Introduction:

Line 58: change Bemisia tabaci x Bemisia tabaci

Line 66: increase "Similarly" Trialeurodes vaporariorum, commonly......

Line 76: increase "resistance in pest populations"

Line 93-94: "In a recent study, *O. majusculus* and *O. laevigatus* were reported as potential natural enemies of *B. tabaci* eggs, nymphs, and adults" reference not very recent (2008), Update.

Line 110: change "......different populations of preyx".... different densities and populations of preys"

Material and Methods

Line 130: increase environmental conditions

Line 144, 147: contradictions "...feed 48 h..." and "......After 24 h, predators were removed..."

Line 197: contradiction with line 144

Results

Line 237: is mentioned "...... that the predator is more efficient at finding whitefly nymphs at low prey densities" however, four prey is the minimum provided. What would happen if you provide 3 prey? Eat all by efficient or by absence of preys? Support their affirmation.

Line 266: change "against" by "when fed of whitefly species"

Line 267: check the table number "table 4" correspond to "developmental characteristics"

Line 268: specific what is N

Line 271: insert the table or fig where is the information (fig 2???)

Line 273: insert the table or fig where is the information (table 5??)

Line 273-275: was nor difference statistic, why is mentioned "highest" and "shorter"

Line 281: change "table 5" by "table 6"

Line 281: delete "respectively"

Line 286: increase "table 6"

Line 287-289: was not difference are similar

Line 292: change "table 6" by" table 7"

Line 295-297: was not difference are similar

Line 299: change "table 6" by "table 7"

Line 301: increase "The" before "Figure"

Table 3: change "a" by "α" in attack rate

Table 3: increase "SD" or "CI"

The fig 3. Change *B.tabaci* by *B. tabaci*

Discussion

Line 338-342: is not necessary (text best in introduction)

Line 351: increase ")"

Line 430: delete "table 4"

Line 445: delete "s" in "factors"

Increase in the discussion support of fig 2 (rate survival when fed different preys) and fig 5 (the fig. showed interest results)

References

Line 469,470: italic scientific name

Line 473: italic scientific name

Line 487,488: italic scientific name

Line 467-756: The scientific names are not in italic in all the references.

Line 503: cite as book or chapter book

Line 512-514: cite with vol and pages

Line 526: Journal with uppercase the first letters

Line 530: Journal with uppercase the first letters

Line 535: Journal with uppercase the first letters

Line 543: Journal with uppercase the first letters

Line 556: Journal with uppercase the first letters

Line 576,577: cite as book or chapter book

Line 586,587: Standardize Cs or C

Line 503: cite as paper, with journal, vol, pages

Line 625: Journal with uppercase the first letters

Line 637: Journal with uppercase the first letters

Line 657: Author with lowercase

Line 670: Journal with uppercase the first letters

Line 672: Journal with uppercase the first letters

Line 687,688: Journal with uppercase the first letters

Line 689,690: cite as book or chapter book

Line 512-514: cite ad journal with vol and pages

Line 698: title is with lowercase except the first letter

Line 702: cite as chapter book

Line 703: Journal with uppercase the first letters

Line 709: Journal with uppercase the first letters

Line 717: Journal with uppercase the first letters

Line 718: cite as chapter book

Line 724: Journal with uppercase the first letters

Line 727: Journal with uppercase the first letters

Line 737: Journal with uppercase the first letters

Line 743: Journal with uppercase the first letters

Line 751: Journal with uppercase the first letters

Line 756: Journal name complete

Bemisia tabaci and Trialeurodes vaporariorum as determined by age-stage, two-sex 2 life table 3 Running Title: development and survival of O. similis fed on two whitefly species 4 5 Shakeel Ur Rehman¹, Xingmiao Zhou^{1*}, Shahzaib Ali¹, Muhammad Asim Rasheed¹, Yasir Islam¹ and Muhammad Hafeez² 6 7 1. Hubei Insect Resources Utilization and Sustainable Pest Management Key Laboratory, 8 College of Plant Science and Technology, Huazhong Agricultural University, Wuhan 9 430070, P. R. China 10 11 2. State Key Laboratory Breeding Base for Zhejiang Sustainable Pest and Disease Control, 12 Institute of Plant Protection and Microbiology, Zhejiang Academy of Agricultural 13 14 Sciences, Hangzhou, Zhejiang, China. 15 *Corresponding Author: Xingmiao Zhou; Email: xmzhou@mail.hzau.edu.cn 16 First author: Shakeel Ur Rehman; Email: Shakeel.entomologist@hotmail.com 17 18 Co-authors Email address: 19 20 shahzaibali@webmail.hzau.edu.cn asim_ento@outlook.com 21 22 yasir_islam@outlook.com hafeez_203@yahoo.com 23 24

Predatory functional response and fitness parameters of Orius similis when fed

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Abstract

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Background: The polyphagous predatory bug Orius similis Zheng is an active predator used to 28 control thrips and aphids. The whitefly species Bemisia tabaci and Trialeurodes vaporariorum 29 30 are voracious pests of different economic agricultural crops and vegetables. Method: In this study, Holling disc equation and age-stage, two-sex life table technique was 31 used to investigate the functional response and biological traits of third instar nymphs and adult 32 female O. similis when presented third instar nymphs of both whitefly species as prey. 33 **Results:** The results showed a type II functional response for each life stage of *O. similis* when 34 35 fed each whitefly species. The calculated prey handling time for different O. similis life stages were shorter when fed T. vaporariorum than when fed B. tabaci nymphs. In contrast, the 36 nymphal development of O. similis was significantly shorter when fed B. tabaci than T. 37 38 vaporariorum nymphs. Additionally, the total pre-oviposition period of adult females was 39 statistically shorter when fed B. tabaci nymphs than T. vaporariorum nymphs. Furthermore, the 40 survival rates and total fecundity of O. similis were higher when fed B. tabaci than T. vaporariorum. There were no significant differences in any population parameters of O. similis 41 42 when fed either whitefly species. These results show that O. similis could survive and maintain 43 its populations on both species of whitefly and could therefore serve as a biological control agent in integrated pest management (IPM). 44

Key words: Orius similis (Anthocoridae), functional response, biological traits, predation,

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whitefly species, biological control.

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Introduction

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Invasive insect pests can significantly disturb native insect communities and cause considerable 50 damage to agriculture and forests (Pimentel et al. 2000). Among these pests, whiteflies 51 52 (Hemiptera: Aleyrodidae) are the most damaging insect pests to agricultural crops globally, 53 including China where more than 1450 species are known (Anderson et al. 2004; Lapidot et al. 54 2014; Martin et al. 2000). Included in these species are the silver-leaf whitefly (*Bemisia tabaci*) 55 and greenhouse whitefly (Trialeurodes vaporariorum), which are generally considered responsible for major economic losses. However, B. tabaci is thought to be a complex of 56 57 morphologically indistinguishable sibling species, referred to as different biotypes (Wraight et al. 2017). Bemisia tabaci is considered to be one of the most significant plant pests and colonizes 58 over 600 host plants, causing significant damage (Polston et al. 2014). Bemisia tabaci is 59 distributed globally (De Barro et al. 2005; De Barro 1995; De Barro et al. 2000) and causes 60 significant damage to crop yield and quality by feeding on plant phloem and secreting honeydew 61 that stimulates the rapid growth of molds (Colvin et al. 2006; Prijović et al. 2013). However, the 62 63 most significant problem associated with outbreaks of B. tabaci is the transmission of plant 64 viruses (Navas-Castillo et al. 2011). Bemisia tabaci has been reported as the vector for the transmission of over 300 viral species in major economically important agricultural and 65 vegetables crops (Gilbertson et al. 2015). Similarly Trialeurodes vaporariorum, commonly 66 67 known as greenhouse whitefly, is also considered an important pest of vegetable and agricultural 68 crops but transmits fewer viruses than does B. tabaci (Brown 2007; Jones 2003; López et al. 69 2012; Navas-Castillo et al. 2011; Wisler et al. 1998). However, because of its short life cycle, this species has been considered a more prevalent insect pest in greenhouse (Simmonds et al. 70 71 2002). Trialeurodes vaporariorum can also adapt to cold climates better than B. tabaci does and

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is common at high elevations (Barboza et al. 2019). Because of its resistance to insecticides such as neonicotinoids, *T. vaporariorum* has received much research attention (Gorman et al. 2007; Karatolos et al. 2011).

Extensive use of pesticides not only causes environmental contamination and ozone layer depletion, but also creates serious health problems in mammals, resistance in pest populations and creates toxic conditions for beneficial insect species (Shaaya et al. 1997; Yoza et al. 2005). Hence, to reduce insecticide use, biological control methods such as use of natural predators, resistant varieties, and plants extracts are important in controlling pests in modern integrated pest management (IPM) programs (Asare-Bediako et al. 2014; Kageyama et al. 2010; Yang et al. 2012; Yazdani & Zarabi 2011).

The genus *Orius* (Hemiptera: Anthocoridae) is the largest group of flower bugs, containing around eighty species globally. They are polyphagous predators of small and softbodied insects considered pests in agriculture and forestry, including spider mites, aphids, thrips, and whiteflies in protected and open-field crops within its native range of Asia (Arnó et al. 2008; Carpintero 2002; Hernández 1999; Herring 1966; Postle et al. 2001; Yamada et al. 2016; Zhao et al. 2017). The artificial introduction of *O. sauteri* as a biological control agent provides potential control against small insect pests on pepper and eggplant, especially under greenhouse conditions (Jiang et al. 2011; Yin et al. 2013). *Orius laevigatus* is an effective biological control species in Europe and is widely used in augmentative release programs (Van Lenteren & Bueno 2003). Studies have been conducted into predation by *O. albidipennis*, *O. insidiosus*, *O. majusculus*, and *O. niger* on different prey species (Fritsche & Tamo 2000; Rutledge & O'Neil 2005; Tommasini et al. 2004). In a recent study, *O. majusculus* and *O. laevigatus* were reported as potential natural enemies of *B. tabaci* eggs, nymphs, and adults (Arnó et al. 2008).

Comentado [Luis1]: 10.1007/s10340-020-01210-0

Comentado [Luis2]: Recent 12 year??

Comentado [Luis3]: Not very recent (12 year)

Orius similis Zheng is a common and effective natural enemy present in cultivated fields in China and is used as a biological control agent against many small pests of economically important agricultural crops (Zhang et al. 2012). Both pre-adult and adult stages prey on lepidopteran insects including eggs or newly hatched larvae of Pectinophora gossypiella, Anomis flava, and Helicoverpa armigera as well as Frankliniella formosae, Aphis gossypii, and Tetranychus cinnabarinus. Large populations of O. similis in cotton fields are useful as biological control agents (Zhou & Lei 2002). This Anthocorid species has many features that make it a good biological control agent, such as high searching efficiency, the ability to increase population levels with outbreaks coinciding with prey density, and an aptitude to aggregate in regions of high prey populations (Hodgson & Aveling 1988). Mass rearing of O. similis and the subsequent augmentative release into crop fields leads to the control of many insect pests, decreasing their populations and hence reducing the use of pesticides (Bonte & De Clercq 2011; Tommasini et al. 2004). However, it is important to estimate the effectiveness of a predator before using it in an integrated pest program (Fathipour et al. 2006).

The potential of a predator to control a pest depends upon its functional response to different populations of prey (Butt & Xaaceph 2015). Therefore, the efficiency of a predator can be assessed by its functional response (i.e., changes in attack rate in response to variations of prey populations and number of prey consumed per unit time in relation to prey density (Riechert & Harp 1987). Four types of functional response have been defined based on the predation rate of a predator as a function of prey density: type I (a linear increase), type II (an increase with a slowdown at high prey densities), type III (a sigmoidal increase) and type IV (a dome shape in prey consumption increase (Holling 1961; Pervez 2005; Sakaki & Sahragard 2011). Similarly, the biological traits of a predator, influenced by changes in prey species, greatly affect its

Comentado [Luis4]: Mainly "densities"

predation activity. Thus, this study aims to investigate the interaction of the predator O. similis with the prey species B. tabaci and T. vaporariorum under controlled conditions. The main aim is to evaluate the functional response parameters, fitness parameters, biological traits, and population parameters of O. similis when fed B. tabaci and T. vaporariorum nymphs separately. **Material and Methods** Insect rearing **Predator** Adults of O. similis were captured from vegetable and cotton fields of the Huazhong Agricultural University (Wuhan, China) and mass reared in an insectarium following the method described by Zhou et al. (2006) with slight modifications. The rearing arenas consisted of transparent boxes $(23.5 \times 22.0 \times 5.5 \text{ cm})$ with ventilation in the lid. Nymphs and adults of O. similis were supplied black aphids (Aphis fabae). Small stems of Vitex negundo (3–4) wrapped with wet cotton over the end were provided as oviposition substrate. Environmental conditions Prey species Adult B. tabaci were collected from vegetables grown in greenhouses and from other crops from open fields located in the campus of the Huazhong Agricultural University. They were then moved to insectaria and released on potted cotton (Gossypium hirsutum) plants (10 cm) to develop the stock culture for the experiments (Khan & Wan 2015; Tomar et al. 2017). The stock culture of T. vaporariorum was maintained from a few adults received from the Southwest University (Chongqing, China). Large screen cages $(65 \times 65 \times 65 \text{ cm})$ were used as arenas for both whitefly species. The obtained T. vaporariorum individuals were released on tobacco

(Nicotiana tabacum) plants (10 cm) for mass rearing (Haiyan et al. 2017; Wei et al. 2018). The

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140 following environmental conditions were maintained inside the insectaria: temperature $26 \pm 1^{\circ}$ C, 141 RH 65 \pm 5%, and a photoperiod of 16 L: 8 D h at a light intensity of 1400–1725 lux. Functional Response of O. similis 142 The third instar nymphs and three-day old adult females of O. similis were collected from the 143 insectaria and fed third instar nymphs of B. tabaci and T. vaporariorum for 48 h separately and 144 starved for 24 h. The predatory bugs were then individually transferred to small petri dishes (9 145 cm in diameter and 2 cm in depth) and supplied separately with third instar nymphs of B. tabaci 146 147 and T. vaporariorum with different densities (4, 6, 8, 10, 12, and 14) per predator. After 24 h, predators were removed, and the prey consumed by both life stages of O. similis counted under 148 149 stereomicroscope. Bugs were used once only. All the dead/empty nymphs of both whitefly species were assumed killed by the predator as preliminary study indicated 100% survival of 150 bugs in the absence of whitefly nymphs. Thirty replications of the experiments involving the 151 152 third instar nymphs and adult females of O. similis were made for each treatment/density with both prey species separately. 153 154 Life table study Nymphal development 155 156 Approximately sixty freshly laid healthy eggs of O. similis were isolated from the insectaria and 157 incubated until hatched. All collected eggs of O. similis were equally distributed to feed on B. tabaci and T. vaporariorum third instar nymphs separately. After hatching, O. similis neonates (≤ 158

24 h) were isolated in small Petri dishes (diameter: 9 cm; depth: 2 cm) firmed with filter paper.

From the results of functional responses, we supplied fifteen third instar nymphs of B. tabaci and

T. vaporariorum separately to each individual of the predatory bug as food. Dead/empty nymphs

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Comentado [Luis6]: Fed 48 h contradictory with line 147

Comentado [Luis7]: Line 144 refer to 24 h. predation

of whitefly were replaced every day. A stem of *V. negundo* was placed in each Petri dish to provide shelter and moisture to the predatory bugs. The end of each stem was wrapped with wet cotton to keep them moist. Thirty nymphs were used in the experiment with three biological replicates for each prey species. Developmental time for each nymphal instar was measured. Individuals that died before reaching the adult stage were also recorded. Sex was confirmed as soon as the adults emerged.

Adult longevity and fecundity

Newly emerged male and female *O. similis* adults were paired for mating. Females that mated for more than 1.5 min were considered to have been mated (Butler and O'Neil, 2006). Each mated female was placed separately in a new cylindrical translucent vial (2.5 × 14 cm diameter and length respectively) enclosed with an adequate mesh nylon screen. A small section of *Vitex negundo* stem was offered to each *O. similis* female as an oviposition substrate. Each stem was wrapped with moist cotton at the end to provide moisture to the stem as well as the bugs using the method of Zhou et al. (2006). *Bemisia tabaci* and *T. vaporariorum* nymphs (N = 15) were supplied into each vial as a source of food for female *O. similis*. Stems of *Vitex negundo* were examined under a stereomicroscope (15×) to confirm egg laying. The stem was changed every day after the female laid the first egg. The total number of eggs laid by each female was counted under a stereomicroscope (15×). All predatory bugs were observed until they died. Development period, survival rate, pre-oviposition and oviposition period, fecundity, and longevity of female and male adults of *O. similis* were recorded.

Data analysis

Functional response

184 To find the functional response type, all the data collected from the experiment were fitted to 185 polynomial function. The polynomial function described the relationship between the proportion of prey consumed (Na) in relation to the density of prey offered (No) (Holling, 1959a; Holling, 186 187 1959b). Hence, a cubic model was applied in a logistic regression analysis (Juliano, 2001): $N_a/N_0 = \exp((P_0 + P_1N_0 + P_2N_0^2 + P_3N_0^3) / [1 + \exp(P_0 + P_1N_0 + P_2N_0^2 + P_3N_0^3)])$ 188 In this equation, P₀, P₁, P₂, and P₃ represent the intercept, linear, quadric, and cubic coefficients, 189 respectively. The negative and positive values of linear coefficient (P₁) define type II and III 190 191 functional responses, respectively. Modifying the Holling disc equation through reciprocal linear 192 transformation, functional response parameters (Th and a) were calculated (Livdahl & Stiven 193 1983). The equation for the linear regression was y = ax + b. Hence, the modified equation

195 $1/N_a = 1/a \cdot 1/TN_o + T_b/T$

obtained was:

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Where $1/N_a$ represents y, 1/a represents a, $1/TN_o$ represents x, and T_h/T represents b. N_a is the number of prey killed by predators during time (T: 24 h in our experiment). N_o is the density of prey and T_h is the predator handling time for one prey item. For each prey density we calculated total handling time ($T_h = T_h \times N_a$), search time ($T_h = T_h \times N_a$), attack rate ($T_h = T_h \times N_a$), and search efficiency ($T_h = T_h \times N_a$) (Hassell 2000; Rocha & Redaelli 2004). All statistical analyses were performed in MINITAB 17.

Life table analysis

All the raw data for the life table of *O. similis* were analyzed based on the age-stage, two-sex life table theory (Chi 1988; Chi & Liu 1985). The developmental period, fecundity of female adults, and male and female longevity/survival of *O. similis* were evaluated using the computer program

Comentado [Luis8]: Contradictory with line 144 (fed 48 h)

two-sex MS-Chart (Akca et al. 2015). To calculate the standard error of pre-adult stage, adult male and female longevity, female fecundity, and population parameters (rate of increase [r], $[\gamma]$, the highest net reproductive rate $[R_0]$, gross reproduction rate [GRR], and mean generation time of individuals [T]), 100,000 bootstrap replicates were run (Akca et al. 2015; Akköprü et al. 2015; Tuan et al. 2016). Based on the confidence intervals of differences, paired bootstrap tests were also used to compare the results of different treatments using two-sex MS-Chart (Akca et al. 2015; PRU et al. 2015). Equations used in the age-stage, two-sex life tables are listed in Table 1. Sigma Plot 12.0 was used to create graphs of survival rates, fecundity, life expectancy, and reproductive value. In Table 1, S_{xi} represents the age-stage specific survival rate, based on the probability that newly hatched individuals will survive to age x and j (Chi & Liu 1985). The l_x and m_x were estimated using equation 1 and 2 respectively. The intrinsic rate of increase (r) was calculated from the Euler-Lotka equation with age indexed from 0 (Goodman 1982), equation 3. Equation 4 was used to calculate the R_0 (mean number of siblings that an individual can produce in its lifespan). The gross reproduction rate (GRR) was estimated using equation 5. The time a population required to rise to R_0 -fold of its size as it acquired a stable age-stage distribution (T) was determined with equation 6. To estimate the rate of increase (λ), Equation 7 was used. Results Functional response of O. similis The results of the logistic regression analysis for third instar nymphs of O. similis were highly

significant (P < 0.05) suggesting a type II functional response as the linear coefficient (P1) was

negative against nymphs of both B. tabaci and T. vaporariorum (Table 2). Similarly, the adult

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females also showed a type II functional response against both prey species, although the parameters were not significant.

The functional response curves of different life stages of *O. similis* to third instar nymphs of *B. tabaci* and *T. vaporariorum* at different densities are show in Fig. 1. The number of nymphs of both prey species consumed by third instar nymphs and adult females of *O. similis* increased with increases in the prey density from 4 to 8 nymphs per predator, but plateaued with no significant increase in prey consumption with densities of more than 8 nymphs per predator. When only four nymphs of *B. tabaci* and *T. vaporariorum* were provided, the third instar nymphs of *O. similis* consumed a mean of 3.8 and 3.3 nymphs per predator per day, respectively, indicating that the predator is more efficient at finding whitefly nymphs at low prey densities.

Comentado [Luis9]: Why this affirmation? Four is the minimum provided preys. Support the affirmation

Similarly, the mean consumption of whitefly nymphs by adult female *O. similis* was higher at lower prey densities (Fig. 1). When provided with third instar nymphs of *B. tabaci* and *T. vaporariorum*, the maximum and minimum prey consumption levels of third instar nymphs of *O. similis* were (95% and 46.4%) and (82.5% and 44%), respectively. However, there were no significant differences found in maximum prey consumption by adult female *O. similis* (92% and 92.5%) when preying on nymphs of *B. tabaci* and *T. vaporariorum*, respectively. The minimum percentage of *B. tabaci* and *T. vaporariorum* nymphs killed by adult female *O. similis* was 48% and 51%, respectively.

Functional response parameters

The parameters of functional response (handling time (T_h) , attack rate (a), and maximum predation rate) of different stages of O. similis against whitefly species are listed in Table 3.

There were no differences estimated in the T_h of third instar nymphs of O. similis against both

250 whitefly species. However, adult females undertook shorter handling times (1.75 h) when 251 pursuing, subduing, and consuming T. vaporariorum third instar nymphs when compared with the handling times for *B. tabaci* (2.45 h). 252 Comentado [Luis10]: Increase in the table 3 the SD or IC with letter for differences or similarities in Th, α 253 In contrast, the coefficient of attack rate was higher, ranging from 0.05 to 0.06, when third instar 254 nymphs and adult females of O. similis were fed nymphs of B. tabaci compared to T. vaporariorum (Table 3). However, there were no significant difference noted for different life 255 256 stages of O. similis in attack rate. The maximum predation rate (T/Th) of third instar nymphs of O. similis per individual was higher (9.82 d⁻¹) for T. vaporariorum nymphs compared to B. 257 tabaci (9.78 d⁻¹). Similarly, the maximum predation rate of adult females was (13.70 d⁻¹) and 258 (11.12 d⁻¹) for T. vaporariorum and B. tabaci nymphs, respectively. The functional response 259 parameters of different life stages of O. similis to different densities of whitefly species are given 260 in Table 4. The T_h for third instar nymphs and adult females of O. similis increased with 261 increasing prey densities. However, the searching time and searching efficiency show an inverse 262 relationship with both prey densities. The attack rates of both life stages of O. similis were 263 264 similar with no significant differences at different densities of both prey species. Growth, development, and longevity of O. similis 265 266 Developmental characteristics of O. similis when fed of against whitefly species (B. tabaci and T. vaporariorum) are listed in Table 4. The nymphal development of O. similis took statistically 267 Comentado [Luis11]: Is correct? longer from N1–N4 when fed T. vaporariorum than when fed B. tabaci. 268 Comentado [Luis12]: What is?? Is Nymph??, specific However, there were no significant differences in development of fifth instar O. similis nymphs 269 against both whitefly species. In addition, higher mortality rates were observed in the fifth instar 270 271 when fed T. vaporariorum nymphs than when fed B. tabaci (fig 2??). The results also showed Formatado: Fonte: Não Itálico

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that longevity of both male and female O. similis was statistically similar when B. tabaci and T. vaporariorum were provided as prey (table 5???). The highest longevity of adult females was recorded when they were fed B. tabaci nymphs (15.47 days), while when presented with T. vaporariorum nymphs, it was comparatively shorter (13.82 days) (table 5????). It was also observed that more individuals of the predatory bug survived and successfully reached the adult stage when fed B. tabaci nymphs (86.66%) than when fed T. vaporariorum nymphs (56.70%). Fecundity and oviposition of female adults No significant effects of the presence of the two whitefly species were observed on adult preovipositional period (APOP) of O. similis (2.35 days for B. tabaci and 2.4 days for T. vaporariorum, respectively; (Table 65). However, the total pre-ovipositional period (TPOP) of O. similis was significantly longer when offered T. vaporariorum nymphs (20.5 days) compared to B. tabaci (19.82 days) (Table 6). It was also observed that the total reproductive days of female adult O. similis were statistically similar with no significant difference for both whitefly species (8.29 and 7.80 days when fed B. tabaci and T. vaporariorum, respectively) (table 6). Furthermore, no significant difference was recorded in total female fecundity against both prey species; however, more eggs were laid by adult females when fed B. tabaci nymphs (54.18 eggs) than T. vaporariorum (49.82 eggs). More females were produced when fed B. tabaci than T. vaporariorum. Population parameters of O. similis The influence of the two prey species on the population parameters of O. similis are listed in Table $\underline{67}$. No significant differences were found in O. similis population parameters when fed B.

tabaci and T. vaporariorum separately. The intrinsic rate of increase (r) and finite rate of

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Comentado [Luis13]: However, was not difference statistic. Are similar, not highest not shorter.

Comentado [Luis14]: Was not difference statistic, are similar

increase (λ) for *O. similis* were (0.13 d⁻¹ and 1.14 d⁻¹, respectively) when fed nymphs of *B. tabaci* and (0.11 d⁻¹ and 1.12 d⁻¹) when fed *T. vaporariorum*. However, the highest net reproductive rate (R_0) occurred when *O. similis* was fed *B. tabaci* (30.70 offspring per female) than when fed *T. vaporariorum* (18.27). Furthermore, the mean generation time of individuals (T) and the values of *GRR* were higher when *B. tabaci* was offered as prey (25.46 d and 54.62) compared to

Comentado [Luis15]: Was not difference statistic

Age-stage and age-specific survival of O. similis

T. vaporariorum (26.61 d and 61.57; Table 76).

The \mathbb{F} fig. 2 explains the age-stage specific survival rate (S_{xj} ; the possibility a newly hatched individual that will successfully survive to age x and stage j) of O. similis when fed B. tabaci and T. vaporariorum. Overlap occurs between stages as a result of variations in the developmental rate of individuals. When fed nymphs of B. tabaci, 86.66% of O. similis eggs successfully survived and reached to the adult stage. However, the survival rate was significantly lower (56.7%) when O. similis were fed T. vaporariorum nymphs.

Age-specific survival rate (l_x ; a simplified form of S_{xy}), age-stage specific fecundity (f_x), age-specific total fecundity of the whole population (m_x), and age-specific maternity ($l_x m_x$; formed on the basis of f_x and m_x) of O. similis when fed B. tabaci and T. vaporariorum are presented in Fig. 3. As age increased, the l_x of O. similis decreased and showed an inverse relationship for both prey species. The peak of the m_x curve was at 26.98 days (5.14 eggs) and 31.09 days (5.72 eggs) when O. similis fed on O. tabaci and O. vaporariorum, respectively. The peak of O0 was at 24.90 days (8.45 eggs) and 30.88 days (7.80 eggs) for O0. tabaci and O1. vaporariorum nymphs respectively.

Life expectancy and reproductive values

The curves for life expectancy (e_{xj}) of O. similis at each stage when presented with B. tabaci and T. vaporariorum are shown in Fig. 4. The life expectancy of a newborn O. similis egg was greater when fed nymphs of B. tabaci (28.57 days) than those of T. vaporariorum (24.91 days). The age-stage reproductive values (v_{xj}) of adult female O. similis when exposed to different species of whitefly are shown in Fig. 5. Age-stage specific reproductive value is a measure of the contribution of an individual (from age x and stage j) to a future population. When adult female O. similis fed on B. tabaci, the highest peak was observed at 21 days, which was greater than when offered T. vaporariorum (19.79 days). The reproductive curve (V_{xi}) indicated that the presence of B. tabaci had a more positive effect on O. similis reproduction than T. vaporariorum. Discussion

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To quantify the ability of a predator to combat agricultural pests, the Holling functional response model has been used for several years (Ganjisaffar & Perring 2015; Yazdani & Keller 2016). Handling time (T_h) and attack rate (a) are considered key parameters in explaining oscillations in predator and prey interactions (Wang et al. 2019). A predator's functional response to its prey plays a significant role in the effect it has on a prey population (Begon et al. 1986). Similarly, life table studies enable us to understand the ecology of an organism and supply some crucial tools to study vital biological functions such as growth, survival, and reproductive rate when an organism is in a diverse environment. Some drawbacks have been found in Jackknife methods; therefore, a bootstrap method using 100,000 resamples was developed to calculate population parameters with more accurate results (Huang & Chi 2012; Huang & Chi 2013). Numerous studies have investigated Orius spp. as a predator of B. tabaci, (Adly 2016; Arnó et al. 2008;

Banihashemi et al. 2017; Shahpouri et al. 2019; Zandi-Sohani et al. 2018). However, to our knowledge, functional response and age-stage life table traits (age-specific survival, age stage survival, female reproductive values and life expectancy) have not been investigated for *O. similis* when preying on *B. tabaci* and *T. vaporariorum*. Thus, our study was designed to determine the predatory potential of *O. similis* against both whitefly species.

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Our results indicate that the third instar nymphs and adult female of O. similis show type II functional responses when fed separately on six different densities of B. tabaci and T. vaporariorum third instar nymphs. With increases in prey density, the net prey consumption of both life stages of O. similis increased until a plateau was reached. Predators with type II and III functional responses have a probability of being a stabilizing force in biological control programs (Fernández-arhex & Corley 2003). Orius spp. have shown a type II functional response in numerous other studies (Holling 1965). A type II functional response was reported for O. albidipennis when it was fed T. tabaci (Madadi et al. 2007), Megalurothrips sjostedti (Thysanoptera: Thripidae) larvae (Gitonga et al. 2002), and Tetranychus turkestani (Acari: Tetranychidae); (Hasanzadeh et al. 2015). In contrast to our results, the adult female O. albidipennis showed a type III functional response when fed B. tabaci third instar nymphs (Shahpouri et al. 2019). Similarly, M. caliginosus showed a type III functional response when presented with nymphs of T. vaporariorum (Enkegaard et al. 2001). These contradictory results may be related to changes in predator species and differences in body size. Supporting our results, O. majusculus and O. laevigatus exhibited a type II functional response when fed nymphs of T. vaporariorum (Montserrat et al. 2000). Predator functional response is influenced by several factors, such size and density of predators and preys (Aljetlawi et al. 2004), temperature (Gitonga et al. 2002; Zamani et al. 2006), occurrence of alternative prey (Abrams

1990), and internal state of the predator (Hassell et al. 1976). In our study, the arena consisted of small Petri dishes. This small experimental arena accelerated the searching efficiency of the predatory bugs and enabled them to repeatedly attack prey that initially escaped (Wiedenmann & O'Neil 1991). The optimal foraging theory of predator-prey relationships has helped reveal the influence of different prey densities on predator handling time, searching time, and predation rate (Cook & Cockrell 1978; Stephens & Krebs 1986). In our study, searching time decreased with increases in prey density for both third instar nymphs and adult female of *O. similis*.

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To estimate the effectiveness of a predator in relation to its prey, handling time is thought to be a key parameter because it shows how long a predator takes to capture, subdue, kill, and digest a single prey item (Atlıhan et al. 2010). In our study, the handling time was shortest when adult female O. similis were offered T. vaporariorum nymphs. In contrast to our study, the handling time was higher when adult female O. albidipennis fed on nymphs of B. tabaci (Shahpouri et al. 2019). However, long handling time enables increased nutrient consumption from prey and hence increases the persistence of predators (Montserrat et al. 2000). Recent studies have shown that handling time is higher when O. albidipennis preys on B. tabaci nymphs than eggs (Shahpouri et al. 2019). Similarly, handling time of O. laevigatus was longer than that for other Orius species when tested again different densities of thrips (Montserrat et al. 2000). Maximum prey consumption enhances the possibility of gaining optimal ratios between predators and pests. Hence it can be useful to accelerate the application of inoculative releases (Wang et al. 2019). Our results show that maximum predation occurred when adult female O. similis fed on T. vaporariorum. Meanwhile, more nymphs were eaten by third instar nymphs of O. similis when fed on T. vaporariorum. Higher prey densities (and thus greater prey availability) or decreases in searching area accelerate predator attack rates while reducing

handling time (Hassell et al. 1976). In a previous study (Opit et al. 1997), lower prey densities induced reduction in predator searching activity to reduce the use of energy. In our study, no significant differences in attack rate were observed when both life stages of *O. similis* preyed on nymphs of *B. tabaci* and *T. vaporariorum* separately, even with six different densities. The attack rate did not therefore depend on prey densities (Holling 1959). However, our results showed higher attack rates with *B. tabaci* than *T. vaporariorum* for both third instar nymphs and adult females of *O. similis*. In contrast to our results, previous work resulted in different values for handling time and attack rate when *O. majuscules* and *O. laevigatus* were fed *T. vaporariorum* (Montserrat et al. 2000). This may be due to changes in experimental design, environment, or predator species (Van Alphen & Jervis 1996). The polyphagous predatory species of *Orius* prey on a broad range of arthropods. Additionally, prey type can significantly alter the activity of their predators (Bonte et al. 2015). The developmental and reproductive performance and fitness of predators in relation to particular prey types highlights their potential as active biological control agents (Grenier & De Clercq 2003).

The results obtained from the age-stage two-sex life table analysis indicate that *O. similis* can survive and build strong populations when feeding on different species of whiteflies (*B. tabaci* and *T. vaporariorum*). However, we found that the nymphal developmental durations of the predator were significantly longer when praying on nymphs of *T. vaporariorum* than on nymphs of *B. tabaci*. Moreover, adult male and female longevity was longer when presented *B. tabaci* relative to *T. vaporariorum*, but a significant difference was not observed. Our results agree with previous reports indicating that the developmental period of *O. similis* pre-adults is greater when they prey on *A. cracivora* than on *C. cephalonica* eggs (Amer et al. 2018). The same interaction was documented in numerous studies where the prey species strongly alter the

developmental duration of pre-adults of O. similis (Kim 1997; Kim 1999; Ohta 2001; Sengonca et al. 2008). Consequently, these fluctuations in the developmental period show that a prey species can strongly influence the developmental time of the pre-adult stages of O. similis (Sengonca et al. 2008). The survival of O. similis individuals was higher when fed on B. tabaci nymphs than T. vaporariorum. Amer et al. (2018) documented that higher mortality rates occurred in the pre-adult stages of O. similis when they fed on C. cephalonica eggs compared to A. cracivora. Similarly, the survival rate of O. laevigatus, O. niger, and O. majusculus individuals during development was very low (i.e., high mortality rates) when fed eggs of E. kuehniella than those of F. occidentalis (Kiman & Yeargan 1985; Tommasini et al. 2004). Based on our results and those of Arnó et al. (2008), the total developmental duration and survival of O. similis nymphs, when fed B. tabaci nymphs, were similar to those of O. majusculus and O. laevigatus. Arnó et al. (2008) also reported results similar to those reported by Riudavets & Castañé (1998), suggesting that whitefly species could be considered suitable prey analogous to F. occidentalis larvae predated upon by Orius spp. Furthermore, our results showed no significant difference in adult-pre-oviposition period for both whitefly species. In contrast to our study, Zhang et al. (2012) found that after mating, the development of the reproductive system of adult female O. similis took longer (5-6 days) compared to our results. Similarly, the number of eggs laid by adult female O. similis were comparatively higher for both whitefly species than that of Tetranychus cinnabarinus (Zhang et al. 2012).

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Chen et al. (2017) found that intrinsic rate of increase (r) is a key population parameter in determining the development, growth, and survival of an organism. Southwood & Henderson (2009) also documented that greater values of (r) i.e., r > 0 highlight the fit of a prey with its host. Comparing our result with these studies shows that the intrinsic rate of increase was more

than (0) and similar for both prey species Table 4. The net reproductive rate (R_0) is also considered an important demographic life table parameter. Values of R_0 of more than 1 indicate an increase in the mean population of an insect (Chen et al. 2017; Southwood & Henderson 2009). Our results agree with this theory, as the highest R_0 was when O. similis was fed B. tabaci relative to T. vaporariorum. In contrast to our study, low reproductive rates have been observed when O. similis is fed T. cinnabarinus (Zhang et al. 2012). The gross reproduction rate (GRR) is thought to be a symbol of a rapid increase of population, which is directly related to adult eclosion and the number of eggs laid and hatched. All of these parameters can be significantly influenced by prey species (Cocuzza et al. 1997; Huang & Chi 2013). In our results, the highest GRR and greatest generation time (T) occurred when O. similis fed on T. vaporariorum when compared to B. tabaci. However, in their study, Zhang et al. (2012) observed longer generation times at three constant temperatures when individuals of O. similis preyed on T. cinnabarinus.

The survival of a predator from the neonate to the adult stage when presented specific prey species highlights its role as an effective biological control agent (Van Lenteren & Woets 1988). Similarly, the searching ability and concomitance of predators and prey in space and time, are thought to be a crucial factors in successful biological control of a pest (Arnó et al. 2008). Montserrat (2001) and Trottin-Caudal et al. (1991) both reported large populations of *Orius* spp. on vegetable crops where thrips and whiteflies coexist. Thus, it can be assumed that if *O. similis* remains on the crop after suppression of the population of their desired prey, they can play a vital role in suppression of both whitefly species and could serve as an effective biological control agent. Supporting our hypothesis, Riudavets (2001) reported more than 20 *Orius* per cucumber plant when the population of *F. occidentalis* was very low, which may have been due to the presence of other pests.

In summary, our results suggest that the predatory bug O. similis has the potential to actively maintain strong populations when in the presence of species of whitefly such as B. tabaci and T. vaporariorum and could serve as a biological control agent in cotton fields and other vegetable crops, as well as in greenhouses where the populations of these species are destructive pests. The results obtained from laboratory experiments should be useful in understanding the biology of O. similis when in association with whitefly species, but field experiments will be required to validate them. Acknowledgments: The authors want to thank Miss. Abida Butt (Institute of Zoology, Punjab University Lahore, Pakistan), Mirza Abid Mahmood (College of Plant Science and Technology, Huazhong Agricultural University, Wuhan) and Fawad Khan (Department of Entomology, University of Georgia, U.S.A.) for technical assistance. The author acknowledge the funding support granted by National Natural Science Foundation of China (Grant No. 31872023), and The National Key R&D Program of China (2017YFD0201000) for the current study. Reference Abrams PA. 1990. The effects of adaptive behavior on the type-2 functional response. Ecology 71:877-885. Adly D. 2016. Use of Predators for Controlling the Whitefly, Bemisia tabaci Genn. and the Two Formatado: Fonte: Itálico Spotted Spider Mite, Tetranychus urticae Koch., in Cucumber Greenhouses in Egypt. Formatado: Fonte: Itálico Egyptian Journal of Biological Pest Control 26. Akca I, Ayvaz T, Yazici E, Smith CL, and Chi H. 2015. Demography and population projection of Aphis fabae (Hemiptera: Aphididae): with additional comments on life table research Formatado: Fonte: Itálico

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