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ALLELOPATHIC INFLUENCE OF *Sorghum bicolor* ON WEEDS DURING GERMINATION AND EARLY DEVELOPMENT OF SEEDLINGS

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Abstract—The allelopathic interaction between sorghum [*Sorghum bicolor* (L.) Moench] and 10 species of grass and broadleaf weeds was investigated. Germination of weed seeds was slightly inhibited or stimulated, depending on species, when incubated in closed Petri dishes with germinating sorghum. Subsequent radicle and hypocotyl or coleoptile elongation of weeds was significantly inhibited by the germinating sorghum. For weeds interplanted with sorghum and grown under greenhouse conditions. The inhibitory effect on some weed species was still evident after 2 months of growth. Significant differences were found in the dry matter per weed plant grown in pots in proximity to sorghum vs. weeds grown in monoculture. Aqueous leachates from pots planted with sorghum alone or from a system in which sorghum roots protruded into water had strong allelopathic activity. These results indicate that water-soluble allelochemicals are produced by germinating sorghum seeds and that production of these substances continues during seedling growth.

Key Words—Allelopathy, *Sorghum bicolor*, weeds, weed control, agroecosystems, phytotoxins, seed germination.

INTRODUCTION

Residues of mature sorghum [*Sorghum bicolor* (L.) Moench] contain water-soluble allelochemicals that inhibit seed germination and seedling development of other species of plants (Lawrence and Kilcher, 1961; Guenzi and McCalla,

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1962; Guenzi et al., 1967; Lehle and Putnam, 1982). Johnson grass [*Sorghum halepense* (L.) Pers.] also produces phytotoxic substances (Abdul-Wahab and Rice, 1967; Nicollier et al., 1983). A number of phenolic acids have been implicated as phytotoxins that can be leached from sorghum residues, but there is insufficient evidence that these are the active or sole allelochemicals (Guenzi and McCalla, 1962). Sorghum and Johnson grass also produce two cyanogenic compounds, dhurrin and taxiphyllin, which upon hydrolysis yield HCN and *p*-hydroxybenzaldehyde, but their potential role as allelochemicals is likewise ambiguous (Nicollier et al., 1983).

While the allelopathic effect of residues of mature sorghum on other plants has been described, studies involving the cogermination of sorghum seeds with seeds of other species are lacking. Allelopathic activity expressed by a species in the earliest stages of development could confer a powerful advantage over other plants. The purpose of our investigation was to determine if sorghum exerts an allelopathic influence on selected weeds when cogerminated with them.

METHODS AND MATERIALS

Seeds. Sorghum seed (cv. Bird-a-boo) was obtained from the Taylor Evans Seed Company,[3] Tulia, Texas. Cress [*Lepidium sativum* (L.) cv. Curlycress] was obtained from the Burpee Seed Company, Warminster, Pennsylania. Weed seeds were collected from the following locations in the years indicated: barnyard grass [*Echinochloa crus-galli* (L.) Beavr.] (Arkansas, 1976), curly dock [*Rumex crispus* (L.)] (California, 1975), green foxtail [*Setaria viridis* (L.) Beavr.] (North Dakota, 1976), Johnson grass (Illinois, 1977), bigroot morning glory [*Impomea pandurata* (L.) G.F.W. Meyer] (Illinois, 1978), redroot pigweed [*Amaranthus retroflexus* (L.)] (Maryland, 1976), red sorrel [*Rumex acetosella* (L.)] (Illionis, 1974), velvetleaf [*Abutilon theophrasti* Medic.] (Mississippi, 1974), and wild mustard [*Sinapsis arvensis* (L.)] (North Dakota, 1976).

Cogermination in Petri Dishes. Seeds of sorghum, weeds, and cress (which was treated as a "weed" in this study) were surface treated with an 800 ppm solution of Roccal to minimize mold growth. Tween 20 (0.5%) was added to the solution to serve as a wetting agent. Seeds were agitated for 15 min with the Roccal solution and then rinsed 10 times with sterile water. Seeds were germinated in sterile plastic Petri dishes (9 cm diameter). The bottom of each dish was lined with a sterilized Whatman No. 3 filter paper disk, and 3 ml of sterile distilled water was added. Controls consisted of 20 evenly spaced weed seeds per dish. Treatments consisted of 20 weed seeds and 20 sorghum seeds, evenly and uniformly interspersed, per dish. For each weed tested, data were

[3] Reference to brand or firm name does not constitute endorsement by the U.S. Department of Agriculture over others of a similar nature not mentioned.

collected from five controls and five treatments. Since data was collected at three days and at seven days, the initial number of controls and treatments was doubled because the measurement made at three days was destructive. As a contingency, one additional control and one additional treatment were included for both the three-day and seven-day trials.

Petri dishes were placed on trays, 12 dishes per tray, so that paired dishes for controls and treatments were side by side. Trays were placed in an environmental growth chamber maintained at 25°C. and 80% relative humidity. The photoperiod was 8 hr per day, and the light intensity at the level of the trays was 9500 lux. Trays were rotated daily to compensate for any minor differences in temperature and light intensity at different locations in the growth chamber.

Daily observations were recorded for germination, leaf formation, root formation, and general health of the developing seedlings. Prior to collecting quantitative data at three days and at seven days, one control and one treatment were discarded from each tray. These were selected visually on the basis of being the least typical (low germination, mold growth, etc.) of the six controls and six treatments on the tray. If none was atypical, the paired treatment and control from the lower left-hand corner of the tray were discarded. For the remaining controls and treatments, the number of ungerminated seeds was counted and, for germination seeds, the length of hypocotyls or coleoptiles and radicals (or primary roots) was measured. Data was statistically analyzed by the Waller-Duncan K-ratio t test to determine the effects and interactions of weed variety, presence or absence of sorghum, and length of trial on three responses: length of hypocotyls or coleoptiles, length of radicles, and percent germination.

Sorghum Root Exudates. A 20-cm-diameter desiccator was autoclaved, and 350 ml sterile distilled water was added to bring the water level to the bottom of the perforated porcelain disk that conventionally holds samples above the desiccant. A layer of sterile, moist cheesecloth was placed on top of the porcelain disk, and 30 g (about 1000 seeds) of sorghum seed, which had been surface-treated with Roccal as described earlier, were evenly distributed on top of the cheesecloth. The lid was placed on the desiccator, and it was located under fluorescent light (about 9000 lux for 15 hr/day) at room temperature. As the sorghum seedlings grew to a height of 9–10 cm with roots of 3–5 cm length protruding into the water, the water was withdrawn from the desiccator and replenished at five-day intervals. Water collected from the desiccator was concentrated on a rotary evaporator to a volume of 30 ml, and 3 ml of this concentrate was tested for allelopathic activity against germinating weed seeds in Petri dish assays as previously described. For control assays, 3 ml of distilled water was used in place of the concentrated sample from the desiccator.

Interplantings in Clay Pots. Seeds of sorghum and selected weeds were planted in sand in 12.7-cm clay pots. Pots were maintained in a greenhouse under conditions of summer sunlight and were watered daily by immersing the

base of the pots in water. Plants were fertilized twice a week by immersing pots in Hoagland and Arnon (1950) nutrient solution with iron content increased to 1.6 times that of the standard solution. Control pots containing only sorghum or a weed and pots interplanted with varying ratios of sorghum and a weed were maintained for periods of up to 60 days. In some cases, leachate was obtained by watering pots planted with 25 sorghum seeds and collecting the runoff. The unconcentrated leachate was used to water an equivalent number of pots planted with weeds or interplanted with sorghum and weeds.

RESULTS

The data on the influence of sorghum on weeds cogerminated in closed Petri dishes is summarized in Table 1. Minor differences in percent germination of weed seeds between weeds germinated alone or in the presence of germinating sorghum seeds were noted. The greatest differences were in the growth and development of seedlings over a period of three or seven days.

Of the 10 species of weeds investigated, statistical analysis of data revealed that cogerminating sorghum in the Petri dish significantly influenced the development of all, except green foxtail and velvetleaf, at the 5% probability level by the seventh day. In several cases, statistically significant differences were noted by the third day. Cogermination with sorghum suppressed the elongation and development of hypocotyls or coleoptiles of weeds with one exception: green foxtail showed a slight but statistically significant increase in average length of coleoptiles. Morning glory, barnyard grass, and velvetleaf exhibited the greatest percent reductions in hypocotyl or coleoptile elongation in the presence of germinating sorghum. Cogermination with sorghum produced significant reduction in the average radicle length of all species tested.

In addition to effects on elongation, weeds germinated in the presence of sorghum generally were less healthy in appearance with higher incidences of necrosis, chlorosis, and retardation of development. Visual observations of differences other than measured growth are presented in Table 2.

Concentrated water samples obtained from desiccators in which the roots of young sorghum seedlings protruded into the water had general effects on the development of selected sprouting weed seeds that were comparable to the effects of proximity to germinating sorghum seeds. In three-day Petri dish assays, the average lengths of barnyard grass coleoptiles and radicles were reduced 30% and 20%, respectively, in comparison with controls. Pigweed hypocotyls and radicles were, on the average, shorter by 20% and 6%, respectively, at three days, and 25% and 30% shorter at seven days. Coleoptile development of green foxtail, as in other tests, was stimulated slightly rather than inhibited and showed an average increase of 11% over controls with a reduction of 8% in radicle length at seven days. The sorghum root leachate reduced the germination of

TABLE 1. INFLUENCE OF GERMINATING SORGHUM SEEDS ON DEVELOPMENT OF GERMINATING WEED SEEDS

	3 Days				7 Days			
	Control		Cogerminated with sorghum		Control		Cogerminated with sorghum	
Weed	Germ[a] (%)	Average length (cm)[b]	Germ[a] (%)	Average length (cm)[b, c]	Germ[a] (%)	Average length (cm)[b]	Germ[a] (%)	Average length (cm)[b, c]
Barnyard grass	76a	1.3 ± 0.5	66b	0.9 ± 0.5†	73a	2.0 ± 0.6	68b	1.4 ± 0.7†
		1.9 ± 1.0		0.9 ± 0.7†		4.5 ± 1.5		1.9 ± 1.2†
Curly dock	74a	0.3 ± 0.1	73a	0.2 ± 0.1†	78b	0.5 ± 0.2	71a	0.4 ± 0.2†
		0.5 ± 0.4		0.4 ± 0.2		1.8 ± 1.0		1.1 ± 0.5†
Green foxtail	83a	1.0 ± 0.6	70b	1.0 ± 0.5	81a	1.5 ± 0.7	74b	1.7 ± 0.8
		2.0 ± 1.0		1.9 ± 0.8		4.2 ± 1.2		3.4 ± 1.4†
Johnson grass	0	0	0	0	47a	2.1 ± 1.0	47a	1.5 ± 1.0†
		0		0		3.2 ± 1.8		2.5 ± 1.6†
Morning glory	74a	1.5 ± 0.8	91b	0.9 ± 0.4†	83c	1.7 ± 1.0	85c	1.2 ± 0.7†
		2.1 ± 0.7		2.1 ± 0.8		2.5 ± 1.0		1.9 ± 1.0†
Pigweed	95ab	0.5 ± 0.1	92b	0.5 ± 0.1	97a	0.7 ± 0.2	92b	0.5 ± 0.2†
		1.1 ± 0.2		0.8 + 0.3†		2.1 ± 0.5		1.3 ± 0.4†
Red sorrel	60a	0.1 ± 0.1	59a	0.1 ± 0.2	72b	0.5 ± 0.1	85c	0.4 ± 0.1†
		0.4 ± 0.2		0.4 ± 0.2		2.0 ± 0.7		1.4 ± 0.5†
Velvetleaf	91a	1.1 ± 0.3	83b	0.7 ± 0.2†	90a	1.2 ± 0.4	92a	0.7 ± 0.2†
		1.6 ± 0.5		1.1 ± 0.5†		1.9 ± 0.8		1.7 ± 0.7
Wild mustard	37a	0.9 ± 0.5	45b	0.9 ± 0.3	32c	1.2 ± 0.4	36ac	0.8 ± 0.3†
		3.6 ± 1.8		2.2 ± 1.0†		4.2 ± 1.7		3.0 ± 1.5†
Cress[d]	100a	1.2 ± 0.3	100a	0.9 ± 0.3†	98a	1.4 ± 0.4	99a	1.1 ± 0.3†
		0.9 ± 0.8		0.8 ± 0.3		1.3 ± 1.2		1.0 ± 0.7†

[a] Two germination means in the same line with a letter in common are not significantly ($P < 0.05$) different.

[b] First value in column is average length of hypocotyl (for dicots) or coleoptile (for monocots). Second value is average length of radicle. Standard deviation is indicated by ± value. Seeds that did not germinate were not included in calculations of average length and standard deviation. The number of measurements was equal to the figure given under % germination, since the beginning number of seeds was 100.

[c] Statistically significant difference between control and treatment indicated by † (5% level).

[d] Not a weed, cultivar Curlycress.

TABLE 2. VISIBLE DIFFERENCES BETWEEN GERMINATING WEED SEEDS IN PRESENCE AND ABSENCE OF GERMINATING SORGHUM SEEDS

Weed	Control	Cogerminated with sorghum
Barnyard grass	At 3 days, radicles appeared healthy with no necrosis. At 7 days, plumules were uncurled and seedlings appeared vigorous with strong, branching roots.	At 3 days, necrosis of some radicles was evident. At 7 days, plumules were not uncurled and most roots had no branching with some necrosis.
Curly dock	At 4 days, radicles had abundant root hair development and 48 green, emerging hypocotyls were visible. At 7 days, roots were hairy and branching and 41 sets of cotyledons were developed with 28 fully open. Cotyledons were deep green in color.	At 4 days, only some radicles had root hairs and only 21 green, emerging hypocotyls were visible. At 7 days, roots had little branching and only 14 sets of cotyledons were developed with only four fully open. Cotyledons were much smaller and less green than controls.
Green foxtail	Overall appearance was healthy.	Overall appearance was healthy. Other than size, not visibly different from controls.
Johnson grass	At 7 days, seedlings were healthy in appearance with branching roots and two developed leaves.	At 7 days, roots were thinner than those of the controls and only five seedlings had developed two leaves.
Morning glory	At 3 days, radicles were branched and cotyledons were open. The general appearance was healthy with no necrosis. At 7 days, seedlings had overall healthy appearance with no necrosis.	At 3 days, radicles had fewer and much smaller side branches than the controls and some were necrotic. Only two seedlings had open, fully developed cotyledons. At 7 days, considerably more necrosis than control including both roots and hypocotyls.

Pigweed	At 3 days, all seedlings appeared healthy. At 7 days, lateral root development was apparent. Roots had no necrosis and 76 sets of cotyledons were fully developed and open.	At 3 days, necrosis of some radicles was evident. At 7 days, roots were brown and necrotic with almost no lateral root development. Only 33 sets of cotyledons were opened.
Red sorrel	At 3 days, seeds were just visibly germinated. At 7 days, seedlings had 56 sets of open cotyledons and all roots were healthy with hairs.	At 3 days, not visibly different from controls. At 7 days, seedlings had 50 sets of open cotyledons but their roots were thinner and less vigorous than controls, and only some roots had hairs.
Velvetleaf	At 3 days, radicles were long and healthy in appearance and seven hypocotyls were formed. At 7 days, 48 sets of cotyledons were fully developed and some roots were branched.	At 3 days, there was pronounced necrosis of radicles and no cotyledons were formed. At 7 days, hypocotyls were thinner than controls and only six sets of cotyledons were developed. Roots were necrotic.
Wild mustard	At 3 days, 32 open, developed sets of cotyledons were visible, radicles were sturdy with some side branches and no necrosis. At 7 days, seedlings were sturdy and healthy with only one underdeveloped set of cotyledons; roots were well developed with much branching.	At 3 days, 40 open, developed hypocotyls were visible. Radicles were less healthy in appearance than controls and some were necrotic. At 7 days, many seedlings were pale and underdeveloped with necrosis of roots and necrosis at juncture of radicles and hypocotyls.
Cress	At 3 days, seedlings were healthy in appearance with large, green cotyledons. At 7 days, some necrosis of radicles and cotyledons was visible.	At 3 days, seedlings were less developed than controls. At 7 days, seedlings had much more necrosis than controls and were, in general, unhealthy in appearance.

barnyard grass by 20% and pigweed by 40%, but increased the germination of green foxtail by as much as 17%. The effect on weed germination was more pronounced for the concentrated root exudate than for cogermination with sorghum seed.

Growth of interplanted sorghum and selected weeds in clay pots for periods of up to two months demonstrated that the inhibitory influence of sorghum extends at least into early stages of sorghum growth. The suppression of weed growth by sorghum was clearly more than competition for light and nutrients as demonstrated by the data presented in Figure 1. As the ratio of barnyard grass seeds to sorghum seeds per pot was decreased, the dry weight per plant of the 5-week-old leaves and roots of barnyard grass decreased (white bars of Figure 1). Alone, this could be interpreted as competition, but when identical interplantings were watered not with tap water, but with the leachate collected from pots planted with 25 sorghum seeds, the dry weight per plant of barnyard grass leaves and roots was reduced greatly in all cases (black bars of Figure 1).

The data presented in Figure 2 show that pigweed also was affected adversely in interplantings with sorghum. The harvested leaves and stems of pigweed plants grown in pots with sorghum for two months contained less than 5% of the amount of dry matter in the controls in the most extreme case where five pigweed seeds were interplanted with 20 sorghum seeds. Root development

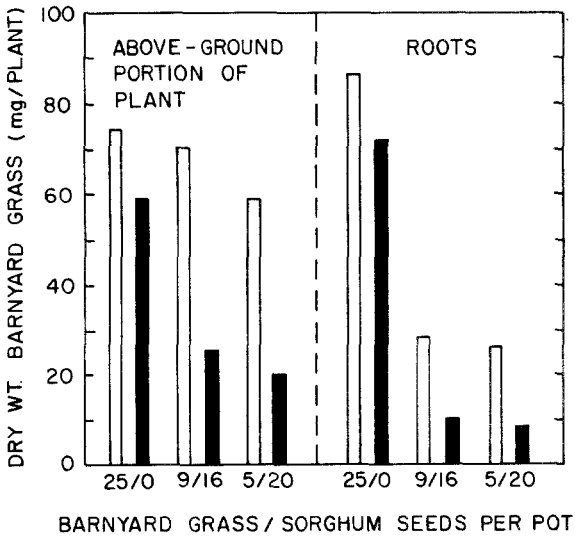

FIG. 1. Dry weights of barnyard grass five weeks after planting in pots by itself or interplanted in two different ratios with sorghum. White bars indicate average weights obtained from pots watered with tap water. Black bars represent average weights from pots watered with leachate obtained from watering pots planted with 25 sorghum seeds.

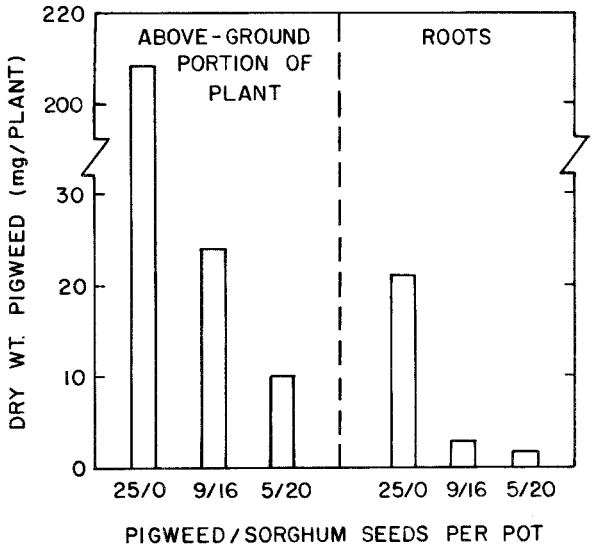

FIG. 2. Average dry weights of pigweed two months after planting in pots by itself or interplanted in two different ratios with sorghum.

of pigweed was suppressed to less than 10% of the growth of controls in the most extreme case.

The growth of red sorrel interplanted with sorghum in pots was inhibited greatly. Two months after planting, the average dry weights of controls (25 red sorrel seeds per pot) was 49.4 and 44.9 mg/plant for the above-ground portion of the red sorrel plants and the roots, respectively. For pots interplanted with 12 red sorrel seeds and 13 sorghum seeds, the harvested average dry weights of the above-ground portions and roots of red sorrel were 23.4 and 15.2 mg/plant (reductions of 53% and 66%), respectively.

In visual observation for periods of up to two months of Jimsonweed and velvetleaf interplanted with sorghum in pots revealed that growth of these weeds also was suppressed greatly by sorghum in comparison to control plantings of the weeds alone. Sorghum had no visible effect on interplanted wild mustard, morning glory, and green foxtail.

DISCUSSION

Our results suggest that young sorghum seedlings produce allelochemicals that can influence the growth and development of a number of weeds. The allelopathic influence of sorghum on some weeds appears to extend from the early stages of germination through at least two months of growth. Other pre-

viously cited investigators have demonstrated that mature sorghum and residues of sorghum plants exert allelopathic effects on other plants, so it is likely that this phenomenon persists throughout the entire growth cycle of sorghum. Under some circumstances, sorghum exerts a pronounced detrimental effect on subsequent planting of sorghum in the same plot (Burgos-Leon et al., 1980), again suggesting the production of allelochemicals that can persist until the next growing season.

In a preliminary experiment, we arranged cress seeds in concentric circles around sorghum seeds germinating on moist filter paper in the center of large, 12-cm Petri dishes. In every trial, the cress seedlings farther removed from sorghum were visibly larger and healthier as they developed. When sorghum was germinated on one side of a divided Petri dish and cress or weeds on the other side, no effect was observed even though the cress or weeds were in close proximity and shared the atmospheric environment of the Petri dish. Taken together, these observations suggested that sorghum produces allelochemicals that are nonvolatile and are water-soluble to the extent that they were able to migrate from sprouting sorghum seeds to nearby locations on a moist filter paper. The reduction in the detrimental effect on cress with increasing distance from the germinating sorghum suggested that the allelochemicals are produced in limited amounts and diffuse slowly from the origin.

Later experiments with root exudates obtained from sorghum potted in sand or grown so that the roots protruded into the water gave further proof that the active allelochemicals are water-soluble. These root exudates suppressed the development of sprouting weed seeds, and the activity could be increased by concentrating the aqueous leachate.

While the allelopathic potential of young sorghum seedlings grown under the laboratory conditions described in this report seems evident, extrapolation of these findings to field conditions is not possible. Our laboratory systems were free of all but a few adventitious microorganisms. In the field, allelochemicals exuded by germinating sorghum seeds or sorghum roots would be subject to possible alteration by soil microorganisms, although the allelopathic effect of mature sorghum residues has been shown to persist from one growing season to the next. Furthermore, our experiments were carried out on moist filter paper, in sand, or with sorghum roots in water while certain soils probably have the capacity to bind and immobilize some allelochemicals. The growth of sorghum following sorghum is markedly decreased in sandy soil, but not at all in soils high in montmorillonite (Burgos-Leon et al., 1980), which supports the contention that allelochemicals may be absorbed on certain types of soil particles, such as clay. Extensive testing would be necessary to document the influence of young sorghum on weeds under various field conditions.

Phenolic compounds, such as *p*-coumaric, *m*-hydroxybenzoic, and protocatechuic acids, have been implicated as the principal allelochemicals produced

by mature sorghum roots (Burgos-Leon et al., 1980). It is likely that these or related phenolics are responsible for the very early allelopathic influence of sorghum on weeds that we have observed. Work is in progress to isolate and identify the active allelochemicals that are produced by sorghum from the time of germination through the first few weeks of growth.

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