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Biocontrol Agent for the Management of *Parthenium hysterophorus* L.

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Abstract
Invasive alien weed *Parthenium hysterophorus* L. is considered as one of the major threat to environment. It has adverse effect on human beings, livestock, crop yield, and biodiversity. In the present study during the survey of biocontrol agent for the management of parthenium number of insects such as *Aphis gossypii, Phenococcus sp., Elasmucha grisea*, leaf miner and *Zygogramma bicolorata* were observed on parthenium. Infestation of *Aphis gossypii* and *Phenococcus sp.* on parthenium was heavy and leaf miner and *Elasmucha grisea* was mild and infrequent respectively. These biocontrol agents also have wide host range thus not considered as suitable biocontrol agent. Due to very heavy infestation of *Zygogramma bicolorata* on parthenium and monophagus feeding habit, it was selected for detailed study. Insects were introduced at rosette stage of plant. After thirty days of continuous feeding, it was evaluated that there is reduction in plant height, shoot length, stem diameter, root length and plant biomass i.e., 65.11%, 56.16%, 23.63%, 12.39%, 71.63% respectively. It was also recorded that such insect fed plant did not attain flowering stage at all.

Keywords: Biocontrol agent, parthenium, infestation, *Zygogramma*

Introduction
Parthenium hysterophorus L. (Asteraceae) is an invasive alien weed species, native to Mexico and adjacent USA (McFadyen, 1981) [14]. It is commonly known as congress grass, carrot weed, parthenium and vernacular name *viz*. chatak chandani, and gajar ghas (Krishnamurthy *et al.* 1977) [12]. It was accidentally introduced in India in 1955 through food grain sent by USA under PL480 and first reported from Pune in 1956 (Rao, 1956) [21]. At present it has spread throughout most of the Indian Subcontinent (Aneja *et al*. 1991) [1]. It is weed of road sides, vacant lots and non-cropped areas, along non disturbed habitat (Singh *et al*., 1993) but also has been recorded as minor pest of cultivation in United States, Brazil, Argentina (Muenscher, 1995) [16] and India (Dhawan and Dhawan, 1994; Krishnamurthy *et al.*, 1993) [3, 11].
The plants are non latiferous and non-aromatic showing two very distinct phase juvenile, rosette or vegetative stage and adult, mature or reproductive stage in life (Dhawan and Dhawan, 1995) [2]. Depending upon the frequency and distribution of rain in year, the plant can complete one, two, or even three generation (Singh *at al.*, 1993) [24]. In India, Parthenium weed causes yield losses of up to 40% in several crops (Khosla and Sobti, 1979) [9] and it is reported to reduce forage production by 90% (Nath 1981) [18]. Indirectly parthenium act as an alternate for many crop pests (Robertson and Kettle, 1994) [22] and also act as secondary host for plant disease (Kishun and Chand, 1988; Jayanarayan *et al*., 1988) [10, 6, 7].
Parthenium causes severe human health problems such as allergy, dermatitis, eczema, black spots and blisters around eyes, burning, rings and blisters over skin and asthma etc (Tower *et al*., 1977) [26]. Parthenium is not palatable to livestock due to the irritating odour, taste and presence of trichomes. Due to parthenium contact animal get rashes on their udders, and whole bodies, loss of hair, depigmentation of skin, ulceration, and development of lesion in mouth and intestine. The bitter and reduced milk yield, contaminated with poisonous parthenin has been reported (Krishnamurthy *et al*. 1977) [12]. Other problems caused by this weed include blockage of common pathways, waterways and reduced aesthetic value of public parks.
In the present study biological control agent (insect) has been applied to control the population of parthenium, as it is permanent, energy efficient, inexpensive, and free from harmful side effects to the environment and the organism habituated in the natural course. Insects constitute the largest group of natural enemies of weeds. Effective biocontrol agents are found in order Hemiptera, Homoptera, Thysanoptera, Coleoptera, Lepidoptera, Diptera, and Hymenoptera.

Mechanism of action of weed feeding insects may include destruction of leaves, flower, stem, seed, tunnelling and feeding of leaf stem and root or formation of gall, destruction of photosynthetic capability and energy storage of weeds. Insect often transfer pathogenic agent that can move through the plant vascular tissue to cause further destruction. More than 350 species including arthropods (insects and mites). Fungi, nematodes, and vertebrates (fish, grazing animal and foul) have been released for biological control of weed worldwide (Julien & Griffiths, 1998) [8].

Materials and Methods:

Extensive survey of parthenium has been carried out in different localities of Allahabad to observe the infestation of insects on parthenium. Insects were collected and brought to laboratory and identified with the help of relevant literatures and guidance available (Walter Lisenmaier, 1972; George C. McGavin, 2000; Rajendra Singh and G.C. Sachan, 2004) [15, 13, 23] and from some expert opinion from Entomology Department of F.R.I. Dehradun.

Of the number of insects *Zygogramma bicolorata* was selected for detail experiments. Healthy and mature seeds of *P. hysterophorus* were sown in pots (25cm deep and 27cm diameter) filled with sterilized soil obtained from garden of Botany Department, Allahabad University. Soil was sterilized by placing moist soil in plastic container (2l.) and then one ampulla Methyl bromide was kept in the middle of container after breaking it and left for 48 hrs. The emerging seedlings with 2-3 leaves were transplanted to 12 earthen pots, 20 plants in each pot.

Adult *Zygogramma bicolorata* beetles were collected from the field and reared on *P. hysterophorus* plants within insect proof cages and newly emerged adults were used in the experiments.

Plants at rosette stage (4 weeks old) with an average of 5 leaves and 4 cm in height were transferred to 12 insect proof cages (64cm deep and 34cm diameter). Among the 12 cages, six cages were selected for 15 days experiment and another six cages for 30 days experiment. In both cases out of 6, 3 were control and 3 were treated.

In each treated cage newly, emerged adult beetles were introduced (10 insect per cage). After the completion of experiment the damage by insect was evaluated by observing the plants under following parameters i.e., total plant height, shoot length, root length, stem diameter and biomass of plants, obtained by drying in an oven at 55 degree Celsius for 72 hrs.

For studying the effect of *Z. bicolorata* beetles on flowering of *P. hysterophorus* plants, the insects were introduced to the plants at pre -flowering stage (8 weeks old) and observation recorded after 30 days.

The percentage of reduction in growth was calculated by following formula (Pant and Mukopadhyay, 2001) [19].

$$Q = a-b/a * 100$$

Q = % reduction in growth
a= average growth in healthy plant
b= average growth in treated plant.

Tests of significance (t value) were determined between healthy and treated plants on different parameters i.e., plants height, shoot length, stem diameter, root length and plant biomass.

Result and Discussion

During the survey number of insects such as aphids (order Hemiptera), Mealybug (Hemiptera), spider (Araneae), Butterflies (Lepidoptera), Flies (Diptera), beetles (Coleoptera), bugs (Hemiptera) and leaf miners were observed on *Parthenium hysterophorus* plants. Biocontrol potentiality of these insects for the management of noxious weed parthenium was also recorded. During the survey the intensity of infestation of most damaging insects such *Aphis gossypii*, *Phenococcus* sp., leaf miner, *Elasmucha grisea* and *Zygogramma bicolorata*, to *Parthenium hysterophorus* was observed and data were recorded in table (1). It is evident from the table (1) infestation of *Aphis gossypii* and *Phenococcus sp.* on parthenium was heavy during observation period. Infestation of *Elasmucha grisea* was infrequent and leaf miner was mild. Infestation of *Zygogramma bicolorata* was very heavy.

Table 1: Intensity of infestation of insects on *Parthenium hysterophorus*

Insects	Intensity of infestation	Mouth parts	Feeding habit
Aphis gossypii	+ ++	Piercing and sucking	Polyphagus
Elasmucha grisea	+	Piercing and sucking	Polyphagus
Phenococcus sp.	+++	Piercing and sucking	Polyphagus
Leaf miner	++	Sucking and biting	Polyphagus
Zygogramma bicolorata	++++	Biting and chewing	Monophagus

+ - infrequent ++ - mild
+++ - heavy ++++ - very heavy

Aphis gossypii commonly observed on stem, lower surface of leaves, inflorescence axis and on flower bud of parthenium. It feeds in cluster. They pierce the parthenium plant with their stylet and suck sap that causes wilting and discoloration of plants. Heavy infestation of aphids causes stunting of shoot growth and delay in production of flowers (Fig 1.A&B). During the survey infestation of A. gossypii was observed on varieties of other plants such as *Hibiscus rosa-sinensis, Abelmoschus esculentus, Ageratum conyzoides, Amaranthus viridis, Colocasia esculenta, Chrysanthemum sp., Helianthus annus, Tagetes erecta, Lycopercicum esculentum, Solanum melongena, Lochnera rosea, Ficus bengalensis, Psidium guajava* etc. Occurrence of *Aphis gossypii, A. spiraecola* and *A. fabae* on parthenium was also reported earlier (Rajendran, 1976; Sundara and Gowri, 1976) [20, 25]

Phenococcus sp. were observed in cluster on stem and leaf whorls of parthenium (Fig. 1 D&E). They generally attack the new growth therefore found near the shoot apex. Feeding damage caused stunting, distortion of new growth, leaf yellowing, premature leaf drop, die back and death of plant in heavy infestation. Infestation of *Phenococcus* was also observed on *Helianthus annus, Hibiscus rosa-sinensis, Lycopercicum esculentum, Solanum melongena, Lochnera rosea, Croton bonplandianum* and members of Cactaceae and Lamiaceae. *Phenococcus sp.* was also reported on parthenium (Hegde and Patil, 1979) [5].

Leaf miners reduced the photosynthetic efficiency of leaf. Mining of leaf tissues causes drying, browning and premature defoliation. Due to light infestation of leaf miners on parthenium, damage was not apparent (Fig.1 C). Severe infestation of serpentine leaf miner was observed on *Lycopercicum esculentum, Solanum nigrum, S.melongena, Pisum sativum, S. tuberosum, Cucumis sativus, Tagetes erecta and Chrysanthemum sp.* Healthy parthenium plants were able to tolerate such type of injury. Damage by *Elasmucha grisea*

on parthenium was also not obvious. Thus, due to wide host range these insects are not suitable bio-control agent for parthenium management.

Insect *Zygogramma bicolorata* was observed feeding ferociously on parthenium and damaging it (Fig 1.F-I).

Literature revealed that *Z. bicolorata* belongs to the beetle family Chrysomelidae subfamily Chrysomelinae. It was imported by Indian Institute of Horticulture Research (IIHR), Bangalore from Mexican substation of Common Wealth Institute of Biological Research (CIBC) in April 1983. Host specificity tests were carried out under strict quarantine measures which confirmed the safety of the insect for liberation under field condition (Jaynath and Nagarkatti, 1987) [6]. It was considered to be monophagus and released in August 1984.

In Present Investigation, the bio- control potential of *Zygogramma bicolorata* was evaluated on different growth stages of parthenium under caged experiments, the result was presented in table 2 and Fig.2. It was evident that feeding by larvae and adult after 15 days of introduction reduced the plant height by 38.16%, shoot length by 35.50%, stem diameter by 28.30%, root length by 10.68% and plant biomass by 40.29%.

In case of 30 days of continuous feeding by both adult and larvae the plant height was reduced by 65.16% shoot length by 56.16%, stem diameter by 23.63%, root length by 12.39% and plant biomass by 71.63%.

Heavy damage was recorded in leaves where the lamina of all the leaves were fed by adult and larvae and only midrib portion remained in some of the plants (Fig. 1 J). When such insect infested plant were left for another 30 days plants did not attain flowering which is significant for control of weed (Fig.1 K). However controlled plant flowered normally within this period .Thus continuous feeding by *Z. bicolorata* at rosette stage up to 30 days caused 100% elimination of flowering. Beetle feed the apical meristem and new emerging leaves, thus causes the reduction in plant height and shoot length. Biomass reduction occurred due to heavy foliage damage and loss of photosynthetic tissue. Earlier studies showed that parthenium is more susceptible to herbivory at rosette stage (Dheleepan and McFadyen, 2001) [4].

The insects were introduced at pre-flowering stage it was found that treated plant did not attain flowering whereas controlled plants flower normally (Fig.1 L). Thus, continuous feeding up to 30 days by beetle at pre-flowering stage caused 100% elimination of flowering.

It can be concluded that *Z. bicolorata* is an effective biocontrol agent for controlling the population of Parthenium. It damaged the all the growth stages of parthenium i.e. in rosette stage it fed the foliage and checked further development and in pre-flowering stage, completely inhibited the flower production .

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Table 2: Impact of *Zygogramma bicolorata* on the growth of *Parthenium hysterophorus* after 15 and 30 days of introduction

Criteria	Days of treatment							
	After 15 Days				After 30 days			
	Control	Treated	% reduction	Calculated "T" value	Control	Treated	% reduction	Calculated "T" value
Plant Height (cm)	10.35	6.4	38.16	8.69	17.2	6.0	65.16	15.65
Shoot length (cm)	8.55	5.6	35.50	12.07	11.59	5.08	56.16	12.63
Stem diameter(cm)	2.65	1.9	28.30	7.39	2.75	2.10	23.63	2.25
Root length (cm)	7.3	6.52	10.68	1.55	8.15	7.14	12.39	2.00
Plant biomass (mg)	479.0	286.0	40.29	580.0	164.0	164.5	71.63	137.44

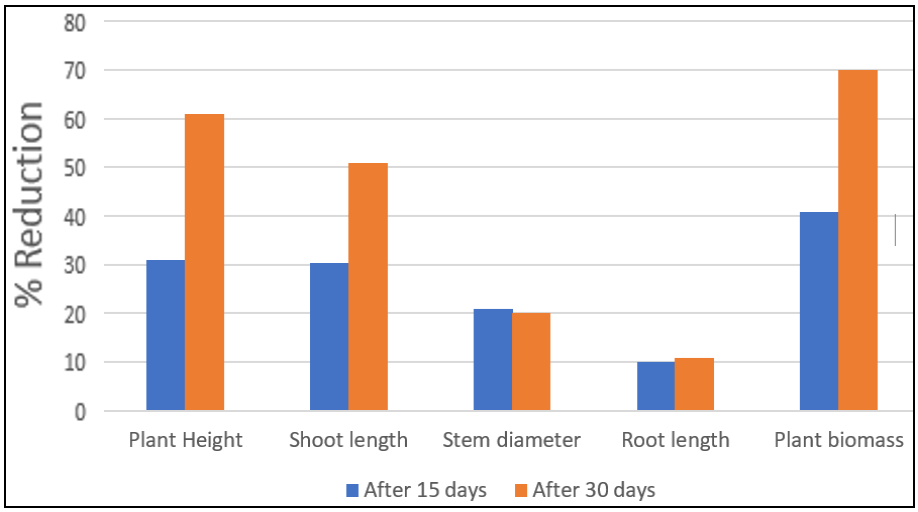

Fig 2: Impact of *Zygogramma bicolorata* on the growth of *Parthenium hysterophorus* after 15 and 30 days of introduction

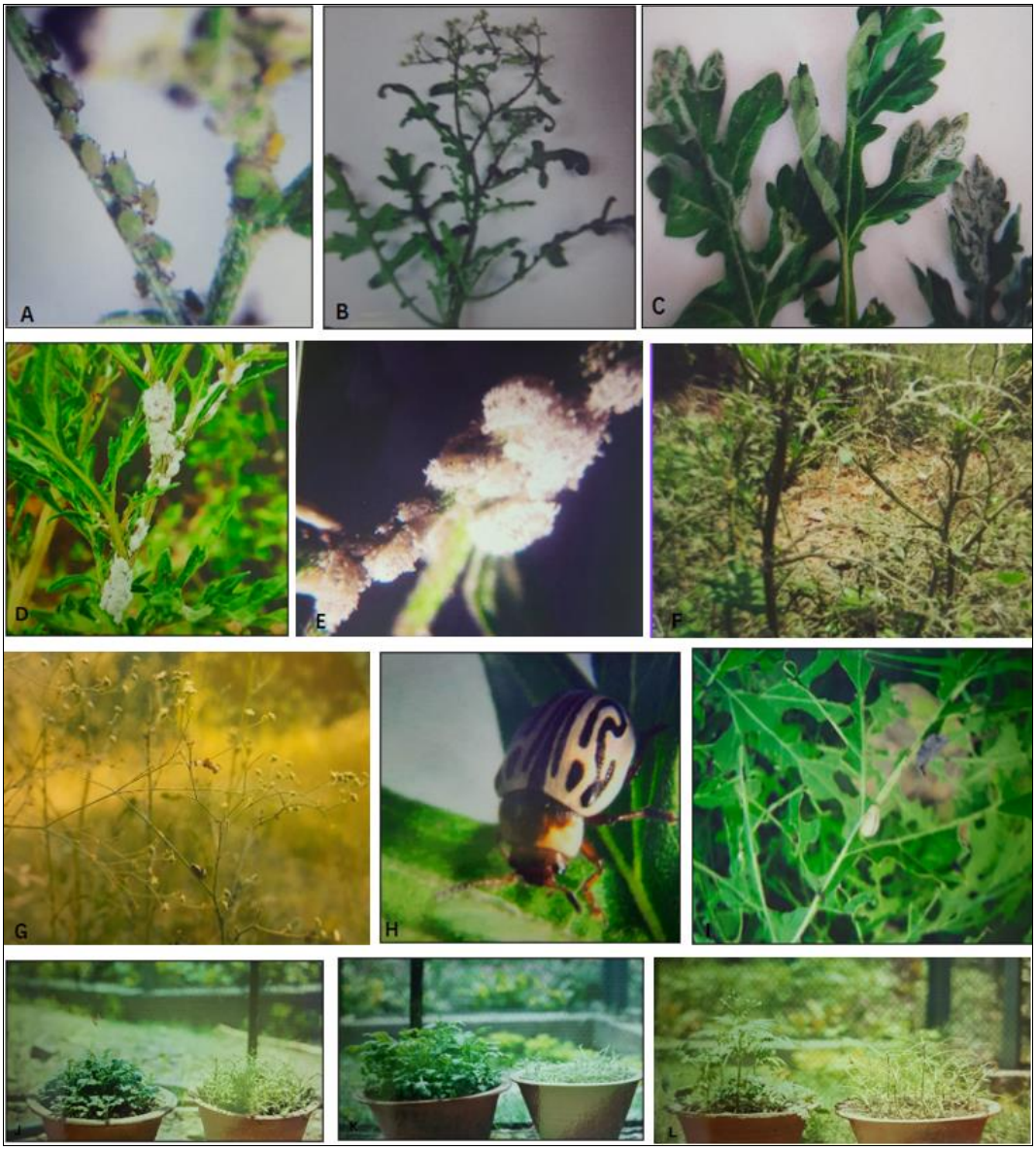

Fig 1: A: *Aphis gossypi* sucking the plant sap from Parthenium. **B**: Curling of leaf and distortion of flower bud due to *Aphis gossypi* infestation and viral infection. **C:** Leaf mine created by Leaf miner. **D&E:** Infestation of *Phenococcus sp*. on Parthenium Plant. **F&G:** Infestation of *Zygogramma bicolorata* on Parthenium plant. **H:** *Zygogramma bicolorata* **I :** Damaged parthenium leaf by larvae of *Z. bicolorata*. **J:** Impact on growth after 15 days of introduction of insects. **K:** Impact on growth after 30 days of introduction of insects **L:** Impact on flowering of plant.

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