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## Determination of structural features of the nest material of *Crematogaster rogenhoferi* (Mayr, 1879), (Hymenoptera: Myrmicinae)

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### Abstract

The aim of this study was to determine the structural features of the nest materials of *Crematogaster rogenhoferi* with some parameters like height above the ground in which the nest carton was built or constructed, weight of the nest material, length, width, diameter of cell, height of trees, element compositions, absorbency capacity and temperature were studied in the month of September 10, 2014. The nest materials are thick and condensed, which constitutes plant fibers and soil particles. Presences of fungal hyphae were seen under SEM. The average height above the ground was  $242.7 \pm 54.42$  cm, weight  $494.4 \pm 72.58$  cm, length  $17.26 \pm 0.15$  cm, width  $7.92 \pm 0.37$  cm, diameter of the cell chamber  $2.06 \pm 0.31$  cm and height of trees was  $9.04 \pm 4.79$  cm respectively. Oxygen and Carbon were high concentrations while K, Hg, Cu, Al and Br were in low concentrations. The total percentage of water absorbency capacity was  $22.92 \pm 9.11$  and the average temperature was about  $22.35 \pm 0.86$  °C.

**Keywords:** *Crematogaster rogenhoferi*, nest material, physical properties, absorbency, temperature, elements

### 1. Introduction

*Crematogaster* ant commonly known as Acrobat ants belongs to Order: Hymenoptera and Family: Formicidae. *Crematogaster* ants are especially dominant in the tropical region where they are remarkably diverse on vegetation [1]. Arboreal nesting is a derived feature in ants [2]. The nest sites are generally found in different types of trees with a height ranges from limited recourses for ants [3]. Ants have a diverse range of nesting habits, from subterranean, soil-nesting species [4] to arboreal species nesting in the upper forest canopy [5]. Arboreal ants nest in naturally occurring tree cavities [6], as well as those created by bark and woodboring insects [7] or excavated by themselves [8]. Alternatively, they may construct nests from materials such as spider silk, larval silk [9] or leaves that are woven together [10]. The presence of domatia can provide a selective advantage to plants by facilitating biotic defense through ant presence [11]. Nests serve many functions including sheltering the queen, providing the most favourable conditions for rearing brood, and as places for tending trophobiotic insects. They provide refuges for workers from fluctuating external environmental conditions, as well as from predators and competitors [12]. For example, nest structure is important for ants in deserts and other arid environments, as the nest provides protection against desiccation and a stable temperature for rearing brood [13]. The availability of nest sites can strongly influence the structure of ant communities and is known to be a limitation for litter, cavity-nesting ants (e.g. within acorns or twigs) and arboreal, twig-nesting ants [14-16]. Where nest sites are seasonally abundant in a lowland tropical rainforest, the coexistence of twig-dwelling ants is possible because nest sites are not a limiting resource [17]. The morphological characteristics of potential nest sites can influence nest site preference in ants, as seen in acorn-nesting *Leptothorax curvispinosus* [18].

Most ant diversity studies take place in tropical forests, where species diversity is particularly high [19-20]. These systems are paradoxical as competition is known to strongly influence arboreal ant communities and would be expected to lead to competitive exclusion and low species diversity. However, spatial partitioning through microhabitat variation can promote coexistence, leading to the high diversity of ant assemblages in these communities [21]. In this study, we determined the nests material in terms of its physical features like height above the ground in which the nest carton was built or constructed, weight of the nest material, length, width, diameter of cell and the height of trees.

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We also determined the concentration of elements, absorption capacity and the temperature of the nest material.

**Material and methods**

**Nest collection**

The nest of *Crematogaster rogenhoferi* was collected from West Jaintia Hills District Meghalaya, India (25°10'0" N, 92°0'0" E, Jowai) on September 10, 2014. Larvae, pupae and eggs were removed from the nest. Small fragments were cut from the comb for observation. The nests were stored in the Entomology Laboratory at Zoology Department of North Eastern Hill University, Shillong.

**Observation of surface and analysis**

The nests and other structures produced by ant colonies can be considered extensions of the superorganism, serving important physiological roles such as thermoregulation and nutrient storage [22]. Ants are dominant social insects that occur in a wide variety of habitats, and exhibit a vast diversity of nesting and feeding habits [23]. Ants are especially dominant in tropical habitats where they are remarkably diverse both on the ground and on vegetation [24]. The plant species used as nest (single or clumped epiphytes) and the height of the nest relative to the ground were recorded for each colony. Social insect colonies are frequently described as super organisms, in which groups of workers play various functional roles, and reproduction occurs at the colony level [25].

Small fragments from nest's wall were observed with a stereomicroscope (OLYMPUS) and scanning electron microscope (JSM – 6360, Jeol). The elemental composition analysis was made by SEM. The edge length and diameter of the nest material were measured by ruler.

**Absorbance**

Small fragments of the nest material were cut down and weighed in the electronic balance. The weight of the nest fragment were recorded and soaked in water about 30second. Each nest fragment was reweighed after immersion in water [26]. The absorption capacity of the nest fragment of ants, expressed as percentage, was estimated using the following formula: Formula 2:

Absorption capacity (%) = [(m2-m1)/ m1] x 100 m1= dried weight of sample before the process; m2 = dried weight of

sample after 30 s.

**Temperature inside the nest material**

The temperatures were measured randomly in different nest location with the help of digital thermometer. The digital thermometer was inserting inside the nest of ant and the readings was made in different replicates of different nest and the parameter was recorded. The optimum temperature is ±200 C to ±260 C.

**Statistical analysis:** Statistical analysis was done using Graph pad prism 4.5 version software.

**Results**

**Nest structure**

The morphology of nests was spheroidal to subspheroidal in shape (Fig.1). The nest were thick and condensed which constitutes plant fibers, soil particles etc. The nests were made up of brownish and grayish cartons. The outer surfaces of the nest were rough which are made from the plant debris and leaf litter. The plant fibers of the nests material were long and thin. The nest was built on different types of tree branches like *Schima wallichii*, *Schima khasiana*, *Aporusa dioica*, *Callicarpa arborea*, *Macaranga peltata*, *Quercus griffithii*, *Shorea robusta*, *Camellia caduca*, *Myrica esculenta* and other unknown species.



**Fig 1:** *Crematogaster* nest on a twig of *Camellia caduca*.



**Fig 2:** Nest internal morphology shows an overlapping arrangement.

**Table 1:** Physical features of nest material of *Crematogaster rogenhoferi* and vegetation type that ant builds their nest.

Nest	Above the ground (cm)	Weight (gm)	Length (cm)	Width (cm)	Diameter of cells (cm)	Tree species	Height of trees (m)
1	155	350	15.27±0.46	6.72±0.34	1.80±0.45	<i>Schima khasiana</i>	12
2	198	525	17.87±0.44	6.58±0.29	2.12±0.30	<i>Camellia caduca</i>	1.75
3	205	505	16.39±0.43	7.08±0.34	1.92±0.38	<i>Macaranga peltata</i>	13
4	235	575	18.73±0.69	7.88±0.66	2.30±0.15	<i>Aporosa dioica</i>	11
5	183	489	15.34±0.28	6.32±0.41	1.80±0.33	<i>Schima wallichii</i>	13
6	295	375	14.71±0.32	8.92±0.29	1.58±0.59	<i>Callicarpa arborea</i>	11
7	270	500	17.20±0.38	8.42±0.37	2.17±0.21	<i>Quercus griffithii</i>	13
8	315	535	18.37±0.14	10.11±0.22	2.20±0.24	<i>Camellia caduca</i>	2
9	321	505	17.27±0.22	6.34±0.38	2.05±0.26	<i>Schima khasiana</i>	12
10	250	585	21.45±0.34	10.88±0.49	2.67±0.19	<i>Shorea robusta</i>	1.65
Total X±SE	242.7±54.42	494.4±72.58	17.26±0.15	7.92±0.37	2.06±0.31		9.04±4.79

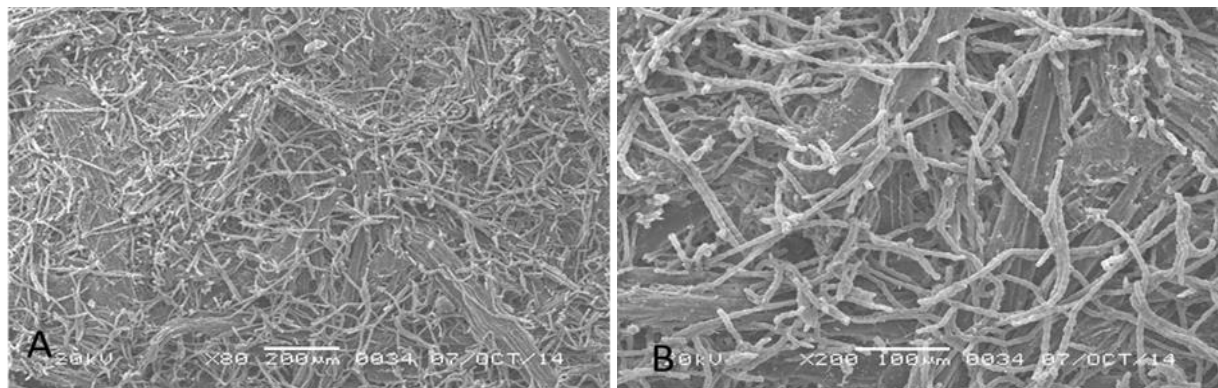
### Internal structure of nest material

The internal morphology showed an overlapping arrangement from the center towards the periphery of the nest. The chambered was arranged in a concentric manner (n=10) which was seen in the vertical and horizontal sections of the nest (Fig.2). The chambered are concavo-convex in shape and concavity which directed towards the center of the nest. The chambered of the nest are aggregated and tapping each other towards the periphery of the nest. The nest chambered are interconnected each other for ant travelling inside the nest. The chambers are quite close together and many are connected by short tunnels. The nests chambered are varying in thickness, shaped and its diameter. The center portion of

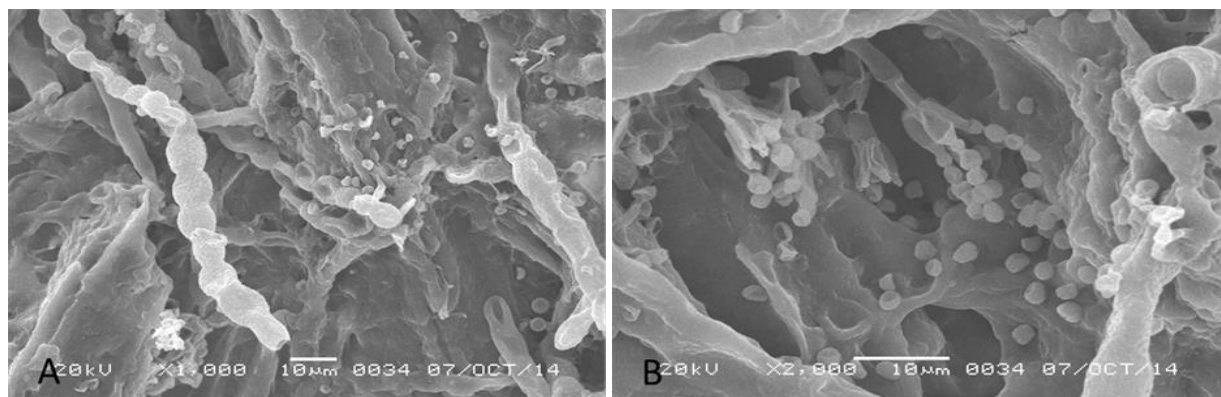
the nest is loosely bound while the periphery is usually thicker and condensed. The average diameter of the chamber was 2.06±0.31cm.

### Observation of nest material under SEM

Small fragments of the nest material were observed under scanning electron microscope (JSM – 6360, Jeol) shows the fungal patch or fungal hyphae present in the nest material (Fig: 4). The fungi grow on inner surfaces of the nest material which play an important role for protection of the larvae, young ones and served as their food materials. This indicates that the ants have a specific feeding behavior.



**Fig 3:** (A and B): Outer surfaces of the nest's wall of *Crematogaster* ant in SEM



**Fig 4:** (A and B). Inner surfaces of the nest's wall of *Crematogaster rogenhoferi* shows the fungal hyphae present in the nest material (SEM).

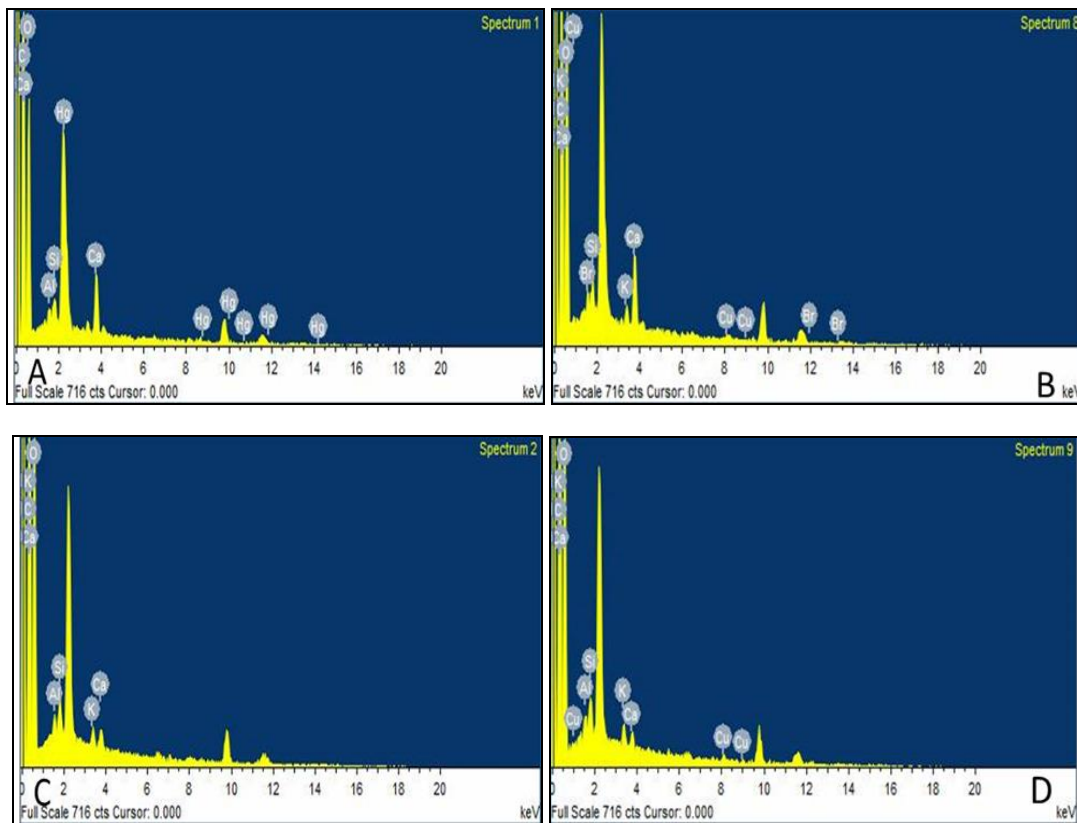
### EDX Analysis

The elements composition in the nest material of *Crematogaster rogenhoferi* was analyzed and found that it consist of different types of elements like oxygen, carbon, silicium, calcium, aluminum, potassium, copper, mercury and bromine were determined in the fragment of the nest wall with EDX analysis. Oxygen and Carbon were the major

elements. Silicium was higher than the other inorganic elements. Silicium was in the state of silicium oxide. Silicium oxide is found in the sand. The soil of the nest was sand. The other inorganic elements were mixed in the sand. K, Hg, Cu, Al and Br were in very low concentrations. The concentrations of elements are shown in Table 2. EDX spectra are shown in Fig. 5.

**Table 2:** Elements and their concentrations in a fragment according to EDX analysis.

Elements	Concentration (%)			
	Outer Surface		Inner Surface	
	1	2	1	2
Carbon (C)	42.17	45.87	50.57	49.26
Oxygen (O)	47.06	49.40	47.30	47.32
Aluminium (Al)	0.74	0.68	0.41	0.37
Silicon (Si)	2.51	0.58	0.64	0.64
Calcium (Ca)	1.17	1.47	0.58	0.65
Potassium (K)	0.45	0.58	0.50	0.57
Copper (Cu)	0.30	0.37	1.20	1.19
Mercury (Hg)	0.34	0.32	0.35	0.34
Bromine (Br)	0.65	0.73	0.74	0.76

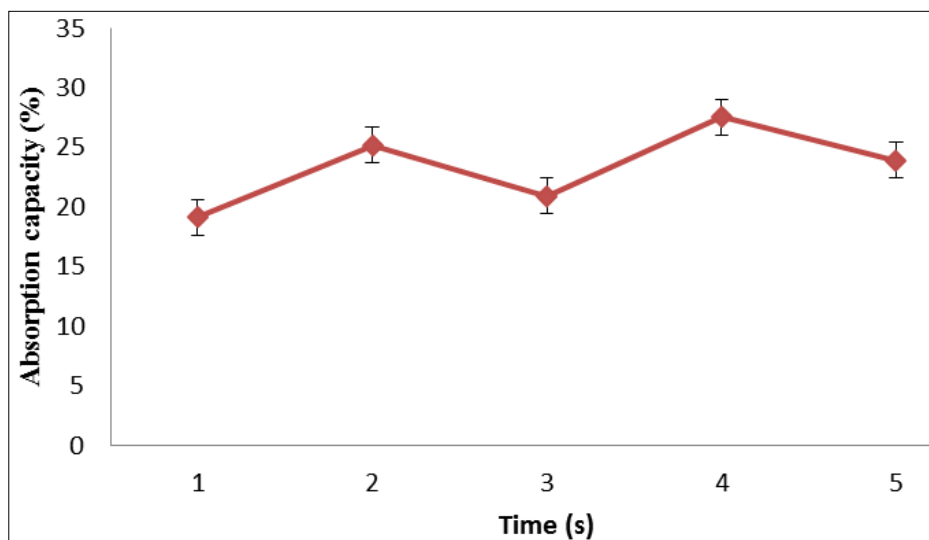


**Fig 5:** (A-D) EDX spectra of elements embedded in the wall of the nest of *Crematogaster sp.*

**Absorption capacity of the nest material**

The water absorption capacity of the nest was calculated in different fragments (n=5). The absorption capacity of the nest

fragment of ants, expressed as percentage, was estimated by using the methods of Curtis *et al.*, 2005. The average absorption capacities of the nest material of were  $22.92 \pm 9.11$ .



**Fig 6:** Figure showing the absorpency capacity of the nest material of *Crematogaster ant.*

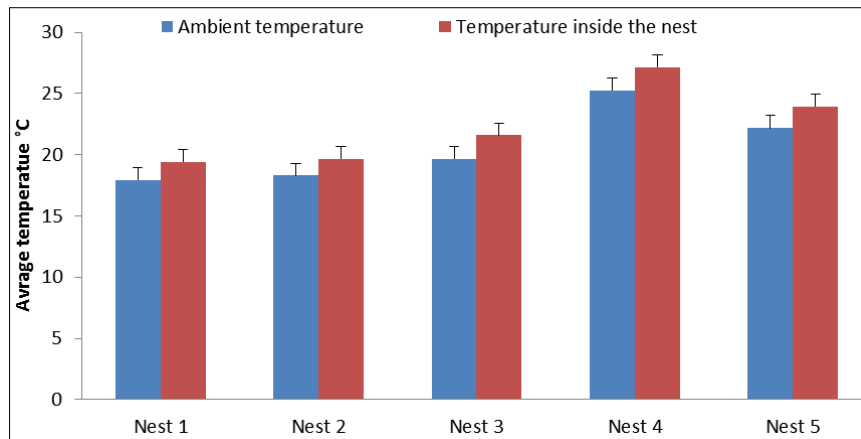
**Temperature inside the Nest of *Crematogaster rogenhoferi*.**  
The temperature of the nest material of *Crematogaster rogenhoferi* was measured and recorded (n=5). The

thermoregulations of the nest material of *Crematogaster* ant are given in the Table 3.

**Table 3:** Differences in ambient and internal temperature ranges of *Crematogaster* dives nests (season)

Seasons	n	Ambient Mean( $\pm$ SE)	n	Internal Mean( $\pm$ SE)	F	P
Spring	10	19.78 $\pm$ 0.78	10	18.75 $\pm$ 3.08	21.8234	0.000101
Summer	10	21.34 $\pm$ 1.19	10	18.83 $\pm$ 3.54	14.6123	0.000608
Autumn	10	17.93 $\pm$ 1.69	10	17.55 $\pm$ 2.38	35.7595	0.000015
Winter	10	13.39 $\pm$ 0.19	10	17.87 $\pm$ 3.58	30.2756	0.000013

The result is significant at  $p < .05$ .



**Fig 7:** Figure showing the temperature inside the nest of *Crematogaster* ant against ambient temperature.

## Conclusions

### Physical structure of the nest material

The ant nests form a shelter that protects the ants from their enemies and the environment around them, including the weather. They also provide a suitable place for the ants to safely rear their brood and to keep their queens protected. Without their nests most ant colonies would die out very quickly. The spheroidal in shape of the nest plays a vital role for resistance rainfalls, winds and cooled. Therefore, this fact suggests that morphology of the nests must have evolved in response to stimuli of the particular places and areas. Thus the spherical shaped and downward slopping of the nest prevents the rainfall from entering inside the nest. The chamber walls are also thoroughly waterproof [27]. The morphological structure of the nest protects their queens, eggs, larvae during heavy rainfall during monsoon and chilled during winter seasons. Insects constructing nests that provide a structure for breeding larvae are termed calichnia [28].

The caste numbers increase with the ant nest width. Workers are the main population numbers in the ant colony as they play such active roles in the ant colony [29]. Therefore, it is necessary for the ant population to enlarge their nest size when the population number of ants in the colony increases to provide more space for rearing the new generation. Holldobler and Wilson (1990) said that as the population of ants' increases, the ant nest size becomes larger.

The element composition in the nest materials of *Crematogaster rogenhoferi* from Meghalaya were composed mainly of Carbon and O with traces of Si, Ca, K, Al, Cu, Br and Hg according to the EDX spectra. In this study, the concentrations of Carbon and Oxygen occupied the highest concentration as compared to the other elements. This is because the nest was made from the plant debris or leaf litter. The inorganic elements like Silicon, aluminum, potassium, copper, mercury and bromine shows the least concentration while calcium was quite high.

The absorption property of the nest material was much slowed and the permeability of the nest membrane was less. Thus it shows that the nest materials play a vital role in water repelling such that it prevent the nest from humid, damp and moisten. During raining seasons the nest become wet only in the periphery part or the external part of the nest while the inner surface it was dry. Nest material strength is highly dependent on its moisture content. There is a relationship between the water absorbency and low moisture content of the nest. The processing of pulp affects the sticking ability of the fibers, the absorbency of nest paper and its durability [30]. Thus, the water absorption capacity of *C. rogenhoferi* nest was considerably high because the plant fibers.

The temperatures were measured with the help of thermometer. The thermometer was inserting inside the nest of ant and the readings was made in different replicates of different nest and the parameter was recorded. Ants regulate the temperature of their brood by carrying it to those parts of the nest with suitable temperatures. Temperature was varied from nest to nest and it depends on the size of the nest. The optimum temperature is  $\pm 200$  C to  $\pm 260$  C. This particular temperature is enhancing the insect development. Nest structure in ants is often designed to optimize the colonies ability to thermoregulation, and this specialization is most highly developed in nest building ant species.

The results of the study showed that there are variations in respect to physical features of the nest materials of *Crematogaster rogenhoferi* in various environmental conditions.

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