paper in the Petri dish. The surface disinfested *Striga* spp. seeds were carefully sprinkled on the filter papers (40−50 seeds per disk). The Petri dish was then sealed with parafilm and incubated at 28 °C in the dark for 10 days. Throughout the conditioning period the filter papers were kept saturated with sterile deionized water.

Preparation of Extracts of *Striga* Spp. Seeds. Dichloromethane (CH_2Cl_2) used for sample preparation was obtained from Ibadan Chemical Co. (Oyo State, Nigeria) and double-distilled before use. Two grams of dry and conditioned seeds of each *Striga* spp. was exhaustively extracted with 6 × 20 mL portions of CH_2Cl_2 for 24 h, and the combined CH_2Cl_2 layers were concentrated in vacuum to afford gummy crude extracts. The crude CH_2Cl_2 extracts were shipped to the chemical laboratory at Louisiana State University and stored in the refrigerator at 4 °C until analysis. Standard stock solutions were prepared by dissolving 20 mg of crude seed extracts in 1 mL of analytical grade CH_2Cl_2 (Aldrich Chemical Company Inc., Milwaukee, WI) and then analyzed by a GC/MS apparatus equipped with a computer.

GC/MS Analysis. A Hewlett-Packard 5971A GC/MS was used with a 5% phenyl−95% methylpolysiloxane capillary column (DB-5 ms, 12 m × 0.20 mm i.d.; 0.33 μm film thickness, J&W Scientific, Folsom, CA) to separate components of CH_2Cl_2 extracts and obtain their electron ionization (70 eV) mass spectra. A GC/MS technique was selected for analysis of CH_2Cl_2 extracts because it provides identification of compounds even when the chromatographic separation is not sufficient to afford an accurate quantification (Wittkowski *et al.*, 1981). To avoid changes in the elution order of some compounds of CH_2Cl_2 extracts, extraction and injection procedures as well as GC/MS conditions were standardized. Samples (1.0 μL) from the standard stock solutions were introduced via split-injection mode (50:1) into a GC inlet at 250 °C. The MS detector temperature was 250 °C. The oven temperature, initially set at 40 °C for 3 min, was then raised to 280 °C at a rate of 20 °C/min, and the interface temperature was set at 280 °C. Helium was used as the carrier gas, and the head pressure was approximately 5 kPa. The mass range scanned was *m*/*z* 50−531, and spectra were acquired at a rate of 1.5 scans/s. Identification of compounds detected was made by comparison of their mass spectra with those in computer libraries (Stenhagen *et al.*, 1974, 1969; Sidow *et al.*, 1970). Sitosterol and 2,6-DMBQ (peaks 16 and 7 in Table 1) were identified by compari-

Table 1. Relative Area of Components in Dichloromethane Extracts of *Striga* Seeds

		relative area (%)[a]					
		S. hermonthica		*S. gesnerioides*		*S. aspera*	
peak[b]	RT[c] (min)	dry	con[d]	dry	con[d]	dry	con[d]
unk[e]	2.8	tr[f]	tr	tr	tr	tr	tr
1	3.6	4	60	tr	13	tr	tr
unk	5.4	nd[g]	nd	nd	tr	nd	nd
2	5.9	6	26	6	5	tr	tr
3	6.1	tr	6	tr	tr	tr	nd
unk	7.0	tr	5	tr	tr	nd	nd
unk	7.2	tr	8	nd	tr	nd	nd
unk	7.5	tr	tr	tr	1	nd	nd
unk	7.9	tr	tr	nd	2	tr	nd
unk	8.5	9	8	tr	tr	1	nd
4	8.8	tr	37	tr	5	1	tr
5	9.1	6	36	tr	tr	1	tr
6	9.3	6	52	tr	19	1	tr
7	9.7	nd	tr	nd	1	1	tr
unk	10.3	tr	tr	nd	tr	tr	nd
unk	11.2	nd	nd	nd	nd	2	n
8	13.3	69	45	44	15	20	6
unk	13.6	tr	7	nd	nd	nd	nd
9	14.18	100	100	100	100	13	100
10	14.20	25	7	33	9	9	5
11	14.9	5	3	3	nd	tr	tr
unk	15.8	tr	10	5	tr	nd	nd
12	15.9	tr	10	5	nd	tr	tr
unk	17.1	nd	nd	tr	nd	nd	nd
13	17.3	tr	21	tr	tr	5	2
14	19.3	tr	14	9	nd	tr	2
unk	19.4	nd	nd	nd	nd	tr	nd
unk	19.9	nd	nd	nd	nd	100	nd
15	22.4	40	11	13	tr	8	5
16	27.4	21	20	tr	tr	tr	tr
unk	28.5	nd	nd	tr	nd	nd	nd
unk	33.4	nd	nd	12	tr	nd	nd

[a] Calculated using relative abundance values (the area of the most abundant peak was assigned a value of 100%). [b] Numbering of peaks is as shown in Figures 1−3. Peak identification: 1, *n*-hexanal; 2, *cis*-2-heptenal; 3, hexanoic acid; 4, *cis*-2-decenal; 5, *trans,trans*-2,4-decadienal; 6, *trans,cis*-2,4-decadienal; 7, 2,6-dimethoxy-*p*-benzoquinone; 8, tetradecanoic acid; 9, *cis,cis*-9,12-octadecadienoic acid; 10, *cis*-9-octadecenoic acid; 11, tricosane; 12, pentacosane; 13, heptacosane; 14, nonacosane; 15, pentatriacontane; 16, sitosterol. [c] Retention time in minutes. [d] Con, conditioned. [e] unk, not identified. [f] tr, traces (<0.05%). [g] nd, not detected.

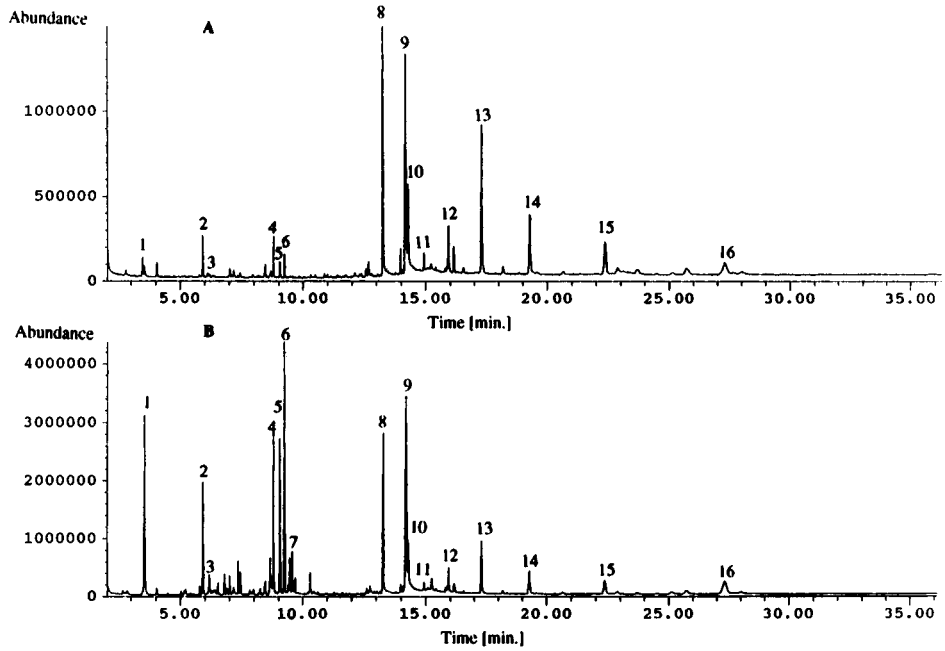

Figure 1. Total ion chromatogram profiles of compounds detected in dichloromethane extracts of dry (A) and conditioned (B) *Striga hermonthica* seeds separated on DB-5ms (12 m × 0.20 mm i.d.) fused silica capillary column. GC/MS conditions are in the text. Numbering of peaks is as shown in Table 1.

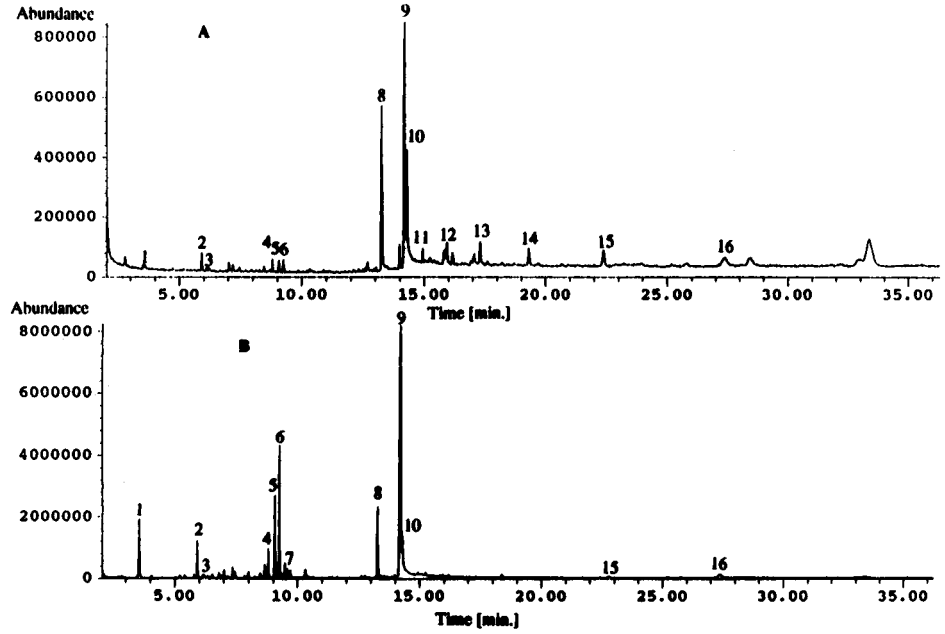

Figure 2. Total ion chromatogram profiles of compounds detected in dichloromethane extracts of dry (A) and conditioned (B) *Striga gesnerioides* seeds separated on DB-5ms (12 m × 0.20 mm i.d.) fused silica capillary column. GC/MS conditions are in the text. Numbering of peaks is as shown in Table 1.

son of retention times and mass spectra with the authentic samples isolated in our laboratory (Rugutt, 1996). The relative area percentages of peaks, which gave an indication of concentrations of various components in *Striga* spp. extracts, were calculated using the abundance values.

RESULTS

Figures 1−3 show typical chromatographic profiles of components detected in dichloromethane (CH_2Cl_2) extracts of dry (panel A) and conditioned (panel B) *Striga* spp. seeds, as separated on a DB-5ms column. Several peaks were detected and their mass spectra and retention times recorded (Table 1). The following 16 types of compounds were identified: 1 quinone, 1 sterol, 4 fatty acids, 5 aldehydes, and 5 long-chain hydrocarbons. The constituents common to all dry and conditioned *Striga* spp. seeds were tetradecanoic acid, *cis*,*cis*-9,12-octadecadienoic acid, *cis*-9-octadecenoic acid, and *n*-hexanal as well as an unidentified component with a retention time of 2.8 min. In the detected hydrocarbon series, the heavier odd-numbered carbon *n*-alkanes *n*-heptacosane ($C_{27}H_{56}$), *n*-nonacosane ($C_{29}H_{60}$), and *n*-pentatricontane ($C_{35}H_{72}$)) predominated in the region above *cis*-9-octadecenoic acid (peak 10).

The distribution patterns of compounds in *S. hermonthica* and *S. gesnerioides* seeds were similar (Figures 1 and 2). Their gas chromatograms indicated that

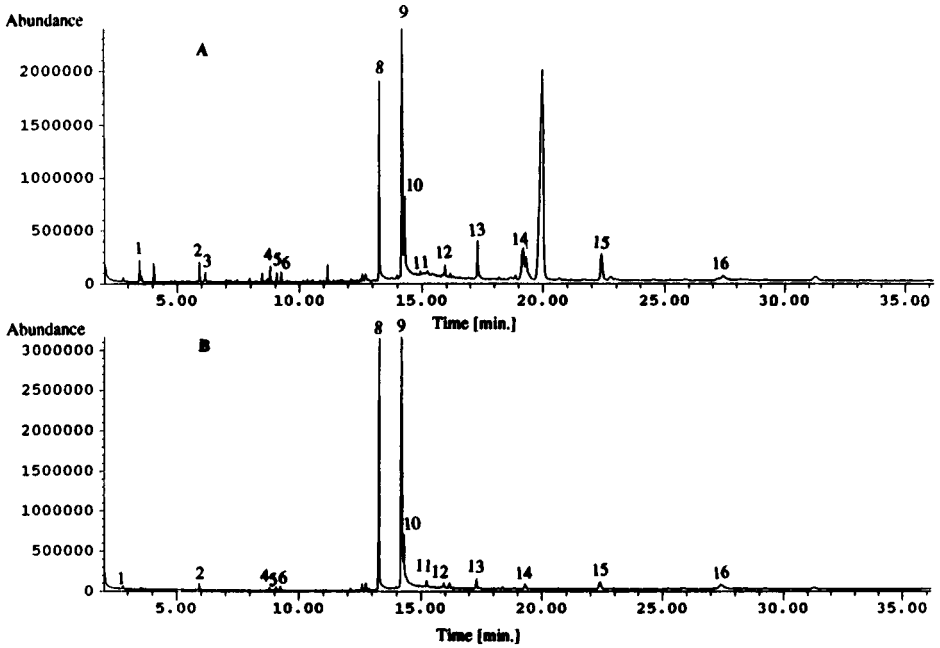

Figure 3. Total ion chromatogram profiles of compounds detected in dichloromethane extracts of dry (A) and conditioned (B) *Striga aspera* seeds separated on DB-5ms (12 m × 0.20 mm i.d.) fused silica capillary column. GC/MS conditions are in the text. Numbering of peaks is as shown in Table 1.

the concentrations of *cis*-2-decenal, *n*-hexanal, and *trans*,*trans*-2,4-decadienal increased during conditioning. Furthermore, the seeds of both species, unlike those of *S. aspera*, contained hexanoic acid and 2,6-DMBQ as well as more of several unidentified components with retention time windows between 2.7 and 11 min. It can be noted from Table 1 that *S. gesnerioides* seeds synthesized several unidentified compounds with retention times of 5.4, 7.2, 7.9, and 10.3 min while compounds with retention times of 14.9, 17.1, and 28.5 min disappeared during conditioning. Unique to dry *S. gesnerioides* extracts was the presence of a compound with the highest retention time of 33.4 min (Figure 2A). Conditioned *S. hermonthica* seeds showed the greatest number of compounds overall and a larger number of compounds in the retention time window between 5 and 10 min (Figure 1B). Clearly, the concentrations of *cis*-2-heptenal, *trans*,*trans*-2,4-decadienal, *n*-pentacosane, *n*-heptacosane, and *n*-nonacosane increased during conditioning.

S. aspera extracts contained the smallest number of compounds (Figure 3). A very dramatic increase in the concentration of *cis*,*cis*-9,12-octadecadienoic acid occurred during conditioning. Moreover, it was found that *S. aspera* seeds exhibited a slight decrease in the level of 2,6-DMBQ and the disappearance of the most abundant component with a retention time of 19.9 min. Compounds with retention times 19.4 and 19.9 min were detected in extracts of dry *S. aspera* seeds but not in extracts of *S. hermonthica* and *S. gesnerioides*.

DISCUSSION

By way of background to our work, a brief review of *Striga* control methods is appropriate. *Striga* spp. control can be effected by stimulating germination in the absence of a host, thus reducing the amount of viable seed inoculum present in the soil. Ethylene gas has been used successfully for *Striga asiatica* eradication program in the United States (Sand and Manley, 1990). By contrast, crop rotations with non-hosts that produce effective germination stimulants are being used as the focus of integrated control programs for *S. hermonthica* in Africa (Berner *et al.*, 1995). However, the exact mechanism through which exudates or active substances from external sources induce *Striga* spp. germination is still largely unknown. There has been an intense effort by our group in recent years to understand the mechanism(s) of germination of *Striga* spp. seeds (Rugutt, 1996). In the present investigation, preliminary identification and quantification of some of the components leached out, synthesized, or retained during conditioning of *Striga* spp. seeds are reported.

As shown in Table 1, plant extracts obtained from dry and conditioned *Striga* spp. seeds differ in their chemical compositions. This probably explains the variable bioassays that have been observed in various isolates of *Striga* spp. (Rugutt, 1996; Pepperman *et al.*, 1982). Our results show that the relative distribution of tetradecanoic, *cis*,*cis*-9,12-octadecadienoic, *cis*-9-octadecenoic, and other fatty acids is an important difference (Vagelos, 1974; Gurr, 1974). The differences could be attributed to environmental factors that influence their compositions in the seeds (Stearns, 1970). The concentrations of aldehydes in *S. hermonthica* and *S. gesnerioides* seeds generally increased during conditioning. Although aldehydes have been known to occur in fruits for a long time, the way they are synthesized is still only solved in part (De Pooter, 1987; Miyake and Shibamoto, 1995; Jennings and Shibamoto, 1980).

An interesting and potentially important observation in this study was the considerably lower levels of 2,6-DMBQ and sitosterol in *Striga* spp. plants in comparison to other plants (Vagelos, 1974; Heftmann, 1971; Handa *et al.*, 1983; Gladu *et al.*, 1991). This indicates that *Striga* spp. seeds may have lost the capability to synthesize these compounds as was suggested by Schopfer (1943). 2,6-DMBQ is not only an inhibitor of mitochondrial respiration (Chappel and Hansford, 1972; Redfearn and Whittaker, 1962) but also an haustoria-inducing principle in *Striga* spp. (Chang, 1986). Using syringaldazine, Chang and Lynn (1986) have detected laccase-type enzymes associated with the roots of *S.*

asiatica (L.) Kuntze and *Agilinis purpurea* (L.) Raf. (Scrophulariaceae). They proposed that such enzymes released from the parasite root may cleave 2,6-DMBQ from the cell wall complex of the host, which then diffuses back to *S. asiatica* and triggers haustorial development. They concluded that zones for both *S. asiatica* germination and haustorial induction are determined by a combination of factors including diffusion rates of promoters, the instability of active compounds, and critical threshold concentrations and exposure times required for induction. The fact that *Striga* spp. seeds usually do not develop haustoria in the absence of 2,6-DMBQ suggest that the level of this quinone is below the threshold concentration necessary to trigger haustorial development. It is possible that this haustorium-inducing factor may have been gradually lost, like the endogenous ACC to ethylene conversion process (Adams and Yang, 1979) during evolution.

The significance of sitosterol in *Striga* seeds can be rationalized from several perspectives. For example, sitosterol is not only one of the biogenetic precursors of steroid hormones, but it may have hormonal activity itself (Heftmann, 1971). Sitosterol is water-soluble and might be leached out of *Striga* seeds in the field by rain or conditioning liquid in the laboratory. This might be the reason why the seeds contained very low levels of this sterol.

The occurrence of inhibitors in seeds is well known (Evenari, 1946; Hermberg, 1949a,b), and studies in several species indicate the physiological importance of these substances (Wareing and Foda, 1956). Khan (1971) hypothesized that dormancy can result from an excess of inhibitor. However, with respect to dry *Striga* spp. seed dormancy, the presence and role of inhibitors has hitherto been obscure. The inhibitory action might possibly be brought about by toxic concentrations of substances that are not specific germination inhibitors. Because germination inhibitors may be leached out of *Striga* spp. seeds during conditioning (Hsiao *et al.*, 1979), it is not unreasonable to conclude that these inhibitors may have the potential to block stimulant-mediated germination. Furthermore, these inhibitors might constitute the (external) seed structures making them impermeable to germination stimulants. From the present results, it is evident that one of the compounds with a retention time of 19.9 min is the primary inhibitor in *S. aspera* seeds (Figure 3A). Inhibitors detected in *S. hermonthica* and *S. gesnerioides* included heavier *n*-hydrocarbons (peaks 11−16 in Figure 2A) in the retention time window between 14 and 30 min.

CONCLUSIONS

In summary, our results indicate that the water-soluble endogenous inhibitors are apparently washed out of *Striga* spp. seeds during conditioning. Though we cannot quantitatively substantiate, it is clear that some compounds are also synthesized during conditioning. Future studies should emphasize investigation of the pattern of accumulation and mechanisms by which the various compounds are accumulated in *Striga* spp. seeds. This will provide important information useful in designing target specific germination stimulants/inhibitors for control of *Striga* spp.

ABBREVIATIONS USED

Spp., species; ACC, 1-aminocyclopropane-1-carboxylic acid; CH_2Cl_2, dichloromethane; 2,6-DMBQ, 2,6-dimethoxy-*p*-benzoquinone; GC/MS, gas chromatography/mass spectrometry.

ACKNOWLEDGMENT

J.K.R. is grateful to the Rockefeller Foundation for the African Dissertation Internship Award.

LITERATURE CITED

Adams, D. O.; Yang, S. F. Ethylene biosynthesis: Identification of 1-aminocyclopropane-1-carboxylic acid as an intermediate in the conversion of methionine to ethylene. *Proc. Natl. Acad. Sci. U.S.A.* **1979**, *76*, 170−174.

Babiker, A. G.; Ejeta, G.; Butler, L. G.; Woodson, W. R. Ethylene biosynthesis and strigol-induced germination of *Striga* asiatica. *Physiol. Plant.* **1993**, *88*, 359−365.

Bebawi, F. F.; Eplee, R. E.; Harris, C. E.; Norris, R. S. Longevity of witchweed (*Striga asiatica*) seed. *Weed Sci.* **1984**, *32*, 494−507.

Berner, D. K.; Kling, J. K.; Singh, B. B. *Striga* research and control: a perspective from Africa. *Plant Dis.* **1995**, *79*, 652−660.

Brown, R.; Edwards, M. The germination of the seeds of *Striga lutea*. II. The effect of time of treatment and of concentration of the host stimulant. *Ann. Bot.* **1946**, *10*, 133−142.

Chang, M. Isolation and characterization of semiochemicals involved in the host recognition in *Striga asiatica*. Ph.D. Thesis University of Chicago, Chicago, 1986.

Chang, M.; Lynn, D. G. The haustorium and the chemistry of host recognition in parasitic angiosperms. *J. Chem. Ecol.* **1986**, *12*, 61−579.

Chappel, J. B.; Hansford, R. G. *Subcellular Components, Preparation and Fractionation*, 2nd ed.; Birnie, G. D., Fox, S. M., Eds.; Butterworths: London, 1972; pp 43−56.

De Pooter, H. L.; Van Acker, M. R.; Schamp, N. M. Aldehyde metabolism and the aroma quality of stored golden delicious apples. *Phytochemistry* **1987**, *26*, 89−92.

Eplee, R. E., Westerbrooks, R. G., Eds. *Abstracts, Meeting of the Weed Science Society of America*; The Society: Champaign, IL, 1990; pp 141−151.

Evenari, M. Germination inhibitors. *Bot. Rev.* **1949**, *15*, 153.

Gladu, P. K.; Patterson, G. W.; Wikfors, G. H.; Lusby, W. R. Free and combined sterols of *Pavlova gyrans*. *Lipids* **1991**, *26*, 656−659.

Gurr, M. I. Biosynthesis of unsaturated fatty acids. In *Biochemistry Series One, Vol. 4, Biochemistry of Lipids*; Goodwin, T. W., Ed.; Butterworth: London, 1974; pp 181−235.

Handa, S. S.; Kinghorn, A. D.; Cordeu, G. A.; Farnsworth, N. R. Plant anticancer agents. XXV1. Constituents of *Peddiea fischeri*. *J. Nat. Prod.* **1983**, *46*, 248−250.

Heftmann, E. Functions of Sterols in Plants. *Lipids* **1971**, *6*, 128−133.

Hermberg, T. Significance of growth-inhibiting substances and auxins for the rest period of potato tuber. *Physiol. Plant.* **1949a**, *2*, 241.

Hermberg, T. Growth inhibitors in terminal buds of *Fraxinus*. *Physiol. Plant.* **1949b**, *2*, 37.

Hsiao, A. I.; Worsham, A. D.; Moreland, D. E. Factors affecting conditioning and germination of witchweed (*Striga asiatica* (L.) Kuntze) seeds under laboratory conditions. In *Proceedings of the Second Symposium on Parasitic Weeds*; Musselman, L. J., Worsham, A. D., Eplee, R. E., Eds.; North Carolina State University: Raleigh, NC, 1979; pp 193−201.

Jennings, W.; Shibamoto, T. *Qualitative Analysis of Flavor and Fragrance Volatiles by Glass Capillary Gas Chromatography*; Academic Press, Inc.: New York, 1980.

Khan, A. A. Cytokinins: permissive role in seed germination. *Science* **1971**, *171*, 853−859.

M'boob, S. S. A regional program for West and Central Africa. In *Proceedings of FAO/OAU All-African Government Consultation on Striga Control*; Maroua, Cameroon, Oct 20−24; Robson, T. D., Broad, H. R., Eds.; Food and Agriculture Organization of the United Nations: Rome, 1988; pp 190−194.

Miyake, T.; Shibamoto, T. Quantitative analysis by gas chromatography of volatile carbonyl compounds in cigarette smoke. *J. Chromatogr.* **1995**, *693*, 376−381.

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*, 562−566.

Redfearn, E. R; Whittaker, P. A. The inhibitory effects of quinones on the succinic oxidase system of the respiratory chain. *Biochim. Biophys. Acta* **1962**, *56*, 440−444.

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.; Weed Science Society of America: Champaign, IL, 1990; pp 141−150.

Saunders, A. R. Studies in phanerogamic parasitism, with particular reference to *Striga lutea* Lour. *S. Afr. Dept. Agric. Sci. Bull.* **1933**, 56−128.

Schopfer, W. H. Classification and Definition of Active Substances. In *Plants and vitamins;* Schopfer, W. H., Robbins, W. J., Eds.; Chronica Botanica Co.: Waltham, MA, 1943; Chapter 4, pp 33−39.

Sidow, E. V.; Anjou, K.; Karlsson, G. *Arch. Mass Spectral Data* **1970**, *1*, 392−578.

Stearns, E. M. Biosynthesis of unsaturated fatty acids in higher plants. *Prog. Chem. Fats Other Lipids* **1970**, *9*, 455−516.

Stenhagen, E.; Abrahamson, S.; McLafferty, F. W. *Atlas of Mass Spectral Data*; Interscience: New York, 1969; Vols. 1−3.

Stenhagen, E.; Abrahamson, S.; McLafferty, F. W. *Registry of Mass Spectral Data*; Wiley: New York, 1974.

Vagelos, P. R. Biosynthesis of Saturated Fatty Acids. In *Biochemistry Series One, Vol. 4, Biochemistry of Lipids*; Goodwin, T. W., Ed.; Butterworth: London, 1974; pp 100−141.

Vallance, K. B. Studies on the germination of the seeds of *Striga hermonthica.* II. The effect of the stimulating solution on seed respiration. *J. Expt. Bot.* **1951**, *2*, 31−40.

Vasudeva Rao, M. I. Techniques for screening sorghum for resistance to *Striga*. Information Bulletin No. 20; International Crop Research Institute for the Semi-Arid Tropics (ICRISAT): Patancheru, P.O. Andra Pradesh 502-324, India, 1985.

Wareing, P. F.; Foda, H. Possible role of growth inhibitors in the dormancy of seed of *Xanthium* and lettuce. *Nature* **1956**, *178*, 908.

Wittkowski, R.; Toth, L.; Baltes, W. F. Collection and analysis of phenolic fraction of smoke for curing. *Lebensm. Unters. Forsch.* **1981**, *173*, 445−457.

Worsham, A. D. Germination of witchweed seeds. In *Parasitic Weeds in Agriculture*; Musselman, L. J., Ed.; CRC Press: Boca Raton, FL, 1987; Vol. 1, pp 45−61.

Received for review February 12, 1996. Revised manuscript received August 21, 1996. Accepted September 16, 1996.[⊗]

JF9600972

[⊗] Abstract published in *Advance ACS Abstracts,* November 1, 1996.