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Rainy Season Food Availability for Anurans of Kimboza Forest Reserve, Tanzania

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

Amphibians are mostly considered to be generalist predators. Some species have a narrow diet or even specialize on certain prey categories. An assessment of the feeding habits of anuran species of Kimboza forest reserve, Tanzania was conducted during rainy season between March 2017 and June 2017. Through a visual encounter survey, 93 species were captured and analyzed for gut contents. About 462 prey items were identified the most abundant component being Isoptera and Hymenoptera. There was no significant difference ($P > 0.05$) in the anurans' diet (composition or diversity or preferences) among different habitats of the forest (along boundaries and within the forest) but significant difference ($P < 0.05$) was found between the upper Stratum and the lower Stratum of the forest. The study provides a preliminary estimation of the actual diet of anurans and establishes a general feeding pattern for these species for the management of the Kimboza forest reserve. The study recommends the intensification of similar assessments in the nearby forest areas to come up with a complete description of the feeding habits for management purposes.

Keywords: Anura; prey; diets; Kimboza; Tanzania.

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1. INTRODUCTION

The differences in anurans' diet composition are due to variation of habitats abundance and diversity of prey [1,2]. The utilization of a broad range of diets provides assurance such that when there is an internal or external influence on the habitat the species tend to respond and cope accordingly. This is different for the highly specialized species which become highly prone to slight or major changes in the environment resulting in their potential elimination and probably extinction [3]. In contrast, species having a broad range of food diets are more flexible and highly adapted to getting a variety of energy sources [4]. Allmon, [5] observed that the distribution patterns of forest leaf litter anurans were linked to prey availability. Thus, one of the factors that influence anurans' distribution in their habitats is diet [6]. Varying exploitation of foraging strategy may as well influence the distribution of anuran species in the same habitat [7]. Knowledge of feeding habits of coexisting anuran species can contribute to the understanding of the trophic level organization within a particular habitat [8]. Feeding provides a means of understanding organism connection to the environmental resources, and to determine its relation to the environment [9]. The information can be very useful in planning management strategies for the amphibians and other vertebrates in an ecosystem.

Kimboza forest reserve located within the slopes of North Uluguru Nature Reserve within the Eastern Arc Mountains is highly influenced by anthropogenic activities which threaten wildlife including amphibians [0]. This study aimed to describe the feeding habits of amphibian (anuran) species in Kimboza Forest Reserve, specifically through identifying the dietary composition of anuran species, and the influence of habitat on their feeding preferences for management purposes.

2. MATERIALS AND METHODS

Study area **-** This study was done in Kimboza Forest Reserve located approximately 50 km southeast of Morogoro district in Morogoro region, Tanzania (Fig. 1). It covers an area of 385ha and lies between 06°59' - 7°02' S and 37°47'- 37° 49'E within the slopes of North Uluguru Nature Reserve [11] within the Eastern Arc Mountains and is categorized as Coastal Forests of Tanzania having IUCN category IV status. The study site has its flora species facing intensified threat due to deforestation activities going on in the forest [12]. The local climate of this area is characterized by bimodal rainfall patterns averaging 1600 mm per year. Most of the rains fall between June and May which is the rain season, while the dry season with little rain is between October and March [13].

Sampling Designs **-** Sampling of anurans was carried out at the end of March which is the beginning of the rainy season and at the end of June 2017 a period of heavy rain (eight days in each month). Visual encounter survey was applied to detect anuran species for capture. The forest was divided into two parts (upper and lower Stratum) separated from each other by a dirt road passing between the two parts while the lower Stratum was more disturbed than the upper Stratum. Then each Stratum was stratified into four habitats: (i) areas along forest and road border, (ii) areas where the forest border with farms whereby each of them was between 0 m to 50 m from the forest borderline to the central part of the forest. The edge effect on amphibians is known to extend 100 m from the forest boundaries to the forest interiors [14], iii) a forest interior between 100 m to 400 m from the forest boundaries transects to the forest interior to avoid edge effect. (iv) along Kimboza forest streams where amphibians usually tend to aggregate [15]. In each Stratum per habitat, one transect of 300 × 4 m was defined. Within each transect, six rectangular plots of 50 × 4 m were established making a total of 48 plots in both strata of the forest for the entire study. The rectangular plots of 50 × 4 m were searched for anurans in 30 minutes by four persons. A total of eight 300×4m transects (0.96 ha of the forest) was searched from 0600 to 1700 hours for diurnal anurans. Each transect was surveyed twice in March and June.

Anurans were searched in all habitats including water streams, water ponds, under logs, within leaf litter, tree openings, and holes as well as under stones. Efforts were made to search tree canopies and bushes near water sources for arboreal amphibians. All animals from their habitats were captured by gloved hands for identification using the keys and field guide books [16,17,18].

Anurans' stomach contents were collected after dissecting an animal from the mouth through the stomach to the anus to get the whole digestive tract for examination. The whole stored food and fecal material were emptied into containers for

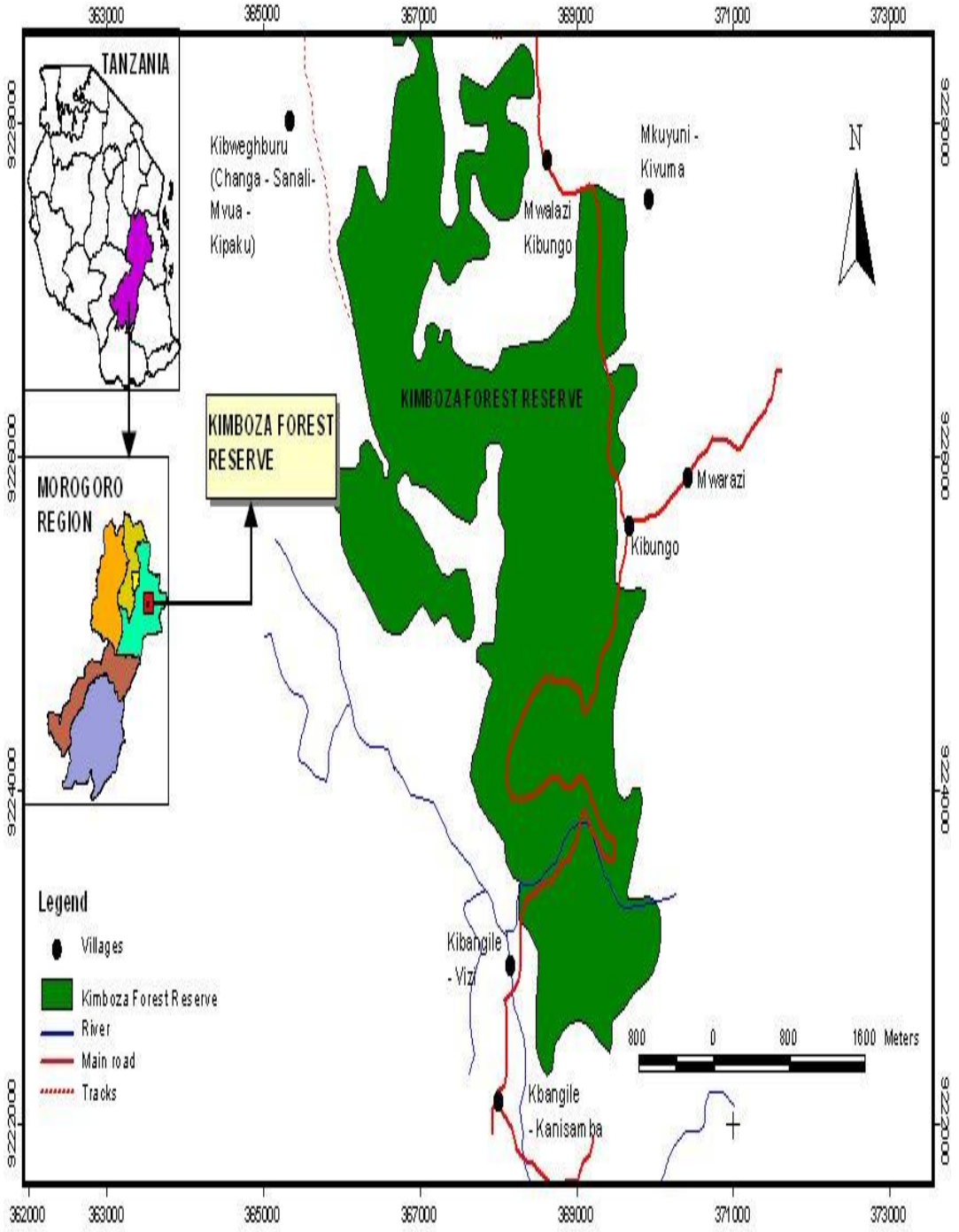


Fig. 1. The map of The Kimboza forest reserve showing the forest location in Tanzania and the surrounding villages. (Source: GIS Unit, Sokoine University of Agriculture)

analysis under dissecting microscopy. All materials found in the digestive tract of each specimen were identified possibly down to the genus level through the help of field guidebooks and those unidentified were marked as unidentified.

Data analysis. -To establish the diet composition of anurans four parameters were analyzed: (i) the rate of feeding activity, (ii) abundance of different food groups, (iii) rate of occurrence of food groups, and (iv) the dietary diversity.

i. The rate of feeding activity was estimated as the percentage of guts containing food for the total number of guts examined.

Equation no 1. Rate of feeding activity = $\frac{100n}{N}$

Where n is the number of guts with food, N is the total number of guts examined.

ii. The abundance of various food groups was estimated from the guts contents (percentage of the total number of individual prey/total number of all prey).
iii. The frequency of occurrence was determined by dividing the number of digestive guts that contained a particular prey by the total number of guts with prey. A food group was then be classified as constant when registered in > 50% of the guts of a particular species, secondary when present in 25 - 50% of guts, or accidental when observed in < 25% of the guts [19].
iv. The dietary diversity was analyzed in two parts: First, dietary diversity per each species, and second, dietary diversity of anurans in each studied habitat. The dietary diversity was estimated with Shannon–wiener diversity index (H').

Equation no 2: $H = -\sum \left[\frac{ni}{N}\right] \ln \left[\frac{ni}{N}\right]$

Where n is the number of prey category i and N is the total number of prey.

Additionally, the degree of food preference was inferred using the index Degree of Food Preference (DFP) developed by Braga [20]. Food groups were ranked by categories from 1 to 4, in the following way: the maximum value (4) was assigned when only one food group was found in a gut. When guts contained more than one group, the value "3" was given to the most abundant, while "2" was given to the second most common, and "1" was attributed to less abundant groups.

DFP was calculated as:

Equation no. 3: DFP = S (i) /N

Where *S* (i) is the sum of values given to a food group *i* in the guts, and *N*, the total number of guts of each species analyzed. Food groups were then categorized as highly preferential (3 < DFP < 4), preferential (2 < DFP < 3), secondary (1 < DFP< 2) or occasional (0 < DFP < 1).

Microsoft office excel 2007 was used to calculate the diet diversity of each anuran species and to calculate the diet diversity of anurans in each habitat. The program of SPSS version 20 was used to perform statistical analysis whereby Kruskal-Wallis test and Mann-Whitney U test (probability level at 0.05) was used to estimate the significance of variation of abundance of food groups, frequency of occurrence, diet diversity index, and Degree of Food Preference index among anuran species in different habitats.

3. RESULTS

A total of 93 adult anurans (24 males and 55 females) were sampled during the months of March to June 2017. The majority of the anurans species occurred along Kimboza streams (70% of all occurrence) and along area where the forest border with farms (70% of all occurrence) followed by forest interiors (60% of all occurrence) in the upper fragment of Kimboza forest reserve. However, areas along the forest and tarmac road border and along Kimboza streams of the lower fragment of Kimboza forest had the lowest occurrence of anurans (only one species occurred in this area) (Table 1). Generally, in the whole forest the variation of total number of occurrences of anurans in different habitats was not significant (Q=11; DF =6; P=0.096) but important as it was almost significant. Nonetheless, the variation of total number of occurrences of anurans in different habitats between upper and lower fragment of the forest was significant (McNemar Test, P<0.05).

Out of 93 adult anurans from 12 species, approximately 65% had full gut contents, while 35% of guts were empty (Table 1). *Arthroleptis xenodactyloides* contributed largely to the number of individuals with full gut contents (40% of all individuals with gut content. Further, *Afrixalus stuhlmanni* was the species that had the highest number of empty guts *(*by 36% of all individuals with empty guts) (Table 2). Most of the species with empty guts were from the upper Stratum of the forest in the areas along the forest and farm border, which contributed to 76% of all individuals with empty guts.

Table 1. The distribution of anuran species among different habitats of Kimboza forest reserve

Species name	HABITATS							% occurrence of species in different microhabitats
	UFI	UFF	UFS	UFR	LFI	LFF	LFRS	
Leptopelis uluguruensis	x			x				29
Leptopelis flavomaculatus	x	x	x	x		x		71
Arthroleptis xenodactyloides	x	x	x	x	x	x		86
Arthroleptis affinis	x		x			x		43
Nectophrynoides tornieri	x			x	x			43
Afrixalus stuhlmanni		x						14
Afrixalus uluguruensis		x	x					29
Hyperolius mitchelli							x	14
Phrynobatrachus acridoides			x					14
Phrynobatrachus natalensis		x				x		29
Xenopus borealis	x	x	x					43
Ptychadena anchietae			x					14
Chiromantis xerampelina		x			x			29
% occurrence of all species in a habitat	60	70	70	40	30	40	10	

UFI= interior of the forest in the upper fragment,
UFF=along the forest and farm border in the upper fragment of the forest,
UFS= along the Kimboza streams in the upper fragment of the forest,
UFR= along the forest and road border in the upper fragment of the forest,
LFI = interior of the forest in lower fragment of the forest,
LFF= along the forest and farm border in thelower fragment of the forest,
LFRS=along the forest border with road and along Kimboza stream in lower fragment of the forest.

Table 2. The rate of feeding activities of anuran species in Kimboza Forest Reserve

Species name	N	Individuals with gut contents (N)	Individuals with Guts content (%)	Individuals with empty guts(N)	Individuals with empty guts (%)
Arthroleptis affinis	4	3	5	1	3
Arthroleptis xenodactyloides	27	24	40	3	9
Leptopelis flavomaculatus	14	11	18	3	9
Leptopelis uluguruensis	3	2	3	1	3
Nectophrynoides tornieri	3	3	5	0	0
Afrixalus stuhlmanni	13	1	2	12	36
Afrixalus uluguruensis	6	0	0	6	18
Phrynobatrachus acridoides	2	2	3	0	0
Phrynobatrachus natalensis	12	5	8	7	21
Xenopus borealis	2	2	3	0	0
Ptychadena anchietae	3	3	5	0	0
Chiromantis xerampelina	4	4	7	0	0
Total number of individuals	93	60		33	
Percent of individuals		65		35	

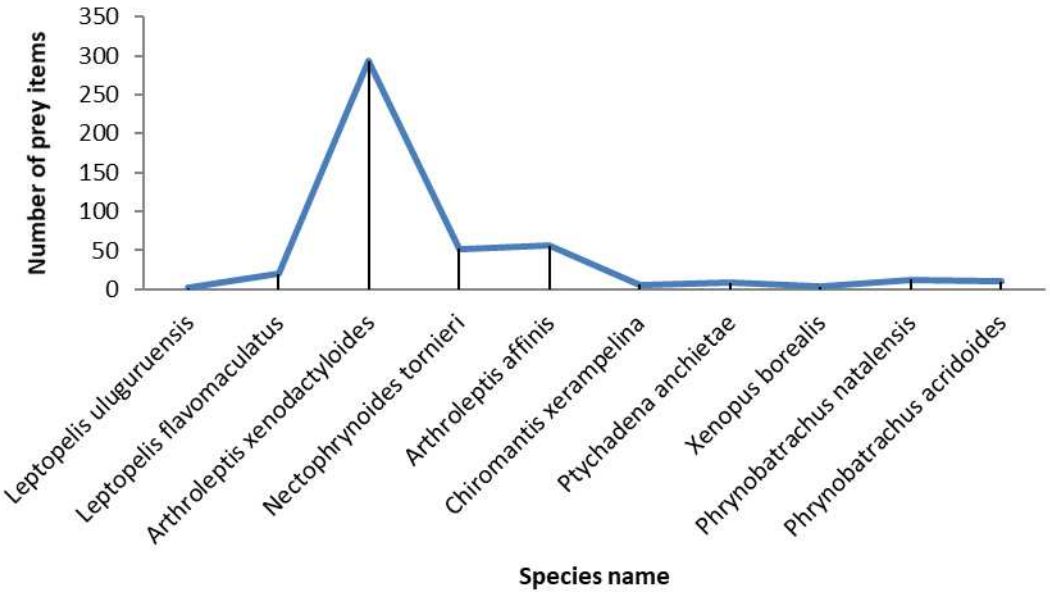


Fig. 2. Number of prey items per each species in Kimboza forest reserve

Diet analysis involved only 10 out of 12 anurans species that were examined. Anurans from two species *Afrixalus stuhlmanni* and *Afrixalus uluguruensis* were not included in the analysis since almost all of them had empty guts (Table 1 The anuran species with highest number of prey items was *Arthroleptis xenodactyloides* (63% of all prey items), while *Leptopelis uluguruensi* (Fig. 2).

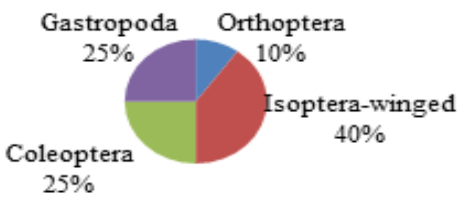
About 462 prey items belonging to 10 categories of food groups were collected from the digestive guts of 60 adult anurans. The categories of the food groups included Isoptera, Orthoptera, Coleoptera, Gastropoda, larvae, Hymenoptera, Araneae, Decapoda, Diptera, and unidentified (Fig. 2). The food groups in the unidentified category consisted of strata from the invertebrate. The most abundant food group was Isoptera (53.25%) followed by Hymenoptera (37.88%), but Decapoda (0.22%) was the least abundant group with only one prey item.

The percentages of prey items eaten by anuras in the upper Stratum were significantly higher than that of the lower Stratum (Mann Whitney U test, $U = 18$, $P < 0.05$). The most frequently occurring food groups were Coleoptera found in the digestive guts of 6 out of 10 anurans species. Coleopterans was a secondary food group in the digestive guts of *Leptopelis flavomaculatus* (27%), *Nectophrynoides tornieri* (33%), *Chiromantis xerampelina* (50%), *Ptychadena anchietae* (33%), and *Phrynobatrachus acridoides* (50%), and lastly was accidental for *Arthroleptis xenodactyloides* (4%) (Fig. 3).

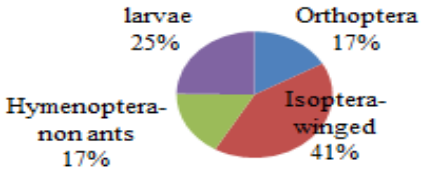
The second frequently occurring food groups were Orthoptera, Hymenoptera and Isoptera were found in the digestive guts of 5 out of 10 anurans species. The least occurring food groups were Decapoda(10%) and Diptera (10%).

However, the variation of frequency of occurrence of food groups among anuran species was not significant (Kruskal-Wallis Test $H'=5$, $P > 0.05$). Further, among species, *Ptychadena anchietae* had the highest diversity of prey items (Shannon-Wiener Function, $H'=1.4942$), while *Leptopelis uluguruensis* (Shannon-Wiener Function, $H=0$), had the lowest diversity of prey items.

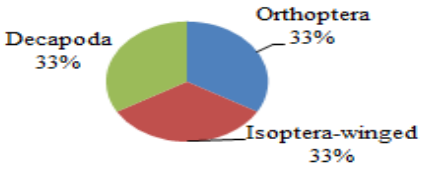
The diversity of prey items eaten by anurans found in different habitats had no significant difference (Kruskal-Wallis test $H=6$, $P > 0.05$). However, the diversity of prey items eaten by anurans in Kimboza forest reserve varied significantly between the upper and lower Stratum of the forest (Mann Whitney U test, $U =22$, $P < 0.05$). The diet diversity index of food groups eaten by anurans in the upper Stratum (1.077) was higher than that of the lower Stratum (0.6048) (Table 3).



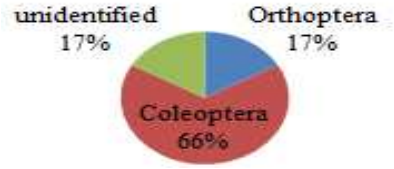
a) *Leptopelis flavomaculatus*



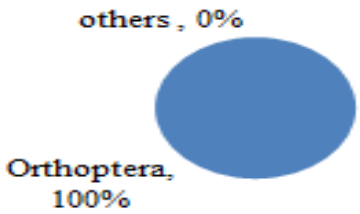
b) *Phrynobatrachus natalensis*



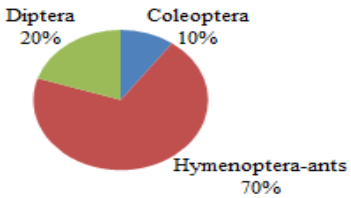
c) *Xenopus Borealis*



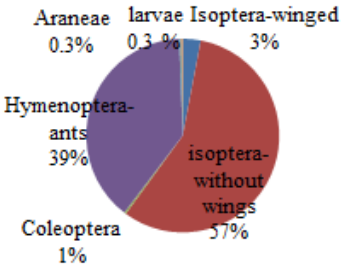
d) *Chiromantis xerampelina*



e) *Leptopelis uluguruensis*

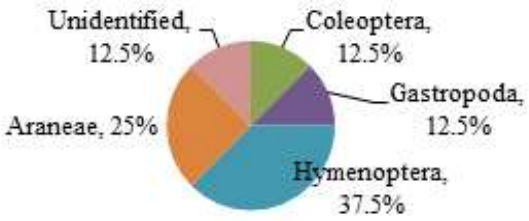


f) *Phrynobatrachus acridoides*

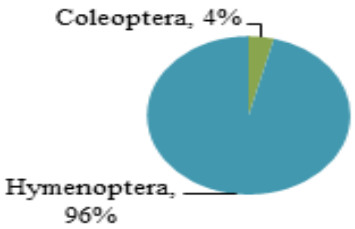


g) *Arthroleptis xenodactyloides*

h) *Arthroleptis affinis*



i) *Ptychadena anchietae*



j) *Nectophrynoides tornieri*

Fig. 3. Food groups eaten by each anuran species of Kimboza Forest Reserve

Table 3. Shannon-Wiener Function, H' of prey items eaten by anurans in different habitats of Kimboza Forest Reserve during March and June 2017

	Diversity of prey items (H')		
Habitat	**Both rounds**	**March**	**June**
Upper Stratum	1.077	0.8755	1.345
Lower Stratum	0.6048	0.4825	0.6365
Upper Stratum and tarmac road border	1.395	1.119	0.673
Lower Stratum and tarmac road border	-	-	-
Upper Stratum interior	1.167	1.167	-
Lower Stratum interior	0.4825	0.4825	-
Upper Stratum along Kimboza streams	0.9623	0.7163	1.06
Lower Stratum along Kimboza streams	-	-	-
Upper Stratum and farms border	0.5536	0.2494	1.517
Lower Stratum and farms border	0.6365	-	0.6365

Table 4. Degree of food preference of food groups eaten by anurans in the upper stratum of Kimboza Forest Reserve

	Degree of Food Preference									
Anuran species	**L.u**	**L.f**	**A.x**	**N.t**	**A.a**	**C.x**	**P.a**	**X.b**	**P.n**	**P.ac**
Food groups										
Orthoptera	2.67^{P}	0.07^{O}	-	-	-	-	-	1.33^{S}	0.36^{O}	-
Isoptera	-	0.79^{O}	2.39^{P}	-	2.75^{P}	-	-	1.33^{S}	0.09^{O}	-
Coleoptera	-	0.71^{O}	0.05^{O}	1.5^{S}	-	4^{H}	1.33^{S}	-	-	0.5^{O}
Gastropoda	-	1^{S}	-	-	-	-	1.33^{S}	-	-	-
Hymenoptera	-	-	0.67^{O}	3.5^{H}	-	-	1^{S}	-	0.55^{O}	3^{H}
Araneae	-	-	0.05^{O}		-	-	0.67^{O}	-	-	-
Insect larvae	-	-	0.05^{O}		0.5^{O}	-		-	0.82^{O}	-
Unidentified	-	-	-	-	-	-	0.33^{O}	-	-	-
Decapoda	-	-	-	-	-	-		1.33^{S}	-	-
Diptera	-	-	-	-	-	-			-	- 2^{P}

H=highly preferential P=preferential S=secondary O= occasional L.u = *Leptopelis uluguruensis,* L.f = *Leptopelis flavomaculatus,* C.x= *Chiromantis xerampelina*, P.a = *Ptychadena anchietae,* A.x = *Arthroleptis xenodactyloides,* N.t = Nectophrynoides *tornieri,* A.a= *Arthroleptis affinis*, X.b= *Xenopus borealis,* P.n= *Phrynobatrachus natalensis,* P.ac=*Phrynobatrachus acridoides*

Furthermore, the Degree of Food Preference differed significantly **(**Mann Whitney U test, U = 23, $P < 0.05$) between anurans in the upper and lower Stratum of the forest. Most of the food groups eaten by anurans found in the upper Stratum of the forest were considered secondary and occasional (Table 3). On the other hand, in the lower Stratum of the forest most of the food groups were considered preferential (Table 3). The upper Stratum had highly preferential as the highest level of DFP given to food groups eaten by anurans. The food group in the highly preferential category was Hymenoptera. Hymenoptera was highly preferential for both *Nectophrynoides tornieri* (3.50) and *Phrynobatrachus acridoides* (3.00). Preferential food groups were Isoptera Orthoptera and Diptera. Occasional food groups were all food groups except Diptera, Gastropoda, and Decapoda while secondary food groups were Isoptera, Orthoptera, Gastropoda, Decapoda, Coleoptera, and Hymenoptera (Table 4). However, the variation of values of Degree of Food Preference (DFP) among anuran species found in the upper Stratum of the forest was not significant (Kruskal-Wallis Test $H = 6.2$, $P > 0.05$).

Food groups from anurans found in the lower Stratum of the forest had preferential as the highest level of DFP. Preferential food groups were Isoptera and Orthoptera. Isoptera and Orthoptera were preferential for *Arthroleptis xenodactyloides* (2.89) and *Chiromantis xerampelina* (2.00) respectively. Hymenopterans in *Arthroleptis xenodactyloides* (1.22) was secondary preferred. The variation of the values of DFP among anuran species found in the lower Stratum of the forest was not significant (Kruskal-Wallis Test, $H = 16.5$, $P > 0.05$).

4. DISCUSSION

The study investigated food contents in the guts of amphibian species of Kimboza Nature Reserve in Morogoro Tanzania. Anurans' diet was found to consist of 10 different food groups and all belonging to the invertebrate category. A study by Anderson *et al.* [21] reveals that the diets of anurans consisted mainly of invertebrates. The diet of anurans of Kimboza forest reserve consisted of a high abundance of termites and ants indicating that their diet reflects prey availability in the forest [22,11,3]. Some species of anurans are known to opportunistic and may consume a huge number of termites if they meet during foraging. For instance, Biavati *et al.* [23] found that anuran species *Ameerega flavopicta (*Dendrobatidae) had taken a relatively big number of termites in central Brazil, where such insects were abundant. Given the evidence, the large consumption of isopterans (termites) and hymenopterans (ants) by anurans in Kimboza forest reserve was due to the high abundance of these insects in this study area [22].

The most frequently collected food group from the guts of anurans was Coleoptera 6 out of 10 anuran species. Coleoptera was found to be the most frequently collected food group, even though it was not the most abundant food group. The tendency of most anurans to consume the less abundant food group is said to be a strategy to save energy, capturing a big prey being more advantageous, and then hunting a high number of small prey [1]. Likewise, Orthoptera, which was the second most frequent food group but was among the less abundant food groups. Klaion *et al.* [24] found that coleopterans and Orthopterans are preyed upon by leaf-litter frogs, as they are abundant and easily captured. The diets of anuran species depend on several factors, but some of them are specialization, foraging mode, and diet plasticity [3].

There were notable differences in diets of anurans between the upper and the lower stratum of the forest. Anurans from the upper Stratum consumed more abundant and diverse food groups than the lower stratum of the forest. This significant difference in the abundance of food group, diversity of preference between the diet of anurans in the upper and lower Stratum may be explained by the difference in the quality of each stratum of the forest. Bogdan et al. (2013; Whitfield and Donnelly, 2006) established that for a habitat to meet the trophic needs of amphibians it must offer abundant and diversified food resources. The upper stratum appears to be content with anurans' food requirement as it offers diets forms different from the lower Stratum which its anurans consumed less abundant and less diversified food groups.

Several studies (e.g. [25,26,27] Tohe *et al.,* 2015) on the diet of anuran species belonging to genus *Ptychadena, Leptopelis, Phrynobatrachus,* and *Xenopus* established that these anurans are opportunistic generalist predators which lack feeding preferences and have a wide diversity of food groups with no constant food in their diets. These results to some extent support the study findings as species like *Ptychadena anchietae, Phrynobatrachus natalensis Leptopelis flavomaculatus* and *Xenopus borealis* had a wide diversity of food more than other anuran species in the forest, and no food group was constant.

5. CONCLUSION

The study provides the first insight into the feeding habits of anurans in Kimboza forest reserve. It provides a preliminary estimation of the actual general feeding pattern for anurans in Kimboza forest reserve. Future studies on the dry season and nocturnal species are recommended to come up with a complete description of the feeding habits of anuran species of Kimboza forest reserve.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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