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Biology and ecology of *Asphondylia coridothymi* (Diptera: Cecidomyiidae) inducing galls on *Coridothymus capitatus* on the island of Samos, Greece

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Abstract. *Asphondylia coridothymi* Skuhravá, 2011 galls the flower buds of *Coridothymus capitatus* (L.) (Reichenb.) (Lamiaceae). Only one generation develops per year. Adults emerge from mid March to mid June. Females usually lay one egg in a flower bud in which the larva develops. Adult life is very short, at most five days. Larvae develop in galls from June to October. Pupae hibernate in galls until spring the following year. Mycelium of a symbiotic fungus of the genus *Aureobasidium* (Basidiomycetes) covers the internal walls of the gall chamber. Seven species of parasitic Hymenoptera, belonging to five families of the superfamily Chalcidoidea, were recorded emerging from galls of this gall midge: *Systasis encyrtoides* Walker, 1834 (Pteromalidae), *Aprostocetus* sp., *Tetrastichus* sp., one unidentified species of the subfamily Tetrastichinae (Eulophidae), *Ormyrus* sp. (Ormyridae), *Eupelmus* sp. (Eupelmidae) and *Eurytoma* sp. (Eurytomidae). These parasitoids attack and kill gall midge larvae in galls. The percentage mortality of larvae of *Asphondylia coridothymi* in galls caused by unfavourable climatic conditions is 23.3% in semi-mountainous and 37.6% in lowland regions. *Asphondylia coridothymi* by galling the flower buds of *Coridothymus capitatus* prevents the development of seed and cause other flowers in galled inflorencesces to be infertile. Based on the reduction in seed production due to galling *Asphondylia coridothymi* is an important pest of *Coridothymus capitatus*. The damage it causes in lowland regions measured in terms of inflorescences bearing galls is from 3.3 to 6% and in semi-mountainous regions from 19 to 30%, which in terms of reduction in the quantity of seed produced by *Coridothymus capitatus* is 0.46 to 3.28% in lowland regions and 17.67 to 25.28% in semi-mountainous regions.

Key words. Diptera, Cecidomyiidae, *Asphondylia coridothymi, Coridothymus capitatus,* ecology, life cycle, gall, plant-insect interactions, symbiotic fungus, parasitic Hymenoptera, mortality, damage, new pest, Samos, Greece.

INTRODUCTION

Coridothymus capitatus (L.) Reichenb.) or *Thymus capitatus* (L.) Hoffmanns & Link., the conehead thyme or Persian-hyssop, is a dwarf shrub of the family Lamiaceae, native to Northern Africa, Western Asia and southwestern Europe. It is an important component of the coastal and island flora in the Mediterranean regions of Greece (Tutin et al. 1964–1980, Pigott 1995). Economically conehead thyme is important for apiculture and in particular for the production of a high quality honey. Recently there has been a decline in the abundance and production of seed by conehead thyme in semi-mountainous regions on the island of Samos, which stimulated us to search for the cause of this decline. Recently a new species of gall midge associated with inflorescences of conehead thyme, *Asphondylia coridothymi,* was described by Skuhravá (2011) on the basis of material collected in Samos. This species severly damages conehead thyme plants so we decided to study its biology and ecology.

Four other species of the genus *Thymus* L. occur on the island of Samos: *T. samius* (Ronniger), *T. sipyleus* (Boiss), *T. ocheus* (Held et Sart) and *T. cilicius* (Boiss). Small populations of these species occur in the mountainous regions of the island. Interestingly, the inflorescences of these plants are not damaged by gall midges. In addition *Thymus vulgaris*, *T. glabrescens* and *T. serpyllum* that occur in southwestern and Central Europe and are host plants of a related species of gall midge, *Asphondylia serpylli* (Kieffer, 1898), which does not occur on the island of Samos (Skuhravá 1986). Galls of *Asphondylia* sp. on *Coridothymus* sp. were recorded for the first time at Galaxidi in the middle of Greece in 1995 by Skuhravá & Skuhravý (1997). Galls of this species were not found during a survey of the family Cecidomyiidae present on the island Samos carried out in 2005 (Skuhravá & Skuhravý 2006).

MATERIAL AND METHODS

The biology and ecology of *Asphondylia coridothymi* on *Coridothymus capitatus* were studied at two locations on the island of Samos in southern Greece from 2007 to 2008. Syrracho that is located in the mountains at round 450 m a. s. l. and Dendrias in the lowlands at round 50 m a. s. l. (Fig. 1).

In order to obtain adult gall flies inflorescences of *Coridothymus capitatus* with galls of *Asphondylia coridothymi* were collected at Syrracho from October to November 2007. Inflorescences were placed in a small glass cylinder attached to a McPhail trap kept illuminated in a laboratory at 20° C and a relative humidity of 67–70%. The gall midges and their parasitoids that emerged from the galls were collected and counted every day. After emergence they were put in 70% ethanol for future morphological and taxonomical studies. Some of these specimens, together with larvae, pupae and exuviae were sent to M. Skuhrava for identification and the rest are in the collection of the author. This study was carried out in the Laboratory of Plant Protection, Epirus Institute of Technology Department of Plant Production in Arta.

In February 2008 samples of conehead thyme plants with galls were collected in the mountains at Syrracho and lowlands at Dendrias and the emergence of gall midges from these samples were compared. The sample of 100 plants collected at Syrracho included 748 inflorescences and that collected at Dendrias 640 inflorescences. Every inflorescence with galls was put in a separate bag made of fine tulle with a mesh of 0.3×0.3 mm. Every bag was opened before being hung individually under glass Mc. Phail traps, which allowed light to enter from above. Every trap plus bag was hung up in low growing bushes at the locations where the inflorescences were collected. The fine tulle allowed the climatic conditions in and outside each bag to quickly equilibiate. Gall midge adults and parasitoids were collected every day. After adults ceased emerging each inflorescence in each bag was examined and the numbers of galls with emergence holes produced by gall midges and parasitoids, and of those that lacked emergence holes, were counted.

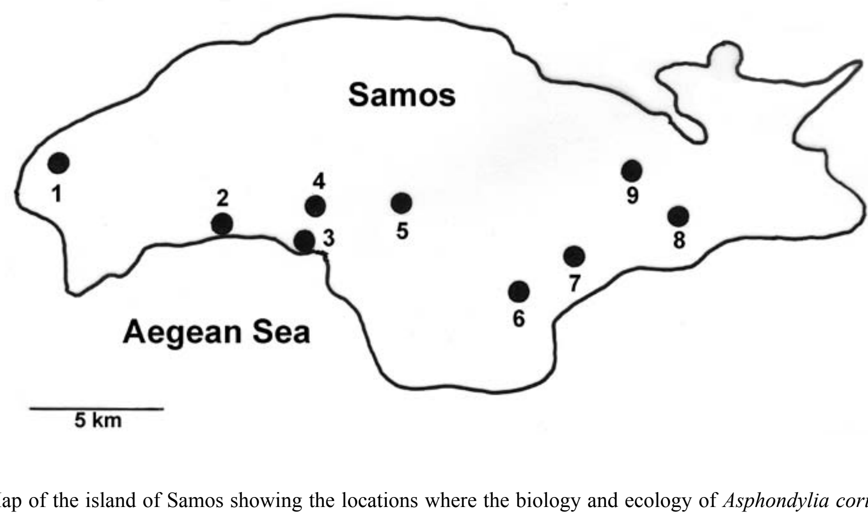


Fig. 1. Map of the island of Samos showing the locations where the biology and ecology of *Asphondylia coridothymi* Skuhravá, 2011 on *Coridothymus capitatus* were studied in 2007 and 2008. 1 – Kallithea, 2 – Ormos, 3 – Belanidia, 4 – Rikla, 5 – Syrracho, 6 – Dendrias, 7 – Chora, 8 – Agios Triada, 9 – Mytilini.

In 2007, from the beginning of May until the end of October, and similarly in 2008 we took samples of 30–40 inflorescences every ten days from ten representative conehead thyme plants. We measured the size of these galls and of the developing larvae and determined the duration of the different developmental stages of the midge.

We evaluated the damage caused to conehead thyme plants on the basis of the percentage of thyme inflorescences that produced galls. Material was collected in three regions in November 2007 and seven regions in November 2008. In each region we cut 1–2 branches of twigs with primary, secondary and subsequent inflorescences from a representative sample of 40 thyme plants and estimated the percentage of inflorescences that were galled. Afterwards we separated the inflorescences without galls from those with galls. We removed the remains of flower calyces separately from the galled and ungalled inflorescences and from each took at random a subsample of 1500 flower calyces and then determined the percentage of infertile flowers (0 seeds) and fertile flowers (with 1, 2, 3 or 4 seeds) in each subsample.

Samples of the fungal flora developing inside galls of *Asphondylia coridothymi* were collected from many galls on various plants in different regions of Samos and observed under a microscope. Samples from the different galls were aseptically incubated separately in Petri dishes on cultivation medium PDA (potato-dextrose agar). The fungus was identified using the key of Barnett & Hunter (1999). Fungus incubation and isolation was done in the Plant Pharmacology Laboratory of Epirus Institute of Technology, Department of Floriculture and Landscape Architecture in Arta. Fungal cultures were sent for verification of our identification to Dr. Laskaris Dimitrios at the Benaki Phytopathological Institute, Department of Mycology, Athens.

RESULTS

During 2007–2008, 972 adults of *Asphondylia coridothymi* were obtained from 1579 galls collected from *Coridothymus capitatus*. After all the gall midge adults and their parasitoids had emerged the galls were examined and the results are shown in Table 1.

Asphondylia coridothymi completed its development and emerged as an adult from about 60% of the galls, parasitic Hymenoptera from 8% and from 24% of the galls no insects emerged and when opened either contained nothing or a dead gall midge pupae.

Life cycle of *Asphondylia coridothymi*

The life cycle of *Asphondylia coridothymi* is synchronized with the biological cycle of its host plant, *Coridothymus capitatus*. The gall midge, *Asphondylia coridothymi,* is a univoltive species. The larval development is restricted to *Coridothymus capitatus* and there is no indication that *A. coridothymi* develops on other species of *Thymus*. Moreover, *Coridothymus capitatus* is the most abundant of the *Thymus* species on the Island of Samos.

Coridothymus capitatus is an herbaceous plant that on Samos starts to produce new shoots from the beginning of March around the coast to the end of March in the semi-mountainous areas. *Coridothymus capitatus* is well adapted to the climatic conditions and growing in stony calcareous soils around the coast and in semimountainous areas on Samos.

Primary shoots stop developing when the primary inflorescence starts to develop at the apex at which time lateral shoots develop and similarly grow and produce seconday inflorescences etc. The primary, secondary and tertiary shoots and their terminal spikelets take the form of a compound cyme. Flower buds successively develop first in the inflorescences on the primary shoots then

Table 1. Summary of the results obtained by monitoring the insects that emerged from galls caused by *Asphondylia coridothymi* on inflorescences of *Coridothymus capitatus* collected on Samos in 2007 and 2008. Legend: NIG – number of inflorescences with galls; TNG – total number of galls; TNA – total number of adults; NM – number of males; NF – number of females; NPG – number of parasited galls; DGP – dead galls and pupae

site	NIG	TNG	TNA	NM	NF	NPG	DGP
Syrracho 2007 (laboratory)	350	311	210	39	171	–	–
Dendrias 2008 (nature)	640	575	294	57	237	65	216
Syrracho 2008 (nature)	748	693	468	98	370	64	161

the secondary and subsequent shoots starting first in the coastal regions and later in the semi-mountainous regions, beginning in May and ending in June. This is the optimum plant stage for females of *Asphondylia coridothymi* to lay their eggs. They use their needle-shaped ovipositor to insert one egg together with the symbiotic fungus in each floral bud. The female usually lays only one egg in one flower bud in each inflorescence. Only exceptionally are two or three flower buds in the same inflorescence galled.

Adults of *A. coridothymi* emerged from galls from the middle March to the middle June in the lowland and coastal regions and from the middle of April up to the middle July in the semi-mountainous regions of the island of Samos (Fig. 2). Of the 972 adults reared from galls, 194 were males and 778 females and the sex ratio is 1:4, i.e. one male to four females (Table 1).

Adults of *Asphondylia coridothymi* are relatively large; their body is 2.7– 2.9 mm long. They have long antennae, large grey wings and dark-brownish coloured abdomen with reddish lateral parts (Fig. 5). They do not feed during adult life although some individuals may take water from dewdrops. Life span of adults under natural conditions is short and lasts about five days. Some individuals may live longer if kept in humid conditions in the laboratory. Females usually live longer than males. This species is nocturnal. Adults emerge after sunset and emergence continues for several hours.

The eggs in female ovaria mature before the adult emerges from a pupa. The primary function of adults is reproduction and dispersal, and of the larvae feeding (Ananthakrishnan 1984). On emergence males swarm over the ground searching for virgin females on the soil surface or on low growing plants, where mating occurs. Males swarm near the host plant where the females

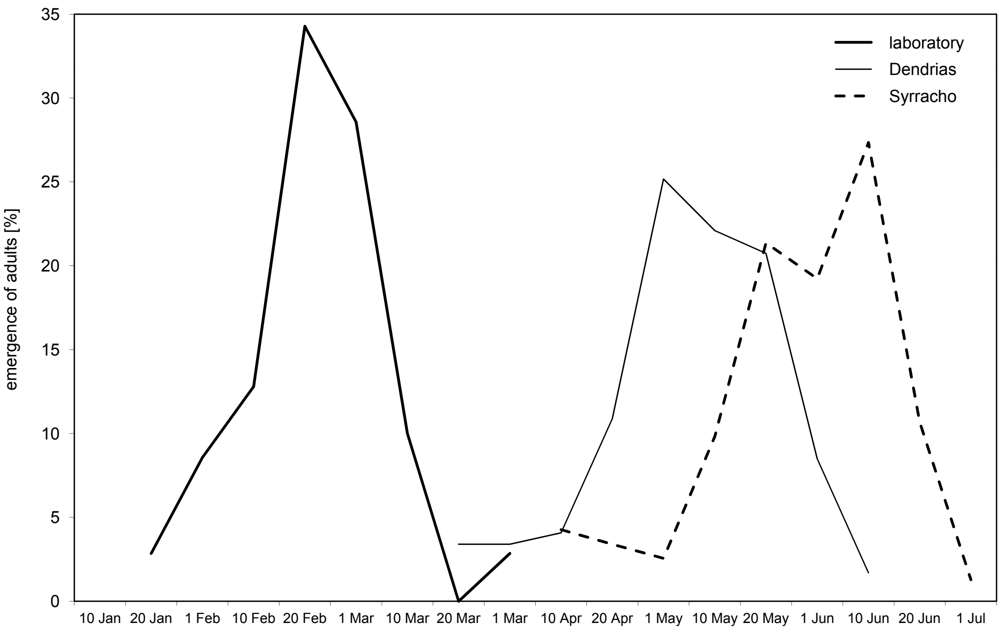


Fig. 2. Flight period of adults of *Asphondylia coridothymi* Skuhravá, 2011 that emerged from galls of *Coridothymus capitatus* in the laboratory and under natural conditions, and the flowering period of its host plant in 2008.

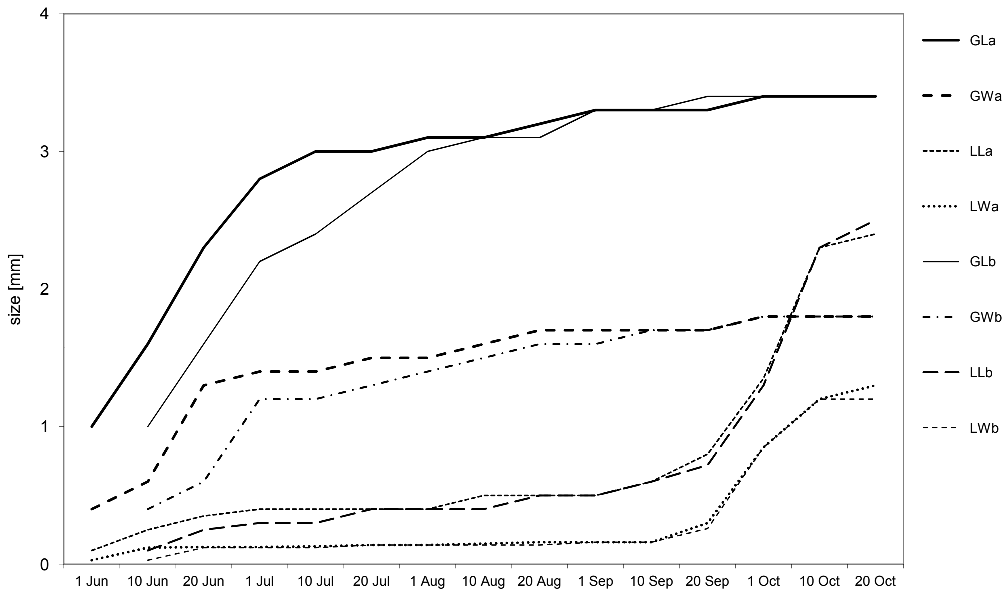


Fig. 3. Growth of galls and larvae of *Asphondylia coridothymi* Skuhravá, 2011 in galls on primary and secondary inflorescences of *Coridothymus capitatus* collected in the Syrracho region during the summer of 2008. Explanations: GLa – all length of primary inflorescences; GWa – gall width of primary inflorescences; LLa – larva length of primary inflorescences; LWa – larva width of primary inflorescences; GLb – gall length of secondary inflorescences; GWb – gall width of secondary inflorescences; LLb – larva length of secondary inflorescences; LWb – larva width of secondary inflorescences.

are also located. Virgin females attract males by extending their ovipositors and releasing sex pheromones. Males recognize the pheromones, locate the females and mate with them. One female usually mates with one male whereas one male can mate with more than one female.

The degree of damage to conehead thyme and the time of year when emergence occurs can vary from year to year. Annual variations in climatic factors affect the host plant phenology and also the time of emergence of adults. However, *Asphondylia coridothymi* responds to thermal effects differently from the host plants. A warm or cold spring may differentially promote the earlier or later emergence of gall midges and may delay or hasten the development of buds on host plants. The gall midge emergence is not always synchronized with the host plant phenology. Synchronization of adult emergence with host plant phenology, however is the critical event for short-lived insects such as gall midges, because a time lag in the synchronization determines the quality and quantity of available floral buds.

In addition, the development of a gall is a complex phenomenon that involves subtle alterations initiated at critical points of time during plant differentiation.

Larva

The larval development of *Asphondylia coridothymi* includes three instars, which can be distinguished by their size, colour and some morphological characters (Figs. 6–7). Each larval instar develops inside a gall of characteristic shape. In 2007 and 2008 the first galls were observed developing in inflorescences of conehead thyme at the beginning of June before the onset of flowering in a semi-mountainous region at Syrracho. Galls were green and of slender elongated pear-shaped form. Only one larva developed inside each gall.

The first instar larva has a vermicular form and is a bright yellow colour, 0.8 mm long and 0.4 mm broad. Larvae of this instar grow very slowly and the duration of this instar is the longest of the three instars. In the Syrracho region this instar lasted from the beginning of June until the end of September, i.e. a total 110 days. The galls at the end of the first larval instar are still green coloured and pear shaped.

The second instar larva is egg-shaped to claviform, bright yellow, 0.8–1.2 mm long and 0.6–0.7 mm broad. Larvae of this instar occur in galls in last decade of September. This instar lasts for only five to six days.

The third instar larva is fusiform, broad anteriorly and tapered posteriorly, bright yellow, 2.1 mm long and 0.9 mm broad. Larva of this instar has a spatula sternalis, a sclerotized organ of characteristic shape, on the ventral part of the prothorax. This larval instar develops in the first decade of October. Larvae in galls develop very quickly and they nearly double their size in a short time. The internal walls of galls are covered with a mycelium of the symbiotic fungus *Aureobasidium* sp., which is white at this time. Occurrence of symbiotic fungus at this stage in the development of the larva is very important for its further development (Fig. 3).

Our observations confirm the fact that larval development is divided into two periods (Mamaev & Krivosheina 1993). During the first the young larva strongly stimules plant tissues to grow. As a result of its activity the gall appears and quickly develops to nearly normal proportions. During the second period the larvae feed intensively whereas the gall grows slowly and its walls start to lignify.

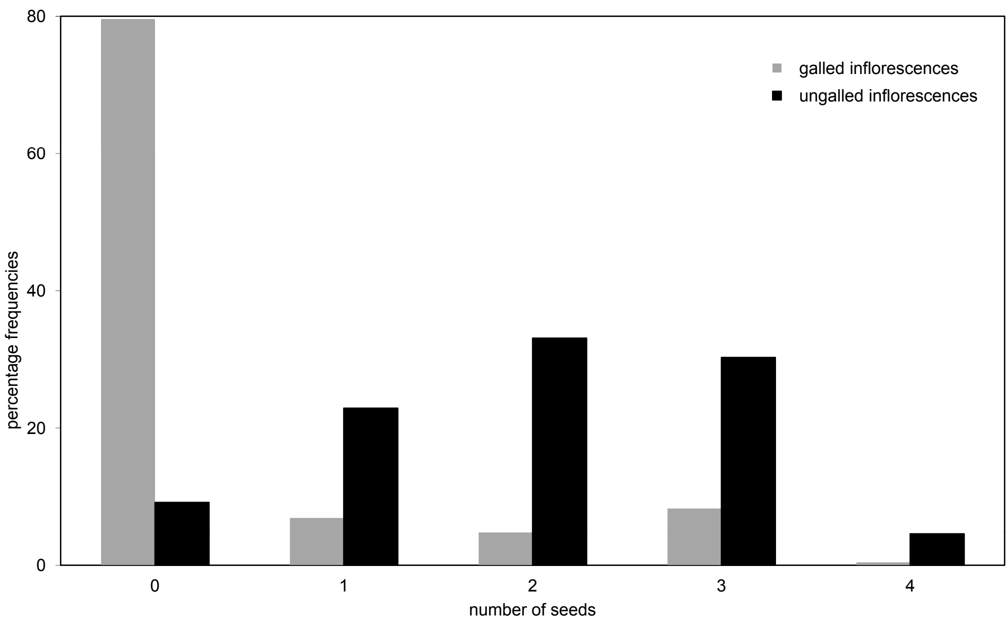
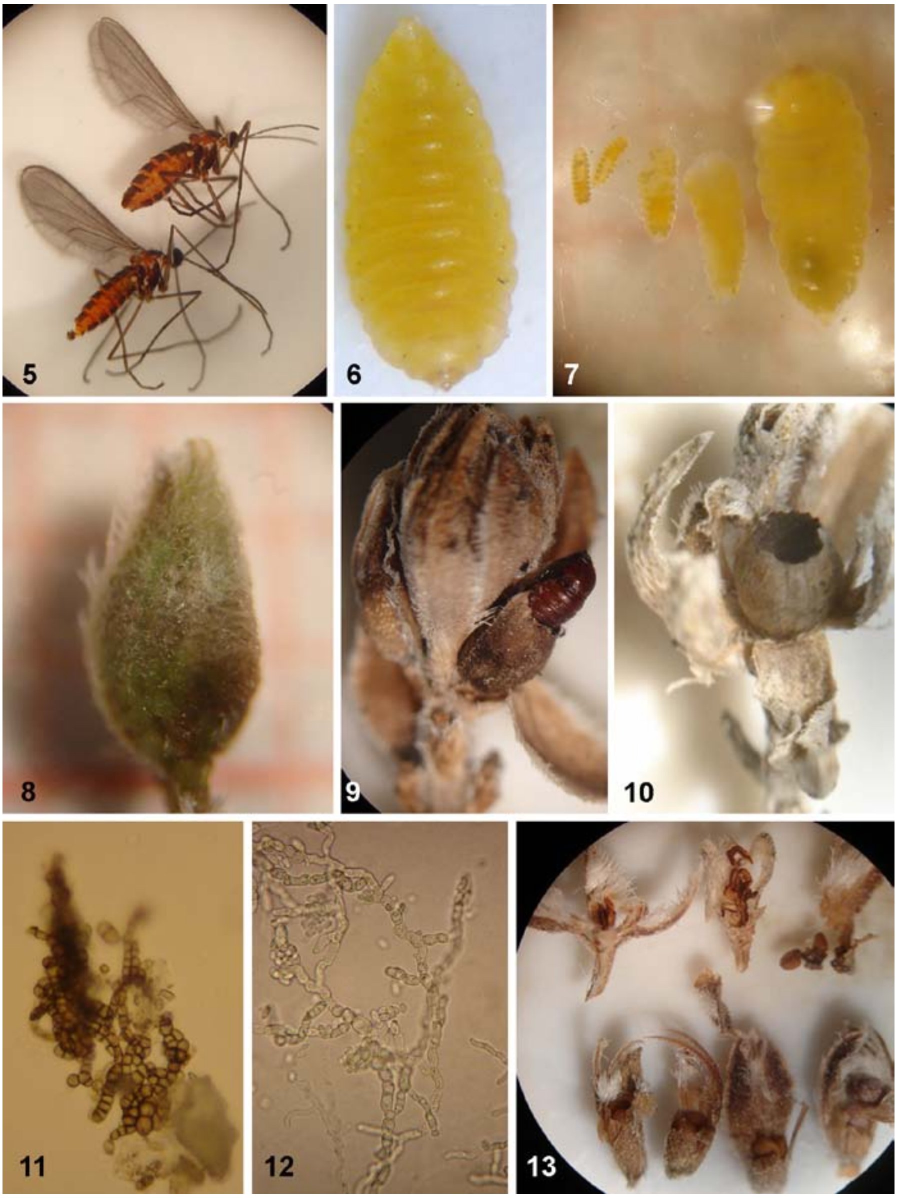


Fig. 4. Percentage frequencies of the number of seeds produced by thyme flower flowers.



Figs 5–13. *Asphondylia coridothymi* Skuhravá, 2011. 5 – female above, male below, 6 – the third instar larva, so-called mature larva, 7 – three larval instars, from the left: two small larvae of the first instar, two larvae of the second instar and one larva of the third instar, 8 – a small gall caused by the larva of the second or third instar in the inflorescence of *Coridothymus capitatus*, 9 – a pupa in the process of cutting its way out of a gall, 10 – gall with an opening cut by a pupa through which the adult emerged, 11 – melanospores of symbiotic fungus of the genus *Aureobasidium* obtained from inner walls of galls, 12 –blastospores of this fungus, 13 – damage caused to flower buds of *Coridothymus capitatus*: above (1) – remnants of flower calyces without seed from galled inflorescences, below (2) – fruits with seed that developed in ungalled inflorescences.

Prepupa

The time spent in the prepupal stage is very short. It occurs usually in the second decade of October. It is an intermediary stage between the third instar larva and pupa. The larva is immobile and its body becomes reddish brown. The galls change colour from green to dark green and their walls harden and lignify. The white mycelium of symbiotic fungus covering the internal walls becomes blackish and greyish.

Pupa

Pupa is the third stadium in gall midge development (Fig. 9). It occurs in galls in the middle of October. *A. coridothymi* overwinters as a pupa in galls until April the following year. The pupa is reddish-brown in colour. If the temperatures in early autumn and late spring are high the pupa may move spasmodically in response to external stimuli. The galls containing pupae usually remain on plants the whole winter.

The main characteristic of a normal inflorescence of conehead thyme are the protective leaves of the flowers in the spikelets (paranthia leaves) and the calyces, which fall to the ground in autumn revealing the sharp ends of each of the spikelets. The exceptions are the spikelets that bear one or more gall on which the protective leaves remain. A hormonal effect of the galls causes an inflorescence retain the protective leaves during winter and spring, which provides additional protection for the galls in unfavorable conditions.

In spring the pupae of *Asphondylia coridothymi* start to cut their way out of the galls. Pupae perform characteristic movements with their abdomen and use their antennal horns, upper frontal horns and lower frontal horns to cut an opening in the wall of the gall (Fig. 10). This process starts slowly in the last decade of March, accelerates in April and finishes in the middle of May. Some pupae remain in the galls, some drop to the ground where they remain until the adult emerges. Some pupae remain exserted from the opening of the galls up to the emergence of the adult. The process of cutting the opening for emergence lasts about 4–7 days depending on temperature. The mature pupae shortly before the emergence of adult become black.

The adult gall midge emerges from the pupal exuvia after it causes the sutura on the dorsal surface of the thorax to crack lengthwise. Adults after emergence remain for a short period stationary while their bodies harden. After several hours the adults are ready to fly.

Gall

First galls, about 1 mm long, develop in the primary inflorescences of conehead thyme at the beginning of June. They are induced by the activity of the first instar larvae. The galls grow quickly and at the beginning of July they are 3 mm long (Fig. 8). Then they grow slowly and at the end of August they cease growing. Mature galls are pear-shaped, 3.4 mm long, 1.8 mm broad and green coloured. Inside the gall is a large chamber the walls of which are covered with the mycelium of the symbiotic fungus. In galls there are never any remains of the organs of the flower from which it developed.

Symbiotic fungus

Gall midges of the group Asphondyliini and Lasiopterini induce so called ambrosia galls and their larvae feed mainly on fungi (Rohfritsch 1992). The females of the genus *Asphondylia* have a characteristically enlarged seventh abdominal sternite and a sharp ovipositor, with which they pierce plant tissue (Yukawa & Rohfritsch 2005). *Asphondylia* females also transfer fungus hyphae on their ovipositor. The females attack the meristomatic tissues of flower buds, introduce a fungus into the host plant tissues during oviposition and induce the formation of ambrosia galls. In these galls the fungal mycelium develops in the larval chamber and is loosely connected to the plant tissue by intracellular haustoriae.

Table 2. Parasitic Hymenoptera reared from galls of *Asphondylia coridothymi* on *Coridothymus capitatus* collected on the island of Samos in 2007 and 2008, and percentage of galls parasitized

family	species	percentage of parasitism
Pteromalidae	*Systasis encyrtoides*	30.76
Eulophidae	*Aprostocetus* sp.	23.07
	Tetrastichus sp.	19.23
	unidentified species	3.87
Ormyridae	*Ormyrus* sp.	7.69
Eupelmidae	*Eupelmus* sp.	7.69
Eurytomidae	*Eurytoma* sp.	7.69

The fungus that is associated with *Asphondylia coridothymi* belongs to the genus *Aureobasidium* (*Pullularia*) of the group Basidiomycetes (Figs 11–12). The fungal mycelium covers the internal walls of the gall chamber but does not attack the larva or pupa. The mycelium covering the walls is at first white and later changes from white to grey-black and shiny. Galls containing pupae are covered with black fungal mycelium.

The mycangium is made up of the setae on the seventh abdominal sternite of *Asphondylia* females but how they collect the right fungus and introduce it into plant tissue remains unexplained (Yukawa & Rohfritsch 2005).

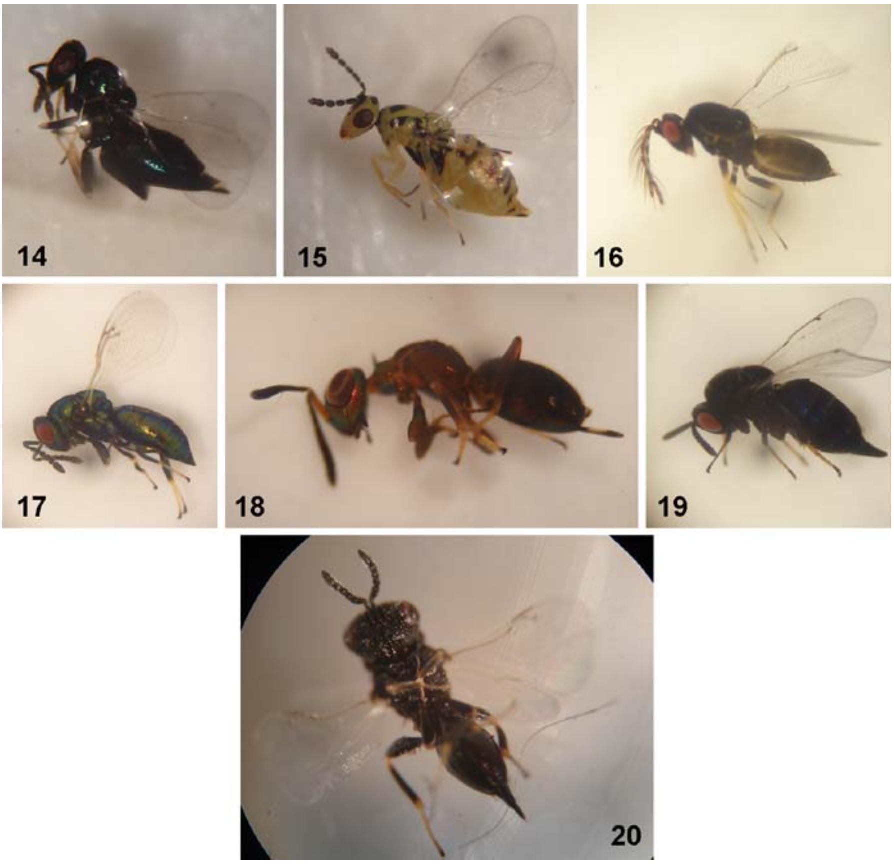
Parasitic Hymenoptera

In the course of this study a total of 762 adult specimens of *Asphondylia coridothymi* were reared from 1268 galls of *Coridothymus capitatus* in 2008. Together with adults of gall midges 129 specimens of parasitic Hymenoptera emerged from galls of *A. coridothymi* (Table 2). They belong to seven species of five families of the superfamily Chalcidoidea, viz. *Systasis encyrtoides* Walker, 1834 (Pteromalidae), *Aprostocetus* sp., *Tetrastichus* sp., an unidentified species of the subfamily Tetrastichinae (Eulophidae), *Ormyrus* sp. (Ormyridae), *Eupelmus* sp. (Eupelmidae) and *Eurytoma* sp. (Eurytomidae) (Figs 14–20). Larvae of these species are ectoparasitoids and attack solitarily the larvae of *A. coridothymi* developing in galls (Gibson et al. 1997, Noyes 2010).

Systasis encyrtoides was the most abundant parasitoid reared from galls of *Asphondylia coridothymi* followed by *Aprostocetus* sp. and *Tetrastichus* sp. Of the 129 parasitoids collected 73% belonged to these three species and the remaining four species were less abundant and together made up only 26%. *Systasis encyrtoides* is the most important parasitoid species and may have a significant role in reducing the population of gall midges. This parasitoid species has also been reared from galls of other gall midges (Parnell 1963, 1964, Simova-Tosic et al. 2007, Skuhravá & Thuroczy 2007).

The mean percentage parasitism of the gall midge based on all the samples collected in various regions of Samos during the summer of 2008 was 10.3%, with the percentage parasitism at altitudes of around 100 m a. s. l in the lowland regions 11.3% and only 9.2% in the semi-mountainous regions.

The galls of *Asphondylia coridothymi* containing larvae of parasitoids, unlike those containing gall midge pupae, were not perforated by large openings. Adult parasitoids only cut, using their mouthparts, a small circular hole in a side wall or near the top of a gall. Larvae of parasitoids start to attack larvae of *Asphondylia coridothymi* in the early stages of development. They suck liquids from bodies of the gall midge larvae. The life cycle of parasitoids is well adapted to the life cycle of *Asphondylia coridothymi*. Adult parasitoids emerge from galls at the same time as adults of *Asphondylia coridothymi*.



Figs 14–20. Parasitic Hymenoptera reared from galls of *Asphondylia coridothymi* Skuhravá, 2011: 14 – *Aprostocetus* sp., 15– *Tetrastichus* sp., 16 – unidentified species of Tetrastichinae, 17 – *Systasis encyrtoides*, 18 – *Eupelmus* sp., 19 – *Ormyrus* sp., 20 – *Eurytoma* sp.

Mortality

About one quarter of the galls of *Asphondylia coridothymi* collected during this study in 2007 and 2008 remained closed and neither gall midge adults nor parasitoid adults emerged from them. There were no emergence holes in these galls and it is likely that the gall midge larvae died in the first instar but the galls continued to develop.

They were probably killed by unfavourable climatic factors, i.e. drought and high temperatures in summer and especially in autumn. The mortality of larvae in galls was 23.3% in the semi--mountainous and 37.6% in lowland regions.

Damage

Damage is measured in terms of any reduction in the quality or quantity of yield that result from injury. Larvae of *Asphondylia coridothymi* develop in flower buds of *Coridothymus capitatus*, change them into galls and prevent the development of seed not only in the attacked flower buds but also in adjacent flowers in the same inflorescence. Thus the injury caused by *A. coridothymi*

Table 3. The results of the statistical analysis of the percentage of inflorescences of *Coridothymus capitatus* that bear galls of *Asphondylia coridothymi* and reduction in seed production in nine areas at various altitudes on the Island of Samos in 2007 and 2008. For positions of localities see map in Figure 1. Legend: A – altitude (m a. s. l.); NT – number of twigs; NI – number of inflorescences; IG – infloescences with galls (%); SE – standard error; SD – standard deviation; SV – sample variance; MaP – max price; MiP – min price; M – mode; RSP – reduction in seed production (%)

region	A	NT	NI	IG	SE	SD	SV	MaP	MiP	M	RSP
2007											
Belanidia	40	45	1025	3.33	0.76	5.2	27.7	25	0	0	2.59
Rikia	150	45	885	6.61	1.59	10.3	106.4	33.3	0	0	5.15
Syrracho	500	45	1390	18.97	1.63	15.4	239.2	66.6	0	0	14.79
2008											
Kallithea	450	43	974	22.66	2.82	18.5	342.8	66.6	0	0	17.67
Ormos	0	38	953	1.13	0.5	3	9.5	12.5	0	0	0.88
Belanidia	40	40	1168	2.34	0.63	4	16	16.6	0	0	1.82
Rikia	150	37	685	1.44	0.57	3.5	12.4	16.6	0	0	1.12
Syrracho	500	48	933	32.42	2.28	15.8	250.3	85.7	0	30	25.28
Dendrias	20	37	872	0.6	0.31	1.9	3.6	7.7	0	0	0.46
Chora	60	33	721	3.37	1.52	8.7	76.8	33.3	0	0	2.62
Agia Triada	150	32	1156	1.3	0.67	3.8	14.4	16.6	0	0	1.01
Mytilini	220	33	849	4.21	1.13	6.5	42.3	21.4	0	0	3.28

results in the loss of seed, which can be used to evaluate the damage caused by this new pest of conehead thyme.

In 2007 and 2008 we evaluated the damage caused by *Asphondylia coridothymi* to inflorescences of *Coridothymus capitatus*. The damage in lowland regions (Dendrias, Belanidia and Marathocampos) was low, as only between 3.3% and 6% of the inflorescences were galled, but in the semi-mountainous regions (Kallithea, Syrracho) it was higher with from 19 to 30% of the inflorescences bearing galls (Table 3).

About 70% of flowers in inflorescences bearing galls are infertile whereas all those in inflorescences that are not galled produce seed (Fig. 4, 13). The large number of infertile flowers may be a result of trophic competition or hormonal manipulation of plant development by the larvae of this gall midge. The seed production of inflorescence bearing galls was 78% of that of ungalled inflorescences. The reduction in seed production of conehead thymes in lowland regions was relatively low and ranged from 0.46 to 3.28%, whereas in semi-mountainous regions it was much higher and ranged from 17.67 to 25.28% (Table 4).

Discussion and conclusions

The negative effect of developing galls of *Asphondylia coridothymi* on the inflorescences of *Coridothymus capitatus* may be a consequence of trophic competition or chemical manipulation by first instar larvae. It is likely that the symbiotic fungus is important for the rapid growth of the third instar larva. Problems with the growth of the symbiotic fungus at this stage in the growth of the larva may result in it dying during the pupal stage.

In lowland regions the damage to thyme inflorescences caused by *Asphondylia coridothymi* is lower (from 3.3% to 6% attacked inflorences) than in semi-mountainous regions (19 to 30%). This corresponds to a decrease in seed production by this plant 0.46 to 3.28 and 25.28%, respectively, in the two regions.

In the semi-mountainous regions on Samos the conditions in summer are better for the survival and growth of galls caused by *Asphondylia coridothymi*. Moreover it seems that in semi-moun-

Table 4. Frequency distribution of the number of seeds produced by flowers in galled and ungalled inflorescences of thyme. Legend: NSF – number of seeds in each flower (a); NFEG – number of flowers examined from galled inflorescences (b); NFEU – number of flowers examined from ungalled inflorescences (c)

NSF	NFEG	NFEU
1	1194	137
2	104	343
3	72	497
4	124	454
5	343	69
total number of flowers	Σ(aXb) 1500	Σ(aXc) 1500
total number of seeds	644	2975
percentage of infertile flowers	79.6	9.13
increase in percentage of infertile flowers	70.47	
percentage reduction in number of seeds	[Σ(aXc)–Σ(aXb)/Σ(aXc)] X100=78%	

tainous areas there is also a better synchronization between the emergence of adult *Asphondylia coridothymi* and the presence of suitable flower buds in the inflorescences of *Coridothymus capitatus*.

The damage to conehead thyme caused by *Asphondylia coridothymi* on the Island of Samos seems to be limited in most localities by the following factors:

(1) The high mortality of larvae and pupae in galls caused by unfavourable climatic conditions during summer and particularly drought in autumn. The percentage mortality varies from 23.3% in semi-mountainous to 37.6% in lowland regions.

(2) Parasitoids attacking larvae of *Asphondylia coridothymi* in galls reduce the abundance of this gall midge. Percentage parasitism is 9.2% in semi-mountainous and 11.3% in lowland regions.

(3) Poor synchronization between the development of host plants and emergence of gall midges. Gall midge females fail to find suitable flower buds for oviposition and subsequent development of larvae. About 26% of the adults failed to find suitable plants in semi-mountainous regions and 42% in lowland regions.

Intensive and extensive attacks of *Asphondylia coridothymi* on *Coridothymus capitatus* plants can have negative consequences for the propagation of thyme by seed and limits the area occupied by thyme. The reduction in the abundance of thyme on the Island of Samos in the last 40 years is obvious but cannot be attributed only to *Asphondylia coridothymi* but also to human activity, in particular burning and land clearances. Moreover it is also necessary to take into consideration the difficulty of cultivating conehead thyme, i.e. low percentage of germination (20–25%), short distance over which seeds are disseminated and unfavourable drought conditions that prevail on Mediterranean islands, particularly recently as a result of reduction in rainfall.

The study and recording of parasitoids may result in the future in their use as biocontrol agents of *Asphondylia coridothymi* in different regions of the country and a reduction in the abundance of this gall midge that damages conehead thyme.

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