# Studies on the host range and ovipositional preference of *Trichogramma* spp. under laboratory conditions, at Raipur, Chhattisgarh

**M. Sc. (Ag.) Thesis**

**by**

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# Studies on the host range and ovipositional preference of *Trichogramma* spp. under laboratory conditions, at Raipur, Chhattisgarh

**Thesis**

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**Indira Gandhi Krishi Vishwavidyalaya, Raipur (C.G.)**

**by**

**Monalisa Mukherjee**

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**in**

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**(Entomology)**

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# CERTIFICATE – I

This is to certify that the thesis entitled **"Studies on the host range and ovipositional preference of *Trichogramma* spp. under laboratory conditions, at Raipur, Chhattisgarh"** submitted in partial fulfilment of the requirements for the degree of **Master of science in Agriculture** of the Indira Gandhi Krishi Vishwavidyalaya, Raipur, is a record of the bonafide research work carried out by **Monalisa Mukherjee** under my guidance and supervision. The subject of the thesis has been approved by the Student's Advisory Committee and the Director of Instructions.

No part of the thesis has been submitted for any other degree or diploma or certificate course. All the assistance and help received during the course of the investigations have been duly acknowledged.

**Dr. (Smt.) J. L. Ganguli**

**(Chairman)**

**THESIS APPROVED BY THE STUDENT'S ADVISORY COMMITTEE**

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(Member from other Department)

**Dr. R.R Saxena**
(Member from supporting Department)

# CERTIFICATE – II

This is to certify that the thesis entitled **"Studies on the host range and ovipositional preference of *Trichogramma spp.* under laboratory conditions, at Raipur, Chhattisgarh"** submitted by **Monalisa Mukherjee** to the Indira Gandhi Krishi Vishwavidyalaya, Raipur, in partial fulfilment of the requirements for the degree of **Master of science in Agriculture** in the Department of Entomology has been approved by the external examiner and Student"s Advisory Committee after oral examination.

**Signature of Head of the Department**

**Date:** 03/08/2021 **(Name)** Dr. D.k. Rana

**Major Advisor** ----------

**Faculty Dean** ----------
28/8/21

**Approved/Not approved**

**Director of Instructions** ----------

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*Date:* 30/06/2021

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| | |
|---|---|
| % | Per cent |
| @ | At the rate of |
| / | Per |
| o C | Degree Celsius |

# LIST OF ABBREVIATIONS

| | |
|---|---|
| CD | Critical Difference |
| CV | Cofficient of variation |
| cm | Centimeter |
| *et al.* | And co-worker/ and others |
| *i.e.* | That is |
| m | meter |
| mm | Milli metre |
| NS | Non-significant |
| S | Significant |
| S. No. | Serial number |
| *viz.* | Namely |
| SEm | Standard error of mean |
| SMW | Standard Meteorological Week |
| MAX T | Maximum Temperature |
| MIN T | Minimum Temperature |
| RH | Relative Humidity |
| Hr | Hours |
| Fig. | Figure |
| Ha | Hactare |
| YSB | Yellow stem borer |

## THESIS ABSTRACT

| | | |
|---|---|---|
| (a) Title of the thesis | : | "Studies on the host range and ovipositional preference of Trichogramma spp. under laboratory conditions, at Raipur, Chhattisgarh". |
| (b)Full name of student | : | Monalisa Mukherjee |
| (c)Major subject | : | Entomology |
| (d)Name and Address of the major advisor | : | Dr. (Smt.) J. L. Ganguli, Professor, Department of Entomology, College of Agriculture, IGKV, Raipur (C.G.). |
| e)Degree to be awarded | : | Master of Science in Agriculture |

Monalisa Mukherjee
Signature of the student

Signature of Major Advisor

Date 30/06/2021

Signature of the head of the department

## ABSTRACT

Biological control is an important part of integrated pest management programs. Use of chemical control is threatening for both growers and consumers; it not only has a detrimental effect on the environment but also adversely affects the soil health as well. The use of bioagents reduces the chemical inputs and conserves the natural enemies of the insect pests and thus plays an important role in maintaining the ecological balance in an eco-friendly manner.

The egg parasitoid, *Trichogramma* spp. (Hymenoptera, Trichogrammatidae) is considered as an important bioagent of several lepidopteran pests. *Trichogramma* are minute wasps, which can be hardly seen by hand lens or microscope. They lay their eggs in lepidopteran eggs, destroying the developing embryo before it hatches, thus avoiding the damaging larval stage. The wasps accomplish their embryonic maturity inside the host eggs and have a broad range of adaptability and host specificity. Females utilize chemical and visual signals, including form, colour, age and size of the eggs to determine the best accommodation for their offsprings to accomplish their metabolic, nutritional and ecological needs while searching their host eggs.

In the present research on the host range and ovipositional preference of *Trichogramma japonicum, T. chilonis* and *T. pretiosum* conducted under laboratory conditions revealed that only lepidopteran insect's egg were parasitized by all the three species of *Trichogramma*. Maximum (9 adults) 7.66 mean number of adult emergence was found from a single host egg of *Daphis nerii* by *T. chilonis* followed by *T. pretiosum* (7 adults) 6.67 mean number of adult emerged from single host egg of *D. nerii*. Minimum number of adult emergence was found in *T. japonicum i.e*. 0.33 from a single host egg of *Spodoptera litura.* No parasitization or emergence was recorded from eggs of insects belonging to Hemiptera, Coleoptera and eggs of Arachnids (non-insect). Statistically, significant positive correlation was found between the mean number of adult emerged from *T. japonicum* (r= 0.704), *T.chilonis* (r= 0.898) and *T. pretiosum* (r= 0.935) which was recorded as r=0.704, r=0.898 and r=0.935 in case of *T. japonicum, T. chilonis* and *T. pretiosum* respectively with size of the eggs.

Studies conducted on testing of the vertical and horizontal movement of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions using transparent PVC pipes of 7.00 mm diameter with different lengths *i.e.* 1m, 2m, 3m and 5m revealed that, all the three species of *Trichogramma* traveled upto 2 meters in both the directions *i.e*. vertically as well as horizontally with the help of the chemical cues released from the egg. It was also observed that when the parasitized cards were

placed without sentinel cards, the *Trichogramma* spp. emerging from the parasitized card showed very little movement in the transparent PVC pipe. Testing of the preferable movement among horizontal and vertical, revealed that *Trichogramma* preferred vertical movement (40-48 number of adults) to horizontal movement (20-28 number of adults).

Studies conducted to test the effect of UV treatment on the parasitization on lepidopteran eggs by *T. japonicum*, *T. chilonis* and *T. pretiosum* under laboratory conditions in embryo of host egg of *Corcyra cephalonica* with 2 UV tubes, one of short wavelength (254nm) and other one, long wavelength (365nm) kept at a distance of 15cm from the source of UV light revealed the emergence of 92.46 % and 15.73% in *T. japonicum* of 5 and 10 minutes of exposure respectively, while the emergence of *T. chilonis* after the same UV treatment was 90.95% and 4.66%, while in case of *T. pretiosum* it was 79.80% and 0.00% respectively. No emergence was found in case of 20 minutes of exposure and more. Thus, it can be concluded that *T. japonicum* had the highest sustainability while *T. pretiosum* was the most vulnerable species in the exposure towards UV radiation.

## शोध ग्रन्थ सारांश

| | | |
|---|---|---|
| (अ) शोध ग्रन्थ का शीर्षक | : | "प्रयोगशाला स्थिति में ट्राइकोग्रामा प्रजाति का पोषक विस्तार एवं अण्डरोपण वरीयता का रायपुर, छत्तीसगढ़ में अध्ययन" |
| (ब) छात्रा का पूरा नाम | : | मोनालिसा मुखर्जी |
| (स) मुख्य विषय | : | कीट विज्ञान |
| (द) मुख्य सलाहकार का नाम और पता | : | डॉ. (श्रीमती) जयालक्ष्मी गांगुली, प्राध्यापक (कीट विज्ञान), कृषि महाविद्यालय, इंदिरा गांधी कृषि विश्वविद्यालय, रायपुर, छ.ग. |
| (इ) उपाधि से सम्मानित किया जाता है | : | एम.एस.सी. (कृषि) |

Monalisa Mukherjee
छात्रा का हस्ताक्षर

मुख्य सलाहकार का हस्ताक्षर

दिनाँक 30/06/2021

विभागाध्यक्ष के हस्ताक्षर

## सारांश


जैविक नियंत्रण एकीकृत कीट प्रबंधन कार्यक्रमों का एक महत्वपूर्ण हिस्सा है। रासायनिक नियंत्रण उत्पादक और उपभोक्ता दोनों के लिए खतरनाक है। इसका पर्यावरण पर हानिकारक प्रभाव पड़ता है यहाँ तक कि यह मिट्टी के स्वास्थ्य को भी प्रभावित करता है। जैव नियंत्रण कारक रासायनिक कीट नाशकों के उपयोग को कम करते हैं और कीट के प्राकृतिक दुश्मनों का संरक्षण करते हैं। इस प्रकार ये पारिस्थितिक संतुलन को पर्यावरण के अनुकूल बनाने में महत्वपमर्ण भूमिका निभाते हैं।

अंड परजीवी, *ट्राइकोग्रामा* प्रजाति (हाइमेनोप्टेरा, ट्राइकोग्रामैटिडी) को लेपिडोप्टेरन कीटों का महत्वपूर्ण जैव नियंत्रक माना जाता है। *ट्राइकोग्रामा* सूक्ष्म ततैया है, जिन्हे हाथ के लेंस या सूक्ष्मदर्शी द्वारा मुश्किल से

देखा जा सकता है। वे अपने अंडे, लेपिडोप्टेरा वर्ग के कीटों के अंडों में देते हैं, और विकासशील भ्रूण को अंडे से इल्ली बनने के पहले नष्ट कर देते हैं, इस प्रकार हानिकारक इल्ली चरण से बचाते हैं। वे अपने भ्रूण विकास को परपोषी अंडे के भीतर पूरा करते हैं। छोटे ततैया में अनुकूलन क्षमता और परपोषी विशिष्टता की एक विस्तृत श्रृंखला होती है। मादा अंडे के रूप, रंग सहित अन्य रासायनिक एवं दृश्य संकेतों का उपयोग परपोषी अंडे खोजने के लिए करती है ताकि उनकी चयापचय, पोषण और पारिस्थितिक आवश्यकताओं के साथ अपनी संतानों के लिए सर्वोत्तम आवास का निर्धारण कर सके।

वर्तमान अध्ययनों में प्रयोगशाला स्थितियों के तहत् *ट्राइकोग्रामा जैपोनिकम, टी. चिलोनिस* तथा *टी. प्रीटियोसम* की परपोषी सीमा और अंडरोपण वरीयता पर किये गए प्रयोगों में पाया कि *ट्राइकोग्रामा* की सभी तीन प्रजातियों द्वारा केवल लेपिडोप्टेरा वर्ग की कीट के अंडों को परजीवीकृत किया। *डैफिस नेरी* के एकल परपोषी अंडे से *टी. चिलोनिस* के अधिकतम (9 वयस्क) 7.66 वयस्क उद्भव की औसत संख्या ततपश्चात् क्रमशः *टी. प्रीटियोसम* में (7 वयस्क) 6.67 औसत संख्या/वयस्क एकल अंडा तथा *स्पोडोप्टेरा लिटुरा* के एकल परपोषी अंडे से 0.33 वयस्क उद्भव की न्यूनतम संख्या पायी गई। हेमिप्टेरा और कोलोप्टेरा से संबंधित कीड़ों के अंडों और एराक्निड (अष्टपदी, गैर–कीट) के अंडों पर कोई परजीवीकरण या उद्भव दर्ज नहीं किया गया। अंडों से निकले वयस्कों की औसत संख्या तथा अंडों के आकार (नाप) के बीच सांख्यिकीय रूप से महत्वपूर्ण सकारात्मक सहसंबंध पाया गया जो कि *टी. जैपोनिकम* (आर = 0.704), *टी. चिलोनिस* (आर = 0.898) और *टी. प्रीटियोसम* (आर = 0.935) पायी गई।

वर्तमान प्रयोगशाला स्थितियों में किये गये अध्ययनों के तहत *ट्राइकोग्रामा जैपोनिकम, टी. चिलोनिस, टी. प्रीटियोसम* के लंबवत और आड़े गति अधिमान ज्ञात करने हेतु में अलग–अलग लंबाई जैसे 1 मीटर, 2 मीटर, 3 मीटर और 5 मीटर के 7 मि.मी. के व्यास वाले पारदर्शी पीवीसी पाइप का उपयोग किया जिससे यह ज्ञात हुआ कि सभी तीन प्रजातियां *ट्राइकोग्रामा* अंडे से निकलने वाले रासायनिक संकेत की मदद से लंबवत और क्षैतिज दोनों दिशाओं में 2 मीटर दूरी तक की यात्रा करती हैं। साथ ही प्रयोग द्वारा यह भी ज्ञात हुआ कि जब परजीवी कार्डो को बिना परजीवीकृत कार्ड के साथ रखा जाता है तो ट्राइकोग्रामा प्रजाति की गति पारदर्शी पीवीसी पाइप में बहुत कम दिखाई देती है। *ट्राइकोग्रामा* प्रजाति के बेहतर दिशा गति की जांच करने पर यह पाया गया कि (40–48) वयस्क लंबवत दिशा में पाये जाते हैं, जबकि (24–30) वयस्क क्षैतिज दिशा में पाए जाते हैं, अतः यह निष्कर्ष निकलता हैं कि *ट्राइकोग्रामा* प्रजाति द्वारा गति हेतु क्षैतिज दिशा की तुलना में लंबवत दिशा अधिक पसंद की जाती है।

पराबैंगनी किरणों के प्रभाव का परीक्षण हेतु ट्राइकोग्रामा के तीन विभिन्न प्रजाति अर्थात *टी. जैपोनिकम, टी. चिलोनिस, टी प्रीटियोसम* पर प्रयोगशाला स्थितियों में का उपयोग किया गया। यह प्रयोग लेपिडोप्टेरान परपोषी कीट, *कॉरसाइरा सेफेलोनिका* के अंडों पर 2 पराबैंगनी किरणों की नली, एक छोटी तरंग दैर्ध्य (254nm) और दूसरी लम्बी तरंग दैर्ध्य (365nm) में 5 मिनट और 10 मिनट रखने पर 92.46 प्रतिशत और 15. 73 प्रतिशत वयस्कों का उद्भव हुआ, जबकि *टी. चिलोनिस,* में 90.94 और 4.66 प्रतिशत और *टी प्रीटियोसम* में

क्रमशः 79.80 प्रतिशत और 0.00 प्रतिशत उद्भव उभरा पाया गया। 20 मिनट तथा 20 मिनट से अधिक पराबैंगनी स्त्रोत में रखने पर में कोई उद्भव नही पाया गया। इस से ज्ञात हुआ कि पराबैंगनी किरणों को सहने की अधिकतम क्षमता *टी. जैपोनिकम* में तथा न्यूनतम क्षमता *टी. प्रीटियोसम* में देखी गई।

# CHAPTER- I
# INTRODUCTION

In order to face the danger of economic pests, biological control is an important component of integrated pest management systems and is also suggested as the initial eco-friendly protection line. A significant factor for effective agricultural pest control is the ecological compatibility of a biological control agent. Chemical control not only raises production prices, but is also detrimental to the health of producers, consumers, non-target wildlife and the ecosystem. The emergence of pesticide resistance in secondary pest outbreaks and tighter pesticide regulation has revived interest in integrated pest management programmes that emphasise biological control.

The use of bioagents decreases chemical spraying, which effectively protects the pest's natural enemies. In addition, bio agents are host-specific and have a strong capacity to scan and spread, leaving no residual effects. While several inundative releases are required to reduce the insect population, sustained suppression of insects is achieved when the bioagent is established in a specific region. For the success of a biological control program, it is important to choose an appropriate type of natural enemy. Therefore, evaluations are necessary, which should mainly involve host preference and suitability, in order to select a species that is efficient for pest control.

Recent research emphasizes the importance of mass rearing of different bio control agents for augmentation and field releases. The important among them are egg parasitoids–*Trichogramma* spp. Trichogrammatids are one of the most important groups of biotic agents for the control of several lepidopteran pests around the world, and one of the most widely distributed egg parasitoid species in India and abroad, partly due to their ease of mass rearing and ability to target a variety of important crop insect pests. Compared to other parasitoid species, *Trichogramma* parasitoids are reported to have great versatility in parasitizing hosts (Li, 1994), and can be mass

grown in the laboratory on artificial hosts and combined inexpensively and readily with the other control methods.

The egg parasitoid, *Trichogramma* spp. (Hymenoptera: Trichogrammatidae) is considered as a significant biological control agent of insect pests in greenhouse and field crops. Trichogrammatid parasitoids have been employed as biological agents to manage a variety of lepidopteron insect pests for over a century. *Trichogramma* are minute wasps and there are approximately 650 species in the family of Trichogrammatidae (Jalali *et al*. 2009), of which about 70 have been used in biological control programs on row crops, fruit and forest trees (Li 1994). Charles V. Riley's first description of a *Trichogramma* species was in North America in 1871. He recognized the miniature wasps that hatched from the Viceroy butterfly's eggs as *Trichogramma minutum*.

Over 200 insect species are reported to be parasitized by various strains of Trichogrammatids. In India about 26 species of Trichogrammatids are recorded of which *T. chilonis, T. japonicum, T. achaeae, T. brassicae, T.pretiosum and T. minutum* are the most effective egg parasitoids for controlling lepidopteran insects (Singh and Jalali, 1994). *Trichogramma* is the most studied egg parasitoids in biological control programs. This technique necessitates a high number of natural enemies at the precise time when insect eggs are present and crop and weather conditions are favourable for release. The greater the release of *Trichogramma*, the faster the spread will be in all crops, rapid establishment is desirable. *Trichogramma's* bio-efficacy has been tested against some lepidopteron pests as they parasitize the larvae, eradicating lepidopteran larval stage before they may cause damage to the host plants.

*Trichogramma* are stout-bodied, minute wasps, which can hardly be seen without a hand lens or microscope. In all crops, rapid establishment is desirable. Within the eggs of other lepidopteran insects, they complete their life cycle and kill the host larvae inside the egg prior to hatching. Male is yellow with blackish abdomen and very strongly tapering and moderately long antennal hairs, forewing with four to six slanting lines setae, fringes about one sixth wing width. Gynobasic triangular

genitalia with dorsal extension, with very prominent lateral lobes and female with the first three abdominal terga black, antennae clubbed having short hairs on flagellum, and the ovipositor is the same length as or slightly longer than hind tibiae (Albo, 1986).

These tiny wasps have a wide range of adaptation and host specificity, are safe to employ, and are inexpensive. They inhibit almost all types of environments, from marshy marshes to scorching, arid deserts, and may be found in low-lying or completely arboreal environments (Nagarakatti and Nagaraja, 1977). Parasitoid reproduction depends on the suitability for immature growth of selected hosts (Vinson and Iwantsch, 1980), and some parasitoids are known to be able to differentiate between host qualities in order to maximize reproduction. The quality of the host changes with parasitoid size, age, and species, and has an impact on parasitoid propensity for parasitism as well as the health of their progeny. Host quality, which includes host size, age, and species, is frequently thought to be the most important element determining parasitisation preference and parasitoid health. Most species of Trichogrammatidae also tend to parasitize freshly laid host eggs (Zhang *et al*. 2014).

When the pupal stage is over, the new adult chews a hole through the egg and then immediately smells and explores the egg they came from. This is how, in order to find the next host egg, they find the requisite signs or smells they should be searching for. Shortly after emergence, adult parasitoids mate. In order to select host eggs, adult females employ chemical and visual signals like as egg shape and colour to choose the optimum accommodation for their progeny to meet their metabolic, nutritional, and ecological demands (Brodeur and Boivin 2004). After finding a suitable egg, an experienced female uses her ovipositor and antennal drumming to try to determine whether the egg has been parasitized previously or not (Tapping on the egg surface). Finally, she decides a suitable host if the host egg is young, robust, healthy and not parasitized by another wasp. In order to assess the size and quality of the target egg, females utilize antennal drumming, which decides amount of eggs the female is going to implant. In a single day, a single female may parasitize up to ten host eggs, and in her adult life period of 3-14 days, a single female wasp is able to parasitize up to 50

eggs. In approximately 24 hours, egg hatches and the parasite larvae matures too. Black melanin granules are accumulated on the internal layer of the egg chorion during the 3rd stage (3 to 4 days after the host egg is parasitized). In 7–14 days, the parasitoid larva consumes the inner material of the egg, pupates, and emerges as an adult wasp. This reduces the damage caused by the future caterpillars, and also breaks the life cycle of the pest, effectively preventing the pest population build up.

*Trichogramma* species typically have a fairly broad host range, with many strains reared on the eggs of stored-product moths due to which mass multiplication has become easier. To get high rate of emergence in laboratory in hot summer, artificial manipulation in temperature is necessary for successful rearing (Rejendran, 1999). The *Trichogramma* wasps emerge from cards in two to five days, depending on temperature, which should ideally be 80º to 90ºF. Emergence can be delayed by holding parasitized moth eggs at cooler temperatures.

The state of Chhattisgarh is fast, paving its way towards organic farming, which has opened new avenues and scope for biological control. Being rice prone area, the crop is challenged by a number of lepidopteran pests, among which the yellow stem borer, *Scripophaga incertulas* is one of the major pest of kharif and summer paddy. Leaf folder, *Cnaphlocrosis medinalis* and case worm, *Nymphula depuntalis,* and rice horn caterpillar, *Melanitis ismene* are few other lepidopteran defoliators attacking the crop commonly. Hence, management of these can be possible by releasing the egg parasitoid, *Trichogramma* through trichocards.

So far, practically no work has been done in this state on the host range and oviposition preference of *Trichogramma* spp. on the pests of paddy or other lepidopteran pests of different crops. Hence, looking to the above facts, the present piece of work was formulated entitled **"Studies on the host range and ovipositional preference of *Trichogramma* spp. under laboratory conditions, at Raipur, Chhattisgarh"** with following objectives:-

1. **To study the host range and ovipositional preference of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions.**

2. **To study the vertical and horizontal movement of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions.**

3. **To study the effect of UV treatment on the parasitization by *Trichogramma japonicum, T. chilonis, T. pretiosum* on lepidopteran eggs under laboratory conditions.**

# CHAPTER- II
# REVIEW AND LITERATURE

In this chapter, review of literature referring to the work done related with the topic entitled **"Studies on the host range and ovipositional preference of *Trichogramma* spp. under laboratory conditions, at Raipur, Chhattisgarh."** by many scholars in India and in other countries, has been briefly mentioned. The literature based on respective aspects is being reviewed under the given heads

1. To study the host range and ovipositional preference of *Trichogramma japonicum, T. chiloni and T. pretiosum* under laboratory conditions.
2. To study the vertical and horizontal movement of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions.
3. To study the effect of UV treatment on the parasitization by *Trichogramma japonicum, T. chilonis, T. pretiosum* on lepidopteran eggs under laboratory conditions.

## 2.1 To study the host range and ovipositional preference of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions.

**Ruberson and Kring (1993)** evaluated the extent to which the age of *Helicoverpa zea* eggs affects their acceptability and suitability for the parasitoid *T. pretiosum*. Three ages of host egg *viz.,* 14, 38 and 62 hour old were selected in which the augmenting time was longer and survival was lower in 62 hour old age than in younger one. The parasitoid preferred 14 hour old host egg over the other two ages on first contact but no preference was exhibited after one egg.

**Grenier *et al.* (2000)** discovered that the length of the body was longer in *T. evanescens* than in *T. pretiosum*, but the breadth was same in all three species. The

size of *T. evanescens* acquired in *Mamestra brassicae* host when three or four insects emerged was comparable to that obtained in *Ephestia kuehniella* host when parasitized singly. The size of the ovipositor was significant as it may impact the potential of *in- vitro* egg laying in artificial *Corcyra cephalonica* and *Helicoverpa armigera* host eggs.

**Bajpai *et al.* (2002)** conducted studies and concluded that the preference of *Trichgramma chilonis* Ishii as parasitoids of *Helicoverpa armigera* (Hubner) eggs in soybean eco-system, with parasitoid host ratio of 1:10 and 1:20 could provide maximum parasitism of 70 and 64 %, respectively. However, the percent adult emergence from the parasitoids eggs was not affected by the different parasitoids-host ratio.

**Bajpai (2002)** further tested the preference of *chilonis,* an egg parasitoid of H. *armigera* in soybean ecosystem with a ratio of 1:10 and 1:20 which resulted in maximum parasitism 70 and 65 percent respectively. Thus, it could be concluded that the emergence of *T. chilonis* was not affected by the parasitoid host ratio.

**Ballal and Singh (2003)** compared the efficiency of three species od *Trichogramma viz., T. chilonis, T. pretiosum* and *T. brasiliense* in the laboratory and screen house settings in Karnataka, India during 1996-97, and found that, *T. chilonis* was the most efficient parasitoid of *H.armigera* eggs on sunflower plants under screen house circumstances when compared to the other two trichogrammatid species. *T. chilonis* parasitized 50.1 and 11.4 percent of *H. armigera* eggs after releasing 50,000 parasitized eggs per ha on sunflower and red grams, respectively. *T. chilonis* parasitism was unaffected by the location of *H. armigera* eggs on different regions of the sunflower plant. However, parasitism by the same trichogrammatid varied dramatically among different red gram plant sections. It parasitized 43.4 and 18.7% of *H. armigera* eggs on leaves and flowers, respectively, but only 3.9 percent of *H. armigera* eggs on pods. The parasitism *T. chilonis* was significantly affected by the

growth stage of red gram plants, with parasitism being intensely low on plants with pods.

**Romeis *et al.* (2005)** mentioned that egg parasitoids of the genus *Trichogramma* were among the most important and best-studied natural enemies worldwide. *Trichogramma* parasitism levels varied substantially depending on the habitat, plant, or plant structure on which the host eggs were found. They compiled a summary of the research on the processes that could explain the observed variation in parasitism rates. Plant spacing, plant structure, plant surface structural chemistry, plant volatiles, and plant colour were among the mechanisms studied. In addition, plants may also influence parasitoid behaviour and activity by supplying carbohydrate food sources such as nectar to adult wasps and influencing the nutritional quality of host eggs for offspring growth, knowledge of plant and habitat factors that affected *Trichogramma* spp. efficacy had important implications for biological control, and for assessing the risks that mass-released *Trichogramma* spp. may pose to non-target insects.

**Moore and Ross (2008)** reported *Daphnis nerii* (L.), the oleander hawk moth, for the first time at Guam in August, 2005 that in the field, larvae were observed feeding only on oleander, *Nerium oleander* L. Thirty out of thirty two *D. nerii* eggs collected from oleander leaves were parasitized. These eggs produced 181 hymenopterans parasitoids belonging to four species: *Trichogramma chilonis* Ishii (Trichogrammatidae), *Eupelmus* sp. (Eupelmidae), *Telenomus (Aholcus)* sp. (Scelionidae), and *Ooencyrtus* nr. *Papilionis* (Encyrtidae).

**Goulart *et al*. (2011)** tested the host preference of the egg parasitoids *Telenomus remus* and *Trichogramma pretiosum*. The tests were conducted in a laboratory setting with well regulated environmental conditions (25°C temperature, 70% relative humidity, and a 14-hour photo phase). It was discovered that *T. remus* reared in *Spodoptera cosmioides* eggs and *T. pretiosum* grown in *A. gemmatalis* eggs preferred to parasitize eggs of the host where the parasitoid was grown. When *T.*

*pretiosum* was reared in *S. frugiperda* eggs and *T. remus* was grown on *A. gemmatali* eggs, no egg preference was detected because more than one pest species may be present in the field, as opposed to those where they were raised in the laboratory, host preference is an essential criterion for biological control strategies.

**Chakraborty (2012*)*** investigated the amount of yellow stem borer (YSB), *Scirpophaga incertulas*, parasitization in an insecticide-free rice field in Raiganj, Uttar Dinajpur, West Bengal, during three years (2008-2010). The three most important YSB egg parasitoids found in the region were *Telonomus rowani*, *Gahan* (Scelionidae), *Tetrastichus schoenobii*, *Ferriere* (Eulophidae), and *Trichogramma chilonis* Ishii (Trichogrammatidae***).***The egg mass was largely parasitized by a single or two parasitoid species. It was unusual to find three parasitoids species in a single egg mass. The prevalence of parasitization by solely *Trichogramma* spp., *Telonomus* spp., and *Tetrastichus* spp. was 6.12 percent, 9.53 percent, and 48.44 percent, respectively. Parasitization by *Trichogramma* spp. + *Telonomus* spp. + *Tetrastichus* spp. was 3.46 percent, 21.06 percent, and 2.35 percent, respectively.

**Bhapkar *et al.* (2015)** studied the natural parasitisation in egg of *Papilio demoleus*. Found that the favourable period of activity of eggs parasite of *P. demoleus* was observed in July, December and November month respectively during July 2010 to February 2011 on different host plant of citrus. The mean egg parasitisation was recorded 11.81, 11.60 and 1.44 % on *Citrus reticulata*; 13.87, 13.07 and 1.29% on *Citrus sinensis* and 15.28, 13.38 and 1.56% on *Citrus aurantifolia*by *Trichogramma chilonis*, *Telonemus* spp. and *Ooencyrtuspapilionis* respectively.

**Saljoqi *et al*. (2015)** observed on extracting of *Spodoptera litura* on various artificial foods *i.e.,* bean based diet (Diet-1) and gram based diet (Diet-2); under controlled laboratory conditions of 27±2°C, 65±5% RH and 14:10 L:D, the paratism of gram based diet with *Trichogramma chilonis* (Ishii) was conducted . The results showed that feeding the 1st, 2nd and 3rd generations of the immature stage of *S. litura*on diet-2 resulted in highest percentage survival rates among all the parameters

of immature stages of *S. litura*. No significant difference was observed among the diets in the percentage parasitism rate of *S. litura* by, *T. chilonis*. Increased trend of parasitism coincided with the increased time of exposure. Highest parasitism rate of 91.1% and 89.5 % was observed in the egg cards exposed to *T. chilonis* for 24 h in Diet-2 and Diet-1, respectively, while after 16 h exposure, 90 and 88.6 % was recorded for Diet-2 and Diet- 1. Comparatively higher parasitism was observed in 16 and 24 h of exposure time of egg cards of *S. litura* to *T. chilonis*.

**Richa Kumari *et al.* (2016)** found that the parasitization capability of *T. chilonis* varied from 58.94 to 68.94% with an average of 64.28% while that of *T. pretiosum* ranged from 41.56 to 46.48% with an average of 43.52%, indicating the superiority of *T. chilonis* over *T. pretiosum.*

**Rui Tang[1] *et al.* (2017)** investigated the efficiency of two species of *Trichogramma* wasps against the yellow stem borer *Scirpophaga incertulas* and found that *T. japonicum* and *T. chilonis* parasitized the yellow stem borer egg masses at a rate of 60.0% - 9.13 % and 40.7 % - 7.11%, respectively, in a laboratory cage test. Both species had high emergence rates after parasitizing the host eggs (95.7% 0.12% for *T. japonicum* and 100% for *T. chilonis*).

**Honnayya and Gawande (2018)** conducted experiments on parasitization and efficacy of the three females of *Trichogramma chilonis* (Ishii) against important lepidopteron pests viz., *H. armigera, E. vitella, P. demoleus, A. janata* and *C. cephalonica*, having 24, 48, and 72 hours old age eggs. The results revealed that, the *T. chilonis* exhibited 86.25%, parasitization on 24 hrs old eggs of *Helicoverpa armigera*, 30.75, average number of adult emergence of *T. chilonis* in 24 hours old eggs of *Papilio demoleus* and 8.50 days required for adult emergence of *T. chilonis* in 72 hours old eggs of *Papilio demoleus*. Hence, it was concluded that the percentage of parasitization was more in *H. armigera* and average number of adult emergence and days required for adult emergence was more in *Papilio demoleus* as compared to all the 5 treatments. Hence, the *H. armigera* and *Papilio demoleus* eggs were more

suitable for rearing of parasitoid to get good development of biological parameters of *T. chilonis*.

**Wen[1] Mei Du *et al.* (2018)** reported that the host quality varied with egg size, age of the eggs, and species which influence the parasitisation preference of parasitoids and the fitness of their offspring. They studied the performance of three *Trichogramma* species, *T. japonicum*, *T. chilonis*, and *T. leucaniae*, on both fertilized and unfertilized eggs of their host *Corcyra cephalonica*at different host ages. In no-choice tests, three *Trichogramma* species parasitized significantly more fertilized eggs than unfertilized eggs when host age was 0 days old. The frequencies of fertilized eggs visited by *Trichogramma* parasitoids were significantly higher than those of unfertilized eggs. The number of unfertilized eggs visited by parasitoids without ovipositing was approximately 3.7 times greater than those of fertilized eggs. Thus, they concluded that *Trichogramma* were able to recognize fertilized and unfertilized host eggs.

**Niranjana *et al.* (2018)** reported that the parasitoids, *T. pretiosum* and *T. embryophagum* had the highest parasitism efficacy against one-day old eggs of the brinjal shoot and fruit borer, *L. orbonalis* during *Rabi*, 2013 (27°C ± 5°C) with 92.01 and 90.02 per cent respectively among nine different egg parasitoids *viz*., *T. pretiosum*, *T. embryophagum*, *T. achaeae*, *T. japonicum*, *T. brassicae*, *T. mwanzai*, *T. chilonis*, *T. dendrolim* and *T. evanescens* .*T.embryophagum* had the maximum emergence rate (90.22 per cent) and greatest ability to parasitize an average of 11.21 eggs of *L. orbonalis.*

**Asha *et al.* (2019)** experimented to determine the host eggs of different ages (1,2,3 and 4 days old) exposed to four gravid females of the all the four different parasitiod in four glass tubes separately with constant 75% relative humidity and 27$^{o}$C temperature. It was observed that under the conditions of the test all the four species of *Trichogramma species* showed maximum percent parasitisation when they were exposed to one day old eggs followed by two days old eggs.

**Masry and Wakeil (2020)** researched that the egg parasitoids have been employed successfully as inundative biological control agents against a wide range of agricultural pests. *Trichogramma evanescens* has been extensively studied in a variety of crops, including maize, cotton, rice, and sugarcane, as well as vegetables such as tomato, cabbage, pepper, and potato, and fruit orchards such as apple, olive, pomegranate, and grape, as well as for controlling forest and stored product insect pests. The parasitoids' efficiency was determined by the factitious hosts on which they are reared: *Sitotroga cerealella*, *Ephestia kuehniella*, and *Corcyra cephalonica***.** It typically rase just a few parasitoid strains or species in a specific time to maintain excellent product quality and minimise contamination with other parasitoid species. The quality of parasitoid wasps needs to be assessed by regulators. Many factors influence the release and disappearance rate in the field, including weather, crop, host, predation, pesticide usage, and parasitoid quality. Releases of several million female wasps/ha throughout the season in field applications across the world have proven to be highly successful in controlling many crops' main lepidopteran pests.

## 2.2 To study the vertical and horizontal movement of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions.

**McDougall and Mills (1997)** studied the dissemination of *Trichogramma platneri* parasitiods from a central release point in an apple orchard in terms of time and parasitiods released. During the first three days, higher *T. platneri* release rates resulted in greater parasitism of sentinel eggs, increasing from around 18 percent for a 5400 release rate to around 36 percent for a 32400 release rate. On the north side of the tree, *T. platneri* parasitized a higher proportion of sentinel eggs. During the first three days following release, parasitism of sentinel eggs decreased from 62% near the point-source to less than 10% 14 m distant.

**Wright *et al.* (2001)** experimented on yellow sticky cards to measure dispersal from central release sites over distances ranging from 35 to 230 m in a square 9 hectare grid of monitoring sites. *Trichogramma ostriniae* was detected using *Ostrinia*

*nubilalis* sentinel egg masses. The wasps dispersed quickly, up to 180 metres in 6 days and 230 metres in 21 days, according to the findings, the amount of *T. ostriniae* caught on sticky cards and sentinel egg mass parasitism reduced with distance from the release sites. Although 11-405 parasitism of egg masses occurred in monitoring sites farthest from the release sites.

**Sarah and Nicholas (2002)** exposed the sentinel eggs of the six physiologically suitable hosts, to *T. plantnerii* during inundative releases in a walnut orchard in California, Sarah and Nicholas and found that all six hosts were attacked equally. Parasitiods were active at all heights within the orchard canopy, ranging from 1.5 to 6.5 metres. *T. platnerii* inundative releases have the potential to affect non-target lepidopteran populations within the release area, as well as naturally occurring populations of green lacewings in walnut orchards.

**Ayvaz *et al.* (2007)** investigated the distance covered by *Trichogramma evanescens* in open field of corn and grapevines. When the host eggs were situated far away from the release site in a corn field and grapevines, the amount of parasitism was inversely associated with distance. At the release location, the parasitization rate on grapevines and maize plants was higher than elsewhere. Parasitization rates were considerably greater in cages with the highest wasp density. The proportion of parasitized host eggs was 11.41 percent when 1000 wasps were released, and this rate climbed to 29.75 percent and 62.06 percent when 2000 and 3000 wasps were released, respectively. A similar pattern was seen in plastic bags, and more parasitoids resulted in a decrease in adult pest emergence.

**Chapman *et al.* (2009)** tested in the summers of 2005 and 2006 in a single release of roughly 0.5 million wasps in two spatially different potato farms. Each release area had 25 monitoring locations that were 5 to 45 meters away from the release point and were equipped with yellow sticky cards and *Ostrinia nubilalis* egg sentinels to detect adult parasitoids and parasitism, respectively. The results demonstrated that adult *T. ostriniae* moved quickly from the release point, with

individual capture at 45 meters within 1 day of emergence. At this distance, high rates of parasitization (20-50%) were also detected, however the levels declined with increasing distance from the releasing point.

**Geetha and Balakrishnan (2011)** assessed the laboratory-reared *Trichogramma chilonis* Ishii for its ability to move in open field through parasitism on sentinel cards of *Corcyra cephalonica* placed at distances of 1, 3, 5, 7, 10, 15, 20, 25 and 30 meters, from the point of release of parasitoids at eight cardinal directions. Sentinel cards were recovered from the field after 1-7 days time at 24 hour interval to observe parasitism and adult emergence. The dispersal rate was affected significantly by the time allowed as well as by the distances from the release point while the impact of direction was not significant. The mean dispersal rate was quite high at 1-10 meter distance which decreased to 7.67 at 30 meter.

**Sharma and Aggarwal (2015)** observed that the dispersal or host searching capacity of *T. chilonis* and *T. japonicum* was negatively correlated with the distance between host eggs and parasitoid released point. The experiment was conducted on organic basmati rice field. Parasitization rate was more (11.39-18.10% and 9.77-15.49% respectively) near the release point (1-3 meter) with maximum parasitism at 1 meter by both the parasitoid species. The *T. chilonis* showed higher parasitism (18-20%) in different distance treatment in comparison to *T. japonicum* (upto 15%).

**Singhamuni *et al.* (2015)** assessed that *T. chilonis* and *T. achaeae* are egg parasitoids, considered as ideal candidates for managing the cabbage looper (*Trichoplusia ni*) through augmentative release. Mass rearing with steady supply of parasitoid is necessary to promote augmentative release. *Corcyra cephalonica* eggs are extensively used for mass rearing. Adult parasitoids should be maintained as a parent stock and they should be fed with suitable food at the right concentration using a suitable feeding technique. These aspects were studied with the objective of improving the existing mass rearing protocol. *T. achaeae* and *T. chilonis* accepted

stored eggs for parasitization, but the level of acceptance significantly varied with storage durations. The eggs of *C. cephalonica* stored for two weeks at 4 or 8 °C were acceptable for parasitization. Pupal stage of both parasitoids within parasitized eggs could be stored upto two weeks at 4°C or four weeks at 8°C while maintaining at least 7 percent adult emergenced. Performance of emerging adults in terms of parasitism significantly varied with storage duration. Bee honey was a better type of food source compared with glucose, fructose and sucrose for feeding parasitoids. The parasitoid adults could be fed with 50% bee honey using drop method successfully.

**Ranjith *et al*. (2018)** assessed the active movement of two Trichogrammatids viz., *Trichogramma chilonis* and *T. pretiosum* towards eggs of *Leucinodes orbonalis* and *Corcyra cephalonica* in 2cm diameter PVC tubes of 1, 2, 3, and 4 meters. The trichogrammatids moved up to 3m towards the eggs of *L. orbonalis* and *C. cephalonica. T. chilonis* showed more active movement than *T. pretiosum*. At a distance of 1m, 50 percent of *T. chilonis* was recaptured on *L. orbonalis* egg cards which got subsequently reduced on the increase in the length of PVC tubes, which were 14.64 percent at 2m and 1.19 percent at 3m and became 0 in a length of 4m. The study also showed that the preference of Trichogrammatids towards the natural host *(L. orbonalis)* was more when compared to the factitious host (*C. cephalonica*). The recapture of *T. chilonis* and *T. pretiosum* at release distance of 1 meter was 50.05 and 28.58 percent in *L. orbonalis* respectively whereas, *C. cephalonica* eggs attracted 41.25 and 9.66 percent parasitoids respectively. Among *T. chilonis, T. japonicum and T. pretiosum, T. pretiosum* showed a higher parasitisation of 90.00 percent and emergence of 91.03 percent on one day old eggs of *L. orbonalis.* The study also showed that the age of host was a matter in parasitisation as well emergence of trichogrammatids. The increase in host age was negatively associated with parasitisation as well as emergence of trichogrammatids irrespective of species.

## 2.3 To study the effect of UV treatment on the parasitization by *Trichogramma japonicum, T. chilonis, T. pretiosum* on lepidopteran eggs under laboratory conditions

**Romeis *et al.* (1997)** irradiated *Helicoverpa armigera* eggs with a 30W ultraviolet (UV)-lamp from a distance of 30cm. Egg mortality increased with duration of exposure to the UV-light source and exposure for 48 min was sufficient to kill 98% of the eggs. Survival of the egg parasitoid *Trichogramma chilonis* was lower on eggs irradiated for 60 and 90 minutes than on untreated control eggs. Clutch size and sex ratio of the parasitoid progeny were similar in irradiated and control eggs. The lower suitability of irradiated host eggs was not recognized by female wasps as they accepted irradiated and control eggs equally in a choice test. The increased parasitoid mortality in irradiated eggs might be attributed to morphological alterations in the host egg chorion. This notion was supported by the observation that the parasitoid's drilling time was much shorter on irradiated eggs than on control eggs.

**Shiranzi (2006)** examined the effects of temperature and photoperiod individually on *T. chilonis*. The fecundity grew as the temperature rose, but the adult's lifetime decreased. At 30°C, adult fecundity is highest and adult lifespan is lowest. *T. chilonis* reared on a 14:10 LD schedule demonstrated significantly higher fecundity per day than those reared on 12:12 or 16:8 LD schedules. Adult lifespan was unaffected by any of the three regimens.

**Faruki *et al.* (2007)** studied s on the eggs of stored grain pests *Tribolium castaneum, T. confusum* (Coleoptera) and *Cadra cautella* (Lepidoptera) belonging to three age groups, 1, 2, and 3 days-old, were exposed to ultraviolet (UV) rays with a wavelength of 254nm (UV-C) for different durations, found that increasing the time of exposure to UV-rays caused a gradual decrease in hatching in 2 and 3 day old eggs of *T. confusum*, and no hatching occurred after 24 minutes of exposure. In all insect tests, all treatment durations resulted in a substantial decrease in adult eclosion.

**Herlinda (2008)** conducted an experiment to evaluate the effect of ultra violet (UV) sterilized eggs of factitious host, *Corcyra cephalonica*, and to observe the effect of frozen eggs of the host on parasitism by *Trichogramma.* The first experiment was done on three levels of UV radiation intensities: 10, 15, and 20 watt combined with three levels of duration of UV radiation (30, 60, and 90 minutes of radiation). The second experiments was done on frozen eggs stored in freezer (– 4$^{o}$C) for 1, 2, and 3 hours. Result showed that the UV radiation intensities did not affect the host parasitism, and *Trichogramma* emergence but the duration of ultraviolet radiation significantly affected the host hatched. The lowest host hatching occurred on 90 minute radiation, but was not significantly different from 60-minute radiation. *Trichogramma* reared on 2 and 3 hour frozen eggs produced more parasitized eggs compared the one reared on 1-hour frozen eggs. Sex ratio was predominantly female based, and most female progenies were produced by *Trichogramma* reared on 2-hour frozen eggs. Longevity of *Trichogramma* reared on 1-hour frozen eggs was significantly lower than those reared on 2 and 3-hour frozen eggs. Sixty-minute UV radiation on the host eggs, and 2-hour frozen eggs was better for the egg parasitoid mass rearing.

**Aydin *et al.* (2012)** investigated that the potential of using gamma and ultraviolet radiation as an alternative treatment to increase the efficiency of *Trichogramma euproctidis* (Girault 1911) (Hymenoptera: Trichogrammatidae) in the laboratory. The developmental and adult stages of *T. euproctidis* were exposed to gamma radiation of different doses (0-30 Gy) and ultraviolet radiation of 254 nm wavelengths (UV-C) for different durations (0-10 min) to assess their effect on each of the instars and their potential in breaking the developmental cycle of the egg parasitoid. The $LD_{50}$ values for eggs, prepupae, pupae and adults were 8.1, 10.0, 22.7 and 9.5 Gy for gamma radiation and 9.5, 0.12, 2.0 and 11.9 min for UV radiation, respectively. The pupa and adult stages were more radiation resistant to both gamma and UV radiation. The most interesting and unexpected result obtained for the

prepupal stage was that UV radiation had a greater effect on prepupal stages than gamma radiation.

**Kyle *et al.* (2012)** investigated the consequences of UV radiation on the behavior and parasitism success of the agriculturally important egg parasitoid wasp, *Trichogramma* spp. Insects are at high risk from UV radiation because of their small size; radiation may penetrate significantly deeper into insect tissues than into larger organisms, there by disproportionately affecting their performance. They found that *Trichogramma* preferred to move toward higher intensities of UV-B radiation and parasitized more eggs in areas with higher UVB radiation. However, higher UV-B radiation reduced the number of adult wasps emerging from host eggs. These results could be of particular importance in the agricultural release of *Trichogramma*, especially in greenhouse settings, where levels of UV-B radiation are low.

**Naggar *et al.* (2013)** assessed that Ultraviolet rays (UV) was used to sterilize the eggs of Angoumois Grain Moth (AGM), *Sitotroga cerealella* (Olivier), the Mediterranean Flour Moth (MFM), *Ephestia kuehniella* Zeller to stop the embryonic development inside the eggs to be favorable for parasitism for longer time. *Trichogramma evanescens* and *T. brassicae* at 25°C (L.S.D at 5% 10.18) reared on AGM and MFM. The data indicated the highest number of black eggs reached 63.5% and 38.1% when in case of *T. brassicae* and *T. evanescens* on eggs of MFM with UV, respectively, while it reached 31.9% and 16.4% for the same species on the eggs of AGM with UV, respectively.

**Ksentini *et al.* (2014)** experimented in four different *Trichogramma* spp., namely *T.-oleae*, *T. cacoeciae*, *T. evanescens* and *T. bourarachae*, are collected from pomegranate orchard in Tunisia, and assessed in order to determine the impact of the *Ephestia kuehniella* egg qualities (ranging from freshly laid to four-day-old hosts) on both their fecundity and survivorship. It was found that UV-killed eggs stored during four days at room temperature (25 °C and 50% RH) were suitable for the development of *T. oleae* and *T. bourarachae*. Also, a combination of fecundity and pre-emergence

survivorship data showed that all egg qualities were to be presented to *T. bourarachae*, while it was advisable to provide *T. oleae* only with three and four-day-old eggs, *T. evanescens* with fresh and one-day-old eggs and finally *T. cacoeciae* with only fresh eggs.

**Edwin *et al.* (2016)** tested the eggs of commonly available hosts, *Spodoptera litura*, *Corcyra cephalonica*, *Plutella xylostella* and *Helicoverpa armigera* exposed to various temperature and nonionizing ultraviolet (UV) radiation treatments, to see if certain combinations of treatments aided *T. chilonis* development. The mean percentage of egg parasitization was highest on *S. litura* egg treatments at constant temperature (24, 28, 32 °C). Ultraviolet radiation treatments (3, 6, or 9 minutes at 254 nm) raised the mean percentage parasitization considerably more than non-UV therapies. In addition, *C. cephalonica* eggs exposed to non-ionizing UV radiation had a higher mean percentage of adult parasitoid emergence and viability than eggs from other hosts in all other treatments.

**Shinde *et al.* (2016)** conducted experiments during 2014-15 and 2015-16 to determine the effect of non-ionizing (UV) radiation of exposure time period for 30 minutes, 45 minutes, 60 minutes, 90 minutes, in chilled deep freezer for 2 hours (-5ºC) and untreated control on host *Corcyra cephalonica* and adult emergence of egg parasitoid, *T. chilonis.* The results revealed that the exposure time of 45 minutes (treatment applied @ 42 cm height from the target site with 30 W UV lamps) found suitable to irradiate the eggs of factitious host, *C. cephalonica* by enhancing parasitoids effectiveness without any detrimental effect of the UV radiation (nonionizing) on *Trichogramma* under laboratory condition.

**Xu[1] *et al.* (2016)** investigated the performance of three *Trichogramma* egg parasitoids, *T. japonicum*, *T. chilonis*, and *T. leucaniae* on fertilised, unfertilized, and UVF (ultraviolet)-irradiated fertilised (UVF) eggs of the rice moth, *Corcyra cephalonica* (Lepidoptera: Pyralidae). UVF hosts were parasitized the greatest by all three *Trichogramma* parasitoids, while unfertilized hosts were parasitized the least.

Adult Emergence was found in equal proportions. They also discovered that in unfertilized hosts, all three *Trichogramma* parasitoids grew more slowly. *Trichogramma* parasitoids preferred to parasitize *C. cephalonica* UVF eggs, with no deleterious consequences for their emergence or sex allocation.

# CHAPTER- III
# MATERIAL AND METHODS

The present investigation **"Studies on the host range and ovipositional preference of *Trichogramma* spp. under laboratory conditions, at Raipur, Chhattisgarh"** was conducted in the Biocontrol Laboratory, Department of Entomology, College of Agriculture, Indira Gandhi Krishi Vishwavidyalaya, Raipur (C.G) during the year 2019-2020 with the following of objectives:

1. To study the host range and ovipositional preference of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions.
2. To study the vertical and horizontal movement of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions.
3. To study the effect of UV treatment on the parasitization by *Trichogramma japonicum, T. chilonis, T. pretiosum* on lepidopteran eggs under laboratory conditions.

## 3.1 Geographical location

Raipur is situated in the central part of Chhattisgarh and lies at 21.6ºN latitude and 81.63º E longitude at an altitude of 298 meters above the sea level under Chhattisgarh plains.

## 3.2 Climatic condition:

The climate of this region is sub-humid to semi-arid. The average annual rainfall ranges from 1200- 1400 mm, of which 85 percent rainfall is received during the middle of June and by the end of September and very little during October to May. The maximum temperature goes as high as 48ºC during the month of May and the minimum as low as 6ºC in the winter months of December-January. The details

regarding materials used and techniques applied during the course of the investigation are described objective wise briefly as under:-

### 3.3 Source of insect cultures

The parasitoids (*Trichogramma japonicum, T. chilonis* and *T. pretiosum*) were bought from NBAIR, Bangalore. The Accession numbers of the different insects used are:-

i. *Trichogramma japonicum* NBA11- MP-TR1-65
ii. *Trichogramma chilonis* NBA11- MP-TR1-13
iii. *Trichgramma pretiosum* NBA11- MP-TR1-70

These were maintained at the Biocontrol Laboratory in controlled conditions (25–35ºC and 75 ± 5% RH) on the eggs of rice moth, *Corcyra cephalonica* (Lepidoptera: Pyralidae).

### 3.4 To study the host range and ovipositional preference of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions.

Under this objective, for testing the host range of *Trichogramma* spp., on eggs of different lepidopteran insects belonging to different families along with Coleopteran, Hemipteran species and eggs of spiders ( Arachnids) were collected from the field of different crops and brought to the Biocontrol laboratory and tested for parasitization by *Trichogramma* spp.

#### 3.4.1 Different species of host eggs tested:-

Various species of eggs were collected from different crops and ornamental plants. Eggs of rice yellow stem borer, rice butterfly, green stink bug, and spiders were collected from the rice field. Eggs of tobacco cut worm, *Spodoptera* spp. were collected from Soybean and Brinjal field. Eggs of fall armyworm, *Spodoptera frugiperda* were collected from maize field. Eggs of the lemon butterfly, *Papilio demoleus* were collected from Lemon plants. Eggs of gram pod borer, *Helicoverpa*

*armigera* were collected from the chickpea field. Eggs of oleander hawk moth and eggs of monarch butterflies were collected from oleander plants from IGKV, Raipur. Eggs of ladybird beetles, Mexican beetles and Reduviid bug were collected from the Biocontrol laboratory of IGKV, Raipur (C.G).

### 3.4.2 Studies on host range and fecundity of *Trichogramma* spp.

For studying the host range and number of eggs laid by *Trichogramma* in a single host egg, fixed numbers of eggs of each species were kept inside the petri-dish or glass vials and a fixed number of parasitized eggs of *Trichogramma japonicum* cut from respective trichocard were released. After 48hrs of release, the parasitized eggs were separated and shifted into new vials labelled and plugged with cotton wool.

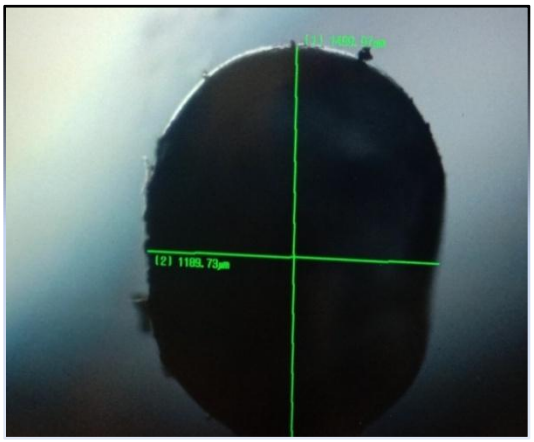
Observations were recorded just after the emergence of *Trichogramma* from the trichocard. The following parameters were considered:-

a. The time taken to locate the host eggs in the glass vial or petridish by the respective species of *Trichogramma*.

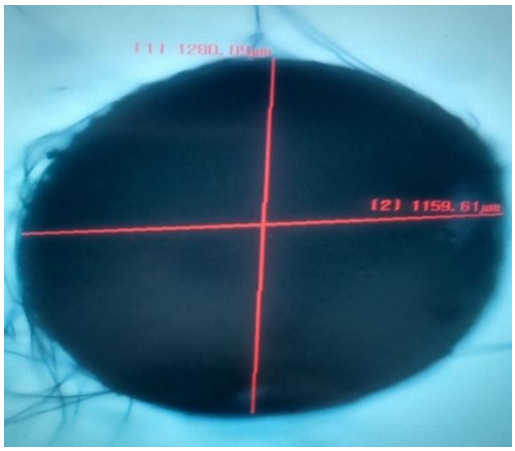
b. The number of parasitized eggs by the respective species of *Trichogramma*.

c. The number of *Trichogramma* emerging from each parasitized host egg.

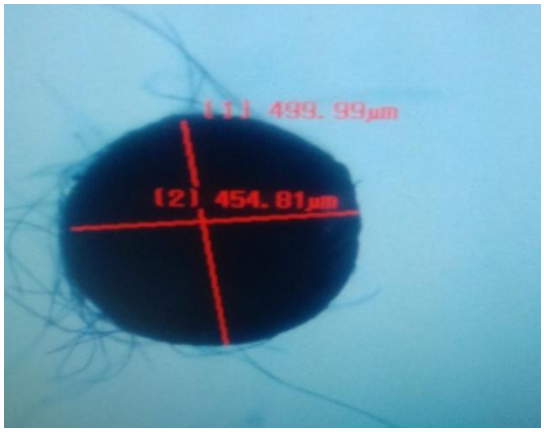
Similarly, it was repeated for *T. chilonis* and *T. pretiosum* as well.



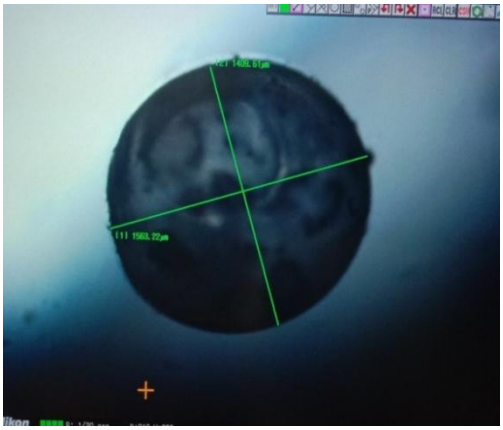

**Egg of *Papilio demoleus***



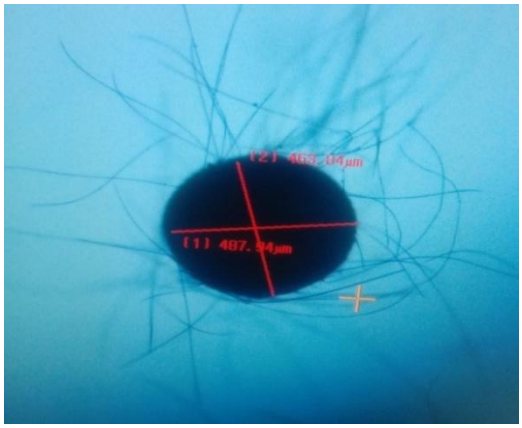

**Egg of *Danaus plexippus***




**Egg of *Spodoptera frugiperda***

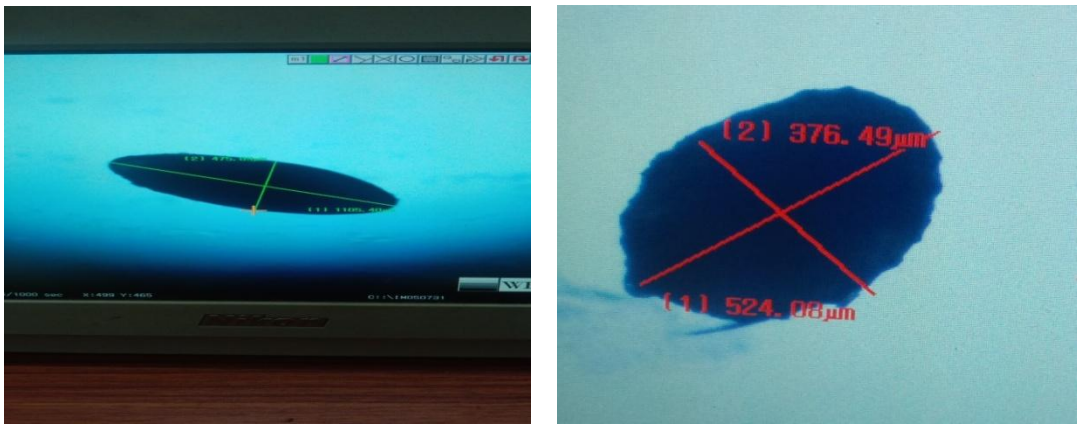


**Egg of *Daphis nerii***



**Egg of *Spodoptera litura***


**Egg of *Corcyra cephalonica***




**Egg of *Zygogramma bicolorata*** **Egg of *Melochinus* spp**

**Fig. 3.1: Single host egg s under Trinocular microscope to measure the size of egg**

**Plate 1: Experimental set up in glass vial for studying parasitisation and adult emergence of *Trichogramma* spp. from single host egg.**

**3.5 To study the vertical and horizontal movement of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions.**

**3.5.1 METHODOLOGY:-**

**3.5.1.1 Experiment 1: Testing the vertical movement of *Trichogramma japonicum.***

Under this experiment testing of the vertical movement of *T. japonicum* was done by using transparent plastic pipes of 6mm diameter, cut into different lengths

such as 1m, 3m, 5m, and so on. Plastic pipes of the above lengths were taken and inserted vertically into the lid of the plastic container (A) of 100ml capacity provided with a parasitized trichocard of 100 eggs placed it and closed with the lid. At the other end of the pipe, another plastic container (B) was connected with the lid having a sentinel card with 100 eggs. The experiment was replicated thrice by changing the lengths of the pipe.

The observations were recorded after 48 hours of the emergence of *Trichogramma* from the parasitized cards of container A to see the vertical distance traveled to reach container B.

A similar experiment was conducted for *T. chilonis* and *T. pretiosum* and replicated thrice by changing the lengths of the pipe.


**Plate 2: Experimental setup for testing the vertical movement of *Trichogramma* spp.**

### 3.5.1.2 Experiment 2: Testing the horizontal movement of *Trichogramma japonicum.*

Testing of the horizontal movement of *T. japonicum* was done using transparent plastic pipes of 6mm diameter of different lengths such as 1m, 3m, 5m, and so on. For this experiment, plastic pipes of the above lengths were taken and

inserted horizontally into the lid of a plastic container (A) of 100ml capacity. A parasitized trichocard of 100 eggs was placed inside the plastic container (A) and closed with the lid. At the other end of the pipe, another plastic container (B) with a lid was connected horizontally, having a sentinel card with 100 eggs. Observations were recorded after 48 hours of the emergence of *Trichogramma* from the parasitized cards of container A, to see the horizontal distance traveled to reach the container B. The experiment was replicated thrice by changing the lengths of the pipe.

A similar experiment was conducted for *T. chilonis* and *T. pretiosum* and replicated thrice by changing the lengths of the pipe.

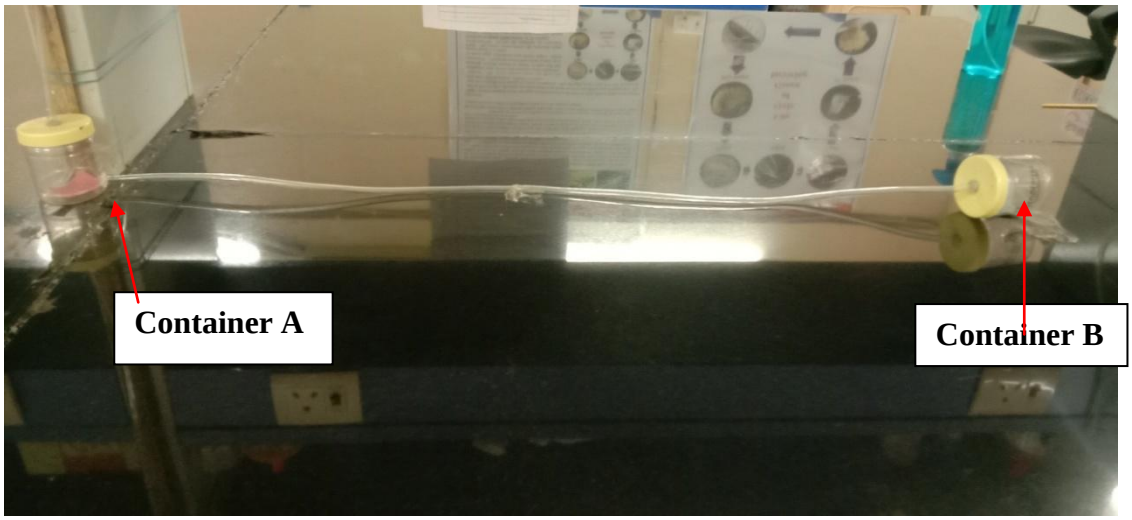



**Plate 3: Experimental setup for testing the horizontal movement of *Trichogramma* spp.**

### 3.5.1.3 Experiment 3: Testing the preferable movement of *Trichogramma* spp. between horizontal and vertical.

Under this experiment, testing of the preferable movement *T. japonicum* was done using transparent plastic pipes of 6mm diameter of 1m length. For this experiment, plastic pipes of 1m lengths were taken and inserted horizontally as well as vertically into the lid and body of a plastic container (A) of 100ml. A parasitized

trichocard of 100 eggs were placed inside the plastic container and closed with the lid. At the other end of both pipes, 2 plastic containers (B&C) with lids were connected, one horizontally, and the other vertically having sentinel cards with 100 eggs. In each container (B&C), observations were recorded after 48 hrs. of the emergence of *Trichogramma* from the parasitized cards of container A, to see the preferable movement between horizontal and vertical distance traveled to reach containers B and C. The experiment was replicated thrice.

A similar experiment was conducted for *T. chilonis* and *T. pretiosum* and replicated thrice.

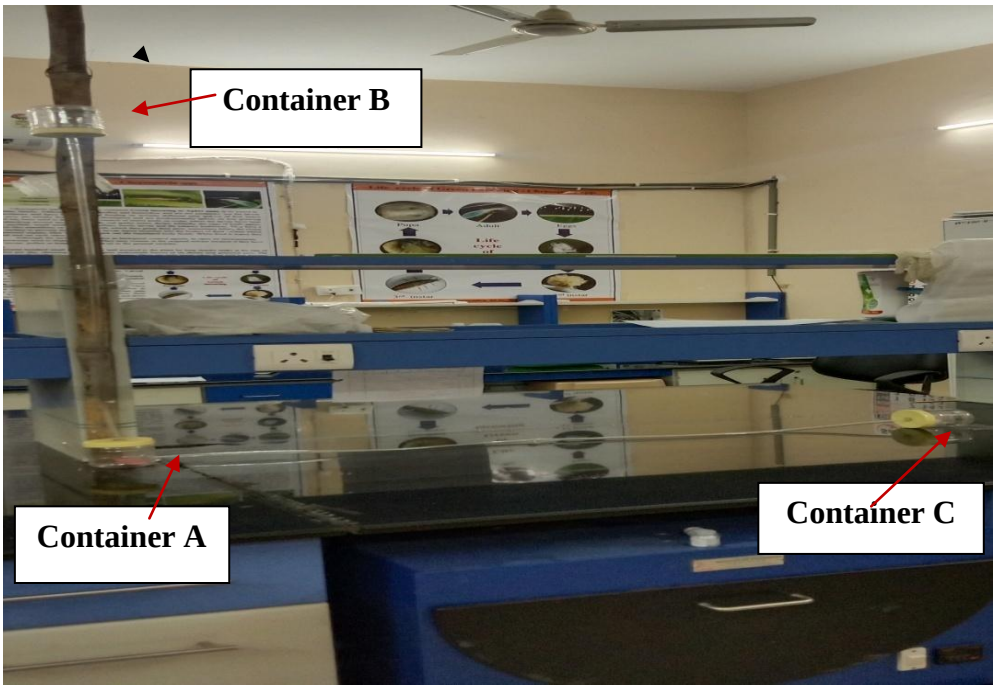



**Plate 4: Experimental setup for testing the ~~V~~vertical and horizontal movement of *Trichogramma* spp.**

## 3.6 To study the effect of UV treatment on the parasitization by *Trichogramma japonicum, T. chilonis, T. pretiosum* on *Corcyra* eggs under laboratory conditions.

### 3.6.1 METHODOLOGY

For this experiment, testing of parasitization by different species of *Trichogramma* was conducted on *Corcyra cephalonica* and oleander hawk moth eggs at various durations of exposure to UV rays.

Fixed numbers of the test eggs were glued to the paper cards. 100 eggs of *Corcyra* and 2 eggs of the hawk moth were irradiated with UV light in the UV chamber at 20cm distance for different durations of time starting from 10min, 20min, 30min, 40min, 50min, and 60 minutes and one untreated as contour of the observations were recorded after 3 days of the experiment in which the number of hatched and unhatched eggs were counted, to see the effect of UV treatment on the percentage of parasitization on eggs at different durations of exposure to UV rays.

The experiment was repeated thrice.

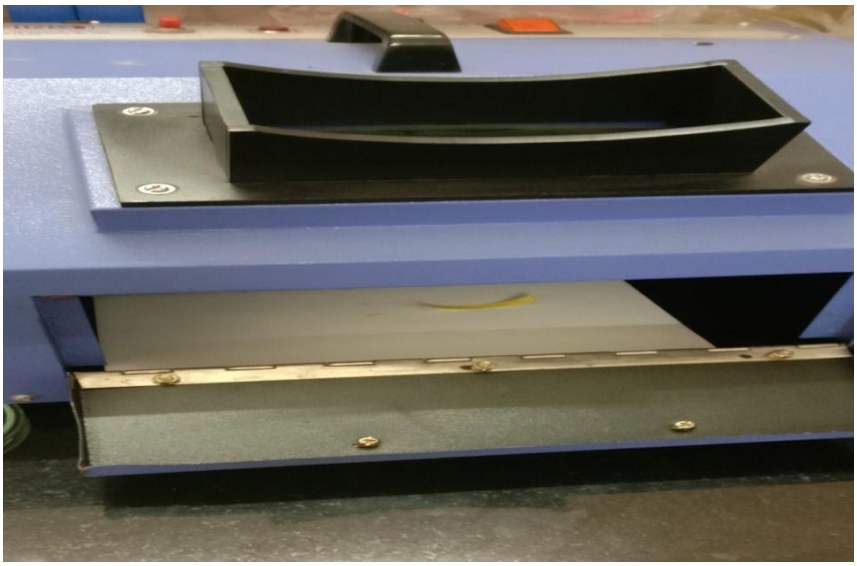


**Plate 5: Experimental setup for treating *Corcyra* eggs under Ultra Violet Cabinet**

# CHAPTER- IV

# RESULT AND DISCUSSION

This chapter deals with the results and discussion of the work done under different objectives of the experiment entitled, **"Studies on the host range and ovipositional preference of *Trichogramma* spp. under laboratory conditions, at Raipur, Chhattisgarh"** conducted in the Biological control laboratory of Department of Entomology, College of Agriculture, Indira Gandhi Krishi Vishwavidyalaya Raipur (Chhattisgarh) during the year 2019-20. The results are presented under the following sub headings.

4.1 To study the host range and ovipositional preference of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions.

4.2 To study the vertical and horizontal movement of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions.

4.3 To study the effect of UV treatment on the parasitization by *Trichogramma japonicum, T. chilonis, T. pretiosum* on lepidopteran eggs under laboratory conditions.

## 4.1 To study the host range and ovipositional preference of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions.

### 4.1.1 Parasitization by three different species of *Trichogramma* on eggs of different insects belonging to different orders.

The experiment was conducted to test the parasitization by different species of *Trichogramma* on the eggs of various insects belonging to different orders such as Lepidoptera, Hemiptera, Coleoptera and Arachnids. The size of the egg was also measured to correlate any preference of parasitization. The results obtained with

regard to average number of parasitoids emerged per egg in laboratory conditions on few important Lepidopteran Coleopteran and Hemipteran insect pests has been presented in Table 4.1.

**Table 4.1: Number of adult emergence in three different species of *Trichogramma* in different host eggs.**

| S. No. | Treatment | Average Size of host Egg(mm) | Mean number of *Trichogamma* spp. emerged /~~per~~ egg | | |
|---|---|---|---|---|---|
| | | | ***T. japonicum*** | ***T. chilonis*** | ***T. pretiosum*** |
| 1 | ***Papilio demoleus*** (Lepidoptera: Papilionidae) | 1.40 | 2.66 | 4.66 | 4.33 |
| | | | (1.91) | (2.36) | (2.29) |
| 2 | ***Daphis nerii*** (Lepidoptera: Sphingidae) | 1.80 | 2.33 | 7.66 | 6.67 |
| | | | (1.82) | (2.94) | (2.76) |
| 3 | ***Danaus plexippus*** (Lepidoptera: Nymplalidae) | 1.2* 0.9 | 0.66 | 2.33 | 2.33 |
| | | | (1.27) | (1.82) | (1.82) |
| 4 | ***Melanitis idea*** (Lepidoptera: Nymplalidae) | 1.30 | 0.66 | 1.66 | 2.33 |
| | | | (1.27) | (1.62) | (1.82) |
| 5 | ***Scripophaga incertulas*** (Lepidoptera: Crambidae) | 0.40 | 1.00 | 0.33 | 0.67 |
| | | | (1.41) | (1.14) | (1.27) |
| 6 | ***Helicoverpa armigera*** (Lepidoptera: Noctuidae) | 0.50 | 0.66 | 0.66 | 1.33 |
| | | | (1.27) | (1.27) | (1.52) |
| 7 | ***Spodoptera frugiperda*** (Lepidoptera: Noctuidae) | 0.49 | 0.66 | 0.67 | 1.00 |
| | | | (1.27) | (1.27) | (1.41) |
| 8 | ***Spodoptera litura*** (Lepidoptera: Noctuidae) | 0.46 | 0.33 | 1.00 | 0.67 |
| | | | (1.13) | (1.41) | (1.27) |
| 9 | ***Corcyra cephalonica*** (Lepidoptera: Pyralidae) | 0.50 | 1.00 | 1.00 | 1.00 |
| | | | (1.41) | (141) | (1.41) |
| 10 | ***Nezara viridula*** (Hemiptera: Pentatomidae) | 1.50 | 0.00 | 0.00 | 0.00 |
| | | | (1.00) | (1.00) | (1.00) |
| 11 | ***Halyomorpha halys*** (Hemiptera: Pentatomidae) | 2.00 | 0.00 | 0.00 | 0.00 |
| | | | (1.00) | (1.00) | (1.00) |
| 12 | ***Rhynocoris marginatus*** (Hemiptera: Reduviidae) | 2.30*0.5 | 0.00 | 0.00 | 0.00 |
| | | | (1.00) | (1.00) | (1.00) |
| 13 | ***Melochinus* spp.** (Coleoptera: Coccinellidae) | 1.1*0.4 | 0.00 | 0.00 | 0.00 |
| | | | (1.00) | (1.00) | (1.00) |
| 14 | ***Zygogramma bicolorata*** (Coleoptera: Chrysomelidae) | 1.1*0.5 | 0.00 | 0.00 | 0.00 |
| | | | (1.00) | (1.00) | (1.00) |
| 15 | **Arachnids** | 2.00 | 0.00 | 0.00 | 0.00 |
| | | | (1.00) | (1.00) | (1.00) |
| | **CD** | | 0.25 | 0.26 | 0.24 |
| | **SE (m) ±** | | 0.086 | 0.091 | 0.084 |
| | **SE (d)** | | 0.122 | 0.129 | 0.118 |
| | **CV** | | 11.901 | 11.164 | 10.04 |

### 4.1.2 Number of parasitoids emerged per egg.

Statistical analysis of the data on average number of parasitoids emerged per egg as presented in (Table 4.1) revealed that, the emergence of *Trichogramma* differed significantly among different size of host egg. Emergence of more number of *Trichogramma* was seen in bigger lepidopteran eggs. Among the three different species of *Trichogramma* tested, maximum number of emergence was recorded in *T.chilonis* with 4.66 and 7.66 number per egg in case of the eggs of *Papilio demoleus* and *Daphis nerii* respectivly.

During parasitization by all the three species of *Trichogramma* only lepidopteran insects were parasitized, no parasitization and emergence were recorded from eggs of insects belonging to Hemipteran, Coleopteran. Eggs of Arachnids (non-insect) were also not parasitized.

### 4.1.3 Number of adults of *Trichogramma japonicum* emerged per host egg.

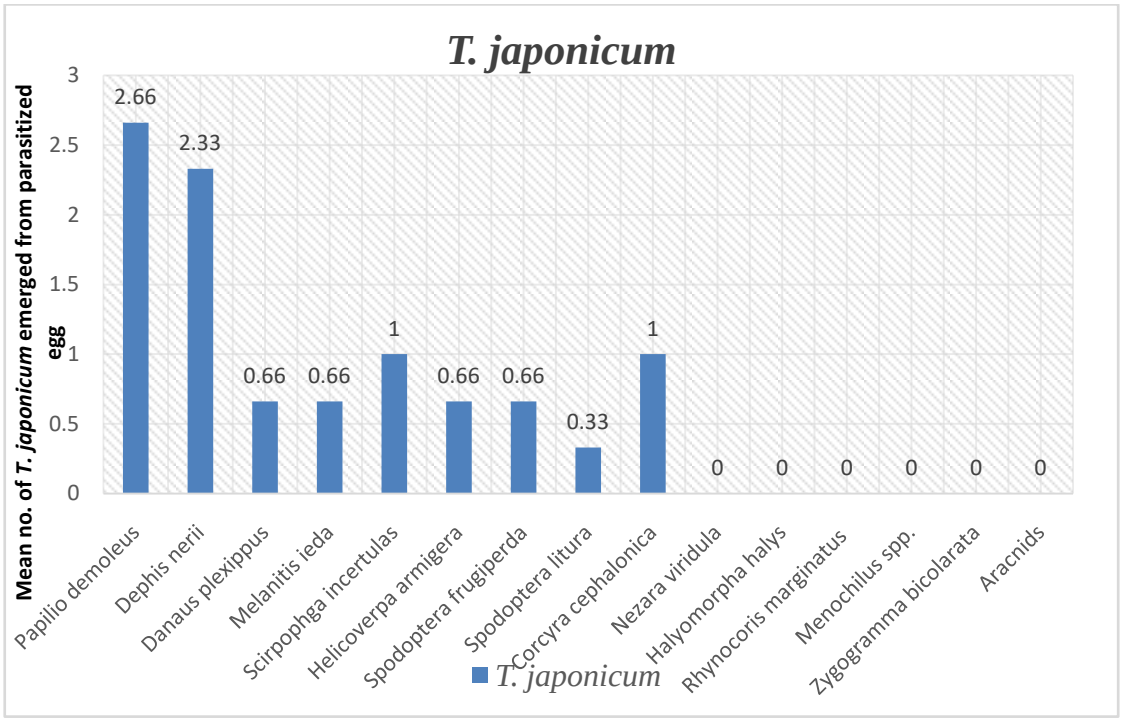



**Fig. 4.1: Number of adults of *T. japonicum* emerged per host egg.**

The statistical data related to the number of *T. japonicum* adult emerged per egg has been presented in the graph fig 4.1 showing that maximum number of adults emerged from the eggs of *Papilio demoleus* (lemon butterfly) with a mean number of 2.66 adults emerged from a single host egg. The shape of egg was spherical and size of egg is 1.2 mm diameter followed by *Daphis nerii* with a mean number 2.33 *Trichogramma* adults emerging from a single host egg. The shape of egg was spherical with a size of egg of 1.8mm diameter. No parasitisation were observed in the eggs of Hemiptera (*Nezara viridula*, *Halyomorapha halys*, *Rhynocoris marginatus)*, Coleoptera (*Melochinus* spp,. *Zygogramma bicolorata)* and the eggs of spiders belonging to class Arachnida.

Thus *Papilio demoleus* was found superior among all treatments of parasitization by *T. japonicum.*

### 4.1.4 Number of *Trichogramma chilonis* emerged per host egg

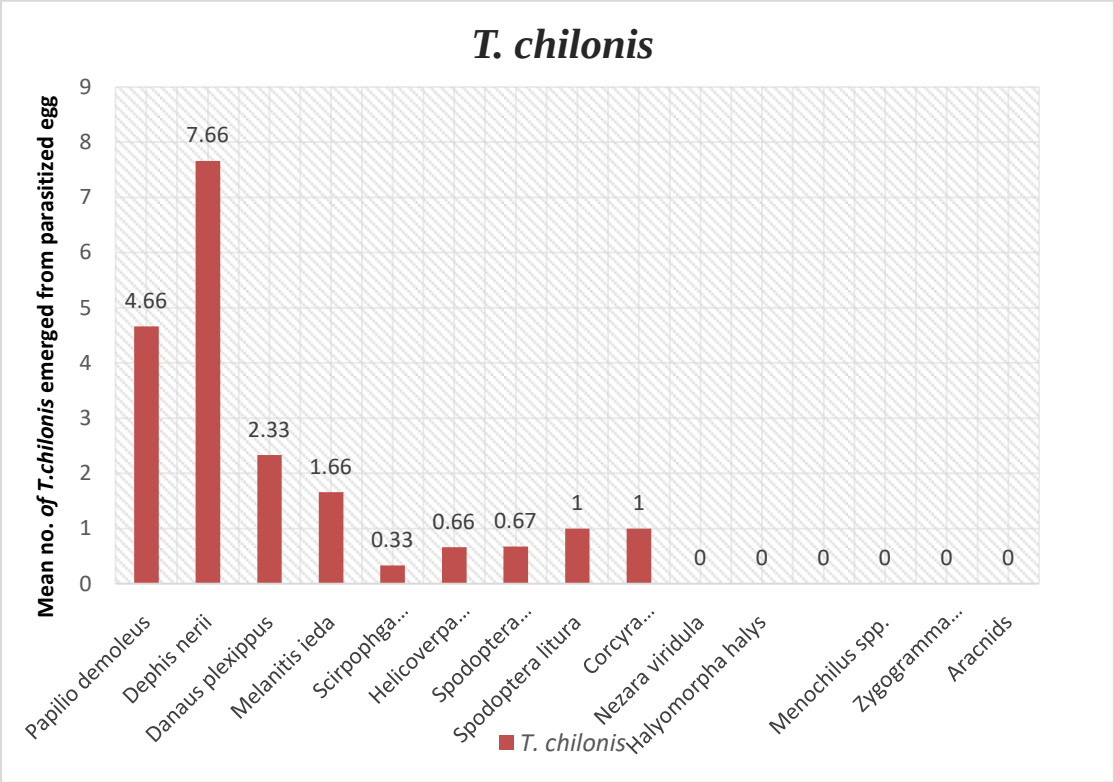



**Fig. 4.2: Number of *T. chilonis* emerged per host egg.**

The result showed that maximum number of adult is emerged from *Daphis nerii* up to (9 adults) 7.66 mean number of adult emerged from single host egg and size of egg is 1.8mm diameter, followed by *Papilia demoleus* (lemon butterfly) up to (6 adults) 4.66 mean number emerged from single host egg and size of egg is 1.4mm diameter.

No parasitisation were observed in the eggs of Hemiptera (*Nezara viridula, Halyomorapha halys, Rhynocoris marginatus),* Coleoptera (*Melochinus* spp,. *Zygogramma bicolorata)* and the eggs of spiders belonging to class Arachnida.

The treatment *Daphis nerii* was found superior among all other treatment because of the large size of the egg preferred by *T. chilonis.*

### 4.1.5 Number of *Trichogramma pretiosum* emerged per host egg

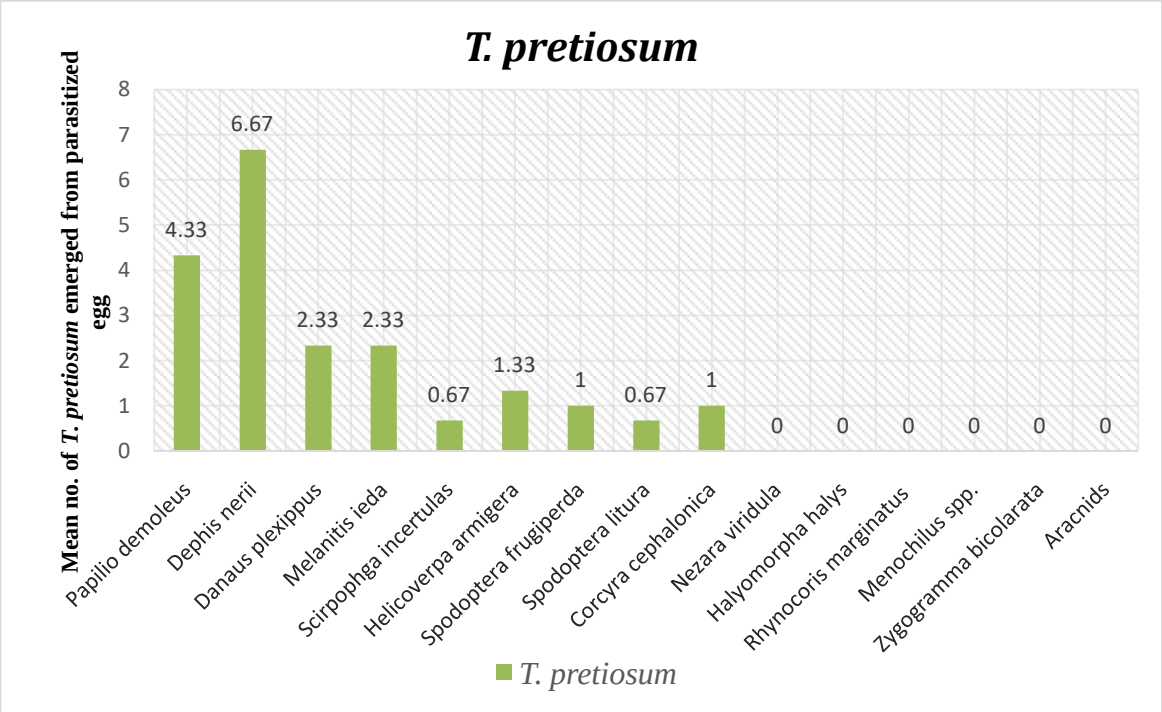



**Fig. 4.3 Number of *T. pretiosum* emerged per host egg.**

The statistical data on emergence of parasitoids from different host egg from *T. pretiosum* is presented in the fig 4.3 which revealed that the number of adults emerged from the host egg depends on size of the egg depicting preference by *T. pretiosum.*

Among the 15 treatments tested in the present experiment only lepidoptern insect's eggs were parasitized by *T. pretiosum.*

The result showed that mean maximum number (7 adults) 6.67 of adults ~~is~~ emerged from *Daphis nerii* from single host egg with an egg size of 1.8mm diameter. Second highest number of adult emerged from the host egg ~~is~~ of *Papili~~a~~o demoleus* (lemon butterfly) mean number up to (6 adults) 4.33 from single host with an egg egg size of 1.4mm diameter.

No parasitisation were observed in the eggs of Hemiptera (*Nezara viridula, Halyomorapha halys, and Rhynocoris marginatus),* Coleoptera (*Melochinus* spp. *Zygogramma bicolorata)* and the eggs of spiders belonging to class Arachnida.

The treatment *Daphis nerii* found superior among all other treatments because of the large size of the egg which was preferred by this species of *Trichogramma*.

### 4.1.6 Correlation between size of the eggs and number of *Trichogramma* species emerged.

**Table 4.2: Correlation between size of eggs and number of *Trichogramma* spp. emerged**

| S.No | *Trichogramma* spp | Size of egg |
|---|---|---|
| 1 | *T. japonicum* | r= 0.704* |
| 2 | *T.chilonis* | r= 0.898* |
| 3 | *T.pretiosum* | r= 0.935* |
| Significant at 5% level (0.632) | | |

**1. Correlation between size of the eggs and number of *T. japonicum* emerged.**

The mean number of *T. japonicum* emerged from single host egg ranged from 2.66-0.33. The size of the egg was considered independent variable whereas the number of adult emergence was the dependent variable. The correlation studies between mean number of *T. japonicum* emerged with the size of the eggs revealed statistically, significant positive correlation (r= 0.704) with the size of the eggs.

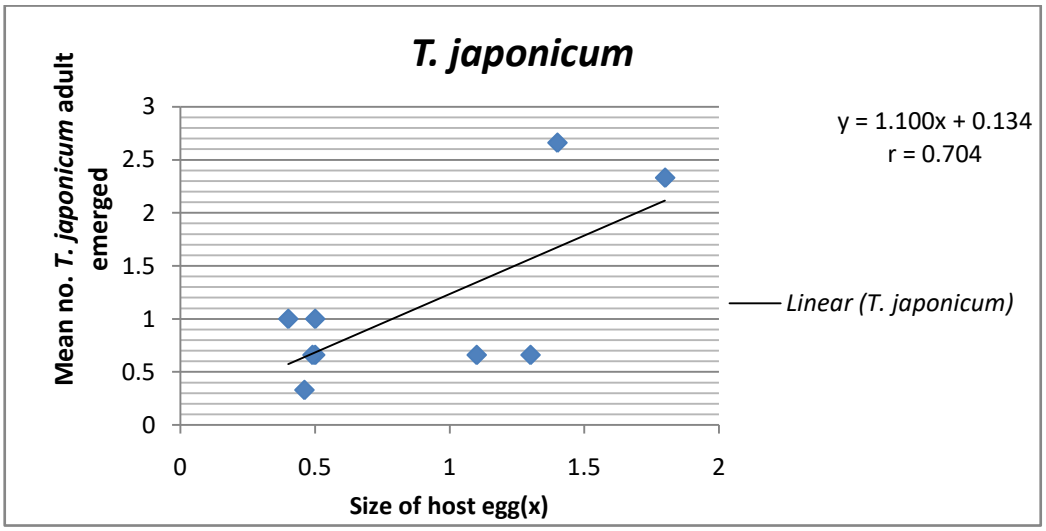



**Fig.4.4 Regression equation between size of the eggs and mean number of *Trichogramma japonicum* emerged.**

**2. Correlation between size of the eggs and number of *T. chilonis emerged*.**

The mean number of *T. chilonis* emerged from single host egg ranged from 7.66-0.33. The size of the egg was taken as independent variable whereas the number of adult emerged was the dependent variable. The correlation studies between mean number of *T.chilonis* emerged with the size of the eggs revealed that mean number of *T.chilonis* emergence highly statistically showed, significant positive correlation (r= 0.898).

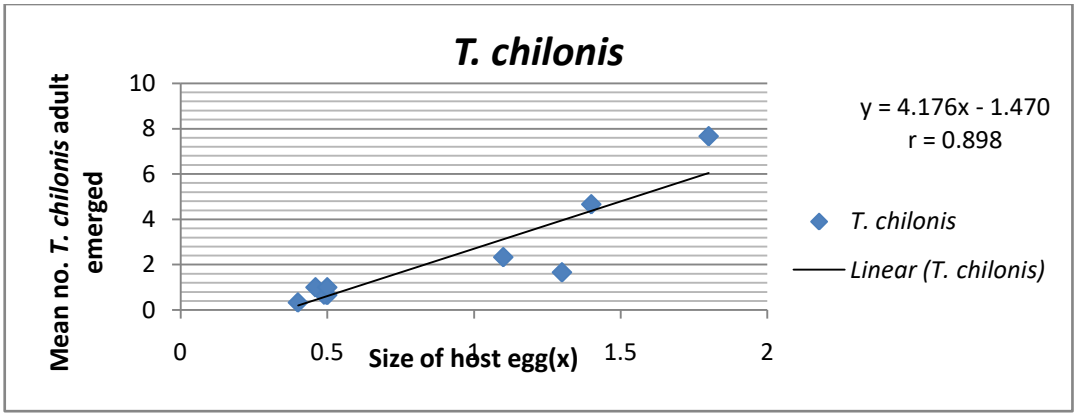



**Fig 4.5 Regression equation between size of the eggs and number of *Trichogramma chilonis* emerged.**

**3. Correlation between size of the eggs and number of *T. pretiosum* emerged.**

The mean number of *T. pretiosum* emerged from single host egg ranged from 2.66-0.33. The size of the egg was considered as independent variable whereas the number of adult emergened was the dependent variable. The correlation studies between mean number of *T. pretiosum* emerged with the size of the eggs revealed that mean number of *T. pretiosum* emerged depicted statistically highly, significant positive correlation (r= 0.935).

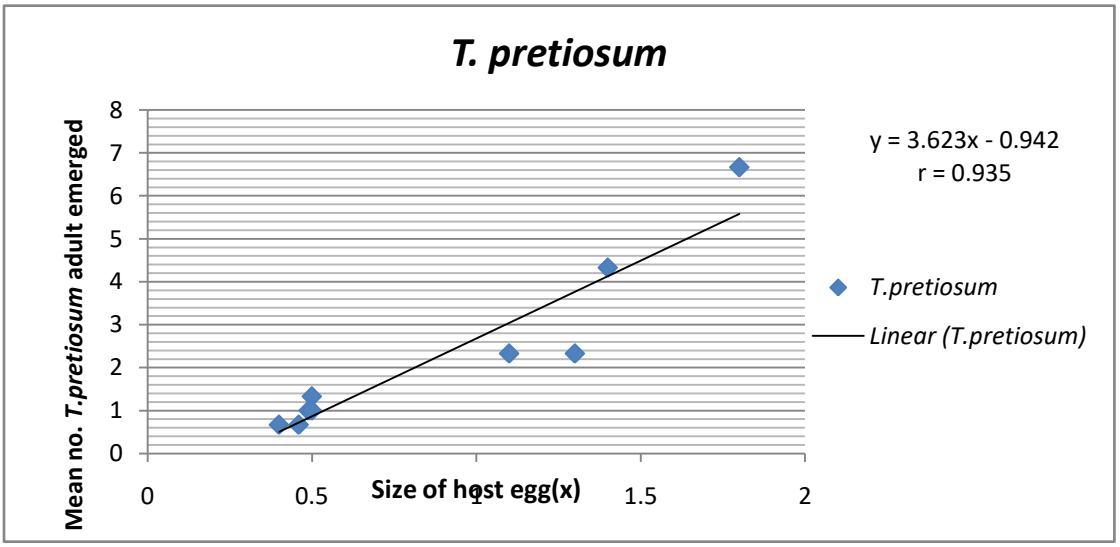



**Fig. 4.6 Regression equation between size of the eggs and number of *Trichogramma pretiosum* emerged.**

### 4.1.7 Comparison of the three species of *Trichogramma* in relation to parasitisation based on the number of adult *Trichogramma* emerged from egg of different species:

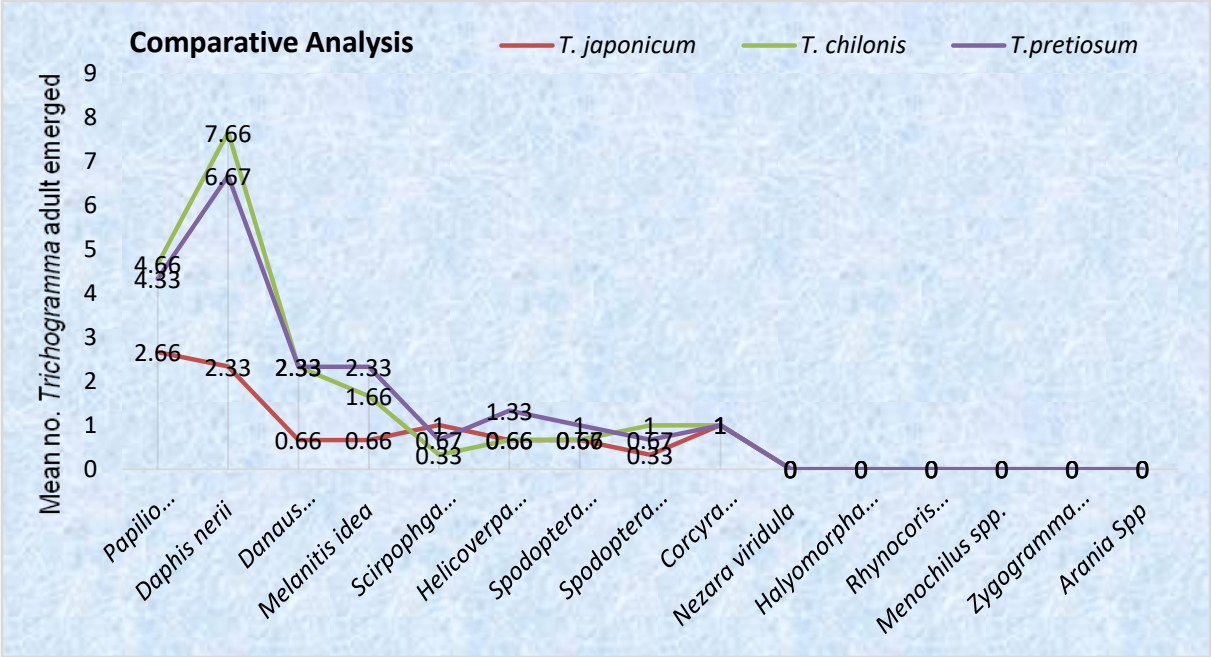



**Fig. 4.7 Comparison of the three species of *Trichogramma* in relation to parasitisation based on the number of adult *Trichogramma* emerged from egg of different species**

Among all the treatments, on the basis of adults of *Trichogramma* spp. emerged from parasitized host egg, revealed that the mean maximum number was found in *T. chilonis i.e* 7.66 from a single host egg of *Daphis nerii*, followed by *T. pretiosum* 6.67 in case of *Daphis nerii* while mean minimum number was recorded in *T. japonicum* with 0.33 emergence from single host egg of *Spodoptera litura*. Thus, *T. chilonis* and *T. pretiosum* can be used to control of most of the lepidopteran eggs. *Daphis nerii* and *Papilio demolus* were the most preffered host eggs in which the maximum emergence were observed.

No parasitization and emergence were recorded from eggs of insects belonging to Hemiptera and Coleoptera. Eggs of Arachnids (non-insect) were also not

parasitized. It was also observed that sperical eggs with no sculpturing on the outer covering were preffered more, as compared to conical shaped with ridged surface or eggs covered with scales or hairs.

These results are in accordance with the findings of earlier studies made by Honnayya and Gawande (2018) who also recorded maximum number of emergence *i.e*. 37.50 and 27.50 adult of *T.chilonis* emerged out of 20 eggs of *P. demoleus* and *H. armigera* respectively in which the parasitization percentage was found to be of 72.50 and 78.75 respectively but differs with the finding of Moore and Ross (2008) who recorded the mean number of *T. chilonis* which emerged from each egg was 12.4 with a range of 2 to 21 which is higher than present findings. Richa Kumari *et al.* (2016) also found that the parasitization capacity of *T. chilonis* was much higher than the parasitization capacity of *T. pretiosum* which is in line with the present results.

**4.2 To study the vertical and horizontal movement of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions.**

This is the maiden attempt to find out the active distance travelled by *Trichogramma* species in confined laboratory conditions (Temp- 25±2°C and RH-56±2%). Transparent PVC pipes of 0.7cm diameter were used for testing the movement of *Trichogramma*. For testing the vertical/ horizontal distance travelled by *Trichogramma* spp., experiment was set as shown in Plate no.2 and plate no. 3. Transparent PVC pipes were cut of 1m, 2m, 3m and 5m in length. One end of the pipe was fitted to a closed container 'A', having parasitized eggs of *Corcyra*, the other end was fitted vertically/ horizontally to another plastic container 'B' having sentinel cards of *Corcyra* eggs. Similar set up was done for different lengths of PVC pipes. This experiment was conducted in the first week of December 2019; and observations were recorded after 6 days on the number of eggs parasitized, confirming on the basis of change in color of eggs from white to black. It was observed in the present studies that a female *Trichogramma* was capable of parasitizing upto 10-150 eggs in her life span.

It was also observed that when the parasitized cards were placed without sentinel cards,–*Trichogramma* spp. emerging from the parasitized card showed very less movement, and they did not reach to the end containers through the pipe, rather they hovered in the container where the parasitized card were kept. By this behavioural observation, it could be concluded that their movement was in response of chemical cues released from the eggs which was the decisive factor for their orientation in a specified direction.

The percent parasitization of different species of *Trichogramma* is presented in the following table below:-

## i. Vertical movement of *Trichogramma* spp.

**Table 4.3: Number of different species of *Trichogramma* moving from container A to B in vertical direction after emerging from parasitized Trichocards through various distances.**

| S. No | Vertical movement (In Meters) | Mean percentage of *Corcyra* egg parasitized | | |
|---|---|---|---|---|
| | | ***T. japonicum*** | ***T. chilonis*** | ***T. pretiosum*** |
| 1 | 1.00 | 86.86 | 90.56 | 77.76 |
| | | (68.85) | (72.14) | (61.39) |
| 2 | 2.00 | 84.3 | 91.23 | 83.76 |
| | | (66.7) | (72.87) | (66.52) |
| 3 | 3.00 | 0.00 | 0.00 | 0.00 |
| | | (0) | (0) | (0) |
| 4 | 5.00 | 0.00 | 0.00 | 0.00 |
| | | (0) | (0) | (0) |
| **C.D.** | | 3.92 | 3.18 | 6.17 |
| **SE (m)±** | | 1.184 | 0.963 | 1.86 |
| **SE (d)** | | 1.674 | 1.362 | 2.63 |
| **C.V.** | | 6.051 | 4.6 | 10.06 |

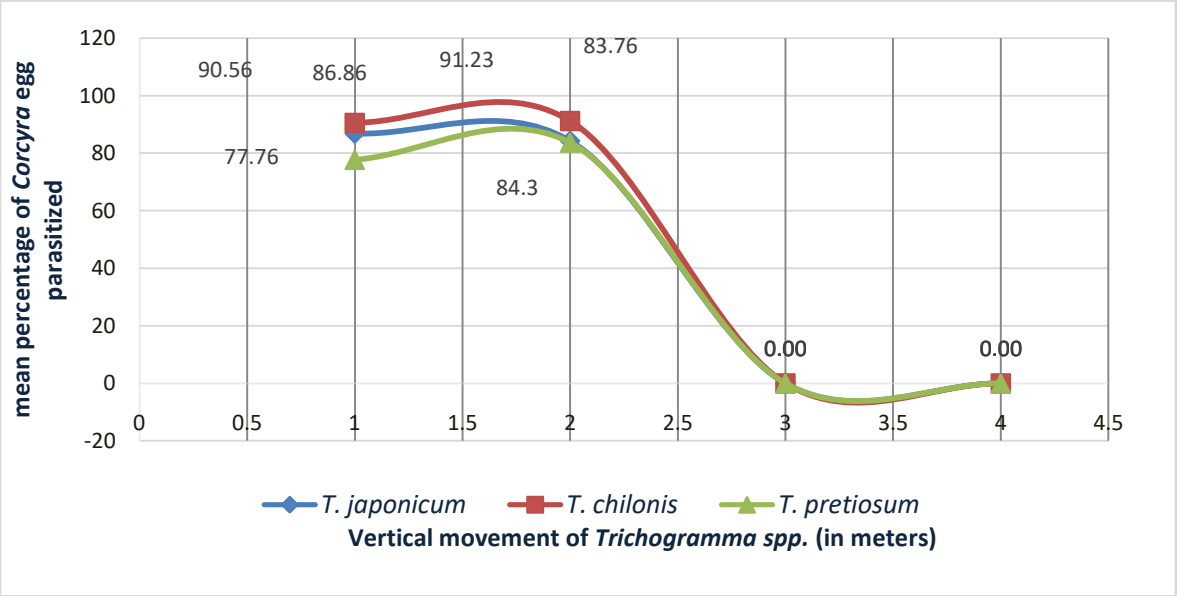



**Fig. 4.8 Mean percentage of *Corcyra* eggs parasitized by *Trichogramma* spp.through vertical movement of various lengths.**

The results from the experiment showed that, *T. japonicum*, *T. chilonis* and *T. pretiosum* travelled upto 2 meters vertically and the percentage of parasitization by different species of *Trichogramma*, differed significantly. Maximum percentage of parasitization was shown by *T. chilonis* (90.56%) followed by *T. japonicum* (86.86%) and *T. pretiosum* (77.76%). At 3m and 5m of distance no parasitization was observed.

## ii. Horizontal movement of *Trichogramma* spp.

**Table 4.4: Mean percentage of *Corcyra* eggs parasitized by *Trichogramma* spp.through horizontal movement of various lengths.**

| S. No | Horizontal movement (In Meters) | Mean percentage of *Corcyra* egg parasitized | | |
|---|---|---|---|---|
| | | *T. japonicum* | *T. chilonis* | *T. pretiosum* |
| 1 | 1m | 82.53 | 82.56 | 75.13 |
| | | (65.31) | (65.36) | (60.08) |
| 2 | 2m | 75.60 | 80.16 | 74.20 |
| | | (60.41) | (63.54) | (59.51) |
| 3 | 3m | 0.00 | 0.00 | 0.00 |
| | | (0) | (0) | (0) |
| 4 | 5m | 0.00 | 0.00 | 0.00 |
| | | (0) | (0) | (0) |
| **C.D.** | | 2.95 | 2.76 | 3.32 |
| **SE (m)±** | | 0.89 | 0.83 | 1.01 |
| **SE (d)** | | 1.26 | 1.18 | 1.41 |
| **C.V.** | | 4.91 | 4.49 | 5.81 |

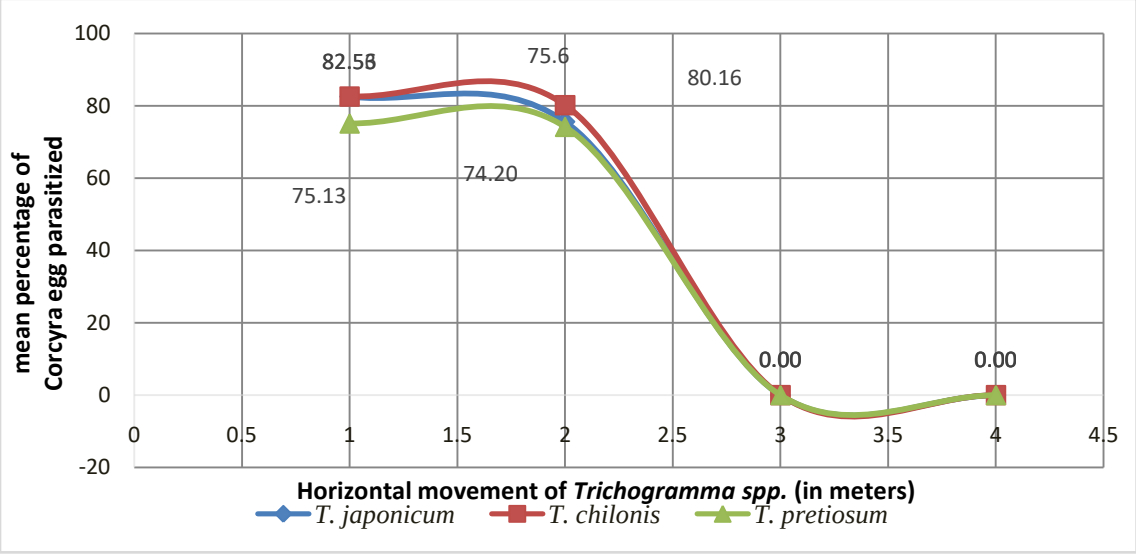



**Fig 4.9 Mean percentage of *Corcyra* eggs parasitized by *Trichogramma* spp.through horizontal movement of various lengths**

The results obtained from the experiment showed that *T. japonicum, T. chilonis* and *T. pretiosum* travelled upto 2 meters horizontally and the percentage of parasitization by different species of *Trichogramma,* differed significantly. Maximum percentage of parasitization was shown by *T. chilonis* (82.56%) followed by *T. japonicum* (82.53%) and *T. pretiosum* (75.13%). At 3m and 5m of distance no parasitization was observed.

Similar observations were made by Ranjith *et al*. (2018) who assessed the active movement of two Trichogrammatids viz., *Trichogramma chilonis* and *T. pretiosum* towards eggs of *Leucinodes orbonalis* and *Corcyra cephalonica* in 2cm diameter PVC tubes of 1, 2, 3, and 4 meters. The Trichogrammatids moved up to 3m towards the eggs of *L. orbonalis* and *C. cephalonica. T. chilonis* showed more active movement than *T. pretiosum*. The present findings are also in line with Chapman *et al*. (2009) also observed decrease in parasitism over increase in release distance in case of *T. ostriniae* on *Ostrinia nubilalis*. Similar result with the present studies were also reported by Sharma and Aggarwal (2015**)** stating that the dispersal or host searching capacity of *T. chilonis* and *T. japonicum* was negatively correlated with the distance between host eggs and parasitoid released point and parasitization rate was more near the release point (1-3m) with maximum parasitism at 1m by both the parasitoid species which is in total agreement with the present results.

### iii. Testing of the preferable movement of *Trichogramma* spp. between vertical and horizontal direction.

**Table 4.5: Preferable movement of *Trichogramma* spp. between vertical and horizontal direction.**

| S. No | Preferable movement (1meter) | *T. japonicum* | *T. chilonis* | *T. pretiosum* |
|---|---|---|---|---|
| 1 | Vertical | 44.67 (6.75) | 43.00 (6.63) | 42.33 (6.58) |
| 2 | Horizontal | 26.66 (5.25) | 29.00 (5.47) | 25.33 (5.13) |
| | CD | 0.619 | 0.479 | 0.348 |
| | SE (m) ± | 0.153 | 0.119 | 0.099 |
| | SE (d) | 0.217 | 0.168 | 0.139 |
| | CV | 4.425 | 3.397 | 2.917 |

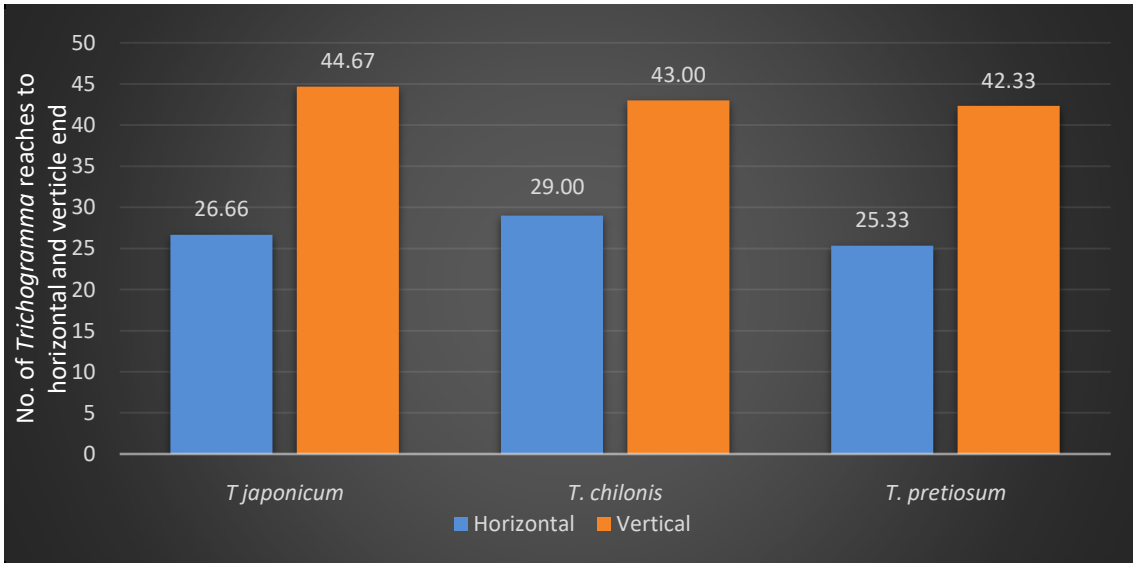



**Fig. 4.10 Preferable movement of *Trichogramma* spp. between vertical and horizontal direction**

This is the maiden attempt to find out the preferable movement by *Trichogramma* species in confined laboratory conditions (Temp- 25±2°C and RH-56±2%). Transparent PVC pipes of 0.7cm diameter were used for testing the movement of *Trichogramma*. Experiment was set as shown in figure 3.5. These pipes were cut of 1m length, one end of the pipe was fitted to a closed container A, having parasitized eggs of *Corcyra*, the other end was fitted vertically to plastic container 'B'. Another pipe was fitted horizontally to a plastic container 'C' as shown in fig. 3.5 having sentinel cards of *Corcyra* eggs. This experiment was conducted in the first week of December 2019 and observations were recorded after 6 days after the adult emerged from parasitized eggs. It was also observed that a single female *Trichogramma* was capable of parasitizing upto 10-150 eggs in her life span.

The mean percentages of *Corcyra* eggs parasitized by *Trichogramma* spp. were nearly equal in both the directions but the number of *Trichogramma* spp. which reached to the end of the vertical and horizontal pipes were different. During the present experiment, after 24 hour of emergence of *Trichogramma* spp., 40-48 numbers of *Trichogramma* were found in vertically fitted container but in the horizontal container the number was lesser and varied between 24-31.Thus, it can be concluded that vertical direction was more preferred than horizontal by *Trichogramma* spp.

### 4.3 To study the effect of UV treatment on the parasitization by *Trichogramma japonicum, T. chilonis, T. pretiosum* on lepidopteran eggs under laboratory conditions.

The influence of different exposure time of UV radiation on parasitized eggs of *Corcyra* by *Trichogramma japonicum, T. chilonis* and *T. pretiosum* was determined by exposing the respective trichocards to UV radiation at different duration of time from 5 minutes to 60 minutes. Ultra violet fluorescence analysis chamber was used; in which 2 UV tubes one of short wavelength (254nm) and other one, long wavelength (365nm) were used.

**Table 4.6: Influence of UV radiation on hatching percentage of parasitized eggs of *Corcyra cephalonica* by different species of *Trichogramma***

| S. No. | Duration of exposure of UV (in Mins) | Mean percentage of *Trichogamma* species emereged | | |
|---|---|---|---|---|
| | | ***T. japonicum*** | ***T. chilonis*** | ***T. pretiosum*** |
| 1 | 5.00 | 92.46 (74.15) | 90.95 (72.5) | 79.80 (63.32) |
| 2 | 10.00 | 15.73 (23.25) | 4.66 (10.99) | 0.00 (0.00) |
| 3 | 20.00 | 0.00 (0.00) | 0.00 (0.00) | 0.00 (0.00) |
| 4 | 30.00 | 0.00 (0.00) | 0.00 (0.00) | 0.00 (0.00) |
| 5 | 40.00 | 0.00 (0.00) | 0.00 (0.00) | 0.00 (0.00) |
| 6 | 50.00 | 0.00 (0.00) | 0.00 (0.00) | 0.00 (0.00) |
| 7 | 60.00 | 0.00 (0.00) | 0.00 (0.00) | 0.00 (0.00) |
| 8 | No treatment | 100.00 (90.00) | 100.00 (90.00) | 100.00 (90.00) |
| | **C.D.** | **2.457** | **5.588** | **1.591** |
| | **SE (m)±** | **0.813** | **1.848** | **0.526** |
| | **SE (d)** | **1.149** | **2.613** | **0.744** |
| | **C.V.** | **6.008** | **14.826** | **4.756** |

### 4.3.1Percentage of adult emerged from *Corcyra cephelonica* eggs parasitized by *Trichogramma japonicum* after exposure to UV rays.

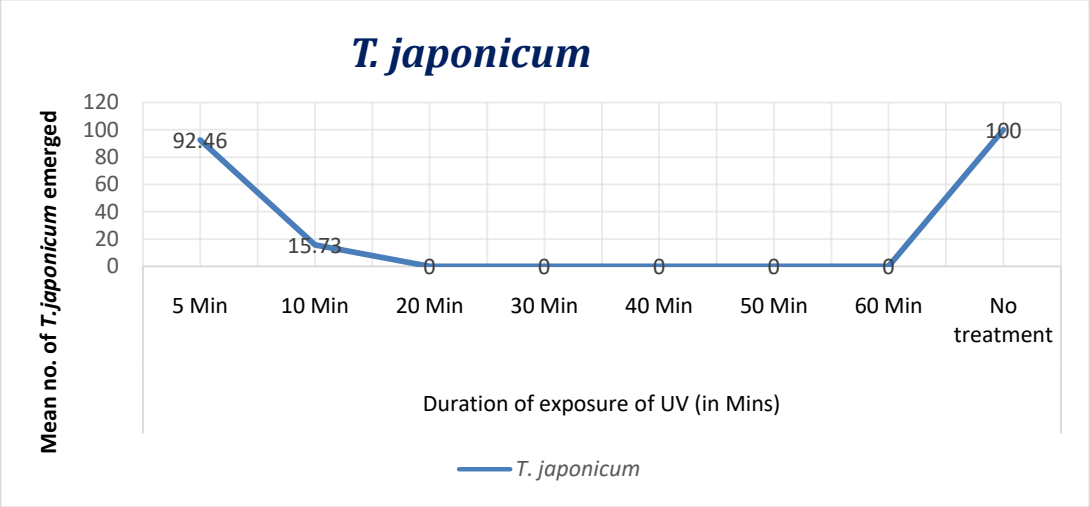



**Fig 4.11 Percentage of adult emerged from *Corcyra cephelonica* eggs parasitized by *Trichogramma japonicum* after exposure to UV rays**

The experiment was conducted in the year 2019-2020 in the mid of October.Data were recorded after 12 hours of adult emergence. The data presented in the table 2.3 showed that the percent of hatching of parasitized host eggs of *C. cephalonica* by *T. japonicum* differed significantly and decreased with the increase in the duration of time of exposure to UV radiation. In 5.00 minutes of exposure, maximum percent (92.46) of adult emergence was observed. In 10.00 minutes of exposure, the percent of emergence was very low (15.73) after which no emergence *i.e.* (0.00) percent was noted on increasing the duration of exposure further. In case of untreated control 100.00 percent of emergence was recorded.

Thus, from the data recorded from the present studies, it can be concluded that 20 minutes of UV radiation exposure was enough to kill the entire embryo inside the parasitized egg.

### 4.3.2 Percentage of adult emerged from *Corcyra cephelonica* eggs parasitized by *Trichogramma chilonis* after exposure to UV rays.

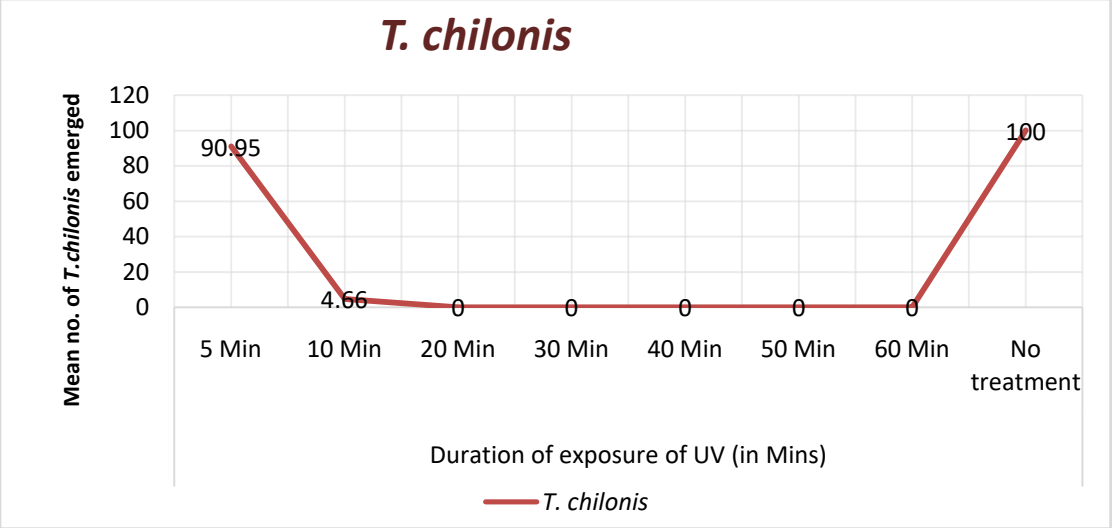



**Fig. 4.12 Percentage of adult emerged from *Corcyra cephelonica* eggs parasitized by *Trichogramma chilonis* after exposure to UV rays**

The experiment was done in the year 2019-2020 in the mid of October and the data were recorded after 12 hours of adult emergence. The data presented in the table 2.3 showed that the percent of hatching of parasitized host egg of *C. cephalonica* by *T. chilonis* differed significantly and decreased with the increase in the duration of time of exposure to UV radiation. In 5.00 minutes of exposure, maximum per cent (90.95) of adult emergence was observed. In 10.00 minutes of exposure, the percent of emergence was very low (4.66) after which no emergence *i.e*. (0.00) per cent was noted on increasing the duration of exposure further. In case of untreated control 100.00 per cent of emergence was recorded.

Thus, from the data recorded from the present studies, it can be concluded that 20 minutes of UV radiation exposure was enough to kill the entire embryo inside the parasitized eggs.

### 4.3.3 Percentage of adult emerged from *Corcyra cephelonica* eggs parasitized by *Trichogramma pretiosum* after exposure to UV rays.

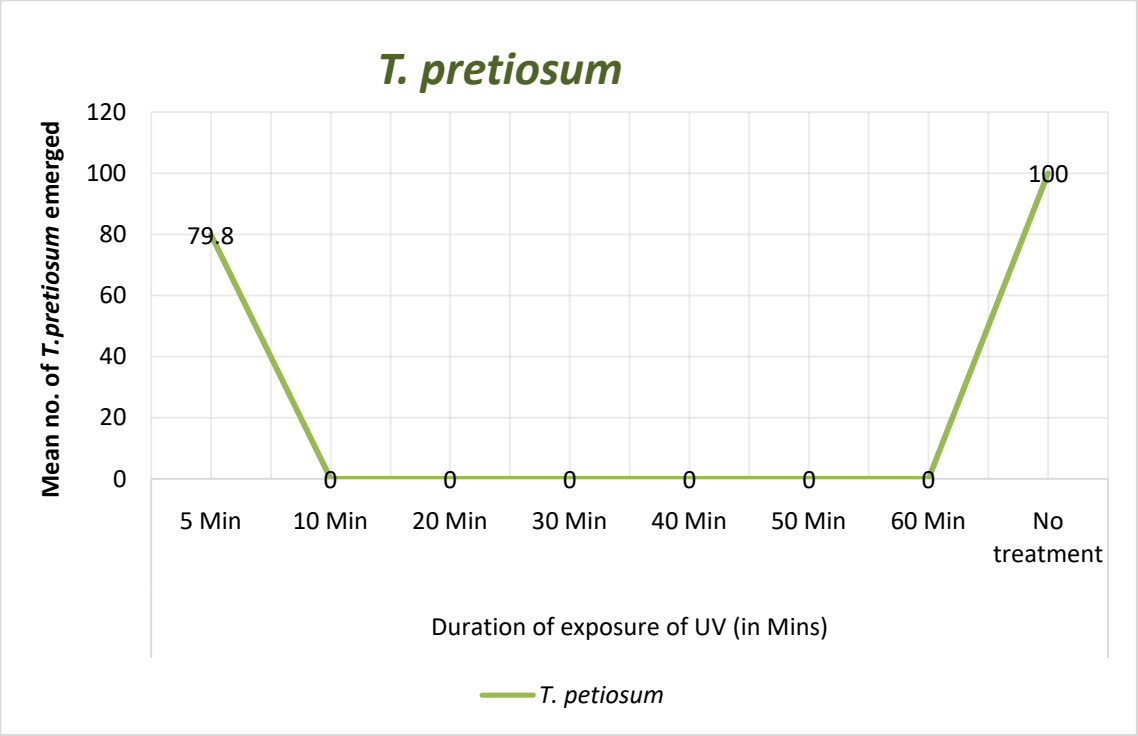



**Fig 4.13 Percentage of adult emerged from *Corcyra cephelonica* eggs parasitized by *Trichogramma pretiosum* after exposure to UV rays**

The experiment was conducted in the year 2019-2020 in the mid of October and the data were recorded in room temperature (26-27 $^{o}$ Celsius) after 12 hours of adult emergence. The data presented in the table 2.3 revealed that the percent of hatching of parasitized host eggs of *C. cephalonica* by *T. pretiosum* differed significantly and decreased with the increase in the duration of time of exposure to UV radiation. In 5.00 minutes of exposure, maximum (79.80) per cent of adult emergence was observed. After which no emergence *i.e.* (0.00) per cent was noted on increasing the duration of exposure further. In case of untreated control 100.00 per cent of emergence was recorded.

Thus, from the data recorded from the present studies, it can be concluded that 20 minutes of exposure to UV radiation was enough to kill the embryo inside the parasitized egg.

### 4.3.4 Comparison s of adult emergence of *Trichogramma* spp.from parasitized eggs of *C. cephalonica* treated with UV radiation

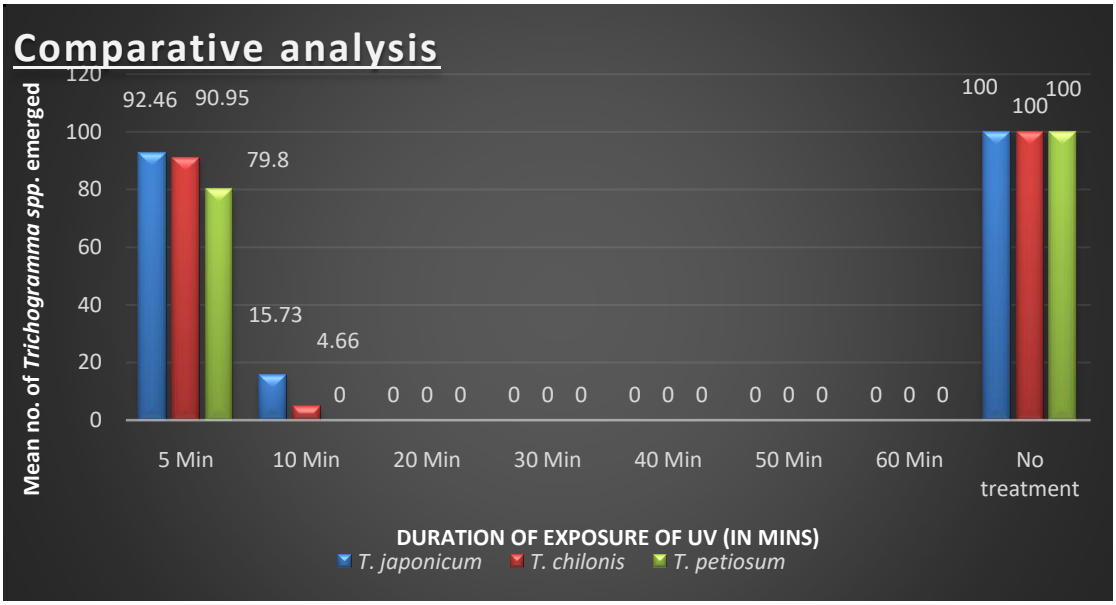



**Fig 4.14 Comparing adult emergence of *Trichogramma* spp.from parasitized eggs of *C. cephalonica* treated with UV radiation**

Comparison of percentage of adult emergence from host eggs of *C. cephalonica* presented in Fig: 4.14 by three species of Trichogrammids namely *T. japonicum, T. chilonis* and *T. pretiosum* revealed that, *T. japonicum* was found to be most tolerant to UV exposure, as maximum per cent of adult emergence (92.46 and 15.73), where as minimum per cent of (79.80 and 0.00) number of adult emergence were observed at 5 and 10 minutes of exposure respectively. In *Corcyra* eggs, an average duration of 20 minutes of exposure to UV, at distance of 15cm was enough to kill the embryo of *Trichogramma* spp. at room temperature (26-27° Celsius).

Comparison of all the three species of *Trichogramma* showed that the most vulnerable species was *T. pretiosum* in which all the embryos were killed within 10 minutes of exposure, followed by *T.chilonis* in which very low per cent of survival (4.66) was seen. Hence, the least effected species of *Trichogrmma* by UV radiation was *T. japonicum* where the maximun emergence was found.

Similar results were reported found by Faruki *et al.* (2007) stating a reduction in egg hatching of *C.cephalonica* of different ages exposed to UV irradiation. They irradiated the eggs with 254nm wavelength (UV-C) for different duration and observed decrease in the percentage of hatching of eggs in all age groups of eggs. No hatching occurred after 24 minutes of exposure of 2-3 days old egg but contradicts with Shinde *et al.* (2016) who correlated the exposure time of 45 minutes (treatment applied @ 42 cm height from the target site with 30 W UV lamp) and found suitable to irradiate the eggs of *C. cephalonica* by enhancing parasitoids effectiveness without any detrimental effect of the UV radiation (non ionizing) on *Trichogramma* under laboratory condition. However the distance of the target with the UV lamp may be the factor in latter.

In the present experiment, Ultra violet fluorescence analysis chamber was used which worked on 230 volts. There were 2 tubes fitted inside of the chamber for testing, sterilizing or killing the embryo of eggs; one ~~is~~ of short wavelength of 254 nm and other was of long wavelength of 365 nm. In sunlight UV light refers to the region of electromagnetic spectrum between visible light and x-rays falling between 400 to10 nm. There are mostly 3 types of UV radiation found in sunlight UVA, UVB and UVC. UVA contains 320-400nm which is of long wavelengths. These are least harmful to insects and their eggs. These are 20 times abundant than UVB reaching into the earth surface. UVB contains 290-320nm which is of short wavelengths. These are harmful to all living organism but 95% are absorbed in ozone. UVC contains 290-320nm which is extremely harmful but absorbed completely by earth atmosphere. According to Kyle *et al.* (2012) *Trichogramma* preferred to move toward higher intensities of UV-B radiation and parasitized more eggs in areas with higher UVB radiation.

However, higher UV-B radiation reduced the number of adult wasps emerging from host eggs. Insects are at high risk from UV radiation because of their small size; radiation may penetrate significantly deeper into insect tissues than into larger organisms.

If 20 minutes of exposure of UV (short wavelength of 254 nm and long wavelength of 365 nm) was given artificially to kill the embryo present in the host egg, it can be assumed that approximately 8 hours of sunshine is required to kill the embryo open areas. Hence, application of trichocards are recommended to be applied on the under surface of the leaves to avoid direct effect of sunlight.

# CHAPTER V

# SUMMARY AND CONCLUSION

The present experiment entitled **"Studies on the host range and ovipositional preference of *Trichogramm* spp. under laboratory conditions, at Raipur, Chhattisgarh"** was conducted in the Biological control laboratory of Department of Entomology, College of Agriculture, Indira Gandhi Krishi Vishwavidyalaya Raipur (Chhattisgarh) during the year 2019-20. Results obtained under different objectives are summarized and concluded below:

1. To study the host range and ovipositional preference of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions.

2. To study the vertical and horizontal movement of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions.

3. To study the effect of UV treatment on the parasitization by *Trichogramma japonicum, T. chilonis, T. pretiosum* on lepidopteran eggs under laboratory conditions.

## 5.1 To study the host range and ovipositional preference of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions.

Only lepidopteran insect were parasitized by all the three species of *Trichogramma,* no parasitization and emergence were recorded from eggs of insects belonging to Hemiptera and Coleoptera. Eggs of Arachnids (non-insect) were also not parasitized.

In *Trichogramma japonicum,* maximum number s of adults were emerged from a single egg of lemon butterfly, *Papilio demoleus* up to (3 adults) 2.66 followed by *Daphis nerii* (3 adults) 2.33 mean number of adults emerged from a single host egg.

In *Trichogramma chilonis*, maximum number of adult emerged were fromthe eggs of *Daphis nerii* with a mean of 7.66 from a single host egg, followed by *P. demoleus* (lemon butterfly) with a mean 4.66 from a single host egg.

In case of *Trichogramma pretiosum*, maximum number of adults were emerged from the eggs of *Daphis nerii* with a mean 6.67 followed by *P. demoleus* with an average of 4.33 from a single host egg.

### 5.1.1 Comparing the adult emergence in all the three species of *Trichogramma*

The results showed that maximum number of adult emergence after parasitization was in *T. chilonis i.e an* average number 7.66 followed by *Daphis nerii*, 6.67 while the minimum adult emergence of 0.33 was recorded in *T. japonicum* from a single host egg of *Spodoptera litura.* Thus, *Daphis nerii* and *Papilio demoleus* were most preffered host eggs by *T. chilonis* in which maximum adult emergence were observed , also *T. chilonis* and *T. pretiosum* could be used to control ~~in~~ most of the lepidopteran eggs.

### 5.1.2 Correlation between size of eggs and number of *T. japonicum, T. chilonis* and *T. pretiosum* emerged

The correlation studies between mean number of *T. japonicum, T.chilonis* and *T. pretiosum* emerged with the size of the eggs revealed statistically, highly significant positive correlation (r= 0.704), (r=0.898) and (r= 0.935) respectively.

## 5.2 To study the vertical and horizontal movement of *Trichogramma japonicum, T. chilonis, T. pretiosum* under laboratory conditions.

### 5.2.1 Testing the vertical and horizontal movement of *Trichogramma* spp.

Studies conducted to test the distance travelled by *Trichogragramma* spp. in transparent PVC pipe with diameter of 7mm under laboratory conditions resulted that all the three species *Trichogramma* travel upto 2 meter in both the direction vertically as well as horizontally with the help of the chemical cue released from the egg. It was

also observed that when the parasitized cards were placed without sentinel cards the *Trichogramma* spp. emerged from the parasitized card showed very less movement in the transparent PVC pipe.

### 5.2.2 Testing the preferable movement of *Trichogramma* spp. between vertical and horizontal direction.

Studies conducted to test the preferable movement of *Trichogramma* spp. between vertical and horizontal direction in transparent PVC pipe with diameter of 7mm under laboratory conditions revealed that vertical direction was more preferred than horizontal direction by *Trichogramma* spp.

### 5.3 To study the effect of UV treatment on the parasitization by *Trichogramma japonicum, T. chilonis, T. pretiosum* on lepidopteran eggs under laboratory conditions.

Experiment conducted to test the effect of UV treatment on embryos of three different species of *Trichogramma* in the host eggs of *Corcyra cephalonica* under laboratory condition exhibited that maximum (92.46 % and 15.73%) adults of *T. japonicum* emerged after exposure of short wavelength (254nm) and long wavelength (365nm) at 5 min and 10 min. while in *T. chilonis* it was (90.95% and 4.66%) where as in case of *T. pretiosum* it was (79.80% and 0.00%) respectively.

No emergence was found in 20 minutes and more than 20 minutes of exposure in UV of short wavelength (254nm) and long wavelength (365nm) where as 100% emergence found in untreated eggs.

Comparison of all the three species of *Trichogramma* tested revealed that, *T. japonicum* had the highest sustainability while *T. pretiosum* was the most vulnerable species towords exposure of UV radiation.

# CONCLUSION

- Only lepidopteran eggs were parasitized by all the three species of *Trichogramma*.
- No parasitization or emergences were recorded from eggs of insects belonging to Hemiptera and Coleoptera.
- Egg of Arachnids (non-insect) was also not parasitized.
- Mean maximum number of adult emergence after parasitization was observed in *T. chilonis* (7.66), followed by *T. pretiosum* ( 6.67) from a single host egg of *Daphis nerii*.
- Minimum (0.33) adults of *T.pretiosum* were emerged from a single host egg of *Spodoptera litura.*
- The correlation studies between mean number of *T. japonicum, T.chilonis* and *T. pretiosum* emerged revealed statistically significant positive correlation of (r= 0.704), (r= 0.898) and (r= 0.935) with the size of the eggs.
- Adults of *Trichogramma* spp. travelled upto 2 meters in both vertical as well as horizontal direction with the help of the chemical cues released from the eggs.
- It was also observed that when the parasitized cards were placed without sentinel cards *Trichogramma* spp. emerged from the parasitized card showed very less movement.
- It was observed that vertical direction was more preferred than horizontal direction by all the three *Trichogramma* spp. tested.
- Maximum per cent of adults of *T. japonicum* (92.46% and 15.73%) emerged after exposure of short wavelength (254nm) and long wavelength (365nm) for 5 and 10 mins. followed by *T. chilonis* (90.95% and 4.66%.) and minimum in *T. pretiosum*(79.80% and 0.00 %) respectively.
- No emergence was found in 20 minutes and more than 20 minutes of exposure in UV of short wavelength (254nm) and other one, long wavelength (365nm). 100% emergence found in untreated eggs.

- Comparison of all three three species of *Trichogramma*, revealed that *T. japonicum* had the highest sustainability while *T. pretiosum* was found to be the most vulnerable species towards exposure of UV radiation.

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# RESUME

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