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Contribution to the systematics and zoogeography of the East-African *Acomys spinosissimus* Peters 1852 species complex and the description of two new species (Rodentia: Muridae)

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[6]*This paper is dedicated to Walter Verheyen and Marco Corti, both passed away before this study was completed*

Abstract

We revised the taxonomic status of the putative *Acomys spinosissimus* complex based on the comparative study of specimen collections from Tanzania, Zambia, Zimbabwe, Mozambique, DR Congo and South Africa, by means of analysis of external morphology, craniometry, enzymes, mitochondrial DNA sequences and karyology. Our results confirm that *A. spinosissimus* represents a complex of species with seemingly non-overlapping distribution ranges. The distribution range of *A. spinosissimus* appears to be restricted between the Zambesi and Limpopo Rivers, while the reinstated *A. selousi* (that includes *A. transvaalensis*) occurs further to the South (*i.e.* northern limit seemingly just north of the Limpopo River). The investigated populations north of the Zambezi River are morphologically and genetically distinct from *A. spinosissimus* and *A. selousi*. Based on this evidence, we described *Acomys muzei* **sp. nov.** and *Acomys ngurui* **sp. nov.,** each one occurring separately along one side of the Eastern Arc Mountains. Finally, we lacked sufficient information to describe a third new species from the area north of the Zambesi River.

Key words: *Acomys spinosissimus*, taxonomy, cytochrome *b*, craniometry, morphology, enzymes

Introduction

Acomys or spiny mice are widespread throughout all of Africa, the near and Middle East, and some Mediterranean islands (Corbet 1978). This genus has been the subject of several molecular and morphological analyses due to their basal phylogenetic position within the Muridae (Sarich 1985; Denys *et al.* 1992 a, b; Chevret *et al.* 1993; Chevret & Hänni 1994; Hänni *et al.* 1995).

Since 1939, when G. M. Allen listed the recorded African species of *Acomys* in his "Checklist of African Mammals" (Allen 1939), African members of the genus have been the subject of a series of studies that resulted in different species lists (*e.g.* Setzer 1975). The most recent annotated checklist recognizes 19 species, but as for many other African rodent genera, their taxonomy requires further study (Musser & Carleton 2005).

To date, the information available for documenting species-limits in *Acomys* includes chromosomal studies and reviews (Matthey 1954, 1956, 1963, 1965a,b, 1968; Volobouev *et al.*1991; Sokolov *et al.* 1992, 1993; Denys *et al.* 1994; Volobouev *et al.* 2007); studies of cranial characters and morphology of molars (Petter 1983; Denys *et al.* 1994), morphology of spermatozoa (Baskevich & Lavrenchenko 1995) and allozyme and mtDNA studies of species-assemblages (Janecek *et al.* 1991; Barome *et al.* 1998, 2000, 2001). The significant contribution by Dippenaar

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and Rautenbach (1986) demonstrated how a combination of morphometric and karyological data could be used to address taxonomic problems in murids (cited in: Musser & Carleton 1993, 2005; Denys *et al.* 1994).

We applied a similar combined approach to identify the samples usually included in *A. spinosissimus* Peters, 1852. Based on the current species status, *A. spinosissimus* is found throughout North East and East Central Tanzania, South East Democratic Republic of the Congo, Zambia, Malawi, Zimbabwe, East Botswana, Central Mozambique, and North and North Eastern South Africa, as it included *A. selousi* De Winton, 1896 and *A. transvaalensis* Roberts, 1926 that were formerly recognized as species (Musser & Carleton 1993, 2005).

Recent studies on Tanzanian and Malawian *A. spinosissimus* compared with samples from farther south within the geographic range demonstrated the monophyly of all samples, but also the distinctiveness of this species compared with *A. subspinosus* and the species occurring farther north (*A. wilsoni, A. russatus, A. ignitus, A. airensis, A. cahirinus*, and *A. dimidiatus*; Barome *et al.* 2000, 2001). The conflicting cytochrome *b*, cytogenetic and morphometric results indicated that the monophyletic *A. spinosissimus* clade could contain two sibling species, a hypothesis that can only be tested by the inclusion of a true *selousi* specimen from the type locality.

The most compelling reason to reinvestigate the taxonomic status of all populations currently assigned to *A. spinosissimus* is the amount of chromosomal variation that has been observed within this taxon; in particular the unusual karyotype of *Acomys* specimens from Tanzania and Malawi that was identified as *A. selousi* (Matthey 1965). This karyotype with a single and exceptionally large X-chromosome in both males and females was seemingly identical to that reported in samples from Berega, Tanzania (Barome *et al.* 2001; Corti *et al.* 2005).

The present study was based on collections of *Acomys* from Tanzania, Zambia, Zimbabwe, Mozambique, DR Congo and South Africa, which we used to unravel the taxonomy and zoogeography of the Tanzanian members of the putative *A. spinosissimus* species complex. Our approach included the comparative study of their external morphology, craniometry, enzyme electrophoresis, mitochondrial DNA sequences and karyology. Whenever possible we included relevant type specimens (*A. selousi* and *A. spinossisimus*) and freshly collected representatives from localities close to the type locality for holotypes that were not available for this study (*A. transvaalensis*, for example). We did not attempt to obtain sequences from types, to avoid damaging the valuable type specimens.

Material and methods

The specimens. This study was mainly based on *A. spinosissimus* specimens collected in Tanzania by the research group on African Rodents (Department of Biology, University of Antwerp, Belgium) and on collections of the Royal Museum for Central Africa (Tervuren, Belgium) (Fig.1, Appendix 1).

We collected 469 specimens using Sherman live traps, Museum special and Victor snap traps. Some specimens (N=22) were transported alive to the laboratories of Sokoine University of Agriculture (Morogoro, Tanzania) for karyotyping, the rest were dissected in the field to sample kidney, liver and testes for enzymes (N=52) and mtDNA (N=44) analyses. Tissues were immediately fixed in liquid nitrogen, and the majority of the specimens were directly preserved in formalin. We measured head and body length (HB); tail-length (TL), hind foot length (-claw) (HF), ear length (EL) and weight (W) on freshly killed specimens as described elsewhere (Van der Straeten 1975).

The collected ecological data indicated that the *A. spinosissimus* species complex was typical for the miombo —woodland and mboga (Swahili)— grasslands, but never at altitudes higher than 1700 meter above sea level. In bush and grassland it was often found in the immediate vicinity of rocky outcrops and ‘koppies’.

We listed the measured specimens and numbers of specimens for each OTU (Operational Taxonomic Unit) in Appendix 1, along with their sex and age. Those individuals that were used for karyotyping, DNA sequencing or enzyme electrophoresis were also identified. The sampling localities (with coordinates) were listed by OTU in Table 1.

We complemented the bulk of our material from Tanzania with selected specimens from the Natural History Museum (London) [BMNH], the American Museum of Natural History (New York) [AMNH], the National Museum of Natural History (Washington, D. C.), [USNM], the Field Museum (Chicago) [FMNH], the Museum of Vertebrate Zoology (University of California, Berkeley) [UCB], the Transvaal Museum (Pretoria) [TM] and the Zoologisches Museum der Humboldt-Universität (Berlin) [ZMB]. The majority of this material represents South African and Mozambican samples of *A. spinosissimus*, as well as type specimens, needed to evaluate the taxonomic status of our Tanzanian samples.

TABLE 1. Localities and geographical coordinates of the specimens studied, as listed in Table 1. (Notes: KILWA is situated in Mtwara province; TETTE (possibly the same locality then BUIO) is the type locality for *A. spinosissimus*; ESSEX FARM is the type locality for *A. selousi;* ZOUTPANSBERG is the type locality for *A. transvaalensis*).

(M)OTU	LOCALITY		COORDINATES
10	DAKAWA	a	06.26S-37.34E
11	AMANI	b	05.06S-38.38E
11	BEREGA	c	06.14S-37.10E
11	KIGURUNYEMBE	d	06.50S-37.39E
11	MBETE	d	06.52S-37.41E
11	MGETA	d	07.03S-37.35E
11	MIKESE MT	d	06.49S-37.51E
11	MKUNDI	d	06.42S-37.39E
11	MLALI	d	06.17S-36.45E
11	MORNINGSIDE	d	06.53S-37.40E
11	MOROGORO	d	06.50S-37.39E
11	MSIMBA	e	07.26S-36.57E
11	NGURU YA NDEGE	d	06.42S-37.37E
11	SANGASANGA	d	06.54S-37.36E
11	ULANGA	f	08.10S-36.57E
11	ULAYA	g	07.02S-36.55E
20	CHINGULUNGULU	h	10.44S-38.33E
20	KILWA	i	09.00S-39.30E
20	MNARA	j	10.07S-39.24E
20	MNIMA	k	10.37S-39.13E
20	MUHUWESI	l	10.54S-37.29E
20	NAKAHUGA	m	10.39S-35.27E
20	PERAMIHO	m	10.38S-35.28E
20	RUANGWA	n	10.04S-38.57E
30	INALA - TABORA	o	05.25S-32.49E
30	KANYELELE	p	02.38S-33.08E
30	KONDOA	q	04.54S-35.46E
30	MAGANGWE	r	07.45S-34.15E
30	MANYONI	s	05.46S-34.50E
30	MATONGOLO	t	05.46S-36.28E
30	MSEMBE	w	07,38S-34,55E
30	MTOWISA	u	08.04S-31.55E
30	MUZE	v	07.45S-31.34E
30	MWANZA	p	02.30S-32.54E
30	MWEYEMBE	w	07.38S-34.55E
30	ZOISSA	t	05.40S-36.25E

(M)OTU	LOCALITY	COORDINATES
31	LUBUMBASHI	11.40S-27.28E
31	MUNAMA	11.47S-27.59E
40	CHIUTA	15.29S-33.20E
40	FINGUE	15.15S-31.48E
40	MUCANHA river	14.58S-31.23E
41	LUANSHYA	13.09S-28.24E
50	VILA PAIVA DE ANDRADA	18.41S-34.04E
60	ASSEN	25.20S-27.25E
60	ELLISRAS	23.45S-27.50E
60	LETSITELE	23.50S-30.30E
60	LEYDSDORP	23.59S-30.30E
60	MARKEN	23.50S-28.30E
60	PAFURI	22.27S-31.21E
60	POTGIETERSRUS	24.15S-28.55E
60	ROOSSENEKAL	25.14S-29.53E
60	RUSTENBURG	25.40S-27.15E
60	SHAMDUNGILA	23.59S-30.30E
60	THABAZIMBI	24.41S-27.21E
60	VAALWATER	24.20S-28.05E
Sequences		
S1	LIWONDE	15.07S-35.23E
S2	MBUGANI - CHUNYA	08.31S-33.30E
S3	MORRUMBALA	17.20S-35.35E
S4	MUTARARA	17.27S-35.04E
S5	MWENERONDO	09.54S-33.55E
S6	PAFURI	22.27S-31.21E
Types		
T1	TETTE	16.10S-33.35E
T2	BUIO	? = Tette
T3	ESSEX FARM	20.21S-29.01E
T4	ZOUTPANSBERG (New Gate)	22.45S-30.00E

In total, we studied 471 specimens from Tanzania, DR Congo, Malawi, Mozambique, South Africa and Zambia, including the two relevant holotypes (*A. spinosissmus* from Buio [ZMB 1711] and *A. selousi* from Essexvale [BMNH 97.1.4.44]) for which the information (type-localities, geographical coordinates, skull-measurements as measured by us) is summarized in Table 2. The type localities are shown in Figure 1.

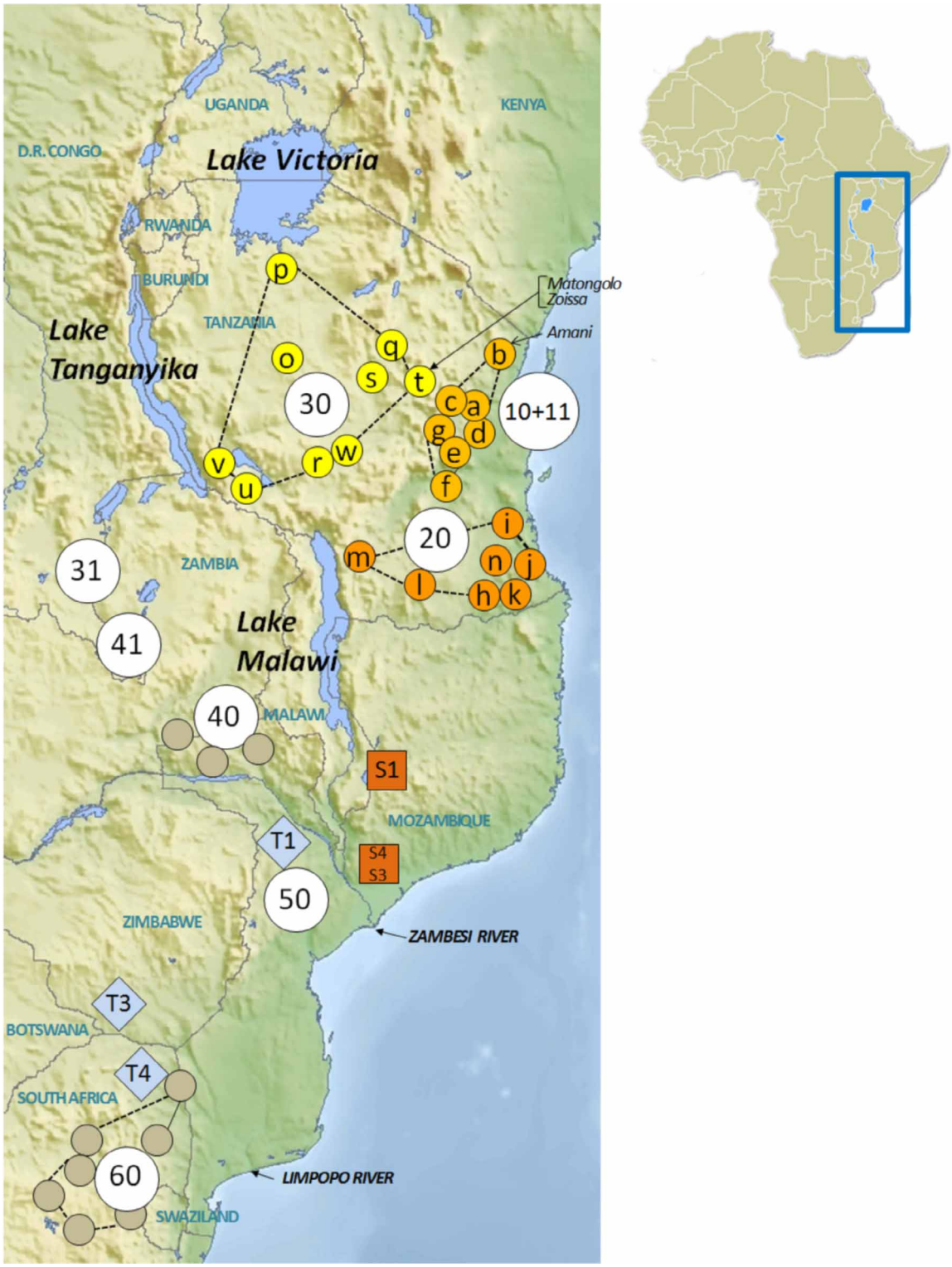


FIGURE 1. East Africa with geographical distribution of the OTU's, MOTU's and relevant type localities of the *A. spinosissimus* species complex. The composition of the individual OTU's are given in Appendix 1; the geographical coordinates of the sampling localities in Table 1; the type specimens (including our own craniometric measurements) in Table 2. Numbers refer to OTU's, Sx refer to additional sequenced specimens and Tx refer to type localities (see Appendix 1 for more details).

TABLE 2. Measurements of the type and paratype specimens of *A. selousi* and *A. spinosissimus*.

LOCALITY	MUSEUM + REG.NR	M1	M2	M3	M4	M5	M6	M7	M8	M9	M10	M11	M12
spinosissimus type													
BUIO	ZMB 1711	-	-	-	-	-	-	-	4.62	-	2.49	4.42	5.56
selousi type													
ESSEXVALE	BMNH 97.1.4.44	-	23.05	19.10	12.30	5.45	6.25	6.85	4.55	12.55	2.70	4.15	5.60

LOCALITY	MUSEUM + REG.NR	M13	M14	M15	M16	M17	M19	M20	M21	M22	M23	M24
spinosissimus type												
BUIO (ZMB)	ZMB 1711	1.21	3.28	2.48	10.22	3.86	4.16	-	1.38	-	3.79	-
selousi type												
ESSEXVALE	BMNH 97.1.4.44	1.35	2.80	2.60	9.70	3.55	4.90	11.30	1.35	5.25	3.90	6.45

Access to the study material: All the specimens used in this study that were collected by the authors and their collaborators, including the measurements can be viewed and accessed at http://projects.biodiversity.be/africanrodentia/ (Terryn *et al.* 2007). All the new sequences were submitted to GenBank under accession numbers JN247671-JN247730.

Morphological methods. The skulls were individually examined to evaluate intraspecific craniological and dental variations. Subsequently, representative specimens were directly compared to the relevant type specimens. The possible influence of age on skull size and shape was evaluated by grouping all the available crania into approximate age classes inferred from the amount of wear on the upper molars (Fig.2). Skulls with M3 not fully erupted (class 0) were excluded from our analyses.

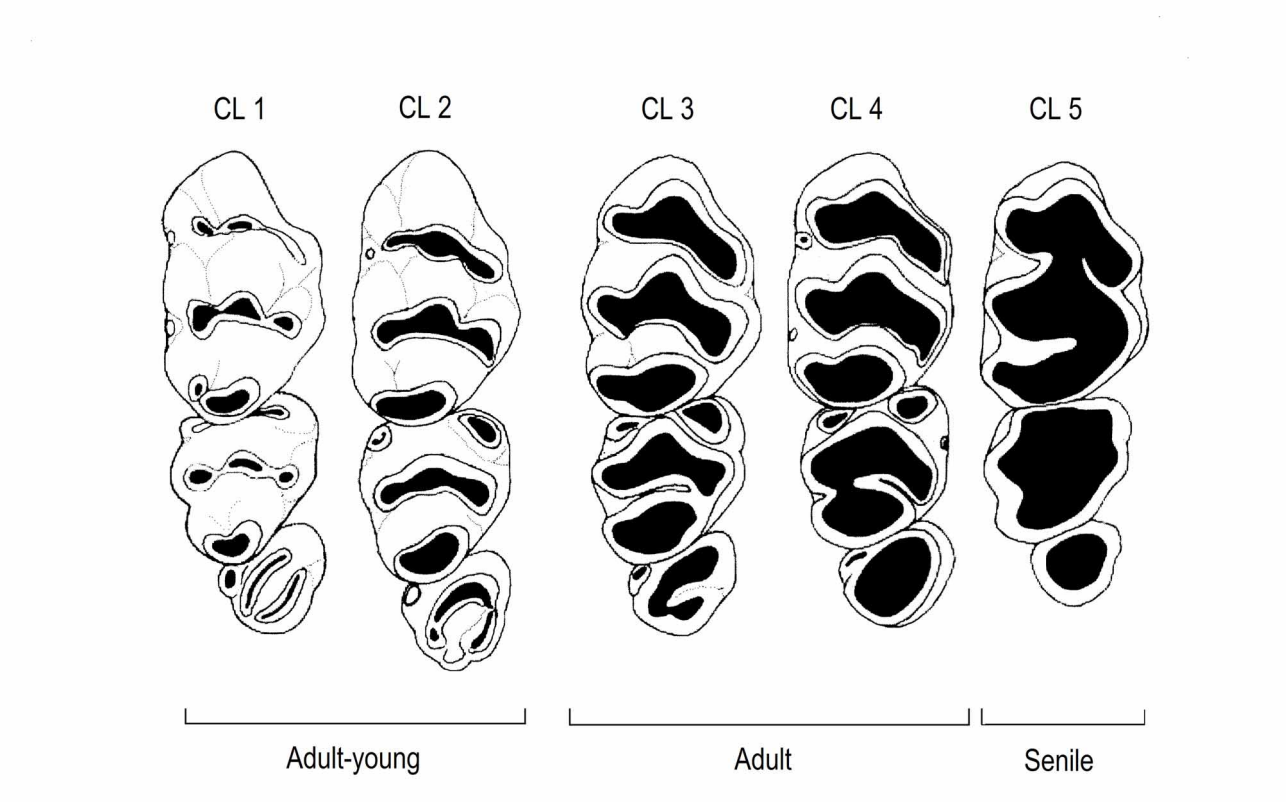


FIGURE 2. The tooth-wear classes distinguished in Tanzanian *A. spinosissimus*. Cl. 1: all teeth fully erupted; wear minimal; M1 and M2: dentine of 2nd cusp-row not continuous. (RMCA 96.036-M-4828), Cl. 2: light wear; M1 and M2: dentine of the 2nd cusp-row continuous, but width of dentine-surface of t5, enamel-rim of t5. (RMCA 96.036-M-4742), Cl. 3: wear obvious but not extensive; M1: dentine-surface of t5 > than enamel-rim of t5; M2: dentine-surface of 1st and 2nd row not continuous. (RMCA96.036-M-4794), Cl. 4: wear extensive; M1: much flattened cusps but still 3 separate dentine-rows; M2: 1st and 2nd dentine-rows communicating. (RMCA 96.036-M-4792), Cl. 5: wear severe; M1: very heavily eroded and at least two dentine-rows continuous; M2: continuous dentine wear surface. (RMCA 96.036-M-4856).

Finally specimens from different sampling localities were grouped into Operational Taxonomical Units (OTU's) to create a working series for further analyses. The OTU numbers have no other significance then that they reflect the order in which they were composed.

Morphometric analyses. We used twenty-three cranial and five external measurements (Fig.3; acronyms as in

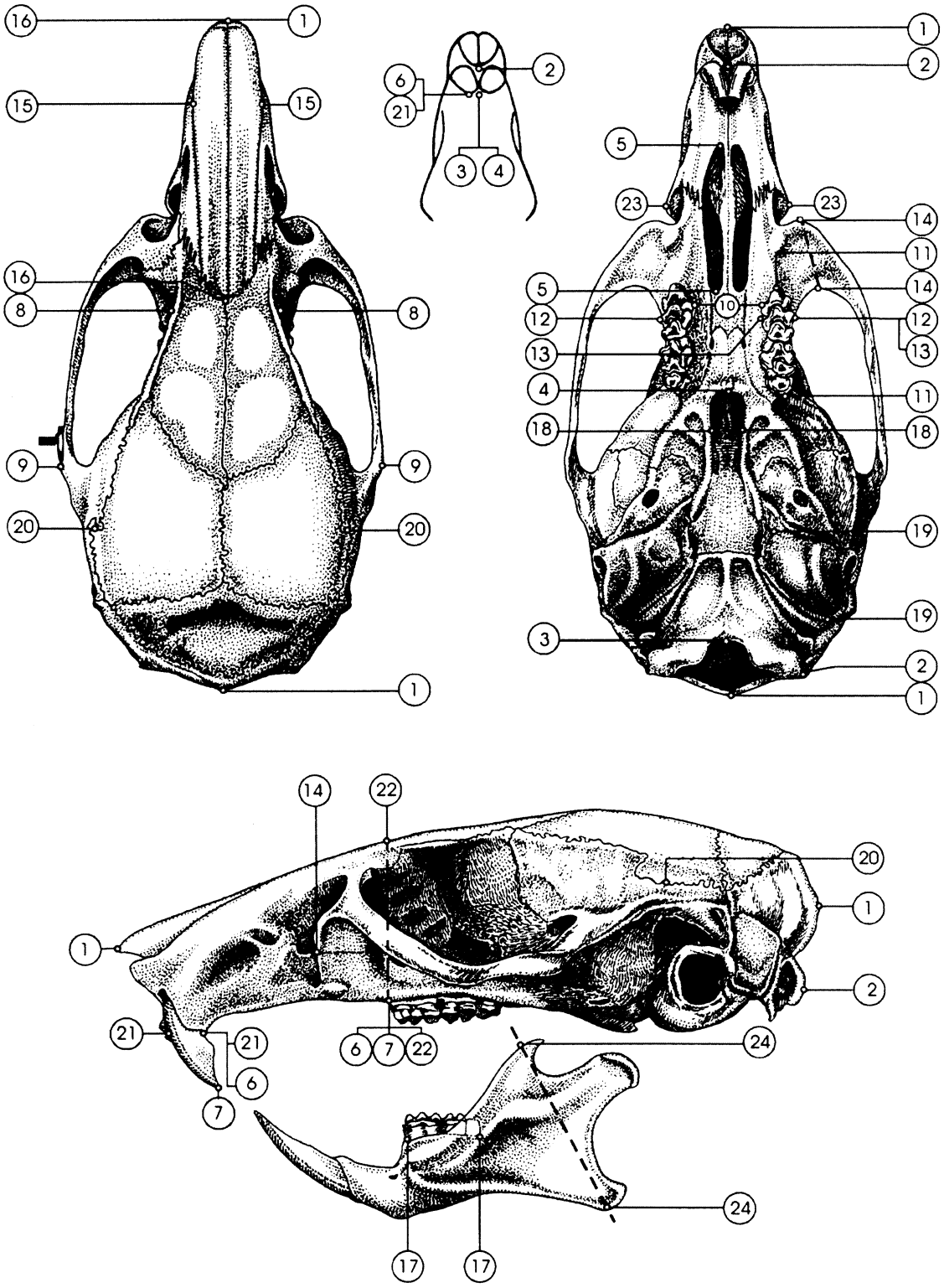


FIGURE 3. Numbers refer to the position of the two points of the caliper when taking the cranial measures . Measure 18 could not be taken for a sufficient number of specimens to be included in our analyses (specimen drawn: *Pelomys* RMCA 12924, Lubumbashi, DR Congo).

Verheyen *et al.* 1996). Craniometrical values were taken with digital calipers and recorded with a precision of 0.05 mm. The following cranial measures were taken : M1 (GRLS), greatest length of skull; M2 (PRCO), condylobasal length; M3 (HEBA), henselion-basion; M4 (HEPA), henselion-palation; M5 (PAFL), length of palatal foramen; M6 (DIA1), length of diastema; M7 (DIA2), distance between alveolus M1 and cutting edge of upper incisor; M8 (INTE), smallest interorbital breadth; M9 (ZYGO), zygomatic breadth; M10 (PALA), smallest palatal breadth; M11 (UPTE), length of upper cheekteeth; M12 (UPDA), breadth of upper dental arch; M13 (M1 BR), greatest breadth of first upper molar; M14 (ZYPL), smallest breadth of zygomatic plate; M15 (BNAS), greatest breadth of nasals; M16 (LNAS), greatest length of nasals; M17 (LOTE), length of mandibular teeth; M19 (BULL), length of auditory bulla; M20 (BRCA), greatest breadth of braincase; M21 (DINC), depth of upper incisor; M22 (ROHE), mediosagittal projection of rostrum height; M23 (ROBR), greatest rostrum breadth; M24 (PCPA), distance between coronoid and angular processes.

Basic statistics, Student's t-tests, 2-way ANOVA and Canonical Analyses were performed with Statistica 6.1 from StatSoft Inc. (2003). All statistical analyses used the whole data set, regardless of sex, but excluding specimens of age-class 0. Univariate statistics and analyses were restricted to summary statistics: n, arithmetic mean, minimum value, maximum value, standard deviation and coefficient of variation. Correlation matrices were used to detect measuring errors; aberrant data or outliers were checked.

Canonical Analyses (Discriminant Function Analysis) were calculated on the variance-covariance matrices using forward stepwise technique, to obtain the best discriminating combination of variables. To enhance the clarity of the results depicted in our multi-group graphs, only the 95% equiprobable ellipses were shown instead of the individual scores. In certain cases, especially when comparing a great number of OTU's, we used the UPGMA method (Sneath & Sokal, 1973) to construct tree-diagrams based on Mahalanobis distances between centroids, an approach that accounted for all the relevant axes in the canonical hyperspace. Since most sample sizes were relatively small, no tests were made to evaluate whether the data were normally distributed. Preferably, in canonical analysis OTU's should have more specimens than variables; this was not the case for OTU 20 (n=23), OTU 50 (n=16) and OTU 60 (n=22).

Chromosomes. We karyotyped 22 specimens of the *A. spinosissimus* species group from 11 localities (Table 3). We obtained chromosome metaphases from the bone marrow, following Hsu and Patton (1969). Fixed cell suspensions were transported to the Universita di Roma "La Sapienza" (Italy) where slides were prepared. We used the Giemsa standard method (pH 7) to stain the metaphases and collected the obtained results with a digital camera Phetometrics Sensys 1600 and the software Iplab (Scanalytics Inc., version 2.420).

TABLE 3. Summary of the studied allozyme frequencies.

LOCUS	MOROG	DAKAW	WILSO	KANYEL	MBETE	MNIMA	MAGA	MBET	MLAL	PERAM
LDH-2	N= 14	N= 3	N=6	N=7	N=13	N=16	N=3	N=2	N=13	N=3
A	1.000	1.000	0.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
B	0.000	0.000	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
LDH-1	14	3	6	7	13	16	3	2	13	3
A	0.000	0.000	0.167	0.000	0.000	0.000	0.000	0.000	0.077	0.000
B	1.000	1.000	0.833	0.000	1.000	1.000	0.000	1.000	0.923	1.000
C	0.000	0.000	0.000	1.000	0.000	0.000	1.000	0.000	0.000	0.000
MDH	14	3	6	7	13	16	3	2	13	3
A	0.000	0.000	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
B	1.000	1.000	0.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
SOD	14	3	6	7	13	16	3	2	13	3
A	0.000	0.000	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
B	1.000	1.000	0.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
GDH	14	3	6	7	13	16	3	2	13	3
A	0.000	0.000	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
B	0.000	0.000	0.000	1.000	0.000	0.000	1.000	0.000	0.000	0.000
C	1.000	1.000	0.000	0.000	1.000	1.000	0.000	1.000	1.000	1.000

continued next page

TABLE 3. (continued)

LOCUS	MOROG	DAKAW	WILSO	KANYEL	MBETE	MNIMA	MAGA	MBET	MLAL	PERAM
DIA	14	1	6	7	13	16	3	2	13	3
A	0.000	0.000	0.700	0.000	0.000	0.000	0.000	0.000	0.000	0.000
B	0.500	0.500	0.300	0.000	0.500	0.700	0.500	0.500	0.500	0.500
C	0.500	0.500	0.000	1.000	0.500	0.300	0.500	0.500	0.500	0.500
PGD	14	3	6	7	13	16	3	2	13	3
A	0.000	0.000	0.000	0.286	0.000	0.000	0.000	0.000	0.000	0.000
B	0.000	0.000	0.000	0.714	0.000	0.000	0.000	0.000	0.000	0.000
C	0.000	0.000	1.000	0.000	0.000	0.333	1.000	1.000	0.000	0.333
D	1.000	1.000	0.000	0.000	1.000	0.667	0.000	0.000	1.000	0.667
GPI	14	3	6	7	13	16	3	2	13	3
A	0.000	0.000	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
B	1.000	1.000	0.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
FUM	14	3	6	7	13	16	3	2	13	3
A	0.000	0.000	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
B	1.000	1.000	0.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
PEP	14	3	6	7	13	16	3	2	13	3
A	0.000	0.000	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
B	1.000	1.000	0.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
GOT	14	3	6	7	13	16	3	2	13	3
A	0.000	0.000	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
B	1.000	1.000	0.000	0.000	1.000	0.967	0.000	0.000	1.000	1.000
C	0.000	0.000	0.000	0.000	0.000	0.033	0.000	0.000	0.000	0.000
PNP	14	3	6	7	13	16	3	2	13	3
A	0.000	0.000	0.000	1.000	0.000	0.000	1.000	1.000	0.000	0.000
B	1.000	1.000	0.000	0.000	1.000	0.000	0.000	0.000	1.000	1.000
C	0.000	0.000	1.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
PGM	14	3	6	7	13	16	3	2	13	3
A	0.000	0.000	0.500	0.000	0.000	0.000	0.000	0.000	0.000	0.000
B	0.500	0.500	0.000	0.500	0.500	0.500	0.500	0.500	0.500	0.500
C	0.500	0.500	0.500	0.500	0.500	0.500	0.500	0.500	0.500	0.500
IDH	14	3	6.000	7	13	14	3	2	13	3
A	0.000	0.000	0.083	0.000	0.000	0.071	0.000	0.000	0.000	0.000
B	0.393	0.167	0.000	0.500	0.500	0.393	0.500	0.500	0.462	0.500
C	0.000	0.000	0.583	0.000	0.000	0.000	0.000	0.000	0.000	0.000
D	0.607	0.833	0.000	0.500	0.500	0.536	0.500	0.500	0.538	0.500
E	0.000	0.000	0.250	0.000	0.000	0.000	0.000	0.000	0.000	0.000
F	0.000	0.000	0.083	0.000	0.000	0.000	0.000	0.000	0.000	0.000

Enzyme electrophoresis. The dissected tissues were transported to the University of Antwerp (Belgium) in liquid nitrogen, where they were stored at -80°C. Prior to electrophoresis, tissue samples were thawed, rinsed in distilled water, weighted and homogenized in a 25 M aqueous sucrose solution (5 µl per mg tissue). All manipulations were done on ice. Crude homogenates were centrifuged during 60 min at 27000 rpm and 4°C. The clear supernatants were used directly or stored at -80°C.

Vertical polyacrylamide gel electrophoresis (PAGE) was performed as described by Van Rompaey (1984). We used three buffer systems: (1) a discontinuous one Tris/Glycine and Tris–HCl, both at pH 9.0); two continuous ones: (2) Tris/Citric acid at pH 8.0; and (3) Tris/EDTA/Boric acid at pH 8.9. Gels were stained for thirteen enzymes: LDH (EC.1.1.1.27), MDH (EC.1.1.1.37) SOD (EC.1.15.1.1), GDH (EC.1.1.1.47), DIA (EC.1.6.*.*), PGD (EC.1.1.1.44), GPI (EC.5.3.1.9), FUM (EC.4.2.1.2), PEP (EC.3.4.*.*), GOT (EC.2.6.1.1), PNP (EC2.4.2.1), PGM (EC.5.4.2.2) and IDH (EC.1.1.1.42).

Alleles were labeled alphabetically according to decreasing electrophoretic mobility (A=fastest allele). Allele frequencies, mean number of alleles per locus (A), proportions of polymorphic loci (P) and mean expected and observed heterozygosities (He and Ho), Nei's (1972) genetic distance (DN) and Wright's (1978) Prevosti distance (PD) were calculated using BIOSYS-1 (Swofford & Selander 1981). To obtain the distance of Reynolds *et al.* (1983) (DR), allele frequencies were also subjected to GENDIST from PHYLIP 3.4 (Felsenstein 1991).

We used BIOSYS-1, UPGMA + TDRAW (Saitou & Nei 1987) and NEIGHBOR (PHYLIP 3.4), to obtain UPGMA, Distance Wagner and neighbor-joining trees. A maximum likelihood tree was inferred based on the obtained allele frequencies and the program CONTML (PHYLIP 3.4). Obtained trees were rooted by midpoint rooting or by designating *A. wilsoni* as outgroup.

DNA methods. Total DNA was extracted from frozen or ethanol preserved muscle or liver tissues using the QuiaAmp DNA Minikit. The complete mitochondrial cytochrome *b* sequences (1140 bp) were amplified using primers L14723 and H15915 (Ducroz *et al.* 1998). PCR amplifications were carried out in 25 µl reaction volumes, containing 10 µM of each primer, 200 µmol dNTPs, 10mM Tris–HCL, 1.5 mM $MgCl_2$, 50 mM KCl (pH 8.3) and 1 unit *Taq* polymerase.

The used PCR conditions were as described elsewhere (Ducroz *et al.* 1998). The PCR products were cleaned using the GFX PCR DNA and Gel band Purification Kit (Amersham Biosciences). Dye terminator cycle sequencing was performed following the manufacturer's instructions. The used primers included L14723, H15915 and two additional primers L15408 and H15553 (Ducroz *et al.* 1998).

The reaction products were either run on an ABI 310 sequencer in the molecular laboratory of the RBINS or at the VIB sequencing facility at the University of Antwerp. To validate the mitochondrial origin of the obtained cytochrome *b* sequences, the corresponding amino acid sequences were screened for the presence of stop codons, deletions or inserts, using MEGA4.0 (Tamura *et al.* 2007).

Phylogenetic analyses. A Bayesian tree was obtained with MrBayes v3.1.2 (Ronquist & Huelsenbeck 2003) based on 92 sequences 1140 bp long cytochrome *b* sequences; 61 of these are new *Acomys* sequences [45 are *A. spinosssisimus*, 12 are *A. wilsoni*, 3 are *A. subspinosus*, one *A. ignitus*] complemented with 28 already published sequences of different *Acomys* species and close and more distant outgroups [*Lophuromys sikapusi* (Barome et al. 1998, 2000, 2001) *Mus musculus* (Bibb *et al.* 1981) and *Rattus rattus* (Suzuki *et al.* 2000)].

AIC, AICc and BIC criteria were tested with jModeltest (Posada 2008). The selected models based on the different criteria were similar, in all cases nst=3 were suggested. The sequence matrix was split into three partitions corresponding to the codon positions; thereby nucleotide substitution parameters for each partition were unlinked. According to jModeltest, a GTR+G model was used for all partitions. Two parallel runs with four chains each were launched for 5 million generations and sampled every 1000th generation until the average split of frequencies reached 0.0062 and the analysis was stopped.

A Bayesian tree was obtained with MrBayes v3.1.2 (Ronquist & Huelsenbeck 2003), other phylogenetic inferences (not shown) were obtained using maximum parsimony (MP) and maximum likelihood (ML) in PAUP*v4b10 (Swofford 2002). For MP, 466 parsimony-informative characters were present. All characters were given equal weight. Further parameters were: starting trees were obtained via stepwise addition (addition sequence: simple, branch-swapping algorithm: tree-bisection-reconnection (TBR)). For ML, starting trees were obtained via stepwise addition. Further parameters were: addition sequence: as-is; branch-swapping algorithm: tree-bisection-reconnection (TBR); jModeltest was followed for parameters. Node support was estimated through bootstrapping with 100 full bootstrap replicates (ML).

Results and discussion

Mitochondrial phylogeny. Regardless of the method used, the Tanzanian representatives of the *A. spinosissimus* complex were clearly divided in clades A and B (Fig.4). Clade A contained 2 subgroups, one that included samples from the north-eastern populations of Tanzania [Molecular Operation Taxonomical Units 10+11, abbreviated as MOTU 10+11], the second one from South East Tanzania (MOTU 20). In contrast, clade B grouped the *A. spinosissimus* populations occurring on the Tanzanian plateau west of the Eastern Arc Mountain zone. The distribution area of this clade extended from northeast Malawi (Mwenerondo) north-eastward into Tanzania (Zoissa).

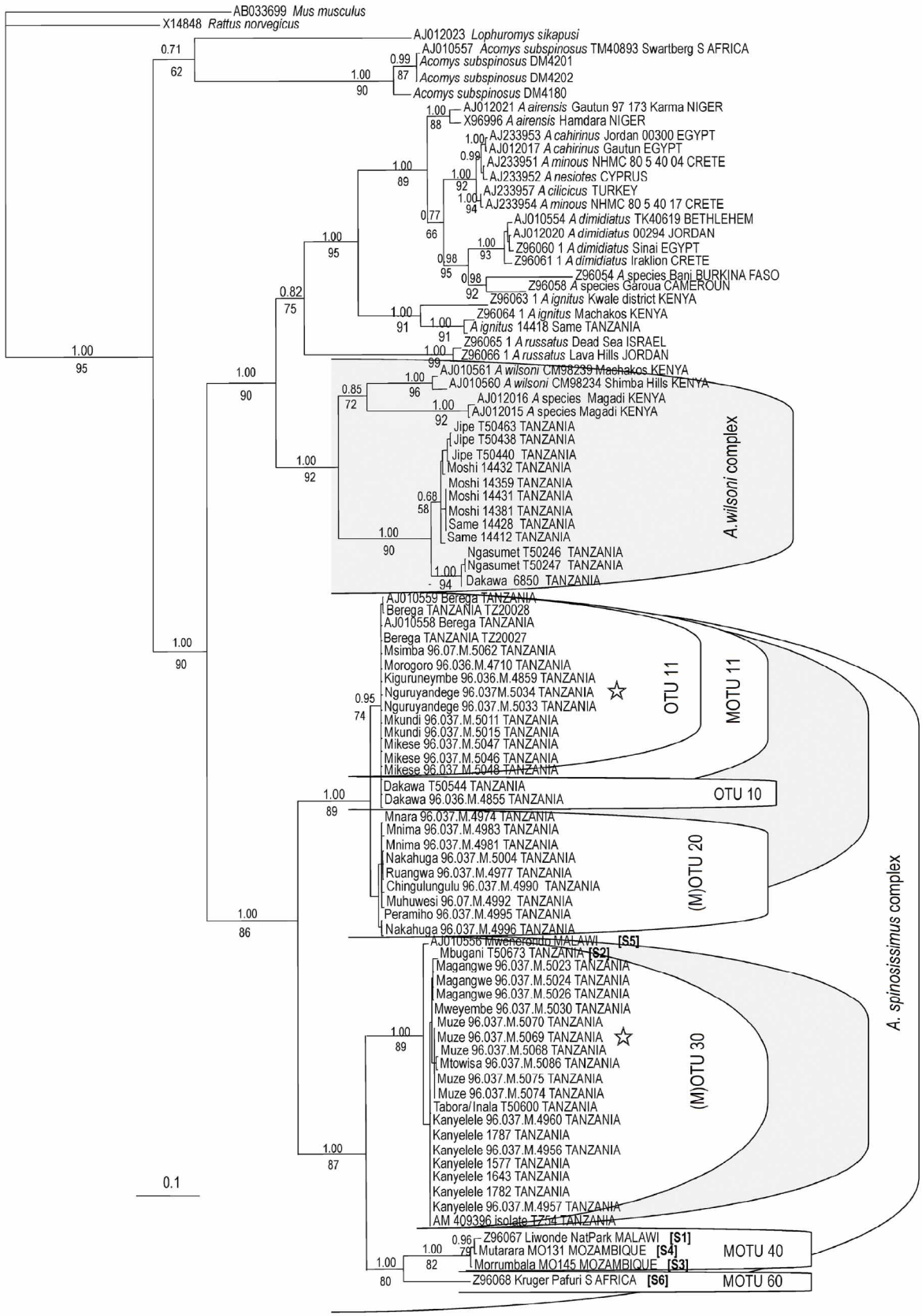


FIGURE 4. Bayesian tree based on complete cyt b sequences for all *A. spinossisimus* populations with corresponding OTU numbers and clade assignations. Species names are preceded by EMBL accession numbers for already published sequences, number preceding a locality name is the museum or specimen number. Support values above the nodes of this phylogeny indicate the degree of support provided by Bayesian analysis, below by ML. Sequenced type specimens are indicated with an asterisk.

The inferred taxonomic implications of the craniometric, morphological and allozyme data (see discussion) were supported by the observation that the genetic distances among these Tanzanian populations and well established species such as *A. cahirinus, A. wilsoni, A. russatus, A. ignitus* and *A. subspinosus* are similarly high (16–19%). Also the included *A. wilsoni* sequences from Tanzania and Kenya suggested that also this taxon was far from homogeneous, and that it is likely that further sampling could reveal even more then the three clades detected here.

Enzyme electrophoresis. The allele frequencies of the fourteen putative loci (two loci for LDH) were summarized in Table 4. The assayed *A. wilsoni* and *A. spinosissimus* samples had alternative alleles at eight out of the fourteen loci (LDH-1, MDH, SOD, GDH, GPI, FUM, PEP and GOT). Hence Nei's (1972) genetic distance between both species was consequently large (D >1.700). We also detected a substantial difference in allele frequencies among the nine studied *A. spinosissimus* populations, yielding genetic distances (DN) ranging from 0.000–0.513.

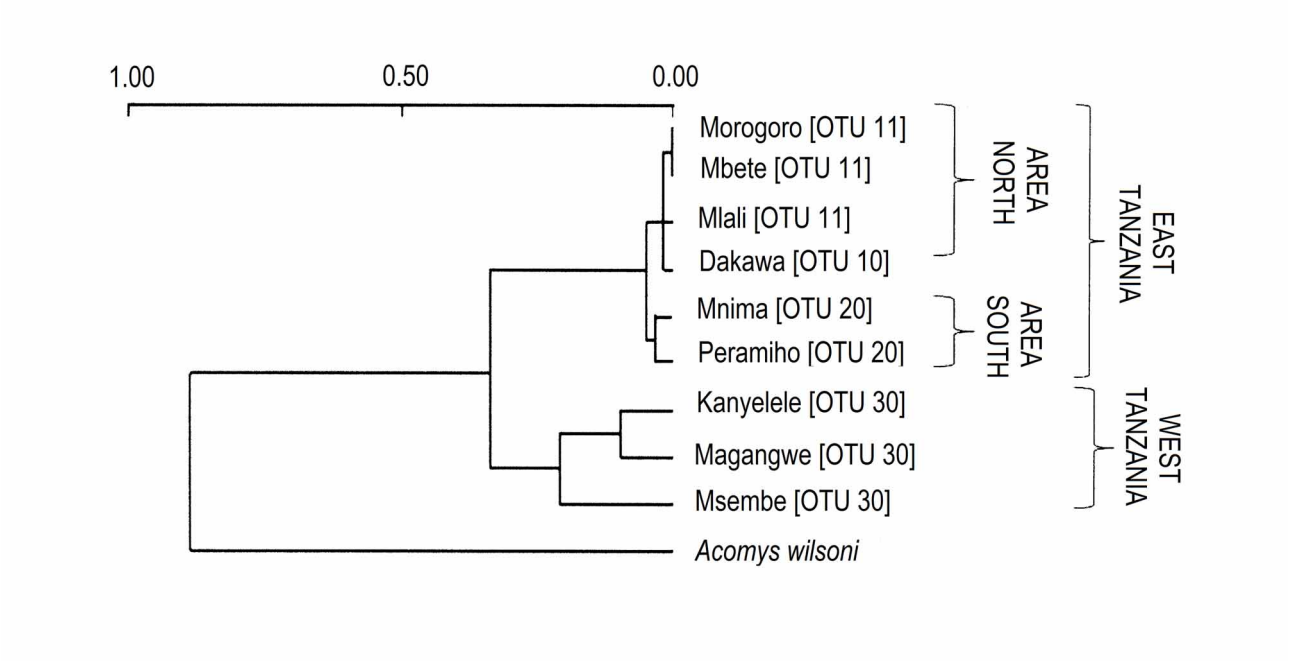


FIGURE 5. UPGMA tree derived from Wright's (1978) Prevosti distances for Tanzanian *Acomys* populations calculated from 14 enzyme-loci.

All distance calculations were rerun without two loci (DIA and PGM) with unusually high numbers of presumed, possibly artificial heterozygotes. This omission usually resulted in even higher genetic distances and lowered estimates for polymorphism and heterozygosity. Regardless of the tree-building method used, the same or very similar topologies were obtained (Fig. 5). Our results suggested that northwest and southeast Tanzanian *A. spinosissimus* populations were genetically clearly differentiated. Although there was substantial genetic differentiation between these two regions, the degree of genetic diversity differs between each region, genetic distances between northwest populations being considerably larger.

Grouping the populations according to regions showed that two loci were diagnostic or nearly so, *viz*. GOT and PNP (Table 4). At these loci, the populations in the northwest region were fixed for single alleles, which are lacking (allele A in PNP) or very rare (allele C in GOT) in the southeast populations (data not shown). In any case, the southeast populations are more homogeneous since they showed significant allele frequency heterogeneity at only one (PGD) out of five polymorphic loci.

Karyotypes. For 19 of the 22 assayed individuals (Table 3) we found a stable karyotype identical to the one described for *A. spinosissimus* from Transvaal, South Africa by Dippenaar and Rautenbach (1986). This karyotype was characterized by a chromosomal mechanism of sex determination of the XX/XY type, a stable diploid number of 60 (within and between individuals), a stable aFN of 68 (10 bi-armed and 48 acrocentric chromosomes) and an X-chromosome representing about 10% of the total length of the diploid set.

Only in two instances (both females) was one of the X-chromosomes missing (2n=59), but at the same locali-

ties (Mkundi and Mbete) other females had two X-chromosomes (2n=60). Interestingly, the only karyotyped female for Dakawa possessed a giant X chromosome and resembled a karyotype previously described by Matthey (1965), which was further characterized by variation in the diploid number among all of the specimens (59–62) and also an important variation in aFN (68–76). In contrast, no chromosomal variation was detected among any of the tested males from Dakawa.

TABLE 4. Karyotype data of the studied Tanzanian *Acomys* populations.

OTU	REG. NR	LOCALITY	SEX	2n	aFN	META CENTRIC	ACRO CENTRIC	
10	RMCA 96.036.M.4837	DAKAWA	M	60	68	5	24	XY
10	RMCA 96.036.M.4838	DAKAWA	M	60	68	5	24	X?
10	RMCA 96.036.M.4848	DAKAWA	M	60	68	5	24	XY
10	RMCA 96.036.M.4849	DAKAWA	M	60	68	5	24	XY
10	ROMA T50544	DAKAWA	F	58–62	68–76			
11	RMCA 96.036.M.5005	MBETE	F	60	68	5	24	XX
11	RMCA 96.036.M.5006	MBETE	F	59	68	5	24	XO
11	RMCA 96.037.M.5009	MKUNDI	F	59	68	5	24	XO
11	RMCA 96.037.M.5011	MKUNDI	F	60	68	5	24	XX
20	RMCA 96.037.M.4990	CHINGULUNGULU	M	60	68	5	24	XY
20	RMCA 96.037.M.4974	MNARA	M	60	68	5	24	XY
20	RMCA 96.037.M.4976	MNARA	M	60	68	5	24	XY
20	RMCA 96.037.M.4981	MNIMA	M	60	68	5	24	XY
20	RMCA 96.037.M.5004	NAKAHUGA	M	60	68	5	24	XY
30	ROMA T50003	MATONGOLO	F	60	68	5	24	
30	ROMA T50673	MBUGANI-CHUNYA	F	60	68	5	24	
30	ROMA T50676	MBUGANI-CHUNYA	F	60	68	5	24	
30	ROMA T50600	TABORA INALA	F	60	68	5	24	
30	ROMA T50087	ZOISSA	M	60	68	5	24	
30	ROMA T50088	ZOISSA	F	60	68	5	24	
30	ROMA T50119	ZOISSA	F	60	68	5	24	
30	ROMA T50202	ZOISSA	M	60	68	5	24	

Morphology and craniometry. First, we evaluated whether the age and sex composition of the OTU's influenced the outcome of our analyses. To evaluate the sexual and growth variation in the skull we used the Dakawa series (OTU 10), with its 147 specimens by far the largest single locality OTU available, including specimens from different age classes as inferred from the wear pattern of the upper molars (Fig.2). We detected no sexual dimorphism in the shape or size in the molars (data not shown).

Student's t-tests across age classes and sexes demonstrated that there is no significant sexual dimorphism in the external and cranial dimensions in age classes (1+2) of OTU 10 (Table 5a). In the age classes (3+4) we observed that the females were slightly larger than the males for M1 (GRLS), M14 (ZYGP), M21 (DINC), M22 (ROHE), M23 (ROBR), but statistically smaller for M19 (BULL). (Table 5b). However, the observed ranges overlapped so widely that we could not infer a significant degree of sexual dimorphism. Moreover, there was no significant craniometrical dimorphism when we pooled the age classes (1–5).

When we evaluated growth between juvenile and adult specimens (respectively age classes 1+2 and 3+4) for both sexes, we found that measures were greater for adult skulls, except for M8 (INTE) (♂and ♀), M10 (PALA) (♂), M13 (M1BR) (♂and ♀), M15 (BNAS) (♂and ♀), M17 (LOTE) (♂and ♀), M19 (BULL) (♂), M20 (BRCA) (♀). For three measurements [M21 (DINC), M14 (ZYGP), M23 (ROBR)] the size difference was less significant in males than in females; the same pattern was observed for HB (Head + Body length) and W (Weight). Assuming

that the tooth wear was a reliable measure for the age of these rodents, our measurements indicated that males grew less than females between the considered age-categories (Appendix 2). Two-Way analyses of variance showed only significant differences for age, none for sex or for age*sex interaction.

Finally, analyses to evaluate the possible effect of differences in composition of our OTU's with regard to age and sex revealed no significant bearing on the outcome of canonical analyses and consequently on our taxonomic interpretation. When the Dakawa sample is split into sex-age classes, the age classes (1+2) and (3+4) cluster together regardless of sex; even the age class (1+2) from another locality (Mbete) clusters with the other age classes (1+2), regardless of locality (Fig.7b).

TABLE 5. (A) Basic statistics and sexual dimorphism in the Dakawa population (OTU 10) for specimens (age classes 1+2). **(B)** Basic statistics and sexual dimorphism in the Dakawa population (OTU 10) for specimens (age classes 3+4). Growth differences in males and females (OTU10, Dakawa) between age (1+2) and age (3+4) are provided in Appendix 2. Significances of the Student's t-tests: ns: not significant; * p<0.05, ** p<0.01, *** p<0.001.

(A)

Dakawa [males age (1+2)] (OTU 10)							Dakawa [females age (1+2)] (OTU 10)							
	N	Mean	Min	Max	SD	CV%	N	Mean	Min	Max	SD	CV%	Sign.	
M1	42	25,08	23,60	26,50	0,701	2,8	23	25,06	24,00	26,10	0,596	2,4	ns	M1
M2	46	22,78	21,50	24,20	0,658	2,9	26	22,56	21,35	23,70	0,591	2,6	ns	M2
M3	46	19,14	18,10	20,75	0,621	3,2	26	18,91	18,15	20,35	0,506	2,7	ns	M3
M4	46	12,18	11,30	13,35	0,529	4,3	27	12,14	11,40	13,15	0,374	3,1	ns	M4
M5	46	5,28	4,75	5,90	0,250	4,7	27	5,29	4,85	5,75	0,257	4,9	ns	M5
M6	46	6,47	5,80	7,15	0,288	4,5	27	6,41	6,00	7,00	0,251	3,9	ns	M6
M7	46	6,92	6,40	7,65	0,287	4,1	27	6,83	6,35	7,45	0,277	4,1	ns	M7
M8	46	4,62	4,30	4,90	0,157	3,4	27	4,63	4,40	4,95	0,146	3,1	ns	M8
M9	46	12,48	11,60	13,30	0,382	3,1	26	12,43	11,60	13,00	0,337	2,7	ns	M9
M10	46	3,00	2,55	3,30	0,168	5,6	27	2,97	2,65	3,30	0,172	5,8	ns	M10
M11	46	4,30	3,95	4,65	0,133	3,1	27	4,26	4,05	4,45	0,106	2,5	ns	M11
M12	46	5,72	5,40	6,10	0,184	3,2	27	5,69	5,35	6,05	0,179	3,1	ns	M12
M13	46	1,38	1,20	1,50	0,072	5,2	27	1,38	1,25	1,50	0,067	4,9	ns	M13
M14	46	3,11	2,80	3,40	0,146	4,7	27	3,08	2,70	3,45	0,166	5,4	ns	M14
M15	46	2,48	2,20	2,75	0,126	5,1	27	2,50	2,25	2,80	0,119	4,8	ns	M15
M16	43	9,72	8,90	10,80	0,473	4,9	25	9,75	9,15	10,65	0,418	4,3	ns	M16
M17	46	3,86	3,45	4,15	0,157	4,1	27	3,84	3,65	4,20	0,126	3,3	ns	M17
M19	46	4,24	3,85	4,80	0,184	4,3	26	4,27	4,00	4,65	0,204	4,8	ns	M19
M20	46	11,39	10,95	12,20	0,256	2,2	26	11,39	10,90	11,95	0,283	2,5	ns	M20
M21	46	1,27	1,10	1,45	0,083	6,5	27	1,25	1,10	1,50	0,098	7,8	ns	M21
M22	46	5,20	4,80	5,70	0,202	3,9	27	5,14	4,85	5,50	0,185	3,6	ns	M22
M23	46	3,76	3,35	4,20	0,184	4,9	27	3,75	3,50	4,15	0,141	3,8	ns	M23
M24	46	6,37	5,75	6,90	0,263	4,1	27	6,29	5,70	6,80	0,226	3,6	ns	M24
W	45	22,8	13	28	3,14	14,2	25	21,1	14	30	3,68	17,4	ns	W
HB	43	89,3	73	104	7,99	8,9	25	88,0	74	100	7,46	8,5	ns	HB
TL	29	75,8	62	90	5,81	7,7	18	73,3	65	86	5,13	7,0	ns	TL
HL	43	15,1	12	17	1,11	7,3	25	15,2	13	18	1,18	7,8	ns	HL
EL	30	13,6	11	16	1,08	7,9	20	13,6	11	15	1,19	8,7	ns	EL

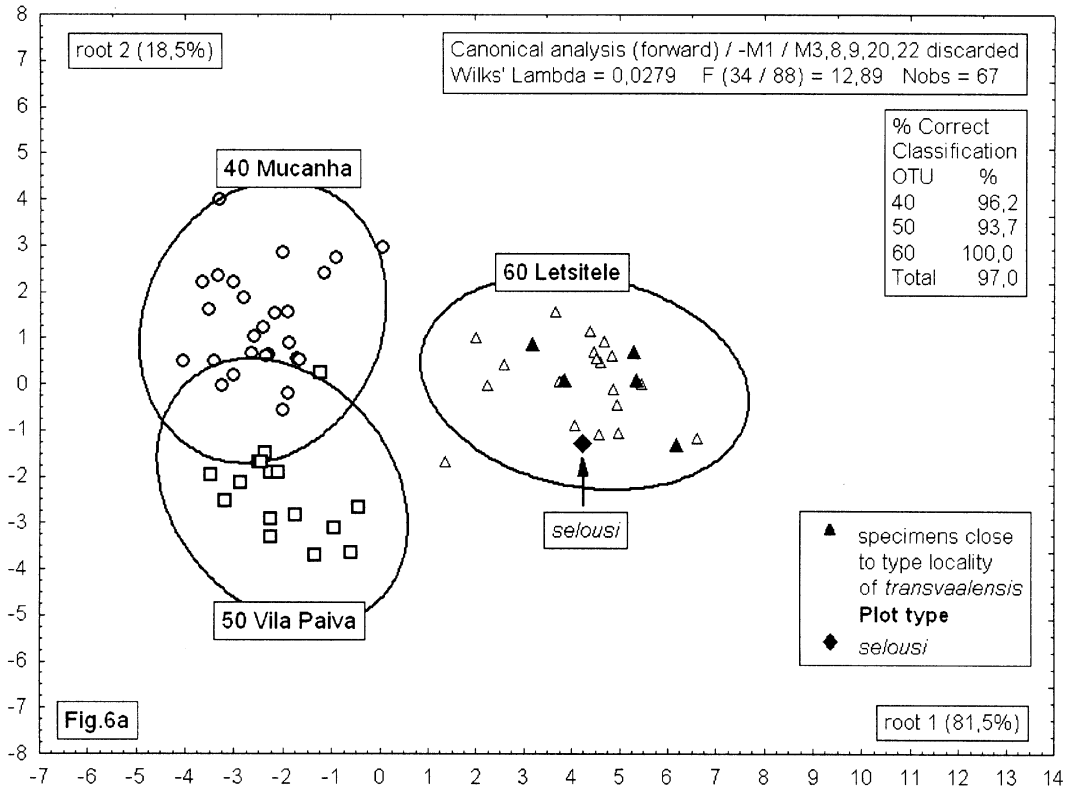
(B)

Dakawa males age (3+4) (OTU 10)							Dakawa females age (3+4) (OTU 10)							
	N	Mean	Min	Max	SD	CV%	N	Mean	Min	Max	SD	CV%	Sign.	
M1	26	25,84	24,80	27,05	0,549	2,1	18	26,28	25,00	27,35	0,571	2,2	*	M1
M2	27	23,70	22,50	25,00	0,595	2,5	18	23,98	22,70	25,50	0,708	3,0	ns	M2
M3	27	20,10	19,15	21,05	0,564	2,8	17	20,20	19,05	20,95	0,499	2,5	ns	M3
M4	25	12,71	11,75	13,45	0,499	3,9	18	12,90	11,35	13,80	0,595	4,6	ns	M4
M5	27	5,60	5,00	6,00	0,265	4,7	18	5,65	5,15	6,15	0,239	4,2	ns	M5
M6	27	6,91	6,25	7,50	0,282	4,1	18	6,98	6,30	7,45	0,281	4,0	ns	M6
M7	27	7,36	6,65	7,90	0,302	4,1	18	7,49	6,90	7,95	0,282	3,8	ns	M7
M8	27	4,68	4,40	4,95	0,138	2,9	18	4,70	4,50	4,85	0,129	2,7	ns	M8
M9	27	12,80	12,00	13,60	0,330	2,6	18	12,87	12,30	13,40	0,346	2,7	ns	M9
M10	27	3,07	2,80	3,40	0,164	5,4	18	3,09	2,75	3,40	0,155	5,0	ns	M10
M11	27	4,40	4,15	4,60	0,138	3,1	18	4,40	4,10	4,70	0,158	3,6	ns	M11
M12	27	5,92	5,65	6,25	0,158	2,7	18	5,94	5,60	6,35	0,173	2,9	ns	M12
M13	27	1,38	1,30	1,50	0,049	3,5	18	1,37	1,30	1,45	0,042	3,1	ns	M13
M14	27	3,18	2,90	3,40	0,139	4,4	18	3,32	2,95	3,60	0,194	5,8	**	M14
M15	26	2,54	2,30	2,80	0,117	4,6	18	2,51	2,35	2,75	0,119	4,7	ns	M15
M16	26	10,21	9,20	10,90	0,363	3,6	18	10,36	9,80	10,85	0,251	2,4	ns	M16
M17	27	3,83	3,50	4,10	0,127	3,3	18	3,80	3,60	4,15	0,143	3,8	ns	M17
M19	27	4,24	4,00	4,75	0,165	3,9	18	4,12	3,90	4,40	0,136	3,3	*	M19
M20	27	11,53	11,05	12,10	0,256	2,2	18	11,54	11,15	11,90	0,218	1,9	ns	M20
M21	27	1,31	1,20	1,40	0,053	4,1	18	1,35	1,25	1,45	0,062	4,6	*	M21
M22	27	5,33	4,95	5,60	0,159	3,0	18	5,44	5,00	5,75	0,182	3,3	*	M22
M23	27	3,85	3,65	4,20	0,134	3,5	18	3,94	3,60	4,20	0,152	3,8	*	M23
M24	26	6,57	6,25	7,05	0,232	3,5	17	6,68	6,30	7,15	0,269	4,0	ns	M24
W	27	24,3	13	38	4,74	19,5	17	26,5	20	33	3,91	14,7	ns	W
HB	26	93,2	81	105	6,52	7,0	16	99,1	89	110	5,47	5,5	**	HB
TL	19	76,5	71	81	2,84	3,7	9	77,2	68	87	5,52	7,1	ns	TL
HL	25	15,2	14	17	0,85	5,6	15	15,5	15	17	0,64	4,1	ns	HL
EL	19	14,3	12	16	1,10	7,7	10	14,3	13	15	0,95	6,6	ns	EL

Canonical analysis of the South African OTU's. A forward canonical analysis clearly separated OTU 60 (Letsitele) from OTU 50 (Vila Paiva de Andrada) and OTU 40 (Mucanha) along the first axis, while root 2 differentiated OTU 50 (Vila Paiva) and OTU 40 (Mucanha) (Fig.6a). Based on the factor structure of the analysis (matrix not shown), the measurements that contributed most to differentiate OTU 60 from Mucanha (OTU 40) and Vila Paiva (OTU 50) were, respectively, M24 (PCPA) and M10 (PALA), M19 (BULL), M21 (DINC).

Linking the *selousi* holotype to the studied OTU's by plotting and the classification of cases identified it as a member of OTU 60 (Letsitele). Our confidence in this conclusion was based on the very high percentages (97%) of correct classification resulting from this analysis. The fact that Letsitele was situated near Zoutpansberg, the type locality of *transvaalensis* (holotype not seen nor measured), confirmed that *transvaalensis* was a synonym of *selousi*.

We repeated the canonical analysis with a reduced set of measurements in order to plot values obtained from the damaged skull of the lectotype of *spinosissimus* (Fig. 6b). As expected, the lower number of included variables resulted in greater overlaps between the OTU's. Nevertheless Wilks' Lambda remained very low (= 0.0983), indicating a high degree of differentiation between groups, which was also reflected in the high total percentage of



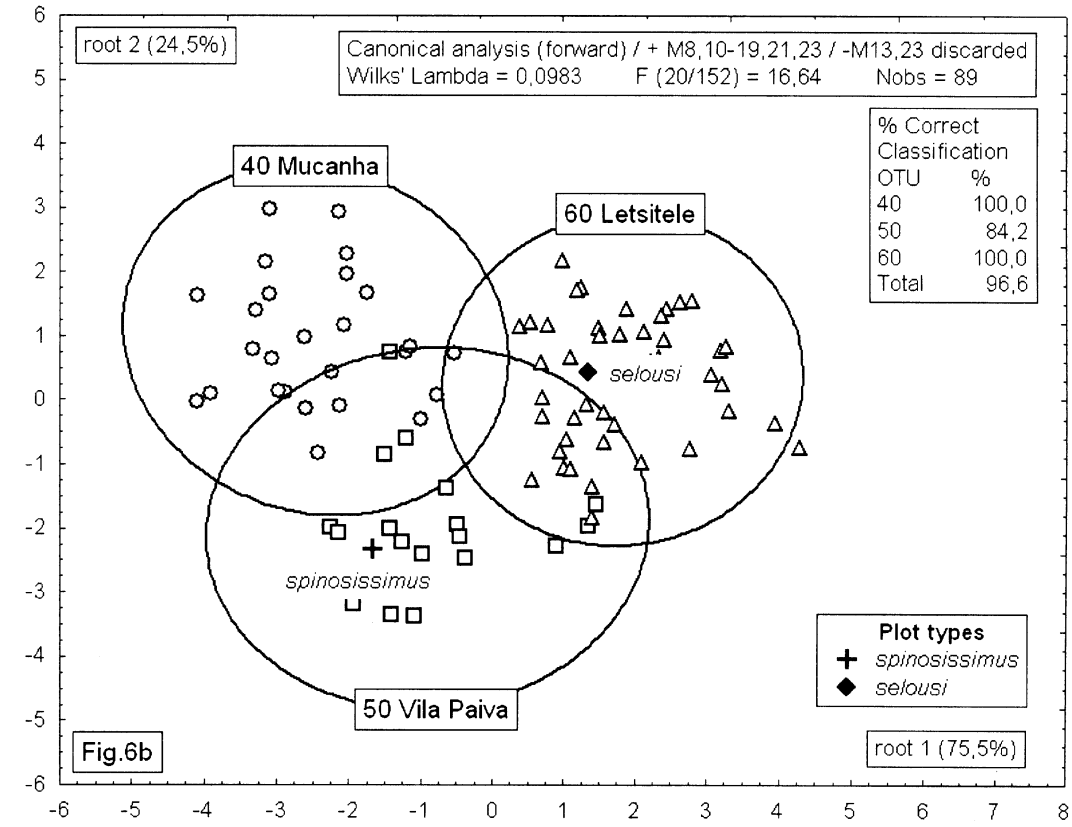
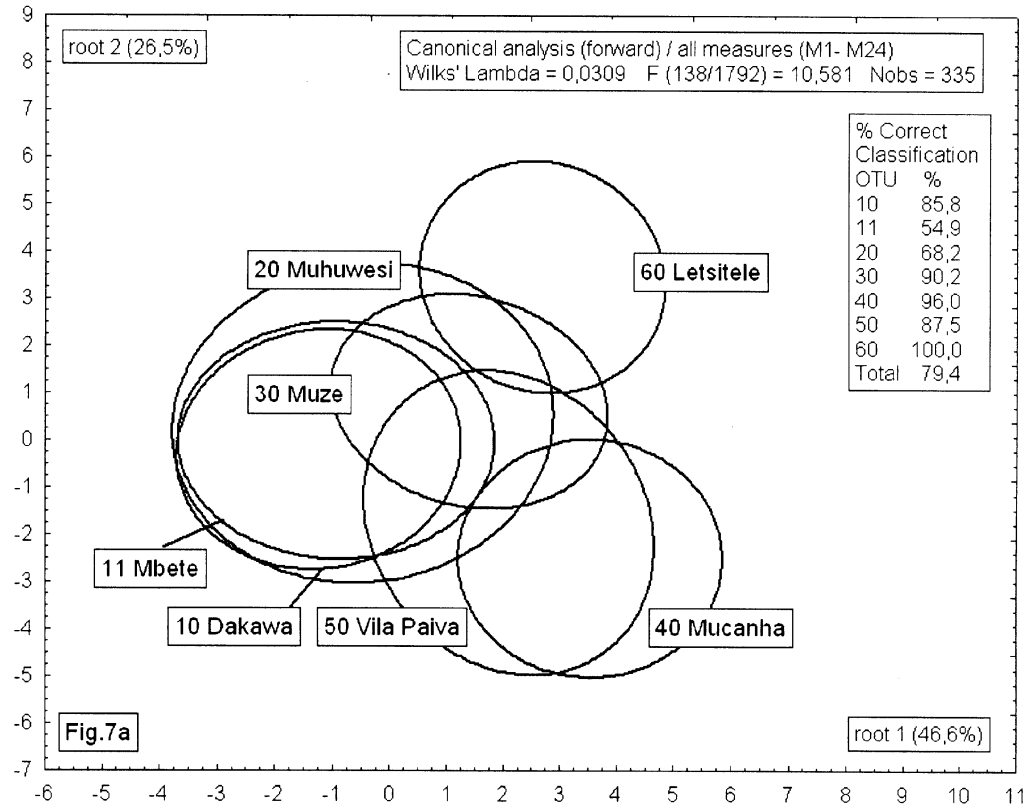


FIGURE 6. (A) Canonical analysis of the southern African *Acomys* OTU 40, 50 and 60 (on which the type of *selousi* is plotted). **(B)** The same canonical analysis with a reduced set of measurements allowed plotting the damaged skull of the *A. spinosissimus* lectotype.



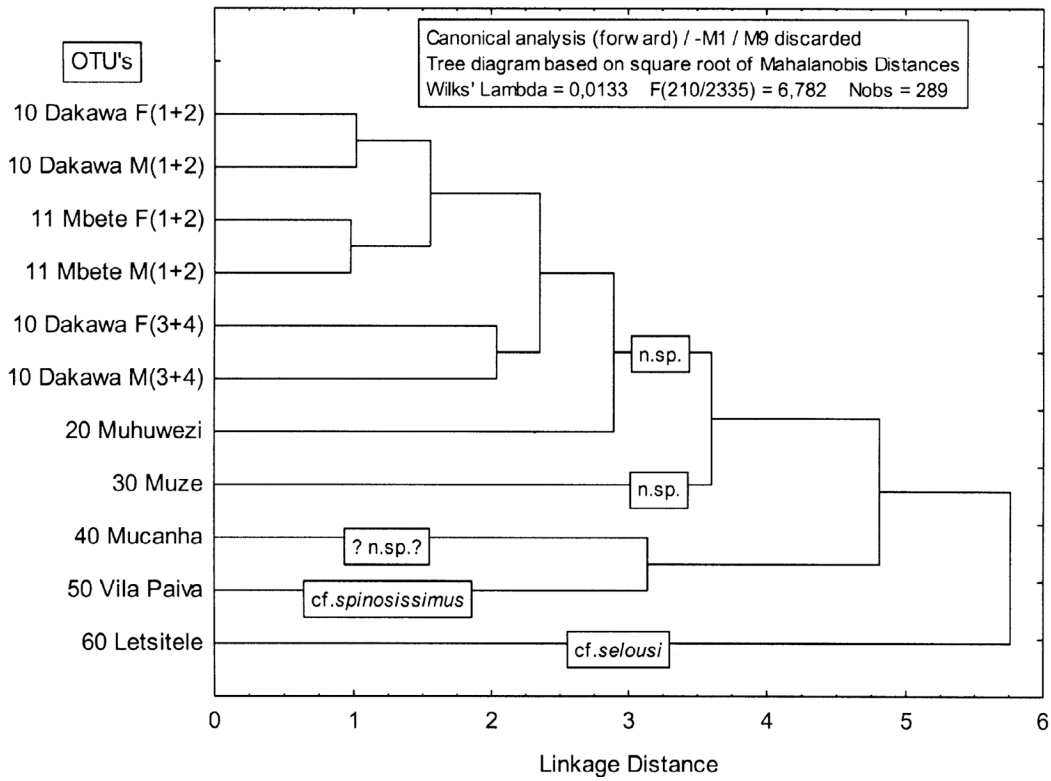
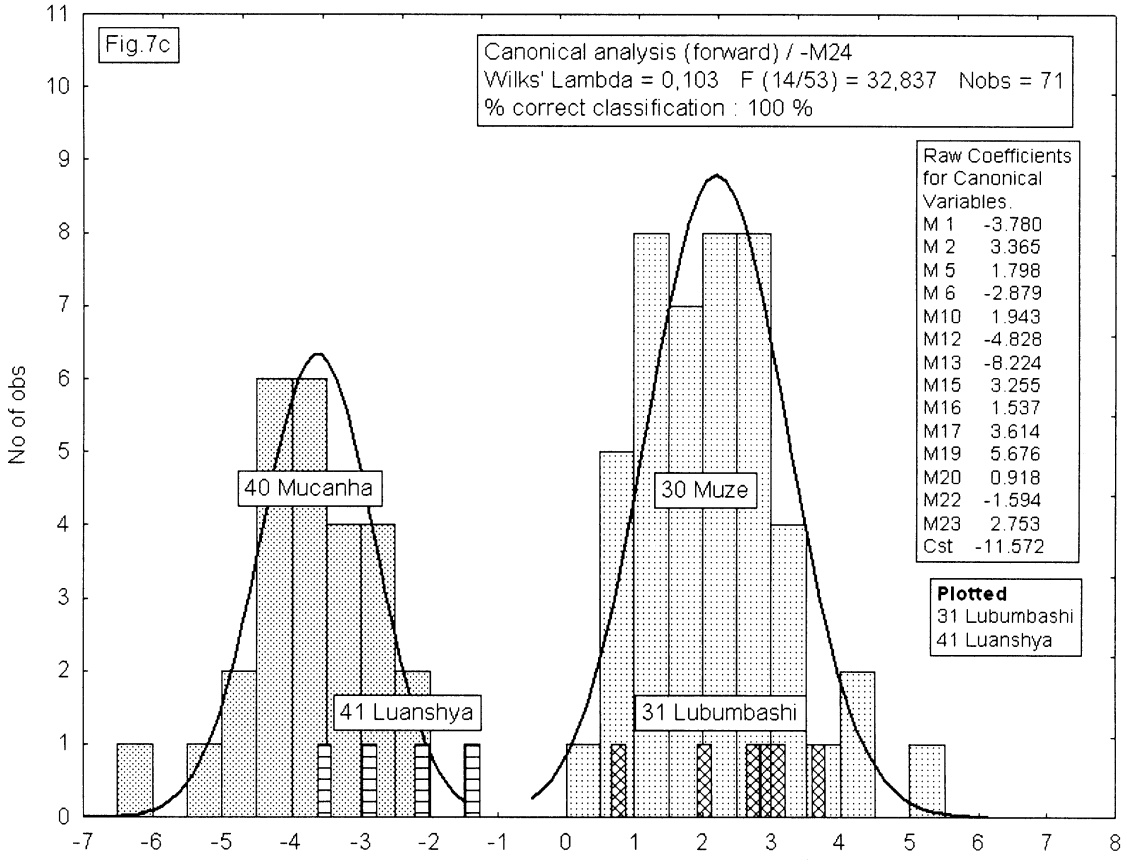


FIGURE 7. (A) Combined canonical analysis of the OTU's that cover most of the geographic range of the *A. spinosissimus species* complex. **(B)** Morphometric UPGMA tree diagram of all the studied OTU's based on the square root of the Mahalanobis squared distances. Type specimens of *A. selousi* and *A. spinosissimus* are represented by OTU60 and OTU50, we labeled them accordingly.



(C) Two group discriminant analysis of OTU 30 and OTU 40, with *a posteriori* plotting of OTU 31 and OTU 41.

correct classification (total=96.6%), OTU's 40 (Mucanha) and 60 (Letsitele) being 100% correctly classified. Plotting of the available type specimens confirmed that *selousi* belonged to OTU 60 (Letsitele) whereas the holotype of *spinosissimus* was assigned to OTU 50 (Vila Paiva), again according to the classification of cases approach. Because we did not have access to a representative series of *spinosissimus* topotypes, these data suggested that OTU 50 from Vila Paiva (relatively closely situated to the *spinosissimus* type locality) was likely to be a valid representative of *A. spinosissimus* on both morphometric and zoogeographical grounds. Finally, these analyses not only indicated that *A. selousi* and *A. spinosissimus* were distinguishable, it appeared that both differ from OTU 40 from Mucanha (north of the Zambezi river), that possibly represented an undescribed taxon.

Canonical analyses of the OTU's from East and South Africa. The outcome of a combined canonical analysis among the seven OTU's that covered most of the geographic range of the *A. spinosissimus* species complex (Fig. 7a) indicated that 73% of the totality of the craniometrical variation was represented by roots 1 and 2. OTU 60 (Letsitele) appeared to be clearly different from all the other OTU's (100% correct classification), whereas OTU 10 (Dakawa), OTU 11 (Mbete) and OTU 20 (Muhuwezi) strongly overlapped. Root 1 was mainly determined by M19 (BULL), M10 (PALA), M17 (LOTE), and M21 (DINC), root 2 was strongly influenced by M24 (PCPA) (factor structure matrix not shown).

Fig. 7b depicted the Mahalanobis distances in a canonical analysis of the OTU's 20, 30, 40, 50, 60, while the original OTU 10 (Dakawa) and OTU 11 (Mbete) have been subdivided into a number of groups according to sex and/or age class. This confirmed that OTU 60 (Letsitele) was clearly differentiated from all the other OTU's. It also showed that OTU 10 (Dakawa) and OTU 11 (Mbete), even when split up into subgroups based upon sex and toothwear, remained associated and were thus craniometrically very similar. Interestingly, our craniometrical measurements suggested that the Tanzanian OTU's (10, 20, and 30) formed a well-defined entity that differed significantly from the Mozambican OTU's (40, 50). In addition, both differed clearly from the South African OTU 60

(Letsitele). Since the type specimens of *A. selousi* and *A. spinosissimus* could be respectively linked to OTU 60 (Letsitele) and OTU 50 (Vila Paiva) (Fig. 6a) we labeled the OTU's accordingly.

In order to establish the affinities of OTU 31 (Lubumbashi) and OTU 41 (Luanshya), we used a two-group discriminant analysis to compare them with their geographically closest neighbors (Fig. 7c). This exercise revealed that the specimens from Luanshya (OTU 41) clustered with OTU 40 (Mucanha), whereas the Lubumbashi (OTU 31) specimens grouped with OTU 30 (Muze). Finally, OTU 30 and OTU 40 appeared to be craniometrically distinct.

Craniometry of the Tanzanian *A. spinosissimus* populations

The results of a canonical analysis performed on all the available Tanzanian *Acomys* skulls of the *spinosissimus* species complex (OTU's 10, 11, 20 and 30) were summarized in Figure 8. Since the type specimens of *selousi* and of *spinosissimus* were previously linked to OTU's 50 and 60, we can use the complete set of 23 measurements in this analysis.

Summarized, this graph indicated that: (i) the representative skull series of the *Acomys* OTU's (10, 11, 20, 30) were craniometrically not clearly differentiated; Wilks' Lambda (=0,233) was rather high compared to other canonical analyses, (ii) OTU's 10 (Dakawa) and 11 (Mbete) overlap; (iii) the OTU 20 ellipse was rather large due to the relatively lower number of specimens it contained (22); (iv) the actual percentage of correct classification for the totality of the examined specimens was very low (71,8%); (v) roots 1 and 2 represented 91% of all observed craniometrical variation; and that (vi) the most north-eastern Usambara population (Amani) completely coincided with OTU's 10 and 11 (not shown).

External measurements. The univariate comparisons with Student's t-tests of the external measurements of OTU's 10+11+20 and OTU 30, and of the Nguru ya Ndege specimens (OTU 11) and the Muze specimens (OTU 30) were summarized in Table 7 a, b. In both comparisons the means for all external measurements differed significantly except for EL (ear length); however, in all cases the ranges overlapped. The values for W and HB were always higher in OTU's 10+11+12 and in the Nguru specimens, while TL and HF were significantly higher in OTU 30 and in the Muze specimens.

Taxonomic implications. Based on the comparative study of specimen collections from Tanzania, Zambia, Zimbabwe, Mozambique, DR Congo and South Africa, we revised the taxonomic status of the Tanzanian *Acomys spinosissimus* populations using external morphology, craniometry, enzyme variation, karyology and mitochondrial DNA sequences. Our results confirmed that *A. spinosissimus* represents a complex of species with seemingly non-overlapping geographic distribution ranges.

Besides the taxonomic implications of our findings for *A. spinossisimus*, the included *A. wilsoni* sequences from Tanzania and Kenya suggested that, also this taxon was far from homogeneous, even when sampled over a relatively limited geographic range. Since the already published information on most *Acomys* species studied to date is based on a limited number of specimens and often relatively few localities, our data on *A. wilsoni* and *A. spinosissimus* indicated that it was likely that further sampling for all *Acomys* may reveal more diversity then was currently accepted, as was evidently the case for *A. spinosissimus* that is the subject of a taxonomic revision resul-ting in the description of two new species.

Implications for *Acomys spinosissimus*. The geographic range of *A. spinosissimus* appeared to be restricted to an area bordered by the Zambezi and Limpopo Rivers. This conclusion was reached despite the limitations imposed by the damaged state of the lectotype of *A. spinossisimus* (ZMB1711-Buio), the fact that it may represent an unusual specimen (a rather large skull for specimens from that region, Dippenaar & Rautenbach, 1986), and that the exact geographical position of the type locality "Buio was imprecisely known (at 17°S fide Peters 1852). This locality (Peters 1852) could be situated close to the escarpment, but also near Tete in the Zambezi Valley (see Thomas 1896: 794).

Our evaluation of the taxonomic status of *A. spinosissimus* was facilitated by the fact that the origin of the studied material (skulls) covered an important proportion of the distribution range of this species complex, including areas in proximity to the *selousi* and *transvaalensis* type localities. From an assignment analysis including the *spinosissimus* lectotype *viz*. OTU 40 and OTU 50 from north and south of the Zambezi River (in the vicinity the *spinosissimus* type locality) and south of the Limpopo River (OTU 60), we concluded that OTU 50 represents *A.*

spinosissimus and that OTU 60 (south of the Limpopo River) and the OTU's from the northern populations represent other taxa (see further).

GenBank lists several *A. spinosissimus* sequences, but none of them characterized this taxon. Indeed, the GenBank mtDNA sequences of two Mozambican localities (Mutarara and Morrumbala, **S4** and **S3** in Fig.1) clustered with the Malawian sequence (Liwonde, **S1** in Fig.1) in OTU 40 (Fig.4). However, craniometrical evidence for MO145 (from Morrumbala) suggested that these sequences should be assigned to OTU 40 (results not shown) [no other craniometrical data was available: specimen MO131 from Mutarara was assigned to age class 0 and cranium of the Liwonde specimen was unavailable (Genbank)]. Our interpretation would clearly be strengthened by the availability of genuine *A. spinosissimus* mtDNA sequences. Finally, it is of interest to mention that the assumed distribution range of *A. spinosissimus* and the *selousi* type locality (Fig.1) suggested that both taxa could have partially overlapping distribution ranges (see below).

Implications for *Acomys selousi.* Our craniometrical analysis assigned the *selousi* holotype as well as specimens from Letsitele (near the type locality of *transvaalensis*) to OTU 60, clustered clearly distinct from OTU 50, which represents *spinosissimus*. These results confirmed earlier reports suggesting that *selousi* and *transvaalensis* were synonyms (Musser & Carleton 2005), but disagreed with the interpretation that *selousi* (the senior synonym) belonged within *A. spinosissimus* (Dippenaar & Rautenbach 1986, Musser & Carleton 2005). As we had no genuine *A. spinosissimus* sequences, our mitochondrial DNA data did not provide additional evidence on this issue. However, the available mtDNA sequences indicated that *A. selousi* (a sequence from Pafuri) was genetically discrete from OTU 40 and that both were clearly differentiated from the northern populations represented by OTU's 10, 11, 20 and 30.

***Acomys* populations north of the Zambezi.** All the investigated populations north of the Zambezi River were genetically distinct from *A. spinosissimus* and *A. selousi* (Fig.4). Following a South-North trajectory, also the craniometrical comparisons revealed that OTU 40 differed significantly from *spinosissimus* (OTU 50) and *selousi* (OTU 60) (Fig.6). Moreover, these two species clearly differed from the strongly overlapping Tanzanian OTU's 10, 11, 20 and 30 (Fig.7a). Further analysis along a West-East axis revealed that OTU's 40 and 41 formed a cluster, as did OTU's 30 and 31, hence providing some additional information on the geographic distribution of these OTU's into respectively Zambia and the DR Congo (Fig.7c). Based upon these observations we drew two conclusions: (i) OTU's 30 and 31 appeared to represent a single species that occurred in western Tanzania as well as in eastern DR Congo; (ii) OTU's 40 and 41 appeared to represent a species that differed from the taxon defined by OTU's 10, 11 and 20 which occupied eastern Tanzania. Although OTU's 10, 11, 20 differed craniometrically from OTU 30, the overlap was considerable. However, other data—such as external morphological features and two alternative measures of genetic cohesion – suggested that the *A. spinossisimus* populations of Tanzania represented two undescribed species.

Except for ear length, all external measurements differed significantly between OTU's 10+11+20 and OTU 30, although in all cases the ranges overlapped (Table 7). The body weight, head and body length were always higher in OTU's 10+11+20, while TL and HF were significantly higher in OTU 30. In addition, enzyme electrophoresis suggested that northwest and southeast Tanzanian *A. spinosissimus* populations (respectively from the same areas as OTU 30 and OTU's 10+11+20) were clearly differentiated (Fig.5). The amount of genetic differentiation among the populations from these two regions differed; the largest genetic distances being recorded between populations in northwest Tanzania. Interestingly, two loci appeared to be diagnostic, or nearly so, between these two groups. The northwest Tanzanian populations displayed fixed alleles, which were lacking (allele A in PNP) or very rare (allele C in GOT) in the southeast populations.

The mtDNA sequences divided the Tanzanian representatives of the *A. spinosissimus* complex in the same manner (Fig.4). Clade A contained two subgroups, one that included the northeast populations of Tanzania (MOTU 10+11) as well as the populations from southeast Tanzania (MOTU 20). Clade B clustered the populations from the Tanzanian plateau, west of the Eastern Arc Mountains. The distribution range of this clade extended further to northeast Malawi (Mwenerondo) and, as shown by craniometrical analysis, eastwards into Tanzania (OTU 30, Zoissa). While the (uncorrected) genetic distances within these clades were very low, the genetic distances between them are surprisingly high (12–13%). The distribution ranges of these two haplogroups were allopatric but in close proximity to each other. For example Berega and Zoissa, localities where respectively clade A and B occurred, were separated by less than 100 km.

Other representatives of the *A. spinosissimus* species complex also appeared to be genetically distinct. The proposed taxonomic implications of these observations were supported by the combination of the detected morphological differences and the observation that the genetic distances between these Tanzanian taxa and well established species such as *A. cahirinus, A. wilsoni, A. russatus, A. ignitus* and *A. subspinosus* were similarly high (16–19%). Although not conclusive, the enzyme and mtDNA sequences indicated that the amount of gene flow between these Tanzanian clades A and B was either extremely low (enzymes) or even undetectable (mtDNA). We argued that the combination of these genetic differences and the observed morphological differentiation warranted the description of two new *Acomys* species that were so far considered to belong to *A. spinossisimus*.

Chromosomal variation within the *A. spinosissimus* species complex. The most often observed karyotype in our Tanzanian samples resembled the karyotype of the Transvaal population as described earlier (Dippenaar & Rautenbach 1986). Occasionally we encountered a karyotype resembling the one attributed to *A. selousi* (Matthey 1965) that was also found and described by Barome (2001) for a specimen from Berega (Tanzania) (Table 4).

We detected this '*selousi*' karyotype (*sensu* Matthey) in only three of the 22 assayed specimens (all three are females: T50544 [OTU 10], RMCA 96.037.M.5006 and RMCA 96.037.M.5009 [both OTU 11]) that originated from geographically neighboring localities (Dakawa, Mbete, Mkundi); whereas six other specimens from the same localities (four males and two females) had the typical '*spinosissimus*' karyotype. As observed by Matthey (1965) it appeared that the '*selousi*' karyotype was rare and that it could co-occur with the 'normal' '*spinosissimus*' karyotype in specimens sampled in the same region. Since the only sequenced female specimen that carried this distinct karyotype was not distinguishable on the basis of its mtDNA sequence (Fig.4, specimen T50544 in clade A) or morphology, we concluded that the available evidence did not allow us to give taxonomic value to this seemingly infrequently occurring chromosomal configuration. This was unexpected since the chromosomal sex determination involved here—with a single and exceptionally large X-chromosome both in males and females – was striking and contrasted with the XX/XY configuration, typical of *A. spinosissimus* and the other *Acomys* species. If future work would confirm this cytogenetic configuration, this would argue against them to belong to a single gene-pool. In that case, (M)OTU's 10, 11 and 20 would contain two taxa that could not be separated on the basis of their morphology and mitochondrial DNA sequences (Table 4).

Our results indicated that the so-called distinguishable '*selousi*' karyotype as described by Matthey (1965) was not typical for *A. selousi*. We have shown that this taxon was only found south of Tanzania, from just north of the Limpopo southward into the Transvaal. Furthermore, the observed chromosomal variation described above appeared to occur among females of OTU's from eastern Tanzania. This agrees with the fact that another specimen with this karyotype was already reported for Berega, Tanzania (Barome *et al.* 2000, 2001).

Our findings warned against relying with too much confidence on karyotypical characterizations of species from poorly known faunas. It did not seem unlikely that also in other cases a particular karyotype was attributed to a certain species name without a firm taxonomical basis. Evidently, in cases where the taxonomy of a group was not sufficiently well known to have reliable information on species distribution ranges, information on karyotypes from specimens collected at localities other than type localities was likely to increase instead of resolve taxonomic problems. This may have been the case here, as numerous descriptions of *Acomys* karyotypes by Matthey (1956, 1963, 1965a,b, 1968) and others (Dippenaar & Rautenbach 1986; Barome 2001; Corti *et al.* 2005) were available, except from the relevant type localities.

Taxonomic conclusions. In contrast to the view that *A. selousi* and *A. transvaalensis* were synonyms of *A. spinosissimus* (Dippenaar & Rautenbach 1986), Barome *et al.* (2000) presented cytogenetic differences suggesting that *spinosissimus* and *selousi* could be different species.

In the absence of topotypes from Zimbabwe (Essexvale), the type specimen of *A. selousi* identified with OTU 60 (Letsitele), which was also at least partly topotypical for *A. transvaalensis*. We considered *selousi* to be the oldest name for a valid species, with *transvaalensis* a synonym, and reaching geographically from deep south (Rustenburg 25.40S –27.15E) up to approximately 20°S.

OTU 50 (Vila Paiva), representing the *Acomys* populations living south of the Zambezi River, clustered with the lectotype of *A. spinosissimus* and agreed with the geographical situation of the topotypical localities of Tete and Buio. Since it was craniometrically well defined and different from both OTU 60 (Letsitele) and OTU 40 (Mucanha), we suggested maintaining *spinosissimus* s. s. as a valid species for that region. Adequate genetic sampling from this region will be needed to corroborate this conclusion.

OTU 40 (Mucanha), representing the *Acomys* population of the northern bank of the Zambezi, was craniometrically differentiated from the populations south of the river (*A. spinosissimus*). Moreover, OTU 40 also differed from OTU 60 (Letsitele) representing *A. selousi* and the Tanzanian populations. The localities Liwonde (S1), Morrumbala (S3) and Mutarara (S4) were situated in the same region as Mucanha (OTU 40) and represented a well defined and separate genetic clade. Should future work confirm that the Liwonde, Morrumbala and Mutarara specimens also belong to OTU40, this unit could very well represent an undescribed taxon with a distribution that stretches westward at least till Luanshya (OTU 41) (see craniometric similarity between OTU's 40 and 41, Fig.7c).

Finally, when plotted on a two-group discriminant analysis of OTU's 30 and 40 (Mucanha), OTU 31 (Lubumbashi), *incertae sedis*, clustered with OTU 30 (Fig.7c), which implied a geographically logical extension of the distribution range of this probably undescribed taxon. We agreed with Barome *et al.* (2000) that a satisfying solution to all taxonomic problems surrounding *A. spinosissimus* can only be reached through cytochrome *b* data from Zimbabwe (preferably topotypical for *selousi*) as well as from Mozambique and intermediate regions. The major conclusion of the current study concerned the taxonomic status of the Tanzanian representatives of the *spinosissimus* complex. Since clade A and clade B were genetically distinct (from each other and from the other clades), we decided to describe them as two new species from the A. *spinosissimus* species complex in Tanzania.

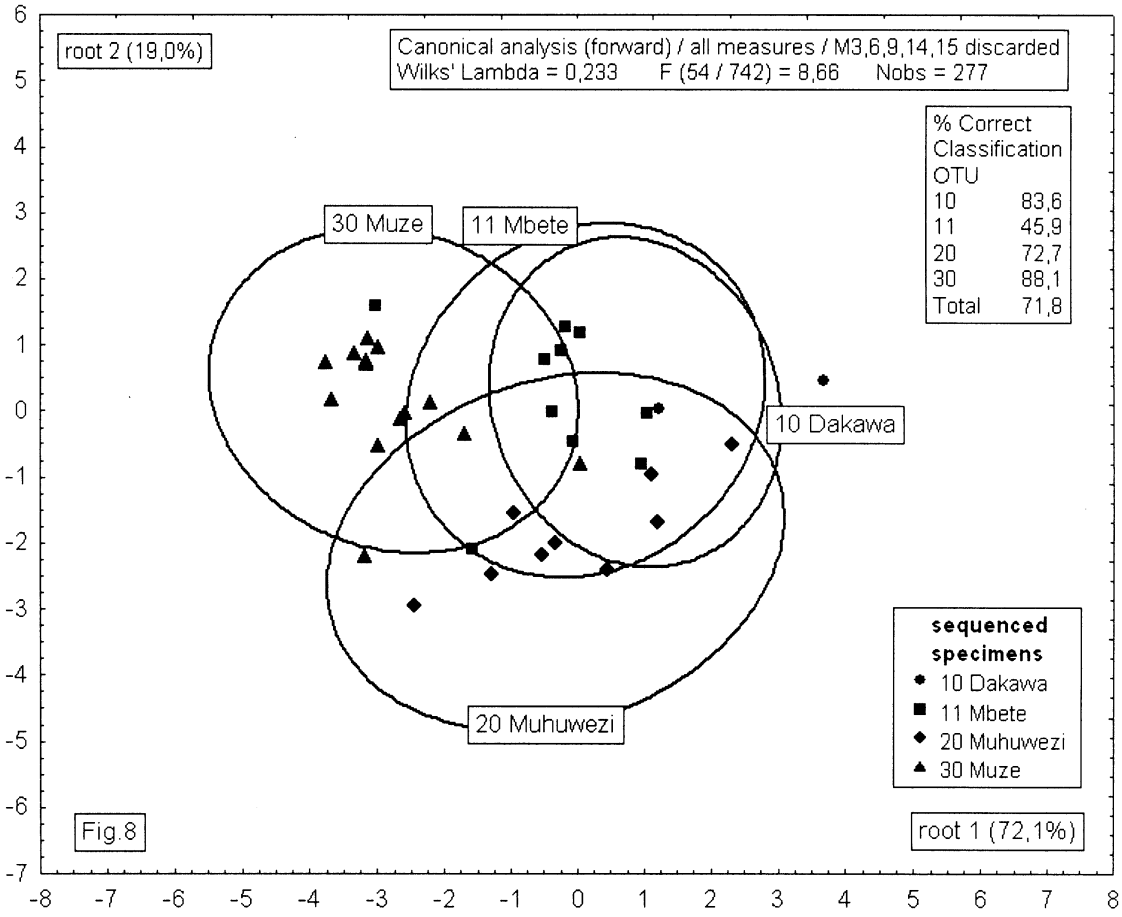


FIGURE 8. Canonical analysis of all the Tanzanian *Acomys* OTU's showing the sequenced specimens.

Species description of *Acomys ngurui* n. sp. (OTU 10, 11, 20)

(Fig. 9a & b)

Type material. Holotype: Adult female collected by Jan Stuyck on 24 October 1988 in Nguru Ya Ndege, Tanzania (RMCA 96.037-5034, ethanol preserved specimen, cranium, alcohol tissue sample, no chromosomal preparation, field number 10340).

Paratypes: 22 specimens from two separate collections listed below **[**alc=specimens in ethanol and cr=cranium; nr= field numbers].

Nguru Ya Ndege, Tanzania

RMCA 96.037-M-5032(ad. fem; alc+cr; nr 10338) (Stuyck; 24/10/1988)
RMCA 96.037-M-5033(ad. male; alc+cr; nr 10339) (Stuyck; 24/10/1988)
RMCA 96.037-M-5035(ad. male; alc+cr; nr 10351) (Stuyck; 25/10/1988)
RMCA 96.037-M-5036(ad. fem; alc+cr; nr 12362) (De Vocht; 18/10/1994)
RMCA 96.037-M-5037(ad. male; alc+cr; nr 12363) (De Vocht; 18/10/1994)
RMCA 96.037-M-5038(ad. male; alc+cr; nr 12364) (De Vocht; 18/10/1994)
RMCA 96.037-M-5039(ad. male; alc+cr; nr 12365) (De Vocht; 18/10/1994)
RMCA 96.037-M-5040(ad. fem; alc+cr; nr 12369) (De Vocht; 19/10/1994)
RMCA 96.037-M-5041(ad. fem; alc+cr; nr 12370) (De Vocht; 19/10/1994)
RMCA 96.037-M-5042(ad. male; alc+cr; nr 12371) (De Vocht; 19/10/1994)
RMCA 96.037-M-5043(ad. fem; alc+cr; nr 12374) (De Vocht; 20/10/1994)
RMCA 96.037-M-5044(ad. male; alc+cr; nr 12377) (De Vocht; 21/10/1994)

Nguru Ya Ndege (Mkundi), Tanzania

RMCA 96.037-M-5008(ad. fem; alc+cr; nr 8674) (Verhagen; 8/06/1988)
RMCA 96.037-M-5009(ad. fem; alc+cr; nr 8675) (Verhagen; 8/06/1988)
RMCA 96.037-M-5010(ad. fem; alc+cr; nr 8679) (Verhagen; 8/06/1988)
RMCA 96.037-M-5011(ad. fem; alc+cr; nr 8726) (Verhagen & Leirs; 8/06/1988)
RMCA 96.037-M-5012(ad. male; alc+cr; nr 8727) (Verhagen & Leirs; 8/06/1988)
RMCA96.037-M-5014(ad. fem; alc+cr; nr 8747) (Verhagen; 5/07/1988)
RMCA 96.037-M-5015(ad. fem; alc+cr; nr 8748) (Verhagen; 5/07/1988)
RMCA 96.037-M-5019(ad. male; cr; nr 9005) (Verhagen & Leirs; 6/07/1988)
RMCA 96.037-M-5020(ad. male; alc+cr; nr 9007) (Verhagen & Leirs; 6/07/1988)
RMCA 96.037-M-5021(ad. not sexed; cr; nr 9028) (Verhagen & Leirs; 7/07/1988)

Habitat. Types were collected at Nguru Ya Ndege, an isolated mountain (highest elevation 1100 m) at a distance of approximately 15 km north of Morogoro. The specimens were collected along the gently sloping side of the mountain that is very dry and covered with wooded savannah. The stony hillside is covered with hilltop forest, scattered miombo and gully forest. This area is frequently exposed to fire. (When specimens were trapped, the area was visibly damaged).

Etymology. named after the type locality, Nguru Ya Ndege.

Diagnosis. This new species was easily distinguished from representatives of the southern *A. spinossisimus* populations (including the type specimen of *selousi* and topo-types of *transvaalensis*) by mtDNA sequences (Fig.4) and craniometric measurements (Fig.8); also see 'Results and Discussion' sections. In addition, craniometric data, allozymes and mtDNA sequences from *Acomys ngurui* allowed us to distinguish it from other Tanzanian *Acomys* populations of the *spinosissimus* group, here represented by OTU 30 that occurred further to the west of the Eastern Arc Mountains (Muze, Inala Tabora, Kanyelele, Kondoa, Magangwe, Manyoni, Matongolo, Mtowisa, Mwanza, Mweneyembe and Zoissa).

While allozyme data and mtDNA sequences indicated that the genetic distance between *A. ngurui* and OTU 30 was significant and comparable with distances among other con-generic murid species, the craniometrical differentiation between *A. ngurui* and OTU 30 was subtle but significant for 13 measurements (analysis of all specimens) or 10 (analysis of type series only) associated with the width of the skull (*ngurui* being the species with the more robust skull).

Interestingly, external body measurements clearly separated *A. ngurui* from other Tanzanian *Acomys* represented by OTU 30 (Table 7). The comparison of all examined specimens reveals that *A. ngurui* was significantly heavier and larger, with a shorter tail and hind foot. Finally, craniometrical and external measurements did not reveal significant levels of sexual dimorphism in *A. ngurui*, the only taxon for which a sufficiently high number of specimens from each sex was available for such an evaluation.

Distribution range. All the specimens listed in OTU's 10, 11 and 20 (Appendix 1) belonged to this species. Figures 1 & 10 illustrated that they mainly occur in Eastern (OTU's 10, 11; East of the Eastern Arc) and Southern Tanzania (OTU 20).

Species description of *Acomys muzei* n. sp. (OTU 30)
(Fig.9a & b)

Type material. Holotype: Adult male collected by Walter N. Verheyen and Jan Stuyck on 20 August 1995 in Muze, Tanzania (RMCA 96.037-M-5069, ethanol preserved specimen, cranium, alcohol tissue sample, chromosomal preparation, field number 13307).

Paratypes: 21 specimens listed below were collected by Walter N. Verheyen and Jan Stuyck [alc=specimens in ethanol and cr=cranium; nr= field numbers].

Muze.

RMCA 96.037-M-5064(ad. male; alc+cr; nr 13302; 20/08/1995)
RMCA 96.037-M-5065(ad. fem; alc+cr; nr 13303; 20/08/1995)
RMCA 96.037-M-5066(ad. fem; alc+cr; nr 13304; 20/08/1995)
RMCA 96.037-M-5067(ad. male; alc+cr; nr 13305; 20/08/1995)
RMCA 96.037-M-5068(ad. not sexed; alc+cr; nr 13306; 20/08/1995)
RMCA 96.037-M-5070(ad. male; alc+cr; nr 13308; 20/08/1995)
RMCA 96.037-M-5071(ad. fem; alc+cr; nr 13309; 20/08/1995)
RMCA 96.037-M-5072(ad. male; alc+cr; nr 13431; 21/08/1995)
RMCA 96.037-M-5073(ad. fem; alc+cr; nr 13435; 21/08/1995)
RMCA 96.037-M-5074(ad. fem; alc+cr; nr 13436; 21/08/1995)
RMCA 96.037-M-5075(ad. fem; alc+cr; nr 13437; 21/08/1995)
RMCA 96.037-M-5076(ad. not sexed; alc+cr; nr 13438; 21/08/1995)
RMCA 96.037-M-5077(ad. fem; alc+cr; nr 13439; 21/08/1995)
RMCA 96.037-M-5078(ad. male; alc+cr; nr 13440; 21/08/1995)
RMCA 96.037-M-5079(ad. not sexed; alc+cr; nr 13441; 21/08/1995)
RMCA 96.037-M-5080(ad. fem; alc+cr; nr 13442; 21/08/1995)
RMCA 96.037-M-5081(ad. male; alc+cr; nr 13443; 21/08/1995)
RMCA 96.037-M-5082(ad. fem; alc+cr; nr 13444; 21/08/1995)
RMCA 96.037-M-5083(ad. fem; alc+cr; nr 13445; 21/08/1995)
RMCA 96.037-M-5084(ad. fem; alc+cr; nr 13446; 21/08/1995)
RMCA 96.037-M-5085(ad. male; alc+cr; nr 13447; 21/08/1995)

Habitat. Muze and other localities where this taxon was collected are typical woodlands and wooded grasslands. The landscape was a hill, which rose above Lake Rukwa (about 10 km from the lake). Vegetation consisted of dry forest (miombo) with open canopy. The plain forming part of the wetlands of Lake Rukwa was dominated by acacia, elephant grass, and typha and cyperus associations towards the lake. At the time of sampling, the area was also partly used for maize and paddy farming.

Etymology. Named after the type locality, Muze.

Diagnosis. This new taxon was easily distinguished from the representatives of the southern *A. spinossisimus* species complex (including the type specimen of *selousi* and topotypes of *transvaalensis*) by mtDNA sequences (Fig.4) and craniometric measurements (Fig. 8); also see 'Results and Discussion' sections.

Moreover, the craniometric data, external body measurements (Table 7) and mtDNA sequences from the new taxon allowed us to distinguish it from other Tanzanian *Acomys* populations occurring to the East of the Eastern Arc Mountains, which we described as *A. ngurui*.

While allozyme data (Fig. 5) and mtDNA sequences (Fig. 4) indicated that the amount of genetic differentiation between *A. muzei* and *A. ngurui* was significant and comparable with observations for other *Acomys* species, the craniometric differentiation between *A. muzei* and *A. ngurui* was subtle but significant for 13 measurements (ana-lysis of all specimens) or 10 (analysis of type series only) associated with the width of the skull (*muzei* being the taxon with the more slender skull).

Nevertheless, the two new species were morphologically distinguishable as traditional external body measurements showed that *A. muzei* is significantly lighter and smaller, with a longer tail and hind foot. Finally, we did not have sufficient specimens to statistically evaluate whether our craniometric and external measurements would indicate sexual dimorphism in *A. muzei*.

Distribution range. All the specimens listed in OTU's 30 and 31 (Appendix 1) belonged to *A. muzei*, as well as the specimens **S2** and **S5**. The available information suggested that *A. muzei* occurred throughout central and western Tanzania, as well as the eastern part of the DR Congo (Fig.1, 10).

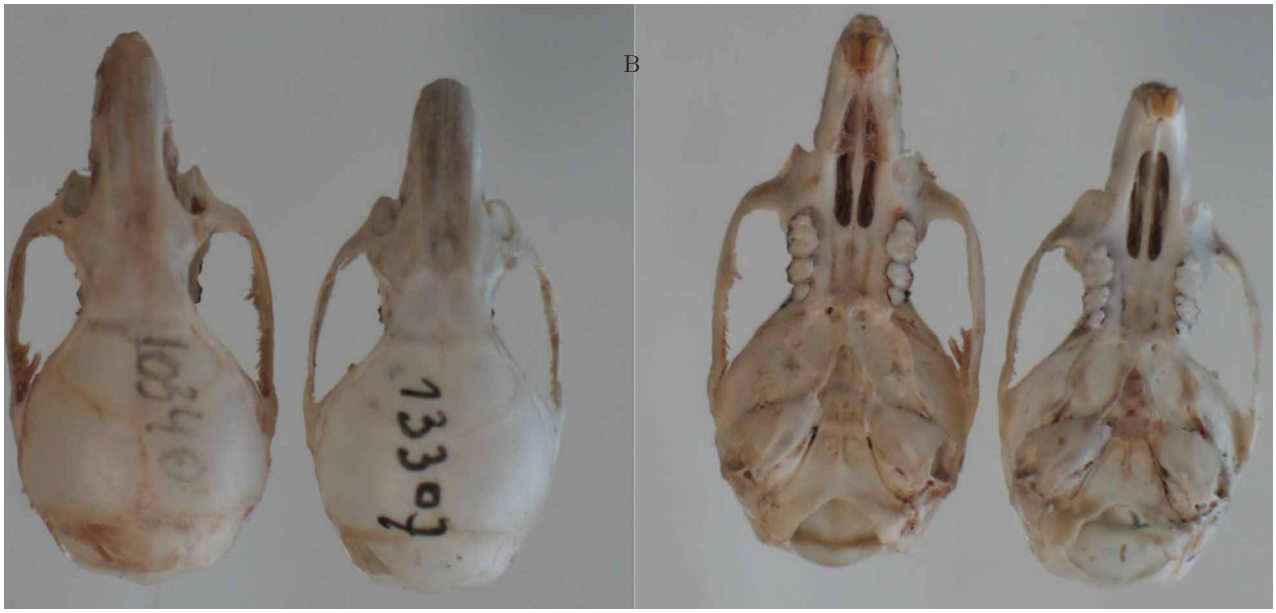


FIGURE 9. (A) Type specimens of *Acomys ngurui* (top) and *Acomys muzei* (bottom). **(B)** Dorsal and ventral view of the skulls of the type specimen of *Acomys ngurui* (left) and *Acomys muzei* (right).

TABLE 6. Measurements of the type and paratype specimens of *A. muzei* **sp. nov**.

LOCALITY	MUSEUM + REG.NR	M1	M2	M3	M4	M5	M6	M7	M8	M9	M10	M11	M12	M13	M14		AGE	SEX
muzei type																		
MUZE	RMCA 96.037-M-5069	24,00	22,05	18,00	11,65	5,10	6,20	6,50	4,45	12,05	2,75	4,15	5,50	1,30	2,70	s	3	M
paratypes																		
MUZE	RMCA 96.037-M-5064	23,90	21,60	17,95	11,15	5,05	6,15	6,40	4,45	11,80	2,55	3,90	5,15	1,30	2,65		2	M
MUZE	RMCA 96.037-M-5065	24,50	22,25	18,80	11,70	5,25	6,40	6,70	4,25	12,15	2,60	3,95	5,30	1,30	2,85		2	F
MUZE	RMCA 96.037-M-5066	24,05	22,05	18,45	11,90	5,15	6,30	6,60	4,25	11,95	2,80	4,05	5,40	1,30	2,80		2	F
MUZE	RMCA 96.037-M-5067	26,75	24,65	20,90	13,35	5,70	7,05	7,55	4,45	13,20	2,90	4,35	5,80	1,35	3,25		3	M
MUZE	RMCA 96.037-M-5068	24,25	22,20	18,80	11,45	5,15	6,30	6,55	4,55	12,40	2,60	4,00	5,45	1,35	2,95	s	2	?
MUZE	RMCA 96.037-M-5070	26,60	24,55	20,60	13,20	5,80	6,90	7,25	4,45	13,00	2,70	4,40	5,70	1,35	3,15	s	3	M
MUZE	RMCA 96.037-M-5071	23,80	21,80	18,05	11,55	5,00	6,00	6,50	4,45	-	2,70	4,10	5,45	1,35	2,75		3	F
MUZE	RMCA 96.037-M-5072	24,45	22,65	18,90	12,35	5,25	6,45	6,80	4,30	12,45	2,75	4,05	5,45	1,35	2,90		2	M
MUZE	RMCA 96.037-M-5073	24,10	21,90	18,25	11,25	5,00	6,30	6,65	4,45	12,20	2,70	4,00	5,50	1,40	2,75		2	F
MUZE	RMCA 96.037-M-5074	24,50	22,05	18,55	12,00	5,35	6,30	6,55	4,45	12,15	2,70	3,90	5,40	1,30	2,75	s	2	F
MUZE	RMCA 96.037-M-5075	24,60	22,40	18,60	11,85	5,25	6,30	6,70	4,35	12,30	2,70	4,00	5,35	1,30	2,80	s	2	F
MUZE	RMCA 96.037-M-5076	25,55	23,55	19,60	12,55	5,35	6,90	7,20	4,55	12,40	2,85	3,90	5,60	1,30	2,95		4	?
MUZE	RMCA 96.037-M-5077	25,90	23,95	20,10	13,10	5,75	7,05	7,45	4,30	12,85	2,85	4,15	5,70	1,35	2,95		3	F
MUZE	RMCA 96.037-M-5078	25,15	23,00	19,20	11,95	5,35	6,60	6,95	4,65	12,70	2,85	4,15	5,60	1,35	2,80		2	M
MUZE	RMCA 96.037-M-5079	24,20	21,75	18,35	11,55	5,35	6,15	6,60	4,55	12,20	2,70	4,00	5,50	1,35	2,75		2	?
MUZE	RMCA 96.037-M-5080	24,65	22,35	18,80	12,10	5,50	6,40	6,70	4,50	12,15	2,65	4,15	5,45	1,40	2,90		2	F
MUZE	RMCA 96.037-M-5081	25,40	23,30	19,50	12,65	5,40	6,55	7,00	4,50	12,65	2,85	4,15	5,45	1,30	2,90		2	M
MUZE	RMCA 96.037-M-5082	23,95	21,75	18,05	11,35	5,00	6,25	6,55	4,40	12,30	2,85	3,95	5,35	1,30	2,70		2	F
MUZE	RMCA 96.037-M-5083	25,90	24,15	20,25	12,95	5,35	6,90	7,40	4,40	12,55	3,00	4,35	5,70	1,35	2,95		4	F
MUZE	RMCA 96.037-M-5084	25,80	23,60	19,95	12,70	5,70	6,85	7,25	4,40	12,75	2,85	4,20	5,75	1,40	2,95		3	?
MUZE	RMCA 96.037-M-5085	24,90	22,60	18,85	11,75	5,45	6,30	6,55	4,45	12,25	2,75	4,15	5,40	1,35	2,75		3	M

continued next page

TABLE 6. (continued)

LOCALITY	MUSEUM + REG.NR	M15	M16	M17	M19	M20	M21	M22	M23	M24	W	HB	TL	HF	EL	AGE	SEX
muzei type																	
MUZE	RMCA 96.037-M-5069	2,40	9,20	3,60	3,85	11,15	1,20	5,05	3,85	6,30	14,0	78	79	16	14	3	M
paratypes																	
MUZE	RMCA 96.037-M-5064	2,15	9,40	3,45	4,00	11,20	1,15	5,00	3,75	6,10	14,0	75	79	16	15	2	M
MUZE	RMCA 96.037-M-5065	2,20	9,75	3,60	4,25	11,25	1,10	5,20	3,80	6,45	17,0	81	95	16	15	2	F
MUZE	RMCA 96.037-M-5066	2,40	9,55	3,55	3,95	11,20	1,20	5,10	3,95	6,15	16,0	82	79	16	14	2	F
MUZE	RMCA 96.037-M-5067	2,50	11,00	3,60	3,95	11,75	1,35	5,65	4,45	6,95	24,0	94	103	17	14	3	M
MUZE	RMCA 96.037-M-5068	2,45	9,50	3,60	4,25	11,50	1,15	5,20	3,95	6,30	16,0	83	87	17	14	2	?
MUZE	RMCA 96.037-M-5070	2,50	10,35	3,70	4,20	11,35	1,35	5,60	4,20	7,05	23,0	97	102	16	13	3	M
MUZE	RMCA 96.037-M-5071	2,30	9,20	3,60	3,90	11,25	1,20	5,10	3,70	6,15	16,0	80	85	16	13	3	F
MUZE	RMCA 96.037-M-5072	2,40	10,00	3,65	4,05	11,30	1,20	5,25	4,00	6,45	18,0	84	84	-	-	2	M
MUZE	RMCA 96.037-M-5073	2,45	9,55	3,50	4,05	11,40	1,25	5,05	3,95	6,20	17,0	84	84	15	14	2	F
MUZE	RMCA 96.037-M-5074	2,40	9,90	3,55	4,10	11,30	1,15	5,05	3,90	6,25	16,0	83	86	15	14	2	F
MUZE	RMCA 96.037-M-5075	2,25	9,70	3,65	4,20	11,55	1,20	5,25	3,80	6,45	17,0	82	85	17	13	2	F
MUZE	RMCA 96.037-M-5076	2,35	10,65	3,50	4,15	11,35	1,20	5,35	4,00	6,35	19,0	86	94	16	14	4	?
MUZE	RMCA 96.037-M-5077	2,55	10,55	3,60	4,10	11,55	1,35	5,35	4,30	6,75	20,0	95	90	16	13	3	F
MUZE	RMCA 96.037-M-5078	2,40	9,85	3,65	4,20	11,85	1,30	5,35	4,20	6,60	18,0	85	93	17	15	2	M
MUZE	RMCA 96.037-M-5079	2,20	9,70	3,60	4,10	11,35	1,15	5,10	3,65	6,10	15,0	84	85	16	13	2	?
MUZE	RMCA 96.037-M-5080	2,40	9,95	3,65	4,05	11,40	1,30	5,20	3,80	6,50	18,0	84	86	16	0	2	F
MUZE	RMCA 96.037-M-5081	2,45	10,20	3,50	4,05	11,65	1,20	5,20	3,95	6,60	18,0	85	87	15	14	2	M
MUZE	RMCA 96.037-M-5082	2,35	9,35	3,55	4,00	11,35	1,15	5,10	3,80	6,10	14,0	75	82	16	13	2	F
MUZE	RMCA 96.037-M-5083	2,45	10,25	3,60	4,30	11,35	1,30	5,35	4,00	6,70	21,0	83	98	16	12	4	F
MUZE	RMCA 96.037-M-5084	2,50	10,15	3,65	4,20	11,50	1,30	5,35	4,20	6,80	-	-	-	-	-	3	?
MUZE	RMCA 96.037-M-5085	2,30	10,05	3,70	4,10	11,45	1,15	5,25	3,80	6,65	16,0	80	87	16	12	3	M

s= sequenced.

FIGURE 10. Inferred distribution ranges of the studied East African *Acomys* species.

TABLE 7. (A) Statistical comparison between the craniometrical and morphological measurements of all *A. ngurui* and *A. muzei*. **(B)** Statistical comparison between the craniometrical and morphological measurements of the types and paratypes of *A. ngurui* and *A. muzei*. Significances of the Student's t-tests: ns: not significant; * p<0.05, ** p<0.01, *** p<0.001.

(A)

ngurui (OTU11)								*muzei* (OTU30)						
	N	Mean	Min.	Max.	SD	CV%	sign.	N	Mean	Min.	Max.	SD	CV%	
M1	228	25,54	23,45	27,35	0,849	3,3	***	47	25,07	23,75	26,75	0,866	3,5	M1
M2	241	23,24	21,20	25,50	0,895	3,9	*	47	22,94	21,55	24,65	0,865	3,8	M2
M3	239	19,57	17,80	21,95	0,844	4,3	*	47	19,26	17,95	20,90	0,813	4,2	M3
M4	241	12,45	10,90	13,80	0,616	4,9	ns	47	12,33	11,15	13,35	0,597	4,8	M4
M5	243	5,48	4,60	6,25	0,327	6,0	ns	47	5,56	5,00	6,40	0,341	6,1	M5
M6	243	6,68	5,80	7,50	0,381	5,7	ns	47	6,58	5,85	7,15	0,319	4,8	M6
M7	242	7,14	6,30	8,60	0,425	6,0	ns	46	7,01	6,40	7,90	0,383	5,5	M7
M8	243	4,65	4,30	5,05	0,146	3,1	***	47	4,49	4,25	4,75	0,114	2,5	M8
M9	238	12,66	11,50	13,90	0,420	3,3	*	45	12,53	11,80	13,20	0,380	3,0	M9
M10	243	3,03	2,55	3,60	0,173	5,7	***	47	2,82	2,55	3,15	0,150	5,3	M10
M11	242	4,32	3,95	4,70	0,147	3,4	***	47	4,20	3,75	4,80	0,195	4,7	M11
M12	243	5,81	5,25	6,40	0,223	3,8	***	46	5,59	5,15	5,90	0,176	3,2	M12
M13	242	1,37	1,20	1,50	0,059	4,3	ns	47	1,36	1,30	1,50	0,058	4,2	M13
M14	243	3,16	2,70	3,65	0,188	5,9	***	47	3,02	2,65	3,55	0,223	7,4	M14
M15	242	2,51	2,10	2,85	0,135	5,4	**	47	2,45	2,15	2,70	0,125	5,1	M15
M16	230	10,01	8,70	11,30	0,514	5,1	ns	47	9,98	9,20	11,05	0,457	4,6	M16
M17	243	3,82	3,45	4,20	0,137	3,6	***	46	3,72	3,45	4,15	0,166	4,5	M17
M19	241	4,24	3,80	4,85	0,172	4,0	***	46	4,09	3,85	4,50	0,147	3,6	M19
M20	242	11,47	10,90	12,20	0,270	2,4	ns	46	11,47	11,05	11,90	0,203	1,8	M20
M21	243	1,30	1,10	1,60	0,092	7,1	ns	46	1,29	1,10	1,55	0,106	8,2	M21
M22	243	5,26	4,80	5,85	0,225	4,3	ns	47	5,23	4,60	5,65	0,217	4,2	M22
M23	242	3,84	3,30	4,30	0,177	4,6	***	47	3,97	3,65	4,45	0,170	4,3	M23
M24	236	6,52	5,70	7,40	0,332	5,1	ns	43	6,43	5,75	7,05	0,279	4,3	M24
W	172	23,2	11	38	4,54	19,5	***	39	19,1	14	30,2	3,43	18,0	W
HB	166	91,4	69	110	7,87	8,6	***	38	86,0	75	97	6,07	7,1	HB
TL	112	76,5	62	90	5,91	7,7	***	35	87,0	78	103	6,78	7,8	TL
HF	165	15,3	12	19	1,03	6,7	***	37	16,5	14	20	1,32	8,0	HF
EL	133	14,0	11	19	1,27	9,1	ns	35	13,6	9	17	1,52	11,2	EL

(B)

ngurui (OTU11, part.)								*muzei* (OTU30, part.)						
	N	Mean	Min.	Max.	SD	CV%	sign.	N	Mean	Min.	Max.	SD	CV%	
M1	22	25,19	23,45	26,60	0,860	3,4	ns	22	24,86	23,80	26,75	0,892	3,6	M1
M2	23	22,87	21,20	24,25	0,946	4,1	ns	22	22,73	21,60	24,65	0,958	4,2	M2
M3	23	19,19	17,80	20,45	0,897	4,7	ns	22	19,02	17,95	20,90	0,878	4,6	M3
M4	23	12,18	11,00	13,25	0,656	5,4	ns	22	12,09	11,15	13,35	0,664	5,5	M4
M5	23	5,41	4,60	6,15	0,359	6,6	ns	22	5,33	5,00	5,80	0,245	4,6	M5
M6	23	6,54	5,90	7,25	0,390	6,0	ns	22	6,48	6,00	7,05	0,318	4,9	M6
M7	22	6,99	6,30	7,70	0,396	5,7	ns	22	6,84	6,40	7,55	0,356	5,2	M7
M8	23	4,62	4,45	4,90	0,121	2,6	***	22	4,43	4,25	4,65	0,100	2,3	M8
M9	21	12,57	11,90	13,65	0,451	3,6	ns	21	12,40	11,80	13,20	0,353	2,8	M9
M10	23	3,04	2,70	3,35	0,164	5,4	***	22	2,76	2,55	3,00	0,111	4,0	M10
M11	23	4,23	3,95	4,55	0,176	4,2	**	22	4,09	3,90	4,40	0,146	3,6	M11
M12	23	5,82	5,45	6,25	0,227	3,9	***	22	5,50	5,15	5,80	0,161	2,9	M12
M13	22	1,39	1,30	1,45	0,044	3,2	***	22	1,34	1,30	1,40	0,035	2,6	M13
M14	23	3,16	2,80	3,55	0,217	6,9	***	22	2,86	2,65	3,25	0,146	5,1	M14
M15	23	2,49	2,10	2,75	0,177	7,1	*	22	2,38	2,15	2,55	0,108	4,5	M15
M16	21	9,87	8,85	10,70	0,490	5,0	ns	22	9,90	9,20	11,00	0,476	4,8	M16
M17	23	3,82	3,55	4,10	0,141	3,7	***	22	3,59	3,45	3,70	0,066	1,8	M17
M19	23	4,25	3,85	4,65	0,183	4,3	**	22	4,09	3,85	4,30	0,120	2,9	M19
M20	23	11,40	10,95	11,75	0,206	1,8	ns	22	11,41	11,15	11,85	0,179	1,6	M20
M21	23	1,36	1,20	1,60	0,096	7,0	***	22	1,22	1,10	1,35	0,077	6,3	M21
M22	23	5,22	4,85	5,80	0,230	4,4	ns	22	5,23	5,00	5,65	0,170	3,3	M22
M23	23	3,85	3,55	4,15	0,176	4,6	ns	22	3,95	3,65	4,45	0,205	5,2	M23
M24	22	6,50	5,70	7,10	0,408	6,3	ns	22	6,45	6,10	7,05	0,283	4,4	M24
W	23	21,8	12	32	5,51	25,3	**	21	17,5	14	24	2,73	15,6	W
HB	21	86,7	69	96	7,99	9,2	ns	21	83,8	75	97	5,68	6,8	HB
TL	16	77,4	62	87	6,74	8,7	***	21	88,1	79	103	7,01	8,0	TL
HF	21	15,4	14	17	0,75	4,8	**	20	16,1	15	17	0,60	3,8	HF
EL	21	13,5	12	15	0,93	6,9	ns	19	13,6	12	15	0,90	6,6	EL

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APPENDIX 1

Measured specimens grouped per Operational Taxonomical Unit (OTU), with indication of the specimens used for electrophoretic studies (e), for karyotyping (k), for sequencing (s), or for which the skull was not measured.

OTU 10
Sex : M (79), F (47), ? (21); age : cl 1 (43), cl2 (40), cl3 (31), cl4 (22), cl5 (10); cl ? (1)

DAKAWA	RMCA	96.036.M.4709(e), 4718(e) - 4736, 4738 - 4776, 4778 - 4783, 4785 - 4788, 4790 - 4799, 4801 - 4802, 4806 - 4816, 4818 - 4821, 4823 - 4826, 4829 - 4836, 4837(k), 4838(k), 4839 - 4843, 4845 - 4848(k), 4849(k), 4850 - 4854, 4855(s), 4856 - 4858, 5483 - 5492, 5494 - 5495, a4.036.M.0047
	ROMA	T50544(s)(k)
	RUCA	6820, 6822, 6825, 6835, 6838 (transferred to Sokoine University of Agriculture)
DAKAWA	RUCA	2273(e) (f)

OTU 11
Sex : M (56), F (40), ? (8); age : cl 1 (10), cl2 (60), cl3 (22), cl4 (6), cl5 (2); cl ? (5)

MKUNDI	RMCA	96.037.M.5008(e), 5009(k)(e), 5010, 5011(s)(k)(e), 5012(e), 5014(e) - 5015(s)(e), 5019(e), 5020(e) - 5021 (type series)
NGURU YA NDEGE	RMCA	96.037.M.5032(e) - 5033(s)(e), 5034(s)(e), 5035(e) - 5044 (type series)
AMANI	BMNH	76.1559 - 63, 340796 - 98
	USNM	340796 - 98
BEREGA	RUCA	TZ 20027(s), 20028(s) - 32,
	RMCA	96.037.M.5063(s)
	MNHN	1998-1611 (s) (f)
KIGURUNYEMBE	RMCA	96.036.M.4859(s)
MBETE	RMCA	96.037.M.4964(e),4965(e), 4966, 4967(e) - 4972(e), 4973, 5005(k) - 5006(k), 5815
	RUCA	3679(e)
MGETA	RMCA	96.037.M.4950 - 4951
MIKESE	RMCA	96.037.M.5045, 5046(s), 5047(s), 5048(s) - 5058
MKUNDI	RMCA	96.037.M.5013(e)(f), 5016(e)(f)-5018(e)(f), 5015(s), 5011(s)
MLALI	RMCA	96.036.M.4959
MORNINGSIDE	RMCA	96.037.M.4952 - 4954, 5813 - 5814
MOROGORO	RMCA	96.036.M.4695(e) - 4701(e), 4702 - 4703, 4704(e) - 4708(e), 4710(s)(e) - 4713, 4715 - 4717(e), 5493
	ROMA	T 998, T 1022, T 2785
MSIMBA	RMCA	96.037.M.5062(s)
SANGASANGA	RMCA	96.037.M.4949(e)
ULANGA	FMNH	57566
ULAYA	FMNH	57561, 57563

OTU 20
Sex : M (13), F (13), ? (3); age : cl 1 (11), cl2 (11), cl3 (4), cl4 (2), cl5 (1); cl ? (0)

CHINGULUNGULU	RMCA	96.037.M.4990(s)(k)(e)
KILWA	AMNH	89756
MNARA	RMCA	96.037.M.4974(s)(k)(e), 4975(e), 4976(k)(e), a4.036.M.0048
MNIMA	RMCA	96.037.M.4980(e) - 4981(s)(e), 4983(s)(k)(e), 4982, 4983(e)(s), 4984, 4985(e), 4987(e) - 4989(e)
MUHUWESI	RMCA	96.037.M.4992(s)(e) - 4993
NAKAHUGA	RMCA	96.037.M.4996(s)(e) - 4999, 5002, 5004(s)(k)(e)
PERAMIHO	RMCA	96.037.M.4994 - 4995(s)(e), 5000, 5003
RUANGWA	RMCA	96.037.M.4977(s)(e) - 79(e)

OTU 30
Sex : M (17), F (23), ? (8); age : cl 1 (1), cl2 (32), cl3 (11), cl4 (3), cl5 (0); cl ? (1)

MUZE	RMCA	96.037.M.5064 - 5067, 5068(s), 5069(s), 5070(s) - 5073, 5074(s), 5075(s), 5085 (type series)
INALA TABORA	ROMA	T50600(s)(k)
KANYELELE	RMCA	96.037.M.4956(s)(e), 4957(s)(e), 4958, 4960(s)(e)
	RUCA	1577(e), 1643(e), 1782(e), 1787(e) no skulls

KONDOA	AMNH	83929
MAGANGWE	RMCA	96.037.M.5023(s)(e), 5024(s)(e), 5025 - 5026(s)(e), 5031
MANYONI	AMNH	205063
MATONGOLO	ROMA	T50003(k)
MTOWISA	RMCA	96.037.M.5086(s)
MWANZA	RMCA	96.037.M.4947 - 4948, 4955
MWEYEMBE	RMCA	96.037.M.5022(e), 5027 - 5028, 5029(s), 5030(e)(s)
ZOISSA	ROMA	T50087(s)(k), T50088(s)(k), T50119(k), T50202(k)

OTU 31
Sex : M (3), F (0), ? (4); age : cl 1 (0), cl2 (3), cl3 (4), cl4 (0), cl5 (0); cl ? (0)

LUBUMBASHI	RMCA	13460, 23361, 23790 - 92, 28930
MUNAMA	RMCA	31535

OTU 40
Sex : M (17), F (10), ? (2); age : cl 1 (0), cl2 (13), cl3 (11), cl4 (1), cl5 (1); cl ? (3)

CHIUTA	USNM	367079, 367081 - 83
FINGUE	USNM	367078, 367349
MUCANHA	USNM	367050 - 55, 367057, 367059, 367061, 367063 - 69, 367071-72, 367074 - 77, 370348

OTU 41
Sex : M (2), F (4), ? (0); age : cl 1 (0), cl2 (4), cl3 (0), cl4 (1), cl5 (0); cl ? (1)

LUANSHYA	UCMVZ	118296 - 98, 118306 - 07, 118320

OTU 50
Sex : M (11), F (8), ? (0); age : cl 1 (0), cl2 (11), cl3 (7), cl4 (1), cl5 (0); cl ? (0)

VILA PAIVA DE ANDRADA	USNM	367120 - 21, 367124, 367127 - 28, 367131, 367135, 367138 - 40, 367143 - 44, 367146 - 50, 367153, 367155

OTU 60
Sex : M (1), F (4), ? (45); age : cl 1 (13), cl2 (15), cl3 (16), cl4 (5), cl5 (1); cl ? (0)

ASSEN	TM	20048
ELLISRAS	TM	19951, 19953, 19984, 19988
LETSITELE	TM	24534 - 38, 24557, 24596 - 97
LEYDSDORP	TM	4835 - 36
MARKEN	TM	24670 - 71, 24676
PAFURI	TM	29821 - 27, 29879
POTGIETERSRUS	TM	23388
ROOSSENEKAL	TM	25353, 25372 - 74, 25388 - 91
RUSTENBURG	TM	23595 - 96, 23611 - 12
SHAMDUNGILA	TM	30577 - 79
THABAZIMBI	TM	20576, 20620 - 21, 20638
VAALWATER	TM	24733, 24786 - 87

Additional sequences

MOTU 11

BEREGA	GenBank	AJ010559 (f) (vide : Baromé *et al.* 2001)
BEREGA	GenBank	AJ010558 (f) (vidé : Baromé *et al.* 2001)

MOTU 30

S2	MBUGANI - CHUNYAROMA		T50673(k), T50676(k) (f)
S5	MWENERONDO	Genbank	AJ010556 (f) (vide : Baromé *et al*. 2001)

MOTU 40

S1	LIWONDE	GenBank	Z96037 (f) (vide : Baromé *et al.* 1998)
S3	MORRUMBALA	RUCA	MO145 (skull; age class 1)
S4	MUTARARA	RUCA	MO131 (skull; age class 0)

MOTU 60

S6	PAFURI	GenBank	Z96068 (f) (vide : Baromé *et al*. 1998)

Type specimens			
Measured			
T1+T2	BUIO	ZMB 1711	*A.spinosissimus*
T3	ESSEX FARM	BMNH 97.1.4.44	*A.selousi*
(=ESSEXVALE)			
Not measured			
T4	ZOUTPANSBERG		*A. transvaalensis*

APPENDIX 2

Relative growth (%) of males and females of OTU 10 (Dakawa) for age classes (1+2) versus (3+4). Formula : 100*(age(3+4)-age(1+2))/age(1+2). Only significant differences are shown.

	M	F
GRLS	3.06	4.90
PRCO	4.05	6.29
HEBA	5.01	6.85
HEPA	4.38	6.29
PAFL	6.07	6.88
DIA1	6.69	8.84
DIA2	6.33	9.79
INTE	ns	ns
ZYGO	2.61	3.55
PALA	2.14	3.77
UPTE	2.39	3.37
UPDA	3.47	4.36
M1BR	ns	ns
ZYPL	2.33	7.85
BNAS	ns	ns
LNAS	4.98	6.26
LOTE	ns	ns
BULL	ns	-3.40
BRCA	1.17	ns
DINC	3.39	8.16
ROHE	2.52	6.02
ROBR	2.39	5.16
PCPA	3.16	6.19