# Morphological variation of Ridged Frogs of the Taita Hills, Kenya

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Submitted on: 2010, 5<sup>th</sup> October; revised on: 2011, 20<sup>th</sup> July; accepted on: 2011, 2<sup>nd</sup> December.

**Abstract.** Comparing of morphological character variation within taxa continues to play an important role in improving species inventories. Using morphometrical and non-meristic morphological adult characters, the diversity of the genus *Ptychadena* in Taita Hills was studied. Comparative material from elsewhere was not used, and therefore species names were only provisionally allocated to the taxa identified. Available names were discussed on the basis of comparisons with morphological data from other regions. The results revealed that female species are larger in size than males. Two species were identified and for each a standardized diagnosis of 32 characters is provided. Comparison of results with morphological data from related studies done elsewhere reveals that certain characters are of critical importance in differentiating the two *Ptychadena* species. The power of these morphological characters is discussed, especially for the background of rapid and easy identification of *Ptychadena* species in the field for conservation purposes.

Keywords. Diversity, Morphological characters, Ptychadena, Taita Hills, Kenya.

# INTRODUCTION

African herpetofauna is quite diverse in terms of species richness. However, geographical sampling and taxonomic inventories are inadequate and most countries are poorly investigated (Poynton, 1999). This study focused on the species diversity of Ridged Frogs, genus *Ptychadena* Boulenger, 1917 (Ptychadenidae), in Taita Hills. These anurans are common across sub-Saharan Africa, Madagascar and some smaller Oceanic islands. Currently, 49 species are recognized (Frost, 2008), which often occur syntopically (Rödel, 2002; Channing and Howell, 2006). Generally, the female species in this genus are larger in size in relation to the males, with most species sharing several morphological characteristics (Channing and Howell, 2006; Bwong et al., 2009). There have been several efforts towards an understanding of *Ptychadena* systematics (Guibé and Lamotte, 1957, 1958, 1960; Lamotte, 1967; Perret, 1979, 1981, 1987, 1994, 1996; Amiet, 1989; Poynton and Broadly, 1985; Largen, 1997, 2000). However, the genus can not be considered as well understood as still the majority of the suggested species are improperly defined. Standardized diagnostic schemes have been applied by few previous researchers but only for a limited number of species (Guibé and Lamotte, 1957; Perret, 1979; Poynton and Broadly, 1985). As a result, several of the suggested taxa are difficult to distinguish and their geographic ranges remain unknown. Moreover, there are only a few comparative studies, and therefore it is difficult to follow the placement of certain nominal species into the synonymy of others (Bwong et al., 2009).

While the genus *Ptychadena* in part has already been studied in other regions of Africa (Channing, 2001; Rödel, 2002; Channing and Howell, 2006), it is apparent that the study of East African *Ptychadena* lags behind. So far, the study on the genus has mainly been based on different morphological characters and in merely part vocalization (Perret, 1996; Rödel, 2002; Bwong et al., 2009). Findings from these studies reveal that most species look quite similar in appearance and cryptic diversity occurs in some species. Therefore, more research is necessary in order to obtain suitable characters to differentiate species. This would also be a step towards reviewing the status of East African *Ptychadena* (Bwong et al., 2009).

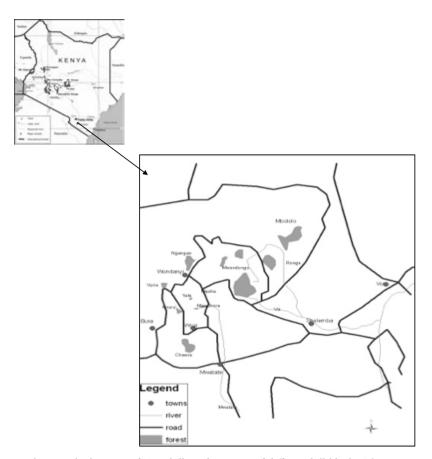
This study aimed at determining the diversity of the genus *Ptychadena* in Taita Hills using morphological characters, and to provide standardized diagnostic schemes for the species identified. Since this study has neither included specimens from elsewhere nor type material, names are only provisionally applied. The results from this study were further compared with findings from other regions where more or less same studies have been conducted, in expectance of comprehensive revisionary action on Ridged Frogs.

## MATERIALS AND METHODS

Taita Hills lie in south eastern Kenya (3°15'S, 3°30'S) and (38°15'E, 38°30'E) in Taita Taveta District of Coast Province (Fig. 1), and form part of the Eastern Arc Mountains (Bennun and Njoroge, 1999). They are at an elevation of 1350–2228 m a.s.l. The climate of the area is under the influence of the innertropical convergence zone, with a bimodal rainfall pattern. The long rains occur in March to June and the short rains in October to December, peaking in April and November respectively. Annual rainfall ranges between 600 mm to 1329 mm per year (Bennun and Njoroge, 1999).

These hills are well known for remarkable fauna and flora with high levels of specific and generic endemism (EAWS, 2005). Therefore, our study on *Ptychadena* species is of special significance, since the hills are Bio-geographically outstanding – i.e. listed as one of the conservation Hot Spots worldwide and displays a high degree of endemism, even in amphibians (Myers et al., 2000; Measey, 2004; Measey and Malonza, 2006).

Opportunistic field sampling, both by day and night (Heyer et al., 1994) was carried out in the field between November 2005 and March 2006. Specimens were collected from 18 different sites in water paddies located at periphery of the different forest blocks, fixed in formalin and pre-



**Fig. 1.** Map showing the location of Taita hills and position of different hill blocks. The specimens were collected from water bodies at forest floors in 18 different sites represented by the names. (Map modified from Bennun and Njoroge, 1999).

served in 70% methanol. Specimens were assigned field numbers and preliminarily identified into two groups based on body colorations and other easily observable non-meristic morphological characters that are distinct. A total of 113 adult *Ptychadena* specimens were used for the analysis, 20 of which had been earlier collections from other researchers (Appendix 1). All the samples were deposited at National Museums of Kenya (NMK)-Department of herpetology, and further used for morphometrical and non-meristic morphological analysis. Sexes were determined through presence (males) versus absence (females) of lateral vocal openings.

Following Guibé and Lamotte, (1957), 1958, 1960), Lamotte (1967), Perret (1979, 1981, 1987, 1994, 1996), Amiet (1989) Largen (1997, 2000), Poynton and Broadley (1985), Rödel (2002), Channing and Howell (2006), 10 morphometrical and 22 non-meristic morphological characters of adults were identified to be useful in defining and diagnosing *Ptychadena* taxa. Using dial callipers,10 body measurements were taken to the nearest 0.1 mm: (1) horizontal eye diameter (ED); (2) distance from anterior corner of eye to posterior nostril (EN); (3) foot length from the proximal edge of the heel to the tip of Toe IV (FL); (4) head length from angle of jaw to tip of snout (HL); (5) head width at broadest (HW); (6) inter-narial distance (ID); (7) distance from tip of snout to anterior

corner of nostril (SN); (8) Snout-vent length (SVL); (9) horizontal tympanum diameter (TD); (10) tibia length (TL). Fifteen non-meristic morphological features were coded as present or absent: (11) Dark bands on posterior face of femur; (12) dark mottling on posterior face of thigh; (13) green or light brown median dorsal band; (14) light bands on posterior face of femur; (15) light tibial line; (16) outermost dorsal ridge coloured whitish; (17) outer metatarsal tubercle; (18) pale triangle on dorsal snout; (19) ridges on lateral sides of body; (20) ridges on the legs; (21) row of tubercles on metatarsus; (22) warts on lateral sides; (23) warts on legs; (24) whitish ring (at least in part) around tympanum; (25) whitish spots on lower lip. Four other non-meristic morphological characters were coded as follows: if dark bands on femurs present, are they continuous or discontinuous from knee to knee (in species definitions below added to (11) if applicable); if outer metatarsal tubercle present is it smaller or larger than inner (in species definitions below added to (17) if applicable); (26) canthus rostralis from eye to nostril concave versus straight; (27) nostrils visible versus invisible from above. Also studied was (28) if the external vocal openings in males are situated above, below or at level of arm insertion: counted were (29) the number of tubercles on Toe IV, (30) the number of dorsal ridges plus (31) the number of incomplete dorsal ridges, and (32) the foot webbing pattern following the manner described by Glaw and Vences (1994).

Diagnoses of species from the Taita Hills covered 32 aspects using characters (1) to (32) listed above. Both variable and non-variable characters were considered in order to provide a standardised diagnostic scheme applicable to the entire genus *Ptychadena*. Information summarized by Frost (2008) was used for the discussion of available names. To compare the variation in morphometry within and between the two species, a Student t-test was used. Further, cluster analysis for the 10 morphometrics and five of the 15 present or absence morphological characters (green or light brown median dorsal band, light tibial line, pale triangle on dorsal snout, ridges on the legs and whitish ring at least in part around tympanum). The five present or absent characters were selected on the basis that a character in consideration is only present in species 1 and not in species 2 and vice versa. Prior to analysis, the combined morphometrics and the morphological data were standardized through log transformation (Anderson et al., 2006). A dissimilarity distance matrix was generated, from which cluster representations of the 113 specimens were constructed. All the analyses were carried out using packages stats and gclus, implemented in R (R Development Core Team, 2010).

#### RESULTS

A standard diagnoses of morphometric and non-meristic morphological characters for each identified species: species 1 and species 2 (L=length: measurements are mean  $\pm$  SD, followed by the range in mm) resulted in to two groupings i.e. species 1 and species 2 described below using 32 characters.

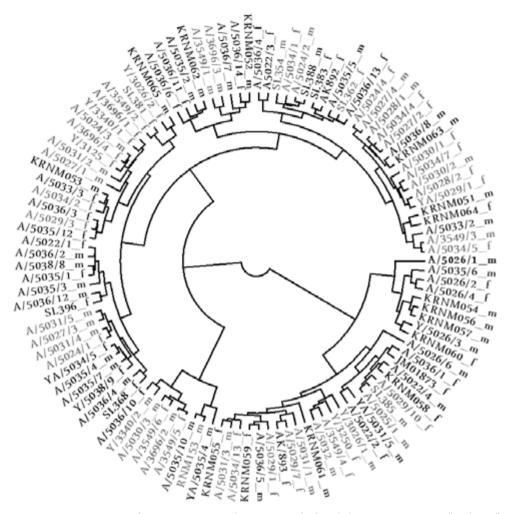
Species 1 "anchietae" diagnosis: (1) ED 4.5  $\pm$  0.6 (5.5–2.6); (2) EN 3.1  $\pm$  0.5 (4.2–2.1); (3) FL 32.6  $\pm$  5.3 (42.0–23.4); (4) HL 12.7  $\pm$  1.7(17.3–9.2); (5) HW 11.4  $\pm$  1.7 (14.8– 8.5); (6) ID 3.1  $\pm$  0.4 (3.9–2.0); (7) SN 3.1  $\pm$  0.5(4.2–2.3); (8) SVL 39.8  $\pm$  6.2 (52.4–27.8, Females > males); (9) TD 3.1  $\pm$  0.5 (2.8–3.9); (10) TL 20.6  $\pm$  3.3 (27.9–14.1); (11) dark bands on posterior femur present, discontinuous from knee to knee; (12) dark mottling on posterior femur absent; (13) light bands on posterior femur present; (14) light or green medial dorsal band absent; (15) canthus rostalis straight; (16) nostril visible from above; (17) pale triangle on snout present; (18) 6–8 dorsal ridges; (19) 2 incomplete ridges; (20) ridges on legs present; (21) ridges on lateral side of body absent; (22) outer dorsal ridge whitish; (23) tibia line not present; (24) outer metatarsal tubercle absent; (25) rows of tubercles absent; (26) warts on legs absent; (27) whitish ring around tympanum present; (28) whitish spots on lower lip present; (29) vocal openings in males positioned below arm insertion; (30) 3 tubercles on toe IV; (31) 1–2 phalanges on toe IV free of web; (32) foot webbing formula 1e (0) 2i/e (0) 3i/e(0) 4i/e(1) 5i(0).

Species 2 "mascareniensis" diagnosis: (1) ED 4.6  $\pm$  0.5 (5.6–2.7); (2) EN 3.1  $\pm$  0.5 (4.2–2.0); (3) FL 34.5  $\pm$  6.4 (48.2–21.3); (4) HL 13.5  $\pm$  2.5 (18.5–8.2); (5) HW 11.8  $\pm$  1.8 (15.1–7.7); (6) ID 2.8  $\pm$  0.5 (4.2–1.7; (7) SN 2.9  $\pm$  0.5 (4.2–1.3); (8) SVL 43.3  $\pm$  7.4 (57.0–26.4, females > males); (9) TD 3.4  $\pm$  0.6 (4.7–2.0); (10) TL 19.9  $\pm$  4.0 (29.3–10.3);). (11) dark and (12) light bands on posterior femur present, continuous or discontinuous from knee to knee; (13) dark mottling on posterior femur absent; (14) canthus rostalis straight; (15) nostril visible from above; (16) pale triangle on snout absent; (17) medial dorsal band present; (18) lateral ridges absent; (19) 6–8 dorsal ridges; (20) if 8, two are short dorsolateral ridges only; (21) outer dorsal ridges whitish; (22) ridges on legs absent; (23) outer metatarsal tubercle absent; (24) row of tubercles absent; (25) tibia line present; (26) warts on lateral sides present or absent; (27) warts on legs absent; (28) whitish spots on lower lips present; (29) vocal openings in males above arm insertion; (30) 3 tubercles on toe 1V; (31) 2 phalanges on toe 1V free of webbing; (32) foot webbing formula 1e (1) 2i/e (1-2) 3i/e (1.5-2.5) 4i/e (2-3) 5i (1-2).

There were a total of 28 females and 24 males of species 1 "anchietae"; and 29 females and 32 males of species 2 "mascareniensis". Some male specimens of species 2 "mascareniensis" showed peculiar aspects in that they lacked certain characters i.e. tibia line and medial dorsal band (ref. numbers: RNM001//A5035/3; RNM002//A5035/2; RNM001//A5035/6; KRNM061//X; KRNM062//X; KRNM065//X from collection sites Wundanyi posta farml for the first three and Shate area for the last three specimens. Two female specimens (ref. SL390//A/5034/5; SL395//A/5055/12 collected from Wundanyi posta farml also showed a similar phenomenon.

Comparisons between morphometry of species 1 "anchietae" female and males revealed high variation in parameters such as EN, FL, HW, SN TD and SVL (P < 0.01). Species 2 "mascareniensis" females and males showed high significant differences in all parameters (P < 0.01) except for ED. There was no significant variation in all the parameters (P = 0.05) between species 1 and 2 females, except for HL that showed a significant variation (P < 0.05). Whereas there was high variation in parameters such as ID, SN, and TL between species 1 and 2 males (P < 0.01), the other parameters did not significantly vary (P = 0.05). Species 1 and 2 females showed high significant differences in parameters such as FL, HW, ID, SN, TD, and TL (P < 0.01); significant variation in HL and SVL (P < 0.05), and no significant difference in ED (P = 0.05). On the other hand, species 1 males and species 2 females highly varied in all parameters (P < 0.01) except for ID and SN that were not significantly variable (P = 0.05).

Cluster representation performed for the 10 morphometrics alone reveal an overlap between the two species (Fig. 2). In contrast, a cluster representation considering five non-meristic morphological characters (Fig. 3) support two distinct clusters of species 1 "*anchietae*" and 2 "*mascareniensis*". Cluster representation of a combination of morphometrics and the five non-meristic morphological characters again advocated for two distinct clusters i.e. of species 1 and 2 (Fig. 4), and the clustering of individual species seems to be influence by sex differences for the two species to some extent.



**Fig. 2.** A representation of 113 specimens in the genus *Ptychadena* belonging to species 1 "*anchietae*" (taxa in grey) and species 2"*mascareniensis*" (taxa in black) using 10 morphometric characters. Two distinct clusters are not formed and the two species are seen to overlap. The letters f and m after the underscore in taxa code number represent sexes for the specimens of the two species respectively.

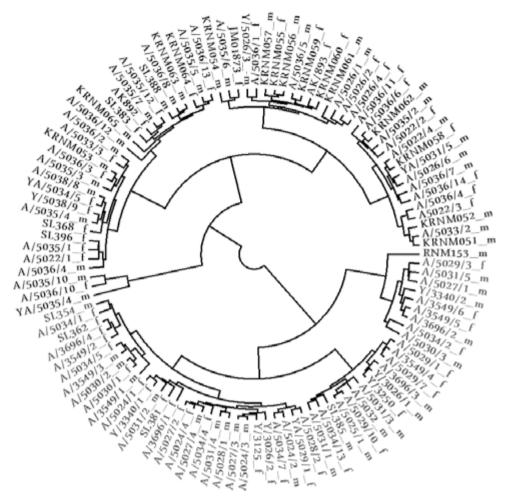
## DISCUSSION

Our results depict that two *Ptychadena* species syntopically occur in Taita Hills. Species 1 is referable to *P. anchietae* (Bocage, 1868 "1867") as it was called by Schick et al. (2005). Originally described from Angola, this ridged frog is suggested to display a distribution from southern Africa north to Eritrea (Channing and Howell, 2006). On the other hand, species 2 "*mascareniensis*" is an unidentified taxon in the species complex behind the name *P. mascareniensis* (Duméril and Bibron, 1841).



**Fig. 3.** A representation of 113 specimens of the frog genus *Ptychadena* for species 1 "*anchietae*"(taxa in grey) and species 2 "*mascareniensis*"(taxa in black) using five present /absent characters. Two distinct clusters are evident, an indication that morphological characters can effectively discriminate the two species. The letters f and m after the underscore in taxa code number represent sexes for the specimens of the two species respectively.

The standard diagnoses of the two species shows that the females are generally larger than their male counterparts, notable either by mere looks at the two species and even as revealed by the morphometrics, with high variation in size notable between the different sexes of species 1 and 2. The two species share and also differ in certain morphological characters. We noted that lateral warts were present in some specimens of species 2 "*mascareniensis*" and absent in others. Species 2 "*mascareniensis*" has a tendency to show variation within its members in non-meristic morphological characters (Bwong et al., 2009). The absence of certain characters e.g. tibia line in some specimens in species 2



**Fig. 4.** A representation of 113 specimens of the frog genus *Ptychadena* for species 1 "*anchietae*"(taxa in grey) and species 2 "*mascareniensis*"(taxa in black) using a combination of 10 morphometric characters and five present /absent characters. Two distinct clusters are evident, an indication that a combination of morphometric and non-meristic morphological characters can effectively discriminate the two species. The letter f and m after the underscore in taxa code number represent sexes for the specimens of the two species respectively.

"*mascareniensis*" that occur in same collection sites may be attributed to breeding between species lacking this characteristic. This aspect is however subject to further research and therefore not featured in the discussion below.

Variation in non-meristic morphological characters between different species plays an important role in entirely discriminating them. Discrimination between species 1"*anchietae*" and species 2 "*mascareniensis*" was mainly achieved on the basis of presence or absence of the morphological character in consideration. Ridges on legs, pale triangle

on the snout and a white ringed tympanum were present only in species 1"anchietae", whereas tibia line and medial dorsal band were only present in species 2 "mascareniensis", except for some few specimens (see results). These characters were noted to be the most informative present or absent non-morphometric characters in discriminating species 1 "anchietae" and 2 "mascareniensis". This is also confirmed by several studies that aimed at discriminating the two species (Stewart, 1967; Channing, 2001; Channing and Howell, 2006; Bwong et al., 2009).

Clustering using morphometry alone was not informative in discriminating the two species, an indication that some of their body proportions are more or less the same between the two species. On the other hand, non-morphometrical morphology and a combined analysis using morphometry and non-morphometrical morphology proved reliable in discriminating the two. For the two species, the differences between sexes tend to influence the clustering of the individual species in the cladogram, notable by presence of same sex specimens in some clades. This may be largely influenced by the morphometric characters. A recent study by Bwong et al. (2009) effectively used combined analysis on this genus but also noted that when a large group of species are in consideration, nonmorphometrical morphology or combining morphometry and non-meristic characters may be ineffective in species discrimination, thus calling for inclusion of other tools such as vocalization and genetic markers.

Snout-vent length is a widely used measure in most morphometric studies on the genus *Ptychadena* and other anurans (Stewart, 1967; Channing and Howell, 2006; Bwong et al., 2009) and can be readily used for a comparative survey. Based on this study, species 1 "anchietae" and 2 "mascareniensis" had SVLs of between 28 mm to 52 mm and 27 mm to 57 mm respectively. Channing and Howell (2006) described *P. anchietae* (species 1) as a medium sized frog, with SVL for males up to 51 mm and females up to 62 mm. On the other hand the females of *P. mascareniensis* (species 2) have SVL of 65 mm and males 53 mm, and their internarial distance is equal to the nostril-snout distance. Recent studies in Kakamega Forest (Bwong et al., 2009) have indicated that the SVL of species 1 "anchietae" range from 22.9 to 60.9 mm, with the females being larger than males while that of Species 2 "mascareniensis" range from, 34.1 to 61.1 mm, with the females being larger than the males. In as much as there is variation on what is the minimum or maximum SVL in different studies, it is clear that all the SVL values discussed above from different studies are within the same range.

In differentiating the two *Ptychadena* species, foot webbing was also informative in that the phalanges in toe 1V free of web were 4ie (1) in species 1 "anchietae" and 4ie (2) in species 2 "mascareniensis"; where (i) represent the Internal and (e) the External part of toe 1V free of webbing. The overall foot web formula for species 1 is (1e (0) 2i/e (0) 3i/e (0) 4i/e (1) 5i (0), which vary from that of species 2; 1e (1) 2i/e (1-2) 3i/e (1.5-2.5) 4i/e (2-3) 5i (1-2), thus differentiating the two species. Similar foot webbing patterns were also noted in the two species from Kakamega forest (Bwong et al., 2009) and Malawi (Stewart, 1967). This shows that foot webbing offers an important character in discriminating not only the two species studied here, but also other species within the genus *Ptychadena* and amphibians as a whole (Stewart, 1967).

This study is unique in that, it is the first of its kind in Taita Hills, and provides a vital basis for future monitoring of *Ptychadena* species. The results depict that two spe-

cies of *Ptychadena* occur in Taita Hills. Our results further propose that morphometrics and non-meristic morphological characters are helpful in species identification and especially when used in combination. Important to mention here is that the possibility of even more species in genus *Ptychadena* occurring in Taita hills cannot be ruled out. Further research has been done to establish the overall diversity of amphibians in these hills, with some species presumed to also belong to the genus *Ptychadena* being discovered at the lowlands. These include *P. schillukorum* & *P. mossambica*, occurring at the bases of Sagalla Hill and Mt. Kasigau (P. Malonza, pers. comm.). However, these have not been subjected to a thorough identification process as is species 1"anchietae" and 2 "mascareniensis" because they are only a few in number.

As shown above, for the identification – not definition – of species, a combination of morphometrical and non-morphometrical morphology characters are suitable. We are optimistic that the existing gaps can be filled because *Ptychadena* species display various external features which should allow for proper diagnoses when applied in a standardized way. Standardised schemes may be especially important as a background of rapid and easy field identifications for conservation purposes in regions where this genus is not yet studied.

#### ACKNOWLEDGEMENTS

We are grateful to the staff of the Department of Herpetology of National Museums of Kenya for logistic support especially P. Malonza and B. Bwong and Wilson. Also to thank is the Forest Department in Taita Hills for allowing us to conduct the research, not forgetting the anonymous referees for their positive comments aimed towards improving this work. This study was funded by Katholischer Akademischier Ausländerdienst, Germany (KAAD), in collaboration with The Biodiversity Monitering Transect Analysis in Africa (BIOTA E08), - part of the BIOLOG campaign under the Federal Ministry of Education and Research (Germany), to whom we are thankful.

#### REFERENCES

- Anderson, M.J., Ellingsen, K.E., McArdle, B.H. (2006): Multivariate dispersion as a measure of beta diversity. Ecol. Lett. 9: 683-693.
- Amiet, J.L. (1989): Quelques aspects de la biologie des amphibians anoures du Cameroun. Ann. Biol. 28: 73-136.
- Bennun, L., Njoroge, P. (1999): Important Bird Areas in Kenya. East Africa Natural History Society.
- Bwong, B. A., Chira, R., Schick, S., Veith, M., Lötters, S. (2009): Diversity of Ridged Frogs (*Ptychadena*) in the easternmost remnant of the Guineo-Congolian rain forest: morphology, bioacoustics, molecular markers. Salamandra 45: 129-146.
- Channing, A. (2001): Amphibians of Central and Southern Africa. Cornell University Press, Ithaca and London.
- Channing, A., Howell, K.M. (2006): Amphibians of East Africa. Cornell University Press, Ithaca and London.

- EAWS (2005): The Taita Biodiversity Conservation Project. [http:// www.easternarc.org/ html/bio.html.]
- Frost, D.R. (2008): Amphibians species of the world: an online reference. Version 5.2. New York: Am. Mus. Nat. Hist. [15 July 2008]. [http://research.amnh.org/herpetology/ amphibia/index.php].
- Glaw, F., Vences, M.A. (1994): Fieldguide to the amphibians and reptiles of Madagascar. Vences & Glaw Verlag GbR, Köln.
- Guibe, J., Lamotte, M. (1957): Rèvision systèmatique des *Ptychadena* (Batraciens, Anoures, Ranidés) d'Afrique occidentale. Bull. Inst. Fondam. Afr. Noire **19**: 937-1003.
- Guibé, J., Lamotte, M. (1958): Les *Ptychadena* (Batraciens, Ranidés) du Cameroun. Bull. Inst. Fondam. Afr. Noire **20**: 1448-1463.
- Guibé, J., Lamotte, M. (1960): Deux espèces affines de batraciens africains longtemps confondues: *Ptychadena oxyrhynchus* (Smith) et *Pt. abyssinica* (Peters). Bull. Mus. Nat. Hist. Nat. **32**: 380-391.
- Heyer, W.R., Rand, A.S., Da Cruz, C.A.G., Peixoto, O.L., Nelson, C.E. (1994): Frogs of Boracéia. Arq. Zool. 31: 237-410.
- Lamotte, M. (1967): Le problème des *Ptychadena* (Fam. Ranidae) du groupe *mascareniensis* dansl'ouest africain. Bull. Mus. Natl. Hist. Nat. **39**: 647-656.
- Largen, J.M. (1997): Two new species of *Ptychadena* Boulenger 1917 (Amphibia Anura Ranidae) from Ethiopia, with observations on the genus recorded from this country and a tentative key for their identification. Trop. Zool. **10**: 223-246.
- Largen, J.M. (2000): A new species of *Ptychadena* Boulenger 1917 from Ethiopia (Amphibia Anura Ranidae). Trop. Zool. **13**: 171-178.
- Measey, J. (2004): Are Caecilians rare? An East African Perspective. J. East Afr. Nat. Hist. Soc. 93: 1-21.
- Measy, G.J., Malonza, P.K. (2006): Life history of an African caecilian: *Boulengerula taitanus* Loveridge 1935 (Amphibia: Gymnophiona: Caecilidae). Trop. Zool. **18**: 49-66.
- Myers, N., Mittermeir, R.A., Mittermeir, C.G., Kent, J. (2000): Biodiversity Hotspots for Conservation Priorities. Nature **403**: 853-858.
- Perret, J.-L. (1979): Remarques et mise au point sur quelques espèces de *Ptychadena* (Amphibia, Ranidae). Bull. Soc. Neuchêt. Sci. Nat. **102**: 5-21.
- Perret, J.-L. (1981): Le statut de *Ptychadena schubotzi* (Sternfeld) (Amphibia, Ranidae). Bull. Soc. Neuchêt. Sci. Nat. **102**: 53-57.
- Perret, J.-L. (1987): À propos de *Ptychadena schillukorum* (Werner, 1907) (Anura, Ranidae). Bull. Soc. Neuchêt. Sci Nat. **110**: 63-70.
- Perret, J.-L. (1994): Description de *Ptychadena largeni* n. sp. (Anura, Ranidae) d'Èthiopie. Bull. Mus. Hist. Nat. Genève, **117**: 67-77.
- Perret, J.-L. (1996): Une nouvelle espèce du genre *Ptychadena* (Anura, Ranidae) du Kenya. Rev. Suisse Zool. **103**: 757-766.
- Poynton, C.J., Broadley, D.G. (1985): Amphibia Zambesiaca 2. Ranidae. Ann. Natal Mus. 27: 115-181.
- Poynton, J.C. (1999): Distribution of amphibians in Sub-Saharan Africa, Madagascar, and Seychelles. In: Patterns of distribution of amphibians. A global perspective, pp. 483-539, W.E., Duellman, Ed. University Press, Baltmore & London.

- R Development Core Team (2010): R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL http://www.R-project.org.
- Rödel, M.-O. (2002): Herpetofauna of West Africa amphibians of the West African savanna. Chimaira, Frankfurt.
- Schick, S., Veith, M., Lötters, S. (2005): Geographical range patterns of amphibians from the Kakamega Forest, Kenya. Afr. J. Herpetol. 54: 185-190.
- Stewart, M.M. (1967): Amphibians of Malawi. Albany. State University of New York Press.

# APPENDIX 1

113 frog species from the genus *Ptychadena* used for the analysis in this study. Letters X represent missing information, yet to be allocated. More details on collectors, collection dates and a complete numbering can be obtained from the National Museums of Kenya, Department of herpetology.

Locality	Field Number	NMK catalogue Number	Supposed name	Collectors
Kasigau	x	Y/3026/1	P. anchietae	-
Kasigau	х	Y/3026/2	P. anchietae	-
Mbololo	х	A/3549/1	P. anchietae	-
Mbololo	Х	A/3549/2	P. anchietae	-
Mbololo	Х	A/3549/3	P. anchietae	-
Mbololo	Х	A/3549/4	P. anchietae	-
Mbololo	Х	A/3549/5	P. anchietae	-
Mbololo	х	A/3549/6	P. anchietae	-
Fururu	Х	Y3250	P. anchietae	-
Kasigau	Х	Y3340/2	P. anchietae	-
Kasigau	Х	Y3340/2	P. anchietae	-
Mbololo	Х	A/3696/1	P. anchietae	-
Mbololo	Х	A/3696/2	P. anchietae	-
chomboke area	YSL201	A/5029/1	P. anchietae	Wilson
Ngarenyi dam	YSL206	A/5029/3	P. anchietae	Wilson
mchome area	YSL203	A/5029/7	P. anchietae	Wilson
chombeke area	YSL066	A/5029/10	P. anchietae	Wilson
Kasigau	Х	Y/3125	P. anchietae	P. Malonza
Mbololo	Х	Y/3696/3	P. anchietae	P. Malonza
Mbololo	Х	Y/3696/4	P. anchietae	P. Malonza
Irido	RNM10	A/5024/1	P. anchietae	Wilson
Irido	RNM011	A/5024/2	P. anchietae	Wilson
Irido	RNM012	A/5024/3	P. anchietae	Wilson
Irido	RNM013	A/5024/4	P. anchietae	Wilson
Wasinyi	SL398	A/5028/1	P. anchietae	Wilson
Wasinyi	SL387	A/5028/2	P. anchietae	Wilson

Locality	Field Number	NMK catalogue Number	Supposed name	Collectors
Wasinyi	SL389	A/5029/1	P. anchietae	Wilson
wundanyi stadium	SL361	A/5034/1	P. anchietae	R. Ng'endo
wundanyi stadium	SL362	Х	P. anchietae	R. Ng'endo
wundanyi stadium	SL363	A/5034/13	P. anchietae	R. Ng'endo
wundanyi stadium	SL364	A/5034/7	P. anchietae	R. Ng'endo
wundanyi stadium	SL381	Х	P. anchietae	R. Ng'endo
wundanyi stadium	SL382	A/5034/2	P. anchietae	R. Ng'endo
wundanyi stadium	SL284	A/5034/4	P. anchietae	R. Ng'endo
wundanyi stadium	SL380	A/5034/5	P. anchietae	R. Ng'endo
wundanyiposta stadium	RNM007	A/5027/4	P. anchietae	R. Ng'endo
wundanyiposta stadium	RNM008	A/5027/2	P. anchietae	R. Ng'endo
wundanyiposta stadium	RNM009	A/5027/3	P. anchietae	R. Ng'endo
Serienyi	SL354	Х	P. anchietae	R. Ng'endo
Serienyi	SL385	Х	P. anchietae	R. Ng'endo
mwatate marshes	SL351	A/5031/5	P. anchietae	R. Ng'endo
mwatate marshes	SL352	A/5031/1	P. anchietae	R. Ng'endo
Mwatate marshes	SL353	A/5031/3	P. anchietae	R. Ng'endo
mwatate marshes	SL356	A/5031/4	P. anchietae	R. Ng'endo
mwatate marshes	SL357	A/5031/2	P. anchietae	R. Ng'endo
mwatate marshes	SL358	A/5032	P. anchietae	R. Ng'endo
nwatate marshes	SL368	A/5025/1	P. anchietae	R. Ng'endo
karangar.falls	RNM014	A/5030/1	P. anchietae	Wilson
karangar.falls	RNM015	A/5030/3	P. anchietae	Wilson
karangar.falls	RNM017	A/5030/2	P. anchietae	Wilson
Wund.opp. bank	RNM153	х	P. anchietae	R. Ng'endo
Wund. Opp. bank	RNM036	A/5027/1	P. anchietae	R. Ng'endo
wundanyi posta farml	RNM001	A/5035/3	P. mascareniensis	R. Ng'endo
wundanyi posta farml	RNM002	A/5035/2	P. mascareniensis	R. Ng'endo
wundanyi posta farml	RNM003	A/5035/6	P. mascareniensis	R. Ng'endo
wundanyi posta farml	RNM004	A/5035/4	P. mascareniensis	R. Ng'endo
wundanyi posta farml	RNM005	A/5035/1	P. mascareniensis	R. Ng'endo
wundanyi posta farml	RNM006	A/5035/5	P. mascareniensis	R. Ng'endo
wundanyi stadium tren	SL396	Х	P. mascareniensis	R. Ng'endo
wundanyi posta farml	SL390	A/5034/5	P. mascareniensis	R. Ng'endo
wundanyi posta farml	SL391	A/5038/8	P. mascareniensis	Wilson
wundanyi posta farml	SL393	A/5035/7	P. mascareniensis	Wilson
wundanyi posta farml	SL394	Y/5038/9	P. mascareniensis	Wilson
wundanyi posta farml	SL395	A/5035/12	P. mascareniensis	Wilson
nwatate dam	SL355	A/5031/5	P. mascareniensis	P. Malonza
mwatate dam	SL359	A/5026/2	P. mascareniensis	R. Ng'endo
mwatate dam	SL360	A/5026/4	P. mascareniensis	R. Ng'endo
mwatate dam	SL365	Y/5026/3	P. mascareniensis	Wilson
mwatate dam	SL366	A/5026/1	P. mascareniensis	Wilson

Locality	Field Number	NMK catalogue Number	Supposed name	Collectors
mwatate dam	SL367	A/5026/6	P. mascareniensis	Wilson
Irido	SL369	A/5022/4	P. mascareniensis	Wilson
wundanyi stadium	SL370	A/5036/11	P. mascareniensis	Wilson
wundanyi stadium	SL371	A/5036/5	P. mascareniensis	Wilson
wundanyi stadium	SL372	A/5036/1	P. mascareniensis	Wilson
wundanyi stadium	SL373	A/5036/12	P. mascareniensis	R. Ng'endo
wundanyi stadium	SL374	A/5036/7	P. mascareniensis	R. Ng'endo
wundanyi stadium	SL375	A/5036/8	P. mascareniensis	R. Ng'endo
wundanyi stadium	SL376	A/5036/3	P. mascareniensis	R. Ng'endo
wundanyi stadium	SL377	A/5036/6	P. mascareniensis	R. Ng'endo
wundanyi stadium	SL378	A/5036/4	P. mascareniensis	R. Ng'endo
wundanyi stadium	SL379	A/5036/10	P. mascareniensis	R. Ng'endo
wundanyi stadium	SL383	Х	P. mascareniensis	R. Ng'endo
wundanyi stadium	SL399	A/5036/13	P. mascareniensis	R. Ng'endo
wundanyi stadium	SL397	A/5036/14	P. mascareniensis	R. Ng'endo
Taita Hills	JM01873	Х	P. mascareniensis	P. Malonza
Faita Hills	AK/893	Х	P. mascareniensis	P. Malonza
Faita Hills	AK892	Х	P. mascareniensis	P. Malonza
Faita Hills	RNM030	A/5033/3	P. mascareniensis	Wilson
Faita Hills	RNM024	A/5033/2	P. mascareniensis	Wilson
Faita hills	SL388	Х	P. mascareniensis	Wilson
Wund. Opp. Bank	SL368	Х	P. mascareniensis	Wilson
Wundanyi Field	RNM072	A/5036/2	P. mascareniensis	Wilson
Wasinyi	RNM078	A/5036/4	P. mascareniensis	Wilson
Wund.opp. Bank	SL392	A/5035/10	P. mascareniensis	Wilson
Wund.opp. bank	RNM122	A/5035/4	P. mascareniensis	Wilson
rido	RNM048	A/5022/1	P. mascareniensis	Wilson
shate area	KRNM049	A/5022/2	P. mascareniensis	R. Ng'endo
nchome area	KRNM050	A5022/3	P. mascareniensis	R. Ng'endo
shate area	KRNM051	Х	P. mascareniensis	R. Ng'endo
nchome area	KRNM052	Х	P. mascareniensis	R. Ng'endo
Ngalenyi dam	KRNM053	Х	P. mascareniensis	R. Ng'endo
nchome area	KRNM054	Х	P. mascareniensis	R. Ngendo
nchome area	KRNM055	Х	P. mascareniensis	R. Ng'endo
nchome area	KRNM056	Х	P. mascareniensis	R. Ngendo
nchome area	KRNM057	Х	P. mascareniensis	R. Ngendo
Madoghodo area	KRNM058	Х	P. mascareniensis	Wilson
hate area	KRNM059	Х	P. mascareniensis	Wilson
shate area	KRNM060	Х	P. mascareniensis	Wilson
shate area	KRNM061	Х	P. mascareniensis	Wilson
shate area	KRNM062	Х	P. mascareniensis	Wilson
Tambaru area	KRNM063	Х	P. mascareniensis	Wilson
Tambaru area	KRNM064	Х	P. mascareniensis	Wilson
shate area	KRNM065	Х	P. mascareniensis	Wilson