

## PROBLEM C- CHEMICAL ESPIONAGE

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From the three problems given to us as a part of SCUDEM 2019, our team chose **Problem-C “Chemical Espionage”**. In this problem, we studied about the large cabbage white butterfly *Pieris brassicae*, its mating process, problems it faces and interference of wasps (both having innate and learned behavior) larvae with the butterfly eggs.

### Introduction

Many experiments have been conducted to study the mating process of butterflies and what factors affect this process. It has been seen that the life span of adult butterflies for virgin and mated are different. Virgin female and male butterfly live for up to 18 and 10 days respectively and mated males for 3-5 days and females for 5-7 days. This indicates that adult butterfly only live long enough to produce next generation.

Mating in adult butterfly occurs one or two days after they have emerged from pupae and are fully mature. Mating period lasts for about three hours. Prior to mating, male patrols for female butterfly. Males are attracted to female butterflies based on their wing color. During mating, males secrete anti-aphrodisiac to prevent other males from trying to mate with the female butterfly and the female can focus on finding a suitable location to lay eggs. One of the problems or factors with population growth is that anti-aphrodisiac can be intercepted by parasitoids like trichogramma wasps and they inject their own eggs which feed on butterfly eggs which eventually kills them.

Female deposits entire batch of eggs on same area of the leaf of host plant it selects. Eggs are mainly laid on underside of leaf to protect them from predators and a batch contains about 50 eggs. Large white eggs appear as pale yellow color and slowly turn into black. After the eggs hatch and larvae is produced, they feed on the host plant and highly damage it.

Various population growth model exist which are used to study population growth rate of *Pieris brassicae* butterfly and the effect of environment and parasitoids on it. Within a year, during summer time two generations of butterfly are produced and if the summer is long enough up to three generations can be produced.

## Basic Model

Instantaneous rate of increase of population (r):-  $r = \ln R_0 / T$

( $R_0$  is carrying capacity and T is mean period between birth of parent and that of offspring)

$R_0 = \sum I_x m_x$  i.e. (Survival rate from birth to age x)  $\times$  (total no of offspring produced at age x)

$T = \sum [ I_x m_x x ] / \sum [ I_x m_x ]$  (x is age) Doubling time :-  $DT = \ln 2 / r$

Finite rate of increase (lambda)  $\lambda = \exp (r)$

This is population multiplication in unit time. From this result we see that according to this model the population keeps on increasing. This does not inculcate any factors of death like natural causes, environmental conditions or attack of parasitoids and thus gives incorrect results and needs modification. For this we included the effects through some tests and probabilistic analysis as mentioned below.

For this experiment, a two chamber olfactometer is used to observe the wasps' response to different butterfly odours. As the aim of this experiment is how the trichogramma wasps' intercept butterfly eggs, we use three types of butterflies: males, virgin females and females which were synthetically treated with Benzyl Cyanide and anti-aphrodisiac. Also, to reduce the butterflies' mobility, they are refrigerated (4°C) for 10 minutes.

**Wilcoxon's matched-pairs-signed-ranks test** is used to calculate the residence time of a wasp in the olfactometer. The wasp is introduced in between the butterflies and observed until it climbs onto one of the 2 butterflies at present in the chamber. **GLM** (General Linear Model) is used to differentiate the residence time for naïve and H+O (Hitchhiking and Oviposition) trichogramma wasps. When a wasp does not mount a butterfly within 300 s, it is recorded as a non-responder. After each third wasp, the butterflies were replaced. For each combination, 40 responding wasps were investigated.

Dependent variable: Residence time Independent variable: Odour of the butterflies

Factors: Wasps' olfactory and visual perception Assumptions: No effect of environment is there (chemicals secreted by the plants, other parasites etc)

Since the data collected during this test is non-parametric (non-normally distributed) we use chi-square test, to calculate errors and to find the goodness of the curve in this test. Chi square test is a test which tells us if there is significant difference between expected and observed values.

The **two tailed binomial test** is used to find the preference of wasps to mount amongst the three kinds of butterflies. Some parasitoids have the ability to mount the host naturally while some learn with experience. To differentiate this, we use GLM where we assign a binary code to the butterflies, i.e., 1 to the synthetically treated females and 0 to virgin females.

Dependent variable: Proportion of first mounts Independent variable: Odour of the butterflies

Factors: Wasps' ability to mount the butterfly, olfactory and visual perception

Assumptions: No effect of environment is there (chemicals secreted by the plants, other parasites etc)

Since the experiments conducted above are done in a simulated environment, there is no interaction between the insects and the plants. This leads to a lot of errors because plants can cause hindrance when a butterfly lays her eggs or when the wasp is trying to detect the chemical signals. Therefore, **two-way ANOVA test** can be used to include this factor.

Dependent variable: Percentage of wasps attracted to the odour source Independent variable: Odour of the plant and eggs Factors: Wasps' olfactory and visual perception

$$\mu_{ij} = \mu + \alpha_i + \beta_j + \gamma_{ij} \qquad Y_{ijk} = \mu_{ij} + \epsilon_{ijk}$$

here i, j, k are for the three parameters no of eggs, type of plant and different kind of wasps.

### **Conclusion**

Differential equation:  $dR / dT = rR - P$  Initial condition: at  $T=0$  butterfly population is  $R_0$

Solved Equation:  $R = (e^{rT}(rR_0 - P) + P) / r$

Variables used: R-butterfly population at time T(carrying capacity) , T- mean generation time, r- birth rate, P- death rate because of wasps (probability of wasps killing eggs  $\times$  wasp population at time T)