

# Investigation the Potential of *Myopopone castanea* Smith (Hymenoptera: Formicidae) as Bio-Agent of *Oryctes rhinoceros* (Coleoptera: Scarabaeidae) in Oil Palm Plantations

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## ABSTRACT

The *Myopopone castanea* Smith (Hymenoptera: Formicidae) is a predator of *Oryctes rhinoceros* Linn larvae, the main pest on oil palm plantations. These predatory ants have potential as biological agents because they have similar niches to their prey. This research aimed to explore the presence and potential of *M. castanea* as bio-agents in oil palm plantations and obtain knowledge about the abiotic environment of *Myopopone castanea* ant nests. The research was conducted in oil palm plantations owned by PTPN 4 Adolina, PTPN 2 Pagar Merbau, and smallholder oil palm plantations in Binjai, North Sumatra Province. Sampling was conducted using the purposive sampling method and as many as 10 sample points in each plantation. The results showed that the average temperature and humidity in the ant nests were 29.4 °C and 70.5%, respectively. The number of individual ants in a colony varied greatly in number and developmental stages. The predation potential of *Myopopone castanea* ants in the field reached 50.3% for five days of exposure in smallholder plantations and 46,9% in Adolina Plantation. This research could be the opportunity to use *Myopopone castanea* ants as a biological control for larvae *Oryctes rhinoceros* in oil palm plantations.

**Keywords:** Biological Control; Predation; Ants; Smallholder Plantation

## INTRODUCTION

Oil palm (*Elaeis guineensis*) is the main commodity of plantation crops in Indonesia. Indonesia is currently the world's largest CPO producer, producing 45.1 million tonnes with a plantation area of 14.62 million Ha ([Badan Pusat Statistik, 2022](#)). One of the obstacles to increasing oil palm production is pest attacks, including *Oryctes rhinoceros* Linn (Coleoptera: Scarabaeidae). This beetle feeds on the canopy of plants and bores through the base of the stem to the point of growth. The leaves that have opened show a V-shape or characteristic of serrate cuts. This symptom is typical of horn beetle attacks. This pest is controlled chemically and mechanically by handpicking ([Pradipta et al., 2020](#)). The use of carbofuran and cypermethrin insecticides is very difficult to do on oil palms that are over one year old and also has an impact on the environment and does not support the demands of sustainable agriculture on RSPO (Roundtable Sustainable Palm Oil) oil palm plantations ([Abd](#)



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[Karim et al., 2019](#))

Biological control is the best alternative method and a more environmentally friendly process to control pestilential organisms (including pathogens, pests, and weeds) for diverse purposes to benefit humans. Biological control that is widely carried out for *O. rhinoceros* pests is to use entomopathogens, such as *Metarhizium anisopliae* ([Indriyanti et al., 2017](#); [Suryanto, 2020](#); [Fauzana et al., 2020](#)), *Bacillus thuringiensis* ([Pujiastuti et al., 2022](#)), *Beauveria bassiana* ([Nasution et al., 2018](#); [Indriyanti et al., 2021](#)), and *Baculovirus oryctes* ([Chalapathi Rao et al., 2018](#); [Rahayuwati et al., 2020](#)). The use of predators is rarely applied, and reports on it are still limited. However, the use of insect predators in biological control is important due to their role in sustainable pest management, ecosystem functioning, reduction of pesticide use, and the specialized behaviors that make them effective natural enemies of pest species ([Marchiori, 2023](#)).

*Myopopone castanea* is one of the biological agents (predators) that consume *O. rhinoceros* larvae ([Widihastuty et al., 2018](#); [Susanti et al., 2017](#)). [Wilson \(1971\)](#) states that these ants are abundant in forests, temperate areas, and the tropics. In oil palm plantations, these ants live in the trunks or stumps of oil palms that have fallen and piled up together, where larvae of *O. rhinoceros* also live and develop. The same living niche between the predatory ants, *M. castanea*, and the larvae of *O. rhinoceros* opens up opportunities for implementing biological control in the pest control of *O. rhinoceros* using predators. To optimally utilize the role of these predatory ants, it is necessary to rear them in the laboratory, and they can be used in biological control programs through augmentation techniques.

A study was conducted to investigate the potential of using *M. castanea* as a biological agent against *O. rhinoceros* by examining the existence of *M. castanea* colonies and various elements of their nest's physical habitat in oil palm farms. Moreover, a predation test was also studied to measure its predation potential in the field to optimize its use as a biological agent.

## **MATERIALS AND METHOD**

The research was conducted in March-September 2019. The exploration research of *M. castanea* ant was carried out in smallholder oil palm plantations in Binjai, Pagar Merbau plantation PTPN 2, Deli Serdang Regency and PTPN 4 plantation, Serdang Bedagai Regency, North Sumatra Province. Field predation research was conducted in the Adolina plantations of PTPN 4. The smallholder oil palm plantations are old, poorly maintained, 34 m above sea level, and overgrown with weeds. The age of the plants is around 20 years, and there are many fallen oil palm trunks. The age of the plants in the PTPN Pagar Merbau plantation ranges from 15 to 20 years, and it has better plantation sanitation than the smallholder plantation. The altitude is 22 m above sea level. PTPN 4's Adolina plantation is a 1-year-old TBM plantation with an altitude of 19 m above sea level.

### **Exploration of *M. castanea* Colonies**

The observation of *M. castanea* colonies was carried out on smallholder and state-owned garden plantations (PTPN). Sampling was carried out using the purposive sampling method. The samples taken were 10 sample points each in smallholder and PTPN-owned plantations. Colonies of *M. cas-*

*tanea* live on the trunks and stumps of fallen and decayed oil palm trunks. The rotten palm trunks or stem stumps piled up were cut into pieces and chopped to find an ant colony (Figure 1). If *M. castanea* colonies were found in the trunk, then a measuring device for temperature and humidity was plugged into the formicary to measure temperature and humidity in the nest of ants. Temperature and abiotic factor measurements are required for mass rearing in the laboratory. After the measurement of such abiotic factors was recorded and completed, the ants were collected by putting them in a plastic container along with pieces of the trunk of the palm where the formicary was taken to the plant pest laboratory for mass rearing at the Faculty of Agriculture, Universitas Muhammadiyah Sumatera Utara Medan.



**Figure 1.** (a) The palm trunk stump as a nest (b) colonies of *M. castanea* in the nest

### **Predation Test in the Field**

A predation test was conducted at PTPN 4's Adolina oil palm plantation in Perbaungan District and the smallholder plantation in Binjai, North Sumatra Province. The age of oil palm plants in the TBM plantation ranged from 1 year (TBM 1), while the field predation test at Binjai area was carried out on smallholder oil palm plantations (TM plantations) between 15-20 years old plants. Predation tests in the field were carried out by placing a log of palm trunks (85cm x 30cm x 5cm), which were made into a symmetric dredge in the middle as a place for laying *O. rhinoceros* larvae. The logs of the palm trunks were placed systematically and evenly at ten sighting points at both the Adolina plantation and smallholder plantation in Binjai, with a range between points ranging from 70m. Fifty worker ants and 20 ant larvae were put into the log, as well as 6 of the 2<sup>nd</sup> instar of *O. rhinoceros* larvae. Then, the log was wrapped using gauze to prevent the ants from escaping from the log. Observations were made five days after application by counting the number of *O. rhinoceros* larvae that died due to predation by *M. castanea*. Predated larvae will have only their cuticle visible, the cuticle color is blackish brown, and the abdomen is wrinkled because the *M. castanea* ant colony has eaten the hemolymph fluid. Differences in predation rate in TM and TBM plantations were statistically tested by t-test.

## Statistical analysis

Ant colony data obtained in the exploratory study were tabulated and statistically described. The temperature and humidity data obtained in the *M. castanea* ant exploration research will be processed using descriptive statistical analysis methods. In contrast, data obtained in field predation tests are processed using descriptive statistical methods and tested using the t-test. The differences in the abiotic environment in the two plantations were statistically tested by t-test.

## RESULTS AND DISCUSSION

### Ant Colonies

Ant colonies found in oil palm plantations were very diverse regarding the number of individuals in the colonies (Table 1). Most colonies did not have a complete developmental stage. [Robinson et al. \(2017\)](#) and [Andersen \(2019\)](#) stated that the abundance of the ant population was strongly influenced by climate change and vegetation coverage, including environmental litter. The more diverse the vegetation structure in the ecosystem, the greater the abundance of ant colonies ([Uhey et al., 2020](#)). The cleanliness and the plants' care in the smallholder plantations are not as good as in PTPN plantations. Stumps, fallen trees, and rubbish are abundantly found in smallholder plantations. Palm leaf fronds can be seen scattered a lot. The presence and abundance of ants usually occur more in places that are not quite disturbed by human activities because ants are insects that are sensitive to disturbances from human activities ([Gordon, 2019](#)). In PTPN plantations, sanitation is better maintained, and there are fewer fallen trees and rotting stumps of oil palm. Sanitation is carried out to eliminate the breeding site for *O. rhinoceros*.

**Table 1.** The colonies of *M. castanea* in oil palm plantations

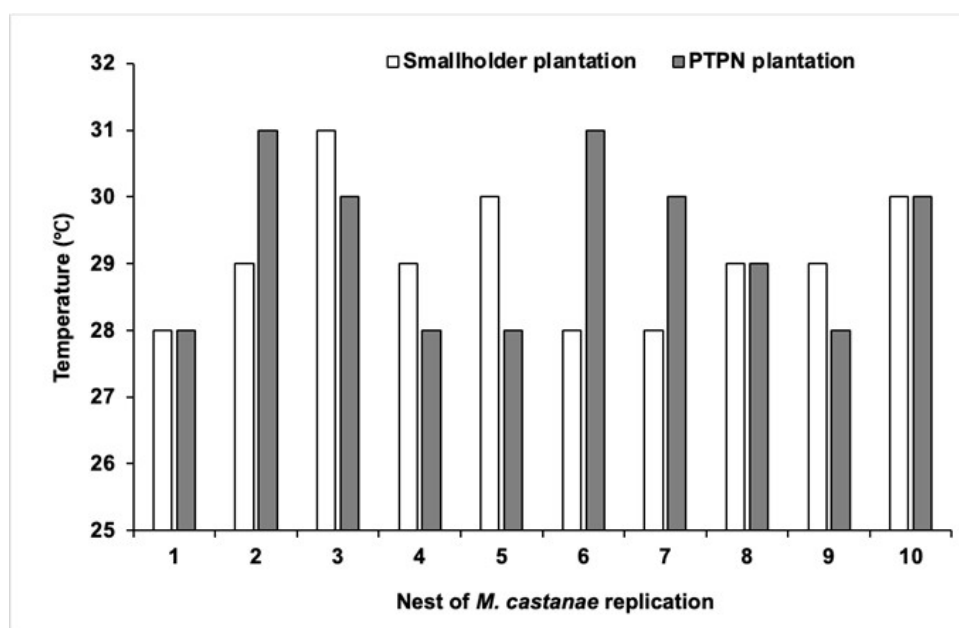
Colonies	Smallholder Plantation					PTPN Plantations				
	Eggs	Larvae	Pupae	Adult	Number of Female and Male in Caste*	Eggs	Larvae	Pupae	Adult	Number of Female and Male in Caste*
1	-	34	16	23		-	58	-	85	
2	-	102	19	121	Male = 3,	-	89	25	117	Male = 3, Female = 2
3	-	65	21	67		-	36	-	62	
4	-	103	5	69		-	128	23	79	Male = 2
5	-	145	5	83		-	58	62	53	
6	-	24	15	96	Male = 2, Female = 3	251	86	45	94	Male = 3, Female = 2
7	-	20	67	74		-	36	43	38	
8	279	63	9	102	Male = 7, Female = 10	-	126	27	82	Male = 5, Female = 3
9	-	52	15	60	Male = 2, Female = 5	-	35	23	65	
10	234	65	10	73	Male = 6, Female = 4	-	20	22	78	
Total	513	673	182	768		251	672	270	753	
Mean	51,3	67,3	18,2	76,8		25,1	67,2	27,0	75,3	
SD	82,08	29,62	10,48	18,96		45,18	32,04	13,08	16,64	

\*Note: Both males and females are present, indicating a full caste.

The number of ants obtained from smallholder plantations and PTPN plantations was not much different because the abiotic environment in both plantations is also relatively the same. The *M. castanea* ant colonies found did not all have reproductive castes (there were male and female ants). This is due to the gamergate phenomenon in *M. castanea* ant colonies. Gamergate is a phenomenon where worker ants can produce eggs. According to the explanation (Ito, 2010; Peeters & Fisher, 2016), more than 100 species of poneroid (subfamilies Amblyoponinae, Ponerinae, and Ektatommine) exhibit the gamergate phenomenon. Ito (2010) explains that *M. castanea* ants are polymorphic ants. *M. castanea* worker ants have varying numbers of ovarioles ranging from 6-22, while female ants (queen) have 24-32 ovarioles. It is suspected that worker ants with several ovarioles close to the number of ovarioles of the queen ant are the candidates for gamergate ants. However, there has been no study explaining the mechanism for the occurrence of this gamergate phenomenon in the Amblyoponinae family ants. Monnin & Peeters (2008) state that gamergate ant colonies generally have fewer colony members than ant colonies that have real queens.

### Abiotic Environment of *M. castanea* Nest

The results showed that the temperature obtained from all the *M. castanea* anthills explored was in the range of 28.0–31.0 °C (Figure 2). The average temperature in the smallholder plantation was  $29,1 \pm 0,994$ , and the mean temperature in the PTPN plantation was  $29,7 \pm 0,949$ . The average temperature obtained in community-owned smallholder and PTPN-owned plantations was not significantly different ( $0,7052 \geq 0,05$ ). This is because the age of the plants in the two gardens at the time of exploration is almost the same, ranging from 15 to 20 years. In these oil palm plantations, which are between 15 and 20 years old, the canopy of plants has protected each other so that the environment of the plantation becomes shadier. The temperature range of the environment during exploration in the field environment is 29-33 °C.



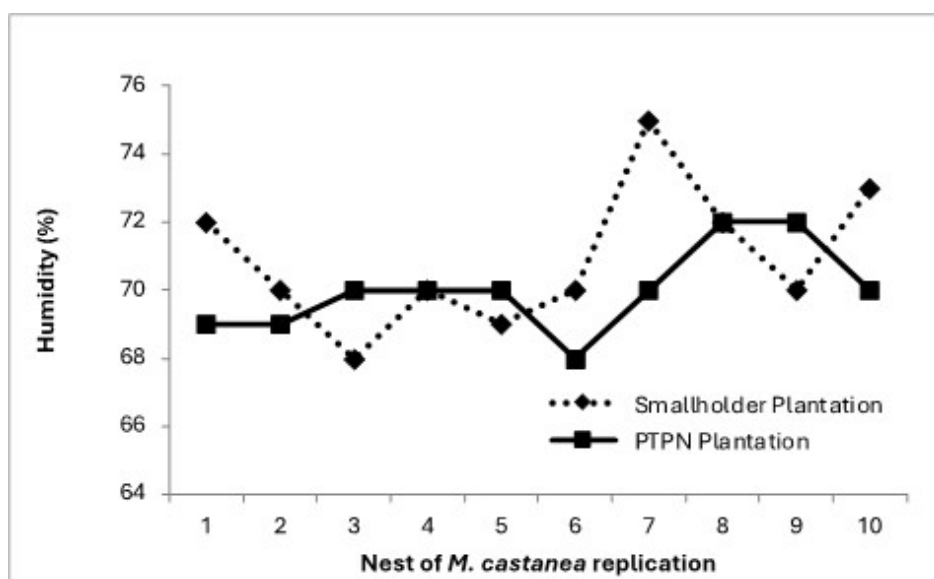
**Figure 2.** The temperature in the nest of *M. castanea*

Abiotic factors such as temperature can regulate the growth and distribution of insects in an ecosystem ([Wills & Landis, 2018](#); [King et al., 2018](#)). Temperature influences all aspects of ant biology. Temperature can affect the condition of ant colonies outside and inside the nest and their foraging activities ([Parr & Bishop, 2022](#)). [Tschá & Pie \(2019\)](#) explain that *Pheidole* sp. is more active in looking for food at higher temperatures, namely around 40°C. The interactions between herbivores and natural enemies in an ecosystem can be influenced by the regulation of abiotic factors of the ecosystem ([Tiede et al., 2017](#)).

Temperature can also affect the activity of ants in foraging. [Lei et al. \(2021\)](#) explain that *Solenopsis invicta* ants actively forage for food in the morning, and as the temperature increases, foraging activity will also decrease. Temperature can also affect the activity of ants in foraging. The results of the study by [Spicer et al. \(2017\)](#) explained that larger-sized ants (*Cephalotes atratus*) were slower in responding to temperature fluctuations than smaller-sized ants (*Azteca trigona*).

Humidity is an important factor affecting insect spread, activity, and development. In general, insects have a water content of about 50-90% in their bodies. This condition can be maintained if the humidity of the environment ranges between these values ([Chong & Lee, 2009](#)). Humid conditions in the habitat can be affected by rainfall conditions, nest material, and shading around the habitat ([Bollazzi & Roces, 2010](#)). *M. castanea* ants belong to ants that like damp places to make their nests ([Wilson, 1971](#)). The measurement of the average humidity of *M. castanea* ant microhabitat obtained in smallholder oil palm plantations was 70.9% (70,9±2,079) and 70.0% (70,0±1,247) for PTPN's oil palm plantations. This result was not significantly different for the two plantations (0,25311 ≥ 0,05). During the exploration of *M. castanea* ants carried out in smallholder plantations and PTPN plantations, *M. castanea* ant colonies were often found on rotten oil palm trunks that looked moist, and ants were usually found on the floor of the fallen and stacked palm trunks (Figure 3).

[Ratnasari \(2017\)](#) explains that the activity of *Paratrechina longicornis* ants that become pests in settlements is more active in foraging during humid air (cloudy weather) than in hot air (sunny

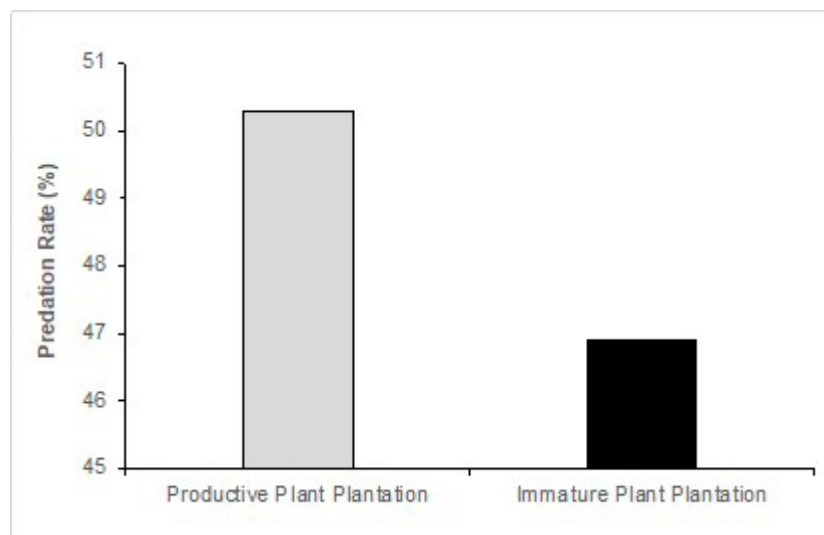


**Figure 3.** Humidity of *M. castanea* nest

weather). [Ronque et al. \(2018\)](#), observing activity in *Camponotus renggeri* and *C. rufipes* ants living in Brazilian savanna areas, stated that the humidity around the nest strongly influenced the activity of these ants. These ants were also found alive and made nests on the stacked logs.

### Predation Tests in The Field

The results of the predation test in the field between the TBM field (Immature Plant Plantation) and the TM field (Productive Plant Plantation) showed that the predation in the TM field was higher than in the TBM field (Figure 4), but statistically, these predation rates were not significantly different ( $P \geq 0.05$ ). The  $P$  value obtained was 0.59190, and the  $t$  value was 0.55578. The correlation value obtained is 0,13755. *M. castanea* ants could prey on the second instar larva of *O. rhinoceros* larvae by 46.87% for 5 days of exposure in TBM fields and 50.3% in TM fields. This difference is generally affected due to differences in the abiotic environment (microhabitat) in the two locations. Differences in the microenvironment of habitats can affect the performance of natural enemies ([Rocha & Fellowes, 2018](#)).



**Figure 4.** Predation of *M. castanea* on field

In TM fields, the existing oil palm plants are, on average, 15-20 years old, so the plant canopy has closed and made the environment below or at ground level more shaded so that the temperature becomes lower and the humidity becomes higher. Meanwhile, in TBM fields, the existing plants are still in the TBM 1 plants or those aged about one year, where the abiotic environment in the TBM fields is more open so that the temperature is hotter and the humidity becomes low. [Luskin & Potts \(2011\)](#) explain that the temperatures in the young and old oil palm plantations differ. The temperature in young plantations averages above 32 °C, while the temperature in old plantations averages below 30 °C. The lower temperature of the environment in these aging plantations is due to the canopy of plants that are close to each other. The abiotic environment greatly affects the abundance and performance of ants as predators ([Damien & Tougeron, 2019](#); [Nascimento et al., 2022](#)).

The process of prey orientation by *M. castanea* ants on prey larvae of *O. rhinoceros* begins with ants touching and circling their prey, biting slightly, and then avoiding, causing the prey to react by

writhing. Usually, the ants will bite the center of the abdomen of their prey a little bit first before stinging them. In this process, the prey usually provides resistance by writhing and trying to catch the ant with its mandibles. If an ant is caught, the prey will also be bitten by the ant, and the ant will also die in the process of this resistance. Ants will shake their bodies and antennae to call other worker ants to help them. [Ito \(2010\)](#) also expressed the same thing, observing the preying behavior of *M. castanea* ants toward *T. molitor* prey that also shook their bodies and antennae to call for help from other worker ants. In observations during the study, about 4-7 ants were working together to immobilize one prey. After the prey looked weak and helpless, it was left alone and temporarily abandoned by the ant. After the prey was completely dead, the ants ate the prey's hemolymph fluid.

## CONCLUSION

Ant colonies found in oil palm plantations differ in the number of individuals in the colony. The average temperature and humidity in the ant nest are 29.4 °C and 70.5%, respectively. Information on this abiotic environment will make it easier to rear these ants in the laboratory for use in biological control augmentation techniques. The predation rate in the field reaches 50.3%. The abiotic environment of ants obtained in the two plantation locations was not statistically different but still had an influence on the presence of *M. castanea* ants in oil palm plantations and still opened up opportunities to be able to utilize *M. castanea* ants as biological agents for *O. rhinoceros*.

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