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A. Entomology

Leaf-litter insects at Amani Nature Reserve, Tanzania: A comparative analysis

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### **ABSTRACT**

Within a forest, the spatial variation in leaf litter composition is expected to affect both the density and diversity of leaf litter insects. In this study, a comparison of insect community structure in the leaf litter has been conducted between three habitats at Amani Nature Reserve: Bamboo shrubs (Bambuseae), gum trees (Eucalyptus sp.) and camphor trees (Cinnamomum camphora). A total number of 1919 individuals were collected from pitfall traps and 658 species from quadrate plots. Collected specimens were found to represent 8 different orders: Blattodea, Coleoptera, Collembolan, Diptera, Hemiptera, Hymenopter, Orthoptera and Thysanura. Diptera had the highest abundance in the overall collection. Statistical analysis did not detect a significant difference in insect composition between the three leaf litter habitats. Likewise, the depth of litter material had no significant influence on insect composition. Furthermore, no significant difference in insect composition was detected between day and night collections. Overall, these results suggest that the community of leaf litter insects at Amani Nature Reserve is characterized by considerable stability against spatial and temporal environmental fluctuations.

**Keywords**: Biodiversity; Tropical rain forest; leaf litter insects; Bambuseae; Eucalyptus; *Cinnamomum* camphora

## **INTRODUCTION**

Leaf litter is defined as the fallen vegetative debris, comprising a mixture of leaves, twigs, wood, fruits and flowers at various states of decay (Fittkau & Klinge, 1973). A wide variety of invertebrates are known to inhabit the leaf litter habitat, including insects, mites, arachnids, nematodes, isopods and centipedes (Anderson, 1975). Leaf litter invertebrates are important component of forest ecosystems, since they contribute to the recycling of nutrients through the decomposition of litter material, together with other soil organisms such as fungi and bacteria. Thus, leaf litter invertebrates help maintaining soil fertility and ecosystem stability (Abbott, 1982). Moreover, leaf litter communities could serve as bioindicators of environmental change and thus they are useful for conservation activities (e.g. Nakamura et al., 2003).

The distribution and the community structure of leaf litter insects are shaped by multiple environmental factors, including the leaf litter pH and moisture content, litter biomass and litter fall rate (Fried, 1990; Broughouts, 1992; Clouse, 1999). An important habitat factor that influences insect communities at the leaf litter is the species composition of the plant material; different insect species may favor different types of plant material. Moreover, the litter of certain plant species may be more attractive to leaf litter insects in general than the litter of other plant species and thus would contain higher insect densities. For example, some insect species were found to exhibit palate preference for plant material containing higher nitrogen levels. A change in the dominant plant species can result in a major shift in leaf litter composition and, consequently, insect composition at the habitat (Anderson, 1975; Olson, 1994; Koivula *et al.*, 1999).

Amani Nature Reserve is a tropical rain forest, located at the East Usambara Mountains in Tanzania; a region characterized by rich plant diversity, with approximately 3450 species of vascular plants; a quarter of them are endemic. Moreover, there are 74 endemic vertebrates in the region, including birds, mammals and amphibians. The insect diversity at the forest surface is extensive containing diverse assemblages of butterflies, moths, ants, termites, crickets and grasshoppers. Tree-associated insects at the forest include a variety of leaf beetles, weevils as well as bugs. The leaf litter insect community at the forest is dominated by beetles, including several species of rove beetles, dried fruit beetles, ground beetles and few dung beetles. Invertebrate fauna at East Usambara region shows high rates of endemism, especially among those with poor dispersal ability such as millipedes (T.B.A., 2010).

In this paper, we present a biodiversity survey for the poorly-known leaf litter insect fauna at Amani forest. We have adopted a comparative rather than a descriptive approach in the analysis of data. Insect composition has been compared between three types of leaf litter; those of Bamboo shrubs (Bambuseae), Gum trees (Eucalyptus) and the Japanese Camphor trees (*Cinnamomum camphora*). Moreover, the effect of the time and the technique of collection on the obtained estimations of insect composition were assessed statistically. The goal was to investigate the potential spatial variations and temporal changes in the diversity and abundance of leaf litter insects at Amani forest.

## **MATERIALS AND METHODS**

## The study site

The study was conducted at Amani Nature Reserve within the region of East Usambara Mountains (5°14′10′′-5°04′30′′ S, 38°30′34′′-38°40′06′′ E). It is a tropical rain forest which covers 8360 Hectares and is approximately 190-1130 m above sea level. There are two rainy seasons; the long season extends between March-May while the short season takes place during the period of November-December. The mean annual rain fall is 1910 mm per annum. Temperature ranges between 16.3°C-24.1°C (T.B.A., 2010).

## **Collection of samples**

Sample collection was conducted during six consecutive days (12th -17th August 2011). Three different monoculture plant habitats (Bamboo shrubs, camphor trees and Gum trees) were selected as sampling locations. The Bamboo Shrubs location lies about 1km away from the Amani Headquarter along Monga Village while the camphor trees site lies about 3km toward Mbomole hill and the gum trees site is about 1.5 km away from Amani Headquarter in the route to Tonga location.

Two methods were applied to collect leaf litter insects: Trap transects and litter quadrates. In the first method, two transects, 50m length each, were set up in the three habitat types. Five pitfall traps were set at about 5m from the transect line and the

traps were 10m apart from each other. Each trap has a length of 7.5cm and a height of 6.0cm. The interval between the two transects in each habitat was 50m. All the pitfall traps were filled with soapy water. Monitoring of transects was conducted twice a day, at morning and evening, during the survey period. Insect samples were preserved in plastic bottles containing 70% ethanol. In the second method (i.e. litter quadrates), a quadrate of 1m² was chosen for insect sampling at each habitat. Each quadrate was located within the interval of the two transects at the habitat, at least 5 meters apart from each transect. Leaf litter material was collected from the quadrates once daily.

## Laboratory analysis

Berlese funnels were used to extract insects from the leaf litter samples collected through the quadrate method. Separate funnels were used for the leaf litter of each habitat. Insect samples were examined in the laboratory using a dissecting microscope and a compound microscope (Vickers®: resolution 30x, eye piece 10x). Using a field guide (Picker *et al.*, 2004), insects were identified to the order level and, in the case of Diptera, to the family level. Samples were then persevered in plastic bottles containing 70% ethanol.

## Statistical analysis

Statistical data analysis was performed using Excel® and Minitab® software packages. The one—way ANOVA test was applied to compare the insect abundance between the three habitats, after the statistical distribution of individuals was proven normal. Non parametric tests, that are Mann Whitney and Kruskal Wallis tests were used when data is not normally distributed, as in the other comparisons performed in this study. Graphical representation of data was applied whenever necessary.

### RESULTS

## A comparison of insect composition between different leaf litter habitats

In this survey, eight orders of insects were identified. These orders are Blattodea, Coleoptera, Collembolan, Diptera, Hemiptera, Hymenopter, Orthoptera and Thysanura. The abundance of insect orders at different habitats is summarized in (Table 1). In addition, the distribution of Dipteran families between habitats is shown in (Table 2) and (Fig.1). Results showed that most insects belong to the order Diptera which had 13 families (560 individuals). The second order in abundance was Collembola (321 individuals) followed by Hymenoptera (255 individuals). The statistical analysis of insect abundance did not detect significant habitat preference (DF=2, F=5.78, P>0.003).

| Table 1: The number | s of insects fro | m different orders | s collected at each habitat |
|---------------------|------------------|--------------------|-----------------------------|
|                     |                  |                    |                             |

| Order       | Bamboo shrubs | Gum trees | Camphor trees | Total |
|-------------|---------------|-----------|---------------|-------|
| Blattodea   | 1             | 0         | 0             | 1     |
| Coleoptera  | 37            | 7         | 18            | 62    |
| Collembola  | 180           | 83        | 58            | 321   |
| Diptera     | 262           | 178       | 120           | 560   |
| Hemiptera   | 2             | 4         | 2             | 8     |
| Hymenoptera | 155           | 49        | 51            | 255   |
| Orthoptera  | 14            | 26        | 12            | 52    |
| Thysanura   | 1             | 0         | 1             | 2     |
| Grand total | 652           | 347       | 262           | 1261  |

| Family         |        | Total |         |     |
|----------------|--------|-------|---------|-----|
|                | Bamboo | Gum   | Camphor |     |
| Mycetophilidae | 37     | 21    | 13      | 71  |
| Diposidae      | 27     | 0     | 0       | 27  |
| Tephritidae    | 42     | 5     | 22      | 69  |
| Drosophiliidae | 7      | 13    | 6       | 26  |
| Calliphoridae  | 10     | 3     | 2       | 15  |
| Nerriidae      | 1      | 0     | 0       | 1   |
| Sepsidae       | 3      | 0     | 2       | 5   |
| Bombyliidae    | 6      | 9     | 5       | 20  |
| Sciaridae      | 28     | 25    | 23      | 76  |
| Dolichopodidae | 3      | 0     | 0       | 3   |
| Muscidae       | 18     | 12    | 2       | 32  |
| Chironomidae   | 10     | 12    | 1       | 23  |
| Ceratopgondae  | 28     | 22    | 12      | 62  |
| Un known       | 42     | 56    | 32      | 130 |
| Total          | 262    | 178   | 120     | 560 |

Table 2: The numbers of insects from the different families of Diptera collected at each habitat

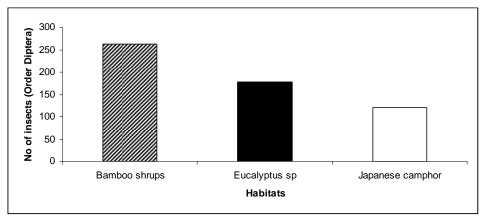


Fig. 1: Abundance of the order Diptera at different habitats

### A comparison of insect composition between different leaf litter depths

The average litter depth was higher at the eucalyptus site (3.08 cm) than at the Bamboo site (2.19cm) and the least depth was recorded at the camphor site (1.58cm). However, results showed no clear pattern of relationship between the leaf litter depth and insect abundance.

## A comparison of insect composition between different sampling methods

The two sampling methods applied during this study that are Trap transects and litter quadrates have yielded different estimations of leaf litter insect composition. Results from the transect method indicates no difference in insect abundance between habitats (Fig. 2). On the contrary, the quadrate method shows higher abundance at the Bamboo site (H = 1.66, DF = 2, P =0.436) compared to the Gum and the camphor sites (Fig. 3). When the two methods were compared for the overall insect abundance at the forest (Fig. 4), the analysis revealed that there is a significant difference in the abundance of individuals based on the sampling method, being higher in the quadrate method than in the transect method (P=0.0329, W = 135022).

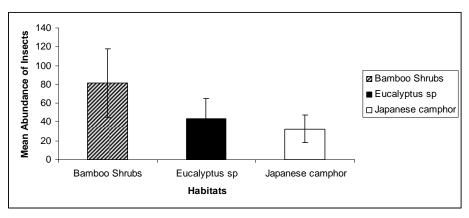


Fig. 2: Abundance of leaf litter insects at different habitats by using pitfall traps

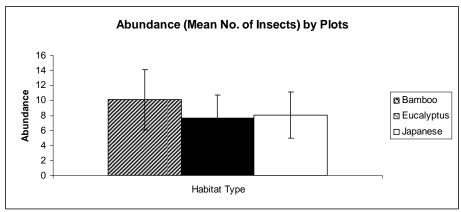


Fig. 3: Abundance of leaf litter insects at different habitats by using quadrate plots

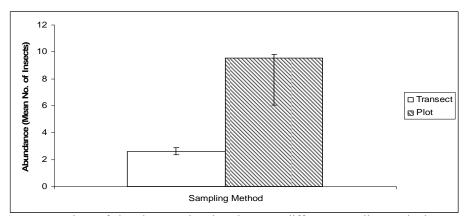


Fig. 4: A comparison of abundance estimations between different sampling methods

## A comparison of insect composition between day and night collections

Leaf litter insects showed no variation in abundance between different collection times (nocturnal: H= 3.14, DF= 2, P=0.208; diurnal: H=0.27, DF=2, P=0.8872), though insects were slightly more active during the night, as indicated by mean densities (Fig. 5). Unlike the other habitats, the number of insects collected during the night at the Bamboo shrubs was more than twice the number collected at the day (Fig. 6).

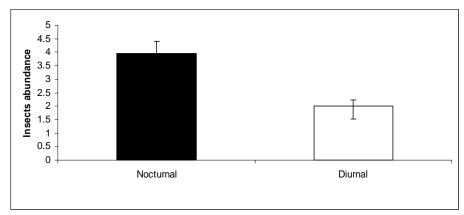


Fig. 5: A comparison of total abundance between the day and the night

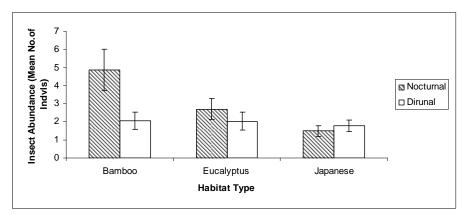


Fig. 6: A comparison of insect abundance between the day and the night at different habitats

### **DISCUSSION**

During this study, the biodiversity of leaf litter insects at Amani Nature Reserve was subjected to preliminary investigation. Overall, a diverse assemblage of insect species was found, with eight different orders of insects being indentified. The order Diptera, in particular, seems to be the most abundant group in the leaf litter community, since it shows both high abundance (44% of the total collection) and diversity (represented by 13 different families). The orders Collembola and Hymenoptera were also considerably abundant (25% and 20% of the total collection, respectively). Interestingly, the order Coleoptera represented only 5% of the total collection, contrary to earlier observational data suggesting that beetles dominate the leaf litter insect fauna at Amani forest (T.B.A., 2010).

Theoretically speaking, the insect community structure at the leaf litter is expected to vary with the habitat type (Hansen, 2000); the reason is that every species have special habitat requirements, thus, individuals from different species would show behavioural preferences toward different habitats. Moreover, the habitat type deeply affects the output of the inter-species competition and thus certain species will tend to dominate the community wherever the habitat requirements that favor them are met. The present survey was designed to investigate the potential influence of the habitat on the leaf litter insect community at Amani forest. A comparative analysis of insect composition has been carried out between three monoculture localities at the forest: Bamboo shrubs, camphor trees and Gum trees. Two habitat factors vary between

these localities: the plant type and the depth of the litter material. The results of the study have shown that insect composition did not differ significantly between the three localities, which suggest that, contrary to expectations, both plant type and litter depth do not affect the diversity and abundance of leaf litter insects at Amani forest.

Despite the absence of general, statistical pattern of habitat preference, it is nevertheless true that certain insect taxa were more abundant (in terms of absolute numbers only) at specific habitats. For example, Dipteran specimens were considerably more abundant at the Bamboo shrubs (262 at the Bamboo location compared to 178 and 120 at the Gum location and the Camphor location, respectively). Furthermore, this pattern was also evident for individual Dipteran families; 10 out of the 13 families showed higher abundance at the Bamboo shrubs. Such habitat preference could result from different factors. For example, the Bamboo site is damp and shady and adult dipterans are naturally attracted to these environmental conditions. Dipteran larvae depend mainly on the leaf litter for feeding (Paradise, 2004), thus, it is possible that the Bamboo site had additional underground vegetation such as berries that attracts larvae. Moreover, the Bamboo site was at the edge of the forest, close to a cultivated area; such environmentally heterogeneous zone might underlay the higher Dipteran diversity observed there.

Most insect species show temporal variation in daily activity (e. g. Diurnal, nocturnal) (Beck, 1968). In order to detect potential temporal fluctuations in the activity of leaf litter insects, a statistical comparison of insect composition has been performed between day and night collections. Once again, no statistically significant difference has emerged out of this analysis. Of course, this result does not imply that these insects have no temporal variation in activity but rather suggests that the leaf litter community at Amani is too diverse to show any distinct, unified pattern of daily activity. This point can be made clearer if we imagine that half of the individuals in the collection are diurnal and the other half is nocturnal. If this is the case, the group as a whole would show no activity pattern at all although every one of its members has its own activity pattern. The only way by which the entire community can show significant temporal variation is if most of its members happen to follow the same direction of daily activity. This condition is unlikely to be met in the studied community since it is composed of taxonomically unrelated species with variable life styles.

The sampling technique is a well-known source of bias in field entomology (e.g. Zink & Rosenheim, 2004). In many cases, highly significant differences between the estimations of independent field studies may result not because of any inherent spatial or temporal variability in the wild community but, rather, due to subtle differences in their collection methodology. In order to avoid potential collection bias in the current study, our estimations of leaf litter insect composition were based on two independent collection techniques: Trap transects and litter quadrates. Furthermore, the collections obtained from each method were isolated and insect composition was independently estimated from each collection. The estimations of the two methods were compared statistically to see whether our caution was justified, that is, whether estimations based on one collection method is different from estimations based on a combination of methods? True enough, the analysis yielded two significant differences that would have biased our estimations of the insect community structure if we adopted one sampling method: first, the quadrate collection estimated higher insect abundance at the Bamboo shrubs although no such pattern was evident from the total collection. Moreover, the overall insect abundance (across all habitats) was higher in the quadrate collection than in the transect collection.

The main conclusion of the present study is that the leaf litter insect community at Amani forest appears highly stable over space and time; no significant spatial or temporal fluctuations in abundance or diversity were detected. However, we think that such conclusion should be interpreted cautiously. There are three reasons for caution: first, the sample size was too small; second, the spatial and temporal scales of the study were too narrow; the third and the most important reason is that the environmental factors that are considered in the study design (i.e. the plant type, the litter depth and the daily variation) are only a tiny fraction of the overall factors that shape the leaf litter ecosystem at Amani forest. Tropical rain forests are ecosystems that are characterized by extreme levels of environmental complexity (Terborgh, 1992). Thus, it is likely that a plethora of environmental factors act harmoniously on the insect fauna at the leaf litter. Due to this complexity, a great deal of variation may be hidden from small-scale surveys that are based on one or few environmental parameters; if more parameters were surveyed, substantial faunal variation might have been unraveled. For example, if the full range of vegetation diversity were considered, including other trees and shrubs as well as the habitats of mixed plant species, it is likely that considerable variability in insect fauna would be found. In addition, the effect of other insect communities, such as those of vegetation-associated insects, might have a great influence on the leaf litter insect community structure. Comparing the insect community between Amani forest and the surrounding, unprotected area that is affected by human activities is likely to yield even more faunal variability. Although no significant variation was detected throughout the day period, extensive temporal variations may occur between successive seasons or years. We strongly recommend future research to investigate these possibilities and we predict that surveys with larger scale and better design will detect considerable spatial and temporal changes in the leaf litter insect fauna at Amani forest.

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#### ARABIC SUMMARY

حشرات المخلفات النباتيه في محمية أماني الطبيعية \_ تنزانيا: تحليل مقارن

اسراء الفكي  $^1$  - فرانسيسكا ماليمبيكا  $^2$  - أليكس تامبا جونسون  $^3$  - شومبي حسن  $^2$  - ايهاب ادريس  $^1$  - جامعة الخرطوم  $^2$  - جامعة سوكوين الزراعيه - تنزانيا  $^3$  - جمعية حياة الطيور العالميه - ليبيريا  $^3$  - جمعية حياة الطيور العالمية - ليبيريا  $^3$  - فسم علم الحيوان - ص. ب 321 الرمز البريدي 11115 الخرطوم - السودان - كلية العلوم

بداخل نطاق الغابه فانه من المتوقع أن يؤثر التباين المكاني في تركيبة البقايا النباتيه علي كثافة و تنوع الحشرات التي تقطن في هذه المواطن. في الدراسه الحاليه تمت مقارنة مجتمعات حشرات البقايا النباتيه بين ثلاثة مواطن في محمية أماني الطبيعيه بتنزانيا: شجيرات البامبو و الأشجار الصمغيه و أشجار الكافور. تم جمع عدد 1919 عينه باستخدام شراك الحشرات و عبر جمع البقايا النباتيه. وجد أن الحشرات التي تم جمعها تنتمي الي 8 رتب مختلفه مثلت بينها ثنائيات الأجنحه الرتبه الأكثر وفرة و تنوعا. لم يجد التحليل الاحصائي فرقا معنويا في تركيبة مجتمع الحشرات بين المواطن الثلاثه مما يدل أن نوع النبات و عمق البقايا النباتيه لا يؤثر بشكل ملحوظ علي وفرة و تنوع الحشرات الجمع التي تمت علي وفرة و تنوع الحشرات و بالاضافه الي ذلك لم يتم رصد فرق معنوي بين نتاج عمليات الجمع التي تميز أثناء النهار و الليل بشكل عام فان نتائج الدراسه تقترح أن مجتمع حشرات البقايا النباتيه في محمية أماني يتميز بثبات ملحوظ في وجه التغيرات المكانيه و الزمانيه في بيئة الموطن.