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First Record of the Predatory Insects, *Androthrips ramachandrai* (Karny) (Thysanoptera: Phlaeothripidae) and *Montandoniola indica* Yamada (Hemiptera: Anthocoridae) on the Pest Thrips, *Gynaikothrips ficorum* (Marchal) (Thysanoptera: Phlaeothripidae) in Turkey

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ABSTRACT

The Cuban laurel thrips, *Gynaikothrips ficorum* (Marchal) (Thysanoptera: Phlaeothripidae), is widely distributed to the eastern Mediterranean region of Turkey and causing damage to *Ficus microcarpa var. nitida* plants. Biological control of *G. ficorum* has a crucial role to maintain the population density of this pest insect at an acceptable level on *Ficus* trees. For this reason, to determine the potential biological control agents of this pest, survey studies have been done in Adana and Mersin provinces. The *Androthrips ramachandarai* (Karny) (Thysanoptera: Phlaeothripidae), and the *Montandoniola indica* Yamada (Hemiptera: Anthocoridae) were reported for the first time in Turkey in 2020 on *G. ficorum* that damaging young leaves of the *F. microcarpa*. Brief diagnoses of *G. ficorum* and both predatory species were provided. A total of 249 adults of *A. ramachandrai* and 461 individuals of *M. indica* were detected in the galled leaves infested with *G. ficorum*. Moreover, 1959 adults, 1335 immatures and 11851 eggs of *G. ficorum* were also recorded. There was a positive relationship between pest eggs-*A. ramachandrai* adults and pest larvae-*M. indica* adults. Ratios of prey/predatory insects indicated that the predators might have the capability in suppressing the pest thrips on *Ficus* trees.

Key words: Ficus, predator, relationship, thrips, Turkey.

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INTRODUCTION

Following the extensive damage of palm trees (*Phoenix canariensis* hort. ex. Chabaud) by the red palm weevil, *Rhynchophorus ferrugineus* (Olivier) (Coleoptera: Curculionidae) to in Adana and Mersin provinces, numerous trees were stubbed, and then considerable numbers of *F. microcarpa* trees have been planted in both cities in the past decade (Atakan & Pehlivan, 2019). With the trade of the *Ficus* trees and an increase in the number of the host plants of Cuban laurel thrips, *Gynaikothrips ficorum* (Marchal) (Thysanoptera: Phlaeothripidae), which was recorded for the first time on *Ficus* sp. in Mersin Province, Turkey (Toros & Tunç, 1988), has widely distributed to the region (Atakan & Pehlivan, 2019) recently. This species is a monophagous, gall-forming thrips (Denmark, Thomas, & Funderburk, 2005) and heavily causes damage to various ornamental figs, *Ficus* spp. (Moraceae), throughout the tropical and temperate climates (Paine, 1992). This thrips reduce the photosynthetic activity of the plants by inducing galls and depreciate their ornamental value due to curled and discolored leaves (Piu et al, 1992).

Control of the pest thrips species mostly relies on using synthetic insecticides (Held & Boyd, 2008). However, applying insecticides is restricted because *Ficus* spp. has decorated both outdoor and indoor landscapes in which these are the people's living areas. Moreover, the use of broad-spectrum insecticides can develop pest resistance and affect the role of natural enemies (Arthurs, Chen, Dogramaci, Ali, & Mannion, 2011; Tavares, Torres, Silva-Torres, & Vacari, 2013). A strong association has been reported with the predators including the thrips egg predator *Androthrips ramachandrai* (Karny) (Thysanoptera: Phlaeothripidae), hemipterans such as *Montandoniola confusa* (=previously identified as moraguesi in some areas) Streito & Matocq, *Montandoniola indica* Yamada (Hemiptera: Anthocoridae). Gall formed by thrips represents a microcosm that hosting all stages of pest thrips and their natural enemies (Paine, 1992; Boyd & Held, 2006; Cambero-Campos, Valenzuela-García, Carvajal-Cazola, Rios-Velasco, & García-Martínez, 2010; Arthurs et al, 2011; Yamada, Bindu, Nasreem, & Nasser, 2011; Tavares et al, 2013).

Although many studies reported the interactions between *G. ficorum* and predatory insects, until this time there is no record from Turkey. With this study, descriptions of the predatory thrips, *A. ramachandarai* (Karny), and the hemipteran predator, *M. indica*, first recorded in Turkey were given. Additionally, the mean and total numbers of pest thrips and predatory insects and their interactions on the *Ficus* trees were summarized.

MATERIAL AND METHODS

Surveying area and insect sampling

Three different sampling locations were chosen for surveys of insects inhabited trees of the *Ficus microcarpa var. nitida* in outdoor landscapes of Adana and Mersin Provinces, Turkey on August 2020. I: University Campus (37°03'41.3"N, 35°21'30.7"E) which is a natural habitat, and *Ficus* trees are grown together with citrus (*Citrus aurantium*) as ornamental; II: Adana City Center (36°59'48.8"N, 35°19'23.7"E) urban area; III: Mersin Province, Turkey (Seaside) (36°05'48.4"N, 33°01'52.7"E) in which woody and herbaceous ornamental plants are grown together. Both Adana and Mersin Provinces are located in the eastern Mediterranean region of Turkey. Mersin Province is about 80 kilometers far from Adana city, and it is a seaside province. Both cities have a typical Mediterranean climate. Ornamental arboreal plants are grown mainly on the University Campus. Plant species diversity is greater on the Campus than in parks of both cities. These woody plants are scattered over the Campus. In the central parks of both provinces, woody and herbaceous ornamental plants are grown together. *Ficus* trees are densely grown in the central parks in Adana than in Mersin. Ten *F. microcarpa* plants in each location were chosen and ten leaves with galls were collected from each plant randomly. The galls were put into the plastic bags and a total of 100 galls for each sampling location were evaluated. Samples were taken from the *Ficus* trees at 08:00-10:00 hours in July. All specimens were kept in 70-80% ethanol just after collecting. Thrips and anthocorid specimens were counted individually under a stereomicroscope (Olympus SZ51) with 45x magnifications.

Identification of insects

Thrips specimens were kept in the AGA (9 part 60% ethyl alcohol, 1 part glacial acetic acid, and 1 part glycerin) solution for two days and then, the specimens were taken into 5% NaOH media, and thrips specimens were kept in this media until a slight color change occurred on their bodies. The specimens were taken to the Hoyer media and slide-mounted.

Following the collection of *M. indica*, the specimens were dried and mounted for observation of their genitalia. For examination and illustration of genitalia, the specimens were macerated in 5% hot KOH solution until the organs became transparent. They were dissected with micro-pins and forceps in glycerin on a glass slide under a binocular microscope (Nikon Stereoscopic Zoom Microscope SMZ1500). Then, the samples were taken to the Hoyer media and mounted. The identification of *M. indica* was made by using the keys Carayon (1961), Yasunaga (1997), and Yamada et al (2011). Thrips specimens (*G. ficorum* and *A. ramachandrai*) were identified by the second author, and dissections of *M. indica* female and male genitalias' were done by the fourth author.

Statistical analysis

Eggs, nymphs, and adults of *G. ficorum* were counted under the stereomicroscope with 45x magnifications. Only adults of *A. ramachandrai* were considered in insect counting because a few numbers of larvae of the predatory thrips were noted. The nymphs and adults of the predatory hemipteran insect were counted and recorded. Seasonal total numbers of each species for each sampling location were given in Table 1. Total prey (pest thrips): predatory insect (predatory thrips or predatory bug) ratios are calculated separately for each predatory insect species and ratios were summarized in Table 2. Generalized Linear Model (GLM) analysis was used to determine the effects of sampling location and trees on the abundance of thrips and predatory insects. According to GLM, different stages of *G. ficorum* were dependable variables. Location was a fixed factor, replicates were a random factor and predatory insects were covariates. Kolmogorov-Smirnov test and Levene's test were used to determine normality and homogeneity of variance, respectively. One-way analysis of variance (ANOVA) was conducted on mean insect numbers at different locations, followed by Turkey's post hoc mean separation. Moreover, Quadratic Regression Analyses were done to determine the relationship between mean numbers of predatory insects and mean numbers of different stages of *G. ficorum* at importance level of P<0.05.

Table 1. Mean and total numbers of *Gynaikothrips ficorum* and their predators on *Ficus microcarpa* per gal in three sampling locations in 2020.

Location	*Gynaikothrips ficorum*						*Androthrips ramachandrai*		*Montandoniola indica*	
	Adults		Nymphs		Eggs					
	Mean	Total	Mean	Total	Mean	Total	Mean	Total	Mean	Total
University Kampüs	10.5 ± 1.47a	1050	7.62 ± 1.66a	762	49.44 ± 4.11b	4944	1.07 ± 0.29b	107	3.16 ± 0.41a	316
Adana City Center	6.07 ± 1.15b	517	5.39 ± 0.95a	539	5.70 ± 1.62c	570	1.42 ± 0.23a	142	0.08 ± 0.03c	8
Mersin (Seaside)	3.92 ± 0.49b	392	0.34 ± 0.17b	34	63.37 ± 4.86a	6337	0.00 ± 0.00c	0	1.37 ± 0.23b	137
Total	-	1959	-	1335	-	11851	-	249	-	461

*Means with same letter in the same column are not statistically significant according to Tukey's HSD test (P < 0.05).

RESULTS

Characterizations of pest, predatory thrips, and predatory bug

Gynaikothrips ficorum was reported for the first time on *Ficus* in Turkey by Toros and Tunç (1988), with descriptions in detail. This published paper and also a key provided by Priesner (1939) were used to identify the specimens of the pest thrips collected in the present study. Adult females of *G. ficorum* are black in color (Fig. 1A). Fore tibiae are yellow, tips of mid and hind tibiae are light yellow, and all tarsus is light yellow. The first and second antenna segments are dark, the rest are light yellow, the apical half of the 7^{th} antenna, and the 8^{th} antennal segments are in gray. The wings are transparent in color. Head length is 1.3 times its width. Postocular setae is short, does not protrude from the side of the head. Setae of the anterior corner of the prothorax vary between 40-50 microns, but epimeral setae between 135-150 microns. Metanotum and anterior abdominal segments are reticulate. The lengths of anal setae in the abdomen are 275-290 microns.

Adult males are smaller than females. The tenth abdominal segment is shorter. The forelegs are weak; the toothed protrusion in the tarsus is smaller than that of the female.

Larvae and pupae are transparent and sometimes light yellowish. Wing protrusions are evident in pupae. Antennas elongated backward (Fig. 1B).

Table 2. Relationship among numbers of *Gynaikothrips ficorum* and their predators.

Locations	Predators	Pest stages	R^2	F	P	Equations
Adana City Center	*Androhrips ramachandrai*	Egg	0.7448	10.217	0.008	$y = 14,22x^2 - 32,838x + 19,426$
		Nymph	0.1488	0.612	0.569	$y = 2,5489x^2 - 8,3724x + 11,381$
		Adult	0.5277	3.911	0.072	$y = -4,052x^2 + 12,653x - 3,4215$
	Mantondoniola indica	Egg	0.402	2.353	0.165	$y = 633,75x^2 - 79,125x + 4,425$
		Nymph	0.1216	0.485	0.635	$y = -98,75x^2 + 9,375x + 5,825$
		Adult	0.0394	0.144	0.869	$y = -16,25x^2 - 1,375x + 5,475$
University Campus (in Adana)	*Androhrips ramachandrai*	Egg	0.4366	2.712	0.134	$y = 39,254x^2 - 128,84x + 120,45$
		Nymph	0.0147	0.052	0.950	$y = 0,4959x^2 - 1,9436x + 8,8551$
		Adult	0.1851	0.795	0.489	$y = -3,0433x^2 + 6,8873x + 8,3134$
	Mantondoniola indica	Egg	0.0022	0.008	0.992	$y = 1,2022x^2 - 8,1087x + 61,695$
		Nymph	0.3521	1.902	0.219	$y = -1,3092x^2 + 9,8097x - 8,8203$
		Adult	0.4655	3.048	0.112	$y = -2,477x^2 + 14,788x - 8,6847$
Mersin (Seaside)	*Androhrips ramachandrai*	Egg	-	-	-	-
		Nymph	-	-	-	-
		Adult	-	-	-	-
	Mantondoniola indica	Egg	0.4834	3.275	0.099	$y = -10,527x^2 + 48,88x + 19,764$
		Nymph	0.5245	2.775	0.034	$y = 0,7343x^2 - 1,3816x + 0,6033$
		Adult	0.3588	1.959	0.211	$y = 0,1566x^2 + 1,4869x + 1,5355$

“-”predatory thrips was not detected.

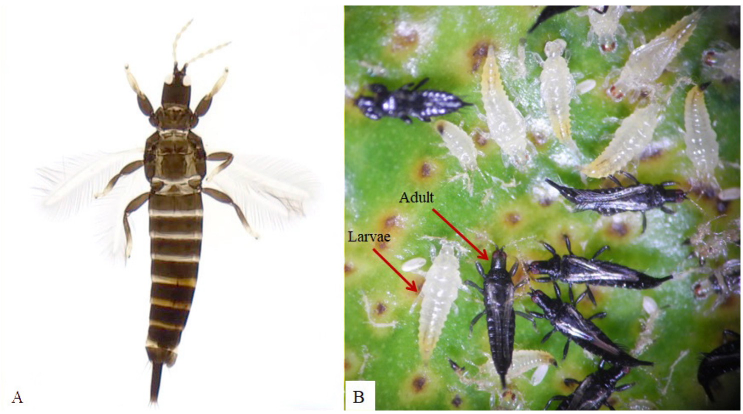

Fig. 1. Dorsal view of *Gynaikothrips ficorum*, (A) slide mounted female, (B) natural view of various biological stages.

There are about 40 species in the genus *Gynaikothrips*, the majority of which have been described in southeastern Asia. This genus is poorly defined and the species distinction is often difficult. *Gynaikothrips ficorum* and *Gynaikothrips uzeli* (Zimmerman, 1990) (Thysanoptera: Thripidae) are very similar species to each other. *Gynaikothrips ficorum* is distinguished from the *G. uzeli* species by having the pronotal posteroangular setae being longer than the discal setae (Mound et al, 1996).

The key provided by Cavalleri, Linder, & Mendonça et al (2016) was used to identify *Androthrips*. Female of the predatory thrips (*A. ramachandrai*): Body and legs brown, tarsi yellow, also fore tibiae and apices of mid and hind tibiae; antennal segment III yellow, IV-VI yellow with apex variably brown; fore wings pale. Antenna with 8 segments, segment III with 3 sensoria, IV with 4 sensoria. Pronotum with no sculpture, 4 pairs of major setae present, anteromarginals minute. Fore femur often enlarged, with rounded tubercle on inner margin near the base; fore tarsal tooth often large (Fig. 2). Male smaller; tergite IX setae S2 short and stout. The genus *Androthrips* includes 12 species, 11 of which have been described in Tropical Asia. The species are morphologically similar to each other. *A. ramachandrai* has brown tibias. In contrast, yellow tibia is found in *A. monsterae* Moulton (Mound & Minaei, 2007).

The general shape and structure of genitalia in obtained specimens from Turkey have almost the same as the type specimens of *M. indica*. The male paramere has a long and very slightly sinuate flagellum which is approximately twice as long as the cone (Fig. 3A). The female mating tube is much longer and its apex is reaching or exceeding the anterior margin of sternum VII (Fig. 3B). Copulatory tube in obtained specimen from Turkey is proximate to the base of the ovipositor; this condition a little differs from those of type specimens (Yamada et al, 2011). The difference in the copulatory tube between Turkish specimens and Indian specimens might be a geographic variation or individual variation. The type specimens have been compared with by the last author; it concluded that these species agree with *M. indica*. Type specimens are now deposited in the Tokushima Prefectural Museum where the last author is.

Total numbers of insect species identified

With this study, totally 249 adults of *A. ramachandrai* and 461 individualls of *M. indica* were detected in the galls infested with *G. ficorum*. Moreover, 1959 adults, 1335 immatures and 11851 eggs of *G. ficorum* were also recorded. Galled leaves collected from trees in Çukurova University Campus had the highest number of prey thrips and predatory insect species. Relatively high number of predatory thrips was recorded on galled leaves collected from *F. microcarpa* in Adana city center. In Mersin, no predatory adult thrips was found in the galls, but hemipteran predators were determined (Table 1).

Prey: predator interactions

According to GLM analysis, location is the most important factor that can affect the abundance of pest thrips and predatory insect species. Moreover, there was a positive interaction between pest eggs-*A. ramachandrai* adults in Adana City Center and pest nymph-*M. indica* adults in Mersin (Seaside) (Table 2). While no *A. ramachandrai* were found in the sampling area in Mersin, the egg numbers of harmful thrips was found to be higher than in the other sampling area. Similarly, in Adana City Center, predator thrips was at the highest level, while the harmful thrips eggs remained at the lowest level.

Fig. 2. Dorsal view of slide mounted female of *Androthrips ramachandrai*.

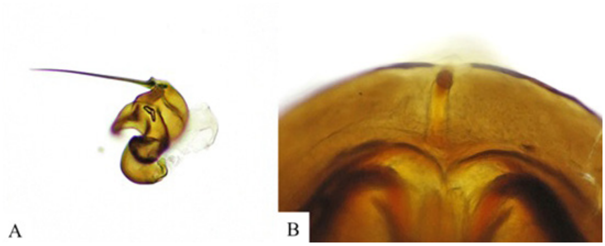

Fig. 3. Male paramere (A) and female copulatory tube (B) of *Montandoniola indica*.

In the current study, the ratios of the thrips per predatory species are shown in Table 3. Since *A. ramachandrai* adults were determined mostly in Adana City Center, the rates of prey per predator were the lowest. Since *M. indica* was detected extremely low in the Adana city center, the ratios of the *G. ficorum* per predator were the highest. *Montandoniola indica* was found with lower rates in the other survey areas. The ratios of mobile stages of *G. ficorum* per predators' *A. ramachandrai* and *M. indica* on galled leaves of *F. microcarpa* in the University Campus were under the 1:10 and 1:5 respectively. In the Adana City Center, these ratios were around 1:5 and 1:71 per predator. And also, the ratios of mobile stages of *G. ficorum* per *M. indica* were under 1:3 in Mersin (Seaside).

Table 3. The ratios of *Gynaikothrips ficorum* per predators (*A. ramachandrai* and *M. indica*) on galled leaves of *F. microcarpa* in three sampling locations in 2020.

Locations	*Androthrips ramachandrai*			*Montandoniola indica*		
	Prey* adults	Prey larvae	Prey eggs	Prey Adults	Prey Larvae	Prey Eggs
University Kampüs	9.81	7.12	46.21	3.32	2.41	15.65
Adana City Center	3.64	3.80	4.01	64.63	67.38	71.25
Mersin (Seaside)	-	-	-	2.86	0.25	46.26

*Prey= *Gynaikothrips ficorum*; "-"predatory thrips was not detected.

DISCUSSION

In The Cuban laurel thrips has been widely distributed with the ornamental plant marketing of decorative *Ficus* spp. throughout all continents, except Antarctica (Mound, 2009). This thrips represents a microcosm and hosts all stages of pest thrips and their natural enemies, such as *A. ramachandrai* and *Montandoniola* spp.. The strong associations were determined with the populations of gall-forming thrips, *G. ficorum*, *G. uzeli* and their natural enemies (Paine, 1992; Boyd & Held, 2006; Cambero-Campos et al, 2010; Arthurs et al, 2011; Yamada et al, 2011; Tavares et al, 2013). Following the trade of the *Ficus* trees and an increase in the number of the host plants of *G. ficorum*, this pest thrips has been widely distributed to the region recently and also cause damage to *F. microcarpa* in outdoor landscapes (& Pehlivan, 2019). Although many studies reported the interactions of the natural enemies of *G. ficorum*, until this time there is no record from Turkey. With the current study, *A. ramachandrai* and *M. indica* were reported for the first time in Turkey in 2020. *Gynaikothrips ficorum* and its natural enemies can be introduced with the horticultural trade of *Ficus* trees in Turkey (Atakan & Pehlivan, 2019). Çukurova University Campus had the highest number of prey thrips and predatory insect species. In University Campus representing rich plant and insects' biodiversity, where no insecticide application against the pest thrips on the ficus trees was performed. Therefore we assumed that *G. ficorum* and its natural enemies were more abundant in comparison to other habitats surveyed.

With the current study, there was a positive relationship between the mean numbers of *G. ficorum* eggs and *A. ramachandrai* adults in Adana City Center. Although the Metropolitan Municipality has applied insecticide to control *G. ficorum*, the predator thrips numbers were the highest. There may be different reasons for the presence of many predatory thrips in the Central park of Adana. At least, the high number of Ficus trees in this sampling unit may have positively affected the population development of predatory thrips as well as the harmful thrips. Genus *Androthrips* includes 12 species (Mound, 2013) and is often considered to be predators on the other gall-forming thrips species in Asia (Varadarasan & Ananthakrishnan, 1981; Sureshkumar & Ananthakrishnan, 1987). *Androthrips ramachandrai* was described from India and found in associate with galls formed by the *Austrothrips cochinchinensis* Karny (Thysanoptera: Phlaeothripidae) on *Calycopteris floribunda* (Roxb.) Lam. (Combretaceae) (Anantakrishnan, 1978). Moreover, this predatory thrips has been reported as an important predator of *Gynaikothrips*, and many researchers have observed *A. ramachandrai* feeding on eggs, larvae, and pupae of *G. uzeli* and *G. ficorum* in galls of *Ficus* spp. (Boyd & Held, 2006; Held & Boyd, 2008; Cavalleri et al 2011; de Melo, Cavalleri, & Mendonça, 2019).

In this study, *M. indica* was widely distributed in the sampling areas with its prey *G. ficorum*. There was a positive interaction between the numbers of pest nymphs and *M. indica* adults in Mersin (Seaside). And also, predator:prey ratios among the mobile stages of the pest thrips and the predatory bug were less than 1:4 in University Campus and Mersin (Seaside) indicating that harmful thrips was at higher risk of predation due to the *M. indica.* The relatively higher numbers of predatory bug in parks in Mersin compared to that of Adana may be related to presence of different ecological factors, such as climate, plant diversity, and whether there is an insecticide application or not.. *Montandoniola* spp. are the predators of family Phlaeothripidae (Insecta: Thysanoptera) including five species belonging to the genus *Gynaikothrips* (Dobbs & Boyd, 2006; Yamada et al, 2011). An anthocorid described as *M. moraguesi* (before taxonomic revision) has been intentionally released for classical biological control of *G. ficorum* from the Philippines to Hawaii and Bermuda, achieved long-term control on outdoor plantings of ornamental *Ficus* spp. (Davis & Krauss, 1966; Funasaki, 1966; Cock, 1985). Despite the availability of different life stages of *G. ficorum* in the gall, *M. confusa* adults preferred to prey upon on thrips eggs, with an estimated 10-fold greater predation on eggs compared to other stages of thrips (Tavares et al 2013). Besides, Arthurs et al (2011) showed that *M. confusa* reproduced throughout the year and reduced *G. uzeli* population's ≥95% and leaf galls by up to 77% within 5 weeks on three *F. benjamina* cultivars in greenhouses. Recently, Yamada et al (2011) examined the Indian specimens of *Montandoniola* and identified *M. indica* as a new species which was associated with the gall-forming thrips, *Liothrips karnyi* Bagnall, 1924 (Thysanoptera: Phlaeothripidae), on black-pepper, *Piper nigrum* Linnaeus (Piperaceae), in southern India. Recently, Ali & Streito (2019) have been detected two heteropterous species *M. indica* and *Geocoris amabilis* Stål, 1855 (Heteroptera: Geocoridae) on the leaves of *Ficus benjamina* which was infested with *G. uzeli*, in the coastal area in Tartous, Syria. This was the first detection of *M. indica* outside India. Tartous province is only 400 km far from Adana province, Turkey. Although there has been limited information about the distribution area of *M. indica*, the predatory bugs are potentially much wider than the only localities reported by Yamada et al (2011). It is not possible to know whether it originates from Turkey, Syria, or is introduced. Moreover, Yamada et al (2011) indicated that 1st instar of the predatory bug preferred eggs and larvae of thrips, 2nd instar nymphs fed on thrips larvae as well, whereas older nymphal stages and adults of *M. indica* fed mainly on adult thrips. Ballal, Gupta, & Sunil (2012) reported that *M. indica* can be successfully reared on the rice moth, *Corcyra cephalonica* (Stainton, 1866) (Lepidoptera: Pyralidae) eggs, and also the mass-produced predator can be evaluated as a biological control agent of *L. karnyi* infesting black pepper in India.

In this study, the ratios of different biological stages of *G. ficorum* per the predators'; *A. ramachandrai* and *M. indica*, on galled leaves of *F. microcarpa* were ranged between 1:3 and 1:71 at different surveying locations. There was no study related to the predator: prey interactions among this species. However, Sabelis & van Rjin (1997) reported that a predator: prey ratio of 1:217 may be sufficient in the long term to control thrips by anthocorid predators, while thrips can be suppressed in a shorter time at a ratio of 1:51. Similarly, Funderburk, Stavisky, & Olson (2000) also determined that the 1:40 ratio was sufficient for *O. insidiosus* to control the *F. occidentalis* population in pepper fields in a short time. In the light of these studies, the predators might be an important factor to control *G. ficorum* on *Ficus* trees in Adana and Mersin Provinces.

In conclusion, this study deals with the first records of *A. ramachandrai* and *M. indica* in galled leaves infested by the *G. ficorum*. However, with the limited number of samplings in the locations, these results indicate that both predatory insect species may play a crucial role in suppressing the pest thrips on *F. microcarpa* growing in natural habitats such as the University Campus. This situation is more important in terms of the protection of the environment and human health. The control of this harmful thrips species on Ficus trees grown in the urban landscapes by using pesticides could be difficult due to its cryptic and feeding behaviors, and pesticides may also pose more risks to the environment and human health. Therefore, the role of natural enemies to achieve long-term control of this pest should be investigated with the laboratory and field experiments such as mass releases of the predatory insects detected in the region. Moreover, the population dynamics and the interactions of the pest thrips and its predators in different habitats hosting Ficus trees should be studied in a more detail in the following years.

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