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ALLELOPATHIC SORGHUM AQUEOUS ROOT EXTRACTS INHIBIT GERMINATION AND SEEDLING GROWTH OF CROPS AND WEEDS

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ABSTRACT

Allelopathic sorghum aqueous extracts can be used as sprays against weeds of arable lands. Water-soluble allelochemicals in the aqueous extracts may also negatively affect crops. Root aqueous extracts from the South African landrace sorghum IS9456 and the Botswanan commercial variety Mahube, with high (584.69 µg mg^{-1} root fresh weight) and low (17.38 µg mg^{-1} root fresh weight) sorgoleone contents respectively, were tested on germination, radicle length, plumule length and dry weight of goosegrass [*Eleusine indica* (L.) Gaertn], blackjack [*Bidens pilosa* (L.)], maize [*Zea mays* (L.)], soya bean [*Glycine max* (L.) Merr.] and wheat [*Triticum aestivum* (L.)]. Factors in five glasshouse experiments were the two sorghum varieties and four root extract solutions (0%, 5%, 10% and 20%) (w/v) arranged in a randomised complete block design with six replications. There was no significant effect ($P>0.05$) of variety and root aqueous extract on germination, radicle length, plumule length and dry weight of maize and on germination of wheat and goosegrass. The sorghum accession IS9456 significantly ($P<0.05$) reduced plumule length and dry weight of wheat and goosegrass and germination, plumule length and dry weight of soya bean and blackjack compared to Mahube. Increasing strength of root aqueous extract solution significantly ($P<0.001$) reduced plumule length and dry weight of wheat and goosegrass as well as germination and dry weight of soya bean and blackjack. Extracts from IS9456 had greater inhibitory effects on crop and weed germination and growth compared to those from Mahube. Due to its low sorgoleone content and weak weed suppression from its root aqueous extracts, Mahube may have low potential for use in allelopathic weed control. The sorghum accession IS9456, which also produces very high sorgoleone content, may be used in integrated weed management exploiting allelopathy from both sorgoleone and water-soluble allelochemicals, although farmers will have to be careful in terms of crop rotations since the aqueous extracts also inhibit germination and growth of some crops. Field studies may be required to further confirm allelopathic effects of root aqueous extracts from IS9456.

Key words: sorghum allelopathy, aqueous extracts, germination, growth, maize, soya bean, goose grass, blackjack

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INTRODUCTION

Sorghum [*Sorghum bicolor* (L.) Moench] is an important cereal crop grown globally for food and feed purposes. It is the fifth most important cereal crop in the world after rice (*Oryza sativa* (L.), wheat (*Triticum aestivum* L.), maize (*Zea mays* L.) and barley (*Hordeum vulgare* L.) [1, 2] and is the main food grain for millions of people in the semi-arid tropics of Africa, Asia, and Latin America [3]. It is therefore a strategic crop for food security, especially in harsh environments [4]. Small-scale subsistence farmers grow most of the crop in many parts of the semiarid tropics [5, 6]. The crop is one of the most important drought tolerant cereal crops that are grown in Africa [7]. Sorghum's tolerance to drought is thought to be aided by its root system [8, 9]. The crop's deep root system increases water extraction depth while high root density increases water extraction area [8, 10].

Sorghum roots can also play a part in the plant's defense system against weeds. The roots can synthesise and exude phytotoxic compounds [11] capable of regulating plant and microbial communities in the rhizosphere [12]. The hydrophobic compound sorgoleone [13], which is exuded from root hairs of sorghum seedlings [14] causes most of the allelopathy that is associated with sorghum [15,16], and reduces sorghum germination and seedling growth through autotoxicity [17]. Apart from sorgoleone, sorghum produces water-soluble compounds, dhurrin and phenolic acids [18,19] which are believed to cause short-term growth suppression of susceptible plants [20,21,22].

Maize, wheat and soya bean, which are important food crops in Zimbabwe, may be grown in rotation, or as intercrops with sorghum [23]. Farmers can also include sorghum as a cover crop [24, 25]. These cropping systems can expose sensitive crops to aqueous allelochemicals that are produced by sorghum, resulting in reduced crop germination, poor seedling growth and reduced competition with weeds. Sensitivity of these crops to water-soluble allelopathic compounds during germination and early growth is not yet known. Research has shown that the herbicidal activity of sorgoleone is high on small-seeded weeds than in weeds with bigger seeds due to a variety of mechanisms that they use to escape its herbicidal effect [26]. It is not clear if crops such as maize, soyabean and wheat, which are large seeded, can also escape the herbicidal activity of water-soluble allelopathic compounds using mechanisms used by small-seeded weeds.

Weeds have the potential to reduce crop productivity through competition for resources [27]. In Zimbabwe, goosegrass [*Eleusine indica* (L.) Gaertn.] and blackjack [*Bidens pilosa* (L.)] are known to be problematic in farming areas. In the last national weed survey that was conducted in the small-scale farming areas of Zimbabwe, Chivinge [28] classified goose grass and blackjack as some of the most aggressive in all of the country's provinces, suggesting that these weeds can potentially infest major crops such as maize, wheat and soya bean. Being aggressive, these weeds will require multiple hand weeding times for effective control, which not only potentially exposes the soil to erosion, but also increases the cost of weeding. Where chemical weed control is practised, these aggressive weeds may require repeated applications of herbicides during the growing season. Heavy reliance on, and repeated use of herbicides, especially when the herbicides have the same mode of action, tends to select for herbicide resistance [29]. In Zimbabwe, weeds found in arable lands comprise broadleaf and grass weeds, offering opportunity to harness sorghum allelopathy for weed control [30]. A number of field crops grown in Zimbabwe have been reported to be allelopathic [31,32,33], but smallholder farmers may not be aware of allelopathy as an alternative weed control technology.

In a study which examined farmer knowledge, attitude and practices on sorghum allelopathy in five sorghum producing districts of Zimbabwe, it was established that about a third of the interviewed farmers knew about the ability of sorghum to inhibit weeds through allelopathy [34]. The study also revealed that some smallholder farmers recognised cultivar differences in allelopathic effects, opening the opportunity for purposive use of such cultivars in integrated weed management in cropping systems that include sorghum. To validate farmer observations and to establish weeds and crops that are susceptible to injury by sorghum allelochemicals, a series of experiments that included quantifying sorgoleone in 353 sorghum accessions from Southern Africa [14], and testing potential involvement of aqueous extracts from local sorghum varieties, were conducted. These studies sought to generate knowledge on how best local smallholder farmers can harness sorghum allelopathy for weed control. The South African landrace sorghum accession IS9456, and the Botswanan commercial sorghum variety Mahube were previously found to produce different quantities of sorgoleone [34], suggesting that they may inhibit germination and growth of other plants through the activity of this lipophilic compound. Their ability to inhibit weed and crop germination and seedling growth through release of water-soluble allelopathic compounds has not been examined. The objectives of the study were to evaluate the potential allelopathic effects of aqueous extracts prepared from roots of IS9456 (a high sorgoleone producer) and Mahube (a low sorgoleone producer) on germination and early growth of three crops (maize, soya bean and wheat) and two weeds (blackjack and goose grass). The study also sought to examine if varying strength of root extract solution had inhibitory or stimulatory effects on germination and growth of weeds and crops.

MATERIALS AND METHODS

The experiment, which was part of a series of trials, was conducted in the Agronomy Laboratory, Department of Crop Science at the University of Zimbabwe (17.7850° S, 31.0546° E). The two sorghum accessions (IS9456 and Mahube), which were grown under supplementary irrigation at Panmure Experiment Station (31°47'E and 17°35'S) in Shamva, Zimbabwe were planted at an inter-row and in-row spacing of 100 cm x 100 cm to allow sufficient working space when harvesting roots at crop maturity. The soil in the plot was a medium grained sandy clay loam classified as Chromic Luvisols (Zimbabwean classification) or Rhodexeralf Alfisols (USDA classification [35]. Weed control was done by hand weeding. Prior to harvesting, the field was irrigated to field capacity to allow easy uprooting of the plants. In order to obtain the maximum quantities of roots, a hand hoe was used to dig a 30 cm radius around each sorghum plant to a depth of about 45 cm. Fifty plants of each sorghum accession were carefully uprooted. Soil around the roots was gently shaken off prior to gentle washing of the roots with running water. Roots were separated from stalks and weighed using a digital scale.

Sorghum water extracts from the sorghum varieties were prepared in solutions of 0%, 1:20 (5%), 1:10 (10%), and 1:5 (20%) (weight to volume) and were applied on weeds (blackjack and goosegrass) and crops (maize, wheat and soya bean). The protocol by Ben-Hammouda *et al*. [36] was used, with minor modifications. Instead of dried herbage, fresh sorghum herbage was used in the current study. Fresh roots from sorghum that was at the late dough stage were separated from stalks, washed gently with water and chopped into 2 cm pieces before being pounded using a mortar and pestle. The pounded roots were put in warm (40 °C) distilled water and kept for 72 hours at room temperature (23±2 °C) to extract aqueous compounds. After 72 hours, residues were filtered using Muslin paper and the supernatant fluid was further filtered using a No. 1 Whatman filter paper. Sorgaab, an aqueous extract of mature sorghum plants is formulated at approximately 10 % residue to water concentration and allowed to ferment naturally over time upon storage [11]. In the current study, each weed and crop was treated as a separate experiment. For each experiment, a 2 × 4 factorial arranged in a randomised complete block design replicated six times was used.

Seeds of blackjack and goosegrass were obtained from the Weed Research Unit at Henderson Research Station, Mazowe, Zimbabwe. Seeds of maize (variety SC727®), soya bean (variety SC Serenade®), and wheat (variety SC Sahai®), all from Seed Co Zimbabwe, were purchased from a local retail outlet. Both weed and crop seeds were sterilised in 5 % sodium hypochlorite solution for 5 minutes and rinsed with distilled water four times prior to planting. Petri dishes measuring 100 mm x 15 mm were lined with No. 2 Whatman filter paper discs. For the weed bioassay, 10 seeds of each weed species were planted in separate petri dishes equidistant from each other. For crops, 5 seeds of each crop were planted in each petri dish. In each petri dish, 6 ml of the sorghum root extract were applied using a pipette. In the control treatment distilled water was used for watering. The average temperature in the laboratory was 25.4°C. No artificial lighting was provided. Both the weed and crop experiments were terminated eight days after germination.

Data on percent germination, radicle length, plumule length, and dry weight were recorded. Seeds were considered germinated after the radicle protruded through the seed coat by at least 2 mm. Germination percentage was calculated by dividing the number of seeds germinated per petri dish by the number of seeds planted and multiplying by 100. Plumule and radicle lengths were measured using a string and a ruler. An electronic top-pan balance (Wensar PGB600) was used to measure plant dry weight. Data were subjected to analysis of variance using GenStat statistical package version 14 (VSN International, Hemel Hempstead, Hertfordshire, UK). The treatment and interaction standard error of differences were used to separate treatment means at the 5% level of significance.

RESULTS AND DISCUSSION

Maize

Table 1 shows the effect of sorghum variety as source of aqueous root extract, root extract concentration and their interaction on % emergence, radicle length, plumule length and dry weight of maize. Main effects of sorghum variety, root extract solution and their interaction were not significant (P>0.05) on percentage germination, radicle length, plumule length and dry weight of maize. Germination was 100% for all treatments. Radicle length, plumule length and dry weight in the 5%, 10% and 20% root extract solutions were not significantly different (P>0.05) to the radicle length, plumule length and dry weight achieved by the control.

Germination percentage, radicle length, plumule length and dry weight of maize were not affected by sorghum allelopathy possibly because the crop is tolerant to sorghum phytotoxins. A study that tested the allelopathic effects of weeds on six plants that included maize found that maize seeds had maximum (100%) germination [37]. In the current study, ability to reduce uptake of allelochemicals at the root surface, lower absorption and translocation or faster metabolic degradation of the allelochemical [13], partitioning of allelochemicals away from molecular target sites or detoxification of allelochemicals [38] possibly made maize avoid the herbicidal effect of allelochemicals from IS9456 and Mahube.

Soya bean

The variety × root extract solution interaction on germination of soya bean was highly significant (P<0.001) (Fig 1). There was a significant effect (P<0.001) of sorghum variety and root extract solution on soya bean germination (Table 2).

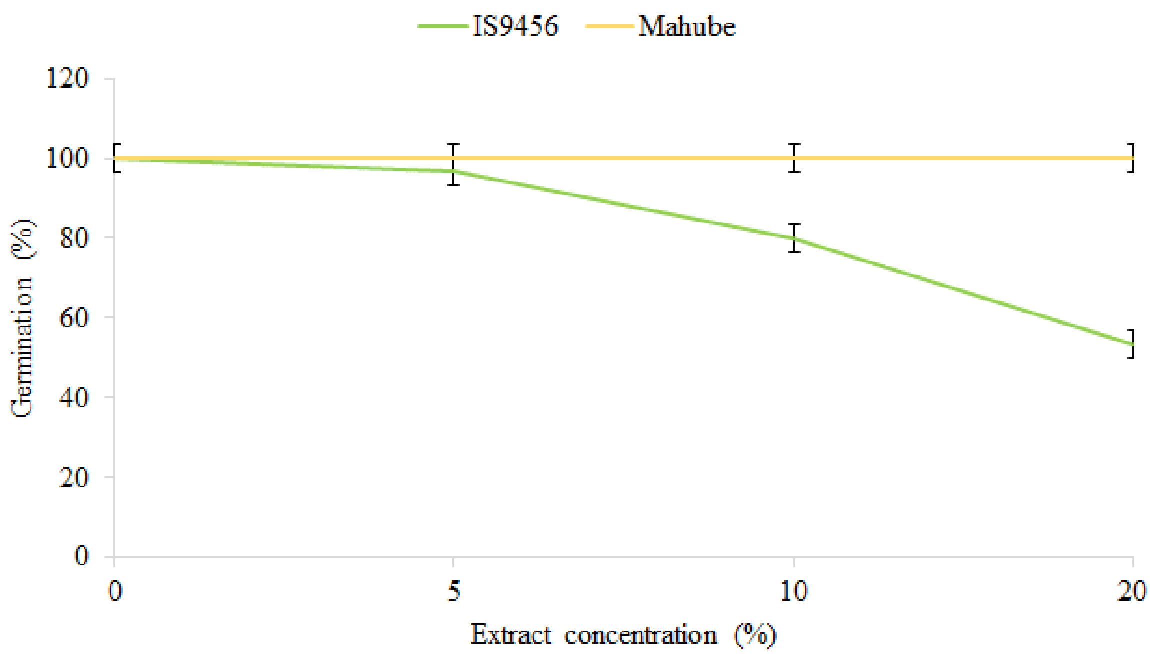

Figure 1: Variety × root extract solution interaction effect on germination of soya bean

There was no significant effect (P>0.05) of root extract solution from Mahube on soya bean germination. In contrast, soya bean germination decreased significantly (P<0.001) as strength of aqueous root extracts from IS9456 increased (Fig 1).

There was no significant effect (P>0.05) of sorghum variety and extract concentration and there was no variety x root extract solution interaction on radicle length of soya bean (Table 2). Extracts from IS9456 significantly (P<0.001) reduced dry weight and plumule length of soya bean by 26.1% and 40.5%, respectively, compared to extracts from Mahube. Plumule length and dry weight of soya bean significantly (P<0.001) decreased as strength of sorghum root extract solution was increased. There was no variety × root extract solution interaction on plumule length and dry weight of soya bean.

Wheat
Table 3 shows the effect of sorghum variety as source of aqueous root extract, root extract concentration and their interaction on % emergence, radicle length, plumule length and dry weight of wheat. There was no significant effect (P>0.05) of sorghum variety and root extract solution on percent germination of wheat. Extracts from IS9456 significantly (P<0.001) reduced radicle length of wheat by 12.9 % compared to extracts from Mahube. Radicle length did not significantly (P>0.05) decrease as strength of root extract solution was increased. There was no variety × root extract solution interaction effect on radicle length of wheat. Extracts from IS9456 significantly (P<0.001) reduced plumule length of wheat by 14.1 % compared to extracts from Mahube. Plumule length significantly (P<0.001) decreased as strength of root extract solution increased. There was no variety × root extract solution interaction effect on plumule length of wheat.

Extracts from IS9456 significantly (P<0.001) reduced dry weight of wheat by 20.2 % compared to extracts from Mahube. Dry weight significantly (P<0.001) decreased as strength of root extract solution increased. There was no variety × root extract solution interaction effect on dry weight of wheat.

Black jack
There was a significant (p<0.001) variety × root extract solution interaction on germination of blackjack (Fig 2). Germination significantly (P<10.001) decreased when strength of root extract solution from IS9456 increased. In contrast, germination of blackjack did not significantly (P>0.05) decrease when strength of root extract solution from Mahube was increased.

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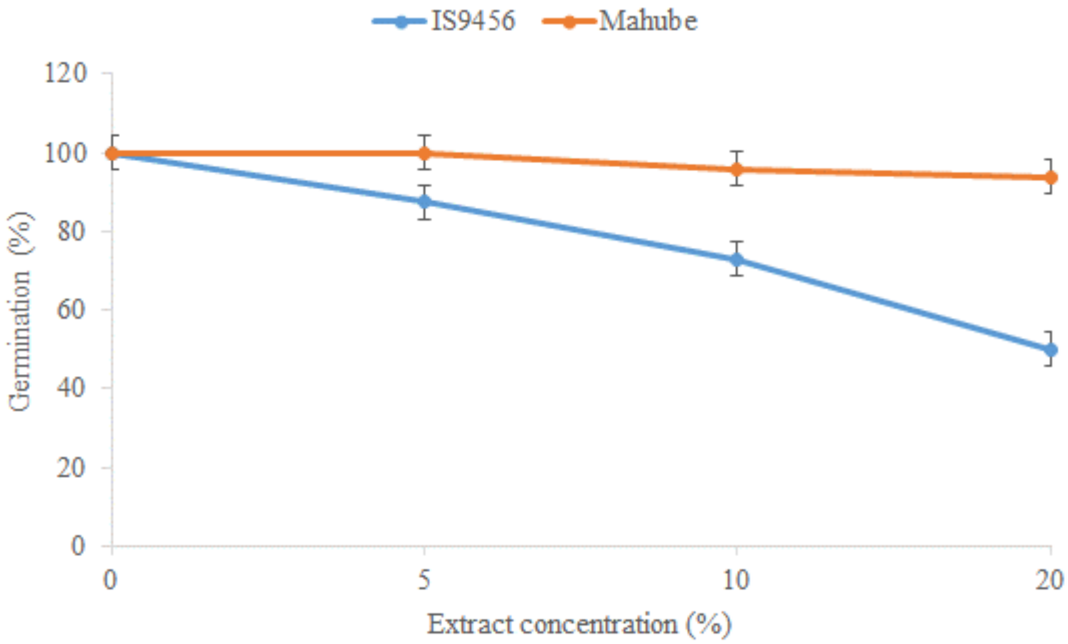

Figure 2: Variety ˣ root extract solution interaction effect on germination of blackjack

There was no significant effect (P>0.05) of sorghum variety and root extract solution on radicle length of blackjack (Table 4). There was no significant effect of sorghum variety ˣ root extract solution interaction on radicle length of blackjack. Extracts from IS9456 significantly (P<0.001) reduced plumule length of blackjack by 61.8 % compared to extracts from Mahube. Plumule length was not significantly (P>0.05) affected by root extract solution. There was no sorghum variety ˣ root extract solution interaction effect on plumule length of blackjack. Extracts from IS9456 significantly (P<0.001) reduced dry weight of blackjack by 33.4 % compared to extracts from Mahube. Dry weight significantly (P<0.001) decreased as strength of root extract solution increased (Table 4). There was no sorghum variety ˣ root extract solution interaction effect on dry weight of blackjack.

Goosegrass

Table 5 shows the effect of sorghum variety as source of aqueous root extract, root extract concentration and their interaction on % emergence, radicle length, plumule length and dry weight of goosegrass. There was no significant effect (P>0.05) of sorghum variety and root extract solution on percent germination of goosegrass. There was no significant variety ˣ root extract solution interaction effect on percent germination of goosegrass. There was no significant effect (P>0.05) of sorghum variety and root extract solution on radicle length of goosegrass. There was no significant variety ˣ root extract solution interaction effect on radicle length. Extracts from IS9456 significantly (P<0.001) reduced plumule length of goosegrass by 56.1 % compared to extracts from Mahube. Plumule length of goosegrass significantly (P<0.001) decreased as strength of root extract solution was increased. There was no variety ˣ root extract solution interaction effect on plumule length of goosegrass. Extracts from IS9456

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significantly ($P<0.05$) reduced dry weight of goosegrass by 11.3 % compared to extracts from Mahube. Dry weight significantly ($P<0.001$) decreased as strength of root extract solution increased. There was no significant variety × root extract solution interaction effect on dry weight of goosegrass.

The result that germination percent of blackjack was significantly affected by sorghum variety as source of extract, whereas that of goosegrass was not significantly affected shows that the two weed species differ in their susceptibility to sorghum water extracts. These results agree with those of Cheema *et al*. [39] who studied the efficacy of sorghum water extract as a natural weed inhibitor in wheat and found that the aqueous extract had species-specific effects on weeds.

The significant interaction between sorghum variety as source of aqueous extract and the extract concentration shows that root extracts from the sorghum variety IS9456 reduced soyabean and goosegrass germination from 100 to 53.3% and 100 to 50%, respectively as aqueous root extract concentration increased from 0 to 20%. In contrast, root extracts from Mahube did not significantly reduce soya bean and goosegrass germination as aqueous root extract concentration increased from 0 to 20%. These results show differences in efficacy of sorghum root aqueous extracts from the two sorghum varieties, with aqueous root extracts from Mahube having no effect on the germination of the broad leaf weed and crop species and those from IS9456 reducing germination of broadleaf weed and crop species as the concentration of extracts increased from 0 to 20%. The results indicate that sorghum varieties differ in their potential allelopathic effects against weeds and crops, and that plant response to allelopathic aqueous extracts is concentration dependent. Randhawa *et al*. [40] found that sorghum water extract at higher concentrations (75 and 100%), suppressed the germination, root and shoot growth of *Trianthema portulacastrum*, and attributed this suppression to the presence of allelochemicals in the sorghum plant. In a related study, Kayode and Ayeni [41] found that germination and growth of cowpea (*Vigna unguiculata* L. Walp.) in extract-treated seeds decreased with the increase in the concentration of the extracts, indicating that the degrees of inhibition were concentration dependent.

Root extracts from IS9456 and Mahube showed differences in their inhibitory capacities against both weeds and crops, and IS9456 generally caused greater inhibitory effects than Mahube. The result suggests that in addition to the high sorgoleone content that was found in IS9456 by Tibugari *et al*. [14], the variety possibly produces allelopathic compounds that can leach out from roots. Since sorgoleone is highly lipophilic [13, 16, 42], it may not have been involved in the allelopathic inhibition that was observed in the current study. Rather, other allelopathic compounds, particularly dhurrin and simple phenolic acids, that have a propensity to leach from plant tissue, were most probably involved.

The result that allelopathic effects that were observed in root extracts from IS9456 differed with those observed in extracts from Mahube suggests that there are varietal differences in allelopathic potential between the two sorghum varieties. Cultivar differences in allelopathic potential have been observed in different studies [23, 43].

Differences in allelochemical production have been attributed to both genetic and environmental factors [42, 44].

The effect of sorghum variety, root extract solution and interaction of variety × root extract solution on germination percentage, radicle length, plumule length and dry weight varied with plant species. Interestingly, germination percent of maize, wheat and goosegrass, which are all members of the Poacea family, was not affected by both source of sorghum aqueous extracts and strength of root extract solution. This result suggests that, in terms of germination, grasses may have some tolerance to the water-soluble allelochemicals produced by roots of IS9456 and Mahube while broadleaf crops and weeds are highly susceptible to the compounds. Except for maize, dry weight of all plants was significantly affected by both source of extract and extract concentration. Under laboratory conditions, IS9456 and Mahube can effectively control the two weeds, goose grass and blackjack.

CONCLUSION

Aqueous root extracts from IS9456 have higher inhibitory effects on both weeds and crops, and this indicates that IS9456 produces higher concentrations of water-soluble allelopathic compounds than Mahube. The sorghum accession IS9456 may be used in integrated weed management exploiting allelopathy, although farmers will have to be careful in terms of crop rotations since the aqueous extracts also inhibit germination and growth of crops. The allelopathic water soluble compounds in roots of IS9456 and Mahube have not been identified and quantified, and therefore identifying and quantifying the compounds that are associated with the inhibitory effects on germination and growth of crops and weeds may be a key area for further studies.

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Table 1: Effects of source of aqueous sorghum root extract and root extract solution on germination and growth of maize

	% germination	Radicle length (mm)	Plumule length (mm)	Dry weight (g plant^{-1}
Sorghum variety (source of aqueous root extract)				
IS9456	100	34.56	42.94	8.28
Mahube	100	36.06	43.00	8.15
P value		P>0.05	P>0.05	P>0.05
± s.e.d.		1.146	1.304	0.390
Root extract solution				
0%	100	35.76	43.71	7.68
5%	100	35.83	43.75	7.93
10%	100	34.33	42.67	8.09
20%	100	35.75	42.50	8.62
P value		P>0.05	P>0.05	P>0.05
± s.e.d.		1.403	1.597	0.478
Variety × root extract solution interaction				
P value		P>0.05	P>0.05	P>0.05
± s.e.d.		1.984	2.259	0.675

Table 2: Effects of variety and root extract solution on germination and growth of soya bean

	% germination	Radicle length (mm)	Plumule length (mm)	Dry weight (g plant^{-1}
Sorghum variety (source of aqueous root extract)				
IS9456	76.7b	35.78	15.50b	5.256b
Mahube	100a	37.89	21.78a	6.622a
P value	P<0.001	P>0.05	P<0.001	P<0.001
± s.e.d.	2.85	1.465	0.645	0.1801
Root extract solution				
0%	100 a	36.36	22.06 a	8.105 a
5%	98.3 a	36.33	21.75 a	8.033 a
10%	90.0 b	36.08	18.42 b	6.042 b
20%	76.7 c	38.08	15.75 c	3.742 c
P value	P<0.001	P>0.05	P<0.001	P<0.001
± s.e.d.	3.50	1.794	0.790	0.2206
Variety × root extract solution interaction				
P value	P<0.001	P>0.05	P>0.05	P>0.05
± s.e.d.	4.94	2.537	1.118	0.3120

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Table 3: Effects of variety and root extract solution on germination and growth of wheat

	% germination	Radicle length (mm)	Plumule length (mm)	Dry weight (g plant^{-1}
Sorghum variety (source of aqueous root extract)				
IS9456	100	41.78b	33.56b	3.789b
Mahube	100	47.17a	38.28a	4.556a
P value		P<0.001	P<0.001	P<0.001
± s.e.d.		1.405	0.741	0.1237
Root extract solution				
0%	100	41.95	39.74 a	4.813 a
5%	100	42.00	39.50 a	4.800 a
10%	100	45.17	35.83 b	4.225 b
20%	100	46.25	32.42 c	3.492 c
P value		P>0.05	P<0.001	P<0.001
± s.e.d.		1.721	0.908	0.1515
Variety × root extract solution interaction				
P value		P>0.05	P>0.05	P>0.05
± s.e.d.		2.433	1.284	0.2143

Table 4: Effects of variety and root extract solution on germination and growth of blackjack

	% germination	Radicle length (mm)	Plumule length (mm)	Dry weight (g plant^{-1})
Sorghum variety (source of aqueous root extract)				
IS9456	70.1b	46.1	19.61b	3.361b
Mahube	96.5a	47.6	31.72a	4.483a
P value	P<0.001	P>0.05	P<0.001	P<0.001
± s.e.d.	2.50	2.41	1.091	0.1538
Root extract solution				
0%	100 a	47.3	27.22	4.581 a
5%	93.8b	47.2	27.17	4.567 a
10%	84.4 c	47.4	25.25	4.050 b
20%	71.9 d	45.8	24.58	3.150 c
P value	P<0.001	P>0.05	P>0.05	P<0.001
± s.e.d.	3.06	2.95	1.336	0.1883
Variety × root extract solution interaction				
P value	P<0.001	P>0.05	P>0.05	P>0.05
± s.e.d.	4.33	4.18	1.889	0.2664

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Table 5: Effects of variety and root extract solution on germination and growth of goosegrass

	% germination	Radicle length (mm)	Plumule length (mm)	Dry weight (g plant^{-1})
Sorghum variety (source of aqueous root extract)				
IS9456	100	43.83	17.33b	4.42b
Mahube	100	42.61	27.06a	4.92a
P value		P>0.05	P<0.001	P<0.05
± s.e.d.		1.913	0.460	0.202
Root extract solution				
0%	100	43.26	25.72 a	5.51 a
5%	100	43.00	25.58 a	5.45 a
10%	100	41.92	22.00 b	4.62 b
20%	100	44.75	19.00 c	3.95 c
P value		P>0.05	P<0.001	P<0.001
± s.e.d.		2.343	0.564	0.247
Variety × root extract solution interaction				
P value		P>0.05	P>0.05	P>0.05
± s.e.d.		3.313	0.797	0.349

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