

Peculiarities of Trophallaxis in the Termite Colony of the *Anacanthotermes Turkestanicus* Family

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Abstract: The article reveals that in termites of *Anacanthotermes* genus of various ages and castes, the degree of self-nutrition and the bolus travel speed differ; also the stomodeal trophallaxis is revealed, namely, mouth-to-mouth. The data provided indicate on the great biological role of the termites trophallaxis in the family and may be the fundamental principle to develop the control policy of the termite population.

Keywords: Termites, *Anacanthotermes*, trophallaxis, castes, termite worker, soldiers, nymph, food

1. Introduction

In all Central Asian countries, one of the harmful pests that considerable damage the buildings and structures, including cultural historic monuments, are the turkestanica (*Anacanthotermes turkestanicus* Jacobs, 1904) and the large Trans-Caspian (*A. ahngerianus* Jacobs, 1904) termites (Khamraev, 2006). It is well-known that such phenomenon as trophallaxis is common with social insects, and in particular in termites. This is the food exchange between the genus members (Waller, 1996). Trophallaxis, or mutual feeding, is the phenomenon that is common in colonial insects to a greater or lesser degree, binding all the family members to a single food chain. Separate castes and/or ages in the family may be interdependent, and individual castes may partially interrelate and some ages of various termite castes may feed themselves independently. Such dependent nutrition allows the scholars to investigate the termite genus as the whole organism (Brian, 1986). The investigation data available today that relate to the food chain in termites is diffused and such data is not available for the termites of *Anacanthotermes* genus of the Central Asian region, yet. The issue on trophallaxis peculiarities is still not investigated on the level of inter-colonial interactions in terms of termites of *Anacanthotermes* genus.

In this view, a question arose to investigate the role of trophallaxis in the colony of termites of *Anacanthotermes* genus.

2. Materials and Methods

All researches were performed in a laboratory environment. Termites (*Anacanthotermes turkestanicus* Jacobs, 1904) were delivered from the Khorezm region in special containers.

They were further classified by caste and age categories (aged I-II, III-IV, V-VI, VII-VIII), the age of termites was defined by the number of joints in horns (Nurzhanov, 2005). The termite workers of all ages and castes were placed in separate Petri dishes in 10 species each (in 3 replications). The filter paper stained in 0.5% neutral red was used as the food. The changes in the color of the hind body served to define the degree of self-nutrition in termites. The number of

termites was accounted to 30 insects for each experiment subject to replications.

Colorability of the termite abdomen was visually assessed as follows: no stain - 0 point; too weak stain - 1 point; weak stain - 2 points; medium stain - 3 points; good stain - 4 points; very good stain - 5 points. The data was tabulated in the end of the experiment based on the total values obtained.

Investigation of the mutual feeding activity. Different termite ages and castes were divided into 2 equal groups (10 species in each group). One of the group was pre-stained (by the use of filter paper treated with 0.5% neutral red solution) and the latter remained without staining. In 2-3 days upon appearance of the stable staining, the stained and unstained termites were mixed and contained in a Petri dish filled in with the porous material, which was moistened with the drinking water on a daily basis. The appearance of the stain on those termites that were no stained earlier in absence of any food was the indication of its transfer from the stained termites by means of trophallaxis.

The activity of food transfer was evaluated by appearance of stains in the unstained termites upon keeping of the latter together with stained termites as per the following point system:

0 - no transfer, 1 point - very poor, 2 points - weak, 3 points - average, 4 points - good and 5 points very good ability for food transfer to species of different castes and ages in the entire family.

Upon completion of the experiment the results were tabulated with a view of the overall number of points obtained.

Investigation of food interaction between the worker termites of different age groups. To investigate the different ways of food transfer (stomodeal or proctodeal trophallaxis) between the worker termites of *Anacanthotermes* genes, these termites were shoot for two days monitoring their activity in Petri dishes with the filter paper placed on the bottom. The termites (10 species - 8 elder worker termites and 2 female termites) were placed under the lens of the video camera. The shoot duration to monitor the behavior of termites was 2 days. Then, the video recording was reviewed and analyzed.

3. Results and Discussion

Trophic activity of termites of certain castes and age groups. It was revealed through the series of experiments that the life span and body weight of termites is sharply reduced when they are kept in isolation (Ganieva Z.A., 2013). Accordingly, this it conveys the suggestion on the vital biological link between the species of its family and colony of termites. To identify the causes of this phenomenon, the age and caste specificity of termites independent feeding was to be specified. The results obtained in this series of observations are shown in the Table 1.

It was found that no stain was detected on the abdomen of the larva. The worker termites of younger (I-II) age that were fed with the filter paper as a food, treated with the phenol red stain slightly although the degree of staining of their bodies was not as intense as in termites of other groups. The worker termites of the middle and elder age groups (III-

IY, Y-YI and YII-YIII age groups) were reported as the most intensely stained in their abdomen, i.e. they were identified as the most active trophically.

According to the lack of stain in the abdomen of soldier termites, they were not involved in independent feeding at all. As for nymph termites, these species had their abdomen stained in both age groups where the staining effect in elder nymph termites was more intense.

Thus, the results of this experiment indicate that the degree of independent feeding in different age groups of termites is not identical. When the sum of points obtained upon the end of the experiment is reviewed, higher values (degree of self-feeding) are reported in groups of middle and elder age, younger worker termites show less ability to self-feeding, it is quite adequate in nymph termites and is absent in larva and soldier termites age group.

Table 1: Degree of self-feeding in different castes and age groups of worker termites

Developmental stage	Age	Degree of staining of termites in points by the days of registration										Total sum of points	
		1	2	3	4	5	6	7	8	9	10		
Larva		0	0	0	0	0	0	0	0	0	0	0	0
Worker termites	I-II age groups	0	1	1	1	2	2	2	3	3	3	18	
	III-IY age groups	4	4	4	4	5	5	5	5	5	5	46	
	Y-YI age groups	4	4	4	5	5	5	5	5	5	5	47	
	YII-YIII age groups	3	3	4	4	4	5	5	5	5	5	43	
Soldier termites	I-II age groups	0	0	0	0	0	0	0	0	0	0	0	
	III-IY age groups	0	0	0	0	0	0	0	0	0	0	0	
Nymph termites	I-II age groups	0	0	2	2	2	3	3	3	3	3	21	
	III-IY age groups	0	2	2	2	2	3	3	4	4	4	26	

Rate of food evacuation in different castes and age groups of termites

The food evacuation rate was determined by the speed of travel of the paper stained with the methylene blue through the gastrointestinal tract of termites until it is completely full and then, after the termites were fed with the white filter paper, until complete disappearance of the stained food.

It was revealed that the alimentary canal was completely stained with the stained food in all groups of worker and nymph termites.

Within 2 hours after the start of the experiment, the alimentary canal in the abdomen of elder worker and nymph termites was filled with stained food. The alimentary canal in the medium age and junior groups of worker termites was filled in with the stained food slightly later.

When the termite were caused to change their food from stained to the white filter paper, it turned out that the stain in the upper part of the abdomen started to fade. And gradually, all the colored food was completely replaced with the uncolored one. However, such a "replacement", i.e. food migration rate in different age groups of termites was non identical.

In the elder group of worker termites the colored food reached the mid-line of the intestine on the 4th -5th day and was completely digested in the alimentary canal on the 6th

day. As for the group of medium age worker termites, the stain on their abdomen half faded on the 7th day of observation and completely disappeared on the 21st -22nd days. And finally, the group of junior age worker termites showed almost the same tendency for food evacuation through the gastrointestinal tract as that in the medium age group, though their intestinal tract completely cleaned up from the stain a bit later - on the 23rd -24th days.

The rate of food evacuation through the gastrointestinal tract in the nymph termites was identical to that in elder worker termites.

Thus, the rate of evacuation of the food bolus is nonidentical in termites of different age and caste. However, it is well manifested in nymphs and elder worker termites. The rate of food evacuation in medium and junior age worker termites decreased 3 times as less as compared with that in the elder age groups.

Mutual feeding in different castes and age groups of termites

The process of mutual feeding or trophollaxis was identified by the stain that appeared on the abdomen of "original" termites when stained and unstained species were kept together and no other food was available. Consequently, in such circumstances the colored food could be transferred to the intact termite only through feeding by the pre-stained termite.

Since previously conducted observations showed that junior age termites (I-II age) can hardly feed themselves, and soldier and larva termites are not able to nourish, it made no sense to investigate their role in process of food transfer to other species. Therefore, investigations were conducted on termites of other age and castes.

The resulting degree of food transfer from stained to unstained species are shown in the Table 2. The stain transfer was reported as too poor in almost all variants investigated, when stained worker termites of III-IY age groups were mixed with unstained worker termites of identical age and elder age groups as well as with nymph and soldier termites. This is to indicate that the worker termites of III-IY age groups are able to participate in the food transfer process but not as actively as species from elder age groups.

Further, we review the food transfer from the worker termites of Y-YI and YII-YIII age groups to termites of other age and castes. It was revealed that the worker termites of Y-YI age managed to pass the food to all the age groups within the same caste, however such a tendency to transfer food was particularly obvious regarding the youngest age groups. Food transfer by worker termites of this group to

soldier and nymph termites was also less intensive as compared with that to worker termites.

By the outcome of our observations, the most active "breadwinners" were worker termites of the Y-YI age groups. In particular, these worker termites were more active to feed worker termites of the I-II age groups and to some weaker extent, to feed worker termites of the III-IY and Y-YI age groups.

Elderst worker termites (YII-YIII age groups) were used to get "stained" by termites of the medium age but not as obviously as it was observed in younger age groups. Since soldier termites were not able to feed themselves, were significantly marked with the colored food delivered to them by the worker termites, while the nymph termites, as expected, were quite slightly stained what responded to their partial ability to feed themselves.

Worker termites of the eldest age group (YII-YIII) are involved in feeding worker termites of all the studied age though the intensity of their participation in mutual feeding is slightly inferior to feeding worker termites of the Y-YI age. It should be noted that the food transfer by the worker termites of that age group to soldier and nymph termites was the most intense.

Table 2: The intensity of mutual feeding between the worker termites of different ages and termites of other castes

Age groups and castes of termites		Degree of staining of termites in points by record days										Total amount of points
Stained	Unstained	1	2	3	4	5	6	7	8	9	10	
Worker termites	Worker termites											
III-IY age groups	I-II age groups	0	1	1	2	2	2	3	3	3	3	20
III-IY age groups	III-IY age groups	0	1	1	1	2	2	2	3	3	3	18
III-IY age groups	Y-YI age groups	0	0	0	1	1	1	2	2	2	2	11
III-IY age groups	YII-YIII age groups	0	0	0	0	0	1	1	1	1	1	5
III-IY age groups	Soldier termites	0	0	0	0	1	1	1	1	2	2	8
III-IY age groups	Nymph termites	0	0	0	0	0	1	1	1	1	1	5
Worker termites	Worker termites											
Y-YI age groups	I-II age groups	0	1	3	4	4	4	4	4	4	5	33
Y-YI age groups	III-IY age groups	1	2	3	3	3	3	3	4	4	4	30
Y-YI age groups	Y-YI age groups	2	2	3	3	3	3	4	4	4	4	30
Y-YI age groups	YII-YIII age groups	2	2	2	3	3	3	3	3	4	4	29
Y-YI age groups	Soldier termites	0	0	1	1	1	3	3	3	3	3	18
Y-YI age groups	Nymph termites	0	0	1	1	1	2	2	3	3	3	16
Worker termites	Worker termites											
YII-YIII age groups	I-II age groups	0	1	1	2	3	3	3	3	4	4	24
YII-YIII age groups	III-IY age groups	2	3	3	3	3	4	4	4	4	4	34
YII-YIII age groups	Y-YI age groups	1	1	2	3	3	3	3	4	4	4	29
YII-YIII age groups	YII-YIII age groups	0	1	1	2	3	3	3	3	3	3	22
YII-YIII age groups	Soldier termites	0	0	1	1	3	3	4	4	4	4	24
YII-YIII age groups	Nymph termites	0	1	1	3	3	4	4	4	4	4	28

Thus, the ability of the worker termites to be engaged in mutual feeding increases as they grow older but at the same time this slightly decreases in the eldest termites. In the chain of trophallaxis, the worker termites of younger age and soldier termites are rather recipients than donors. Worker termites of the medium age and elder age feed on their own and are actively involved in nourishing worker termites of other ages as well as termites of castes, in particular, nymph and soldier termites. Mutual feeding between termites of different castes takes place even in case of abundant availability of food and the ability of "donors" and "recipients" to feed themselves.

This suggests that in the process of trophallaxis not only food is transmitted but also other biologically important information which allows preservation of the termite family and colony as a whole as well as the main determinant of the prosperity of the species.

Food transfer method

The described researches above suggest that certain termite castes cannot feed on their own at all (larva, soldiers) or the nourishment is reported as too passive (I-II age group worker and nymph termites). Furthermore, they show that the "chain" food transfer exists between species of termites.

Therefore, it was necessary to identify the methods by which the termites transfer food to each other, either by stomodeal (mouth to mouth) or proctodeal (licking of feces) method.

Video recording of termite behavior in the laboratory environment showed that the main way of the worker termites feeding is independent manifested as eating away at the paper by termites. However, the colony of termites is known for the trophallaxis known as the mutual feeding. It turned out that the main way of mutual feeding is stomodeal, i.e. mouth-to-mouth (Fig. A).

As for the proctodeal method known for other types of termites, it was not so often observed within a two-night observation. This is probably due to the fact that this method of feeding is usually used by termites of *Anacanthotermes* genes for "inoculation" of the sterile intestine observed upon the rejection of the horny intestinal epithelium of the relevant microflora upon its molting.

In addition, the grooming process, that is brushing, licking, is often reported in termites (Figure D). Perhaps grooming not only helps cleanse one individual but also is a source of nutrient and biologically active material, bacteria, chemical information for another individual.

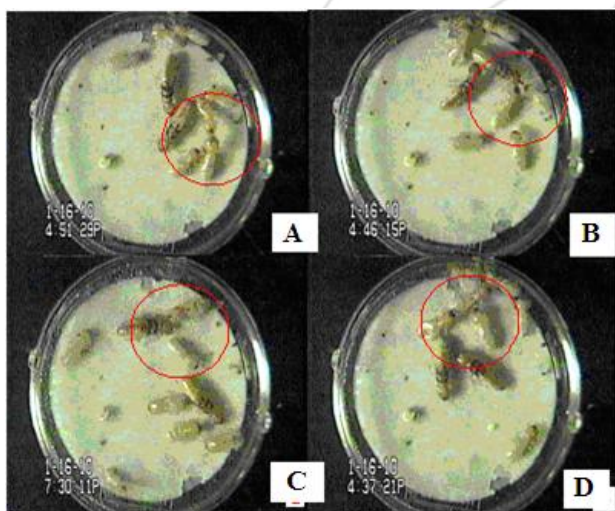


Figure: Some types of termite feeding behavior. Mutual feeding: A - between two worker termites; B - between two worker termites and a female termite; C - between a female termite and one worker termite; D - grooming: two worker termites 'clean' the third one.

It was proven in the laboratory conditions that worker termites of the middle and older age groups feed on their own, however, there are cases of a food transfer between the said groups. In mutual feeding the main mode of a food transfer is stomodeal method, ie, mouth-to-mouth.

Our observations have shown that this method is used by the worker termites for a food transfer between each other (Figure A), from the worker termites to the nymph termites (Figure A, B, C.) and from the worker termites to the soldier termites (Table 2).

Thus, phenomenon of trophallaxis can be considered as one of the key elements in maintaining the family and colonies of termites, including the species of the *Anacanthotermes*

kind. This biological feature of the colony of termites can be used to develop control measures against them.

4. Conclusion

The degree of independent feeding is non-identical in different groups and ages of termites, i.e., it is the most strongly pronounced in the middle and older age groups, slightly expressed in the younger age groups of worker termites, quite noticeable in nymph termites and is absent in larvae and soldier termites.

- 1) It was found that the rate of movement of food bolus is non-identical for different age groups and castes of termites, ie, expressed in the groups of nymph and older age worker termites, and in the middle and younger age groups of the worker termites the rate of food movement decreased for 3 times in comparison with the older age groups.
- 2) It was found that termites of *Anacanthotermes* kind use a mutual feeding - trophallaxis, and the main way of a mutual feeding is stomodeal method, i.e. mouth-to-mouth.
- 3) Also, it was determined that the ability of the worker termites to engage in mutual feeding increases as they grow older but the ability of individuals of the most older age groups is somewhat weakened.

In the chain of trophallaxis the worker termites of the younger age and soldier groups are mostly recipients than donors. The worker termites of the middle and older groups feed on their own and are actively involved in feeding of worker termites of other ages as well as the castes, especially the nymph and soldier termites.

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