

UGC sponsored Major Research Project

on

“Relevance of Pollinating Insects in propagation of flowering plants in Calcutta and its surrounding areas, including college campus in Park Circus. : Attempts at DNA typing of pollinators and Biochemical characterization of their hosts”

**UGC Reference No. & Date F 41-56/2012 (SR), 13 July, 2012**

**Tenure: July 2012 to December 2015 (including 6 months extension)**

Principal Investigator:

Dr. Anuradha Chaudhuri, (Retd. Associate Professor), Department of Zoology

Lady Brabourne College, Kolkata, 700 054

#### **SUMMARY:**

Pollination is an ecosystem function that is vital to agriculture and to the maintenance of biodiversity. Commercial production of most crops, are accomplished through pollinators.

Important pollinators such as bees, butterflies and beetles have declined due to shortage of suitable food plants and nesting sites. Pollination systems are under increasing threat mainly from anthropogenic sources, including fragmentation and alteration of habitat, changes in land use pattern, modern agri practices, move towards monocultures, use of chemicals such as pesticides and invasions of alien species. Butterflies apart from being important pollinators play a big role as indicators of a healthy environment.

Decline of biodiversity as a result of urbanization and anthropogenic actions is now well documented. Much of our native flora and fauna are disappearing due to unplanned developmental activities in urban areas. Large scale construction, destruction of greens have contributed to the decline. There is urgent need to restore our lost habitats.

Role of butterfly gardens for *in situ* conservation and promoting environmental education is well recognized. As a business-cum-educational enterprise, butterfly gardens and butterfly houses are becoming increasingly popular in many countries including India. To some extent it can offset the damages done by disturbance of habitat.

The present project was aimed at conservation and restoration of lost habitat of pollinators specially butterflies in an urban scenario. An unmanaged butterfly garden within our college campus (Lady Brabourne College, Kolkata) had many species of butterflies. Developmental activities caused a sharp decline in their population causing us to relocate our garden in another area of college campus. Survey of butterflies found in the locality was performed prior to

construction activity. A butterfly garden was created in an alternative site in the campus area through landscaping. Based on already available data on host plant preferences of the local butterflies, plants were planted. Prior to construction activity, this garden was frequented by about 34 species of butterflies. The disturbed habitat saw a sharp decline in sightings. After the restoration of the garden, there was slow increase in sightings over a study period of 3 years. 39 species were recorded in the garden including nine species which were not recorded previously but were sighted in the new location for the first time and 3 introduced species. 8 species completing their life cycles. Presently the garden is flourishing but problems of increase in natural enemies as birds need to be addressed.

Considering that habitat loss is a primary driver of species extinctions, detailed habitat assessments are among the most important first steps guiding conservation efforts for imperiled species.

To know how the butterflies behave in the habitat of the newly established garden, behavioral allocation of different activities in different species of butterflies was done over a period of 5 months. Various activities such as basking, courtship behavior, chasing, mating etc was studied for the different species found in the garden. Butterflies are seasonal in their occurrence. The months when adults are active are called “flight periods”. Seasonality depends on availability of the larval host plants, availability of the plant in correct growth stage, suitable temperature and humidity. Their flight periods during different seasons in natural habitat were studied. It helped in planting the suitable plants in succession.

Some flowering plants such as *Tabernemontana* (Tagar), *Ravenia* (Laboni) were already growing at the study site. Several nectar plants were freshly planted which were to flower all through the year.

Some species of butterflies not seen before but common in surrounding areas were introduced in the relocated garden by captive breeding.

Common rose butterfly found abundantly in outskirts had not been sighted before the establishment of the garden. Plants of Aristolochiaceae family, *Aristolochia tagada*, *A. bracteolata*, *A. indica* (Rudrajota or Isher mul in Bengali) are larval host plant of Common Rose. The caterpillars, and hence the butterflies are protected because of the pungent smelling aristolochic acid found in their host plants. *A. indica* were planted to attract the Common Rose. Males of Blue tiger and other Tiger species need an alkaloid source to maintain their stock of mate-attracting chemicals or “sex pheromones”. These sources are certain plants as *Crotalaria*, *Heliotropium*, *Ageratum* which contain the alkaloids (pyrrolizidine) in a relatively high amount. The Blue tiger, along with Crows and other tigers swarm over damaged parts of these plants from where the sap oozes out. Plants belonging to these species and larval host plant like *Wattakaka* (*Dregea*) *volubilis* (Blue Tiger), *Oleum* (common Crow) were planted.

Passion flower ( wild variety) was planted for Tawny Coster. Adults and larvae of butterfly species like Tawny Coster, Blue Tiger and Common Rose were collected from different areas including established butterfly garden at Science City and propagated through captive breeding. Nectar plants and host plants of the respective butterflies were kept within the enclosed area. Adults mated, laid eggs and progeny after emergence were released in the garden. Butterflies were reared in the laboratory and life cycles studied.

Information on nectar preferences of butterflies in natural conditions is scanty.

Nectaring activity was studied in the butterflies in the garden. Butterflies as nectar foragers, largely feed on flowers to get the nutrition for their survival and reproduction. Study was carried out to understand the feeding preferences, the nectar sources visited by butterflies in the Lady Brabourne College campus. The main objective was to help create a better habitat conducive to breeding and survival of pollinators in an urban surrounding such as our campus and its surroundings.

Out of the 29 species studied, 23 species preferred weeds.

This study indicates preference of weeds as nectar source by the different families of butterflies. Out of 25 plant species, 14 species which are commonly known as weeds are preferred over the cultivated species.

Although the butterflies, individually visits nectar on a diversity of flowers, but as a group they showed similar patterns in their use and preference of nectar flower. Among the five different families of butterflies, only Papilionidae shows the maximum preference for the cultivated flowering plants. The other families of butterflies do nectaring in cultivated plants, but they clearly show a definite preference towards the so called neglected plant – ‘Weeds’ like *Lantana*, *Mikania*, *Tridax*, *Vernonia* etc

Study is preliminary and shows certain trends commensurate with known behavior. But some behavioral activities not seen by no means indicate lack of it but needs observations over a longer time and in different seasons.

Phytophagous insects respond to plant allelochemicals by production of enzymes for detoxification. Approximately 20 different enzyme systems are involved in detoxification of various allelochemicals. Of these, esterases, and glutathione transferases are most generally important

The object of studying esterase enzyme activity in the some species of butterfly in the study area was to find the least toxic of food plant among the different host plants available to them. Caterpillars fed on host plant was used for enzyme activity staining. Enzyme activity could be visualized. Conditions for the assay was thus standardized. Further work on other detoxifying enzymes need to be done to find out suitable host plants.

Due to existence of closely related species, morphological attributes that change as a function of environment and the prevalence of biotypes and their sibling species, the identification of butterflies on the basis of morphology has become problematic. One of the goals of the project was to explore the possibilities of using molecular techniques to find out genetic relatedness or distance between the species of butterflies found locally.

Preliminary studies were done on feasibility of using Random amplified polymorphic DNA-polymerase chain reaction, (RAPD-PCR) with locally found butterflies (specially mimicking species) using primers reportedly used for Indian butterflies for molecular taxonomy studies

We are specially interested to find the relatedness of polymorphic species. *Papilio polytes* also known as “Common Mormon” is a mimic swallowtail butterfly having a wide range of distribution in the world including India. Three forms of polymorphic females : *Stichius*, *Cyrus* and *Romulus* are found in an area depending on the presence of the model unpalatable species they mimick.

Out of the four primers used, two were found to be suitable for RAPD-PCR. Female and male specific bands could be found. The method of DNA isolation, gel electrophoresis and RAPD-PCR have been standardized. Future studies can be conducted in this area.

This is a short time study which has focussed on Status survey of pollinators before and after relocation of their habitat. Specific behaviour pattern like feeding, foraging, basking, flight, etc as also nectaring preferences of local butterflies for local plants was studied. The study had several aspects so as to have a holistic view of the relevance of pollinators in an urban scenario. Preliminary molecular and biochemical studies were performed to standardize the methodology. Methods relevant to the enzyme activity staining, DNA isolation, electrophoresis, RAPD-PCR have been standardized for further studies in this field.

Finally, many butterflies have been seen in the relocated garden, some choosing it as their home for completing their life cycle. Prior to construction activity, this garden was frequented by about 35 species of butterflies. The disturbed habitat saw a sharp decline in sightings. After the restoration of the garden, there was slow increase in sightings over the study period . Overall, 46 species were recorded in the garden including 9 not seen before, with 8 species completing their life cycles. 3 species were introduced which are now seen regularly. They are completing their life cycles in this habitat. To this end, it can be said that the butterfly garden established in an urban surrounding has to some extent restored the habitat and offset effects of anthropogenic disturbance. This study reiterates and emphasizes the importance of saving some land in a garden for the so-called weeds.

Proper management and maintenance of the relocated garden and floral quality and quantity within and surrounding areas is the prime requirement for its healthy existence, so as to accommodate diverse flora and fauna throughout the year. During urban garden planning, emphasis is laid on ornamental plants but to save the pollinators it is of utmost importance that space be allotted for the so called weeds in messy growth.

It is hoped that a sound knowledge of plant preferences will help in planning and restoring the habitat of pollinators and will be useful in conservation.

Space is a limiting factor. Host plants of butterflies whose adults were sighted but who did not find enough plants to lay eggs need to be planted. If more area in surrounding areas can be made available, it is hoped more butterflies will choose this relocated garden as their home. Care has to be taken to decrease effects of street lighting, predation by birds and anthropogenic disturbance.