

FIRST CHECKLIST, SPECIES RICHNESS AND DIVERSITY OF LEAF-LITTER DWELLING ANTS (HYMENOPTERA: FORMICIDAE) IN ANCIENT BENIN MOAT, NIGERIA

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ABSTRACT

Encircling Benin City, in Nigeria, lies an enormous excavation commonly known as a moat or 'Iya' by the people of Benin or the Great Benin Kingdom, which dates back several centuries and holds diverse, but poorly known faunal and floral species. To catalogue its leaf-litter ant species, 24 10-m² quadrats randomly sited within the moat were sampled with a leaf-litter sampler (sifter) and the collected ants were euthanised in properly labelled enclosed jars containing a ball of cotton charged with ethyl acetate. Results revealed eight species of ants distributed among eight genera and four subfamilies (Dolichoderinae, Formicinae, Myrmicinae and Ponerinae). Pheidole megacephala was the most abundant species followed by Odontomachus troglodytes. Diversity indices revealed a heterogeneous community of ants at microhabitat levels notably dominated by P. megacephala, while five species were rare. The study predicts an additional ant collection of 27 to 45 % to realise the complete inventory of litter ants in the moat, which typifies an isolated green space capable of retaining several species despite pressure from urbanisation.

Keywords: Ants inventory, Distribution, Abundance, Historic sites, Benin kingdom

INTRODUCTION

Urbanisation is one of the key threats continually eroding biodiversity, worldwide. In sub-Saharan Africa, species around urban areas are directly affected by urbanisation; however, one of Nigeria's unaltered urban spaces endowed with an ancient regrowth of terrestrial habitat cocooned within it is Benin moat. It is known by the indigenous people of Benin Kingdom, partly present-day Benin City, as 'Iya' (Kingdom of Benin, 2022). The moat was dug between 800 AD and 1460 AD stretching several kilometres and several metres deep (Darling and Agbontaen-Eghafona, 2014; Onwuanyi *et al.*, 2021; Kingdom of Benin, 2022). Benin kingdom, hitherto ruled by successive Kings –

addressed as 'Oba'– built and managed the moat in its heyday for defence, but is now appallingly metamorphosing into a receptacle for runoffs and municipal waste (Akujieze and Irabor, 2014; Onwuanyi *et al.*, 2021).

Presumably, the moat houses diverse species isolated for centuries - a yet-to-be-quantified ecological value. Although Onwuanyi *et al.* (2021) earlier interrogated the moat's value to infer that it is a non-functional urban void; naturalists cannot discount its ecological role of sheltering species (e.g., flora, flightless crawling fauna, etc.), whose founding populations once dominated the surrounding habitats now occupied by the city. Such species remain undocumented to date, and not even the most distributed animals – ants are

documented. For the invaluable ecological roles as a species sanctuary, it would be intriguing to know the resident species of Benin moat, particularly the ants. Given the increasing anthropogenic pressures on the moat arising from its atrocious misuse as either a waste receptacle or a flood-control conduit, it is noteworthy to catalogue its resident species; for which ant composition and their richness caught our interest.

Few records of myrmecological surveys in Nigeria abound in literature (Taylor, 1976; 1977; 1978; 1979; 1980a,b; Taylor and Adedoyin, 1978; Ewuim *et al.*, 1997; Taylor *et al.*, 2018; Okpanachi and Yaro, 2019), but the knowledge of resident ants within the Benin moat - a peculiar urban-isolated relic of ancient culture - is poorly understood. Meanwhile, ants provide numerous ecosystem services in pollination, soil bioturbation, biomonitoring, seed dispersal, food and biomedical services (Hölldobler and Wilson, 1990; Crozier *et al.*, 2010; Dossey, 2010; Chaves-Campos, 2011; Del Toro *et al.*, 2012; Diamé *et al.*, 2018). Despite evidence of complex ant-plant interactions in our megadiverse rainforest, only a handful of ants e.g., *Tetraponera aethiops* Smith, F., 1877 (Hymenoptera: Formicidae) and *T. latifrons* Emery, 1912 (Hymenoptera: Formicidae) have thus far been reported in Benin, Edo State, Nigeria (Janzen, 1972; Ward, 2022). Given the age-long isolated vegetation, the Benin moat provides an avenue to study the ant species whose founding populations once dominated the surrounding area until urbanisation crept in. Thus, the survey here provides a checklist, species richness and diversity of ants exploring the leaf litter in the moat's forest floor with the hope of preserving the moat's ecosystem as a relic of natural history.

MATERIALS AND METHODS

Survey Site: The myrmecological survey was conducted on January 15 and 22, 2022 within an aspect of Benin moat with a geo-reference of N 06.33172, E 5.65002; 53.4 ± 6.6 m above sea level as obtained using a GARMIN 72H handheld device. The sites were close to Edo College situated at Third East Circular, Benin City. The

moat is a massive relic of the ancient Benin Kingdom and has enclosed evergreen vegetation within its valley but is surrounded by an intensively urbanised human settlement as illustrated in Figure 1.

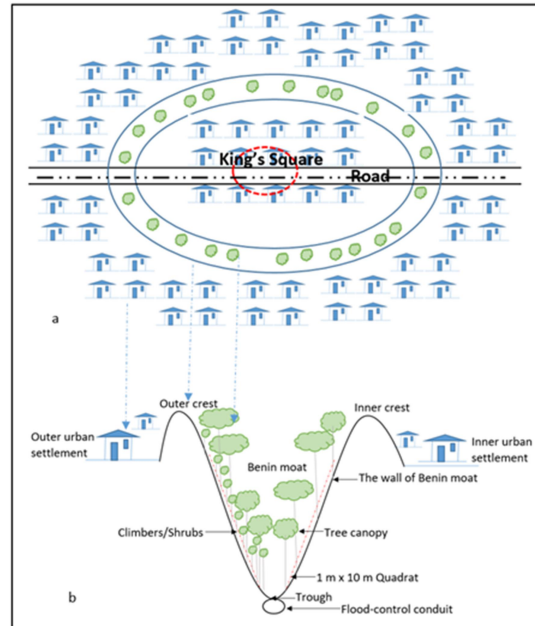


Figure 1(a and b): Schematic representation of Benin moat and its adjoining urbanised inner and outer area delineated by the two crests which form two concentric rings around the city as seen in the topmost schema, while b) represents its cross-sectional view and the vegetation. Note: the red central circle in 1a) represents the King's square – the seat of power in the kingdom; the middle horizontal bar signifies the roads that cut across several parts of the moat linking up human settlements on both sides, and at such points, the moat is divided into sections (left and right of the roads) as the moat was landfilled from the trough to the top by levelling off the crest at those spots to pave way for vehicular traffic

The moat was accessed through its lower end at Ogiso community near Shaka Polytechnic. The entire region of southern Nigeria, where the moat is situated, falls within the tropical rainforest belt characterised by two seasons – the wet and dry seasons. While the latter starts in November and ends in February, the former starts in March and ends in October. The survey was conducted towards the end of the dry spell that started in 2021 to ebb out in 2022.

Hitherto, the moat's vegetation is part of the tropical rainforest belt. The trough at the lower end of the moat (near Ikpoba River

bank) is used by locals for some small-scale farming - notably plantain and maize farming. Deep into the moat lies aspects that have been deliberately inundated with municipal waste from the moat's crest through its wall to the trough, while the majority of the interiors where our samplings were conducted have steep slopes on both sides of the moat wall comprising flora with variable heights and girths. The plants are randomly scattered along its crests, walls and trough, and with a largely continuous canopy with allowance for incident light rays. The forest floor may enjoy an all-year-round litter fall; however, the litter density on the wall is poor, perhaps caused by the downward gravitational pull down the steep wall. Aspects of the moat's flora revealed the presence of several trees, shrubs and herbs, whose origins are of different phytogeography which includes invasive alien plants such as *Chromolaena odorata* (Linn.) R. M. King and H. Robinson (Asterales: Asteraceae).

Sampling Techniques and Identification of Specimens:

The leaf litter within a quadrat of 1 x 10 m² was sampled on either side of the moat to constitute a sample using a leaf-litter sampler equipped with a sifter with a mesh size of 0.5 x 0.5 cm². The leaf litter sampling method facilitates the collection of leaf litter from the forest floor and through its mesh; small debris and leaf-litter dwelling organisms fall into the debris-holding section of the sampler. Upon completely sieving the leaf litter within each quadrat, the debris-holding section was emptied into separate but properly labelled bags. The bags (of dimension 40 x 30 cm²) were made from satin affixed with strings to seal off the bags once the samples were released into them for onward extraction of ants in the laboratory using an aspirator. Collected ants per sample were euthanised with ethyl acetate. Thereafter, the lifeless ants were sorted into morphospecies (a group of similar forms) and counted with the ensuing data, tabulated.

Samplings were conducted between 8:00 am and 4.00 pm on sampling days. Collected specimens were prepared for

mounting in an entomology laboratory situated in the Department of Animal and Environmental Biology at the University of Benin, Nigeria. With proper labels containing their locations, collection method, species and collector's names, the specimens were mounted in an entomological box and stored in an insect cabinet within the laboratory and duplicate specimens will be deposited in our newly commissioned, yet to be operational, museum of natural history for proper curation. The specimens' identities were determined using an AmScope Stereomicroscope at magnification X40 using relevant identification keys (Bolton, 1973; Hölldobler and Wilson, 1990; Borowiec and Salata, 2018) and pictorial keys available at AntWeb (2022) for comparing their diagnostic characters.

Statistical Analyses: Tabulated taxa counts were summarised, while the diversity indices of the samples and taxa rarefaction curve were computed using PAST software version 4.03. For the estimation of the efficiencies of the non-parametric species, Chao 2 and second-order Jackknife (Jackknife 2) that rely on presence and absence data were used (Smith and van Belle, 1984; Chao, 1987; Colwell and Coddington, 1994; Hammer and Harper, 2006), to compute the probable number of species in the study area. The models' parameters: S_{obs} represents the absolute number of observed species; L represents the species number in precisely a sample, and M represents those in precisely two samples, while n represents the sample size. The model for Chao 2 = $S_{obs} + L^2/2M$, and for Jackknife 2 = $S_{obs} + L(2n - 3)/n - M(n - 2)^2/(n^2 - n)$ (Hammer and Harper, 2006). The diversity indices were taxa count represented as S ; Shannon-Wiener H' index, represented as $H' = -\sum p_i \ln p_i$ (where \ln stands for natural logarithm). H' depends on relative abundance and taxa count, and it is lowest for a single taxon (i.e., $H' = 0$) and the highest, $H_{max} = \ln S$ (Hammer and Harper, 2006). Evenness varies from 0 to 1 as obtained from H'/H_{max} ; Dominance is the notional inverse of Evenness (Hammer and Harper, 2006).

RESULTS

A total of 359 individual ants distributed among eight genera and eight species, belonging to four sub-families (Dolichoderinae, Formicidae, Myrmicinae and Ponerinae) were found in the forest litter of Benin moat within a total area of 24 x 240 m² area. Though eight distinct species were obtained, the study could not identify one to the species level. The composition of the observed ants (Hymenoptera: Formicidae) includes *Dolichoderus* species Lund 1831, *Camponotus acvapimensis* Mayr 1862, *Paratrechina longicornis* Latreille 1802, and *Pheidole megacephala* Fabricius 1793. Others were *Brachyponera sennaarensis* Mayr 1862, *Mesoponera ambigua* André 1890, *Odontomachus troglodytes* Santschi 1914 and *Palthothyreus tarsatus* Fabricius 1798 (Table 1). Among the four subfamilies, the least speciose were Dolichoderinae (*Dolichoderus* sp.) and Myrmicinae (*P. megacephala*), each representing 12.5 % of the observed species, while Ponerinae was the most speciose (50 % representation, i.e., four species) (Table 2).

The species' relative abundances were heterogeneous with a remarkable dominance by the myrmicine ant, *P. megacephala*, which constituted 82 % (i.e., 293 individuals), followed by the ponerine ants, *O. troglodytes* 12.5 % (45 individuals) and *B. sennaarensis* 2.8 % (10 individuals). Coincidentally, in a similar manner, these ants were also the three most encountered ants in the samples, where they occurred 22 times (91.7 %), 15 times (62.5 %) and 10 times (41.7 %), respectively, out of the entire 24 samples (Table 2). Apart from *P. longicornis* which occurred only in two samples, others (*Dolichoderus* sp., *C. acvapimensis*, *M. ambigua* and *P. tarsatus*) occurred only once. While *Dolichoderus* sp. and *P. tarsatus* occurred as singletons, *M. ambigua* occurred as a doubleton - with only two individuals. Aside from some evidence of species rarity per sample, others e.g., *O. troglodytes* had as high as six individuals and as low as an individual (or none at all) in some samples. Also, *P. megacephala* had a range of 1 – 43

(minimum-maximum) individuals per sampled plot (Table 2).

The non-parametric species estimators (Chao 2 and Jackknife 2), estimated 10.9 and 14.6 species as opposed to the observed count (of eight species); suggesting that only about 55 and 73 % of the true ant species resident in the litter were seen in this study. The sample-based species rarefaction curve (Figure 2) also supported these estimates given that the curve had not reached its asymptote when sampling ended. Intuitively, it indicates that more species would have been found if sampling continued. The species count per sample averaged at 2.0 ± 0.8 with as low as one species per sample but as high as four (Table 3). The individual ants per sample had a median of 10.5 ± 12.3 , with as low as two individuals and as high as 44 individuals of ants per sample (Table 3).

From the four diversity indices viz.: Dominance (D), Evenness (E), and Shannon-Weiner (H) were reported in Table 3, species dominance had a median of 0.66 ± 0.20 with a range of 0.38 to 1.00. Complete dominance is reported when D equals 1. Shannon-Weiner H index ranged from 0 to 1.03 with a median of 0.56 ± 0.3 . Some sampled areas (10 m²) were less diverse as they had only a species while others were relatively diverse. The evenness indices ranged from 0.45 to 1.

DISCUSSION

In providing an inventory of the leaf-litter ants enclosed within the moat system in Benin (Nigeria), three dominant species of ants were encountered with a sampling technique that utilised active sampling (litter sifting method), and outcomes were compared with a passive one (pitfall trap) used elsewhere; as will be discussed shortly alongside the economic importance of ants to man. Here, our study of leaf litter ants occupying the forest floor of the Benin moat's atypically closed vegetation after 24 repeated samples, yielded a total of 359 individuals, belonging to eight genera, eight species, and four subfamilies in the family Formicidae.

Table 1: Composition and abundance of ant species collected per sample obtained from the forest of Benin moat using a leaf-litter sifter for sampling

| Observed Ants | Leaf-litter Samples | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | |
|---|---------------------|----|---|---|----|---|----|---|---|----|----|----|----|----|----|----|----|----|----|----|----|----|----|----|---|---|---|---|---|---|--|--|--|--|--|--|--|--|--|--|---|---|---|
| | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 | 15 | 16 | 17 | 18 | 19 | 20 | 21 | 22 | 23 | 24 | | | | | | | | | | | | | | | | | | | |
| <i>Dolichoderus</i> species Lund, 1831 | 1 | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| <i>Camponotus acvapimensis</i> Mayr, 1862 | 4 | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| <i>Paratrechina longicornis</i> Latreille, 1802 | 1 | | | | | | | | | | | | | | | | | | | | | | 2 | | | | | | | | | | | | | | | | | | | | |
| <i>Pheidole megacephala</i> Fabricius, 1793 | 11 | 17 | 3 | 6 | 11 | 3 | 43 | 8 | 5 | 15 | 11 | 2 | 2 | | 3 | 19 | | 6 | 7 | 34 | 1 | 34 | 14 | 38 | | | | | | | | | | | | | | | | | | | |
| <i>Brachyponera sennaarensis</i> Mayr, 1862 | 1 | | | | | | | | | | | | | | | | | | | | | | 1 | | 1 | 1 | 1 | 1 | 1 | | | | | | | | | | | | 1 | 1 | 1 |
| <i>Mesoponera ambigua</i> André, 1890 | 2 | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | |
| <i>Odontomachus troglodytes</i> Santschi, 1914 | 4 | 6 | 4 | 1 | 3 | 5 | 1 | | | | | | | | | | | | 5 | | 3 | 4 | 1 | 2 | 2 | | | | 2 | 2 | | | | | | | | | | | | | |
| <i>Palthothyreus tarsatus</i> Fabricius, 1798 | 1 | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | |

Each sample was collected from a specific area with dimensions 1 x 10 m² and replicated 24 times

Table 2: Relative abundance and frequency of occurrence of leaf-litter ants collected in the Benin moat [samples, n = 24].

| S/N | Reference Code* | Subfamily | Ant species | Counts | | |
|-----|-----------------|---------------------------|---|------------------------------|-----------------|---------------------------------------|
| | | | | Total (% Relative Abundance) | Range (min-max) | Occurrence (% of total samples, n=24) |
| 1 | EgC0001 | Dolichoderinae (1, 12.5%) | <i>Dolichoderus</i> species Lund, 1831 | 1(0.28) | Singleton | 1 (4.17) |
| 2 | EgC0002 | Formicinae (2, 25%) | <i>Camponotus acvapimensis</i> Mayr, 1862 | 4(1.11) | 4 | 1 (4.17) |
| 3 | EgC0003 | Formicinae (2, 25%) | <i>Paratrechina longicornis</i> Latreille, 1802 | 3(0.83) | 1 to 2 | 2 (8.33) |
| 4 | EgC0004 | Myrmicinae (1, 12.5%) | <i>Pheidole megacephala</i> Fabricius, 1793 | 293(81.62) | 1 to 43 | 22 (91.67) |
| 5 | EgC0005 | Ponerinae (4, 50%) | <i>Brachyponera sennaarensis</i> Mayr, 1862 | 10(2.79) | 1 to 1 | 10 (41.67) |
| 6 | EgC0006 | Ponerinae (4, 50%) | <i>Mesoponera ambigua</i> André, 1890 | 2(0.56) | Doubleton | 1 (4.17) |
| 7 | EgC0007 | Ponerinae (4, 50%) | <i>Odontomachus troglodytes</i> Santschi, 1914 | 45(12.53) | 1 to 6 | 15 (62.50) |
| 8 | EgC0008 | Ponerinae (4, 50%) | <i>Palthothyreus tarsatus</i> Fabricius, 1798 | 1(0.28) | Singleton | 1 (4.17) |

*Personal collection codes and corresponding duplicate specimens are to be deposited at our museum of natural history, the 'Museum and Biodiversity Building' once it becomes operational

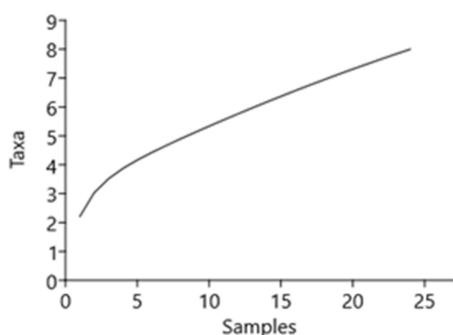


Figure 2: Sample-based rarefaction curve for ants sampled from the forest floor of Benin moat

Table 3: Central tendencies and range of species richness and diversity indices of ant species recovered from leaf litters in Benin moat within a ten-meter square area replicated 24 times

| Diversity indices | Mean \pm SD (Median) | Range (Minimum-Maximum) |
|-----------------------------|------------------------|-------------------------|
| Taxa, S | 2.21 \pm 0.8 (2.0) | 1 – 4 |
| Individuals | 15 \pm 12.3 (10.5) | 2 – 44 |
| Dominance, D | 0.72 \pm 0.2 (0.66) | 0.38 – 1.00 |
| Shannon-Weiner H | 0.46 \pm 0.3 (0.56) | 0.00 – 1.03 |
| Evenness, e ^H /S | 0.79 \pm 0.2 (0.86) | 0.45 – 1.00 |

This study does not only offers the preliminary checklist, diversity and species richness of leaf-litter ants that occupy Benin moat (an ancient man-made trench whose long-standing vegetation is surrounded and threatened by urbanisation), but it is also the first faunistic account of ants in Benin City, the metropolis where the moat is situated. The three most abundant species recorded in the leaf litter were *P. megacephala* (293, 81.6%), *O. troglodytes* (45, 12.5%) and *B. sennaarensis* (10, 2.8%), and these belonged to three genera and two subfamilies viz.: Myrmicinae and Ponerinae.

P. megacephala and *O. troglodytes* dominance were unsurprising, given the similar reports of studies elsewhere (Ewuim *et al.*, 1997; Ewuim and Osondu, 2008; Ryder Wilkie

et al., 2010). Our findings were akin to those of Ewuim *et al.* (1997), whose pitfall-trapped ants in forest and fallow sites at Ile-Ife, Nigeria, trapped a slightly higher count of ten ant species as opposed to the eight from leaf-litter sampling reported here. While *P. crassinoda*, *Camponotus maculatus* and *O. troglodytes* dominated the forest and fallow plots at Ile-Ife, *P. megacephala*, *O. troglodytes* and *B. sennaarensis* dominate the leaf litter in Benin moat, and the only *Camponotus* species found in this current study occurred in one sample. From both studies and another study by Ewuim and Osondu (2008), who also noted *Pheidole*'s dominance, it would suffice to say that *Pheidole* species are the key species among leaf-litter and/or ground-dwelling ants in Nigeria's tropical forest belt until otherwise refuted. The sole drivers of species abundance, distribution and impact are their environment-linked preferences. For instance, while the *Pheidole* species are dominant ground and subterranean dwellers as seen in western Amazonian forest, *Camponotus* and *Crematogaster* dominate tree canopy (Ewuim *et al.*, 1997; Ryder Wilkie *et al.*, 2010). The soil-mobilising proclivity of *P. megacephala* when building its nest on plant canopies and its affinity for homopterans' honeydew predispose the plants to diseases, as evident in the transmission of cocoa black-pod disease -a source of worry to cocoa farmers (Taylor *et al.*, 2018).

Although there are peculiarities in collection techniques, the outcomes of those used here and other related studies were comparable. For instance, while Ewuim *et al.* (1997) used a passive technique (pitfall), ours was an active one (sifting), yet both outcomes demonstrated the dominance of *P. megacephala* and *O. troglodytes*, and the rarity of *P. tarsatus* - the solitary stink ant. Nonetheless, Ewuim *et al.* (1997) had earlier identified the shortcomings of pitfalls in adjudging the relative abundance of ants, citing the absence of army ants in their traps despite evidence of their trail within their study site. The sifting technique revealed not only a similarity in proportion of these ants, but it also captured similar species as reported by Ewuim *et al.* (1997). These were much more than those obtained from a baited

trap where four species, including *Pheidole* and *Paratrechina* species were encountered around human habitation (Ewuim and Osondu, 2008).

Interestingly, Chao 2 and Jackknife 2 estimates, and the rarefaction curve indicated that there were more ant species unaccounted for in this study. Perhaps, adopting multiple sampling techniques such as baited traps or increasing sampling efforts could help capture such species. Studies by Egbon *et al.* (2019a,b), Begum *et al.* (2021) and Subedi *et al.* (2021) recognise the import of sampling efforts in improving inventory completeness. Recall that the non-parametric estimates from this current study suggest that about half the true species richness in the studied area was yet to be sampled. Thus, further sampling studies are required either by employing other techniques or by increasing sampling intensity to improve the checklist and have a rarefaction curve that approaches or reaches its plateau, where it will be near impossible to discover additional species -i.e., the point of a complete inventory.

Conclusion: In summary, this study presents an elementary inventory of leaf-litter ant species (and their relative abundance) in the several-centuries-old Benin moat, which typifies the ecological relevance of having (a) forest patch(es) or green spaces (e.g., gardens, green parks, ponds, among others) that accommodate different species, albeit enclosed within a metropolis. Benin moat not only provide a unique space for assessing the effects of urbanisation on closed communities of flora and fauna, it also creates a means for other animals to thrive amidst human settlements.

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