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Correspondence analysis of shell morphology in the African freshwater snail *Biomphalaria pfeifferi* (Kraus 1848) (Pulmonata: Gastropoda)

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A correspondence analysis of shell measurements taken from 521 widely dispersed specimens of the African aquatic pulmonate snail *Biomphalaria pfeifferi* suggests the existence of eight morphological groups. These groups appear to relate to either ecophysiological factors or to factors associated with the stability of the freshwater system rather than to aspects of geographic distribution and genetic isolation.

ADDITIONAL KEY WORDS:—Morphometry – Africa – climatic cline.

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INTRODUCTION

The evolutionary development of the Afro- and Neotropical taxon *Biomphalaria* probably occurred 135 million years ago (Myr) (Meier-Brook, 1984). Two main scales of adaptive radiation are possible. Firstly, there is a scale of space, exemplified by a widely spread species such as *Biomphalaria pfeifferi* (Krauss 1848). Secondly, there is a scale of time, exemplified by species which occur in habitats of widely varying stability; in the stable African Great Lakes for example, *Biomphalaria* has diverged into a wide range of species including *Biomphalaria smithi* Preston 1910, *Biomphalaria choanomphala* (Martens 1879) and *Biomphalaria stanleyi* (Smith 1888).

There is however, disagreement about the evolutionary processes involved in the radiation of such freshwater snails. For example, interpretations based on both ecophenotypic differentiation and punctuated equilibria have been proposed for the evolution of prosobranchs and pulmonate snails in the African Great Lakes (Williamson, 1983; Fryer, Greenwood & Peake, 1983; Gould, 1983).

The genus *Biomphalaria* has been taxonomically well-worked (Mandahl Barth, 1958; Brown, 1980; Henriksen & Jelnes, 1980; Jelnes, 1980). At subspecies level however, much is still not understood. For example, African *B. pfeifferi* show signs of genetic stabilization (Jelnes, 1980) whereas infra-specific differentiation appears to be occurring in the *Bulinus africanus* group of freshwater snails (Kristensen, Frandsen & Christensen, 1987).

It is possible that the *Biomphalaria* genus shows morphological variation firstly in relation to geographical distribution (*B. pfeifferi*) and secondly in relation to time (endemic species). Our aim was therefore, to investigate the degree of spatial and temporal genetic distancing of *B. pfeifferi* across a climatic cline in Africa, as exemplified by shell morphometry.

MATERIAL AND METHODS

A total of 521 specimens of *B. pfeifferi* in the collections of the Museum National d'Histoire Naturelle de Paris were examined; they comprised samples of approximately 30 individuals identified by D.S. Brown of the British Museum of Natural History in London (see Appendix).

Thirteen variables (Fig. 1) were measured on the shell (Rousseau, 1985) and comprised: HTA LTA—height and width of the aperture; LTC—total shell width; ARS and ARI—heights of the upper and lower parts of the whorl opposite the aperture; aa′, cc′, ee′, dd′, a′c′ and d′b describe the aperture, where HTA = cc′ + d′d and LTA = a′c′ + c′b. Measurement took place at U.A. C.N.R.S. 157 in Dijon by means of an orientable stage, binocular lens connected to a video camera, T.V. monitor, Numonics digitizer and a Victor 256K microcomputer programmed in BASIC. Data files were transferred to the Centre de Calcul de l'Université de Bourgogne, where multivariate statistical methods

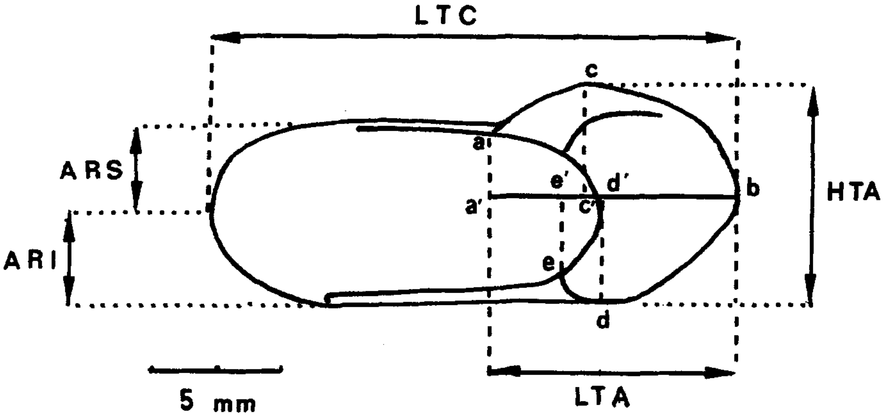

Figure 1. Morphometric parameters measured on each shell of *Biomphalaria pfeifferi*.

including correspondence analysis (Benzecri, 1973) were used. The mean and standard deviation of each variable is given in Table 1.

RESULTS

Correspondence analysis allows the columns (in this case, morphometric variables) and rows (in this case, 521 shells) of a data matrix to be studied. Two point clouds are obtained; they have identical factorial axes and can therefore be superimposed. It is always possible that the results reflect a relatively small number of samples taken over wide geographic area. However, the first three factorial axes explained 72.38% of the total variance, comprising respectively 44.95, 14.06 and 13.38%; this implies that much of the variability is statistically explained in the analysis.

Assuming that all 13 morphometric variables were identically involved, their theoretical contribution to the general variability is 1/13 = 0.077 or 7.7%. All correspondence values above this theoretical threshold contribute disproportionately to the total variability. Table 2 shows the contribution of each variable as a percentage. The closer the correlation coefficient gets to unity,

TABLE 1. Mean (mm) and standard deviation of each variable

	Mean	Standard deviation
ARS	1.956	0.426
ARI	1.222	0.477
LTC	14.689	3.401
HTA	5.482	1.217
LTA	5.643	1.304
a'c'	2.868	0.937
c'b	2.766	0.734
aa'	1.313	0.520
cc'	2.081	0.538
ee'	2.323	0.775
dd'	3.392	0.876
e'd'	1.949	0.658
d'b	3.812	0.914

TABLE 2. Correspondence analysis of *B. pfeifferi*. Only values higher than the theoretical threshold (1/13 = 7.7%) are reported. The degree of relationship is expressed by the correlation coefficient (r). Values close to unity indicate good representation of the parameters on each factorial axis. CTR = percentage contribution

	Factor 1		Factor 2		Factor 3	
	CTR%	r	CTR%	r	CTR%	r
aa′	−21.7	0.735				
c′b′	−8.8	0.408				
e′d′	−8.4	0.428				
ee′	+27.8	0.888				
dd′	+10.9	0.752				
a′c′		0.208	−18.2	0.249	−37.7	0.492
LTA		0.034	−15.3	0.603		
cc′		0.289	−13.6	0.385		
LTC		0.053	+25.1	0.659		
d′b′		0.019	+15.3	0.449		
ARI		0.198		0.096	−8.8	0.139
c′b		0.408		0.022	+26.1	0.363

the better is the representation of each variable on the factorial axis. The relatively high proportion of significant correlation coefficients again suggests that the results are not an artefact of sample size.

B. pfeifferi shows great morphological variability, especially in the region of the aperture, which can be strongly asymmetric. This can be a centrifugal asymmetry, where the dimension c′b is proportionately larger than a′c; conversely, it can be centripetal where a′c is relatively greater than c′b. On factorial axis 3 (Fig. 1), the orientation of the cloud of points is governed by a′c′ for the negative part of the coordinate (negative pole), and by c′b for the positive part of the coordinate (positive pole). On axis 2, the orientation of the cloud of points is due to a strong contribution from widths d′b and LTC on the positive pole, and widths LTA, a′c′ and height cc′ on the negative pole. Shells with high values of LTA (width of the aperture) contribute proportionately higher values of a′c′ and cc′. On axis 1, there is opposition between aa′ on the negative pole, and ee′ and dd′ on the positive pole. These three variables characterize the aperture, whose form may result from either a regular or variable ontogenetic development (Fig. 2).

Variables which make the strongest contribution can be used to form groups in which the majority of individuals show relatively strong values (Table 3). These results can be decomposed into a vectoral space of eight morphological

TABLE 3. Distribution of the parameters on the first three factors determining the eight morphological groups

1	ee′ dd′	(1+)	LTC d′b	(2+)	c′b	(3+)
2	ee′ dd′	(1+)	LTC d′b	(2+)	a′c′	(3−)
3	ee′ dd′	(1+)	LTA a′c′ cc′	(2−)	c′b	(3+)
4	ee′ dd′	(1+)	LTA a′c′ cc′	(2−)	a′c′	(3−)
5	aa′	(1−)	LTC d′b	(2+)	c′b	(3+)
6	aa′	(1−)	LTC d′b	(2+)	a′c′	(3−)
7	aa′	(1−)	LTA a′c′ cc′	(2−)	c′b	(3+)
8	aa′	(1−)	LTA a′c′ cc′	(2−)	a′c′	(3−)

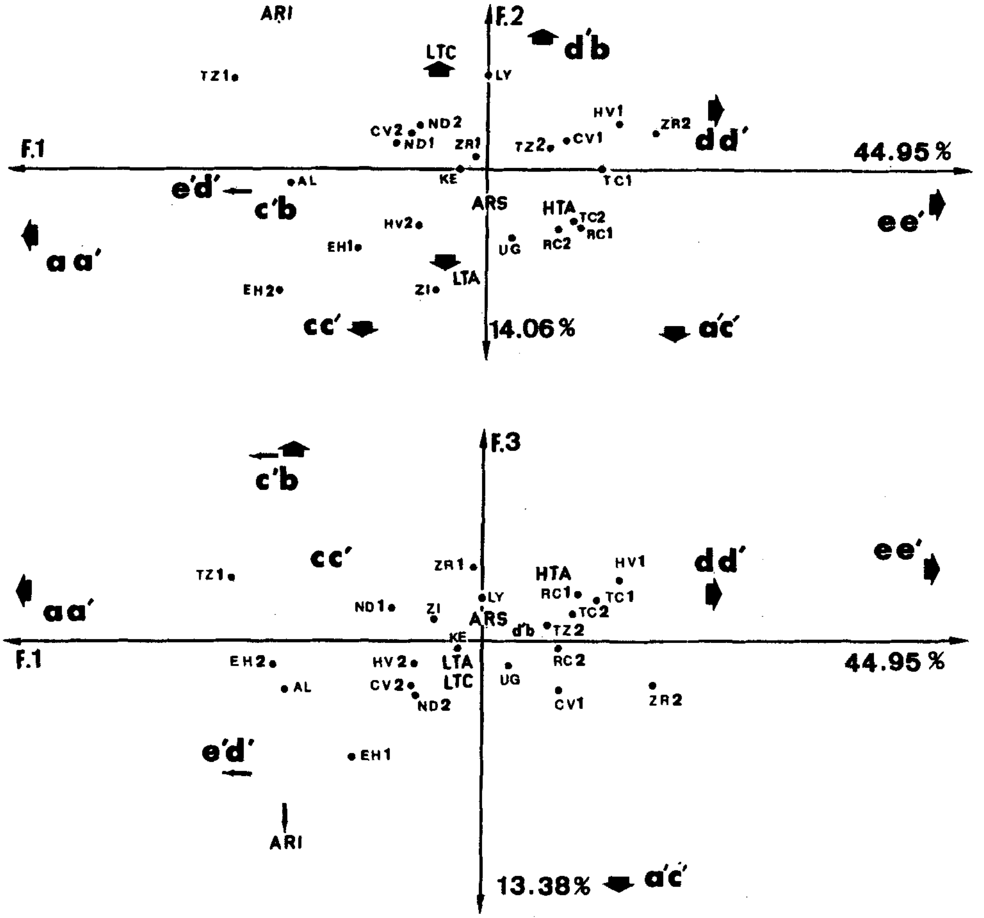

Figure 2. Results of correspondence analysis of *Biomphalaria pfeifferi* populations showing a plot of the morphological parameters (columns) and of the centre of gravity of each population on the first two factor planes (axes 1–2 and 1–3). Wide arrows correspond to the highest contribution to the general variability.

assemblages in three dimensions, made up of positive and negative co-ordinates for each factor. This is achieved by taking account of the average centre of gravity of each population, and the total variance explained by the three major axes. A summary of the partition of the centres of gravity is shown in Table 4.

Table 4. Geographical distribution of the morphological groups. These results should be compared with Fig. 3.

Morphological group	Factor	Location
1	1+,2+,3+	Upper Volta, Tibesti, Tanzania
2	1+,2+,3−	Ivory Coast, Zaire
3	1+,2−,3+	CAR, Chad, Libya
4	1+,2−,3−	CAR, Uganda
5	1−,2+,3+	N. Cameroon, Zaire, Tanzania, Kenya
6	1−,2+,3−	Ivory Coast, North Cameroon
7	1−,2−,3+	Zimbabwe
8	1−,2−,3−	Upper Volta, Ethiopia, Algeria

DISCUSSION

Morphologial variation of the shell in relation to spatial distribution

The eight morphological groups of *B. pfeifferi* show a patternless geographic distribution. At neither the continental nor local level can a cline of variation be seen (Fig. 3). For example, the morphological groupings include geographically separate and distant populations, such as those of Upper Volta and Tanzania. Conversely, a local geographic association often includes groups with quite different morphology. For example, Group 8 includes representatives of Upper Volta and Central Saharan relict populations such as those of Algeria, which

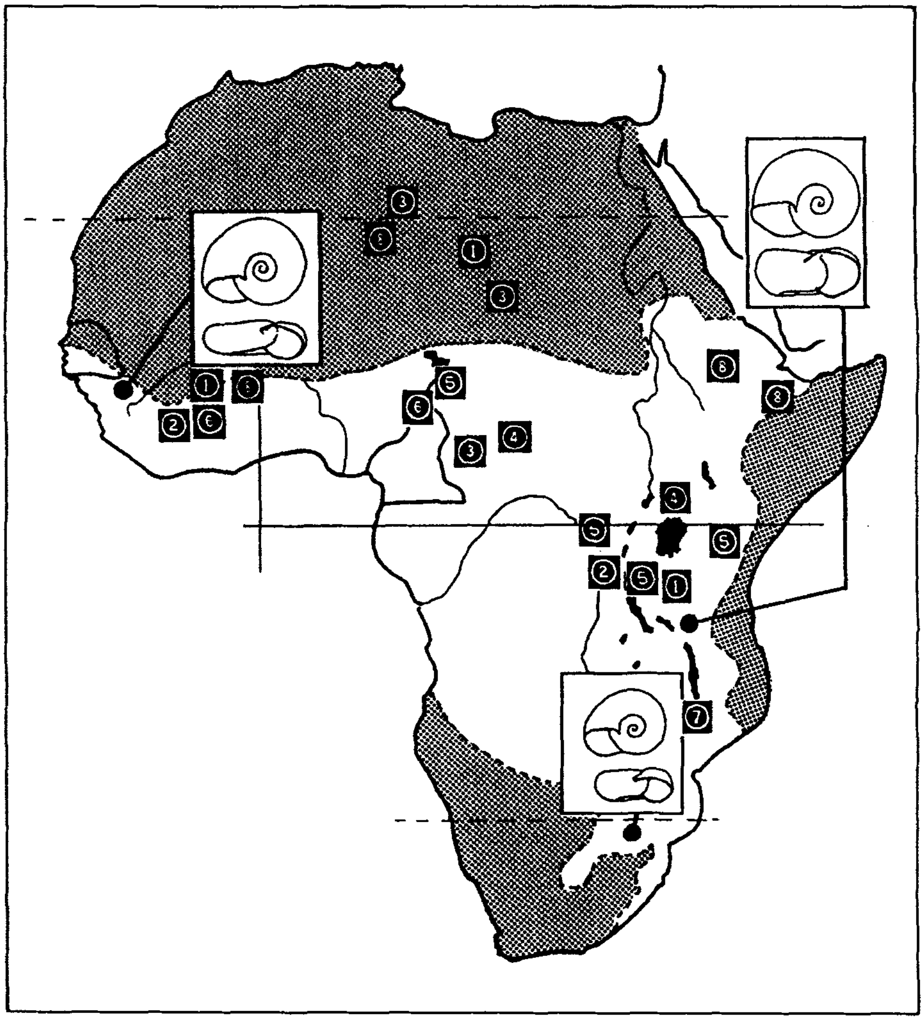

Figure 3. Geographic locations of the populations measured within the current distribution of *Biomphalaria pfeifferi* (modified after Brown, 1980). The numbers correspond to the different morphological groups determined by the correspondence analysis. Inset shell shapes show three typical morphotypes and their locations. Note the sub-keel or the slight deflection of the aperture. Except for the isolated Saharan relict populations, the distribution is mainly Central African (the white area).

have become well-adapted to Sahelian conditions since the withdrawal of the *Biomphalaria* taxon at the end of the Pliocene (Meier Brook, 1984).

The Sudanese domain has a wider range of niches than the Sahelian domain and more morphotypes appear to have evolved in the former. This suggests either reproductive isolation of some populations or ecophysiological responses to a greater range of ecological conditions.

In the Yaounde area of Cameroon, geographical variation in shell thickness and aperture height appears to be an ecophysiological response to the degree of mineralization in the catchments, and it does not correlate with genetic structure and differentiation estimated from protein polymorphisms (Dupouy & Mimpfoundi, 1986; Mimpfoundi *et al.*, 1986).

There is a cluster of morphological groupings in the area of the African Great Lakes, where Group 5 is the most frequently represented morphological group. Certain shell characters, such as the dorso-ventral blunt keel and the deflection of the aperture in relation to the axis of coiling (Fig. 3), are also shown by endemic, sympatric species of *Biomphalaria* species in the Great African Lakes. For example, *B. stanleyi* Mandahl Barth 1958 and *B. choanomphala* Mandahl Barth 1958 of Lakes Victoria and Albert have a sub-keel and *B. smithi* Preston 1910 of Lake Edward has a deflected aperture. These may be adaptations to wave action on the shores of the Lakes since for example, aquatic gastropods are known to show changes in shell morphology which might be related to wave action (Arthur, 1982).

Morphological variation in the shell in relation to time

The general distribution of *Biomphalaria* is inter-tropical but the group appears to have become extinct in Europe at the end of the Pliocene. Since then, *Biomphalaria* species have been limited to regions south of the Tropic of Cancer, on the borders of the Saharan desert and on the high, arid Sudan-Ethiopian plateaux. These refuges emphasize this period of contraction in distribution, especially in the central Sahara (Tassili, Tibesti—Fig. 4). Thus the original Gondwanaland stock appears to be more diverse in the equatorial regions than towards the tropics. This gradient of species reduction can also be seen in other groups of continental molluscs (Brown, 1980). Aridity and frost have helped to push the taxon south of the Sahara but its ecological plasticity allows it to survive in specific Sahelian habitats, particularly in geographically temporary water pools. In this way, *B. pfeifferi* is a good representative of its taxon; its high ecological valency is mainly due to its capacity to aestivate and also self-fertilize (Mimpfoundi, 1990). It capitalizes on the best aspects of the poor resources available to it in oligotrophic environments, although this leads to a low level of heterozygosity (Jelnes, 1980; Mimpfoundi *et al.*, 1986). Conversely, it has adapted to low levels of mineralization in the perennial rain forest (Dupouy & Mimpfoundi, 1986) and has colonized the margins of the Great African Lakes.

The parallel evolution of the shell (carination and deflection of the aperture) between species of *Biomphalaria* may result from two selection pressures. On one hand, the permanence of the East African lakes may have produced stable shell morphology in endemic species. In this way, time could be a major factor in the development of endemicity in the Great African Lakes. On the other hand, in *B.*

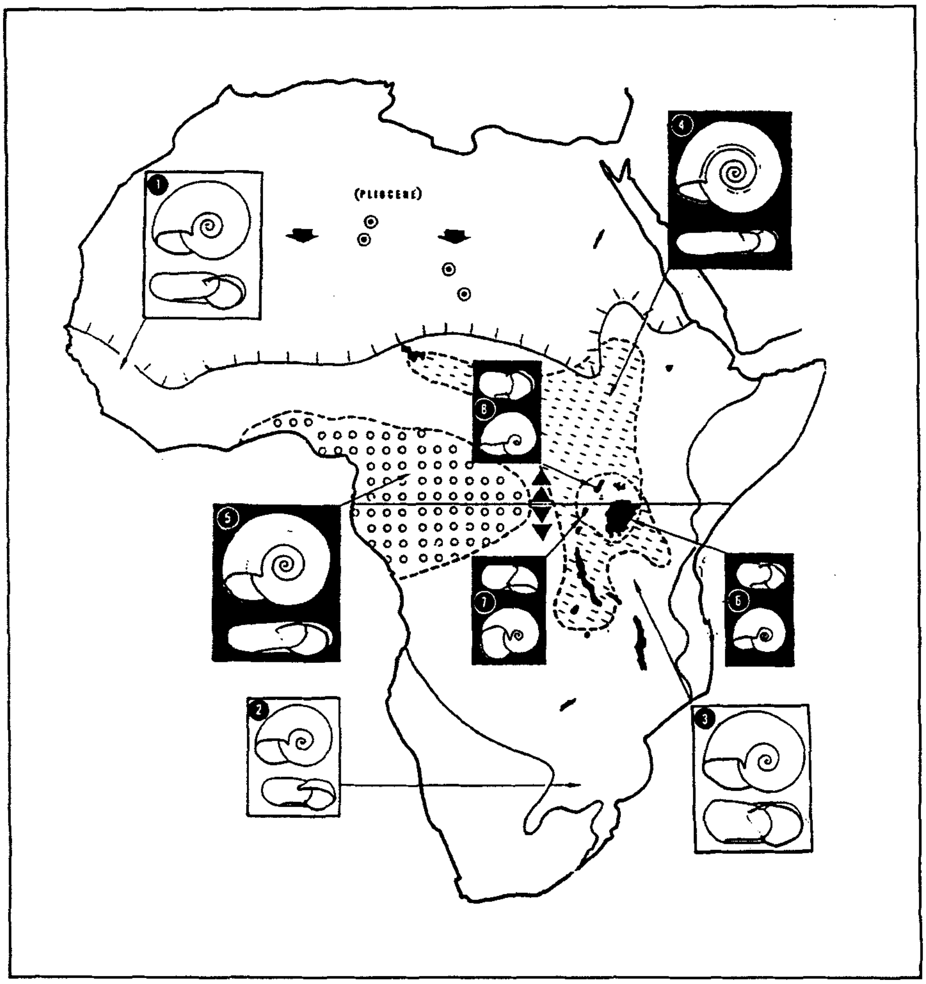

Figure 4. Distribution of the sympatric *Biomphalaria* species in Africa.
1,2,3 = *B. pfeifferi*. The geographic area is limited by a continuous line. The withdrawal of the species during the Pliocene is indicated by large, wide arrows. Saharan refuges are shown as dotted circles.
4 = *B. sudanica*. The geographic area is indicated by broken lines.
5 = *B. camerunensis*. The geographic area is indicated by circles.
6,7,8 = Great Lakes species including *B. choanomphala* (6) Lake Victoria); *B. smithi* (7) (Lake Edward); *B. stanleyi* (8) (Lake Albert). Note the accentuated keels or deflection of the aperture. Snail species diversity increases from the equator (black arrows).

pfeifferi, selection pressures may have caused morphological divergence into eight groups. However, the situation is further complicated by the apparent absence of correlation between shell morphology and a genetic component in certain demes of *B. pfeifferi* studied in Cameroon (Mimpfoundi *et al.*, 1986; Dupouy & Mimpfoundi, 1986). Unfortunately, the functional relationships of snail shells are not well understood. For example, there is little relationship between shell amino acid composition and shell shape (Dussart, 1983), and Evans (1989) found little relationship between calcium content of freshwater gastropod shells and the calcium concentration of the local environment.

CONCLUSION

The data from these shells of *B. pfeifferi* show no obvious morphometric pattern across the African continent. However, there are eight morphological groups whose distribution in certain geographical areas suggests a relationship between shell morphology and ecophysiological adaptation. Ecological factors may affect shell morphology through phenotypic plasticity or through effects of local natural selection. In addition, environments which have been stable for long periods appear to have species and populations with their own characteristics, such as blunt keels and deflected apertures in the aquatic pulmonates of the African Great Lakes. The high ecological valency and morphological variability of *B. pfeifferi* shown in this study emphasizes that this parasitologically important freshwater snail has not yet given a clear picture of its genetic and environmental relationships.

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APPENDIX

Sources of material

Date of identification by D. S. Brown is shown in brackets. Second bracket shows number of specimens measured.

Libya (Fezzan Ghat): collector Dr Jacquemin 1951 (10/9/80) (30)

Kenya (Nairobi region): coll. J. Millet 1952 (8/9/80) (30)

Zaire
  (1) Bukawu: coll. G. Ranson, May 1957; (10/9/80) (29)
  (2) Katana, near Iwins by Bukawu: coll. G. Ranson May 1957 (10/9/80) (29)

Chad
  (1) Faya Largeau: coll. Dr Taufflieb 5/8/60 (10/9/80) (30)
  (2) Tibesti: example of *Biomphalaria rupellii* (10/9/80) (30)

Central African Republic
  (1) Bossangoa district: coll. Bouthillier (31/3/81) (30)
  (2) Oubangui-Chari (Marigot Koyali Bozoum): coll. Dr Gand (11/9/81) (24)

Uganda (Jinja): coll. Cridland 1954 (8/4/81) (15)

North Cameroon
  (1) Maroua-Mogazang: coll. Mouchet 1960 (6/4/81) (11)
  (2) Maroua-Djidoura: Mouchet 1960 (6/4/81) (30)

Ivory Coast
  (1) Fer Kessedougou: coll. Binson 1956 (1/9/82) (30)
  (2) Korhogo: coll. Binson (2/7/82) (30)

Upper Volta
  (1) Houndje Village, Bodo-Dioulasso Circle: coll. Dr Sanssaricq 1958 (29/6/82) (28)
  (2) Santidougou village, Bodo-Dioulasso Circle: coll. Dr Sanssaricq 1958 (29/6/82) (30)

Ethiopia
  (1) Gondar stream (nr. Public Health College): coll. A. Najjan 18/3/56 (11/9/82) (9)
  (2) Harrare Creek: coll. A. Najjan (10/9/82) (5)

Zimbabwe (Inyayadzi irrigation canal): coll. Alves 21/10/52 (2/4/81) (25)

Algeria (S. East of Tassili des Ajjer): coll. Vermeil 1951 (10/9/80) (16)

Tanzania
  (1) Tanganyika stream 13 miles N. of Tunduma: coll. Alves 1955 (1/4/81) (30)
  (2) 13 miles N. of Tunduma on the Great North Rd: coll. Le Roux, Gray, Clarke (8/4/81) (30)