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Integration of Plant Product and Insect Agents for Control of Water Hyacinth (Eichhornia crassipes(Mart.) Solms)

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Abstract: Classical biological control of *Eichhornia crassipes* using insects agents *Neochetina* spp. is constrained in many tropical watershed environs with interrupted host range due to season 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 taken to explore the possibility of integrating the insect agents and the plant product of dried leaf power of *Coleus amboinicus/aromaticus* that was shown to be extremely allelopathic on water hyacinth. Two sequences of integration viz., application of the plant product to water body first followed by the release of the insect agents on to weed and another with releasing the insect first on the weed followed by spraying of the plant product on the weed were compared. The first sequence of plant product–insect agents failed to produce additive response as insects migrated to untreated healthy plants. However, second one with the sequence of insect agents – foliar spray of plant product showed additive or synergistic response with efficient control water hyacinth in a season. The integrated approach comprising the release of insect agents on the weed first followed by the foliar spray of 25 per cent plant product on 10 days after releasing the insects was exhibiting a higher degree of inhibition with cent per cent reduction in fresh weight and chlorophyll content on 25 days after releasing the insects, respectively. No insect mortality was observed with plant product spray. Application of plant product as foliar spray on *E. crassipes* without exposing to insect agents did not show any significant influence on fresh weight reduction.

Key words: *Eichhornia crassipes,* Integrated biological control, *Neochetina* sp., *Coleus amboinicus/aromaticus*

INTRODUCTION

Water hyacinth (*Eichhornia crassipes* (Mart.) Solms - Laubach: Pontederiaceae) is one of the most troublesome aquatic weeds all over the world. It was introduced in India first time in West Bengal in 1889 as an ornamental plant and by now it has been recorded from all types of water bodies like ponds, canal and drainage in all most of the cities, villages including major river systems Brahmabutra, Cauvery, Ganga etc, in India. Excessive infestations of the weed deleteriously affect water traffic, water quality, fishing potential, and infrastructure for pumping, hydro electricity generation, water use and biodiversity. Other damages include water loss due to evapotranspiration and an increased population of vectors of human diseases like malaria, encephalitis, schistosomiasis, filariasis, etc.[6]. Several methods have been used to control water hyacinth viz., mechanical, chemical and biological. Frequent mechanical removal of this weed is highly expensive, labour intensive, time consuming and is unsatisfactory as repeated weeding are needed. Chemical herbicides, even though effective, are not popular because of their high cost and pollution hazards. Options such as classical biological control using insects offer satisfactory control but over a longer period of time. Biological control requires a minimum of several years, usually 3 to 5 years, for insect population to increase to a density that could bring down the weed stand to a substantial decline[1]. Integrated control of *E.crassipes* has been achieved by integrating bio-control agents *Neochetina eichhorniae* and *N.bruchi* with other bio-control agents and plant pathogens. Based on the above facts, the present study is taken up to explore the possibility of integrating the insect bio-control agent *N.eichhorniae*/*bruchi* with the plant product *C.amboinicus/aromaticus* (that has been reported to be allelopathic on water hyacinth) for effective control of the weed.

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MATERIALS AND METHODS

Experiments were conducted at Department of Agronomy, Faculty of Agriculture, Annamalai University, and Tamilnadu, India. Two different sequences that are possible for integrating the bio-control tools were compared. The first sequence involves application of plant product in to the water body with the intention of weakening the weed plant and pre-disposing it for rapid destruction by the insect agents later. The second sequence involves releasing the insects first followed by spraying the plant product on the weed canopy with expectation that the absorption of plant product by the weed canopy could be assisted by the insect damage on the weed foliage. For studying the efficacy of the first sequence, water hyacinth plants were taken in plastic container of size 16 x 12x 7 cm and treated with dried leaf powder of *C. amboinicus/aromaticus* at varying doses viz., 5, 10, 15, 20 and 25 g l^{-1}, in two sets. In one sets of these plant product treated containers with water hyacinth, insect agents *(Neochetina eichhorniae/bruchi)* were released @ three plant^{-1} on the same day, where as the other set is kept as such. Another container with water hyacinth was released with the insect agents alone and one more was retained as an untreated control. These treatments were replicated three times under completely randomized design.

The second sequence of a possible integration of both bio-control tools was studied by releasing the insect agents @ three plant^{-1} on to the water hyacinth plants in plastic containers and spraying the plant product at varying concentration viz., 25, 20, 15, 10 and 5 per cent aqueous extracts 10 days after releasing the insects. These treatments were compared with treatment that accommodated a water hyacinth plant, released with insect alone without plant product sprays with three replication under completely randomized design. The observations recorded were percentage reduction in fresh weight and chlorophyll content at 10 days intervals, insect migration and mortality rate at 1,2,3,4,7 and 14 DAS (days after spraying) and nitrogen(N), phosphorus(P), potassium(K) content of water hyacinth. The reduction in fresh weight was recorded at 10 days intervals (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)[2]. The mortality rates of insects were 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 dimension 35 x 30 x 30 cm. A white marking was made on the back of the insect prior to release into plants. The numbers of insects moved to the pot kept by the side (without insect release or any other control treatment originally) were counted at regular intervals and were considered as the insects migrated from the pots subjected to treatment. Weed nutrient content of nitrogen, phosphorus and potassium were analyzed in the laboratory.

Insect mortality rate (%) =

$$\frac{\text{Total no. of insets released into the pot - No. of insects alive in the pot}}{\text{Total no. of insects released into the pot}} \times 100$$

RESULTS AND DISCUSSIONS

The approach or sequence of treating the water content first with the plant product *C. amboinicus/ aromaticus* followed by the release of insect agents on the weed failed to produce any positive interaction with additive bio-control impact. The lethality observed on the weed with a near full reduction in fresh weight and chlorophyll content of the weed on 10 and 5 DAT, respectively in the treatment comprising 25 g l^{-1} of plant product, could be solely attributed to the allelopathic injury suffered by the weed due to plant product. It was the reflection of more an independent allelopathic effect of the plant product on the weed rather than an enhanced activity due to combined or integrated mode of injury by both the insect agents and plant product in other treatment involving lesser doses of plant product viz., 20, 15 g l^{-1} as well. In these treatments weed lethality and biomass reduction to non-traceable levels were observed over a prolonged duration of exposure (Tables 1 and 2). The plant product interrupted membrane permeability and caused electrolyte leakage and root dysfunction, there by leading to weed lethality and biomass reduction[4]. Biochemical constituents in the plant product reported to be responsible for several of its insecticidal, microbicidal functions were α-humulene, carvacrol, thymol, α-pinene and a-terpene[7]. Lack of additive or synergistic interaction between the plant product and insect agents in controlling the weed particularly in this sequence of treating the water body first with plant product followed by release of insect agents is due to migration of insect agents from treated partially dying plants to untreated healthy plants as their preferred choice of feed is not available in plants whose physiology is struck by plant product, lacking electrolytes. This could be appreciated

Table 1: Impact of the integrated approach of treating water with the plant product followed by the release of insect agents on *E.crassipes*

Treatments	Percentage reduction in fresh weight of *E.crassipes*								
	5 DAT	10 DAT	15 DAT	20 DAT	25 DAT	30 DAT	35 DAT	40 DAT	45 DAT
Control	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)
5 g plant product	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)
10 g plant product	21.13 (13.00)	25.56 (18.61)	29.71 (24.56)	33.80 (30.95)	35.67 (34.00)	38.06 (38.00)	39.84 (41.05)	41.32 (43.60)	41.64 (44.15)
15 g plant product	35.19 (33.21)	50.93 (60.28)	56.16 (69.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)
20 g plant product	43.06 (46.62)	61.34 (77.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)
25 g plant product	52.82 (63.48)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)
Insects alone	16.97 (8.52)	25.84 (19.00)	28.88 (23.33)	33.83 (31.00)	35.67 (34.00)	42.71 (46.00)	45.80 (51.40)	62.03 (78.00)	90.00 (100.00)
5 g plant product + insects	17.73 (9.27)	27.97 (22.00)	29.31 (23.97)	35.26 (33.33)	37.45 (36.97)	44.46 (49.06)	50.45 (59.47)	67.21 (85.00)	90.00 (100.00)
10 g plant product + insects	27.88 (21.86)	36.32 (35.08)	42.88 (46.30)	54.92 (66.97)	59.93 (74.90)	70.59 (88.96)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)
15 g plant product + insects	36.71 (35.74)	51.98 (62.07)	56.79 (70.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)
20 g plant product + insects	43.59 (47.55)	62.64 (78.88)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)
25 g plant product + insects	52.99 (63.78)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)
SE_D	1.25	1.01	0.63	1.32	1.45	1.35	1.53	0.76	0.91
CD (p=0.05)	2.52	2.03	1.25	2.65	2.89	2.70	3.05	1.52	1.80

Figures in parentheses are original values before angular transformation
DAT-days after treatment

Table 2: Impact of the integrated approach of treating water with the plant product followed by the release of insect agents on *E.crassipes*

Treatments	Percentage reduction in chlorophyll content						
	5 DAT	10 DAT	15 DAT	20 DAT	25 DAT	30 DAT	35 DAT
Control	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)
5 g plant product	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)
10 g plant product	22.91 (15.16)	27.56 (21.41)	31.78 (27.74)	33.26 (30.08)	37.73 (37.46)	41.28 (43.51)	44.39 (49.15)
15 g plant product	53.13 (64.00)	68.86 (87.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)
20 g plant product	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)
25 g plant product	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)

Table 2: Continued.

Insects alone	21.35 (13.25)	29.97 (24.95)	36.52 (35.42)	52.43 (62.83)	61.09 (76.64)	67.21 (85.00)	90.00 (100.00)
5 g plant product + insects	28.55 (22.84)	33.72 (30.82)	39.54 (40.53)	55.47 (67.83)	64.53 (81.51)	70.54 (88.91)	90.00 (100.00)
10 g plant product + insects	31.18 (26.81)	36.98 (36.18)	44.04 (48.34)	56.76 (69.96)	66.42 (84.00)	90.00 (100.00)	90.00 (100.00)
15 g plant product + insects	53.94 (65.35)	71.41 (89.84)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)
20 g plant product + insects	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)
25 g plant product + insects	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)
SE_D	1.06	1.70	1.81	2.05	2.11	2.11	4.28
CD (p=0.05)	2.11	3.44	3.62	4.11	4.21	4.33	4.56

Figures in parentheses are original values before angular transformation
DAT-days after treatment

Table 3: Impact of the integrated approach of treating water with the plant product followed by the release of insect agents on E.crassipes

Treatments	Insect migration rate (%)						Nutrient content of *E. crassipes*(%)		
	1 DAT	2 DA t	3 DAT	4 DAT	7 DAT	14 DAT	N	P	K
Control	-	-	-	-	-	-	8.931 (2.421)	3.729 (0.423)	11.495 (3.972)
5 g plant product	-	-	-	-	-	-	8.332 (2.100)	3.662 (0.408)	11.263 (3.815)
10 g plant product	-	-	-	-	-	-	7.375 (1.648)	3.022 (0.278)	8.780 (2.330)
15 g plant product	-	-	-	-	-	-	7.058 (1.510)	2.871 (0.251)	8.368 (2.118)
20 g plant product	-	-	-	-	-	-	6.797 (1.401)	2.737 (0.228)	7.715 (1.802)
25 g plant product	-	-	-	-	-	-	6.597 (1.320)	2.569 (0.201)	7.678 (1.785)
Insects alone	0.01 (0.00)	21.41 (13.33)	21.41 (13.33)	25.56 (20.00)	26.56 (20.00)	31.09 (26.66)	7.142 (1.546)	3.181 (0.308)	10.107 (3.080)
5 g plant product + Insects	21.41 (13.33)	25.56 (20.00)	25.56 (20.00)	31.09 (26.66)	35.26 (33.33)	46.90 (53.33)	7.072 (1.156)	3.071 (0.287)	9.795 (2.894)
10 g plant product + Insects	25.56 (20.00)	25.56 (20.00)	31.09 (26.66)	31.09 (26.66)	35.26 (33.33)	50.77 (60.00)	6.945 (1.462)	2.917 (0.259)	8.755 (2.317)
15 g plant product + Insects	46.90 (53.33)	54.53 (66.66)	58.91 (73.33)	58.91 (73.33)	75.03 (93.33)	90.00 (100.00)	6.827 (1.413)	2.785 (0.236)	8.146 (2.008)
20 g plant product + Insects	58.91 (73.33)	63.43 (80.00)	75.03 (93.33)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	6.711 (1.366)	2.682 (0.219)	7.682 (1.787)
25 g plant product + Insects	68.38 (86.66)	75.03 (93.33)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	6.615 (1.327)	2.627 (0.210)	7.682 (1.787)
SE_D	3.72	2.70	2.38	1.74	2.20	2.04	0.125	0.023	0.210
CD (p=0.05)	7.45	5.41	4.77	3.48	4.40	4.08	0.250	0.046	0.415

Figures in parentheses are original values before angular transformation N- Nitrogen: P- Phosphorus: K- Potassium
DAT-days after treatment

Table 4: Impact of the integrated approach with the sequence of releasing the insect agents first followed by plant product as foliar spray on *E.crassipes*

Treatments	Percentage reduction in fresh weight				Percentage reduction in chlorophyll content		
	15 DAS	25 DAS	35 DAS	45 DAS	15 DAS	25 DAS	35 DAS
Control	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)
5% plant product spray	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)
10% plant product spray	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)
15% plant product spray	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)
20% plant product spray	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)
25% plant product spray	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)	0.01 (0.00)
Insects alone	31.37 (27.10)	46.91 (53.33)	54.11 (65.63)	90.00 (100.00)	41.83 (44.47)	59.32 (73.96)	90.00 (100.00)
Insects + 5% plant product spray	34.79 (32.57)	47.47 (54.30)	61.11 (76.66)	90.00 (100.00)	42.42 (45.50)	65.17 (82.36)	(82.36) (100.00)
Insects + 10% plant product spray	40.84 (42.77)	53.73 (65.00)	67.21 (85.00)	90.00 (100.00)	46.62 (52.84)	68.95 (87.10)	90.00 (100.00)
Insects + 15% plant product spray	42.13 (45.00)	56.37 (69.33)	90.00 (100.00)	90.00 (100.00)	53.33 (64.34)	73.57 (92.00)	90.00 (100.00)
Insects + 20% plant product spray	46.52 (52.66)	62.03 (78.00)	90.00 (78.00)	90.00 (100.00)	60.21 (75.31)	78.46 (96.00)	90.00 (100.00)
Insects + 25% plant product spray	54.74 (66.67)	90.00 (100.00)	90.00 (100.00)	90.00 (100.00)	68.04 (86.06)	90.00 (100.00)	90.00 (100.00)
SE_D	2.34	2.85	1.85	2.00	2.29	2.01	1.58
CD (p=0.05)	4.68	5.71	3.71	4.01	4.61	4.02	3.20

Figures in parentheses are original values before angular transformation
DAS- days after spraying

Table 5: Impact of the integrated approach with the sequence of releasing the insect agents first followed by plant product as foliar spray on *E.crassipes*

Treatments	I nsect migration rate (%)						Nutrient content of *E. crassipes*(%)		
	1 DAS	2 DAS	3 DAS	4 DAS	7 DAS	14 DAS	N (%)	P (%)	K (%)
Control	-	-	-	-	-	-	9.875 (2.941)	3.612 (0.397)	11.694 (4.108)
5 % plant product spray	-	-	-	-	-	-	9.657 (2.814)	3.622 (0.399)	11.530 (4.000)
10 % plant product spray	-	-	-	-	-	-	9.456 (2.699)	3.552 (0.384)	11.513 (3.984)
15 % plant product spray	-	-	-	-	-	-	9.542 (2.748)	3.477 (0.368)	11.883 (4.240)
20 % plant product spray	-	-	-	-	-	-	9.705 (2.842)	3.506 (0.374)	11.303 (3.842)
25 % plant product spray	-	-	-	-	-	-	9.570 (2.764)	3.571 (0.388)	11.210 (3.782)

Table 5: Continued.

Insects alone	21.41 (13.33)	21.41 (13.33)	26.56 (20.00)	26.50 (20.00)	31.09 (26.66)	31.09 (26.66)	6.872 (1.432)	3.135 (0.299)	10.244 (3.163)
Insects + 5 % plant product spray	21.41 (13.33)	21.41 (13.33)	26.56 (20.00)	26.56 (20.00)	31.09 (26.66)	35.26 (33.33)	6.771 (1.390)	3.049 (0.283)	9.978 (3.002)
Insects + 10 % plant product spray	21.41 (13.33)	26.56 (13.33)	26.56 (20.00)	31.09 (26.66)	39.23 (40.00)	46.90 (53.33)	6.597 (1.320)	2.906 (0.257)	2.867 (2.501)
Insects + 15 % plant product spray	26.56 (20.00)	26.56 (20.00)	31.09 (26.66)	35.26 (33.33)	46.90 (53.33)	75.03 (73.33)	6.552 (1.302)	2.883 (0.253)	8.722 (2.300)
Insects + 20 % plant product spray	26.56 (20.00)	26.56 (20.00)	31.09 (26.66)	35.26 (33.33)	50.76x (33.33)	90.00 (100.00)	6.473 (1.271)	2.731 (0.227)	8.073 (1.972)
Insects + 25 % plant product spray	26.56 (20.00)	26.56 (20.00)	31.09 (26.66)	39.23 (40.00)	54.53 (66.66)	90.00 (100.00)	6.349 (1.223)	2.688 (0.220)	7.732 (1.810)
SE_D	2.91	3.04	1.85	1.43	3.15	2.91	0.073	0.027	0.166
CD (p=0.05)	5.85	6.08	3.70	2.86	6.32	5.83	0.146	0.054	0.535

Figures in parentheses are original values before angular transformation DAS- days after spraying N- Nitrogen: P- Phosphorus: K- Potassium

from higher insect migration of 86.66 per cent recorded with the plant product @ 25 g l^{-1} + insect agents, on 1DAT itself (Table 3). However, no insect mortality rate was observed in any of the treatments. The least weed nutrient content of nitrogen, phosphorus and potassium were recorded at 25 g l^{-1}.Untreated control recorded the highest weed nutrient contents.

However, the approach or sequence of releasing the insect agents first on the weed and spraying the plant product on the weed canopy later was observed to impart an additive or synergistic impact in controlling the weed. The integrated approach comprising the release of insect agents on the weed first followed by foliar spray of 25 per cent plant product was exhibiting a higher degree of inhibition with cent per cent reduction in fresh weight and chlorophyll content on 25 days after releasing the insects (Table 4). The insect by virtue of their feeding behaviour, imparted leaf scrapping, wherein cuticular lining was removed exposing the inner soft parenchymatous tissue beneath. These scrapings enabled better absorption of the allelochemicals in the plant product sprayed over the foliage and thus the insects in addition to partially damaging the weed vigour and physical stature also served as vehicle or penetrant helping absorption and translocation of the plant product. Further, the plant product also did not deter the insects either by antifeeding or repulsive mode as seen from the least 20 per cent insect migration in treatment that included the plant product spray at the higher concentration tried (Table 5). However, no insect mortality was observed in any of the treatments. The least weed nutrient contents were recorded with the releasing of the insect agents and followed by 25 per cent foliar spray of plant product. Longevity of the weeds in the system in treatments that exerted a slow paced control *viz*., plant product spray with lesser concentrations after releasing the insects, contributed for increased nutrient uptake by the weeds in such treatments. Application of plant product as foliar spray on *E. crassipes* without exposing to insect agents did not show any significant influence on reduction in fresh weight and chlorophyll content and nutrient content of the weed. This is because the plant product could not get through the citicular barrier of the weed host[3]. No insect mortality was observed in any of the treatments. This is probably due to the fact that many of the active principles in the plant product are only allelopathic and not allelomediatory in terms of toxicity to adult insects. This is in line with the findings of[5].

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