

Biodiversity of water-filled tree holes in Kibale Forest, a fragmented Ugandan tropical moist forest

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Abstract

Water-filled tree holes are important temporal ecosystems for a number of different aquatic and semi-aquatic organisms. In this study we tried to assess the biodiversity of these habitats and the abiotic factors that determine this diversity. The results showed high values for Simpson biodiversity index and Shannon evenness at family levels. Furthermore the investigated biotic and abiotic factors including hole volume, depth, temperature, pH, mass of detritus as well as tree dbh and canopy cover explained the species numbers and family assembly of the majority of holes. Against expectations the dissolved oxygen levels do not have any impact on community structures.

INTRODUCTION

Aquatic tree hole habitats (pythothelmata) are small natural containers formed by living or dead plant parts when rainwater is collected in bark depressions. They are most commonly found when main branches fork or brake, behind scar tissues and behind outgrowths of bracket fungi (McCafferty 1998). In tropical regions many tree holes are also found in buttresses.

Pythothelmata are mainly temporal ecosystems, strongly dependant on the lengths of the wet period and dry season (Williams, 1987). In addition to water, they also contain leaf litter and other detritus compounds, accumulated over longer periods of time, which are considered as the most import food source. Consequently, catabolic processes predominate in tree holes and often lead to anoxic conditions.

Hence the animals found in such tree holes can be characterized by three major traits. Firstly, they mainly filter the water for bacteria or feed directly from detritus, although there are some predatory species present.

Secondly, they must have adaptations for low oxygen levels, common in many holes. They may use haemoglobin to maximize their oxygen gleaning, like many Chironomidae, or use atmospheric oxygen, like Culicidae and Stratiomyidae.

Furthermore, they must develop strategies to recolonize or survive in temporal habitats. Some families pass the dry periods as adults but for others detritus plays a key role as refuge for dormant larvae and substrate for eggs and cysts (Jeffries & Mills, 1990).

In contrast to European and North American water-filled plant reservoirs, tropical tree holes are not well studied. Investigations in South America have shown a distinctly different species and family assembly from temperate phytothelmata. Little is known about paleotropical tree holes, although they might be of high importance as breeding sites for mosquitoes that transmit diseases such as malaria (Jeffries & Mills, 1990).

This study aimed to collect more information about aquatic African tree hole habitats, in a tropical moist forest, and answer the following research questions:

- i) What are the community structures of phytothelmata in unlogged tropical moist forests and how do tree holes differ in their community structure?
- ii) Which biotic and abiotic factors influence biodiversity and abundances in these ecosystems and in what way do they interact?

METHODOLOGY

The study site was the tropical moist forest of Kibale National Park situated in western rift valley in Uganda. Our base camp was the Makerere University Biological Field Station (MUBFS; 0°13'0"14"N, 30°19'30"32"E). The climate of the site is characterized by two rainy seasons with an average annual amount of 1500 mm of rainfall (Chapman *et al.*, 1997, Skorupa, 1988, Struhsaker, 1975, 1997: in Chapman, 2000a). The sampling occurred between 14th and 19th September, during the main rainy season.

Three transects were established in unlogged forest in the surroundings of the field station. They were searched randomly and 38 tree holes, with at least 10 ml of water, were located and sampled.

For each hole, a set of external biotic and abiotic parameters were measured in situ. Height of the hole, diameter of opening and depth of hole were recorded as well as diameter at breast height (dbh) and tree species. Canopy cover was estimated using a spherical densiometer. Temperature was determined using a thermometer and pH was measured using a pHep device (Hanna).

All water was withdrawn from the tree holes using firstly a pipe of 10 mm diameter and then a pipe of 2 mm diameter for the remaining water. The water was filtered (mesh size of 200 nm) and its volume recorded. A small volume of the filtered water was used to determine the oxygen level using a dissolved oxygen kit (Hanna). If possible, up to 120 ml of the filtered water was stored and brought back to the field station, along with the total amount of detritus found in the holes and the particulate organic matter filtered out of the water.

In the laboratory the filtered water samples were allowed to settle for 24 hours and the upper water layers were used to determine transparency using a 100 ml test tube with 17 mm diameter (adapted as a transparency cylinder). A small white disk (10 mm diameter) with a black dot (6 mm diameter) was inserted into the tube. The height of the water column was recorded at the point in which the disk was no longer visible.

Detritus samples were weighed and a 10 g sample taken. The remains of the detritus were put to dry in a 70 °C climatic chamber. After 48 hours, dry weight was determined.

For each hole, the filtered material and the 10 g sample of detritus were analyzed and all meso- and macro-plankton and benthos organisms were collected. Afterwards, the animals were measured (length and width) and identified to family level. Vertebrates were not included in the analysis.

RESULTS

Abiotic measurement

The results of the abiotic measurements show a very high heterogeneity between the tree holes (Table 1). In many parameters, such as detritus weight and hole volume, the standard deviation is very high. The average dissolved oxygen value of 2.5 mg/l is low and 53% of the values are below 2.3 mg/l. Temperature and pH measurements did not show high levels of variation among phytothemata. The average diameter of trees at breast height (DBH) is 52.14 cm \pm 65.87 (S.D.).

Table 1. Abiotic and biotic factors measured

	tree height [cm]	tree DBH [cm]	canopy cover [%]	hole diameter [cm]	hole depth [cm]	hole volume [ml]	detritus wet weight [g]	detritus dry weight [g]
Mean	153.58	52.14	91.34	7.35	9.94	272.26	399.57	100.90
Sd	119.50	65.87	5.26	4.67	11.69	316.64	824.40	234.05
Min	19.00	6.00	78.16	1.00	1.00	10.00	0.00	0.00
Max	545.00	270.00	100.00	22.00	65.00	1190.00	4508.00	1184.76
	T[°C]	pH	dissolved O ₂ [mg/l]	transparency [ml]	number of individuals	number of families	no. pelagic animals	no. of benthic animals
Mean	17.39	6.77	2.56	54.44	24.42	3.82	18.66	5.76
Sd	1.57	0.95	1.37	32.06	21.90	1.77	20.72	6.06
Min	16.00	4.00	0.40	6.00	3.00	1.00	0.00	1.00
Max	22.00	8.80	6.60	106.00	87.00	11.00	83.00	30.00

Biotic analysis

We found 929 individual invertebrate, belonging to 32 different taxa. 737 of them were associated with the 21 taxa as shown in Table 2. Over 60% of the individuals found belonged to two diptera families (Culicidae and Chironomidae). The majority of the taxa identified had very few individuals and occurred only in a small number of tree holes. Culicidae were very common and occurred in 92% of the tree holes.

Three vertebrates (two frog species and one lizard) were found in three different locations.

Table 2. Number of individuals per identified taxa and their occurrence

Taxa no.	Taxa	No. of Indv.	Individuals [%]	Occurrence in holes [%]	Taxa no.	Taxa	No. of Indv.	Individuals [%]	Occurrence in holes [%]
1	Ceratopogonida	2	0.27	2.63	12	Colembola	4	0.54	7.89
2	Cordulida	3	0.41	2.63	13	Dixida	7	0.95	7.89
3	Empidida	2	0.27	2.63	14	Ephydrida	3	0.41	7.89
4	Gastropoda	1	0.14	2.63	15	Nematoda	6	0.81	10.53
5	Isopoda	1	0.14	2.63	16	Hydracarina	22	2.99	15.79
6	Lepidoptera	1	0.14	2.63	17	Tipulida	8	1.09	15.79
7	Nepida	1	0.14	2.63	18	Stratiomyida	45	6.11	23.68
8	Sciomycida	1	0.14	2.63	19	Helodida	128	17.37	47.37
9	Halipida	2	0.27	5.26	20	Chironomida	208	28.22	68.42
10	Ostracoda	28	3.80	5.26	21	Culicida	261	35.41	92.11
11	Psychodida	3	0.41	5.26					

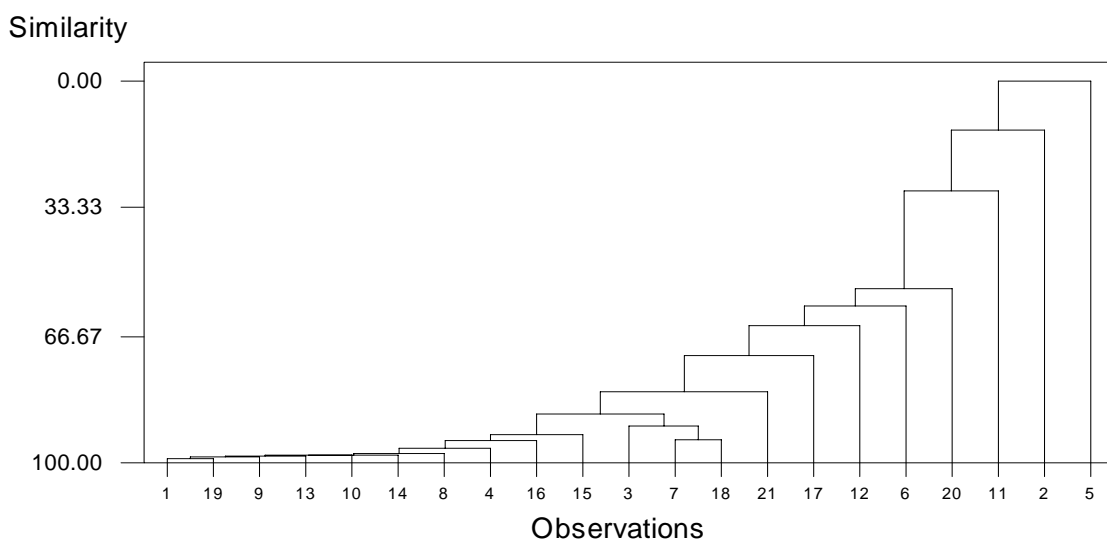


Fig. 1 Similarity among the taxa’s distribution in the tree holes. Number representing taxa according to table 2.

Cluster analysis, using single linkage and a Pearson distance measure, were calculated to determine the similarity among the taxa’s distribution in the tree holes (Fig. 1). The analysis shows that there is a group of 13 taxa with very similar distribution patterns (over 90%), including the two numerous taxa of Stratiomyida and Helodida. In contrast, the family of Chironomida shows quite a distinct distribution.

Biodiversity analysis

Looking at the tree holes as one single population, we calculated two diversity indices: Simpson's index value was 0.828 (sd 0.006) and Shannon's index value was 2.18. Shannon's evenness value was calculated as 0.591. Considering each phytothelmata separately, the Simpson's index varies from 0 to 1 (Table 3).

Table 3. Number of individuals and families and Simpson diversity index per tree hole

Tree Hole No.	No. of individuals	No. of families	Simpson diversity	Sd of Simpson diversity	Tree Hole No.	No. of individuals	No. of families	Simpson diversity	Sd of Simpson diversity
1	11	3	0.655	0.095	20	6	3	0.806	0.051
2	46	6	0.802	0.021	21	20	3	0.333	0.123
3	11	6	0.846	0.075	22	13	2	0.590	0.098
4	54	6	0.588	0.054	23	9	2	0.691	0.059
5	29	4	0.842	0.018	24	49	4	0.188	0.073
6	3	3	1.000	0.000	25	43	6	0.767	0.035
7	5	3	0.700	0.175	26	18	4	0.673	0.084
8	83	11	0.709	0.034	27	37	5	0.667	0.045
9	3	2	0.667	0.181	28	7	2	0.286	0.189
10	16	4	0.484	0.136	29	26	3	0.218	0.103
11	29	5	0.667	0.051	30	2	2	0.667	0.000
12	1	2	1.000	0.000	31	3	1	0.000	0.000
13	5	4	0.905	0.053	32	18	5	0.800	0.046
14	47	4	0.345	0.083	33	19	5	0.674	0.096
15	10	3	0.673	0.110	34	6	3	0.836	0.071
16	1	2	0.500	0.217	35	22	5	0.740	0.041
17	6	3	0.821	0.058	36	31	4	0.747	0.041
18	10	4	0.803	0.076	37	9	4	0.800	0.079
19	21	4	0.348	0.127	38	9	3	0.418	0.077

Cluster analysis, using complete linkage and an Euclidean distance measure, was applied to determine similarity among the tree holes assemblages (Fig. 2). It shows that there is a large group of holes which share a very similar taxa assembly. In contrast, there are also several clusters that show distinctive biodiversity patterns, especially the cluster formed by holes 14 and 24. Holes 4 and 8 are also colonized by a quite unique assemblage of taxa.

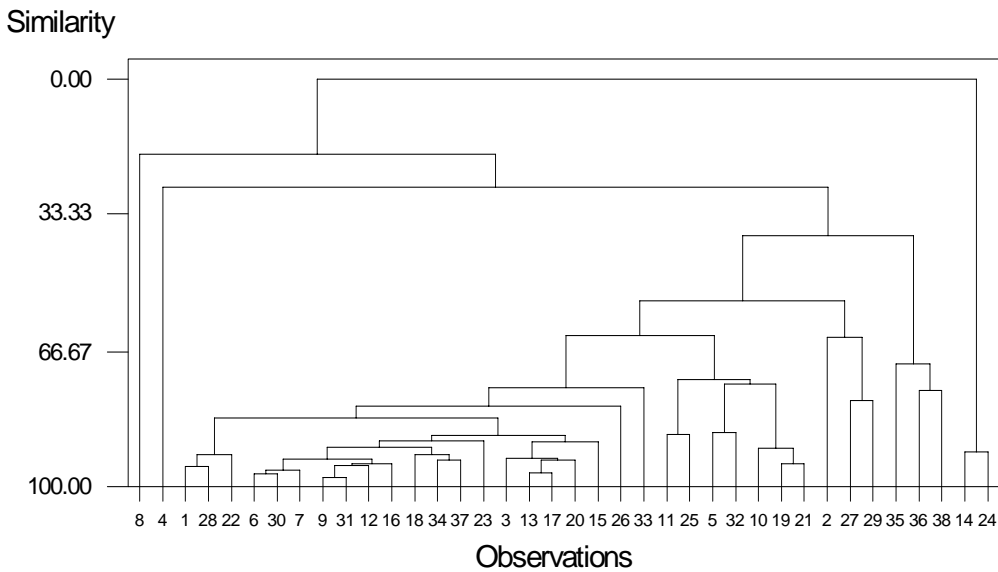


Fig. 2 Similarity among the tree holes assemblages. Numbers representing different tree holes.

Relations within abiotic factors

Using complete linkage and an Euclidean distance measure, we did a cluster analysis to determine similarity among the tree holes regarding the abiotic factors (Fig. 3). 79% of the holes form a cluster with more than 80% similarity. Two other clusters were formed that differ more widely according to the abiotic factors.

All biotic and abiotic parameters have been tested for correlations. Significant correlations are shown in Table 4. The analysis showed that Dbh correlates positively with hole diameter, volume and depth as well as with both detritus weight categories. Furthermore positive correlations between the pH level and hole depth, as well as between water transparency and dissolved oxygen were significant.

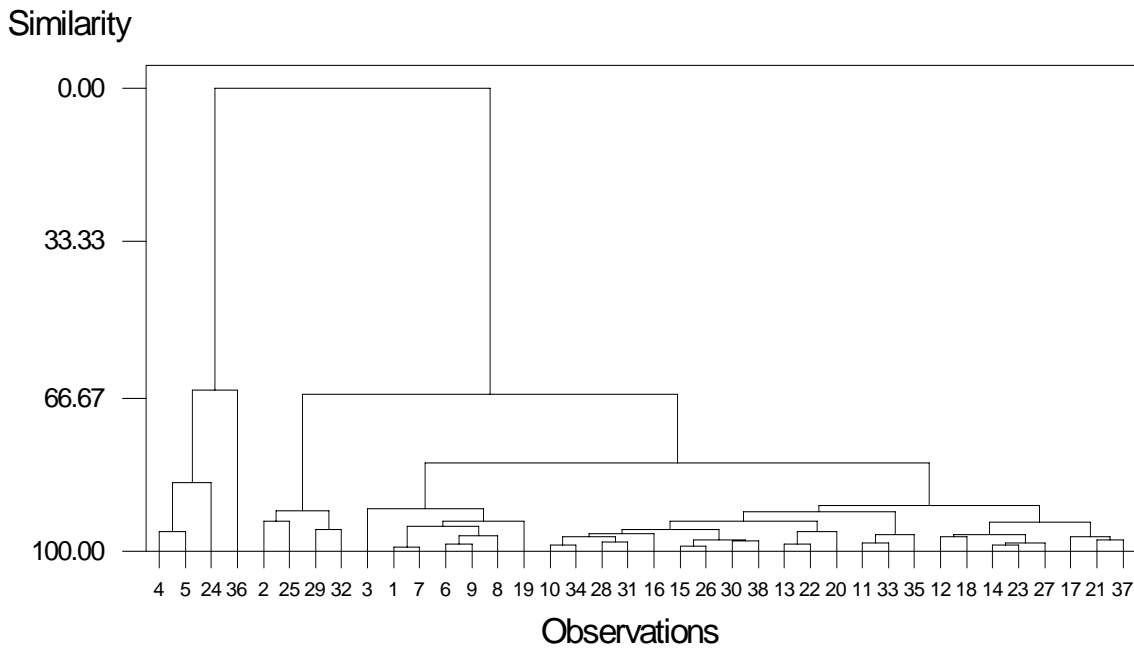


Fig. 3 Similarity among the tree holes regarding the abiotic factors. Numbers representing different tree holes.

Table 4. Significant correlations between abiotic factors

Variables	Pearson's correlation	p-value	Variables	Spearman's correlation	p-value
pH & hole depth	0.454	0.007	Dbh & detritus wet weight	0.557	<0.001
hole diameter & hole volume	0.675	<0.001	Dbh & detritus dry weight	0.543	0.001
hole volume & hole depth	0.492	0.002	Dbh & hole diameter	0.525	0.001
detritus wet weight & hole diameter	0.858	<0.001	Dbh & hole volume	0.567	<0.001
detritus wet weight & hole volume	0.722	<0.001	Dbh & hole depth	0.377	0.02
detritus dry weight & hole diameter	0.796	<0.001			
detritus dry weight & hole volume	0.724	<0.001			
detritus dry weight & detritus wet weight	0.965	<0.001			
water transparency & dissolved oxygen	0.467	0.033			

Finally we built two different regression models (Table 5), one to predict the number of families and one to predict the number of individuals per tree hole. The R-Sq Value for the regression model for number of individuals explains 60% of the variance, while the R-Sq Value for the regression model for number of families explains 81% of the variance. Both models are significant.

Table 5. Regression models for number of individuals and number of families

Regression equation	R-Sq	F	p-value
number of individuals = - 2.71 pH - 0.94 diameter of hole [cm] - 10.3 tree height [cm] + 19.4 hole depth [cm] + 15.9 hole volume [ml] + 0.84 dissolved oxygen [mg/l] + 1.30 canopy cover [%] - 39.0 detritus wet weight [g] + 39.3 detritus dry weight - 71.8	59.40%	2.93	0.025
number of families = - 1.11 pH - 0.262 hole diameter [cm] - 2.11 tree height [cm] + 1.68 hole depth [cm] + 1.01 tree volume [ml] - 0.101 dissolved oxygen [mg/l] + 2.72 detritus dry weight [g] - 0.0150 transparency [ml] + 8.21	81.30%	3.47	0.045

DISCUSSION

Taxa assemblage and biodiversity

Identification of the individuals resulted in a large number of different taxa (Table 2). Most of them show adaptations to recolonise or survive in temporal habitats as well as to the low dissolved oxygen levels found in most holes. As expected (Jeffries & Mills, 1990) Diptera were the most common inhabitants of the sampled reservoirs. Within the order of Diptera the families of Culicidae and Chironomidae were dominant. These taxa are commonly found in phytothelmata (e.g., Lounibos, 1981; Louton *et al.*, 1996) and there are even some specialized species which are only found in these habitats (McCafferty 1998).

In contrast only two different taxa of Crustacea, which could be identified as two morphospecies, were found in the samples. This is a low number of taxa considering the ability to adapt to temporal aquatic systems (Bayliss, 2002).

Other interesting groups of invertebrate taxa were Cordulidae and Nepidae larvae, which must have specially adapted to these ecosystems. Furthermore the sighting of two frog species and of one lizard under 5 cm of water and a 7 cm detritus layer shows the importance of tree holes as a refuge and food source for various vertebrates.

The cluster analysis of distribution patterns of the taxa (Fig. 1) mapped the two dominant families of Culicidae and Chironomidae apart from all other taxa. The analysis also suggests that they share little similarity between themselves. The reason for this result might be that these two families are each found in high number in some holes that otherwise showed a very low biodiversity (for example high number of Culicidae in hole 14 and 24).

In general the results of Simpson and of Shannon indices (Table 3) showed high biodiversity levels for most holes and for the total number of individuals found. This is remarkable, especially as species were only identified to family level and a species-identification would produce a much higher result. This confirms the trend that biodiversity is higher in tropical ecosystems.

Relations within different biotic and abiotic parameters

Many of the expected correlations between abiotic factors were confirmed by the data analysis. Large trees with a high Dbh are expected to form tree holes with greater hole volume, depth and diameter as well as with a higher detritus weight (Table 4). Moreover the detritus mass increases with the volume and the diameter of the hole.

Dissolved oxygen and transparency show a significant negative correlation. This might be because opaque water contains a greater amount of fine particulate and dissolved organic matter. Therefore cloudy water might be a better habitat for heterotrophic bacteria and protozoa which have a negative impact on the oxygen level. The positive correlation between pH and hole depth is an interesting finding and needs further investigation.

The cluster analyses of the similarity of tree holes according to families and the similarity analysis of holes according to their abiotic factors (Figs. 2 and 3) showed similar distribution pattern. Both analyses created one cluster containing over 50% of the sampled habitats within a similarity of at least 80%.

A comparison of the two models shows that all holes that were clustered together in the similarity analysis of holes according to families are also in the main group of the second analysis. This confirms that the most important external factors for most holes were investigated. However some holes that have a very unique species assemblage (for example hole 8) are, according to the abiotic cluster analysis, closely related to other holes with completely different species assemblages. This suggests that there must be some biotic or abiotic parameters, which vary strongly between these holes.

The models established by the regression analysis (Table 5) seem to lead to quite accurate results. The model for estimation of the number of families has an especially high R-sq value (81%) but as the cluster analysis of the holes demonstrates, there will be problems with some unusual holes.

Simpler regression models were established but they were found to be far less accurate. Against expectations, during this process the dissolved oxygen level turned out to be the least important parameter in both regressions. Two proposals have been raised to explain this finding.

Firstly the dissolved oxygen level could also vary strongly during rainy season due to drought periods. Therefore all animals in tree holes have to develop strategies to cope with anoxic conditions and are consequently not affected by intermediate changes of oxygen levels.

Secondly high oxygen levels can be toxic for many different heterotrophic bacteria and protozoa (Jeffries & Mills, 1990). Hence a high level of oxygen, though favourable for macroscopic organisms, will have a negative impact on the food availability of those communities.

We would recommend further investigation of these issues to confirm or decline our proposals. Furthermore we would suggest long term studies of abiotic factors influencing tropical temporal habitats. It is of high importance to understand these ecosystems more clearly as a comparison between the two rainy seasons and factors like duration of drought could not be measured during this project.

Subsequent to the field course, we will measure the total biomass of the sampled tree holes and include them in our analysis. Additionally we will continue with the identification process as due to lack of time some taxa were only identified to Class and only a low accuracy level could be achieved.

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