Herbicidal activity and crop injury of aqueous extracts of sorghum leaves

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수수 식물체 추출물의 제초활성 및 작물 선택성

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Received on 11 April 2011, revised on 2 May 2011, accepted on 20 June 2011

Abstract : Herbicidal effects and crop selectivity of aqueous leaf extracts of sorghum (*Sorghum bicolor* L.) were evaluated against several weed species for developing sustainable weed management in organic farming. Aqueous sorghum leaf extracts were highly phytotoxic to different weed species. No broadleaf weeds were germinated in the concentration of 5 fold or higher concentrated sorghum leaf extracts and 90% of seed germination was inhibited within that range in grass species. Sorghum leaf extracts strongly inhibited the growth of different weeds by pre-emergence and foliar applications in greenhouse condition. Foliar application of sorghum leaf extracts had a higher inhibitory effect than the pre-emergence application. Broadleaf weed species were more susceptible than grasses to the application of sorghum leaf extract in foliar applications than grasses. *Galium spurium*, *Erigeron candensis*, *and Rumex japonicus* were completely killed at the highest concentrated sorghum leaf extract both in pre-emergence and foliar application. Most broadleaf weed species were inhibited more than 80% at pre-emergence application at 50 fold concentrated sorghum leaf extract. *G. spurium*, *E. candensis*, *R. japonicus*, *Eclipta alba*, *Plantago asiatica* and *Portulaca oleracea* were most susceptible to sorghum leaf extract in foliar application. Growth of most broadleaf weed species was suppressed by greater than 90% at 50 fold concentrated sorghum leaf extract. Most crop species were tolerant to sorghum leaf extract but shoot growth was slightly reduced by the application of 40~50 fold concentrated extracts. Sorghum leaf extract may used to control weeds in organic farming without affecting the growth of crop.

Key words **:** Aqueous extracts, Crop selectivity, Herbicidal activity, Organic farming, *Sorghum bicolor*

I. Introduction

Weed control by mechanical method is very expensive and is almost out of reach to the resource poor farmers. Chemical control, on the other hand, exhibits residual effects of herbicides in crop produces and leads to environmental pollution. Current agricultural practices involved in weed management rely strongly on the use of synthetic herbicides. Weed management with herbicides, although effective, can be costly and is increasingly problematic due to public concerns about health and environmental issues (Cheema and Khaliq 2000).

The search for natural weed control methods is emphasized all over the world. Allelopathy has been recognized as natural weed control approach (Hardwood 1979; Rice 1984). Different crops possess allelochemicals, which could be utilized for suppressing weeds (Putnam and Defrank 1979).

Sorghum species are known to us as cover crops and green manures and also for the production of grain and molasses. Besides, this crop species has a potential

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for allelopathic effect, which was first showed in crops grown in rotation with sorghum (Breazeale 1924) and later confirmed in several studies (Lehle and Putnam 1982; Putnam *et al*. 1983; Forney *et al*. 1985; Einhellig and Rasmussen 1989). The suppressive effects of sorghum on different weed species such as *Chenopodium album, Phalaris minor, Cyperus rotundus, Senebiera didyma* and *Rumex dentatus* have been observed (Cheema and Ahmad 1992). Mature sorghum herbage possesses a number of watersoluble all-elochemicals (Putnam and Duke 1974; Cheema 1988). The use of sorghum water extract (sorgaab) as a foliar weed inhibitor in wheat has been reported (Cheema *et al*. 1997). Similarly, Iqbal (1997) reported that sorgaab (sorghum shoot extract) reduced weed biomass by 25 ~35% and increased wheat yield by 18.6%. The effects of sorghum allelochemicals are selective, species specific and concentration dependent (Cheema 1988).

Modern agriculture is now challenged to reduce environmental damage and health hazards from chemical inputs and yet maintain a high level of production. The potential for undesirable environmental contamination from herbicides is relatively high, and there is a need for environmentally safe herbicides that are equally or more effective and selective than currently available synthetic herbicides. Hence, an alternative way is needed to control weeds in crop fields which have no adverse effect on the environment. Sorghum is a potential allelopathic crop, which possesses a number of allelochemicals. Accordingly, the present study was done to investigate herbicidal effects and crop injury of aqueous extracts of sorghum leaves for developing weed management practices in organic farming.

II. Materials and methods

1. Plant materials and growing condition

Sorghum seeds (SS-450) were collected from National Institute of Animal Science, Sung Hwan, Korea. Seeds were sown in upland field, Experimental Farm of Chungnam National University and allowed to grow for one month.

2. Preparation and extraction of samples

Sorghum leaves were collected from one month old seedlings. The fresh leaves were allowed to dry under shade condition in a vinylhouse for one week. The well dried leaves were brought to the laboratory and chaffed with fodder cutter (2~3 cm pieces). Chaffed sorghum leaves were soaked in water in the ratio of 1:20 (w/v) for 24 hours at ambient room temperature. This mixture (leaves + water) was boiled for 3~4 hours and then water extract was filtered using a coarse mesh to remove the plant residue. The filter extract was evaporated under vacuum for desire concentration of leaf extract.

3. Bioassay of leaf extract of sorghum in a growth chamber

The herbicidal activity of leaf extract of sorghum was tested on weed species (i.e., *Echinochloa crus-galli*; *Digitaria sanguinalis*; *Cyperus nipponicus*; *Chenopodium album*; *Amaranthus retroflexus*; *Plantago asiatica*; *Portulaca oleracea*; *Eclipta alba*; *Galium spurium*; *Erigeron canadensis*; *Polygonum hydropiper*; *Rumex japonicas*) in Petri dishes. Crops (corn, cucumber, red pepper, tomato, lettuce and perilla) injury of sorghum leaf extract were investigated in Petri dish culture system. Fifty seeds of each weed species and 10 seeds of each crop species were allowed to germinate in sterile Petri dishes (100×40 mm) on the surface of sterile Whatman #1 filter paper at concentrations of 0, 1, 2.5, 5 and 10 fold concentrated leaf extracts for 2 wk. A 10-ml solution of each concentration was added to individual Petri dishes. Afterwards, for a 2 wk period, the Petri dishes were placed in a growth chamber at 25℃ under standard cool-white fluorescent tubes with a flux rate of 550 μmol $s^{-1}m^{-2}$ and a 16-h photoperiod. Each treatment was replicated 4 times.

4. Effect of pre-emergence application of sorghum leaf extract on control of weeds and crop injury in greenhouse

The herbicidal efficacy of soil applied aqueous leaf extract of sorghum was evaluated as a pre-emergence compound against several weed species. The concentrated concentrations of sorghum shoot extract was 0, 10, 20, 30, 40 and 50 fold and was applied to different weeds and crop species as pre-emergence applications. Twelve weed species (i.e., *G. spurium; E. candensis*; *R. japonicus*; *E. alba*; *P. asiatica*; *P. oleracea*; *A. retroflexus*; *C. album*; *P. hydropiper*; *E. crus-galli*; *D. sanguinalis*; *C. nipponicus*) and twelve crop species (rice, barley, wheat, corn, soybean, lettuce, tomato, red pepper, radish, perilla, cucumber and Chinese cabbage) were grown in plastic pots (15×12 cm). Two thirds of the pots were filled with upland cultivated soil, and the pot was seeded with soil. The aqueous leaf extract was applied at concentrations at 0, 10, 20, 30, 40 and 50 fold concentrations using a knapsack CO_2 sprayer at a water volume of 1000 L ha^{-1}. Seedlings were grown in the greenhouse at 25 ± 5℃ for a 5-wk period. Treatments were replicated 4 times and arranged by completely randomized design in the greenhouse. Five weeks after the treatment, the shoot portion of each weed species was collected. The efficacy of sorghum leaf extract was measured from the shoot dry matter content of each weed species.

5. Effect of foliar application of sorghum leaf extract on control of weeds and crop injury in greenhouse

To determine the effect of foliar applied sorghum leaf extract, a post-emergence application test was performed on the same weeds and crop species used in the pre-emergence assay. Seedlings were grown in the greenhouse as described above for a 3-wk period. At 21 d after seeding, sorghum leaf extract was applied to the foliage of weed seedlings at 0, 10, 20, 30, 40 and 50 fold concentrations using a knapsack CO_2 sprayer at a water volume of 1000 L ha^{-1}. Three weeks after treatment, the shoot portion of each weed species was collected to determine the biomass.

6. Statistical analysis

Data were analyzed using the SAS Software release 9.2 (SAS Institute Inc., Cary, NC, USA). Means were separated with Tukey's studentized range test.

III. Results

1. Bioassay of sorghum leaf extract in a growth chamber

Aqueous sorghum leaf extracts were highly phytotoxic to different weed species. All the applied concentrations of leaf extract significantly suppressed the germination of all weed species (Table 1). The inhibition of germination was higher in the broadleaf weed species than in grasses. No broadleaf weeds were germinated following the application of sorghum leaf extract at or above 5 fold concentration and seed germination inhibited by 90% in grass species. The germination was inhibited by 50% in most broadleaf weed species at the original concentration (1 fold).

2. Pre-emergence herbicidal activity under greenhouse condition

After a 5-wk growth period, the presence of leaf extract resulted in significant reduction in shoot biomass of weeds. The growth inhibition of *R. japonicus* was 92.5% at 30 fold concentration of sorghum leaf

Table 1. Effect of different concentrations of sorghum shoot extract on germination inhibition of weed seeds.

Sorghum leaf extract (fold)[a]	% inhibition of germination											
	Ec[b]	Ds	Cn	Ca	Ar	Pa	Po	Ea	Gs	Ec	Ph	Rj
1	20.0d[c]	30.0c	33.3d	40.0c	22.2c	30.0c	50.0c	50.0b	50.0c	30.0c	50.0c	20.0c
2.5	50.0c	60.0b	66.7c	80.0b	77.8b	70.0b	75.0b	100a	83.3b	70.0b	75.0b	60.0b
5	80.0b	90.0a	88.9b	100a	100a	100a	100a	100a	100a	100a	100a	100a
10	100a	100a	100a	100a	100a	100a	100a	100a	100a	100a	100a	100a

[a]times concentrated than the original extract concentration.
[b]Ec, *Echinochloa crus-galli*; Ds, *Digitaria sanguinalis*; Cn, *Cyperus nipponicus* Ca, *Chenopodium album*; Ar, *Amaranthus retroflexus*; Pa, *Plantago asiatica*; Po, *Portulaca oleracea*; Ea, *Eclipta alba*; Gs, *Galium spurium*; Ph, *Polygonum hydropiper*; Rj, *Rumex japonicus*.
[c]Mean values indicated by the same letter in a coloum did not differ significantly at the 5% level by Tukey's studentized range test.

Table 2. Effect of pre-emergence application of sorghum shoot extract on control of weeds by species in greenhouse.

Sorghum leaf extract (fold)[a]	Efficacy of weed control (%)											
	Gs[b]	Eca	Rj	Ea	Pa	Po	Ar	Ca	Ph	Ec	Ds	Cn
10	24.2d[c]	34.8c	48.6b	11.4de	12.0bc	13.3de	15.0de	16.2de	8.9d	11.1de	15.9cd	17.6d
20	61.5c	53.7b	88.4a	22.4d	33.4bc	29.2cd	35.6cd	39.1cd	27.9c	30.5cd	22.2cd	27.6d
30	78.1b	66.2b	92.5a	38.7c	54.0ab	39.8bc	47.6bc	54.3bc	36.3c	45.8bc	31.8bc	41.2c
40	93.5a	89.5a	100a	64.2b	60.6ab	50.4b	65.3ab	69.5ab	53.6b	53.8ab	51.6ab	52.9b
50	100a	100a	100a	82.0a	89.4a	74.3a	72.2a	89.7a	79.3a	74.7a	62.9a	86.5a

[a]times concentrated than the original extract concentration.
[b]Gs, *Galium spurium*; Eca, *Erigeron candensis*; Rj, *Rumex japonicus*; Ea, *Eclipta alba*; Pa, *Plantago asiatica*; Po, *Portulaca oleracea*; Ar, *Amaranthus retroflexus*; Ca, *Chenopodium album*; Ph, *Polygonum hydropiper*; Ec, *Echinochloa crus-galli*; Ds, *Digitaria sanguinalis*; Cn, *Cyperus nipponicus*.
[c]Mean values indicated by the same letter in a coloum did not differ significantly at the 5% level by Tukey's studentized range test.

extract followed by *G. spurium* and *E. candensis* that showed greater than 90% growth inhibition at 30 fold concentration of sorghum leaf extract (Table 2). At the highest sorghum leaf extract (50 fold concentration), *R. japonicus, G. spurium* and *E. candensis* were fully suppressed and the other broadleaf weed species was inhibited by more than 80% at pre-emergence application. *E. crus-galli* and *D. sanguinalis* showed 60~75% of growth inhibition at 50 fold concentration of sorghum leaf extract.

3. Post-emergence herbicidal activity under greenhouse condition

Aqueous extracts of sorghum applied as foliar spray exhibited significant phytotoxic activity against different weed species (Table 3). The foliar application of leaf extract, which was evaluated 21 days after treatment when seedlings were 6 weeks of age, resulted in significant growth reduction in broadleaf weed species showing greater susceptibility than grass weed species. Foliar application of leaf extract had a greater inhibitory effect on broadleaf weeds than the pre-emergence application. But, the growth inhibition of grass species was slightly less in foliar application. *G. spurium*, *E. candensis*, *R. japonicus*, *E. alba*, *P. asiatica* and *P. oleracea* were most susceptible to sorghum leaf extract. Growth of most broadleaf weed species was suppressed by greater than 90% at 50 fold concentration of sorghum leaf extract. *G. spurium*, *E. candensis* and *R. japonicus* were completely killed at the highest concentration of sorghum leaf extract. The growth reduction of *E. alba, P. asiatica, P. oleracea*, and *A. retroflexus* were 92.8, 89.6, 86.5 and 84.6%, respectively at 50 fold concentration of sorghum leaf extract (Table 3).

4. Crop injury of sorghum leaf extract

Most crop seeds germinated with higher percentage at highest concentrated sorghum leaf extract (Table 4). The growth inhibition of crop species as measured by shoot biomass was very low. Most crop species were tolerant to sorghum leaf extract showing a slight shoot growth reduction at 40~50 fold concentration. Among them, lettuce, cucumber and perilla were slightly susceptible to sorghum leaf extract that did not exceed more than 10% (Table 5). Growth inhibition in crop species was slightly higher at foliar application

Table 3. Effect of foliar application of sorghum shoot extract on control of weeds by species in greenhouse.

Sorghum leaf extract (fold)[a]	Efficacy of weed control (%)											
	Gs[b]	Eca	Rj	Ea	Pa	Po	Ar	Ca	Ph	Ec	Ds	Cn
10	60.5d[c]	25.6c	25.9e	21.1e	10.0c	10.0e	13.0cd	19.4cd	9.0c	13.9c	13.9d	13.7e
20	73.8c	67.6b	46.8d	40.7d	42.4b	26.2d	30.6bc	27.1c	24.3bc	32.9b	27.0cd	28.9d
30	89.3b	82.5ab	65.0c	54.1c	53.3b	41.4c	44.5b	34.1bc	49.2ab	40.2ab	40.7bc	37.6c
40	100a	90.3ab	87.7b	78.0b	71.0ab	67.6b	68.5a	52.4ab	60.3a	48.1ab	45.4ab	50.8b
50	100a	100a	100a	92.8a	89.6a	86.5a	84.6a	62.4a	65.7a	52.3a	57.7a	59.4a

[a]times concentrated than the original extract concentration.
[b]Gs, *Galium spurium*; Eca, *Erigeron candensis*; Rj, *Rumex japonicus*; Ea, *Eclipta alba*; Pa, *Plantago asiatica*; Po, *Portulaca oleracea*; Ar, *Amaranthus retroflexus*; Ca, *Chenopodium album*; Ph, *Polygonum hydropiper*; Ec, *Echinochloa crus-galli*; Ds, *Digitaria sanguinalis*; Cn, *Cyperus nipponicus*.
[c]Mean values indicated by the same letter in a coloum did not differ significantly at the 5% level by Tukey's studentized range test.

Table 4. Effect of different concentration of sorghum shoot extract on emergence of crops in growth chamber.

Sorghum leaf extract (fold)[a]	% emergence					
	Corn	Cucumber	Red pepper	Tomato	lettuce	Perilla
0	100.0	100.0	80.0	100.0	80.0	100.0
1	100.0	100.0	70.0	100.0	70.0	100.0
2.5	90.0	95.0	70.0	80.0	60.0	90.0
5	80.0	70.0	60.0	70.0	60.0	90.0

[a]times concentrated than the original extract concentration.

Table 5. Effect of pre-emergence application of sorghum shoot extract on dry weight of crops.

Sorghum leaf extract (fold)[a]	Dry weight (mg/pot)											
	Rice	Barley	Wheat	Corn	Soybean	Lettuce	Tomato	Red pepper	Radish	Perilla	Cucumber	Chinese cabbage
0	87.6 (0)[b]	711.4 (0)	457.7 (0)	1343.0 (0)	2196.5 (0)	214.0 (0)	50.2 (0)	45.0 (0)	1310.2 (0)	76.2 (0)	1448.0 (0)	735.3 (0)
10	86.0 (1.8)	715.6 ((0)	459.6 ((0)	1338.1 (0.4)	2200.0 ((0)	211.3 (1.3)	49.8 (0.8)	45.2 ((0)	1315.4 ((0)	75.8 (0.5)	1435.6 (0.9)	744 .6 ((0)
20	85.5 (2.4)	708.3 (0.4)	455.3 (0.5)	1322.5 (1.5)	2180.6 (0.7)	210.3 (1.7)	49.3 (1.8)	44.8 (0.4)	1301.6 (0.7)	74.1 (2.8)	1410.5 (2.6)	725.1 (1.4)
30	86.0 (1.8)	702.6 (1.2)	446.3 (2.5)	1298.6 (3.3)	2170.2 (1.2)	205.6 (3.9)	48.0 (4.4)	43.1 (4.2)	1284.5 (2.0)	72.6 (4.7)	1390.1 (4.0)	710.5 (3.4)
40	85.1 (2.9)	695.8 (2.2)	439.1 (4.1)	1287.6 (4.1)	2154.2 (1.9)	194.3 (9.2)	46.8 (6.8)	42.0 (6.7)	1260.0 (3.8)	70.3 (7.7)	1354.5 (6.5)	700.2 (4.8)
50	85.0 (3.0)	689.2 (3.1)	432.3 (5.5)	1280.0 (4.7)	2140.6 (2.5)	187.5 (12.4)	45.1 (10.2)	41.0 (8.9)	1254.2 (4.3)	69.5 (8.8)	1325.4 (8.5)	680.2 (7.5)

[a]times concentrated than the original extract concentration.
[b]Values in parentheses are % growth inhibition.

Table 6. Effect of foliar application of sorghum shoot extract on dry weight of crops.

Sorghum leaf extract (fold)[a]	Dry weight (mg/pot)											
	Rice	Barley	Wheat	Corn	Soybean	Lettuce	Tomato	Red pepper	Radish	Perilla	Cucumber	Chinese cabbage
0	98.6 (0)[b]	975.2 (0)	618.2 (0)	1502.3 (0)	2425.4 (0)	222.8 (0)	49.4 (0)	50.3 (0)	1682.4 (0)	82.7 (0)	1588.9 (0)	964.5 (0)
10	98.4 (0.2)	980.1 ((0)	620.4 ((0)	1514.3 ((0)	2404.3 (0.9)	210.4 (5.6)	48.7 (1.4)	49.8 (1.0)	1688.2 ((0)	81.4 (1.6)	1574.3 (0.9)	970.3 (0.6)
20	97.6 (1.0)	960.4 (1.5)	611.3 (1.1)	1501.3 (0.1)	2361.3 (2.6)	202.1 (9.3)	47.2 (4.5)	48.7 (3.2)	1676.2 (0.4)	78.6 (5.0)	1508.6 (5.1)	950.4 (1.5)
30	96.1 (2.5)	924.6 (5.2)	585.5 (5.3)	1488.4 (0.9)	2270.4 (6.4)	195.4 (12.3)	45.6 (7.7)	46.2 (8.2)	1635.4 (2.8)	70.4 (14.9)	1450.3 (8.7)	928.6 (3.7)
40	92.4 (6.3)	901.4 (7.6)	551.4 (10.8)	1483.2 (1.3)	2237.9 (7.7)	178.2 (20.0)	44.0 (10.9)	45.0 (10.5)	1615.4 (4.0)	67.6 (18.3)	1402.5 (11.7)	906.7 (6.0)
50	90.8 (7.9)	875.6 (10.2)	540.3 (12.6)	1425.2 (5.1)	2195.6 (9.5)	169.8 (23.8)	43.0 (13.0)	44.3 (11.9)	1567.2 (6.8)	65.7 (20.6)	1350.6 (15.0)	887.6 (8.0)

[a]times concentrated than the original extract concentration.
[b]Values in parentheses are % growth inhibition.

of sorghum leaf extract than pre-emergence application. The crop species such as rice, barley, wheat, corn, soybean, tomato, perilla and Chinese cabbage were very tolerant to sorghum leaf extract. Among the crop species studied here, lettuce, perilla and cucumber showed susceptible response in post-emergence application of sorghum leaf extract showing approximately 10% growth inhibition at 50 fold concentration (Table 6).

IV. Discussion

An increasing number of studies have shown that for managing weeds, allelopathic interactions can be utilized directly or indirectly through the use of allelochemicals as alternatives to herbicides (Dayan *et al*. 2000; Duke *et al*. 2002). From our study, significant growth reduction as measured by shoot growth in several broadleaf and grass species was observed after both pre-emergence and post-emergence applications of sorghum leaf extract, with the greatest phytotoxicity and growth reduction noted for the broadleaf weeds. In particular, *R. japonicus*, *G. spurium*, *E. candensis*, *P. asiatica*, and *A. retroflexus* were greatly inhibited (Table 3).

Einhellig and Rasmussen (1989) noted that in field grain sorghum residues were located the previous year, the inhibitory effects of grain sorghum were primarily on broadleaf weeds, with little activity observed on grass weeds. Putnam and DeFrank (1983) found that residues of sorghum reduce the number and biomass of common purslane and smooth crabgrass (*D. ischaemum* [Schr.] Muhl.) in the fields by 70 and 98%, respectively. All parts of sorghum like roots, herbage and germinating seeds release allelochemicals reducing the growth of grass and broadleaf species such as green foxtail, velvetleaf, and smooth pigweed (Panasiuk *et al*. 1986, Hoffman *et al*. 1996). Sorghum residues release sorgoleone, cyanogenic glycosides-dhurrin, and a number of breakdown products of phenolics that bring about weed suppression (Nicollier *et al*. 1983; Weston *et al*. 1989).

Foliar application of sorghum leaf extract resulted in significant growth reduction of weed species in all the studies. Affected species exhibited severe injuries in post-emergence application. Burning and growth inhibition were visually observed immediately after application in sensitive species, and significant stunting of growth was also measured by 21 days after treatment. The use of sorgaab as a foliar weed inhibitor has been reported in mung bean (Moosavi *et al*. 2011), soybean (Khaliq *et al*. 1999), horse purslane (Randhawa *et al*. 2002), in wheat (Cheema *et al*. 1997). The effects of sorghum allelochemicals are selective, species specific and concentration dependent (Cheema 1988).

Most crop species i.e., rice, barley, wheat, corn, soybean, red pepper, radish, tomato, and Chinese cabbage were tolerant to sorghum leaf extract. The growth inhibition was very low in crops; meaning that sorghum leaf extract contained allelopathic compounds and that their phytotoxicity remains lower in crop species.

V. Conclusions

In this study we found that aqueous sorghum leaf extracts were highly phytotoxic to the growth of various weed species. Significant growth reduction in several broadleaf and grass species was determined by both pre-emergence and post-emergence applications of sorghum leaf extract, with the greatest phytotoxicity and growth reduction noted for the broadleaf weeds at 50 fold concentrated concentration of sorghum leaf extract. However, most crop species were tolerant to sorghum leaf extract showing a slight shoot growth reduction at 40-50 fold concentration of sorghum leaves. The strong herbicidal ability of sorghum leaf extract might offer the possibility for effective biorational weed management.

Acknowledgments

This research was financially supported by the research grant (PJ005868 201004) from Rural Development Administration in 2010.

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