



Original article

## Host Specificity of Sudan Witchweed (*Striga Hermonthica* [Del.] Benth.) Seed Germination and Haustorium Initiation in Response to Sorghum Root Exudates and Extracts <sup>1</sup>

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### Abstract

Field surveys and laboratory experiments were conducted during the season 2009/10 and 2010/11 in witchweed (*Striga hermonthica* [Del.] Benth.) endemic areas in Sudan to investigate the host specificity of witchweed populations collected from different locations with respect seed germination and haustorium initiation in response to sorghum root exudates and extracts. Field surveys were conducted to collect seeds from witchweed plants growing under their respective hosts; sorghum and millet. A total of fifteen witchweed populations were collected. Two *in vitro* experiments were conducted at the Phytopathology Center and Biology Laboratory, Faculty of Agricultural Sciences, University of Gezira, Sudan to study the effects of root exudates and root extracts of sorghum cv. Abu-70, cv. Wad Ahmed and cv. Hakika on percentage of seed germination and haustorium initiation. Treatments (fifteen witchweed populations and three sorghum cultivars) were arranged in a factorial completely randomized design with three replicates. Data were collected and subjected to analysis of variance. Means were separated for significant using Duncan's Multiple Range Test (at  $p \leq 0.5$ ). The results showed that, there were significant differences in seed germination and haustorium initiation of witchweed in response to root exudates and root extracts among sorghum cultivars and among the witchweed populations. However, the highest seed germination and haustorium initiation percentages attained by each of the witchweed population were on their respective hosts. This study suggests two levels of physiological specialization in witchweed in Sudan: intercrop specialization and intra-crop specialization. Moreover, two strains of witchweed are suggested, one specific to sorghum and the other, to millet. The existence of variability and host specificity within witchweed populations are suggested to be based almost entirely on differential response of *Striga* isolates to root exudates and root extracts from host.

**Keywords:** Variability, Specificity, Host, Witchweed, *Striga*, Germination, Haustorium, Sorghum, Exudates And Extracts.

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## INTRODUCTION

*Striga* spp., Orobanchaceae, are obligate root parasites on important cereals and leguminous crops in Sub-Saharan Africa. Existence of physiological strains, ecological variants and races of the parasite together with variability in size of the seed bank offer serious obstacles for development of simple and effective control measures. Co-evolution of the parasite with its hosts has resulted in both specificity and non-specificity within the genus (Babiker, 2007). Existence of biological strains was first reported by Lewin (1932). However, this area of host specificity and host specialization in *Striga* is not fully explored (Musselman, 1987). Two levels of physiological specialization have been suggested in *Striga*: inter-crop specialization (strain specificity to a crop species) and intra-crop specialization (strain specialization to a cultivar of a crop species) (Ramaiah and Parker, 1982). Ramaiah and Parker (1982) suggested the existence of physiological strains of witchweed in Africa, where it was observed that varieties of *Sorghum* resistant in one location were susceptible in others. King and Zummo (1977) reported the existence of physiological specialization in witchweed from West Africa following their analysis of parasite virulence on different host crops.

Several experiments were undertaken to study variability and host specificity in within witchweed populations and its interaction with selected hosts, however, little work has been done in Sudan. Therefore, this study was conducted to investigate the host specificity of witchweed populations collected from different locations in Sudan with respect seed germination and haustorium initiation in response to sorghum root exudates and extracts.

### Material and Methods

These experiments were conducted at the Phytopathology Centre and Biology Laboratory, Faculty of Agricultural Sciences, University of Gezira, Sudan in season 2010/11.

#### *Collection of witchweed seeds*

Field surveys were conducted during the rainy season 2009/10 in witchweed endemic areas in Gadarif, Gezira and Kordofan, i.e. Eastern, Central and Western Sudan, respectively. Field surveys were conducted to collect seeds from *Striga* plants growing under their respective hosts (sorghum and millet). A total of fifteen witchweed populations were collected. Twelve witchweed populations, one each, were collected from under sorghum in Galabat, Sumsum, Gadarif, Butana, El Fau (Gadarif area); Hasaheisa, Abu-Haraz, Hag-Abdalla, Barakat and Wad-Rabia (Gezira area); Um-Rawaba and El-Rahad (Kordofan area). Three witchweed populations, one each, were collected from under millet in Kadugli, Khour-Tagat and El Obied (Kordofan area). The seeds were surface sterilized by sodium hypochlorite, (NaOCl) 1% solution, for 3 min with continuous agitation. Subsequently the seeds were thoroughly washed with sterilized distilled water for several times. Floating seeds were discarded and the remaining seeds were stored at room temperature till used.

### ***Seed conditioning***

In these experiments, the disc technique described by Dafaallah (2006) was used. About 80-100, Glass Fiber Filter Paper (GFFP) (Whatman GF/C) discs (0.5 mm diameter) were placed one layer GFFP in a glass Petri-dish (GPD), 9 cm internal diameter (i.d). Striga seeds (25-50) were sprinkled on each disc and the GPDs, moistened with distilled sterilized water (4.5 ml), sealed with Parafilm, covered with black polyethylene bag, were incubated at 30°C in the dark for 12 days.

### ***Effects of sorghum root exudates on germination and haustorium initiation of witchweed***

An *in vitro* experiment was designed to study the effects of sorghum root exudates on germination and haustorium initiation of witchweed. Sorghum cultivars (cv. Abu-70, cv. Wad Ahmed and cv. Hakika) obtained from the Sorghum Programme, Agricultural Research Corporation (ARC), were used in this experiment. Crops seedlings, grown in Petri dishes for 24 hour, were transferred each to rockwool in plastic pots. In each case rockwool was moistened with 200 ml of sterilized distilled water and incubated in a growth chamber for 10 days prior to collection of root exudates under suction using a suction pump. Aliquots of each root exudates (30µl) were applied to each pair of discs containing conditioned witchweed seeds placed on glass fiber discs (8 mm i.d.). The Petri dishes were sealed with Para film, placed in black polyethylene bags and incubated at 30°C in the dark. Treatments (fifteen witchweed populations and three sorghum cultivars) were arranged in a factorial completely randomized design with three replicates. The seeds were examined for germination and haustorium initiation 24 and 72 hour (h) after incubation, respectively.

### ***Effects of sorghum root extract on germination and haustorium initiation of witchweed***

An *in vitro* experiment was designed to study the effects of sorghum root extracts on germination and haustorium initiation of witchweed. Sorghum cultivars (cv. Abu-70, cv. Wad Ahmed and cv. Hakika) were sown on filter paper rolls placed in plastic pots and watered daily for 6 days. Root samples (1g) each was obtained from crop seedlings and placed in a mortar. Five ml of distilled water were added and the roots were crushed using a mortar and pestle, subsequently centrifuged and the supernatant was collected. Conditioned witchweed seeds were treated with aliquots of the root extract (30µl each). Treatments (fifteen witchweed populations and three sorghum cultivars) were arranged in a factorial completely randomized design with three replicates. Seeds were examined for germination and haustorium initiation 24 and 72 h after incubation, respectively.

### ***Statistical analysis***

Data were collected and subjected to analysis of variance procedure. Means were separated for significance using DMRT at  $p \leq 0.05$ .

## Results

### *Effects of sorghum root exudates on germination*

Irrespective of witchweed collection, root exudates from sorghum cv. Wad Ahmed and Abu-70 induced significantly high germination (71.4 and 76.4%), respectively (Fig. 1). While, root exudates from sorghum cv. Hakika induced significantly low germination (41.8%).

On treatment with root exudates from sorghum cv. Abu – 70, germination of witchweed, sorghum populations, ranged between 60.0% for seeds collected from El Fau to 87.3% for seeds collected from Butana and Likewise from Um-Rawaba (Table 1). On treatment with root exudates from sorghum cv. Wad Ahmed, germination of witchweed, sorghum populations, ranged between 60.0% for seeds collected from EL Fau to 82.3% for seeds collected from Um-Rawaba. On treatment with root exudates from cv. Hakika, germination of witchweed, sorghum populations, ranged between 37.3% for population from EL Fau to 52.3% for population from Gadarif.

On treatment with root exudates from sorghum cv. Abu – 70, germination of millet populations, ranged between 53.0% for seeds from El Obied to 55.3% for seeds from Kadugli (Table 1). On treatment with root exudates from sorghum cv. Wad Ahmed, germination of millet populations ranged between 40.0% for population from El Obied to 41.7% for population from Khour-Tagat. On treatment with root exudates from cv. Hakika, germination of millet populations, in response to root exudates from Hakika, ranged between 20.3% for seeds collected from El Obied to 26.3% seeds collected from Khour-Tagat.

### *Effects of sorghum root exudates on haustorium initiation*

Root exudates from sorghum cv. Abu-70 and cv. Wad Ahmed Attained significantly moderate haustorium initiation (41.1 and 47.3%, respectively), while, root exudates from sorghum cv. Hakika showed slightly low haustorium initiation (33.6%) (Fig. 2).

On treatment with root exudates from sorghum cv. Abu- 70, haustorium initiation of witchweed, sorghum populations, ranged between 49.9% for seeds collected from Abu-Haraz and likewise from El-Rahad to 55.9% for seeds collected from Gadarif, with exceptional cause of Um-Rawaba population that achieved low germination 17.3% (Table 2). When *Striga* germilings were challenged with root exudates from sorghum cv. Wad Ahmed, haustorium initiation of witchweed, sorghum populations, ranged between 50.0% for seeds collected from Butana and similarly from El Fau and Wad-Rabia to 54.0% for seeds collected from Galabat and likewise from Barakat, with exceptional cause of Um-Rawaba that illustrated slightly low haustorium initiation 30.3%. On treatment with root exudates from sorghum cv. Hakika, haustorium initiation of witchweed, sorghum populations, ranged between 7.3% for population collected from El-Rahad to 47.3% for population collected from Barakat.

On treatment with root exudates from sorghum cv. Abu-70, haustorium initiation of witchweed, millet populations, ranged between 16.1% for population from El Obied to 57.1% for population collected from Khour-Tagat (Table 2). When *Striga* germilings were challenged with root exudates from sorghum cv. Wad Ahmed, haustorium initiation of witchweed, millet populations, ranged between 27.7% for population collected from El Obied to 50.0% for population collected from Khour-Tagat. On treatment with root exudates from sorghum cv. Hakika, witchweed, millet populations achieved low germination or less than 20.0%.

#### ***Effects of sorghum root extract on germination***

Root extracts from sorghum cv. Abu-70 induced relatively high germination (61.7%) (Fig. 3). Root extracts from sorghum cv. Wad Ahmed displayed moderate germination (48.8%), while root extracts from sorghum cv. Hakika was, significantly, less effective and induced (40.8%) germination.

On treatment with root extracts from sorghum cv. Abu-70, germination of witchweed, sorghum populations, was relatively high and ranged between 59.9% for seeds collected from El Fau to 72.8% for seeds collected from Gadarif (Table 3). There were no significant differences in germination between sorghum populations from Galabat, Sumsum, Gadarif and Um-Rawaba in response to root extracts for sorghum cv. Abu-70. On treatment with root extracts from sorghum cv. Wad Ahmed, germination of witchweed, sorghum populations, was moderate and ranged between 40.7% for population collected from Wad Rabia to 59.7% for populations from Galabat, Sumsum and Um-Rawaba (Table 3). In addition, treatment with root extracts from sorghum cv. Hakika, germination of witchweed, sorghum populations was low to moderate and ranged between 35.0% for seeds collected from Wad-Rabia to 53.3 for seeds collected from Galabat and likewise from Sumsum and Gadarif.

On treatment with root extracts from sorghum cv. Abu-70, germination of witchweed, millet populations, in response to root extracts from sorghum cv. Abu-70 was low and ranged between 37.7% for population collected from El Obied to 39.0% for populations from Khour-Tagat (Table 3). On treatment with root extracts from sorghum cv. Wad Ahmed, germination of witchweed, millet populations, in response to root extracts from sorghum cv. Wad Ahmed was rather low and ranged between 32.7% for population collected from El Obied to 35.3% for population from Kadugli. Germination of witchweed, millet populations, in response to root extract from sorghum cv. Hakika was poor and ranged between 20.7 for population collected from El Obied to 25.7% for population from Kadugli.

#### ***Effects of sorghum root extract on haustorium initiation***

Haustorium initiation in response to root extracts from sorghum cv. Hakika, cv. Abu-70 and cv. Wad Ahmed was 35.7, 47.3 and 46.4%, respectively (Fig. 4).

On treatment with root extracts from sorghum cv. Abu-70, haustorium initiation, of witchweed, sorghum populations, was moderate and ranged between 47.0% for seeds collected from Wad Rabia to 55.3% for seeds collected from Gadarif (Table 4). There were no significant differences in haustorium initiation between sorghum populations collected from Galabat, Sumsum, Gadarif and Um-Rawaba. When witchweed, sorghum populations, treated with root extracts from sorghum cv. Wad Ahmed, haustorium initiation was moderate and ranged between 43.7% for population collected from El Fau to 55.7% for population collected from Um-Rawaba. On treatment with root extracts from sorghum cv. Hakika, haustorium initiation of witchweed, sorghum populations, was relatively low to moderate and ranged between 33.7% for population collected from Wad-Rabia to 43.0% for population collected from Sumsum.

On treatment with root extracts from sorghum cv. Abu-70, haustorium initiation of witchweed, millet populations was poor and ranged between 15.1 – 17.3% (Table 4). When witchweed, millet populations, treated with root extracts from sorghum cv. Wad Ahmed, haustorium initiation, was low and ranged between 27.3% for seeds collected from El Obied to 33.7% for seeds collected from Kadugli. On treatment with root extracts from sorghum cv. Hakika, haustorium initiation of witchweed, millet populations, was low and ranged between 20.0% for populations collected from Khour-Tagat and El Obied to 25.0% for population collected from Kadugli.

### **Discussion**

The results showed that, there were significant differences in seed germination and haustorium initiation of witchweed in response to root exudates and root extracts from sorghum cultivars and among the parasite populations. The highest seed germination and haustorium initiation was induced by root exudates and root extracts of sorghum cv. Abu-70 followed by sorghum cv. Wad Ahmed, while, root exudates and root extracts of sorghum cv. Hakika induced the lowest seed germination and haustorium initiation. Also, the results revealed that, seed germination and haustorium initiation of witchweed collected from under sorghum in response to sorghum root exudates and root extracts was significantly high. However, germination and haustorium initiation of witchweed collected from under millet in response to sorghum root exudates and root extracts was significantly low.

These findings are in agreement with those of Ali, (2008) who reported that, root extracts and exudates from all plants tested were able to induce germination and haustorium initiation. However, the magnitude of germination and haustorium initiation varied with the parasite population and the host in question. These findings are, also, in agreement with the results of Rao (1982) who suggested that the earlier stages of parasite establishment may have greater importance in determining host specificity.

These findings are consistent with observation made by Wilson-Jones (1955) that two strains of witchweed exist in Sudan, one prevailing in Eastern and Central Sudan and only attacks sorghum while

in Western Sudan, both millet and sorghum were attacked. Furthermore, the strain on millet did not attack sorghum and vice versa. Sorghum was usually heavily attacked by witchweed in the clay soils of Central Sudan whereas millet was particularly immune, but the reverse was true on sandy soils.

The observed differential response of the two *Striga* strains to haustorium inducing factor(s) from sorghum and millet may indicate specificity of the haustorium factors. Such specificity may be related to differences in quality, identity and/or quantity of the haustorium factor. The observed differential response is consistent with a previous report by Astatt and Hansen (1978) who reported that the potential number of haustoria is a product of the concentration and/or quality of haustoria inducing factor and the parasite individual ability to respond.

### Conclusions

- The results showed that seed germination and haustorium initiation of the witchweed, sorghum populations, were invariably the highest on the true host.
- This study suggests two levels of physiological specialization in witchweed in Sudan: intercrop specialization and intra-crop specialization. Furthermore, the results confirm the existence of two host-specific strains.

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**Table 1.** Effects of sorghum root exudates on germination of witchweed seed populations

Witchweed population		Germination (%)		
		Sorghum cultivar		
Area	Location	cv. Abu-70	cv. Wad Ahmed	cv. Hakika
Gadarif	Galabat*	86.9 a	80.0 fg	50.3 lm
	Sumsum*	86.2 ab	80.0 fg	51.3 klm
	Gadarif*	86.6 a	81.0 efg	52.3 jkl
	Butana*	87.3 a	81.0 efg	50.3 lm
	El Fau*	60.1 h	60.0 h	37.3 q
Gezira	Hasaheisa*	83.4 bc	80.7 efg	45.3 n
	Abu-Haraz*	84.0 cde	79.3 g	40.3 op
	Hag-Abdalla*	83.2 cd	81.7 defg	44.3 n
	Barakat*	83.0 cd	80.7 efg	42.0 o
	Wad-Rabia*	81.5 defg	80.0fg	50.0 m
Kordofan	Kadugli**	55.3 i	41.0 op	25.0 r
	Khour-Tagat**	53.4 ij	41.7 op	26.3 r
	Um-Rawaba*	87.3 a	82.3 cdef	51.0 klm
	El Obied**	53.3 jk	40.0 op	20.3 s
	El-Rahad*	79.9 fg	82.0 cdef	39.7 pq
SE ±		0.71		
CV %		4.2		

\*, \*\*= *Striga* populations collected from under sorghum and millet, respectively.

Means in columns and rows followed by the same letter(s) are not significantly different according to Duncan's Multiple Range Test ( $P \leq 0.05$ ).



**Table 2.** Effect of sorghum root exudates on haustorium initiation of witchweed populations

Witchweed population		Haustorium initiation (%)		
		Sorghum cultivar		
Area	Location	cv. Abu-70	cv. Wad Ahmed	cv. Hakika
Gadarif	Galabat*	55.1 bc	54.0 cd	40.3 kl
	Sumsum*	53.9 cd	52.0 def	41.0 jk
	Gadarif*	55.9 ab	51.0 fg	42.3 j
	Butana*	53.0 cde	50.0 g	45.0 i
	El Fau*	53.8 cd	50.0 g	42.0 jk
Gezira	Hasaheisa*	51.1 fg	51.0 fg	42.3 j
	Abu-Haraz*	59.9 g	53.3 cde	34.7 m
	Hag-Abdalla*	55.0 bc	53.7 cd	38.7 l
	Barakat*	51.8 efg	54.0 cd	47.3 h
	Wad-Rabia*	53.7 cd	50.0 g	34.7 m
Kordofan	Kadugli**	17.9 q	29.0 no	20.0 p
	Khour-Tagat**	57.1 a	50.0 g	20.0 p
	Um-Rawaba*	17.3 qr	30.3 n	19.0 pq
	El Obied**	16.1 r	27.7 o	19.0 pq
	El-Rahad*	49.9 g	54.3 bc	17.3 qr
SE ±		0.62		
CV %		4.7		

\*, \*\* = *Striga* populations collected from under sorghum and millet, respectively.

Means in columns and rows followed by the same letter(s) are not significantly different according to Duncan's Multiple Range Test ( $P \leq 0.05$ ).

**Table 3.** Effect of sorghum root extracts on germination of witchweed seed populations

Witchweed population		Germination (%)		
		Sorghum cultivar		
Area	Location	cv.Abu-70	cv. Wad Ahmed	cv. Hakika
Gadarif	Galabat*	71.9 ab	59.7 gh	53.3 j
	Sumsum*	71.1 abc	59.7 gh	53.3 j
	Gadarif*	72.8 a	58.3 hi	53.3 j
	Butana*	67.8 d	49.7 kl	52.3 j
	El Fau*	59.9 gh	42.7 n	41.3 nop
Gezira	Hasaheisa*	69.2 cd	56.3 i	40.3 opq
	Abu-Haraz*	65.0 e	50.3 k	42.0 no
	Hag-Abdalla*	67.3 d	53.0 j	42.3 no
	Barakat*	60.1 fg	42.3 no	34.7 st
	Wad-Rabia*	62.3 f	40.7 nopq	35.0 s
Kordofan	Kadugli**	38.9 pqr	35.3 s	25.7 u
	Khour-Tagat**	39.0 qr	33.7 st	24.0 u
	Um-Rawaba*	71.0 abc	59.7 gh	48.0 l
	El Obied**	37.8 r	32.7 t	20.7 v
	El-Rahad*	70.3 bc	57.3 i	45.3 m
SE ±		0.71		
CV %		5.2		

\*, \*\*= *Striga* populations collected from under sorghum and millet, respectively.

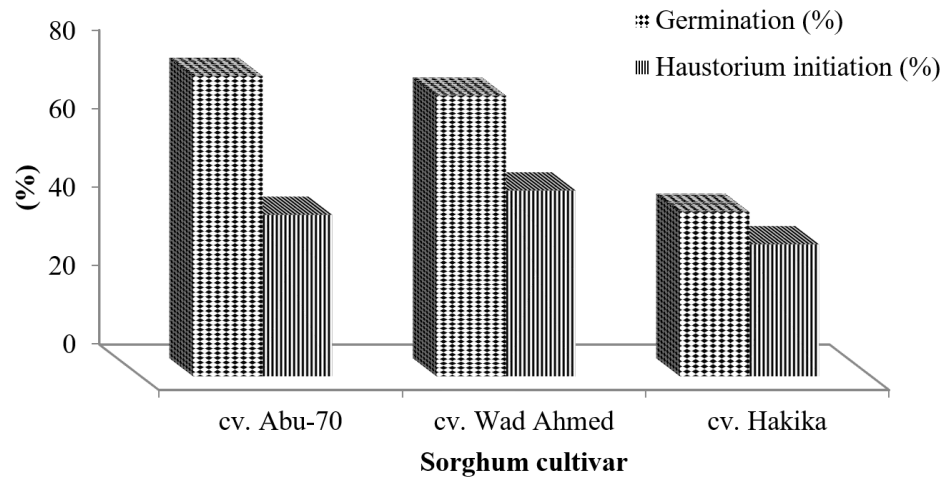
Means in columns and rows followed by the same letter(s) are not significantly different according to Duncan's Multiple Range Test ( $P \leq 0.05$ ).

**Table 4.** Effect of sorghum root extracts on haustorium initiation of witchweed populations

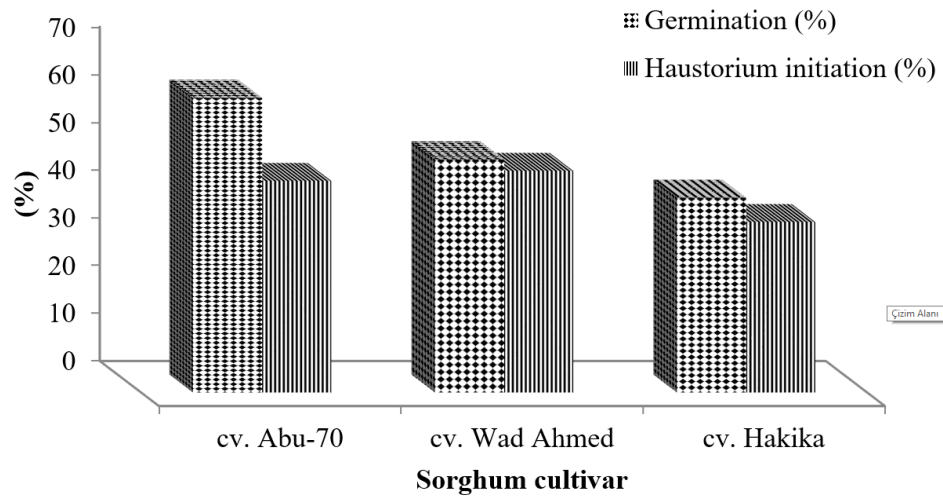
Witchweed population		Haustorium initiation (%)		
		Sorghum cultivar		
Area	Location	cv. Abu-70	cv. Wad Ahmed	cv. Hakika
Gadarif	Galabat*	54.9 abc	53.3 bc	42.0 kl
	Sumsum*	54.9 abc	54.0 abc	43.0 jk
	Gadarif*	55.3 ab	55.3 ab	40.3 lm
	Butana*	50.1 ef	50.3 ef	39.3 m
	El Fau*	49.1 fg	43.7 jk	35.3 no
Gezira	Hasaheisa*	51.2 de	51.3 de	41.0 lm
	Abu-Haraz*	50.0 ef	47.7 gh	42.3 kl
	Hag-Abdalla*	48.1 gh	53.0 cd	40.0 m
	Barakat*	47.7 gh	44.7 ij	35.3 no
	Wad-Rabia*	47.0 gh	46.0 hi	33.7 o
Kordofan	Kadugli**	17.3 t	33.7 o	25.0 r
	Khour-Tagat**	16.3 tu	29.3 p	20.3 s
	Um-Rawaba*	54.0 abc	55.7 a	36.3 n
	El Obied**	15.1 u	27.3 q	20.0 s
	El-Rahad*	53.2 bc	50.7 ef	40.7 lm
SE ±		0.65		
CV %		5.2		

\*, \*\*= *Striga* populations collected from under sorghum and millet, respectively.

Means in columns and rows followed by the same letter(s) are not significantly different according to Duncan's Multiple Range Test ( $P \leq 0.05$ ).



**Fig. 1.** Effect of root exudates of sorghum cultivars on germination and haustorium initiation of witchweed



**Fig. 2.** Effect of root extracts of sorghum cultivars on germination and haustorium initiation of witchweed