

EFFECTS OF CARBOSULFAN ON THE BIOLOGY OF BIRD CHERRY OAT APHID

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Abstract: Aphids are the most commonly occurring, destructive, sap sucking and serious threat to cereal crops especially wheat (*Triticum aestivum*). Bird Cherry Oat aphid *Rhopalosiphum padi* (L.), is one of the most important aphids on *T. aestivum* which is one of the most consumed food and a source of nutrition in Pakistan. It causes considerable yield loss in wheat. Carbosulfan, a carbamate, is of the most commonly used pesticide against *R. padi*. The effects of Carbosulfan on generations of *R. padi* were performed under standard lab conditions by exposing adult aphids to three different concentrations (1.4×10^{-7} ppm, 1.4×10^{-10} ppm, 1.4×10^{-13} ppm) of Carbosulfan (Advantage® EC). Based on the results, all three concentrations noticeably reduced the pre-adult survival rate. 1.4×10^{-13} significantly extended the development duration of 1st instar, 2nd instar and 3rd instar nymphs. 1.4×10^{-13} ppm also extended the total pre-adult period and female longevity of *R. padi*. The total longevity was increased by 1.4×10^{-10} ppm. However, the fecundity decreased the most at 1.4×10^{-10} ppm. While the TPRP and APRP increased the most at 1.4×10^{-13} ppm. In the life table parameters, both the intrinsic rate of increase (r) and the finite rate of increase (λ) decreased at 1.4×10^{-7} ppm and 1.4×10^{-10} ppm, as well as the net reproductive rate (R_0) also decreased at 1.4×10^{-7} ppm and 1.4×10^{-10} ppm, while mean generation time (T) showed increase at 1.4×10^{-13} ppm. Thus, at the concentrations of Carbosulfan tested here, there were negative impacts on *R. padi* fitness and biology by decreased pre-adult survival rate, λ , r , and R_0 . The concentrations also slowed down the development of some stages and extended *T*. My results would be helpful in assessing the overall effects of Carbosulfan on *R. padi* and should be taken into consideration when use Carbosulfan as a seed dressing insecticide for management of *R. padi* in wheat crop.

Keywords: Bird cherry oat aphid, *Rhopalosiphum padi*, *Triticum aestivum*, cereals, Carbosulfan

Introduction

Wheat is a staple food in Pakistan satisfying 80% of dietary needs with 38% share in caloric intake. Pakistan ranks eighth in terms of production in the world and is presently reaping production of over 25 million tons due to consolidated efforts of the farmers, policymakers and scientists. Over 152 varieties of wheat have been launched in Pakistan so far and a significant number of share from Punjab in the last decade; Punjab (69), Sindh (25), KP (44), Baluchistan (8), and PARC (6), respectively (Ahmad *et al.*, 2017). Bird cherry-oat aphid, *R. padi*, is the most implicated aphid in the transmission of the barley yellow dwarf virus, among cereal aphids (Watson and Mulligan, 1960) which can dangerously reduce the yield of cereals (Watson, 1959).

Bird cherry oat aphid *R. padi* (L.), is one of the most financially important aphids on wheat (Akhtar *et al.*, 2007). It is widely distributed over Europe, Asia, Africa and other countries such as Mexico, Canada,

Australia, Brazil and Uruguay (CABI, 2019). Aphids are nearly transparent sucking insects with soft bodies. Aphids can induce premature death of leaves and yellowing when they are present in sufficient numbers. They secrete drops of sugary liquid known as "honeydew", which encourages the development of sooty molds on the leaves and causes tiny scorch. The life cycle of aphids involves wingless (apterous), sexual, winged (alates), and asexual forms. The females of most aphid species reproduce asexually (without being fertilized) when it feeds on cereals, giving rise to nymphs rather than eggs (Prescott *et al.*, 1986). Attack of aphids also reduce canopy dry matter. Infestation of aphids also causes continuous yellowing in wheat from tillering stage to the end of flowering stage. Yellowing symptoms can be observed at the flower stage in wheat when aphids feed for seven days (Roza-Gomes, 2008). *R. padi* damages plants both by removing their sap and by transmitting plant viruses (Close and Lamb, 1961; Greene, 1966; Harper and Blakeley, 1968). Usually

aphid feeding causes poor root growth and a decrease in tiller number but in spring heavy infestations can kill young plants (Russell, 1978). They can reduce grain yield, thousand-kernel mass, and quality also (Rautapaa, 1968). Feeding during the seedling (2-leaf) stage causes largest reductions in yield causing a yield loss upto 40-60%, with mean densities of 10-20 aphids/tiller (Kantack and Kieckhefer, 1979). Plant physiology is affected in different ways by sieve diversion by numerous aphids, depending on the plant growth stage at time of infestation. The most common reaction in fruit trees is twig stunting due to reduced growth. Bad fructification in flowers is also a result of aphid feeding (flower abortion). Fruits can be smaller if aphids' colonies develop later. Most of these injuries by aphids are 'asymptomatic' (Quisenberry and Ni 2007). Carbosulfan has been observed to significantly lower the population of *R. padi* on wheat (Faheem *et al.*, 2016). Carbosulfan has also been shown to cause highest population reduction when compared with other insecticides such as imidacloprid, acetamiprid and thiamethoxam etc. (Ahmad *et al.*, 2017). Hence, this approach was adopted to study the population responses of *R. padi* when exposed to Carbosulfan. The main objective of the current study was to gain a better understanding about how very low concentrations of Carbosulfan affect the development, reproduction, and survival of *R. padi*.

Materials and methods

Insects

R. padi adults were collected from wheat fields at Ayub Agricultural Research Institute (coordinates), Faisalabad, Pakistan. Collection sites were selected based on insecticide usage history, seed dressing history and varietal differences. Field collected aphids before exposing, were kept in laboratory for few hours to attain homogeneity and to ensure pathogen and ectoparasites free culture.

Insecticide

Carbosulfan (Advantage® 20EC) by FMC® was used in the experiment.

Bioassay

Leaf dip method was used for bioassay with some modifications (Sawicki and Rice 1978). Insecticide solution was prepared by taking 5 µL amount of insecticide from insecticide bottle and added in 250ml water to attain required volume. Serial dilution was done to attain required concentrations. Experimental design CRD was adapted under controlled lab conditions: $23 \pm 1^\circ\text{C}$, $60 \pm 10\%$ relative humidity (RH). There were three concentrations, every concentration had three replicates. Fresh leaves were collected from wheat field at AARI, washed and cleaned. After that leaves

were cut in smaller sizes, dipped into insecticide solution and left to dry for 1-2 hours. The dried leaves were placed into Petri plates on filter papers. 30 insects were exposed at every concentration (1.4×10^{-7} ppm, 1.4×10^{-10} ppm, 1.4×10^{-13} ppm). Data was collected after every 24 hours till 8-10 days. Every specimen of adult aphids and their newly young ones were separated after 24 hours, one specimen was kept into one petri plate to check proper effects of the insecticide. The aphids were observed under microscope, the live aphids were given fresh food and the dead aphids were disposed of.

Analysis

The basic life parameter such as age-specific fecundity (m_x), age-specific survival rate (l_x), age-specific maternity ($l_x m_x$), intrinsic rate of increase (r), finite rate of increase (λ), mean generation time (T), net reproductive rate (R_0), TPRP and APRP were calculated using the computer program TWOSEX-MS Chart (Chi, 2017).

Results

According to biological parameters of *R. padi* (Table 1), 1.4×10^{-13} ppm could significantly increase the survival period of 1st instar nymphs (N1), 2nd instar (N2) nymphs and 4th instar (N4) nymphs. In contrast 1.4×10^{-10} ppm significantly increased the survival period of 3rd nymph instar (N3). Total pre adult period increased significantly in 1.4×10^{-13} ppm. There was significant increase in pre-adult survival rate and the female longevity at 1.4×10^{-13} ppm. However, total longevity rose more at 1.4×10^{-10} ppm (5.06 ± 0.46) as compared to 1.4×10^{-7} ppm (4.09 ± 0.53) and 1.4×10^{-13} ppm (4.80 ± 0.73). According to (Table 2), fecundity (9.45 ± 1.99) was significantly increased at 1.4×10^{-13} ppm. The Total pre reproductive period (TPRP) and the Adult pre reproductive period (APRP) increased the most at 1.4×10^{-13} ppm. However, there was a significant decrease in Total pre reproductive period (TPRP) (6.78 ± 0.28) and Adult pre reproductive period (APRP) (0.11 ± 0.11) at 1.4×10^{-7} ppm. The reproductive duration of female aphids increased the most at 1.4×10^{-7} ppm. According to the life table parameters of *R. padi* (Table 3), the mean generation time (T) at 1.4×10^{-13} ppm (10.61 ± 0.48) was longer as compared to 1.4×10^{-7} ppm (9.15 ± 0.92) and 1.4×10^{-10} ppm (9.37 ± 1.39). Net reproductive rate (R_0) was significantly decreased by 1.4×10^{-7} ppm (1.66 ± 0.55) and 1.4×10^{-10} ppm (1.18 ± 0.33) in comparison to 1.4×10^{-13} ppm (2.6 ± 0.84). The intrinsic rate of increase (r) and Finite rate of increase (λ) of aphids at 1.4×10^{-7} ppm and at 1.4×10^{-10} ppm was significantly reduced as compared to 1.4×10^{-13} ppm where they increased distinctively.

Table 1: Effect of different concentrations of carbosulfan on preadult developmental duration, preadult survival and longevity of *R. padi*

Parameter (days)	n	1.4×10^{-7} ppm	N	1.4×10^{-10} ppm	n	1.4×10^{-13} ppm
N ₁	19	1.89±0.2	30	1.83±0.13	17	1.94±0.16
N ₂	14	1.93±0.13	20	1.7±0.15	12	2.00±0.25
N ₃	9	1.56±0.18	15	2.07±0.15	11	1.73±0.24
N ₄	9	1.67±0.33	11	1.55±0.21	11	2.36±0.20
Total pre-adult	9	6.67±0.29	11	6.91±0.16	11	8.27±0.24
Pre-adult survival		0.2±0.05		0.22±0.05		0.27±0.07
Female longevity	9	10.22±0.62	11	10.27±0.3	11	12±1.26
Total longevity	45	4.09±0.53	50	5.06±0.46	40	4.80±0.73

Where= N₁-N₄ indicates the 1st to 4th instar nymphs of *R. padi*

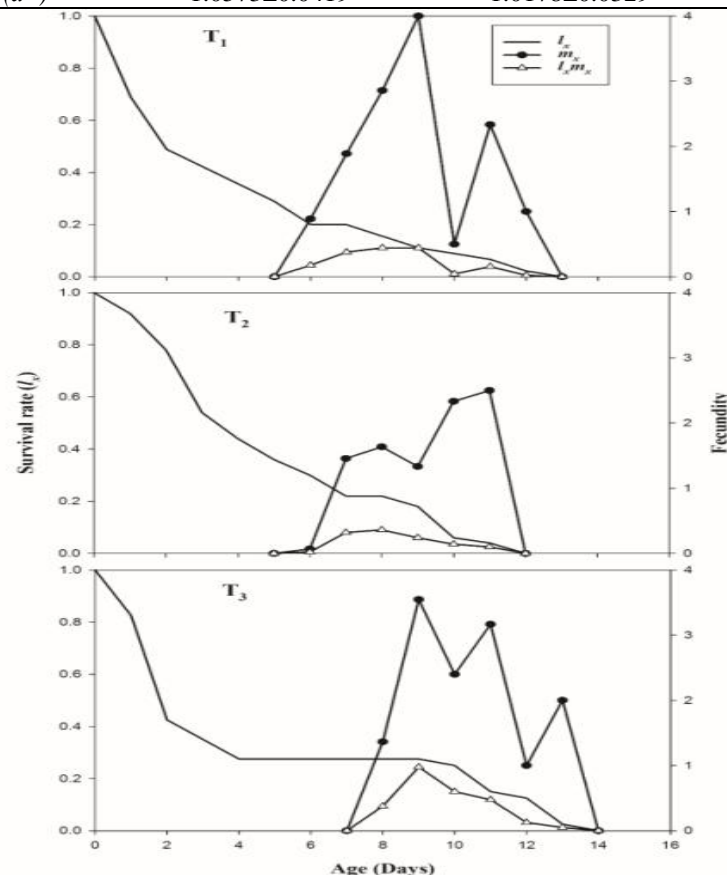
Table 2: Effect of different concentrations of carbosulfan on fecundity and reproduction of *R. padi*

Parameter	1.4×10^{-7} ppm	1.4×10^{-10} ppm	1.4×10^{-13} ppm
Fecundity (nymph/female)	8.33±1.3	5.36±0.56	9.45±1.99
TPRP	6.78±0.28	7.27±0.24	8.73±0.27
APRP	0.11±0.11	0.36±0.15	0.45±0.16
Reproductive duration	3.22±0.32	2.91±0.28	3.18±0.35

Where, TPRP= Total pre-reproductive period, APRP= Adult pre-reproductive period

Table 3: Effect of different concentrations of carbosulfan on life table parameters of *R. padi*

Parameter	1.4×10^{-7} ppm	1.4×10^{-10} ppm	1.4×10^{-13} ppm
Mean generation time T (day)	9.15±0.92	9.37±1.39	10.61±0.48
Net reproductive rate R_0	1.66±0.55	1.18±0.33	2.6±0.84
(offspring/individual)			
Intrinsic rate of increase r (d^{-1})	0.0557±0.0407	0.0176±0.0329	0.0900±0.0331
Finite rate of increase λ (d^{-1})	1.0573±0.0419	1.0178±0.0329	1.0942±0.0357

**Figure 1: Age specific survival rate (l_x), age-specific fecundity (m_x) and net maternity ($l_x m_x$) of *R. padi* were tested on different concentrations of carbosulfan**

The age-specific fecundity, age-specific survival rate and net maternity for each of the three treatments are shown in Fig. 1. At concentration 1.4×10^{-7} ppm there was significant decrease in the age specific survival rate (l_x), on the other hand the age-specific fecundity rate (m_x) showed some fluctuation as it increased at one point but dropped and then increased again. The age specific maternity ($l_x m_x$) increased uniformly for some time but then started to fluctuate. At concentration 1.4×10^{-11} ppm age-specific survival rate (l_x) of *R. padi* decreased with the passage of time and age-specific fecundity (m_x) rate showed minor fluctuations and did not change drastically. The age specific maternity ($l_x m_x$) displayed a flat curve. At concentration 1.4×10^{-14} ppm age-specific survival rate (l_x) of the aphids decreased but then became constant for some time and then slowly decreased again, while the age-specific fecundity (m_x) rate showed large fluctuations. The age-specific maternity ($l_x m_x$) increased drastically and then decreased periodically (Fig.1).

Discussion

Carbosulfan [2,3-dihydro-2,2-dimethylbenzofuran-7-yl (dibutylaminothio) methylcarbamate] is a broad-spectrum carbamate pesticide which inhibits the activity of acetylcholinesterase. It is used to control mites, insects and nematodes by foliar, seed and soil treatment applications, foliar pests can be controlled via systemic action by soil applications, and through direct contact or stomach ingestion is said to be effective (FAO/WHO 1984), 1984). Carbosulfan (Advantage 20EC) acts as the most effective insecticide for the control of aphids on Wheat. The present study was to check the severity of effects of various concentrations of carbosulfan (advantage 20EC) on *R. padi* in terms of the fecundity rate, survival rate, adult longevity and age specific maternity.

My results of bioassay in laboratory showed significant differences in mortality and fecundity of *R. padi* feeding on the plants. Total pre adult period and the preadult survival showed maximum increase at 1.4×10^{-13} ppm. The female longevity and the total longevity increased the most at 1.4×10^{-13} ppm. The nymph instars exposed to 1.4×10^{-7} ppm and 1.4×10^{-10} ppm showed lower values of TPRP and APRP. At lower concentration (1.4×10^{-13} ppm) net reproductive rate (R_0) is more as compared to other two concentrations. Effect on biology with respect to fecundity (5.36 ± 0.56) was recorded highest in exposure to concentration (1.4×10^{-10} ppm) at it decreased the most at that concentration. In life table parameters measurement, it was noted that intrinsic rate of increase and finite rate of increase were highest at 3rd concentration (1.4×10^{-13} ppm) in the larval life span. The mean generation time (T) and the

net reproductive rate (R_0) also increased at 1.4×10^{-13} ppm. Torres and Ruberson (2004) support my results that, all nymph instars (N1, N2, N3, N4) are affected more at higher concentration and their survival rate is more at lower. All three concentrations used in experiment had significant impact on aphid population when compared with control plot. Maximum population reduction was observed in T2 with application of Advantage 20 EC @ 300 ml/acre and proved to be more effective insecticide against the canola aphid, *L. erysimi* Kalt. as compared to other insecticides succeeded by T1 with Actara 25 WP @ 24 g/acre, T3 with Confidor 200 SL @ 150 ml/acre and T4 with Mospilan 20 SP @ 80 g/acre (Ahmad *et al.*, 2017). Sublethal concentrations of insecticides like carbofuran and bifenthrin are shown to have negative effects on aphid's population growth and biology as the concentrations increase (Kerns and Stewart 2000). Calafiori *et al.* (1999) reported that carbosulfan @ 0.6 L ha⁻¹ provided more than 80% control of aphids and thrip in cotton. Chinnaiah and Asif (1999) reported that carbosulfan @ 25 g a.i kg⁻¹ seed reduced the number of sucking pests of cotton such as leafhoppers and aphids after sowing up to 45 days. Moreover, *R. padi* was more susceptible to all insecticides than *S. avenae* and *S. graminum* and all the insecticides were more effective against *R. padi* than *S. avenae* and *S. graminum*. However, malathion, bifenthrin, imidacloprid, pymetrozine, carbosulfan and thiamethoxam significantly decreased the population of *R. padi* (Faheem *et al.*, 2016). When *R. padi* aphids fed on leaves of wheat treated with sublethal concentrations of insecticides, their xylem feeding was reduced. Moreover, the feeding on sublethal concentrations also reduced their body water contents and at reproductive maturity the aphids were small, light in weight and has less fecundity. The honeydew secretion was also affected negatively (Daniels *et al.*, 2009). Amer and his colleagues tested toxicity of different conventional and neonicotinoid insecticides such as bifenthrin, carbosulfan, methamidophos, thiamethoxam and imidacloprid on aphids and there was a clear difference in effected aphids and control plots (Amer *et al.*, 2010). When generations of *R. padi* were exposed to sublethal doses of insecticides it was noted that their, both fecundity and longevity were significantly decreased after exposure (Lu *et al.*, 2016). This study will be helpful to understand how Carbosulfan changes the biology of aphids at low concentrations and the susceptibility of nymphs of aphids to lower concentrations of insecticide.

Conflict of interest

The author declared that they have no conflicts of interest to this interest. The authors declare that they do not have any associative or commercial interest

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that represents a conflict of interest in connection with the work submitted.

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