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Integrated Biological Control of *Eichhornia crassipes* (Mart.) Solms at Different Growth Stages

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ABSTRACT

Classical biological control of water hyacinth using insect agent *Neochetina* spp. is constrained in many tropical watershed environments with interrupted host range due to seasonal water flow and complete drying of water during the hot summer months. Accordingly, the need for reinforcing the classical bio-control approach with sustainable short term measures has been realized and studies were undertaken to explore the possibility of integrating the insect agents and the plant product of dried leaf powder of *Coleus amboinicus/aromaticus* in controlling water hyacinth at different growth stages based on discriminate analysis using data on plant height, leaf area and fresh weight. Among the different inoculation loads of insect agents, releasing the insect agents @ three/ plant registered the highest reduction in fresh weight, chlorophyll and N, P, K content with small (I stage) and medium (II stage) growth stages of *Eichhornia crassipes*. At large growth stage (III stage) of *E. crassipes* none of the loads of insect agents was effective in reducing the fresh weight and chlorophyll content. Among the different concentrations of plant product, spraying at 25% registered the cent per cent reduction in fresh weight, chlorophyll and least N, P and K content at both the stages I and II of *E. crassipes*. No insect mortality was observed in any of the treatments compared.

INTRODUCTION

The invasive alien aquatic weed, water hyacinth, is among the top 10 weeds worldwide (Holm *et al.,* 1977) and is one of the most successful colonizers of the plant kingdom. Rapid growth, vegetative reproduction and ability to reinfest via the seed bank or flood-borne plants have resulted in excessive infestations in Africa, South Asia and the USA. In Tamil Nadu state in India, the Veeranum Lake and its distributaries form the major irrigation source that covers a large proportion of the rice tract of the state with a command area of 18,000 ha. This lake and its distributaries in recent years have been infested with *Eichhornia crassipes.* Its mechanical control is expensive and less efficient. Herbicides, even though effective, are not popular because of higher cost and pollution hazards. Biological control of water hyacinth was reported to be the preferred option in such situation (Andres, 1977) particularly when considering ecological safety. *Neochetina eichhorniae/bruchi* (Julien, 1987) has been reported as successful biological agents. It, however, requires a minimum of 3 to 5 years for insect population to increase to a density that can bring substantial decline in weed stand (Harley *et al.*, 1996). Further, the success of biological control depends on the availability of continued availability of weed host. In India, most of the water bodies are constrained with seasonal water flow and interrupted host range, as weed dries off in hot summer. Effective control of *E. crassipes* has been achieved by integrating insect bio-control agents *N. eichhorniae* and *N. bruchi* with other bio-control agent. An Indian medicinal herb *Coleus amboinicus*/ *aromaticus* showed remarkable allelopathic inhibition of *E. crassipes* when applied in water bodies @ 30 g/l, imparting complete weed mortality within 24 h. The plant product was active when absorbed through roots and spray of plant product over foliage. However, the plant product showed remarkable activity even at lower dose over cut leaves of the weed in a specific bio-assay (Kathiresan, 2000). This observation brought out the fact that, if introduced into weed system, the botanical herbicide could work effectively. Based on the above facts, the present study was taken up to explore the possibility of integrating the plant product *C. amboinicus/ aromaticus* with the insect bio-control agent *N. eichhorniae*/*bruchi* for effective control of water hyacinth at different growth stages.

MATERIALS AND METHODS

The experiments were conducted during 2003 at Department of Agronomy, Faculty of Agriculture, Annamalai University, Tamil Nadu, India to assess the response of different growth stages of *E. crassipes* to integrated technology. This study was taken up based on the results obtained in the preliminary experiments. The preliminary study revealed that releasing the insect agents first, followed by spraying the plant product *C. amboinicus/aromaticus* at different concentrations after 10 days proved effective in controlling the weed with a higher degree of inhibition in fresh weight and chlorophyll content of water hyacinth. Three different growth stages viz., (i) small (with the fresh weight of 50-60 g, leaf area of 400-420 cm^2 and plant height of 10-12 cm, (ii) medium (with the fresh weight of 100-120 g, leaf area of 500-520 cm^2 and plant height of 20-24 cm) and (iii) large sizes of *E. crassipes* (with the fresh weight of 260-270 g, leaf area of 700-720 cm^2 and plant height of 30-34 cm) as identified by Kannan and Kathiresan (1999), were selected for the experiments to assess the impact of integrating the insect agents and plant product for biological control. For each of the different growth stages of *E. crassipes* identified, three different inoculation loads of insect agents *Neochetina* spp. were tried viz., three insects/plant, two insects/plant and one insect/plant as main treatments and different concentrations of plant product *C. amboinicus/aromaticus* were tried viz., 25, 20, 15, 10 and 5% alongwith untreated control as sub treatments. The foliar spray of plant product was applied 10 days after releasing the insects (DAIR) in the respective treatments (corroborating with the results of the preliminary experiment). The experiment was planned in a split plot design with five replications, separately for each of the three different growth stages of *E. crassipes*. The observations recorded were percentage reduction in fresh weight and chlorophyll content at 10 days interval, insect migration and mortality rate at 1, 2, 3, 4, 7 and 14 days after spraying (DAS) and nitrogen (N), phosphorus (P) and potassium (K) content of water hyacinth. The reduction in fresh weight was recorded at 10 days interval (in comparison with initial fresh weight of plants in the same treatment). Chlorophyll content of *E. crassipes* was estimated at 10 days interval by extracting the leaf tissue using dimethyl sulphoxide (DMSO) (Hiscox and Israeltam, 1979). The mortality rate of insects was calculated based on the number of insects died per pot. In order to trace the migrational behaviour of insect agents, every treatment container was accompanied by another container with untreated *E. crassipes* plants (without plant product or insect) and both these containers were covered by fish net stretched over steel frames of 35 x 30 x 30 cm dimension. A white marking was made on the back of the insect prior to release into plants. The number of insects moved to the pot kept by the side (without insect release or any other control treatment originally) was counted at regular intervals and was considered as the insects migrated from the pots subjected to treatment. *E. crassipes* plants were collected from each treatment on 75 DAS; oven-dried at 65°±5°C for 24 h and were ground to fine powder. The powder was taken for analysis of nitrogen (Yoshida *et al.*, 1976), phosphorus and potassium content (Jackson, 1973). The data were statistically analyzed using the methods described (Panse and Sukhatme, 1978). After subjecting the data to analysis of variance, least significant difference was worked out at 5% probability level.

$$\text{Insect mortality rate (\%)} = \frac{\text{Total No. of insets released into the pot} - \text{No. of insects alive in the pot}}{\text{Total No. of insects released into the pot}} \times 100$$

RESULTS AND DISCUSSION

All the treatments registered significant influence on the fresh weight and chlorophyll content, insect migration rate and nutrient content of the weed. Among the different inoculation loads of insect agents, releasing the insect agents @ three/plant registered the highest reduction in fresh weight and chlorophyll content (Tables 1 and 2), least weed nutrient content (Table 4) and lowest insect migration rate (Table 3) with both small (I stage) and medium (II stage) growth stages of *E. crassipes*. The cent per cent reduction in chlorophyll content and fresh weight was recorded on 30 and 50 DAS, respectively, at stage I *E. crassipes* and the highest reduction in chlorophyll content (78.92%) and fresh weight (82.50% t) on 30 and 70 DAS, respectively, were recorded with stage II *E. crassipes*. The least fresh weight and chlorophyll content reduction and highest weed nutrient content were recorded with the insect agents @ one/plant. At stage III *E. crassipes* none of the loads of insect agent was effective in reducing the fresh weight and chlorophyll content.

Among the different concentrations of plant product, spraying at 25% registered the highest reduction in fresh weight and chlorophyll content and least weed nutrient content at both I and II stages of *E. crassipes*. The cent per cent reduction in chlorophyll content and fresh weight of the weed in smaller growth stage (I)

Table 1. Per cent reduction in fresh weight of *E. crassipes* to integrated approach at different growth stages

Treatment	I Stage					II stage						
	10 DAS	20 DAS	30 DAS	40 DAS	50 DAS	10 DAS	20 DAS	30 DAS	40 DAS	50 DAS	60 DAS	70 DAS
Main treatments												
One insect/plant	22.58	32.46	42.22	53.11	53.94	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Two insects/plant	39.23	51.67	64.14	78.15	81.74	30.15	36.92	42.61	52.73	59.81	61.35	62.35
Three insects/plant	46.14	60.59	81.39	92.33	100.00	36.83	45.95	54.74	66.74	71.01	79.17	82.50
LSD (P=0.05)	2.64	1.79	2.49	2.26	1.98	2.11	2.54	3.02	3.46	2.82	3.08	2.20
Sub-treatments												
25% plant product spray	56.77	71.82	92.48	100.00	100.00	35.32	42.87	50.33	64.14	66.66	66.66	66.66
20% plant product spray	48.32	63.18	91.61	100.00	100.00	30.88	36.93	44.20	59.42	66.66	66.66	66.66
15% plant product spray	35.48	50.77	69.35	82.33	82.75	20.92	25.33	30.67	36.00	40.67	50.33	51.33
10% plant product spray	32.62	44.95	59.19	76.61	82.33	18.00	24.35	28.82	33.47	37.41	43.36	49.69
5% plant product spray	21.60	29.80	37.82	45.28	53.48	14.56	18.88	21.16	24.02	26.33	28.33	29.01
Control (insect alone)	21.11	28.93	37.07	42.95	52.79	14.28	17..38	19.50	21.90	23.90	25.66	30.83
LSD (P=0.05)	2.91	2.26	3.08	2.93	2.22	2.69	2.83	3.46	3.82	3.06	3.42	2.62

Table 2. Per cent reduction in chlorophyll content of *E. crassipes* to integrated approach at different growth stages

Treatment	I Stage			II stage		
	10 DAS	20 DAS	30 DAS	10 DAS	20 DAS	30 DAS
Main treatments						
One insect/plant	28.81	44.58	54.66	0.00	0.00	0.00
Two insects/plant	49.09	70.69	79.62	38.76	52.51	58.05
Three insects/plant	59.77	83.96	100.00	47.49	69.16	78.92
LSD (P=0.05)	3.00	3.11	2.80	2.06	1.56	2.36
Sub-treatments						
25% plant product spray	72.38	94.45	100.00	43.25	66.66	66.66
20% plant product spray	65.83	92.85	100.00	38.86	59.65	66.66
15% plant product spray	49.41	75.63	82.00	29.53	40.06	48.27
10% plant product spray	38.55	59.71	81.61	25.99	37.35	47.55
5% plant product spray	24.95	38.55	52.68	17.00	21.93	24.14
Control (insect alone)	23.87	37.27	52.26	15.86	17.74	20.64
LSD (P=0.05)	3.28	3.38	2.96	2.46	1.92	2.60

Table 3. Insect migration rate by integrated bio-control involving the use of insect agents followed by foliar spray of plant product on *E. crassipes*

Treatment	I Stage						II stage					
	1 DAS	2 DAS	3 DAS	4 DAS	7 DAS	14 DAS	1 DAS	2 DAS	3 DAS	4 DAS	7 DAS	14 DAS
Main treatments												
25% plant product spray	22.22	31.11	46.66	55.55	75.55	100.00	22.22	27.77	41.11	53.33	74.44	86.67
20% plant product spray	22.22	31.11	42.22	47.78	75.55	100.00	22.22	27.77	41.11	50.00	74.44	86.67
15% plant product spray	20.00	25.55	35.55	47.78	70.00	86.67	13.33	22.22	33.55	42.22	58.89	76.67
10% plant product spray	20.00	22.22	33.33	35.55	61.11	86.67	10.00	13.33	30.00	30.00	51.11	65.55
5% plant product spray	7.78	7.78	27.78	30.00	32.22	47.78	4.44	7.78	24.44	30.00	35.55	47.78
Control (insect alone)	7.78	7.78	22.22	22.22	24.44	43.33	4.44	4.44	16.67	25.55	27.78	35.55
LSD (P=0.05)	2.02	2.71	3.26	3.60	4.96	3.20	2.00	2.42	3.71	3.81	4.52	2.46
Sub-treatments												
One insect/plant	13.33	13.33	23.33	30.01	40.00	66.67	6.67	10.00	15.00	30.00	33.33	46.66
Two insects/plant	16.66	21.66	35.00	41.66	58.33	81.66	11.67	20.00	38.33	40.00	56.66	71.67
Three insects/plant	20.00	24.44	41.11	47.66	67.77	82.22	19.99	23.33	40.00	45.55	71.11	81.11
LSD (P=0.05)	2.80	2.38	3.20	3.26	4.20	2.89	2.82	2.22	3.20	3.62	2.11	2.55

Table 4. N, P and K content (mg/g fresh weight) of *E. crassipes* to integrated approach at different growth stages

Treatment	I Stage			II stage		
	N	P	K	N	P	K
Main treatments						
One insect/plant	3.148	0.430	3.957	2.921	0.416	3.894
Two insects/plant	2.635	0.372	3.444	2.559	0.389	3.425
Three insects/plant	2.459	0.349	3.392	2.284	0.369	3.230
LSD (P=0.05)	0.148	0.016	0.181	0.123	0.019	0.183
Sub-treatments						
25% plant product spray	2.500	0.358	3.473	2.207	0.355	2.794
20% plant product spray	2.551	0.359	3.495	2.251	0.362	2.993
15% plant product spray	2.694	0.379	3.552	2.487	0.380	3.286
10% plant product spray	2.763	0.385	3.603	2.566	0.395	3.743
5% plant product spray	2.962	0.407	3.656	2.968	0.424	4.087
Control (insect alone)	3.015	0.415	3.810	3.048	0.432	4.196
LSD (P=0.05)	0.172	0.020	0.290	0.168	0.032	0.260

N–Nitrogen, P–Phosphorus, K–Potassium.

was recorded on 30 and 40 DAS, respectively. The highest reduction in chlorophyll content (66.66%) and fresh weight (66.66%) of the medium growth stage (II) weed were recorded on 30 and 70 DAS, respectively. The least fresh weight and chlorophyll content reduction and highest weed nutrient content were recorded with insect alone (control) at both stages I and II of *E. crassipes*. Young plants (I stage) of *E. crassipes* were very efficiently controlled by the integrated approach, succumbing to 25 and 20% plant product spray even after the least inoculation load of one insect/plant. If tried with an inoculation load of two insects/plant, the plant product spray concentration could even be reduced to 15%. Medium plants (II stage) of *E. crassipes* were also efficiently controlled by the integrated approach but only with the insect inoculation loads of two/plant and at plant product spray concentration of 25 and 20%. Large plants (III stage) of *E. crassipes* showed appreciable tolerance to the insect scraping and hence, the integrated approach failed to control such large and hardy plants at any of the inoculation loads and plant product spray concentrations tried. The variable response of different morphological stature of *E. crassipes* to the integrated approach is mainly because of its tolerance or deterrence to insect feeding and adequate amount of feeding scars produced by the insects on the weeds foliage that favoured absorption and translocation of the applied plant product into the weed. As the weed grows large, the biochemical constituents such as ascorbic acid and specific sugars that offer host specificity get altered. This altering biochemical nature deters the insect and repels them from feeding and making feeding scars. When the insects fail in making feeding scars, the plant product sprayed on the foliage couldn't penetrate through the cuticle and hence prove inefficient. The present observation is in line with the reports of Kannan and Kathiresan (1999) regarding the resistance of large plants of *E. crassipes* to *N. eichhorniae/bruchi*.

No insect mortality rate was observed in any of the treatments. However, highest insect migratory behaviour was observed with the most effective treatment of 25% coleus leaf extract spray (Table 3). The least insect migration was observed with spray of 5% leaf powder extract. The higher percentage of insect migration could be attributed to the degree of injury suffered by the weed as a result of combined action of both leaf powder extract and insects rather than to the direct repulsion of the insects by the leaf powder extract and surfactant. The fact that depletion of food reserve is the cause for the insect migration rather than deterrence by the plant product.

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