Cuticular Permeability of Two Sympatric Species of *Macrotermes* (Blattodea: Termitidae)

by

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Abstract

The ability of water reserve may influence the foraging behavior of termites. Cuticular permeability (CP) and water loss of *Macrotermes gilvus* (Hagen) and *M. carbonarius* (Hagen) were examined in laboratory. The CP values of both species did not differ significantly ($P > 0.05$). However, *M. carbonarius* had higher body water content and lower ratios of surface area to volume than those recorded in *M. gilvus.* This suggested that *M. carbonarius* is more tolerant to drier environment and allow moderate desiccation than *M. gilvus*.

Key words: Cuticular permeability, sympatric species, foraging above-ground, *Macrotermes gilvus*, *Macrotermes carbonarius,*

Introduction

Water reserve is important to subterranean termites, because they have very little resistance to dehydration with relatively high cuticular permeability (CP) (Arquette et al. 2006, Sponsler and Appel 1990). Termites forage for food above-ground with mud tubes protecting themselves from water loss. Different termite species have been found to exhibit different mud tubes geometries or patterns (Campora and Grace 2001, Su 2001). However, not all foraging termite species required the protection of mud tubes.

The mound-building termites *Macrotermes gilvus* (Hagen) and *M. carbonarius* (Hagen) are abundant in Southeast Asia, especially in plantations. Both species feed on cellulose debris (e.g., plant litters). Colonies of both species consist of a or multiple pairs of queen and kings, nymphs, minor and major workers, as well as minor and major soldiers (Neoh et al. 2010). One of the most conspicuous behavioral differences between these two sympatric species is that *M. gilvus* forages below ground or concealed within mud foraging tubes, whereas *M. carbonarius* forages above ground in open air with extensive trails that may reach up to a distance of 23 m. Furthermore, *M. carbonarius* could forage within a broad range of relative humidity from 69 - 86% above ground without mud tube protection (Hu, J., unpublished data). The abilities to reduce desiccation are crucial to subterranean termites and may influence termite routine, particularly their foraging activity.

Previous studies reported that *M. gilvus* often forages below ground or in a concealed mud foraging tubes, while *M. carbonarius* often forage in the open (Sugio 1995). In this study, we determined the cuticular permeability of both species. We hypothesized that *M. carbonarius* has lower cuticular permeability than *M. gilvus*.

Materials and methods

Cuticular permeability was determined gravimetrically. A total of 6 minor workers, 6 major workers, 6 minor soldiers and 3 major soldiers from 3 colonies of each species were selected. The experiment was replicated 7 times for each colony. Only healthy termites were used and were handled using filter paper into uncovered 22 ml glass vials. Initial masses were determined and then vials held at 29 ± 1^{o}C in an 11-l glass desiccator containing 1 kg anhydrous $CaSO_4$ (≈0 - 2% RH, resulted in saturation deficit of 29.764 mmHg in the desiccator). The desiccant was dried at 200^{o}C for a minimum of 1 h. At 2 h after placing the vials in the desiccator, mass loss was determined with a 0.01 mg sensitivity balance (Appel 1993, Appel and Tanley 1999, Shelton and Grace 2003). Following the mass determination, vials of termite were transferred to an oven at temperature of 55^{o}C for 72 h to be dried to constant mass, and then reweighed. Because water loss during the first 2 h of desiccation accounts for the cuticular permeability than during the other time periods (Appel et al. 1983, Mazer and Appel 2001), then CP values were calculated using the difference between the initial and 2 h masses as μg of H_2O lost per surface area (cm^2) per unit time (h) per saturation deficit (mmHg)(Edney 1977). Surface areas were estimated for each specimen using Meeh (1897) formula: $S=12M^{2/3}$, where *S*, body surface area (cm^2) and *M*, initial mass (g). To test if Meeh formula provided a biased estimating of surface area, CP values were regressed against fresh mass (Shelton and Grace 2003). Water mass was calculated as the difference between fresh mass and dry mass.

Statistical analyses were performed using Statistix 9.0 (Analytical Software 2008). Body mass, water mass and CP value water were analyzed by ANOVA, followed by Tukey HSD tests for post-hoc comparison of means.

Results and discussion

CP values using Meeh (1897) surface area model estimates were not significantly ($P > 0.05$) related to initial mass for all castes, indicating that surface area estimation model did not cause biasness to the CP values. CP values ranged from 14.96 to 47.62 μg cm^{-2} mm Hg^{-1} h^{-1} for *M. gilvus* and from 14.74 to 49.52 μg cm^{-2} mm Hg^{-1} h^{-1} for *M. carbonarius* (Table 1). There were significant differences within *M. gilvus* castes (F=285.15, d.f.=3, P<0.001) and within *M. carbonarius* castes (F=67.97, d.f.=3, P<0.001). The CP of the minor soldier CP was significantly less than that of the minor worker, major worker and major soldier in both *M. gilvus* and in *M. carbonarius* ($P < 0.05$). However, there was not a significant difference between species of the corresponding caste. This indicated that the CP of caste. i.e. the water loss rate per area under the same condition was similar between *M. gilvus* and *M. carbonarius*.

Fresh mass of termites ranged from 5.14 to 26.49 mg for *M. gilvus* and from 11.25 to 69.17 mg for *M. carbonarius* (Table 1). For both species, major soldiers had the greatest fresh body mass, and it was significantly different from minor workers, major workers and minor soldiers ($P < 0.05$); minor workers had the least fresh body mass. Between species, *M. carbonarius* had significantly greater body mass than in *M. gilvus* ($P < 0.05$). Although surface area is associated with fresh mass, the increase of surface area of *M. carbonarius* is less than that in its fresh mass (Table 1).

Body water mass ranged from 4.08 to 19.67 mg for *M. gilvus* and from 9.11 to 52.92 mg for *M. carbonarius* (Table 1). In both species, major soldiers water mass was significantly higher than in other castes (Table 1). There was also significant difference between the two species in corresponding caste. This suggested that *M. carbonarius* allows relatively higher amount of water to desiccate compared to *M. gilvus.*

Table 1 Mean (± S.E) CP values, fresh mass, surface area and body water mass of *M. gilvus* and *M. carbonarius.*

Species	Caste	n	CP	Fresh mass (mg)	Surface area (cm^2)	Body water mass (mg)
M. gilvus	Minor workers	112	47.62± 0.78b	5.14± 0.14c*	0.36 ±0.01c*	4.08 ± 0.09c*
	Major workers	112	51.51± 0.93a	9.30± 0.32b**	0.53±0.01b**	7.55 ± 0.24b**
	Minor soldiers	112	14.96±0.45d	5.69± 0.12c***	0.38± 0.01c***	4.50 ± 0.08c***
	Major soldiers	56	28.97± 1.54c	26.49± 0.31a†	1.07± 0.01a†	19.67 ± 0.30a†
M. carbonarius	Minor workers	112	47.49± 1.34x	11.25 ± 0.22z*	0.60± 0.01z*	9.11 ± 0.17z*
	Major workers	112	49.52 ± 2.15x	24.91 ± 0.71y**	1.02± 0.02y**	20.73 ± 0.53y**
	Minor soldiers	112	14.74 ± 0.97z	23.49 ± 0.49y***	0.98± 0.01y***	19.25 ± 0.52y***
	Major soldiers	56	27.86 ± 2.98y	69.17 ± 1.00x†	2.02 ±0.02x†	52.92 ± 0.81x†

Means followed by the different letters within a group (abcd or xyz) are significantly different ($P < 0.05$, Tukey's HSD). Means followed by the same symbol (*, **, *** or †) are significantly different within the same caste ($P < 0.05$, Student t-Test, two tails).

CP is defined as μg of H_2O lost per surface area (cm^2) per unit time (h) per saturation deficit (mmHg) and is a means of describing and comparing water loss from insect and other arthropods (Edney 1977, Shelton and Appel 2001). A number of authors have used a relative lower CP values to explain arthropods adapted to xeric environments, suggesting that arthropods can conserve a great amount of water may tolerate to drier environmental condition (Appel et al. 1983, Hull-Sanders et al. 2003, Mack and Appel 1986). In addition, Arthropods living in wet or humid habitats usually have much higher CP values (Edney 1977, Hadley 1994). However, the situation differed from what was observed for *M. gilvus* and *M. carbonarius* as there was no significant difference in CP value between both species (Table 1). Similar result in beetles, *Megetra cancellata* and *Pleuropasta reticulate*, was also found by Cohen and Pinto (1977). One proximate explanation is that *M. carbonarius* has larger body size (≈3 times as *M. gilvus*) as well as larger body water mass (≈3 times as *M. gilvus*) (Table 1) compared to *M. gilvus*. Furthermore, *M. carbonarius* has lower ratio of surface area to volume. Thus, this allows *M. carbonarius* more tolerate to moderate desiccation but this not in the case of *M. gilvus*.

Conclusions

This study rejected the hypothesis that *M. carbonarius* has greater CP values than in *M. gilvus*. Instead, the lower ratios of surface are to volume and the body water mass in *M. carbonarius* plays an important role in encountering desiccation stress, particularly during foraging activity. Other physiological parameters such as percentage of body water, rate of water loss, body lipid content and compression strength (Suzuki et al. 2009) may also have significant roles and are currently undertaken.

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