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پارازیتیسم و هیپرپارازیتیسم فصلی شته‌ی گردو *Chromaphis juglandicola* (Hom.: Aphididae) در استان تهران

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چکیده

پارازیتیسم و هیپرپارازیتیسم فصلی شته‌ی گردو *Chrompahis juglandicola* (Kalt.) در دو منطقه از استان تهران مورد بررسی قرار گرفت. در مجموع دو زنبور پارازیتویید *Trioxys pallidus* (Haliday) (Hym.: Braconidae) و *Aphelinus asychis* Walker (Hym.: Aphelinidae) و سه زنبور هیپرپارازیتویید *Alloxysta citripes* (Thompson) (Hym.: Cynipidae)، *Pachyneuron aphidis* (Bouche) (Hym.: Pteromalidae) و *Syrphophagus aphidivorus* (Mayr) (Hym.: Encyrtidae) شناسایی و تغییرات فصلی جمعیت آنها ثبت گردید. زنبور *A. asychis* تنها به صورت موردی جمع آوری شد و ظاهراً سازگاری کمی با شتهٔ گردو یا زیستگاه آن دارد، اما زنبور پارازیتویید *T. pallidus* تطابق کاملی به شتهٔ *C. juglandicola* داشته و در کل دورهٔ فعالیت شته حضور داشت. جمعیت این زنبور در طول فصل تغییرات زیادی داشت. در هر دو منطقه، جمعیت زنبور همراه با تغییرات جمعیت شتهٔ میزبان در بهار بسیار بالا رفت و در تابستان ناپدید شد و در پاییز اندکی افزایش یافت. زنبور *T. pallidus* داخل شته‌های مومیایی شده به صورت دیاپوز تابستانه ـ زمستانه یا دیاپوز زمستانه روی برگهای بـه زمین

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ریخته زمستان‌گذرانی می‌کند. فعالیت زنبور *T. pallidus* به شدت تحت کنترل سه گونه زنبور هیپرپارازیتویید بود. زنبور *A. citripes* وابستگی زیادی به زنبور *T. pallidus* دارد، اما دو گونه‌ی دیگر به صورت غیر اختصاصی عمل می‌کنند.

واژگان کلیدی: شته‌ی گردو، پارازیتیسم، هیپرپارازیتیسم، تهران.

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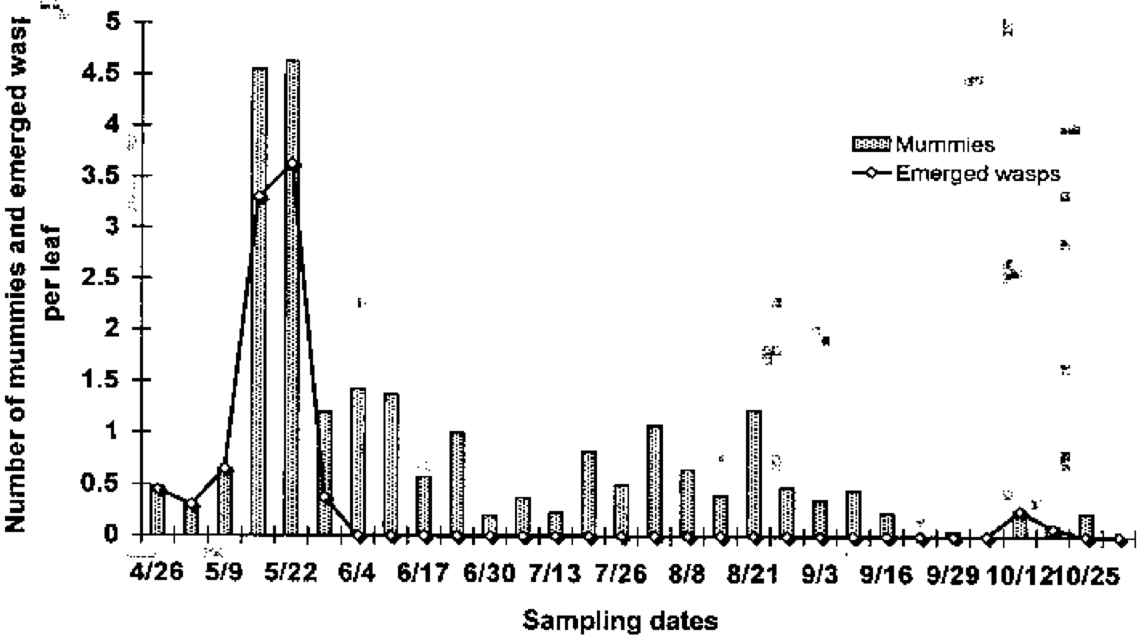

Fig. 5: Seasonal fluctuation in number of mummied aphids and emerged wasps in ARC

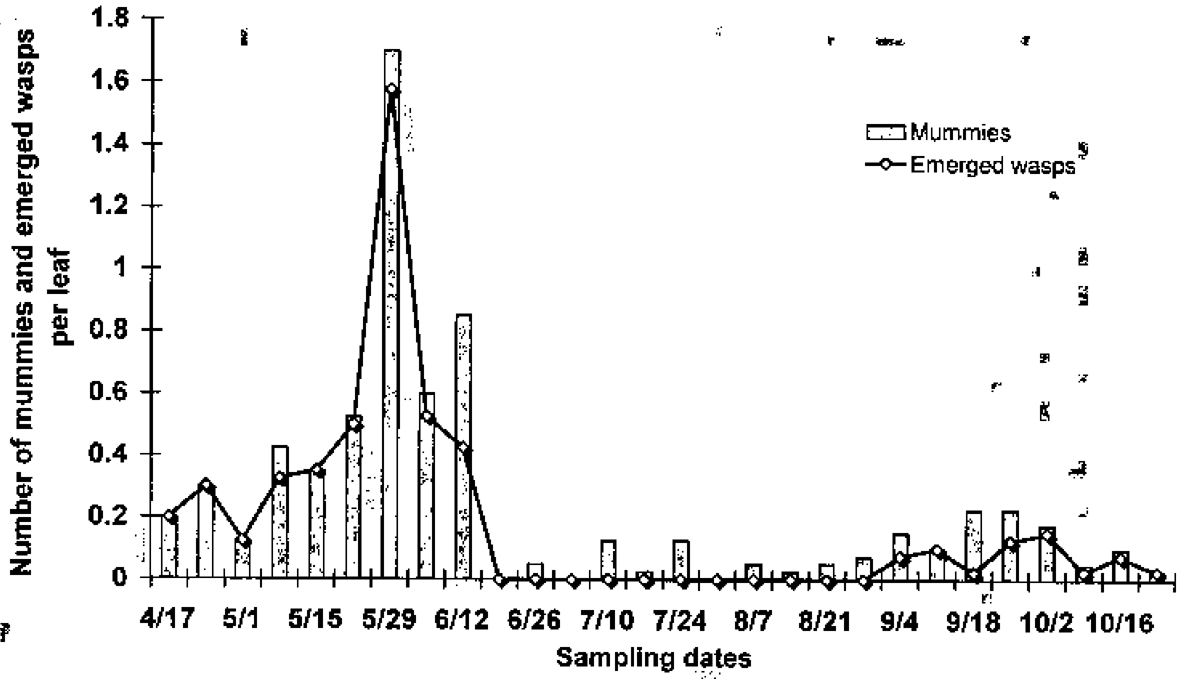

Fig. 6: Seasonal fluctuation in number of mummied aphids and emerged wasps in TCA

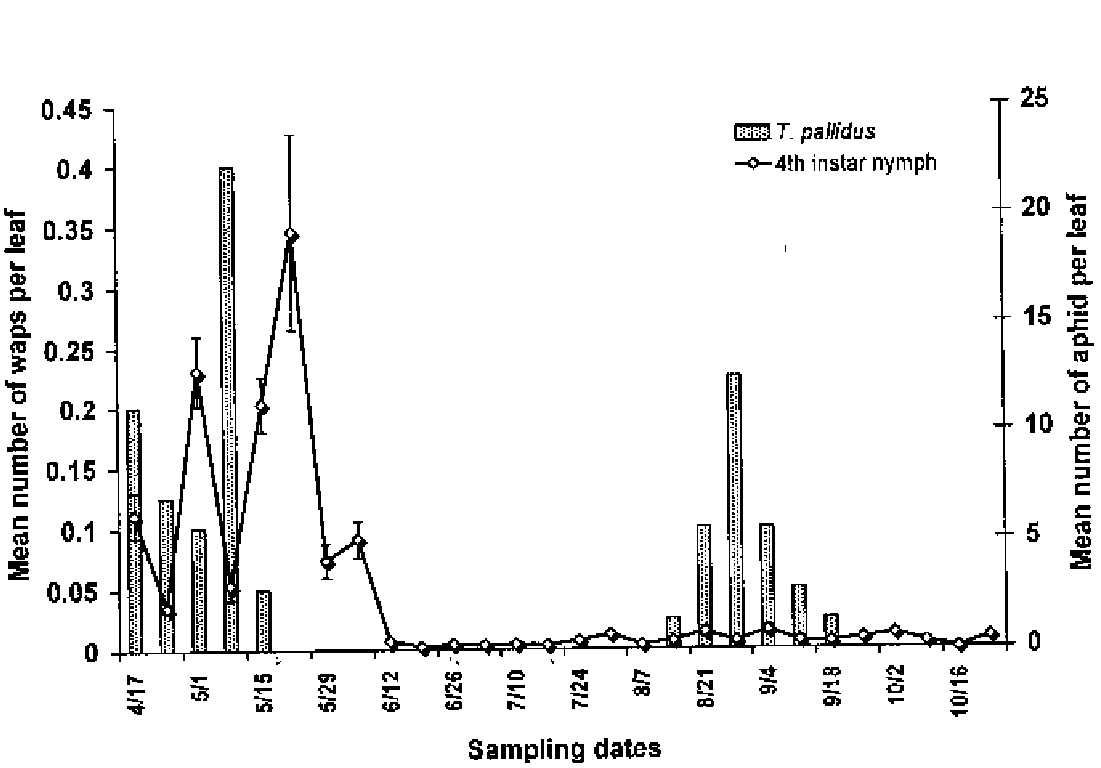

Fig. 3: Seasonal fluctuation in number of 4th aphid instars nymphs and *T. pallidus* in TCA

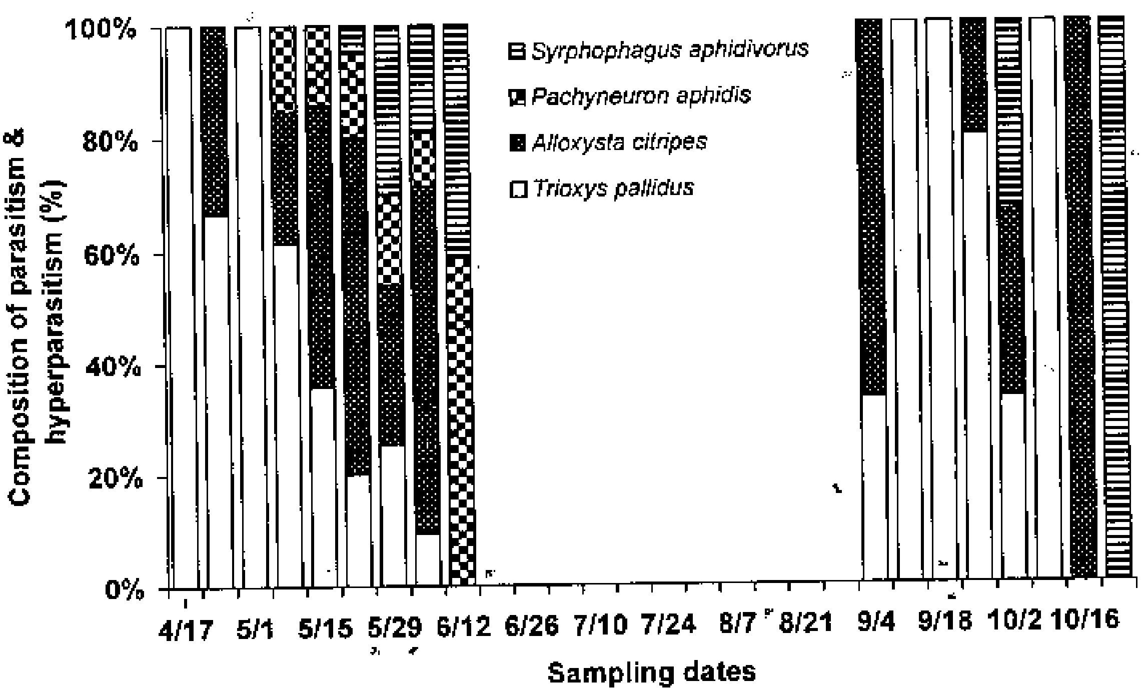

Fig. 4: Seasonal fluctuation in composition of parasitoid and hyperparasitoids of walnut aphid in TCA

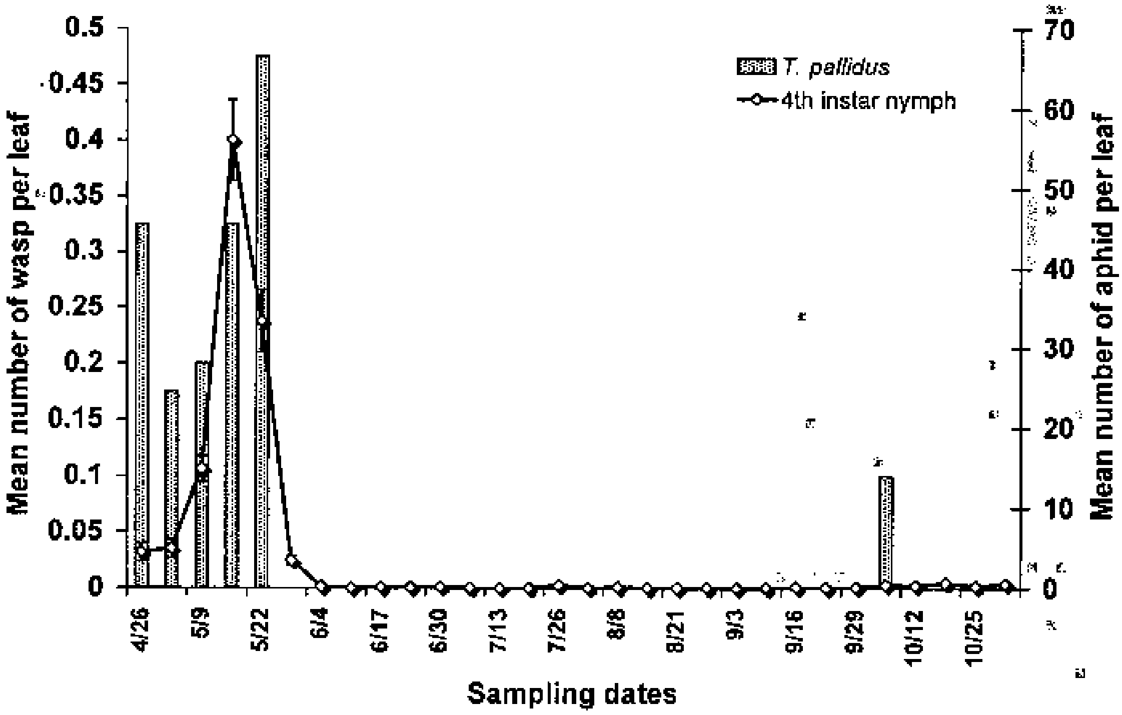

Fig.1: Seasonal fluctuation in number of 4th aphid instars nymphs and *T. pallidus* in ARC

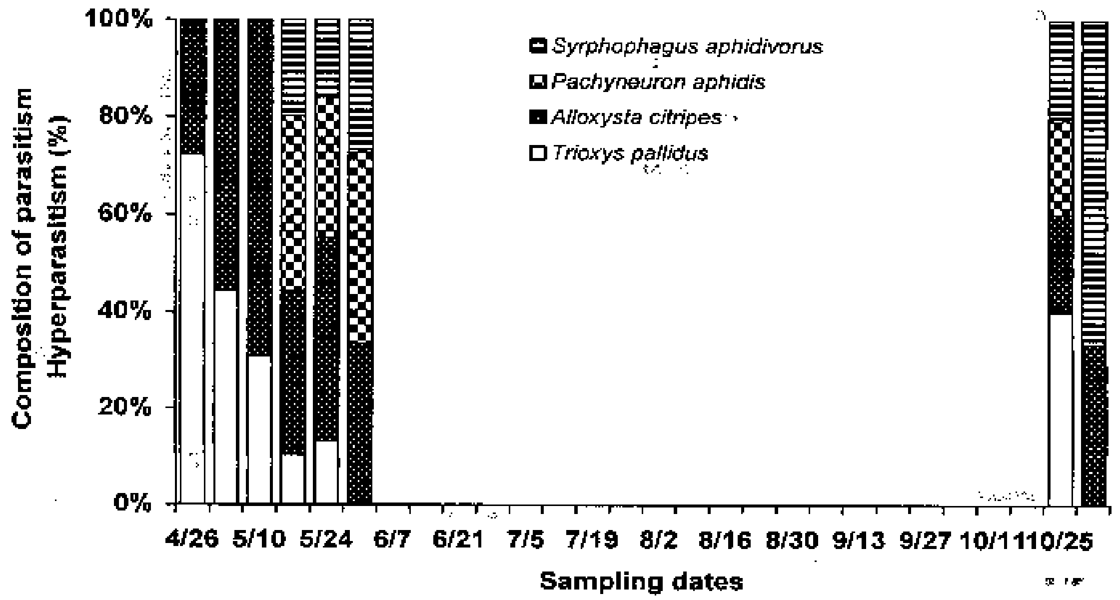

Fig.2: Seasonal fluctuation in composition of parasitoid and hyperparasitoids of walnut aphid in ARC

of primary parasitoid is predictable. Delays in the emergence of the two other hyperparasitoids, *P. aphidis* and *S. aphidivorus* could be described as the low dependency on *T. pallidus*. Both species attack a wide range of hosts and have been observed to emerge from several aphid parasitoids (17).

P. aphidis has been known as a cosmopolitan and polyphagus secondary parasitoid (13) which has been also reported as a hyperparasitoid of walnut aphid in California (14) and Portugal (5). It is worth to note that this species has also been reported previously as a primary parasitoid of olive psylla, *Euphyllura olivina* (Costa) in Iran (10). The third hyperparasitoid was *S. aphidivorus*, which has previously been reported as a hyperparasitoid of walnut aphid from California (14, 15, 16) and Portugal (5).

Generaly, hyperparasitoids seemed to have a negative effect on both population and activity of *T. pallidus*. Kavallieratos *et al.* (9) also showed that the parasitism rate of *Binodoxys angelicae* (Haliday) was negatively influenced by the activity of hyperparasitoids on *Aphis gossypii* Glover, a fact that resulted in the reduced efficiency of parasitism in the *A. gossypii* populations on citrus. Further experimentation is required in order to investigate the role of hyperparasitoids in relation to the populations of *C. juglandicola* and its complex of primary parasitoids.

Diapause is an important factor in the ecology of *T. pallidus*, an effective parasitoid of aphids. Aestival diapause in *T. pallidus* could cause some physiological changes in the host (14). We found that the collected parasitoids in the late spring, remained as mummies and did not produce the next generation in laboratory condition (temperature: 26 ± 2 °C, RH: 60 ± 5 % and a 16L:8D photoperiod). The sharp decline of aphid density and occurrence of diapause in *T. pallidus* caused the disappearance of hyperparasitoids during the summer. Temporal synchronization is largely a function of diapause (14). Synchronization of *A. citripes* and *T. pallidus* is result of diapause in this hyperparasitoid. Our observations of diapause in this hyperparazitoid supported the Brodeur and McNeil's (2) idea: *"Diapause is adaptation of multivoltine species with a short developmental time, permitting the parasitoid to assess adequately seasonal changes in the environment"*.

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T. pallidus. *S. aphidivorus* and *P. aphidis* disappeared from early summer and emerged occasionally from mummies in fall. furthermore, only *T. pallidus* and *A. citripes* emerged from several collected mummies in the next spring.

Discussion

T. pallidus was revealed to be the most important parasitoid of walnut aphid. The host plants of aphids that could be parasitized by *T. pallidus* were not observed in the near surrounding of the walnut orchard studies. It has great degree of efficacy that it is derived from its full spatial and temporal synchronization with its specific host, *C. juglandicola* (14). In our study areas, *T. pallidus* was active during the whole period of the walnut aphid activity in spring and autumn. The early spring population of parasitoid had a significant effect on the aphid density. *A. asychis* was recovered rarely from walnut aphids, therefore, it seemed to be poorly adapted to *C. juglandicola* or its habitat. It has been the first time that *A. asychis* reported as a parasitoid of walnut aphid. Although *C. juglandicola* and *A. asychis* are native to Iran, the interrelation of this host-parasitoid was very weak and parasitism rate was extremely low, which indicated that this aphid was not a suitable host. *Aphelinus perpallidus* Gahan has previously been reported from California as an unadapted parasitoid of walnut aphid (14). Carver and Woolcock (4) demonstrated that *A. asychis* parasitized several genera of Aphididae. In both of our study regions the parasitism by *T. pallidus* was impaired by hyperparasitoids. Among three hyperparasitoids, presented in our study areas, *A. citripes* observed to have the main effect on the efficacy of *T. pallidus*. Our experimental results supported the previous studies by Cecilio and Ilhraco (5), Frazer and van den Bosch (7) and Ozkan and Turkyilmaz (11) which reported hyperparasitism of *C. juglandicola* as an important factor for reducing the efficacy of *T. pallidus*. Evenhuis (6) reared *A. citripes* on *T. pallidus*, and reported this hyperparasitoid species as a specialized parasitoid of *T. pallidus*. Carver (3) mentioned alloxystines as the only true hyperparasitoids of aphids. Furthermore, Andrews (1) also reported *Alloxysta xanthopsis* (Ashmead) as hyperparasitoid emerging from *C. juglandicola* aphids as well as many other hosts. Sullivan (13) mentioned that, in Palearctic regions in which walnut aphid and *T. pallidus* are native, the dominant hyperparasitoids belong to Alloxystinae, whereas in California in which walnut aphid was introduced, the Alloxystinae were rare and their ecological niche were replaced by species of *Syrphophagus*. Since walnut orchards are permanent ecosystems, therefore the high degree of dependency between *A. citripes*, *T. pallidus* and the regulating effects of the hyperparasitoid on population occurred on May and then declined sharply because of the occurrence of the aestival diapause. In contrast to the population of *T. pallidus* occurring in other orchard, this species seemed to have a moderate population in our study area. There was no significant resurgence in the aphid and parasitoid population during summer. Following an increase in the population of aphids in autumn, parasitoids began to emerge from the mummies (figure 3). The presence of parasitoids in autumn was partly overlapped by the appearance of sexual aphids. The mean number of mummified aphids and *T. pallidus* were observed to be 0.24±0.03 and 0.05±0.02 per leaf, respectively, in each season.

The population fluctuation of *T. pallidus* and three hyperparasitoids are shown in figure 4. As other orchard (ARC), hyperparasitism had significant effects on efficacy of *T. pallidus*. *A. citripes* was present during *T. pallidus* activity period. Population of this hyperparasitoid remained in a low density in the first weeks and then increased gradually, to generate up to more than 50% hyperparasitism in mid May. Two other hyperparasitoids, *S. aphidivorus* and *P. aphidis* appeared much later (figure 4). The population of hyperparasitoids appeared in autumn when there was a slight increase as other orchard. The mean percentage of hyperparasitism was observed to be 45.81% and 28.79% in spring and autumn, respectively.

Diapause

In our study regions, *T. pallidus* had two steps of diapause. The first step occurred late in spring with a sharp decrease in the population of walnut aphids. The second step occurred in autumn and last until next spring. Disappearance of *T. pallidus* in late spring seemed to be the result of diapause. All of the collected mummies from both regions had an aestival diapause. Most of the collected mummies remained in diapause until the next spring. As a result, many of the parasitoids did not emerge from the collected mummies in summer and autumn (figures 5 and 6). *T. pallidus* appeared in autumn, when the aphid population was developing again. Hibernal diapause occurred in both parthenogenetic and sexual aphid, but the number of sexual aphids containing the parasitoids was very low. Emergence of *T. pallidus* from diapause occurred in the spring time coincident with the initial activity of aphid, which resulted in an increase in proportion of attacks on aphids

The aestival diapause of *T. pallidus* started when hyperparasitoids began to disappear. Hyperparasitoids became active in fall, shortly after *T. pallidus*. Among the three hyperparasitoid species, *A. citripes* was remarkably synchronized with *T. pallidus*. Occurrence of aestival diapause and emergence in autumn were happened in the presence of

Results

Five species in total were recorded from the studying regions and their population fluctuations were evaluated. Two primary parasitoids including *Trioxys pallidus* (Haliday) (Braconidae: Aphidiinae) and *Aphelinus asychis* Walker (Aphelinidae), and three hyperparasitoids including *Alloxysta citripes* (Thomson) (Cynipidae); *Pachyneuron aphidis* (Bouche) (Pteromalidae) and *Syrphophagus aphidivorus* (Mayr) (Encyrtidae) were identified. *A. asychis* collected occasionally from the TCA samples, but they produced many offspring under the laboratory condition.

Alborz Research Center (ARC): The parasitoids that had emerged from diapause earlier in the spring showed a high activity that increased gradually with increase in aphid population. *Trioxys pallidus* performed a very low activity during the summer and the mummies remained until autumn when a few number of the wasps emerged (figure 1). The mean number of mummied aphids and *T. pallidus* during the season were calculated to be 0.79±0.06 and 0.05±0.02 per leaf, respectively. The seasonal fluctuation of the population of *T. pallidus* and its hyperparasitoids is shown in fig 2. The trend of hyperparasitism was markedly upward. The activity of *T. pallidus* in this region was obviously affected by three hyperparasitoids, *P. aphidis*, *S. aphidivorus* and *A. citripes*, so that the number of emerged hyperparasitoids was often significantly higher than *T. pallidus*. Among the hyperparasitoid wasps, *A. citripes* was strict to *T. pallidus* and was active during the whole activity preiod of *T. pallidus*. The significant effect of *A. citripes* on the population of *T. pallidus* was observed early in the season. The highest population of *A. citripes* occurred (about 1.52 wasps/leaf) shortly after the peak of the aphid population (figure 2). Two other hyperparasitoids appeared with a longer interval, three weeks after the emergence of *T. pallidus* and *A. citripes*. The activation period of this hyperparasitoids continued until end of May. The highest population occurred in the first week of emergence. No parasitoid and hyperparasitoid was emerged from the mummies during the summer and early autumn (figure 2). After this time all of wasps appeared with low densities. The total percentage of hyperparasitism was calculated to be 51.71% and 67.5% in spring and autumn, respectively.

College of Agriculture (TCA): The Seasonal fluctuation in number of 4th instars aphid nymphs and *T. pallidus* is illustrated in figure 3. The population trend of *T. pallidus* was synchronized with the population of the walnut aphid. The peak population of the parasitoid

Introduction

The walnut aphid, *Chromaphis juglandicola* (Kaltenbach) has long been recognized as a serious pest of Persian walnut (*Juglans regia* L.) in Iran and some other parts of the world (5, 7, 11, 12, 14, 15). An ecotype of *Trioxys pallidus* (Haliday) was successfully imported from Iran to California, as a biological control agent, and rapidly decreases the aphid population (7, 16). Although van den (14) reported only *T. pallidus*, as a parasitoid of walnut aphid, there were several active hyperparasitoids present. Moreover, Cecilio & Ilharco (5) also reported some hyperparasitoids of walnut aphid from Portugal. Although there are well established data concerning the relations between *T. pallidus* and its related aphid hosts, very little information is available for estimating the population of parasitoids on *C. juglandicola* since these data might be useful for evaluating their efficiency in biological control programs.

In this study, we experimentally investigated the seasonal abundance of the primary parasitoids of *C. juglandicola* as well as hyperparasitoids, attacking those primary parasitoids.

Materials and Methods

The study was conducted in two walnut orchards, Alborz Research Center (ARC) and College of Agriculture of Tarbiat Modarres University (TCA) which located in Karadj and the west of Tehran respectively. These two stations are 40 km. far from each other. Ten trees, randomly chosen, were sampled weekly. The number of the healthy and parasitized aphids was counted for four middle compound leaves, randomly chosen from cardinal points of each tree. The aphid infested leaves were collected and transferred to the laboratory. The aphids were reared in growth chamber (temperature: 26 ± 2 °C, RH: 60 ± 5 % and a 16L:8D photoperiod) until parasitoids emerged. The emerged wasps, except for *T. pallidus*, were examined for the determination of parasitoid or hyperparasitoid type. Aphids, wasps and walnut leaves were caged together, using plastic-clip cages (10 cm in diameter), to allow parasitization. If parasitism was observed, then the wasp was considered to be a primary parasitoid; otherwise the experiments were continued. Groups of different stages of *C. juglandicola* nymphs which were parasitized by *T. pallidus* prepared. Two, four and six days after the initial parasitiziation, the aphids were transferred to petri dishes, and a female tentative wasp was introduced into the petri dishes and its behaviour was observed under a stereomicroscope for up to 30 minutes. If oviposition behaviour was observed and offspring were produced, then the examined parasitoid considered to be a hyperparasitoid.

Seasonal Parasitism and Hyperparasitism of Walnut Aphid, *Chromaphis juglandicola* (Hom.: Aphididae) in Tehran Province

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Abstract

Seasonal parasitism and hyperparasitism of walnut aphid, *Chromaphis juglandicola* (Kaltenbach) were studied in Tehran. Five species including two primary parasitoids, *Trioxys pallidus* Haliday (Hym.: Braconidae: Aphidiinae) and *Aphelinus asychis* Walker (Hym.: Aphelinidae) as well as three hyperparasitoids, *Alloxysta citripes* Thompson (Hym.: Cynipidae), *Pachyneuron aphidis* (Bouche) (Hym.: Pteromalidae) and *Syrphophagus aphidivorus* (Mayr) (Hym.: Encyrtidae) were identified and the seasonal fluctuations in their population were accounted. *Trioxys pallidus* appeared to be fully synchronized and adapted to *C. juglandicola*, but *A. asychis* was recovered on occasion, and it seems poorly adapted to walnut aphid or its habitat. The population of *T. pallidus* was observed to be fluctuated during the season. Population fluctuation of the parasitoid marked by a spring population peak, following changes in its host densities, and a summer disappearance, which was followed by a weak autumnal increase. *Trioxys pallidus* over winters as summer-winter or winter diapause mummies on the soil after leaf fall. The efficacy of *T. pallidus* revealed to be restricted mainly by *Alloxysta citripes* which seemed to be a specialized hyperparasitoid of this wasp species. The two other hyperparasitoids, *Pachyneuron aphidis* and *Syrphophagus aphidivorus* were seemed to be more generalist.

Key words: *Chromaphis juglandicola*, Parasitism, Hyperparasitism, Tehran.

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