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Host preference and potency of *Altica cyanea* as a bio-control agent of major rice field weeds, *Ludwigia* spp.

Nayan Roy
Ecology Research Unit, Department of Zoology, M.U.C. Women's College, Burdwan, West Bengal, India

Abstract
The water primrose, *Ludwigia* spp. (*L. adscendens, L. prostate, L. hyssopifolia*) (Onagraceae) are wide spread decumbent weeds commonly found in rice field and wetland ecosystem abundantly throughout India and elsewhere. They are highly invasive and commonest form of weed species in the Indian rice field ecosystems including West Bengal. They induce nutrient partitioning with the rice plants and thus reduce their production along with short and long term allelopathy on the other subsequent economic crops cultivated after rice. Usually, herbivores are used as bio-control agent to regulate weeds without interfering the cropping system. It is well known that chemical signals and nutritional constituents play an important role in the interactions between plants and phytophagous insects. The weeds attract the flae beetle, *Altica cyanea* Weber (Coleoptera: Chrysomelidae) for defoliation by means of chemical signals. The life history and host-preference of the flea beetles were studied to determine its potential value as a biological control agent for one of the major rice field weed, *L. adscendens* over the other two species of *Ludwigia*. The phytochemical constituents including nutritional and some anti-nutritional factors were correlated with such type of preference and survivability of this bio-control agent. Thus the study support the potency of the flae beetle, *A. cyanea*, as a bio-control agent of the major rice field weed, *L. adscendens* over the other two *Ludwigia* spp. (*L. prostate*, *L. hyssopifolia*) and they can be used for better production of rice as well as other subsequent crops cultivated after rice.

Keywords: *Ludwigia adscendens, L. prostate, L. hyssopifolia, Altica cyanea*, bio-control agent, allelopathy

1. Introduction

Ludwigia spp. (Myrtales: Onagraceae) is a troublesome weed growing abundantly in Indian rice fields and elsewhere [1-6]. Generally *Ludwigia* spp. occurs in moist places while a few is predominantly aquatic, ranging from annual herbs to large shrubs [3-5]. It is generally common across different districts of West Bengal and also widespread over Bihar and Assam of India [1, 3, 4]. They compete with rice plants for nourishment by nutrient partitioning and disturb crop production [1-4]. This weed starts their life cycle with the germination of rice and completes many generations up to rice harvesting [2-4]. At the same time, they also inhibit germination and seedling growth of rice due to allelochemicals released by leaching of the weed parts, decomposing weed residues [7]. They also showed long and short term allelopathic effects on different subsequent crops cultivated after rice by deposition of allelochemicals in the rice fields [3, 4].

Today, the control of weeds through herbivorous bio-control agents without interfering the cropping system is a most effective eco-friendly approach rather than exception. A number of coleopteran chrysomelid insects have been found in association with aquatic weeds in areas of the native range and can serve the purpose of regulating weeds [2, 8, 9]. The chrysomelid, *Altica foeveicollis*, is very effective in the natural control of *Ludwigia* spp. in Thailand [2]. The search for suitable biological control agents for marshy herbaceous weeds common in rice fields has been conducted by many researchers from the oriental and southeast Asian countries [1, 6, 9-13]. The beetle, *Altica cyanea* (Weber) (Coleoptera: Chrysomelidae) also attracted in large numbers for defoliation and feeding on *L. adscendens* [1, 9, 13]. The larvae and adults of this beetles caused considerable damage to different *Ludwigia* spp., *Trapa natans*, *T. bisponsis*, *Rotala indica*, etc. but no feeding occurred on rice [1, 8]. They are multivoltine in nature and completed a generation within 80–84 days [1, 8]. One of the predatory species, *Zicrona coerulea* L. [Hemiptera: Pentatomidae] along with other natural predators preyed upon *A. cyanea* larvae to reduce their natural population [1]. The beetles may also decline by the usual light traps or bated traps present in the field to control the other insect pests.

But, still there is no report regarding the preferential potency of *A. cyanea* as a bio-control agent against the selected three *Ludwigia* spp. (*L. adscendens, L. prostate, L. hyssopifolia*). It is well known that, host-preference by herbivorous insects depends on chemical cues including nutritional requirements of insects and to some extent on secondary metabolites of their host- plants [14-27]. In this study, the host-preference of the flea beetle, *A. cyanea* were studied to determine its potential value as a biological control agent against the three *Ludwigia* spp. (*L. adscendens, L. prostate, L. hyssopifolia*) for their ecologically sustainable management in near future.

2. Materials and Methods

2.1. Plant Materials

Three types of Fresh *Ludwigia* spp. leaves were collected randomly growing in rice fields near Chinsurah Rice Research Center adjacent to the district Hooghly (22°53´N, 88°23´E),West Bengal, India. Leaves were initially rinsed with distilled water and dried by paper toweling for phytochemical analysis.

2.2 Insect Rearing

The insects used in this study were collected by light trap from the weed growing in a rice field near Chinsurah Rice Research Center. The insects were maintained in 1 l glass jars, containing three types of weed leaves, and covered with fine-mesh nylon nets at 27±1°C temperature, 65±10% relative humidity, and a 12L:12D photoperiod in a BOD incubator as described by Roy *et al*., and Roy [9, 13]. The F2 *A. cyanea* adults were kept for oviposition separately in different sterilized glass jars. Fresh leaves were given daily by replacing the previous one until eggs were laid by the test insects, and the eggs with each host-plant leaves were placed in new sterilized glass jars separately. To study the duration of larval and post larval development, the eggs were separated and reared separately in sterilized glass jars containing 20 larvae on each kind of host-plant, and observations were noted during their respective development with five replicates for each host plants.

2.3 Biochemical Analysis

The freshly harvested leaves of the three host plants (*L. adscendens, L. prostate, L. hyssopifolia*) were dipped in different solvents for extraction of different primary and secondary biochemicals. The variability in nutritional quality of three host-plants was estimated by subjecting the fresh undamaged leaves to various biochemical analyses, such as total carbohydrates [28] total proteins [29], total lipids [30], total amino acids [31], total phenols [32] and moisture [22]. Determination of each biochemical analysis was repeated for five times and expressed in mg/g or percent dry weight basis.

2.4 Statistical Analysis

All the data of biochemical analyses of the three host plants as well as the data on growth duration (i.e. larval and post larval duration and adult longevity),survivability and fecundityof *A. cyanea* reared on three host-plants were subjected to one way analysis of variance (ANOVA) to compare the effects of the diet regimes as described by Roy [9, 24-27] as well as by Roy and Barik [22, 23]. Means associated with all the data for each variable were separated using Tukey's test ("honestly significant difference test") when significant values were obtained [33, 34].

3. Results

The host preference of the flea beetle, *A. cyanea*, was studied to determine its potential as a biological control agent for the three *Ludwigia* spp. (*L. adscendens*, *L. prostate*, *L. hyssopifolia*). The biochemical analyses of the three host-plants are presented in Figure 1. Total carbohydrates, proteins, lipids and moisture content was varied significantly among the three host-plants, which can be arranged in order of *L. adscendens* > *L. prostate* > *L. hyssopifolia* ($P < 0.0001$). Total amino acid content was highest and significantly differed ($P < 0.001$) in *L. adscendens* than the other two host plants. Phenol concentration was also differed significantly among the three host-plants ($P < 0.0001$) and can be arranged in order of *L. adscendens* > *L. prostate* > *L. hyssopifolia*. The females laid yellowish eggs in masses on the abaxial surface of the weed leaves and larvae passed through three distinct instars along with pre-pupal and pupal stages. The developmental duration (i.e., total larval, post larval duration and adult longevity) of the different stages of *A. cyanea* on the three species of *Ludwigia* was varied significantly ($P < 0.001$) and they could be arranged as *L. adscendens* < *L. prostate* < *L. hyssopifolia* whereas, the adult longevity was in the reverse order i.e., *L. adscendens* > *L. prostate* > *L. hyssopifolia* (Figure 2). The fecundity was always higher in *L. adscendens* and significantly differed ($p < 0.001$) than the other two host plants (*L. prostate* and *L. hyssopifolia*) (Figure 3). The hatchability percent, larval survivability and adult emergence were significantly differed ($p < 0.001$) among the three selected host plants (Figure 4). The beetle showed lower hatchability, larval survivability and adult emergence when they were fed on *L. hyssopifolia* and *L. prostate* instead of *L. adscendens* during the study (Figure 4).

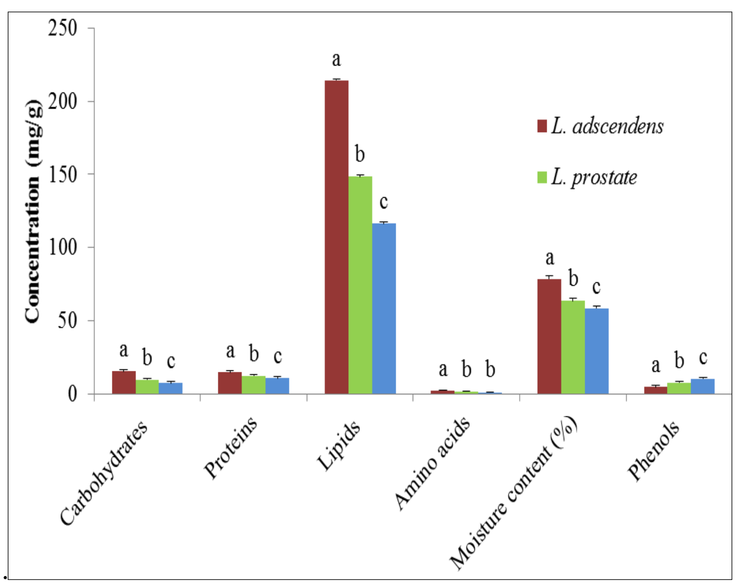

Fig 1: Phytochemical variations of the three *Ludwigia* spp. Different letters over the bars indicate that the means (Mean ± SE of 5 observations) are significantly different ($P < 0.05$), while comparing one type of plants with the other within the column.

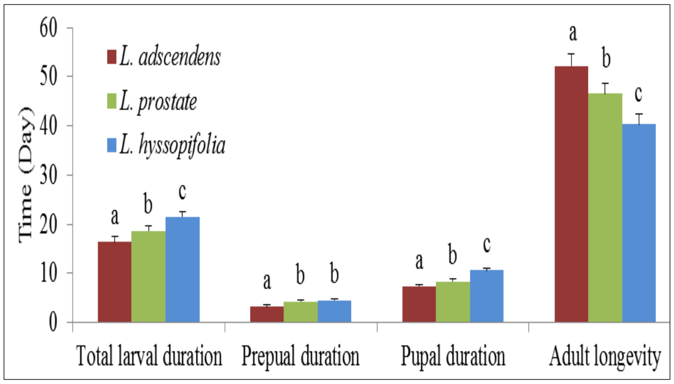

Fig 2: Developmental duration of the different stages of *A.cyanea* on the three species of *Ludwigia.* Different letters over the bars indicate that the means (Mean ± SE of 5 observations) are significantly different (*P* < 0.05), while comparing one type of plants with the other within the column.

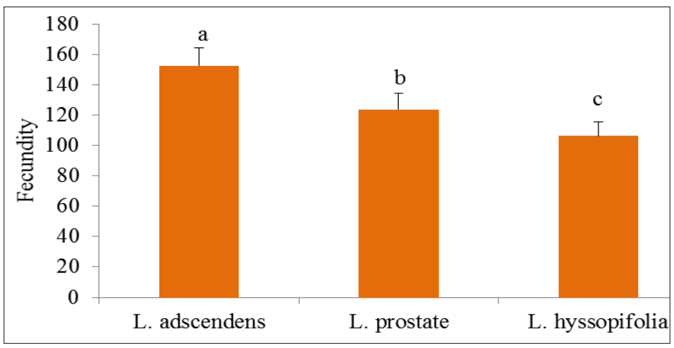

Fig 3: Fecundity of *A.cyanea* on the three species of *Ludwigia.* Different letters over the bars indicate that the means (Mean ± SE of 5 observations) are significantly different (*P* < 0.05), while comparing one type of plants with the other within the column.

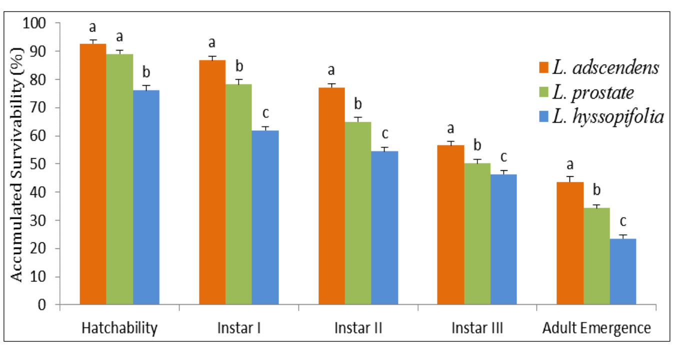

Fig 4: Accumulated survivability (%) of *A. cyanea* on the three species of *Ludwigia.* Different letters over the bars indicate that the means (Mean ± SE of 5 observations) are significantly different (*P* < 0.05), while comparing one type of plants with the other within the column.

4. Discussion

In this modern era with increasing human population there is a need to increase rice production per unit of land through economically and environmentally sustainable strategies. India is the second largest producer of rice in the world. Rice consumes almost 50 per cent of irrigation water and the water crisis is the greater threat to rice cultivation in India (Hosmani *et al*. 2010) [35]. Another threat is the various types of weeds in the rice fields throughout the world including tropical and subtropical Asia (*Ludwigia* spp., *Echinochloa* spp. *Paniculum* spp. *Cyperus* spp. etc.) grown in and around paddy fields [3, 36, 37]. Control strategies in current use against the weeds are largely based on chemical herbicides but intensive use creates an ecological imbalance through destruction of non-target beneficial insects, and accumulation of toxic residues in the environment [38-44]. The role of host-plant is an important factor in regulating herbivorous insect populations as the concentrations and proportion of nutrients differ greatly among species [21, 45]. The chemical signals and nutritional constituents play an important role in the interactions between plants and phytophagous insects [13, 16, 22-24, 46-48]. Nutritional requirements for insect growth and reproduction depend on the ability of the insect to ingest, assimilate and convert food into body tissue [15, 18]. The growth duration such as, developmental time, longevity, fecundity and survival of insectsshowed significant differences among the three host-plants tested with respect to their food quality [14, 20, 23]. Fecundity of herbivorous insects also depends on host-plant quality, which influences growth rates, developmental time and survivability of herbivores [20]. Clear variation was observed in food consumption and development of *A. cyanea* when fed with these three host-plants. The possible explanation for variation in food consumption and development of this insect is due to significant differences in carbohydrates, proteins, lipids, amino acids and moisture content in the respective host plants. This study indicated highergrowth rate, lower developmental time, higher adult longevity, survivability and fecundity of *A. cyanea* when fed with *L. adscendens*, which may be due to the better nutritional quality relative to the other two host plants. Phenols determine the suitability of the substrate for exploitation by the herbivores and thus govern host preferences and acceptability [21, 45]. Further, an increase in phenol content indicates reduction in adult longevity and fecundity, and retardation of larval growth [45]. Higher phenol content was observed in *L. hyssopifolia* followed by *L. prostate*, which probably indicates greater resistance in this plant against herbivory. Further, greater content of phenol in *L. hyssopifolia* and *L. prostate* probably caused reduction in fecundity and retardation of larval growth of *A. cyanea* when compared to the host-plant *L. adscendens* used in this study. In this study, higher levels of carbohydrates, proteins, lipids including amino acids, and lower levels of phenol in *L. adscendens* might be the possible explanation for relatively fastest larval development, higher fecundity and survivability when the insects were fed with this host plant relative to the other two host species. Thus in this case, *A. cyanea* show a preference towards *L. adscendens* over *L. prostate* and *L. hyssopifolia* due to higher nutritional quality relative to secondary metabolites. These suggest that, *A. cyanea* may be a promising biological control agent for *L. adscendens* growing in rice-paddies rather than the other two host species.

5. Conclusion

In the modern industrial agricultural system long persistent broad spectrum herbicides are still using indiscriminately against the weeds in nature to increase agricultural productivity in order to ensure food security. This injudicious application of herbicides as well as pesticides obviously leads to the destruction of ecological biodiversity including beneficial natural enemies, essential pollinators and foragers. This actually hampers the sustainability and normal functioning of the food chains by toxic effect and biomagnifications through trophic interactions in our ecosystem. In this study, host-preference and population growth of the flea beetles were studied to determine its potential value as a biological control agent for ecological management of the beetle against the weed species. The host preference and population survivality of the beetle, *A. cyanea*, on *L. adscendens* has showed superiority relative to the other (*L. prostate* and *L. hyssopifolia*) host plants. The phytochemical constituents were correlated with such type of preference and survivability of this beetle. Thus, by knowing such variations in their host susceptibility, one can make ecological management by time based application of the beetle in the field rather than toxic herbicides against the notorious weed species. It will be the most important biointensive tool towards the judicious use of different control measures mainly environmentally benign chemical herbicides for ecofriendly management of the weed species in our agro ecosystem for better cultivation of rice well as other subsequent crops cultivated rotationally. There may be few limitations in the methodical scientific study but this particular study somehow has triple- E (Environmental, Ecological and Economical) sustainability for any kind of weed management by any biocontrol agent in near future.

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