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EFFECTS OF 2, 4-D, GR24 AND CONDITIONING PERIOD ON *STRIGA HERMONTHICA*

RASHIDA MOHAMMED AHMED ABUSIN*
University of Bahri, Sudan

Department of Pests and Plant Health,
College of Agriculture,
University of Bahri,
Alkadaro, Khartoum North, Sudan
Tel: +00249916680095
E-mail: rashidaabusin@hotmail.com

ELFATIH ABDELAZIZ AHMED MOHAMMED
Crop Protection Department, Sudan

Agricultural Research Corporation, Sinnar,
Crop Protection Department, Sudan
E-mail: fatihaziz@yahoo.com

HAMZA AZIBER OUSMAN
University of N'Djamena, Chad

Department of Agricultural and Veterinary Sciences,
College of Applied Sciences,
University of N'Djamena, Chad
E-mail: aziber1977@yahoo.fr

MOHAMMED MAHGOUB HASSAN
National Centre for Research, Sudan

Bio-pesticides and Bio-fertilizers Department,
National Centre for Research, Khartoum, Sudan
E-mail: mohkadis@yahoo.com

*Corresponding author

AMANI HAMAD ELTAYEB HAMAD
Sudan University of Science and Technology, Sudan

College of Agricultural Studies,
Sudan University of Science and Technology, Sudan
E-mail: amani.hamad@yahoo.com

TILAL SAYED ABDELHALIM MUSA
Agricultural Research Corporation, Sudan

Horticulture Research Center,
Agricultural Research Corporation, Kosti, Sudan
E-mail: tilalkosti@yahoo.com

ABDELGABBAR ELTAYEB BABIKER
Sudan University of Science and Technology, Sudan

College of Agricultural Studies,
Sudan University of Science and Technology, Sudan
E-mail: agbabiker@yahoo.com

ABSTRACT

Purpose: This study was set to examine the effects of 2, 4-D on the early developmental stages of the root parasitic weed *Striga hermonthica*.

Design/Methodology/Approach: *S. hermonthica* seeds, sprinkled on 8 mm glass fibre discs, in Petri dishes, were treated with 2, 4-D, incubated, treated with GR24, examined for germination radicle extension and response to the haustorium inducer Dimethoxy-p-Benzoquinone (DMBQ).

Findings: *Striga* seeds conditioned in Distilled Water (DW), and subsequently treated with the synthetic germination stimulant GR24, displayed 53.7–87.4% germination. Seeds conditioned in 2, 4-D prior to treatment with GR24 exhibited 2–64.4% germination. Germilings from seeds conditioned in DW exhibited 8.8–45.4 mm radicle extension. Seeds conditioned in 2, 4-D displayed shorter radicles (0.4–6.2 mm). DMBQ applied to *Striga* germilings from seeds conditioned in DW effected 73.1% haustorial initiation. Germilings from seeds conditioned in 2, 4-D developed significantly less haustoria.

Originality/value: Germination, radicle extension and haustorium induction were negatively correlated with 2, 4-D concentration.

Keywords: 2, 4-D; *Striga*; GR24; germination; Dimethoxy-p-Benzoquinone; DMBQ; haustorium; radicle; Sudan.

INTRODUCTION

Witchweeds are the greatest biological constraint to cereals production in Africa. Of the all cereals, sorghum [Sorghum bicolor (Monech) L.], maize (*Zea mays* L.), pearl millet (*pennisetum glaucum* (L.) R. Br.) and rice (*Oryza sativa* L.) are the most affected and of all the species, *S. hermonthica* is the most important (Parker and Riches, 1993). *Striga* seeds germinate in response to stimulants exuded by hosts and some non-host plants (Jamil et al., 2010; Matusova et al., 2005). Following germination, the radicle elongates and, in response to a second root-derived signal, a haustorium is induced. A perquisite for successful parasitism is haustorium attachment, penetration of the host

roots, and establishment of connection with the host xylem. Subsequent to the establishment of a connection with the host xylem the parasite stays subterranean for 6–8 weeks prior to emergence above the ground. During the subterranean phase the parasite is totally dependent on its host and inflicts most of its damage (Babiker, 2007). Decreasing germination or radicle elongation and haustorium initiation would be expected to curtail contact, attachment and penetration of the host roots and, consequently, parasitism.

LITERATURE REVIEW

Prodigious seed production and prolonged viability of seeds coupled with the subterranean nature of early parasitism make *Striga* difficult to control (Babiker, 2007). Accordingly manipulation of seed germination and subsequent early developmental stages, together with prevention of seed replenishment, are important objectives in *Striga* management strategies (Babiker, 2007). Various chemicals, including synthetic germination stimulants, fumigants, antitranspirants and herbicides, have been reported as a means of control for *Striga* (Egley, 1990).

The 2, 4-D is reported to be one of the most widely used herbicides for *Striga* control, both as soil and foliar applied treatments (Aly, 2007). However, a review of the effects of soil applied 2, 4-D on *Striga* revealed contrasting performances. The herbicide at 2 kg a.e. ha^{-1} was reported to curtail *Striga* emergence and increase sorghum yield significantly (Carsky et al., 1994; Lagoke et al., 1994). However, Parker and Riches (1993) enunciated an erratic performance for 2, 4-D as they reported occasional success of 2, 4-D in prevention of witchweed attack and reduction of crop damage. The success of soil applied herbicides, including 2, 4-D, is influenced by a multitude of factors including those that affect availability of the herbicide in the soil solution and the soil layer where the host roots are parasitised (Eizenberg et al., 2013; Parker and Riches, 1993). This is further compounded by environmental factors and temporal and spatial variability in root growth, positioning and distribution of the parasite seeds.

The present laboratory investigation was conducted to study the effects of the herbicide 2, 4-D on the early developmental stages of *Striga*, including germination, radicle extension and haustorium initiation.

METHODOLOGY

Experiments were conducted at the *Striga* Research Laboratory, College of Agricultural Studies, Sudan University of Science and Technology at Shambat. *Striga* seeds were collected from under sorghum from the Gezira, Sudan in 2011. A 2, 4-D, as Decbore, the synthetic *Striga* germination stimulant GR24 and Dimethoxy-p-Benzoquinone (DMBQ) were used. Treatments were laid in a Complete Randomized Design (CRD) with five replicates.

Effects of 2, 4-D on Striga seeds conditioning

S. hermonthica seeds were surface sterilised by immersion, for 3 min, in a sodium hypochlorite solution (1%), obtained by the appropriate dilution of a commercial sodium hypochlorite solution (Bleash containing 5% NaOCl). The seeds were sprinkled on 8 mm Glass Fibre Filter Papers (GFFPD) and the discs (25–30 seeds/disc) were placed in Petri dishes. Aliquots (5 ml) of aqueous solution of the herbicide at 10, 20, 40, 60 and 80 μM were added, each, to a Petri dish. The Petri dishes were sealed with Parafilm, wrapped in aluminium foil and incubated in the dark at 30°C $\pm$ 2 for 14 days. A control in which the seeds were conditioned in water was included. Subsequent to conditioning,

the seeds were treated with 20 μl (aliquots) of GR24 at 0.01 and 0.1 ppm per disc. The Petri dishes were sealed with parafilm, wrapped in aluminium foil and incubated in the dark at 30°C ± 2 for 24 hr prior to examination for germination and radicle extension. The germilings were treated with DMBQ (20 μl per disc) and examined for haustorium initiation 24 hr later.

RESULTS AND DISCUSSION

Effects of preconditioning witchweed seeds in 2, 4-D in response to GR24

Witchweed seeds pre-conditioned in water (control) displayed high germination (53.7–87.4%) irrespective of the conditioning period and GR24 concentration (Figures 1 and 2). A 2, 4-D applied during conditioning, irrespective of concentration or conditioning period, reduced seed germination significantly. Seeds conditioned in 2, 4-D at 10 and 20 μM for five days prior to treatment with GR24 at 0.01 and 0.1 ppm, displayed significant reductions in germination when compared with the aqueous control. Conditioning in 2, 4-D at 10, 20 and 40 μM for 10 days prior to treatment with GR24 at 0.01 and 0.1 ppm, reduced germination by 3.3–64.4% (Figures 1 and 2). Seeds conditioned in 2, 4-D at 10 μM for 15 days and subsequently treated with GR24 at 0.01 and 0.1 ppm displayed 26.8–30.3% germination. A further increase in concentration to 40 μM or more effected 3.2–10.7% germination (Figures 1 and 2).

The results of these investigations indicate that pre-conditioning witchweed seeds in 2, 4-D suppressed germination irrespective of GR24 concentration. Germination strategy in *Striga* is based on ethylene biosynthesis and action (Logan and Stewart, 1991). In conditioned *Striga* seeds, germination stimulants induce ACC synthase and increased the capacity of the seeds to oxidize ACC to ethylene (Babiker et al., 1993, 2000). Inhibition and promotion of *Striga* germination can be achieved by the manipulation of ethylene biosynthesis, ethylene action, or by promotion of ethylene metabolism or that of its immediate precursor ACC. A notable decline in germination occurred when 2, 4-D was applied at 10 and 15 days prior to stimulant treatment. It would appear that the action of 2, 4-D on witchweed germination is complex and is influenced by a multitude of factors, including penetration and accumulation of 2, 4-D in the seeds, inhibition, induction and/or activation of enzymes, as well as toxic effects.

Effects on radicle extension

Germilings from seeds conditioned in Distilled Water (DW) showed an increase in radicle extension with a conditioning period from 5 to 10 days, and a subsequent decline in further increases of the conditioning period to 15 days (Tables 1 and 2). Germilings from seeds conditioned in DW and treated with GR24 at 0.1 ppm displayed greater extension than those similarly conditioned and treated with GR24 at 0.01 ppm (Tables 1 and 2). Germilings from seeds conditioned in 2, 4-D at 10 μM, irrespective of the conditioning period, displayed a sharp decline in radicle extension (78.3–90.7%). A further increase in 2, 4-D in the conditioning medium to 20–80 μM decreased radicle extension by 84–99.1% (Table 1).

Effect of 2, 4-D on S. hermonthica haustorium initiation

DMBQ applied to *Striga* germilings resulting from seeds previously conditioned in water and treated with GR24, induced haustoria in 73.1% of the germilings (Table 3). Seed conditioned in 2, 4-D at 10 and 20 μM, stimulated to germinate with GR24 and subsequently challenged with

Table 1 Effects of 2, 4-D on *S. hermonthica* radicle extension

2,4-D (μM)	*Radicle extension (10^{-3} mm) (GR24 0.1 ppm)*		
	5	10	15
DW	28.6 ± 0.1^{a}	45.4 ± 0.1^{a}	15.2 ± 0.1^{a}
10	6.2 ± 0.1^{b}	4.2 ± 0.0^{b}	3.2 ± 0.0^{b}
20	4.6 ± 0.0^{bc}	2.6 ± 0.0^{bc}	2.0 ± 0.0^{bc}
40	3.6 ± 0.0^{bc}	2.0 ± 0.0^{bc}	1.6 ± 0.0^{bc}
60	3.2 ± 0.0^{c}	0.4 ± 0.0^{c}	1.6 ± 0.0^{bc}
80	2.0 ± 0.0^{c}	0.6 ± 0.1^{c}	0.8 ± 0.1^{c}
3-way ANOVA			
2, 4-D	1434.5^{***}		
Days	167.1^{***}		
2, 4-D *Days	148.8^{***}		

Note: $\pm$ standard errors of means. Means within a column having the same superscript letter(s) are not significantly different according to DMRT. *** = $P \leq 0.001$.

Table 2 Effects of 2,4-D on *S. hermonthica* radicle extension

2,4-D (μM)	*Radicle extension (10^{-3} mm) GR24 0.01*		
	5	10	15
DW	20.4 ± 0.1^{a}	$22.8 \pm 0^{a}1^{a}$	8.8 ± 0.1^{a}
10	4.4 ± 0.0^{b}	2.8 ± 0.0^{b}	2.6 ± 0.0^{b}
20	4.6 ± 0.0^{bc}	2.4 ± 0.0^{b}	1.6 ± 0.0^{b}
40	3.0 ± 0.0^{bcd}	0.8 ± 0.1^{b}	1.6 ± 0.0^{b}
60	2.6 ± 0.0^{cd}	1.2 ± 0.1^{b}	0.8 ± 0.1^{b}
80	2.0 ± 0.0^{d}	0.8 ± 0.1^{b}	0.8 ± 0.1^{b}
3-way ANOVA			
2, 4-D	556.2^{***}		
Days	90.0^{***}		
2, 4-D *Days	40.8^{***}		

Note: $\pm$ standard errors of means. Means within a column having the same superscript letter(s) are not significantly different according to DMRT. *** = $P \leq 0.001$.

Table 3 Effects of 2, 4-D on *S. hermonthica* haustorium initiation

2, 4-D (μM)	*Haustorium initiation (%)*
DW	73.1^{a}
10	44.9^{b}
40	36.7^{b}
SE$\pm$	5.7
LSD	17.5

DMBQ 24 hr post-germination, displayed 36.7% and 44.9% haustoria, respectively. These findings are in line with those reported by Keyes et al. (2000) on the inhibition of haustorium initiation in *S. hermonthica* by auxins and auxin-like herbicides.

The observed reductions in germination, radicle extension and haustorium initiation showed negative correlation with 2, 4-D concentration ($r = -0.8$ to -0.5). This correlation is in line with the suggestion made by Parker and Riches (1993) and Eizenberg et al. (2013) that there was availability of the herbicide in the soil solution and the soil layer where the host roots are parasitised. Availability and movement of herbicides in soil solution and their movement are functions of the herbicide, the environment and soil type. Irrigation water and rainfall may thus be determinant factors in determining performance of soil applied herbicides. Based on the results of this study, the use of soil acting herbicides must be restricted to high rainfall areas.

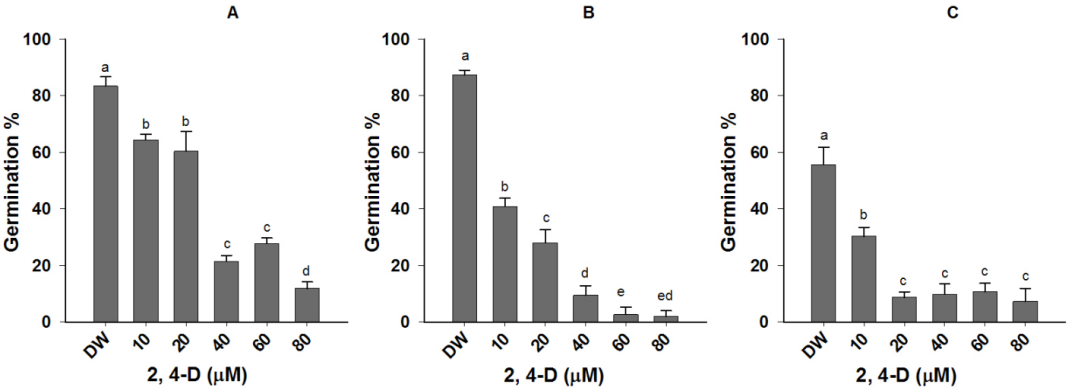

Figure 1 **Effects of 2, 4-D, applied during conditioning, on *S. hermonthica* seeds germination in response to GR24. Conditioning for A) 5 days (GR24 0.1); B) 10 days (GR24 0.1); C) 15 days (GR24 0.1). Bars are means ± standard errors according to DMRT; $P \leq 0.05$**

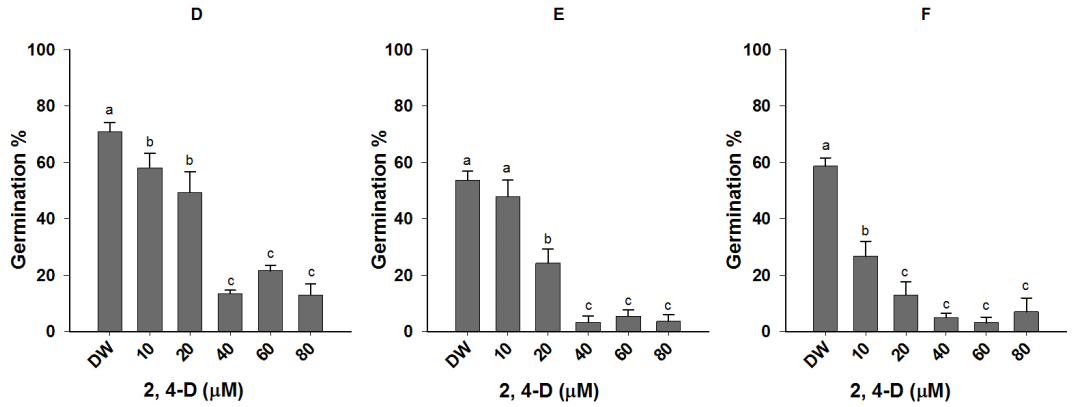

Figure 2 **Effects of 2, 4-D, applied during conditioning, on germination of *S. hermonthica* seeds in response to GR24. Conditioning for D) 5 days (GR24 0.01); E) 10 days (GR24 0.01); F) 15 days (GR24 0.01). Bars are means ± standard errors according to DMRT; $P \leq 0.05$**

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BIOGRAPHICAL NOTES

Rashida Mohammed Ahmed Abusin is an Assistant Professor of Weed Science, currently working as a Lecturer and a researcher of Weed Science and Plant Pathology, College of Agriculture, University of Bahri (formerly University of Juba), Khartoum, Sudan. She holds the headship of the Department of Pests and Plant Health. Her research interests deal with the integrated management of parasitic higher plants.

Elfatih Abdelaziz Ahmed Mohammed is an Associate Professor of Weed Science, Agricultural Research Corporation (ARC), Sennar, Blue Nile State. Currently, he is working as a Researcher of Weed Science especially in the Bio-pesticides and Bio-fertilizers.

Hamza Aziber Ousman is a Lecturer at the University of N'Djamena, College of Applied Sciences, Department of Agricultural and Veterinary Sciences. He has participated in designing the intermediate and secondary schools' science syllabi according to the programme of the Ministry of Education. He is presently on a part-time job as a lecturer at the Higher Institute for the Preparation of Teachers.

Mohammed Mahgoub Hassan is an Associate Professor of Parasitic Weeds Bio-Control, currently working as a researcher at the Bio-pesticides and Bio-fertilizers Department, Environment, Natural Resources and Desertification Research Institute, National Centre for Research, Khartoum, Sudan. He was awarded AU-TWAS young Scientist National Awards in Life Sciences Prize (2012) as well as distinguished PhD thesis, Sudan Academy of Sciences (2010). He has published more than 30 papers in peer-reviewed journals.

Amani Hamad Eltayeb Hamad is an Assistant Professor at the College of Agricultural Studies, Sudan University of Science and Technology, Khartoum, Sudan. She is the Head of Department of Agronomy. She is a research member of the project "Improvement of Food Security in Semi-Arid Regions of Sudan through Management of Root Parasitic Weeds" in collaboration with Japan Science and Technology (JST), Kobe University and Sudan University of Science and Technology.

Tilal Sayed Abdelhalim Musa is an Assistant Professor, Weed Research Scientist at the Agricultural Research Corporation (ARC), Kosti, White Nile State. Currently, he is working as a Senior Scientist. He has been a partner of the Research Project Sorghum Genotypes, Tillage Systems and Cover Crops for the Promotion of Mycorrhiza between (ARC) Sudan and schareika.uni-goettingen, Germany. He has published a book entitled Development and Integration of Biocontrol Products in Branched Broomrape (Phelipanche ramosa) management in Tomato.

Abdelgabbar Eltayeb Babiker is a Professor of Weed Science, Sudan University of Science and Technology, Khartoum, Sudan. He was a regional coordinator to *Striga* Research East Africa and to the International Sorghum and Millet Research (INTSORMIL) funded by USAID. He was a visiting Professor, Department of Agronomy, Purdue University, USA for 12 months conducting research on *Striga*. He was also a visiting professor at the Arid Land Research Center, Tottori University, Japan and University of Hohenheim, Germany. He has published hundreds of articles in reputable journals.