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Original Research Article

Host specificity of *Gibbago trianthemae* Simmons, a Phaeodictyoconidial Hyphomycetes fungus on *Trianthema portulacastrum* L. (Horse purslane)

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K e y w o r d s

Agricultural Crops, *Trianthema portulacastrum*, *Gibbago trianthemae*, Koch's postulates,

A B S T R A C T

A systematic field study was conducted in agricultural fields such as Food crops, Pulses, Vegetable crops, Oil crops and Commercial crops to estimate infestation of *Trianthema portulacastrum* L. (Horse purslane), a common terrestrial weed belonging to family Aizoaceae. The *in vitro* pathogencity studies on Horse purslane were conducted using spore inoculum ($5x10^4$/ml) of an indigenous fungus, *Gibbago trianthemae*. The pathogen was re-isolated from inoculated plants to fulfil Koch's postulates and confirmed its host specificity on Horse purslane. The disease caused by the isolate was critically analyzed and the results revealed that *Gibbago trianthemae* is a potential agent to biological control of Horse purslane.

Introduction

Trianthema portulacastrum L.[Horse purslane (Aizoaceae)] is a branched, prostrate, succulent, annual herb indigenous to South Africa (Adamson, 1962; Jeffrey, 1960, 1961), but is widely distributed in northern India and several other tropical and subtropical areas, including West Asia, Africa, and tropical America, as an invasive weed of cultivated fields and wastelands (Duthie ,1960; Holm *et al*., 1997).

It is considered as a major weed in various agricultural and vegetable crops, such as *Brassica spp*. (Mustard), *Zea mays* L.(Corn), *Cajanus cajan* (L.) Millsp. (Pigeonpea), *Glycine max* (L.) Merr. (Soybean), *Solanum lycopersicon* L. (Tomato), *Solanum tuberosum* L. (Potato), *Allium cepa* L. (Onion), and *Gossypium hirsutum* L. (Cotton). It has become a noxious weed due to competition for yields in many crops like *Pennisetum glaucum* L. (Millet), *Sorghum bicolor* L. (Sorghum), *Zea mays* L. (Maize), *Triticum aestivum* L. (Wheat), *Vigna mungo* L. (Mash), *Vigna radiata* L. (Mungbean), *Cyamopsis tetragonoloba* L. (Guar) and

Helianthus annuus L. (Sunflower) and causing significant reduction in the yield (Nayyar *et al.,* 2001).

In India, Horse purslane has been reported in the states of Uttar Pradesh, Punjab, Haryana, Rajasthan and Delhi and considered as a problematic terrestrial weed by virtue of its infestation in various agricultural and vegetable crops such as mustard, maize, pigeon pea, mung bean, potato, onion, cotton, soybean, pearl millet and sugarcane, especially during the rainy seasons (Balyan and Bhan, 1986; Simmons, 1986).Up to 60-70% infestation of this weed has been reported in pigeon pea and soybean fields and 80-90% in maize and brassica fields (Aneja *et al*.,2000).

Control measures against this weed include hand weeding and chemical herbicides as the most effective and immediate solution although these measures can control the weed on a small scale, they are not feasible for large infestations. Mycoherbicides are primarily attractive because they can be weed specific, have low environmental impact, and are often cost effective (TeBeest *et al*., 1992).

Biocontrol agents are generally perceived by the public to be more environmentally friendly and safer for users and consumers. Earlier studies (Aneja and Kaushal 1999; Aneja *et al.,* 2000; Bohra *et al.*. 2005; Boyette *et al.,* 2007; Mitchell, 1988) indicate the potential of biological control of Horse purslane using plant pathogens.

Boyette and Abbas (2001) showed that the bioherbicide *Myrothecium verrucaria* (Albertini & Schwein) Ditmar. Fr., isolated from sicklepod [*Senna obtusifolia* (L.) H.S. Irwin & Barneby], eliminated Horse purslane and several other weeds that have seriously interfere with commercially grown tomatoes. Similarly, Babu *et al.,* (2004) reported *Paecilomyces varioti* Bain. as mycoherbicide against horse purslane in south India.

Mitchell (1988) and Aneja and Kaushal (1999) reported on the herbicidal potential of *Gibbago trianthemae* Simmons against Horse purslane. The fungus has been recently described by Simmons (1986) as a new genus and species. Gibbago trianthemae has several characteristics similar to those of the genera Stemphylium and Alternaria but is distinct from them (Mitchell, 1988). Between 1989 and 1998 a series of surveys of plant pathogenic fungi associated with naturally infected Horse purslane were conducted in the states of Haryana and Punjab.

Infected leaves collected from various sites, yielded a species of *Gibbago,* identified as *Gibbago trianthemae* Simmons, a phaeodictyoconidial hyphomycetes fungus reported on horse purslane for the first time in India (Aneja *et al.,* 2000). Excepting work of Mitchell (1988) at the University of Arkansas, USA no other work has been done on controlling of *Trianthema portulacastrum* by biological means around the world.

The pathological study on Horse purslane with indigenous fungi is economically important to control of this common weed in many agricultural crops. The findings will be useful to develop mycoherbicide strategies for sustainable agricultural in India and all over regions of the world. Horse purslane is a harmful weed, infested in many vegetable crops like Brinjal, Okra, and other vegetables. The control of Horse purslane in field crops is very essential due to the increase of loss in yield of many crops in every year and also many farmers depended on these food and vegetable crops for their economy.

Materials and Methods

Field study

During the field study the common weed Horse purslane was heavily competing with agricultural crops such as Paddy, Jowar, Maize, Sugarcane, Ground nut, Brinjal, Tomato and Okra etc. The natural infection on leaves of *Trianthema portulacastrum* was observed and identified as leaf spot disease in study area. The infection of Leaf spot and Leaf blight caused by fungal pathogens were critically studied and photographs were taken with Nikon 12.1 Mega pixils Camera. The study includes different agricultural crops classifieds as Food crops, Pulses, Vegetable crops, Oil crops and Commercial crops in various agricultural regions of Vishakhapatnam District. The weed infestation was studied using random sampling method in all agricultural fields and some valuable information about the weed was gathered from local formers at Hanumanthuvaka (Experimental field of Dept. of .Botany, Andhra University), Madhuravada, Boyapalem, PM palem, Thagarapuvalasa regions and Mandals of Chodavaram and Devarapalli in Visakhapatnam District.

Field study was conducted based on three seasons (i) Kharif or South West Monsoon season (July to October, 2013), the major period of growth and shorter day length for flowering of rice, maize, castor and groundnut. (ii) Rabi /post Monsoon season (November to February, 2014),the major growth period and longer day length for flowering of wheat, mustard, barley, oats, potato, Bengal gram, cabbage and cauliflower.(iii) Zaid or summer season (March to June, 2014), the major growth period and longer day length for flowering of black gram, green gram, sesame and cowpea.

Collection of samples

The disease symptoms on leaves and stems were examined as round to oval straw colored spots with maroon margins. The diseased plants and propgules were collected in sterilized polythene bags and brought to the laboratory for the study of symptoms, isolation and pathogenicity test of the organism (s) involved in leaf spot disease. The disease symptoms on leaves and stem were critically observed and studied in plant pathology laboratory, Department of Botany, Andhra University, Visakhapatnam.

Isolation and identification of fungal pathogens

The diseased leaves were washed thoroughly in running tap water to remove soil particles and the infected portions of the leaves were cut into 1.0 – 1.5 cm. fragments. The pieces were surface sterilised by 70% ethyl alcohol for 1-2 minutes and then rinsed in sterile distilled water for six to seven times. Finally the leaf bits were rinsed in 0.01% mercuric chloride for 1 or 2 minutes fallowed by washing with sterile autoclaved double distilled water for 2 or 3 times. These fragments were transferred on to Potato dextrose agar (PDA) plates supplemented with 1% streptomycin sulphate (antibiotic) under sterile conditions in an inoculation chamber. After inoculation plates were incubated at 28 ±2°C for 21 days on a 12 h light/dark photoperiod. Pure cultures of fungi were maintained for the harvesting of spores in different growth media such as Potato Dextrose Agar (PDA), Czapek,s Dox Agar (CZA), Sabouraud's Dextrose Agar (SDA), and Trianthema extract dextrose agar (TeDA) [Fresh horse purslane leaves 200.0 g; Dextrose 15.0 g; Agar–agar 20.0 g; and Distilled water 1000.0ml.(Aneja *et al*.,2000)] supplemented with 1% Streptomycin. Identification of the

fungal isolates was made with help of the relevant literature (Simmons, 1986; Mitchell, 1988).

Test Plants

Seeds and seedlings of Horse purslane (*Trianthema portulacastrum*) were collected from agricultural fields during the field study. The collected seeds were dried and maintained in healthy condition without any contamination. The plants for further studies were grown by sowing the seeds in 10 cm diameter plastic pots containing sterilized, black soil. The pots containing seed lings of weed plants were maintained in a green house with a 12 h light/dark photoperiod. For host–range studies, each weed was maintained in four replicates along with control plant. The plants in the greenhouse were watered daily and fertilized farmyard manure when required.

Preparation of Spore inoculum

The pathogen isolated from diseased leaves of *Trianthema portulacastrum* was cultured in 10 cm diameter petri dishes containing PDA and CZA media and incubated for 14 days at 28 ± 2°C with a 12 h light/dark photoperiod. After that, conidia and mycelium were harvested with a sterilized spatula by flooding the plates with sterile 10 ml distilled water and then scraping the mycelial mass slowly for conidial suspension. The suspension was then filtered through sterile, muslin cloth folded in four layers and the final inoculum was taken into 100 ml conical flasks containing sterile distilled water and 5 ml of 0.01% (v/v) Tween 20 (Merck). The Inoculum concentration was adjusted to $5x10^4$ spores/ml using Improved Neubauer haemocytometer (Depth = 0.1mm).

Disease intensity (DI)

Inoculum was applied onto the test plants of *Trianthema portulacastrum* within 2 hours of sunset to avoid drying and to allow for a natural dew period shortly afterwards. Plants were observed three days after treatment (DAT) for disease symptoms. The intensity of infection was determined visually, based on the initiation of disease and increase in disease area on the leaves, stems of test plants every day. The disease intensity of pathogen on test plants was determined using a score chart (-, no symptoms, a healthy plant; +, mild symptoms, a plant showing slight symptoms on ≤15% of the leaf area; ++, moderate symptoms, a plant showing definitely bigger patches of diseased areas on 16 to 59% of the leaf area; and +++, severe symptoms, enlarged lesions covering 60 to 80% of the leaf area) (Ray and Hill, 2012).

Results and Discussion

Infestation of horse purslane in crop fields

Horse purslane is an introduced terrestrial weed in India; it has become a noxious weed due to competition for yields in various agricultural and vegetables crops (Table-1). It was considered as a major weed in field crops of *Zea mays* L. (Corn), *Cajanus cajan* (L.) Millsp. (Pigeonpea), *Solanum lycopersicon* L. (Tomato), and *Gossypium hirsutum* L.(Cotton) and noxious to many crops like *Pennisetum glaucum* L. (Millet), *Sorghum bicolor* L. (Sorghum), *Vigna radiata* L. (Mungbean), and *Helianthus annuus* L. (Sunflower) and causing significant reduction in the yield . 60-70% infestation of this weed was reported in pigeon pea and other pulses. 80-90% infestation of Horse purslane was observed in maize and vegetable crops.

Identification of pathogen

After approximately 5–7 days dark grey

velvety mold growth was observed on culture plates. Observations under a light microscope by staining of mycelial fragments confirmed that conidiophores of the isolated fungus were stemphylioid in general appearance , Conidia simple, pale straw colored with 1–4 transeptation and very slightly swollen at apex. Conidiophores solitary or 2-4 loosely fasciculate, erect, rarely distantly branched, simple with a single apical conidiogenous locus, then often proliferating by means of a secondary conidiophore that arises immediately bellow the apical cell of the existing conidiophores; septate, slightly pigmented. Conidia initially solitary, ellipsoid, beakless, pigmented, becoming transversely and longitudinally septate; with apical cells swelling slightly and producing secondary conidia similar to initial ones. Conidia were clear pale yellow–brown, smooth, broadly ellipsoid to broadly sub ovate-ellipsoid, having 1–4 complete or partial transverse septa (Figure-1). On the basis of these morphological and cultural characteristics isolated pathogen was identified as *Gibbago trianthemae* Simmons (1986), a phaeodictyoconidial hyphomycetes fungus. A pathogenicity test was performed and Koch's postulates were fulfilled by re-isolation of the fungus *G. trianthemae* from diseased tissues of *T.portulacastrum.*

Host specificity of *Gibbago trianthemae*

To confirm the pathogenicity, a wild isolate of *G. trianthemae* was multiplied on CZA and PDA culture plates. After 14 days, conidial suspensions were made using 10 ml (per plate) of sterilized distilled water and adjusted to $5x10^4$ conidia per ml using a heamocytometer after sieving through a muslin cloth. The conidial suspension was sprayed on leaves of 10 healthy test plants of *T. portulacastrum* using a hand sprayer. After inoculation plants were covered with polyethylene bags for 72 h to maintain at high humidity and then incubated in a glasshouse at 25–30 °C. Three days after inoculation numerous pinpoint maroon spots were examined on the leaves of inoculated plants (Figure 2). After 7–9 days these spots expanded and most of them coalesced causing chlorosis and defoliation, similar to the symptoms examined under field condition. In experimental pots, defoliation started after 18 days of inoculum spraying. Application of inoculum significantly reduced the production of leaves, height and biomass per plant as compared to control. Germination of *Gibbago* conidia took place within 6 to 12 hours therefore symptoms on leaves initiated as small pin point lesions, 3-4 days after spraying of inoculum. In moist chambers, growth of the fungus with conidiophores and conidia was observed on infected portions, 3 days after incubation. Lesions often coalesced and defoliation of leaves started 20 days post inoculation. Up to 85-95% infection was observed on the leaves and stems of test plants 30 days post inoculation.

Leaf spot disease on test plants

Symptoms on leaves and stems of test plants were examined as round to oval straw colored spots with maroon margins. As the disease progressed, affected leaves became chlorotic and dried up causing severe defoliation and withering of stems. Disease symptoms on leaves (upper and lower surfaces) were found as pinpoint round to oval maroon spots at initial stage of the disease. Spots expanded with the passage of time and became sunken and straw colored with maroon borders. Later, few spots coalesced and the whole leaf became chlorotic and dried up causing severe defoliation, leaving only a few new leaves on the stem tip. Under severe attack quite similar lesions were also examined around the stems causing withering.

Table.1 Diversity of Horse purslane (*Trianthema portulacastrum* L.) in agricultural crops

Type of Crop	English Name	Scientific Name	Weed Status
Food Crops	Paddy/Rice	*Oryza sativa* L.	Occasional
	Jowar/Great millet	*Sorghum bicolor* (L.) Moench	Common
	Bajra/Pearl millet	*Pennisetum glaucum* (L.) R.Br.	Common
	Maize/Corn	*Zea mays* L.	Common
	Ragi/Finger millet	*Eleusina coracana* Gaertner	Rare
Pulses	Red gram/Pigeon pea	*Cajanus cajan* (L.) Millsp.	Common
	Black gram	*Vigna mungo* (L.) Hepper	Common
	Green gram/Mung bean	*Vigna radiata* (L.) Wilczek	Occasional
	Horsegram	*Macrotyloma uniflorum* (Lam.) Verdc.	Occasional
Vegetables	Brinja/Egg plant	*Solanum melongena* L.	Common
	Okra/Ladies finger	*Abelmoschus esculentus* (L.) Moench	Common
	Tomato	*Lycopersicon esculentum* Miller.	Common
	Pepper/Capsicum	*Capsicum annuum* L.	Common
	Ridge gourd	*Luffa acutangula*(L.) Roxb.	Common
Oil Crops	Ground nut/Pea nut	*Arachis hypogaea* L.	Common
	Sesamum /Gingelly	*Sesamum indicum* L.	Occasional
	Sunflower	*Helianthus annuus* L.	Common
	Castor	*Ricinus communis* L.	Rare
Commercial Crops	Sugarcane	*Saccharum officinarum* L.	Rare
	Cotton	*Gossypium arboreum* L.	Common
	Tobacco	*Nicotiana tabacum*	Occasional
	Jute	*Corchorus olitorius* L.	Common

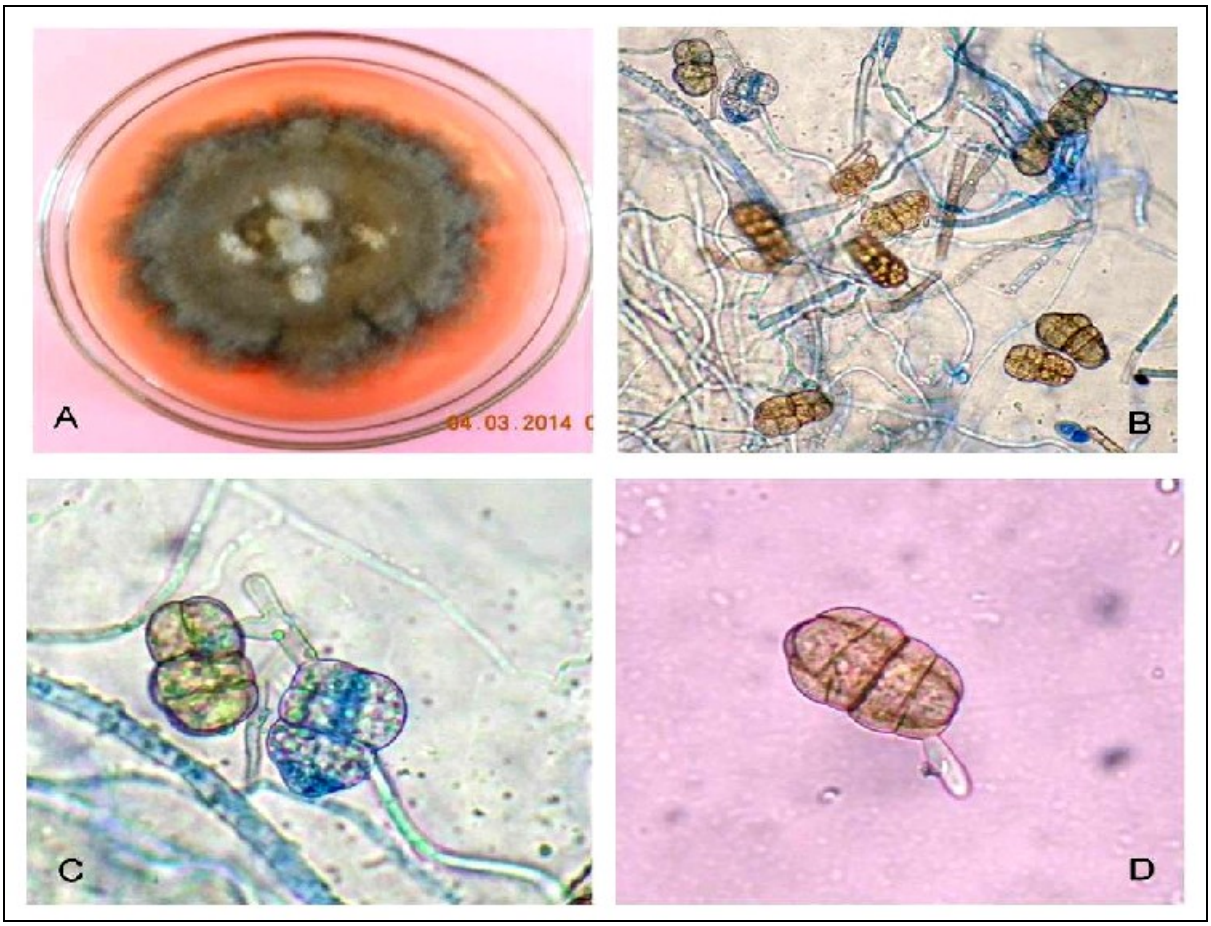
Table.2 Disease intensity on test plants inoculated with spore suspension ($5x10^4$/ml) of *Gibbago trianthemae*

Days After Treatment (DAT)	Disease intensity (DI)	
	Control plants [a]	Test plants [b]
3rd Day	–, no symptoms;	+, mild symptoms on 10% of the leaf area;
6th Day	–, no symptoms;	+, mild symptoms on 20% of the leaf area;
9th Day	–, no symptoms;	++, moderate symptoms on 35% of the leaf area;
12th Day	–, no symptoms;	++, moderate symptoms on 59% of the leaf area;
15th Day	–, no symptoms;	+++, severe symptoms, enlarged lesions covering 80% of the leaf area
18th Day	–, no symptoms;	Affected leaves became chlorotic and dried up causing severe defoliation and withering of stems.

'a' inoculated with sterilized distilled water

'b' inoculated with spore suspension of isolate

Figure.1 Macro and Microscopic observations of *Gibbago trianthemae*



A. Two week old culture of Gibbago trianthemae on CZA medium
B. Pale yellow– brown, smooth Conidia with 1–4 complete or partial transverse septa.
C. Phaeodictyoconidia of Gibbago trianthemae
D. Spore germination of Gibbago trianthemae

Figure.2 Pathological studies of Horse purslane (*Trianthema portulacastrum* L.)

A. Disease symptoms on leaves of Horse purslane in field study
B. Disease symptoms on leaves of Horse purslane in vitro study
C. Round to oval straw colored spots with maroon margins and chlorotic symptoms on test plant
D. Defoliation of Horse purslane after 14 days of treatment

Appearance of symptoms on leaves started after 3-4 days of spraying of inoculum. Initially symptoms were pinpoint, black with maroon margins upto 1 mm in diameter. The lesions became sunken and necrotic after 7-9 days of inoculum spraying (Table-2). Lesions often coalesced and abscised resulting in premature defoliation on test plants. As the lesions elongate they also expanded around the stem of inoculated plants.

***Gibbago trianthemae* as a potential Biocontrol agent**

The fungus *G. trianthemae* with similar cultural and morphological characteristics was re-isolated consistently from these inoculated leaves for the conformation of host specificity. The inoculated pathogen was re-isolated and found similar to the original isolate in cultural characteristics thus confirmed the pathogenicity of *G.*

Trianthemae to *Trianthema portulacastrum* and the Koch's postulates was fulfilled. The re-isolation of these isolate conformed that the pathogen Gibbago trianthemae causes leaf spot disease of *Trianthema portulacastrum* and a potential agent to biological control of Horse purslane in many agricultural crops of India.

G. trianthemae caused similar symptoms on *T. portulacastrum* in USA, Cuba, Venezuela and India a new phaeodictyoconidial genus of Hyphomycetes (Simmons 1986; Aneja and Kaushal 1999). Host range studies performed by Mitchell (1988) and Aneja et al. (2000) suggest that G. trianthemae is only pathogenic to T. portulacastrum, and has been suggested as a possible candidate for inundative mycoherbicide to control *T. portulacastrum*. *Gibbago trianthema*e on *Trianthema portulacastrum* is a new record for India (Aneja and Kaushal, 1999) and second for the world.

The present data suggest that *G. trianthemae* was approved as highly aggressive towards Horse purslane and has certain characteristics suggested by various workers that make it a desirable candidate as biological control agent of a weed, such as: capable of limiting population without eliminating the species; can be easily cultured on natural host; good sporulation capacity; infection can take place from conidia and/or mycelial fragments , narrow host range, fast growth rate and hence can be mass produced in a short time.

Weeds compete with crop for space, sunlight, moisture and nutrients thus decrease the crop yield. Horse purslane (*Trianthema portulacastrum* L.) is a common weed of maize, cotton and vegetables all over India. It is found as a major weed in maize, cotton, potato, sugarcane, and summer vegetables due to indeterminate habit, vegetative and reproductive growth continues for the entire life span. Keeping this in view, the pathological study was conducted on horse purslane *in vivo* and *in vitro* conditions for controlling of weed and investigation of indigenous pathogens as potential bio control agents to Horse purslane. Our observations revealed that Gibbago trianthemae is highly pathogenic to horse purslane as evidenced by the rapid rate of infection and colonization of the host in field and *in vitro* condition. Intensive work is still needed on the impact of the field environment and application technology on the efficacy of this pathogen as a mycoherbicide.

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References

Adamson, R. S. 1962. The South African species of Aizoaceae, 12: Sesuvium, Trianthema, Zaleya. J. S. Afr. Bot. 28:243–253.

Aneja, K. R. and S. Kaushal. 1999. Occurrence of Gibbago trianthemae on horse purslane in India. J. Biol. Control 12:157–159.

Aneja, K. R., S. A. Khan, and S. Kaushal. 2000. Management of horse purslane (*Trianthema portulacastrum* L.) with Gibbago trianthemae Simmons in India. Pages 27–33 in N. R. Spencer, ed. Proc. of the 10th Intl. Symp. on

Biol. Control of Weeds. Bozeman, MT: Montana State University.

Babu, R. M., A. Sajeena, and K. Seetharaman. 2004. Paecilomyces varioti as bioherbicide on Trianthema portulacastrum (L.) - a new host record. J. Mycol. Plant Pathol. 34:142–143.

Balyan, R.S. and V.M. Bhan. 1986. Emergence, growth and reproduction of horse purslane (*Trianthema portulacastrum*) as influenced by environmental conditions. Weed Sci. 34:516-519.

Bohra, B., B. N. Vyas, N. B. Godrej, and K. B. Mistry. 2005. Evaluation of Alternaria alternata (Fr.) Keissler for biological control of Trianthema portulacastrum L. Indian Phytopathol. 58:184–188.

Boyette, C. D. and H. K. Abbas. 2001. New bioherbicides whacks weeds.Vegetable Prod. & Marketing News. Pp. 9–10.

Boyette, C. D., R. E. Hoagland, and H. K. Abbas. 2007. Evaluation of the bioherbicide Myrothecium verrucaria for weed control in tomato (Lycopersicon esculentum). Biocontrol Sci. Technol. 17:171–178.

Duthie, J. F. 1960. Flora of the Upper Gangetic Plain. Delhi, India: Periodical Experts. 500 p.

Holm, L., J. Doll, E. Holm, J. Pancho, and J. Herberger. 1997. World Weeds: Natural Histories and Distribution. New York: J. Wiley. 1129 p.

Jeffrey, C. 1960. Notes on tropical African Aizoaceae. Kew Bull. 14:235–238.

Jeffrey, C. 1961. Aizoaceae (including Molluginaceae and Tetragoniaceae). Page 37 in O.B.E. Hubbard and E. Milne-Redhead, eds. Flora of Tropical East Africa. London: Crown Agents for Oversea Governments.

K. R. Aneja, S. A. Khan, and S. Kaushal. 2000. Management of Horse purslane (*Trianthema portulacastrum* L.) with *Gibbago trianthemae* Simmons in India. *Proceedings of the X International Symposium on Biological Control of Weeds* 4-14 July 1999, Montana State University, Bozeman, Montana, USA Neal R. Spencer [ed.]. Pp. 27-33.

Mitchell, J. K. 1988. Gibbago trianthemae, a recently described hyphomycete with bioherbicide potential for the control of horse purslane (*Trianthema portulacastrum*). Plant Dis. 72:354–355.

Mitchell, J.K. 1988. *Gibbago trianthemae*, a recently described hyphomycete with bioherbicide potential for control of horse purslane (*Trianthema portulacastrum*). Plant Disease, 72: 354-355.

Nayyar, M.N., M. Ashiq and I. Ahmad. 2001. Manual on Punjab Weeds 1:52 Directorate of Agronomy. Ayub Agricultural Institute, Faisalabad, Pakistan.

Ray, P. and M. P. Hill. 2012. Impact of feeding by Neochetina weevils on pathogenicity of fungi associated with waterhyacinth in South Africa. J. Aquat. Plant Manag. 50:79–84

Simmons, E.G. 1986. *Gibbago*, a new phaeodictyoconidial genus of hyphomycetes. Mycotaxon 27: 107-111.

TeBeest, D. O., X. B. Yang, and C. R. Cisar. 1992. The status of biological control of weeds with fungal pathogens. Annu. Rev. Phytopathol. 30:637–57.