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Performance of the parasitoid species *Aphelinus asychis* Walker (Hymenoptera: Aphelinidae), *Aphidius ervi* (Haliday) (Hymenoptera: Braconidae) and *Diaeretiella rapae* (McIntosh) (Hymenoptera: Braconidae), using *Myzus persicae* (Sulzer) (Hemiptera: Aphididae) as host

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Abstract

Three parasitoid species *viz. Aphelinus asychis* Walker (Hymenoptera: Aphelinidae), *Aphidius ervi* (Haliday) (Hymenoptera: Braconidae) and *Diaeretiella rapae* (McIntosh) (Hymenoptera: Braconidae) parasitizing the aphid species *Myzus persicae* (Sulzer) (Hemiptera: Aphididae) in mid-hills of north India were studied. At different locations and times of the year, the parasitization by *A. asychis*, *A. ervi*, and *D. rapae* ranged from 7.53 to 37.58, 4.26 to 80.45, and 74.25 to 80.48%, respectively. All the 3 parasitoids successfully completed their development on different nymphal instars of the aphid host and the total developmental duration of *A. asychis*, *A. ervi*, and *D. rapae* ranged 10.4–14.6, 24.2–29.6, and 10.2–15.2 days, respectively. It was significantly longer on the 1st nymphal instar of the host. The longevity of the female parasitoids was significantly longer than their counterparts. Differences in host age significantly influenced the longevity of female parasitoids and it was more on 1 to 2-day-old nymphs than that on 4 to 5-day-old nymphs. Fecundity and ovipositional periods of the parasitoids on younger (1–2 days old) host age group were considerably prolonged than on the older ages of the aphid. Average total fecundity of *A. asychis* and *D. rapae* was significantly higher when parasitizing 1–2-day-old nymphs. In *A. asychis*, host feeding behavior was also observed by a total host feeding of 89.2 aphids (1–2 days old) and 43.4 aphids (4–5 days old) during its life span. It is concluded that *A. asychis*, *A. ervi*, and *D. rapae* can be mass reared using *M. persicae* as host and can be utilized successfully in augmentative biological control program.

Keywords: *Aphelinus asychis*, *Aphidius ervi*, *Diaeretiella rapae*, *Myzus persicae*, Parasitization, Biological parameters

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Background

Myzus persicae (Sulzer) (Hemiptera: Aphididae) feeds on over 500 host plants belonging to 40 unlike families and various chief agricultural crops cultivated in natural fields and protected environments (Byeon et al. 2011). It is an important pest of cucumber, capsicum, carnation, and gerbera grown under protected environment and oilseed brassicas under open field conditions (Sanchez et al. 2010 and Kumar and Gavkare 2014). Among various management practices, the use of chemical insecticides is common among farmers. However, the extensive use of these chemicals has ensued in several problems like insecticide resistance, resurgence of minor insect pests, and mortality of beneficial organisms. All these harmful consequences of injudicious use of pesticides have guided to the evolution of integrated pest management, wherein biological control is a very important component. Many biocontrol agents (parasitoids, predators, and entomopathogens) associated with *M. persicae* have been reported from distinct parts of the world (van Emden et al. 1969 and Perdikis et al. 2008).

Aphid parasitoids are often used in biological control programs in polyhouses and open fields (Boivin et al. 2012). Several hymenopteran parasitoids *viz. Aphelinus gossypii* Timberlake, *Aphidius colemani* Vierek, *Binodoxis indicus* (Subba Rao and Sharma), *A. ervi* (Haliday), and *Lipolexis oregmae* Ghan have been reported to restrain the population growth of *M. persicae* under open field conditions (Kumar 2013). *Diaeretiella rapae* (McIntosh) is a potential bio-agent for biological control against aphids in different nations (Zaki et al. 1999 and Maghraby 2012).

In the present study, main biological parameters of 3 predominant parasitoid species namely *A. asychis*, *A. ervi*, and *D. rapae* were estimated when parasitizing the aphid species, *M. persicae*.

Methods

Sampling of parasitoids

Samples of the *M. persicae* infested host plants (*Capsicum annuum* L. from polyhouses and *Brassica* oilseeds from open fields) were collected during the winter months of December 2014 and March 2015 from 4 districts *viz.* Kangra (32°, 15.987′ N, 76°, 28.412′ E), Kullu (31°, 84.646′ N, 77°, 16.053′ E), Mandi (31°, 53.841′ N, 76°, 89.634′ E), and Solan (30°, 86,004′ N, 77°, 17.303′ E) under humid subtropical mid-hills of Himachal Pradesh, India. Ten samples of aphid-infested leaves of *C. annuum* were collected from each of the 5 polyhouses selected at different locations of each district and also from *Brassica* oilseed crops from open fields. Fifty samples were collected from each district under polyhouse and open fields. The aphid infested leaves of *C. annuum* and apical twigs of *Brassica* oilseed plants were transferred to the laboratory in paper bags. The samples were examined under stereo-zoom microscope to count total numbers of healthy and mummified aphids. From these counts, percent parasitization was estimated, using the formula of Walton (1986):

$$\text{Parasitization }(\%)\frac{\text{Number of mummies}}{\text{No.of mummies} + \text{No.of live aphids}} \times 100$$

Insect rearing

The stock culture of *M. persicae* was reared on *C. annuum* in an insectary at 25 ± 1 °C, 70 ± 5% RH, and a L16:D8 photoperiod as per the method adopted by Kumar et al. (2019). Mummies of the 2 parasitoid species *A. asychis* and *A. ervi* were collected from the polyhouses at different locations, whereas the ones of *D. rapae* was obtained from *Brassica* oilseed farms in the locality. The stock cultures of parasitoids were reared on *M. persicae* maintained on *C. annuum* under caged conditions in small plastic containers (50 mm × 80 mm) as per the method of Sengonca et al. (2008). The identity of parasitoid was ascertained by comparing its morphological attributes with taxonomic keys (Takada 2002 and Rakhshani et al. 2015) under stereo-zoom microscope in the laboratory. Fresh parasitoids individuals were used in experimentation. Plastic containers having mummified aphids were examined daily for the emergence of adult parasitoids. The emerged adults were separated by sex and the females were allowed to mate for 6 h in glass containers before initiating the experiments. The mated female wasps were transferred to plastic pots contained aphid colony. As a food source to the adult parasitoids, cotton swabs saturated with honey-water solution (30%) were provided.

Developmental parameters of the parasitoids

Developmental duration

Potted plants of *C. annuum* infested with *M. persicae* (n = 150) were exposed to mated female parasitoids (n = 50) for oviposition. After 24 h, the plants were placed in the insectary for further development. After emergence, the parasitoids were collected and reared in separate cages on the respective day. One leaf of capsicum, with aphid nymphs (n = 10) of each instar, was offered to one mated female in separate glass vials (15 × 150 mm). The vials were examined at 24 h interval from oviposition till mummification.

Forewing width of the parasitoids

Forewings of males (n = 10) and females (n = 10) of the parasitoids *viz. A. asychis* and *A. ervi* and *D. rapae* were mounted on slides using a Canada balsam. The prepared slides were then placed in the oven for 4–5 days at 25 °C. The slides were examined under a stereo-zoom microscope fitted with imaging software to measure the forewing widths.

Longevity of male and female parasitoids

After mating (for 6 h), the emerged adult parasitoids were separated into males and females. Twenty nymphs of each host age group were offered daily to each mated male (n = 10) and female (n = 10) (1 day old) in individual rearing glass jars throughout their life span. The male parasitoids were removed on 3rd day, while the females were placed daily into new glass jars with fresh aphids (from their stock culture) until death. Longevity of adult males and females of the parasitoids were estimated.

Age-specific fecundity and ovipositional periods of the parasitoids

These parameters of female parasitoids were studied in 2 different age groups (1–2 and 4–5 days old) of *M. persicae* as per the method of Sengonca et al. (2008). Ten mated females of the parasitoid provided daily, until their death, to 20 nymphs of aphid of each host age group. After 12 h exposure, the female parasitoids were daily transferred into glass containers having fresh aphids. After 3–4 days, parasitized aphids were dissected and the number of parasitoid larvae was counted. Mummified aphids in the old jars were observed for adult emergence. In case of *A. asychis*, data on age-specific host feeding was also recorded by counting the number of aphids killed by the parasitoid (shrunken and decapacitated aphids). This behavior of host feeding was observed in case of the *A. asychis* females on both the host age groups of the aphid and the data were recorded throughout the entire life span of the parasitoid. The males did not show host feeding behavior like females. All the experiments were repeated 10 times.

Statistical analysis

Data collected were subjected to completely randomize one-way ANOVA at 5% level of significance following the method of Gomez and Gomez (1984). Data on the natural parasitism were subjected to arcsine transformation before ANOVA. The means that differed significantly were separated by least significant difference (LSD) at p = 0.05. Differences among the means of ovipositional, post ovipositional period, fecundity, and host feeding (in *A. asychis*) of the parasitoids on 2 different host age groups were compared using t test ($p \leq 0.05$). Two-way ANOVA was used to analyze the effect of parasitoid species on mean longevity of male and female parasitoids and mean longevity of female parasitoids parasitizing different age groups of *M. persicae*.

Results and discussion

Natural parasitism

Survey revealed the prevalence of 3 main parasitoid species *viz. A. asychis*, *A. ervi*, and *D. rapae* infesting *M. persicae* in mid-hills of north India. *A. asychis* and *A. ervi* were recorded parasitizing *M. persicae* infesting *C. annuum* grown in polyhouses, whereas *D. rapae* was recorded on 3 aphid species *viz. B. brassicae*, *L. erysimi*, and *M. persicae* infesting *Brassica* oilseed crops under open field conditions. Data on parasitization % of the aphids by these parasitoids (Table 1) revealed that during December, the parasitization % by *A. asychis* was 37.58 + 0.84 and 36.95 + 0.71 at Kullu and Solan, which was significantly higher than other locations (LSD = 0.64, $F_{3, 36}$ = 14.73, p < 0.00001). Similarly, in March, the parasitization % at Kullu (10.85 + 1.60) was highest and differed significantly than other locations (LSD = 1.13, $F_{3, 36}$ = 12.71, p < 0.00001). In case of *A. ervi*, the parasitization % was (6.67 + 0.48) significantly higher at Kullu and Solan (6.39 + 0.50) during December (LSD = 0.75, $F_{3, 36}$ = 28.64, p < 0.00001) and at Kullu (80.45 + 1.40) during March (LSD = 1.24, $F_{3, 36}$ = 8.02, p = 0.00032). There was no parasitization with *D. rapae* during December. However, during March, the parasitization % at Kullu was 80.48 + 1.32 and it was significantly more than that at Mandi (78.65 + 1.39) and Solan (78.66 + 0.89) (LSD = 1.22, $F_{3, 36}$ = 18.62, p < 0.00001).

Sampling for the parasitoids was carried out during December and March and high parasitization by *A. ervi* and

Table 1 Important parasitoids parasitizing of *M. persicae* in mid-hills of north India

District	Percent parasitization*					
	Aphelinus asychis		*Aphidius ervi*		*Diaeretiella rapae*	
	December	March	December	March	December	March
Kangra	34.25 ± 0.89 (35.85)c	7.53 ± 0.60 (15.98)c	4.26 ± 0.35 (11.95)c	76.25 ± 2.07 (60.84)c	–	74.25 ± 1.81 (59.50)c
Kullu	37.58 ± 0.84 (37.82)a	10.85 ± 1.60 (19.20)a	6.67 ± 0.48 (14.99)a	80.45 ± 1.40 (63.82)a	–	80.48 ± 1.32 (63.82)a
Mandi	35.86 ± 0.92 (36.81)b	9.22 ± 0.56 (17.64)b	5.20 ± 0.56 (13.15)b	78.11 ± 1.04 (62.11)b	–	78.65 ± 1.39 (62.53)b
Solan	36.95 ± 0.71 (37.40)ab	8.19 ± 0.56 (16.62)bc	6.39 ± 0.50 (14.64)a	78.66 ± 0.84 (62.45)b	–	78.66 ± 0.89 (62.52)b
LSD (p = 0.05)	0.64	1.13	0.75	1.24	–	1.22
$F_{3, 36}$	14.73	12.71	28.64	8.02	–	18.62
p value	< 0.00001	< 0.00001	< 0.00001	0.00032	–	< 0.00001

Figures in parentheses are arc sine transformed values
Mean values within the columns bearing the same letters are not significantly different—LSD (p = 0.05)
*Average of 50 samples

Table 2 Mean developmental duration (mean ± S.E.) of *A. asychis*, *A. ervi*, and *D. rapae* on different nymphal instars of *M. persicae*

Aphid Instar	Developmental duration (days)								
	A. asychis			*A. ervi*			*D. rapae*		
	Oviposition to mummification	**Mummification to adult emergence**	**Oviposition to adult emergence**	**Oviposition to mummification**	**Mummification to adult emergence**	**Oviposition to adult emergence**	**Oviposition to mummification**	**Mummification to adult emergence**	**Oviposition to adult emergence**
I	6.80 ± 1.04^{a} (6–8)	7.80 ± 0.56^{a} (7–8)	14.60 ± 1.11^{a} (14–16)	15.20 ± 0.56^{a} (15–16)	14.40 ± 0.68^{a} (14–15)	29.60 ± 0.68^{a} (29–30)	7.20 ± 1.04^{a} (6–8)	8.00 ± 0.88^{a} (7–9)	15.20 ± 1.36^{a} (13–16)
II	6.60 ± 0.68^{ab} (5–7)	5.00 ± 0.88^{b} (4–6)	11.60 ± 1.89^{b} (10–13)	15.00 ± 0.88^{a} (15–16)	13.80 ± 1.04^{ab} (13–15)	28.80 ± 1.04^{a} (28–30)	6.60 ± 0.68^{ab} (5–7)	6.60 ± 0.68^{b} (6–8)	13.20 ± 1.04^{b} (13–14)
III	5.80 ± 1.04^{bc} (5–6)	4.80 ± 1.04^{b} (4–6)	10.60 ± 0.68^{b} (9–12)	13.40 ± 0.68^{b} (13–15)	13.40 ± 0.68^{b} (13–14)	26.80 ± 1.04^{b} (26–29)	6.00 ± 0.88^{bc} (5–6)	5.60 ± 0.68^{c} (5–7)	11.40 ± 0.68^{c} (11–12)
IV	5.60 ± 0.68^{c} (5–6)	4.60 ± 0.68^{b} (4–5)	10.40 ± 0.68^{b} (9–11)	12.20 ± 0.56^{c} (12–13)	12.00 ± 0.88^{c} (11–13)	24.20 ± 1.04^{c} (24–25)	5.60 ± 0.68^{c} (5–6)	4.60 ± 1.11^{d} (4–5)	10.20 ± 1.36^{c} (9–11)
LSD (p = 0.05)	0.95	0.87	1.29	0.73	0.90	1.04	0.90	0.92	1.24
$F_{3,\ 16}$	3.47	26.78	20.32	33.50	11.56	48.31	5.44	22.18	28.00
p value	0.041	< 0.00001	0.000011	< 0.00001	0.000337	< 0.00001	0.009	< 0.00001	< 0.00001

Figures in parentheses indicate range
Mean values within the columns bearing the same letters are not significantly different—LSD (P = 0.05)
All values are mean of 10 observations

D. rapae, during March, was due to the favorable temperature. The temperature remained low in December (5.7–18.8 °C) as compared to that in March (8.9–21.2 °C) at all locations. This is in agreement with the findings of Malina and Praslicka (2008) who reported that the relationship between parasitism with *A. ervi* and temperature increased at the temperature range of 15 to 25 °C. However, *A. asychis* was well adapted to the temperatures ranged from 18 to 30 °C (Sengonca et al. 2008); thus, temperature did not influence the parasitoid population in March.

Developmental duration of the parasitoids

Parasitoids completed their developmental periods reaching the adult stage in all instar of *M. persicae* (Table 2). In case of *A. asychis*, developmental duration from oviposition to mummification was longer in 1st (6.80 ± 1.04 days) and 2nd nymphal instars (6.60 ± 0.68 days) than in the 4th instar (5.60 ± 0.68 days) (LSD = 0.95, $F_{3,\ 16}$ = 3.47, p = 0.041). The developmental duration from mummification to adult emergence was longer (7.80 ± 0.56 days) in 1st instar and differed significantly among other instars (LSD = 0.87, $F_{3,\ 16}$ = 26.78, p < 0.00001). Similarly, the total developmental duration from oviposition to adult emergence was significantly longer in 1st instar (14.60 ± 1.11 days) (LSD = 1.29, $F_{3,\ 16}$ = 20.32, p = 0.000011). In case of *A. ervi*, the developmental duration from oviposition to mummification in 1st (15.20 ± 0.56 days) and 2nd instars (15.00 ± 0.88 days) did not differ significantly. However, it was significantly longer than that observed in 3rd (13.40 ± 0.68 days) and 4th (12.20 ± 0.56 days) instars (LSD = 0.73, $F_{3,\ 16}$ = 33.50, p < 0.00001). Duration from mummification to adult emergence was longer in 1st instar (14.40 ± 0.68 days) and shorter in 4th instar (12.00 ± 0.88 days) (LSD = 0.90, $F_{3,\ 16}$ = 11.56, p = 0.000337). The mean developmental duration from oviposition to adult emergence in 1st (29.60 ± 0.68 days) and 2nd instars (28.80 ± 1.04 days) did not differ significantly; however, it was significantly longer than that observed in 3rd (26.80 ± 1.04 days) and 4th instars (24.20 ± 1.04 days) (LSD = 1.04, $F_{3,\ 16}$ = 48.31, p < 0.00001). Similarly, in *D. rapae*, the developmental duration from oviposition to mummification was significantly longer in younger instars (first and second) than that in later instars (third and fourth) (LSD = 0.90, $F_{3,\ 16}$ = 5.44, p = 0.009). The developmental duration from mummification to adult emergence ranged from 4.60 ± 1.11 to 8.00 ± 0.88 days, and the differences amongst the instars were significant (LSD = 0.92, $F_{3,\ 16}$ = 22.18, p < 0.00001). Total developmental duration from oviposition to adult emergence was longer in 1st instar (15.20 ± 1.36 days) than in all other instars of the aphid (LSD = 1.24, $F_{3,\ 16}$ = 28.00, p < 0.00001).

The present investigation gave important information on biological attributes of native strains of *A. asychis*, *A. ervi*, and *D. rapae* using *M. persicae* as host. Selecting excellent bio-control agent is critical for any successful bio-control program. The choice of suitable aphid instar

Table 3 Forewing width (mean ± S.E.) of the parasitoids parasitizing *M. persicae*

Parasitoids	Forewing width* Mean (mm) ± SE	
	Female	Male
A. asychis	0.32 ± 0.05[c] (0.26–0.39)	0.25 ± 0.02[c] (0.22–0.28)
A. ervi	0.86 ± 0.07[a] (0.74–0.97)	0.77 ± 0.09[a] (0.63–0.95)
D. rapae	0.67 ± 0.011[b] (0.53–0.79)	0.59 ± 0.01[b] (0.52–0.74)
LSD (P = 0.05)	0.06	0.08
$F_{2,\ 27}$	190.23	158.47
p value	< 0.00001	< 0.00001

Figures in parentheses indicate range
Mean values within the columns bearing the same letters are not significantly different—LSD (p = 0.05)
*All values are mean of 10 observations

Table 4 Mean longevity (mean ± S.E.) of *A. asychis*, *A. ervi*, and *D. rapae* males and females parasitizing *M. persicae*

Parasitoids	Mean longevity of male and female parasitoids parasitizing *M. persicae*	
	Longevity in days* (mean ± SE)	
	Male	Female
A. asychis	13.00 ± 1.49[a] (10–15)	25.00 ± 3.01[a] (20–30)
A. ervi	7.10 ± 2.42[b] (5–11)	10.00 ± 2.16[b] (7–14)
D. rapae	6.80 ± 1.25[b] (4–9)	9.40 ± 1.22[b] (7–13)
LSD (p = 0.05)	2.21	2.22
$F_{2,\ 27}$	21.08	133.16
p value	< 0.00001	< 0.00001

Figures in parentheses indicate range
Mean values within the columns bearing the same letters are not significantly different—LSD (p = 0.05)
*All values are mean of 10 observations

Table 5 Mean longevity (mean ± S.E.) of *A. asychis*, *A. ervi*, and *D. rapae* females parasitizing different age groups of *M. persicae*

Parasitoids	Mean longevity of female parasitoids parasitizing different age groups of *M. persicae*	
	Longevity in days* (mean ± SE)	
	Host age of 1–2 days	Host age of 4–5 days
A. asychis	25.80 ± 3.96[a] (20–30)	20.40 ± 1.52[a] (19–22)
A. ervi	11.20 ± 1.92[b] (9–14)	7.20 ± 1.48[b] (5–9)
D. rapae	9.50 ± 0.91[b] (7–11)	5.50 ± 1.13[c] (3–8)
LSD (p = 0.05)	2.04	1.37
$F_{2,\ 27}$	162.14	296.88
p value	< 0.00001	< 0.00001

Figures in parentheses indicate range
Mean values within the columns bearing the same letters are not significantly different—LSD (p = 0.05)
*All values are mean of 10 observations

Table 6 ANOVA results for *A. asychis*, *A. ervi*, and *D. rapae* comparing forewing width, mean longevity of male and female parasitoids, and mean longevity of female parasitoids parasitizing different age groups of *M. persicae*

Source	Degrees of freedom (df)	*F* value	*p* value
Parasitoid sp.	2,59	347.33	1.476E−31
Sex of parasitoid	1,59	22.49	1.58281E−05
Parasitoid sp. × sex of parasitoid	2,59	0.28	0.75
Longevity of male and female parasitoids parasitizing *M. persicae*	1,59	87.53	6.84E−13
Parasitoid sp. × longevity of male and female parasitoids parasitizing *M. persicae*	2,59	24.47	2.72E−08
Longevity of female parasitoids parasitizing different age groups of *M. persicae*	1,59	83.22	1.59E−12
Parasitoid sp. × longevity of female parasitoids parasitizing different age groups of *M. persicae*	2,59	0.91	0.41

can affect considerably the population development of both host and parasitoid. Developmental period is important for mass rearing of the parasitoids. From these results, it can be concluded that the developmental duration of the parasitoids affected with the host age as it was significantly longer on early instars than in the later ones. Prior studies showed that the competence of the parasitoid was influenced by the host age, hence, can be enhanced by choosing the most preferred stage for augmentative biological control (Li et al. 2006). Sengonca et al. (2008) also observed significantly shorter developmental duration of *A. asychis* on the older instars of *Aphis gossypii* (Glov.). The present findings also corroborate with Guigo et al. (2012) who reported that the developmental duration of *D. rapae* varied from 15.65 to 16.41 days on *M. persicae* reared on *Brassica* species.

Host age is important for optimizing the mass rearing; therefore, these results are also consistent with the findings of Schirmer et al. (2008) who reported that under the same climatic conditions, *A. asychis* completed its development in a significantly shorter period when parasitizing 4–5-day-old rather than 1–2-day-old *A. gossypii* nymphs.

Forewing width of the emerging parasitoids

The mean forewing width of *A. asychis*, *A. ervi*, and *D. rapae* females was measured as 0.32, 0.86, and 0.67 mm, while for males, it was 0.25, 0.77, and 0.59 mm, respectively (Table 3). Based on two-way ANOVA (Table 6), the forewings of females were wider than those of the males (df = 1,59; F = 22.49; p = 1.58281E−05). Among the parasitoids, the forewing width of *A. ervi* was greater than that of *A. asychis* and *D. rapae* (df = 2,59; F = 347.33; p = 1.476E−31). Further, analysis showed no interaction between forewing width of parasitoid species and sex (df = 2,59; F = 0.28; p = 0.75).

Tatsumi and Takada (2005) measured the forewing width of *A. asychis* and *A. albipodus* emerging from different aphids *viz. A. gossypii*, *M. persicae*, and *M. euphorbiae* and found that the females of both parasitoids had broader forewing width than the males. He and Wang (2008) observed that the females of *A. ervi* preferred to oviposit fertilized eggs in large hosts, while unfertilized eggs in the smaller host, which resulted in the large-sized females and smaller sized males, respectively. This behavior of females directly affects the forewing width of the parasitoids.

Adult longevity

Among the parasitoids, mean longevity of *A. asychis* was significantly longer than that of *A. ervi* and *D. rapae* both in males (LSD = 2.21, $F_{2,\ 27}$ = 21.08, p < 0.00001) and females (LSD = 2.22, $F_{2,\ 27}$ = 133.16, p < 0.00001) (Table 4). Mean female longevity was significantly longer among parasitoid species (df = 1,59; F = 87.53; p = 6.84E-13) and the ANOVA table also showed an interaction between parasitoid species and mean longevity of parasitoids parasitizing *M. persicae* (df = 2,59; F = 24.47; p = 2.72E-08).

The longevity of *A. asychis* females was significantly longer (25.8 ± 3.96 days), whereas the longevity of *A. ervi*

Table 7 Developmental parameters (mean ± SE) of *A. asychis* females parasitizing different age groups of *M. persicae*

Aphid age groups	*A. asychis*					
	Oviposition period (days)	Post oviposition period (days)	Daily eggs laid/female	Total fecundity (eggs/female)	Daily host/ female	Total host feeding (aphids/female)
1–2 days	19.00 ± 0.45 (19–20)	6.80 ± 0.66 (6–8)	7.00 ± 2.87 (2.8–11)	138.60 ± 7.40 (122–152)	4.0 ± 1.53	89.2
4–5 days	15.00 ± 0.45 (15–16)	5.40 ± 0.69 (4–7)	3.20 ± 1.43 (1.8–6.6)	49.80 ± 3.53 (39–55)	2.3 ± 0.65	43.4
t value	19.88*	3.21*	3.80*	–	4.41*	–
p value	< 0.00001	< 0.00001	< 0.00001	–	< 0.00001	–

Figures in parentheses represent the range
All values are mean of 10 observations
*Significant at $p \leq 0.05$ and df = 18

Table 8 Developmental parameters (mean ± SE) of *A. ervi* females parasitizing different age groups of *M. persicae*

Aphid age groups	*A. ervi*			
	Oviposition period (days)	**Post oviposition period (days)**	**Daily eggs laid/female**	**Total fecundity (eggs/female)**
1–2 days	10.00 ± 1.4 (8–13)	1.20 ± 0.45 (1–2)	4.28 ± 1.53 (4.1–6.46)	49.00 ± 2.99 (43–55)
4–5 days	7.00 ± 1.01 (5–9)	1.10 ± 0.23 (1–2)	7.06 ± 3.26 (6.38–7.67)	50.00 ± 2.88 (44–57)
t value	4.47*	4.10*	2.20*	–
p value	< 0.00001	< 0.00001	< 0.00001	–

Figures in parentheses represent the range
All values are mean of 10 observations
*Significant at $p \leq 0.05$ and df = 18

and *D. rapae* females did not differ significantly, while parasitizing 1–2-day-old nymphs (Table 5) (LSD = 2.04, $F_{2,\ 27}$ = 162.14, p < 0.00001). In case of 4–5-day-old aphid hosts, significant differences in the longevity of female parasitoids were observed (LSD = 1.37, $F_{2,\ 27}$ = 296.88, p < 0.00001). Results of ANOVA (Table 6) showed a significant variation in mean longevity of the parasitoid between different age groups of the aphid (df = 1,59; F = 83.22; p = 1.59E−12), but showed no interaction between parasitoid species and mean longevity of parasitoids parasitizing different aphid age groups (df = 2,59; F = 0.91; p = 0.41).

It is clear from these results that the mean longevity of female parasitoids was longer, while parasitizing nymphs of younger age groups than the older age groups. Nutritional and physiological attributes of the host significantly influence the development, mortality, longevity, and fertility of parasitoids (Roitberg et al. 2001). Sengonca et al. (2008) made similar observations on the longevity of female parasitoids on different host ages and reported that the mean longevity of *A. asychis* females, when parasitizing and host feeding on 1–2-day-old nymphs of *A. gossypii*, was 32.8 days, while on host age groups of 4–5-day-old nymphs and adults the obtained values were 25.2 and 24.2 days, respectively.

Ovipositional period and fecundity of the parasitoids

A. asychis when parasitizing younger nymphs (1–2 day old) of *M. persicae* had a mean ovipositional period of 19.00 ± 0.45 days and total fecundity of 138.60 ± 7.40 eggs (Table 7). On an average, a single female of the parasitoid was capable of host feeding 4.0 ± 1.53 nymphs (1–2 days old) of the aphid. The parasitoid exhibited its host feeding behavior from the very 1st day of its emergence and continued until its death. During its entire average life span of 25.8 days as an adult, the parasitoid killed 89.2 aphids by its host feeding behavior. On 4–5-day-old nymphs, the parasitoid killed 2.3 ± 0.65 nymphs per day by host feeding and a single parasitoid killed 43.4 nymphs in about 20 days of its adult life span. It is clear from the results that the ovipositional period (t = 19.88, p < 0.00001, df = 18), post ovipositional period (t = 3.21, p < 0.00001, df = 18), and daily fecundity (t = 3.80, p < 0.00001, df = 18) of the parasitoid were greater when parasitizing 1–2-day-old nymphs of *M. persicae*.

A. ervi when parasitizing younger nymphs (1–2 days old) of *M. persicae* had a mean ovipositional period of 10.00 ± 1.40 days and a total fecundity of 49.00 ± 2.99 eggs (Table 8). In case of the 4–5-day-old nymphs, the ovipositional period and total fecundity of *A. ervi* females were 7.00 ± 1.01 days and 50.00 ± 2.88 eggs/female, respectively. The results showed that age of aphid host had an influence on the ovipositional period (t = 4.47, p < 0.00001, df = 18), post ovipositional period (t = 4.10, p < 0.00001, df = 18), and daily fecundity (t = 2.20, p < 0.00001, df = 18) of the parasitoids.

D. rapae when parasitizing 1–2-day-old nymphs of the *M. persicae* had a mean oviposition period of 8.40 ± 0.84 days and a total fecundity of 54.10 ± 3.17 eggs (Table 9). In case of 4–5-day-old nymphs, the ovipositional period of female parasitoids was 7.80 ± 1.05 days. The ovipositional period (t = 27.16, p < 0.00001, df =

Table 9 Developmental parameters (mean ± SE) of *D. rapae* females parasitizing different age groups of *M. persicae*

Aphid age groups	*D. rapae*			
	Oviposition period (days)	**Post oviposition period (days)**	**Daily eggs laid/female**	**Total fecundity (eggs/female)**
1–2 days	8.40 ± 0.84 (7–10)	1.40 ± 0.37 (1–2)	5.20 ± 1.21 (4–9)	54.10 ± 3.17 (48–60)
4–5 days	7.80 ± 1.05 (6–10)	1.30 ± 0.35 (1–2)	4.40 ± 1.02 (3–7)	31.60 ± 3.90 (25–40)
t value	27.16*	12.34*	13.62*	15.31*
p value	< 0.00001	< 0.00001	< 0.00001	< 0.00001

Figures in parentheses represent the range
All values are mean of 10 observations
*Significant at $p \leq 0.05$ and df = 18

18), post ovipositional period (t = 12.34, p < 0.00001, df = 18), daily fecundity (t = 13.62, p < 0.00001, df = 18), and total fecundity (t = 15.31, p < 0.00001, df = 18) of the parasitoid was greater when parasitizing younger nymphs of *M. persicae*.

The ovipositional period and fecundity of the parasitoids were less when parasitizing 4- to 5-day-old nymphs than the 1- to 2-day-old nymphs of *M. persicae*. Observations on the ovipositional behavior of *A. asychis* revealed that the older nymphs (4–5 days old) could defend themselves against ovipositing females of the parasitoid more effectively than the younger nymphs (1–2 days old). Therefore, it can be inferred that the younger instars of *M. persicae* were the most suitable age for oviposition to produce the fittest progeny of the parasitoid. Environmental factors like temperature, day length, and size of the adult female also affects the fecundity of the parasitoids (Stary 1988 and Hagvar and Hofsvang 1991). Sengonca et al. (2008) observed longer ovipositional period and high fecundity of *A. asychis* on younger nymphs (1–2 days old) of *A. gossypii* than those on the older nymphs (4–5 days old and adults) of the aphid. Similarly, host feeding by *A. asychis* was significantly more on the younger instars than on the older instars of *M. persicae*. Tatsumi and Takada (2005) found similar results on the cotton aphid, *A. gossypii*, where highly significant differences in host feeding behavior of *A. asychis* were found in distinct age groups (1–2 and 4–5 days old) of the aphid. In accordance to present results, net fecundity rates of *D. rapae* were 40.82 eggs on *B. brassicae* (Hosseini-Gharalari et al. 2003) and 238.7 eggs on *M. persicae* (Fukui and Takada 1988). The differences between the fecundity rates in previous and present studies could be attributed to the difference in the host's and parasitoid strain, host plants, temperature, and photoperiod conditions (Reed et al. 1992 and Bernal and Gonzalez 1993).

Conclusions

Selection of a suitable host age is critical for mass rearing protocols of parasitoids and their utilization in augmentative biological control programs. Biological parameters of the parasitoids were better when parasitized younger aphids. This information can be useful for standardizing the mass rearing and release protocols of these parasitoids to supplement the biological control programs of *M. persicae* in India.

Abbreviations

viz.: Videlicet (namely); RH: Relative humidity; L: Light; D: Dark; ANOVA: Analysis of variance; LSD: Least significant difference; df: Degrees of freedom; i.e.: That is

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Authors' contributions

The author SK carried out the study and designed and performed all the experimental work. SS wrote the manuscript and helped in the statistical analysis of the data. The lead author SK supervised and participated in the planning and implementation of the experiments and reviewed the manuscript. All authors read and approved the final version of the manuscript.

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