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New association between *Cecidochares connexa* (M.) (Diptera: Tephritidae) and local parasitoids: Revisiting classical biological control

Asosiasi baru antara *Cecidochares connexa* (M) (Diptera: Tephritidae) dan parasitoid lokal: Sebuah tinjauan terhadap pengendalian hayati klasik

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ABSTRACT

Lalat argentina *Cecidochares connexa* (L.) (Diptera: Tephritidae) diintroduksikan pada tahun 1993 di Bantimurung, Sulawesi Selatan untuk mengendalikan gulma siam *Chromolaena odorata* yang merupakan gulma invasif dari Florida, Amerika Serikat. Namun, beberapa penelitian sebelumnya menunjukkan bahwa *C. connexa* dapat terserang oleh predator dan parasitoid lokal sehingga dapat memengaruhi efektifitas *C. connexa* di lapang. Penelitian ini bertujuan mempelajari asosiasi baru antara hama eksotik *C. connexa* dan parasitoid lokal. Penelitian dilakukan dari bulan April–Desember 2021 pada dua habitat, yaitu lahan terbuka dan perkebunan kakao, di empat desa di Kabupaten Polewali Mandar. Gulma siam diambil secara *purposive sampling*, yaitu *C. odorata* yang berpuru. Tiga puluh *C. odorata* berpuru dikoleksi pada tiga lokasi berbeda di setiap habitat dan setiap desa. Puru dari setiap sampel dikelompokkan menjadi dua macam, yaitu puru yang tidak berlubang dan puru yang berlubang. Puru yang tidak berlubang dipelihara sampai serangga keluar dan spesimen serangga yang didapatkan diidentifikasi. Hasil penelitian menunjukkan bahwa rata-rata jumlah puru (baik puru sehat maupun puru terparasit) lebih tinggi pada lahan terbuka dibandingkan pada perkebunan kakao. Fenomena parasitasi mengindikasikan adanya interaksi dan asosiasi baru antara *C. connexa* dengan parasitoid lokal. Tingkat parasitasi *C. connexa* di lahan terbuka (23,24%) tidak berbeda jika dibandingkan dengan parasitisasi di perkebunan kakao (28,84%). Ada empat famili parasitoid yang ditemukan yaitu Eulophidae, Braconidae, Eupelmidae, dan Ormyridae. Pada lahan terbuka ditemukan enam jenis morfospesies parasitoid dan pada perkebunan kakao sebanyak tiga jenis. *Quadrastichus* sp.1 merupakan parasitoid dominan dengan persentase parasitisasi 14% pada lahan terbuka dan 10% pada perkebunan kakao.

Key words: gall insects, invasive species, new association, parasitoid

ABSTRAK

Lalat argentina *Cecidochares connexa* (L.) (Diptera: Tephritidae) diintroduksikan pada tahun 1993 di Bantimurung, Sulawesi Selatan untuk mengendalikan gulma Siam *Chromolaena*

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Kata kunci: interaksi baru, parasitoid, serangga puru, spesies invasif

INTRODUCTION

Biological control is defined as the activity of parasitoids, predators, or diseases in maintaining the other organism's population density at a lower average than would occur in their absence (De Bach 1964). Classical biological control involves the introduction of a biocontrol agent to a new habitat to control pests that have spread beyond their original habitat. According to Caltagirone (1981), classical biological control is the regulation of exotic pest populations (insects, mites, mammals, weeds, pathogens) by exotic natural enemies due to the lack of local natural enemies that can suppress the exotic pests, thus creating a favorable situation for invasive species to spread. It works on the premise that old associations between the host (pests) and its biocontrol agent have already been established (co-evolved), and thus should be an effective control agent (Hokkanen & Pimentel 1998)

The most famous classical biological control was the introduction of the vedalia beetle predator *Rodolia cardinalis* (Coleoptera: Coccinellidae) to control the cottony cushion scale *Icerya purchasi* (Hemiptera: Margarodidae) in California (Clausen, 1978; Caltagirone & Doutt 1989). The introduction of *R. cardinalis* is regarded as the beginning of classical biological control. The use of biological agents for weed management started in 1795 when the cochineal insect *Dactylopius ceylonicus* from Brazil was introduced to India to control the invasive weed *Opuntia monacantha* (Willd.) Haw. (Cactaceae) (Winston et al. 2014). By 2012, a recorded number of 1,555 cases of biological control of weeds were reported in 90 countries in which 468 biocontrol agents were introduced to manage 175 species of weeds in 48 plant groups (Schwarzlander et al. 2018). However, only 982 (63.2%) introductions demonstrated stability, with a total of 332 (70.9%) biological agents used (Schwarzlander et al. 2018).

In Indonesia, C*ecidochares connexa* (M.) (Diptera: Tephritidae) is a gall fly that has been used as a biocontrol agent to suppress the population of the invasive species *Chromolaena odorata* (L) R.M. King and H. ob (Asteraceae). *C. odorata* is a fast-growing invasive species that entered Indonesia in 1934 at the Lubuk Pakan tobacco plantation in North Sumatra and spread rapidly to other major Indonesian islands (Tjitrosoedirdjo 2005; Setyawati et al. 2015; Padmanaba et al. 2017). *C. connexa* has been established in several countries, including the Philippines, India, Guam, and Papua New Guinea (Aterrado & Bachiller 2002; Bhumannavar & Ramani 2006; Reddy et al. 2010; Day et al. 2013). The success of its establishment was one of the reasons for its introduction to Indonesia. McFadyen et al. (2003) reported that *C. connexa* causes galls on the terminal and axillary vegetative meristems of *C. odorata,* and can decrease seed production by 50% and reduce *C. odorata* population by up to 37,2% within two years (Tjitrosemito 1999).

C. connexa was first introduced in Indonesia in 1993 from Colombia, USA (Tjitrosemito 1999). In 1995-2001, releases were carried out in several areas, including eastern Indonesia, one of which was South Sulawesi. The first release of *C. connexa* in Sulawesi was conducted in the Bantimurung sub-district (60 km northeast of Makassar), South Sulawesi, in March 1999, with a total of 240 galls. A second release was carried out in February 2000 at two different locations, namely the Bantimurung sub-district (60 km northeast of Makassar) with a total of 300 galls and Camba sub-district (25 km northeast of Bantimurung) with 232 galls (Wilson & Widayanto 2004). In April 2002, the establishment of *C. connexa* was evaluated at release sites. *C. odorata* infested by *C. connexa* was found approximately 10 km from the release point (Wilson & Widayanto 2004).

Since 2002, no studies have evaluated the effectiveness *C. connexa* in Bantimurung, South Sulawesi, nor have any studies been conducted to see the changes in the food web interaction due to the introduction. As mentioned by Pearson & Callaway (2005), biocontrol agents introduced in new areas could indirectly change the interactions in the ecosystem and attack non-targets through food web subsidies or host change. This interaction can change the overall food web and interactions of many species in an ecosystem. One example of research related to food web subsidies is the introduction of *Urophora affinis* and *U. quadrifasciata* to control *Centaurea maculosa* and *C. diffusa* in western North America (Winston et al. 2014). Their research showed that the two gall flies that were introduced became an additional food source for populations of generalist predators, deer mouse *Peromyscus maniculatus* (Pearson et al. 2000; Ortega et al. 2004; Winston et al. 2014). Another study on ecosystem changes was conducted by Jaya (2006) and Lukvitasari et al. (2021), who showed that *C. connexa* could be attacked by local parasitoids and predators. Several types of parasitoids have been found to attack *C. connexa* including those from the Families Ormyridae, Eupelmidae, Bethylidae, Braconidae, Ichneumonidae, Encyrtidae, Eucoilidae, Eulophidae, Eurytomidae, Pteromalidae, and Torymidae (McFadyen et al. 2003; Safi 2006; Buchori et al. 2020; Lukvitasari et al. 2021). These findings show that there is a new association between *C. connexa* and local parasitoids, which invokes the old hypothesis of Hokkanen & Pimentel (1989) regarding new associations that can be formed between biological control agents and local natural enemies. Thus, biological agents introduced in new areas can potentially become new hosts for natural enemies.

The aim of this research is to study community interactions and possible new associations between *C. connexa* and local parasitoids in Polewali Mandar, West Sulawesi. Does the establishment of the bioagent *C. connexa* in West Sulawesi cause an array of new interactions with local parasitoid communities? Does the complex of host parasite interactions similar as was found in the Western part of Indonesia? These questions are important since Sulawesi is located on the Eastern side of the Wallace line, thus may have different biodiversity compared to the western part of Indonesia. These are among the questions raised to understand the impact of *C connexa* released in the field at Polewali Mandar, West Sulawesi.

MATERIAL AND METHOD

Time and location of research

The research was conducted from April to December 2021. Sampling and rearing were conducted at Polewali Mandar Regency, West Sulawesi Province (Figure 1), while the specimens were identified at the Biological Control Laboratory, Department of Plant Protection, Faculty of Agriculture, IPB University.

Sampling was conducted using a purposive sampling method, namely infestation of *C. odorata* by gall-made flies. Sampling locations were in two different habitats, that is, the open fields at three fillages and the cocoa plantations in three villages in Polewali Mandar Regency, West Sulawesi (Table 1 and Figure 1).

Sampling, rearing, and observation of *Cecidochares connexa* galls

Galls were collected from 30 *C. odorata* sample plants that were purposively selected from each of the three sampling locations. Each gall was grouped into two categories: those with and without

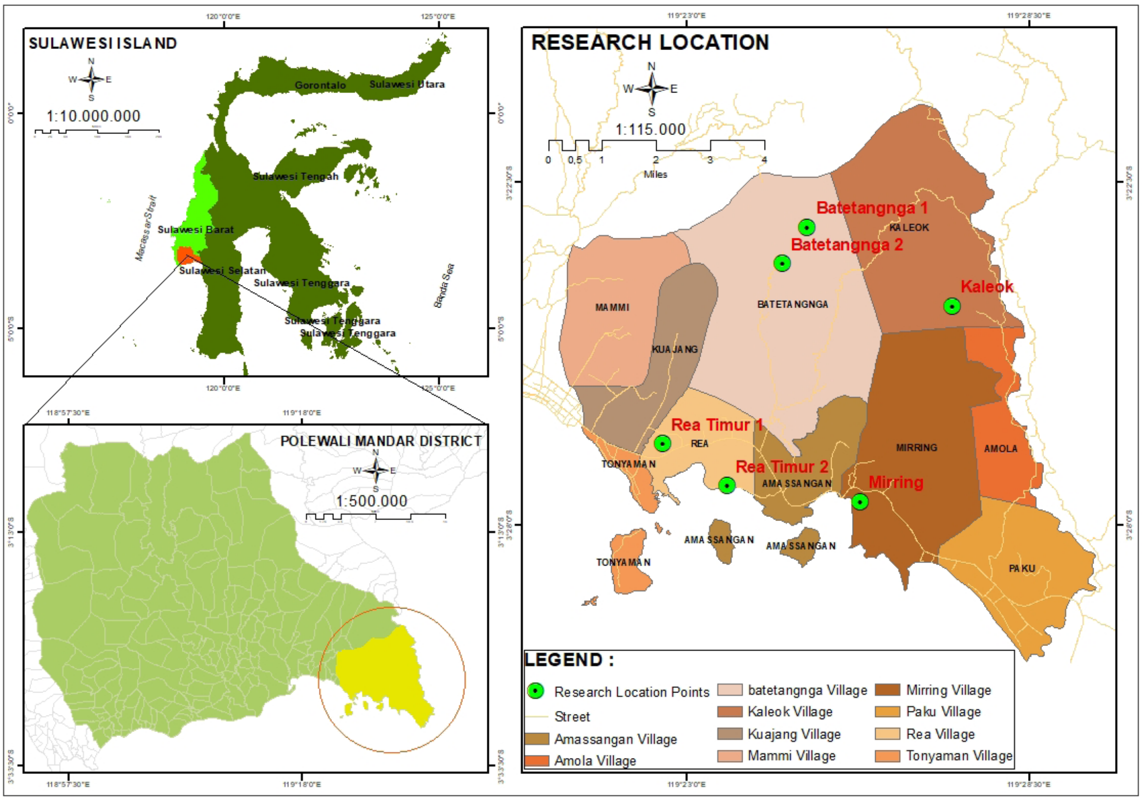

Figure 1. Research location.

Table 1. Locations of galls sampling at open field and cocoa plantation habitats in Polewali Mandar Regency, West Sulawesi Province

Sampling locations	Habitat types	Coordinate points (latitude and longitude)	Altitude (m asl)
Mirring	Open field	3.46019°S 119.42999°E	±89
Rea Timur 1	Open field	3.44482°S 119.37697°E	±64
Rea Timur 2	Open field	3.45604°S 119.39420°E	±67
Batetangnga 1	Cocoa plantation	3.38692°S 119.41557°E	±640
Batetangnga 2	Cocoa plantation	3.39664°S 119.40894°E	±375
Kaleok	Cocoa plantation	3.40793°S 119.45488°E	±705

holes (Figure 2). All collected sample galls were labelled and brought to the laboratory for further culture and identification of insect specimens. Galls with bigger holes, about 1.45 mm to 1.6 mm in diameter (Figure 3A) were counted as *C. connexa* emergence, whereas galls with small holes, about 0.4 mm to 0.6 mm in diameter (Figure 3B) were counted as parasitoid emergence. Galls without exit holes (Figure 2) were observed every two days for emerging insects (either *C. connexa* or its parasitoids). After 30 days, each gall without holes reared in the laboratory was dissected to observe the internal conditions. The number of *C. connexa* and parasitization rates were calculated for each replicate per habitat. Each insect that came out was placed into a 1.5 ml microtube containing 70% alcohol for identification.

Identification

Identification was performed using a stereo microscope (Olympus SZ61). Identification keys were found in the Insect of Australia Vol 1-2 (Naumann *et al.* 1991), Manual of Nearctic Diptera Vol 1-2 (McAlpine et al. 1987), and Hymenoptera of The World: An Identification Guides to Families (Goulet & Huber 1993) at the Laboratory of Biological Control, Department of Plant Protection, Faculty of Agriculture, Bogor Agricultural University.

Data analysis

The parasitization rate was calculated from emerged parasitoids in the laboratory, and small exit holes were found in the field. For every parasitoid exit hole found in the field, we assumed

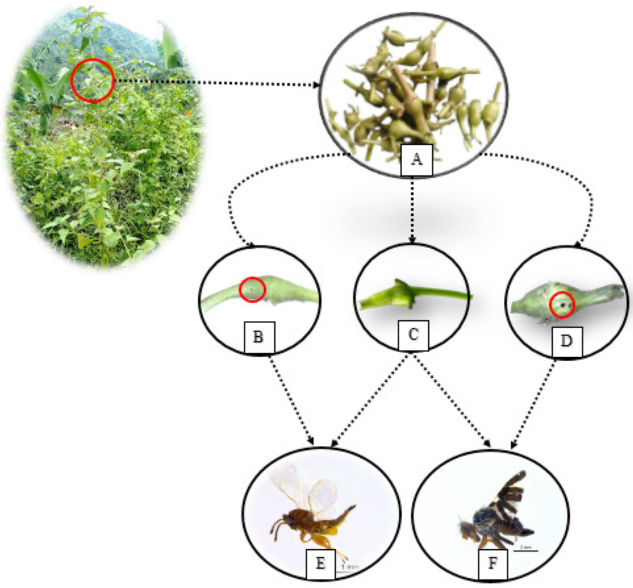

Figure 2. A: Galls formed by *Cecidochares connexa*; B: gall with parasitoid hole; C: gall without hole; D: gall with *C. connexa* hole; E: parasitoid; F: *C. connexa*.

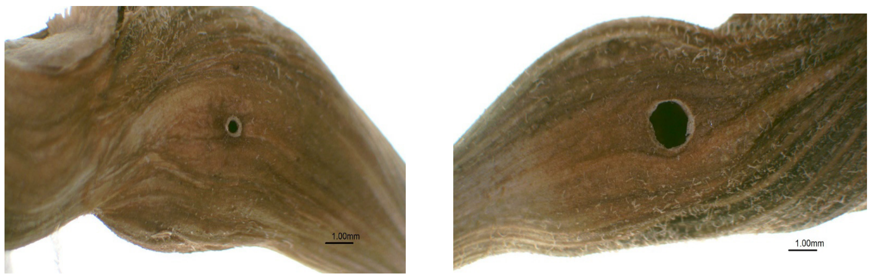

Figure 3. Gall with a small insect exit hole of *Cecidochares connexa* (A) and gall with a big insect hole of parasitoids (B).

that the parasitoid was solitary and that one *C. connexa* was parasitized.

The parasitization rate on gall is calculated using the formula:

$$\text{Parasitization} = \frac{\text{Number of gall parasitized}}{\text{Total number of gall}} \times 100\%$$

The percentage of parasitization on C. connexa was calculated using the formula:

$$\text{Parasitization of } C.\ connexa = \frac{\text{Number of } C. connexa \text{ A}}{\text{Number of } C.\ connexa \text{ A} + C.\ connexa \text{ B}} \times 100\%$$

The percentage of parasitization of each parasitoid species on C. connexa from the results of closed-gall rearing was calculated using the following formula:

$$\text{Parasitization of parasitoid} = \frac{\text{Number of } C.\ connexa \text{ C}}{\text{Number of } C.\ connexa \text{ D}} \times 100\%$$

wich in A: parasitized; B: not parasitized; C: parasitized by 1 type of parasitoid; D: parasitized by all types of parasitoids

The data obtained were tabulated using Microsoft Excel 2013 software. Data on the number of galls, parasitization of galls, and parasitization of *C. connexa* were processed by t-test analysis using Minitab 16.0. Data on parasitization of each parasitoid type on *C. connexa* were processed by analysis of variance (ANOVA) and further tested by Tukey's significant difference test at the 5% level using Minitab 16.0. Box plots were generated using the package ggplot2 (Wickham 2005) with R-Studio software version 4.2.2 (R Core Team 2022).

RESULTS

The presence of the gall fly *C. connexa*

The results showed that *C. connexa* can be found in diverse habitats (open fields and cacao plantations) where the studies were conducted. This shows that the gall flies were dispersed in

Polewali Mandar, West Sulawesi. These findings suggest that the gall fly *C. connexa* can disperse at a distance of more than 250 km from the release point (assuming the release point is Bantimurung) within 20 years or less. This is the first report of the current presence of *C. connexa* in Polewali Mandar.

Establishment of *C. connexa* in two different habitats

The gall fly *C. connexa* attacks *C. odorata* in different habitats. On average, approximately 1-10 galls per plant were found in *C odorata* in the open field and cacao plantations; however, the number can vary from 1 to 80 galls per plant (Figure 4). Two plants were found with more than 90 galls per plant in the open field. Compared to the galls found in *C. odorata* from cocoa plantations, galls from open fields exhibited more variance. More than one *C. connexa* individuals can found in each gall (Table 2) in both habitats. There are even galls that can house up to nine individuals, although this is a very rare case.

Based on the results of the T-test at the 5% level, the type of habitat had a significant effect on the average number of galls (*P-value* = 0.0001) (Figure 5A) and *C. connexa* (*P-value* = 0.0001) (Figure 5B) per *C. odorata* with the average gall *C. connexa* in open fields being higher than that in cocoa plantations.

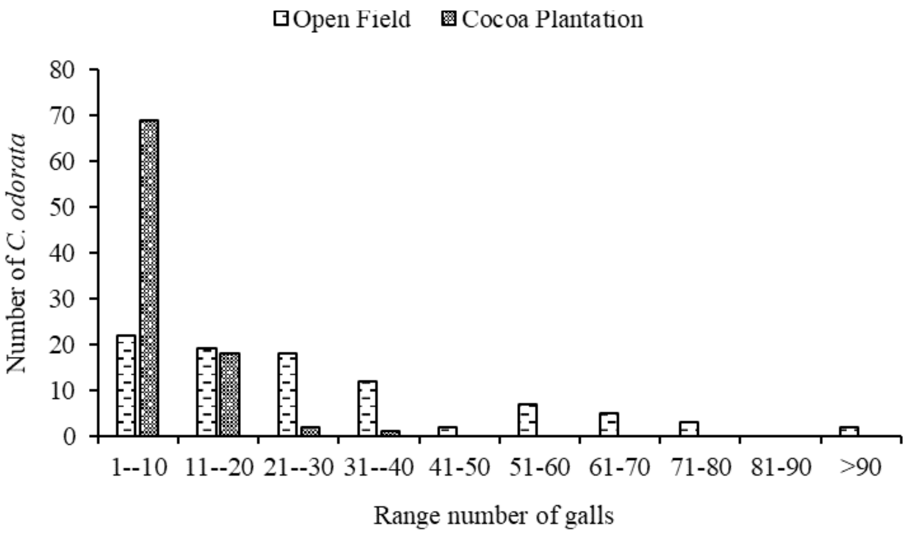

Figure 4. The number of galls per *Chromolaena odorata* sample plant in two different habitats (N = 90); N: number of *C. odorata* per habitat.

Table 2. The number of *Cecidochares connexa* per gall from *Chromolaena odorata* plants collected at opened field and cocoa plantation habitats

Number of *C. connexa* per gall	n (locations)		Total numbers of gall from 3 locations		Galls per location (x̅ ± SD)		%	
	OF	CP	OF	CP	OF	CP	OF	CP
1	3	3	2,285	614	761,67 ± 347,00	204,67 ± 36,35	89,57	90,96
2	3	3	191	56	63,67 ± 11,06	18,67 ± 1,53	7,49	8,30
3	3	3	60	5	20,00 ± 3,61	1,67 ± 1,15	2,35	0,74
4	3	3	10	0	3,33 ± 2,52	0	0,39	0
5	3	3	4	0	1,33 ± 0,58	0	0,16	0
6	3	3	0	0	0	0	0	0
7	3	3	0	0	0	0	0	0
8	3	3	0	0	0	0	0	0
9	3	3	1	0	0,33 ± 0,58	0	0,04	0
Total	-	-	2,551	675	-	-	100	100

OF: opened field; CP: cocoa plantation; x̅: average; SD: standard deviation.

New association between *C. connexa* and local parasitoids

The presence of *C. connexa* in *C. odorata* triggered the formation of a "new association" with local parasitoid insects (Figure 6). *C. connexa* is parasitized by several species of parasitoids. The parasitoids found were from the order Hymenoptera, consisting of four families: Eulophidae, Braconidae, Ormyridae, and Eupelidae. Six morphospecies of parasitoids were found in the open field: *Aprostocetus* sp.1 (Eulophidae), *Aprostocetus* sp.2 (Eulophidae), *Doryctobracon* sp. (Braconidae), *Ormyrus* sp.1 (Ormyridae), *Ormyrus* sp.2, and *Eupelmus* sp. (Eupelidae) (Figure 6A). In the cocoa plantations, only three morphospecies of parasitoids were found: *Aprostocetus* sp.1 (Eulophidae), *Doryctobracon* sp. (Braconidae), and *Ormyrus* sp.1 (Ormyridae) (Figure 6B).

Habitat differences had a significant effect, based on a t-test at the 5% level, on the average number of parasitized galls (P-value = 0.0027) and *C. connexa* (P-value = 0.002) (Figure 7). The average number of galls and *C. connexa* parasitized in open fields was also higher than that in the cocoa plantations. However, habitat did not significantly affect the parasitism rates of gall (P-value = 0.2404) and *C. connexa* (P-value = 0.865) (Figure 8).

As many as 319 out of a total of 1.799 *C. connexa* individuals found in galls (reared in the laboratory) obtained from open fields were parasitized (17,89%). Meanwhile, the parasitization rate in cocoa plantations was higher, with 100 out of 461 *C. connexa* individuals (21,69%). Compared to cocoa plantations, open fields included more types of parasitoid species with the number of *C. connexa* parasitized by *Aprostocetus* sp.1 being more than the other parasitoids in both habitats, whereas the lowest parasitoid species in the open field was *Ormyrus* sp.2 and in cocoa plantations was *Ormyrus* sp.1. The parasitization rate of *C. connexa* in the cocoa plantations was higher than that in the open field (Table 3).

Analyais of Variance (ANOVA) revealed that the type of parasitoid had a significant effect (*P-value* = 0.002) on the average number of parasitized *C. connexa*, as well as the parasitization of *C. connexa* (*P-value* = 0.003) in the open field habitat. However, in cocoa plantations, it did not significantly affect the average number of parasitized *C. connexa* (*P-value*= 0.131) or parasitization rate (*P-value*= 0.521) (Table 3).

DISCUSSION

Our result showed that *C. connexa* are already established in West Sulawesi and can be found in open fields and cocoa plantations. This study is the first to report that *C. connexa* are able to adapt and become established in Polewali Mandar, West Sulawesi. This finding is in line with Tjitrosemito (2002) which stated that *C. connexa* has been established on most of the larger Indonesian islands and that *C. connexa* can spread widely over short periods of time. According to Harjaka & Mangoendihardjo (2010), *C. connexa* can

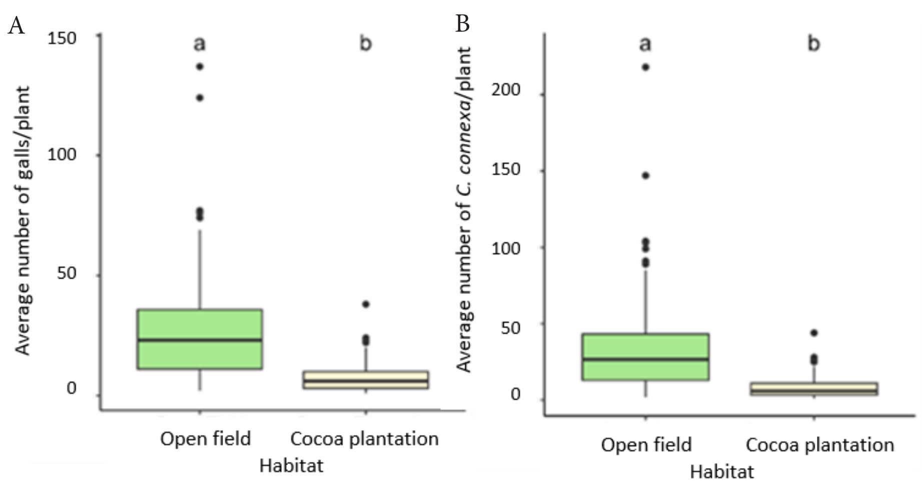

Figure 5. Average number of gall (A) and *Cecidochares connexa* (B) per sample plant of *Chromolaena odorata* (N = 90) in different habitat. N: number of *C. odorata* per habitat.

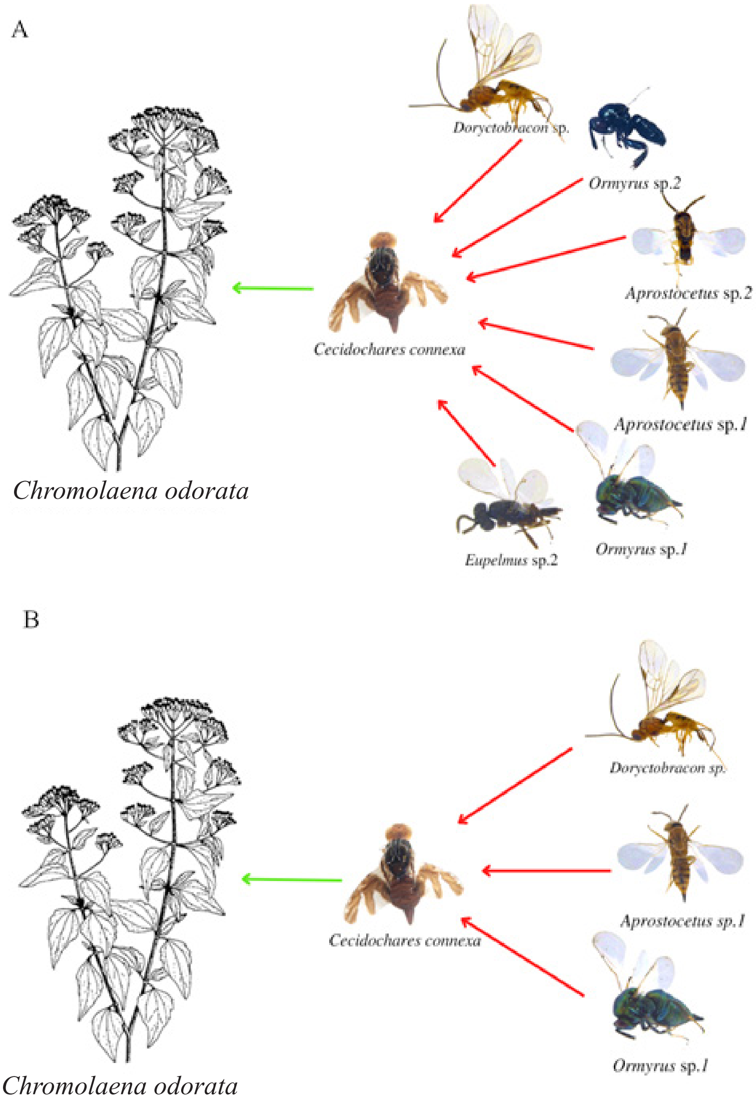

Figure 6. Interactions between *Chromolaena odorata*, *Cecidochares connexa*, and parasitoids in two different habitats. A: open field; B: cocoa plantation. The green line indicates herbivore insects associated with *C. odorata*. Red lines indicate parasitoids associated with *C. connexa*.

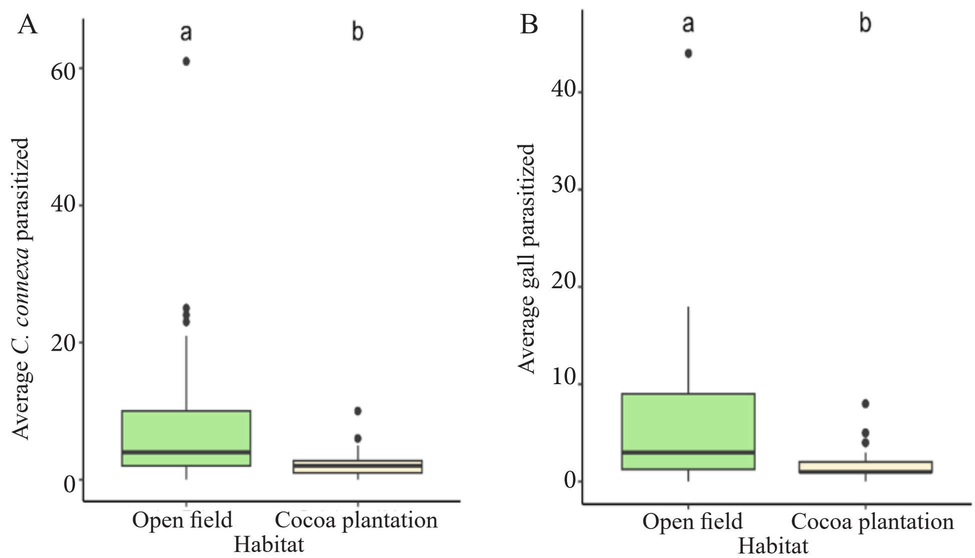

Figure 7. The average number of parasitized *Cecidochares connexa* (A) and gall (B) per habitat (N = 90). N: number of *Chromolaena odorata* per habitat.

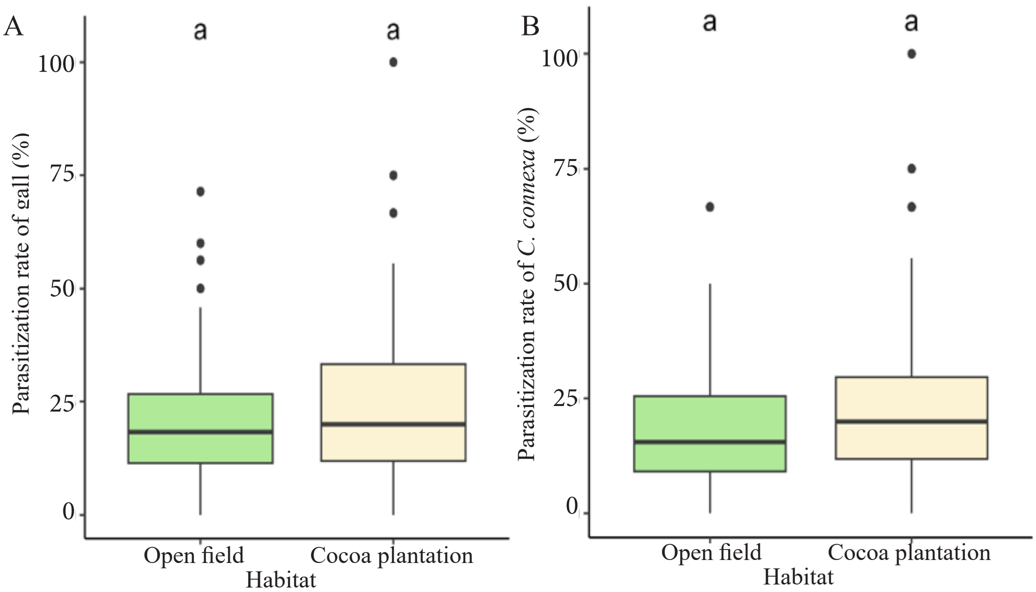

Figure 8. The percentage parasitization of galls (A) and *Cecidochares connexa* (B) per habitat (N = 90). N: number of *Chromolaena odorata* per habitat.

Table 3. The average number of *Cecidochare connexa* parasitized and percentage parasitization by each type of parasitoid per habitat (N1 = 1799; N2 = 461)

Parasitoid Famili Species	Open field			Cocoa plantation		
	X̄ ± SD	N	% parasitization	X̄ ± SD	N	% parasitization
Braconidae *Doryctobracon* sp.	11,33 ± 13,32 b	34	1,91 b	12,67 ± 13,58 a	38	8,24 a
Eulophidae *Aprostocetus* sp.1	80,67 ± 46,06 a	242	13,45 a	16,33 ± 14,57 a	49	10,63 a
Aprostocetus sp.2	3,33 ± 5,77 b	10	0,56 b	0,00 ± 0,00	0	0,00
Eupelmidae *Eupelmus* spp.	3,33 ± 5,77 b	10	0,56 b	0,00 ± 0,00	0	0,00
Ormyridae *Ormyrus* sp.1	6,33 ± 5,51 b	19	1,06 b	4,33 ± 5,13 a	13	2,82 a
Ormyrus sp.2	1,33 ± 2,31 b	4	0,22 b	0,00 ± 0,00	0	0,00
Total		319	17,89		100	21,69
Mean		53,67	2,96		16,66	3,64

Numbers followed by the same letter are not significantly different based on Tukey's test with a level of 5%; x̄: the average number of *C. connexa* parasitized; SD: standard deviation; N1: the number of *C. connexa* in open field; N2: the number of *C. connexa* in cocoa plantation.

spread over a radius of more than 200 km from the release site in Gunungkidul, Yogyakarta, to several areas in East Java within 10 years. Day et al. (2013) also reported that *C. connexa* had spread more than 100 km from most of the release points in Papua New Guinea within seven years. It is likely that *C. connexa* can also be found in areas more than 250 km from the release point. However, the introduction of *C. connexa* did not seem to reduce the population of *C. odorata*, as indicated by its abundant population at the study site. Kenis et al. (2019) reported that the success of biological control is classically characterized by a decrease of the target population. However, our observation found *C. odorata* to be abundant in the field, hence raising the question of *C. connexa* effectiveness in as biological control agent. Field observations showed that the attacked branches do not instantly die but continue to develop and it even create new branches. Thus attack of *C. odorata* seems to increase the branch production.

Our observation showed that *C. connexa* could lay more than one egg on each shoot of C. *odorata*. In addition, we also found more than one *C. connexa* larvae in one gall that can emerge as imago. McFadyen et al. (2003) reported that *C. connexa* could lay up to 16 eggs per shoot on the shoots of *C. odorata*. However, the number of larvae that can survive to become imago is between 2-4 individuals.

Compared to open fields, cocoa plantations have fewer galls and *C. connexa*. This is because fewer *C. odorata* plants may be found in cocoa plantations, which leads to fewer galls being discovered. Pruning and spraying of herbicides routinely on *C. odorata* around cocoa plantations may affect to the population of *C. odorata*, resulting in a decrease in the number of galls formed. The presence of shade is also believed to be a factor that slows the growth of *C. odorata* in addition to pruning and spraying. This has an impact on *C. connexa* which can parasitize parasitoids.

Overall we found six species of parasitoids associated with *C. connexa* with Aprostocetus sp 1 being the most abundant . The parasitoid complex is different than the community of parasiotids found in West Java (Buchori et al. (2020). *Quadrastichus* sp was the most abundant parasitoid found in Bogor and Lampung. The difference may seem be due to the different diversity that are present in the island of Java, Sumatra and Sulawesi. This study however, strengthens the findings of Buchori et al. (2020), Lukvitasari et al. (2021), Jaya 2006 and Pahlevi 2006; that new associations have been formed from exotic bioagents introduced to new habitat*s*. The fact that *C. connexa* from different islands are attacked by different parasitoid complex has shown that adaptation of local parasitoids to invasive/exotic pests can happen. McFayden (2003) also reported the attack of *C. connexa* by parasitoid insects five years after release and the first report of a new association between *C. connexa* and a parasitoid. Boughton et al. (2012) also reported that *Neomusotima conspurcatalis* Warren (Lepidoptera: Crambidae) was introduced to Florida in 2008 to control *Lygodium microphyllum* (Schizaeales: Lygodiaceae) and are then attacked by local parasitoids in just a few years. These findinga are interesting because it reveals the local adapatation of local parasitoids with exotic herbivore species, thus strengthening Hokkanen Pimentel (1989) argument to use local parasitoids as bioontrol agent, since new associates can happen quite rapdily in the field. Our findings raise other questions e.g. does the presence of *C. connexa* change the interaction dynamics of the parasitoid with its original hosts? Are there shifts in the preference of the parasitoid from its original hosts, and if there is a shift, what are the implications for the original hosts? Will there be a natural enemy-free space that results in the explosion of new pests? Further investigation is required to corroborate some of the assertions made above because there is a shortage of knowledge on the subject.

One important question that can be raised from this study is whether *C. connexa* only functions as an additional host for parasitoids or actually functions as a subsidy that can increase the population of parasitoid insects and can indirectly increase the role of these parasitoids to attack other hosts. The parasitoids found attacking *C. connexa* in this study are most likely generalist parasitoids that attack a variety of gall-forming insects in several habitats in the tropics. Thus, when *C. connexa* attacks *C. odorata* and form galls, the local parasitoids can shift and atttack *C. connexa* as a new host. Parasitoids of the Genus *Aprostocetus* were the most common and had the highest parasitization rate. This is different from what was previously reported by Buchori et al. (2020) and Lukvitasari et al. (2021) that the dominant parasitoid found was *Quadrastichus* sp. *Aprostocetus* parasitoids are likely common parasitoids that commonly attack other insects such as *Orseolia javanica* (Diptera: Cecidomyiidae) on rice in Bogor, West Java (Maqsalina 2021), *Ophelimus eucalypti* (Hymenoptera: Eulophidae) on eucalyptus plants Medan, North Sumatra (Anisa et al. 2023).

Parasitoids are generally considered beneficial for pest control as they parasitize insect pests. In this case, they have a detrimental impact, as they attack the biological control agent of *C. connexa* which is used to control *C. odorata* populations. Overall, the parasitoid discovered in the cocoa plantations was also present in the open fields. The parasitoid observed to attack *C. connexa* in this study is most likely a generalist parasitoid that targets different forming-gall insects in several habitats in tropics. The introduced *C. connexa* which was established in release areas to control *C. odorata* weed, is a new host for local parasitoids. According to Pearson and Callway (2005), biological agents introduced into new areas to control invasive pests have the potential to become hosts for local natural enemies. If they are unable to control the population of invasive weeds, it results in an abundance of weed populations.

One reason for the ineffectiveness of *C. connexa* in suppressing *C. odorata* populations is the presence of parasitoids that parasitize *C. connexa*. Although the level of parasitization is still relatively low, the presence of parasitoids on *C. odorata* can reduce attacks from *C. connexa*. Abdala-Robert et al. (2019) stated that one of the indirect effects of the presence of natural enemies is that they can reduce herbivore populations and indirectly affect the plants that are attacked.

Classical biological control using *C. connexa* to suppress populations of the invasive weed *C. odorata* has been unsuccessful. Although *C. connexa* is widespread and established, its presence is insufficient to control *C. odorata* because it is attacked by a variety of parasitoids in a variety of habitats, including industrial plantations, forests, open fields in Bogor (Jaya 2006; Safi'I 2006), palm oil plantations and open fields in Bogor and Lampung (Lukvitasari et al. 2021), cocoa plantations, and open fields in Polewali Mandar. Consequently, its effectiveness is reduced. In addition, *C. odorata* endured *C. connexa* attacks by producing additional branches in response to the attack.

CONCLUSION

The gall fly *C. connexa* was found in open fields and cocoa plantations in Polewali Mandar, West Sulawesi, more than 250 km from the release point. This is the first report of *C. connexa* in Polewali Mandar, West Sulawesi. The abundance of *C. connexa* in *C. odorata* was influenced by habitat type. There is a new association between *C. connexa* and local parasitoids with the discovery of six types of parasitoid morpho-species from four different families. *Aprostocetus* sp.1 was the dominant parasitoid, with a parasitization percentage of 13.57% in open land and 10.63% in cocoa plantations.

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