An updated distribution map and additional museum voucher specimens of the genus *Cistugo* (Chiroptera: Cistugonidae), with a review of the distinction between *Cistugo seabrae* Thomas, 1912 and *C. lesueuri* Roberts, 1919, and aspects concerning their allopatric distribution

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While recent work focused on resolution at the family level, and led to the suggestion of a new family for the genus *Cistugo,* the validity of *Cistugo lesueuri* still remains unresolved. We provide a review of the classification of *C. lesueuri* and *C. seabrae,* and an updated distribution map for the genus including new localities for each species. Two measurements initially provided to distinguish the species are revisited, and suggest the distributions of *C. lesueuri* and *C. seabrae* may not be allopatric, as had previously been assumed.

Keywords: *Cistugo lesueuri, Cistugo seabrae,* Species Classification, Distribution.

INTRODUCTION

The results of recent molecular work by Lack *et al.* (2010) led them to propose that *Cistugo* Thomas, 1912 should be placed in its own family, the Cistugidae. The African Chiroptera Report (ACR) (2011), however, points out the correct form of the new family name is Cistugonidae. Combining mitochondrial and nuclear DNA information with more complete taxonomic sampling enabled Lack *et al.* (2010) to confirm earlier karyology (Rautenbach *et al.,* 1993) and molecular results (Bickham *et al.,* 2004; Stadelmann *et al.,* 2004; Eick *et al.,* 2005) that had indicated the genus *Cistugo* occurs basally to all other vespertilionids and is separated from other vespertilionid genera by a primitive karyotype and a high genetic distance. Historically, *Cistugo* has been synonymized with *Myotis* Kaup, 1829, albeit in some instances as a separate subgenus to *Myotis,* in the following species checklists: Ellerman and Morrison-Scott (1951), Ellerman *et al.* (1953), Ellerman (1954), Ellerman and Morrison-Scott (1966), Hayman and Hill (1971), Honacki *et al.* (1978), Honacki *et al.* (1982), Meester *et al.* (1986), Corbet and Hill (1991), Koopman (1993), Herselman and Norton (1985), Koopman (1994), and Taylor (2000). However, more recently, on the basis of diploid and fundamental chromosome number (Rautenbach *et al.,* 1993) and mitochondrial cytochrome-*b* data (Bickham *et al.,* 2004; Stadelmann *et al.,* 2004, Rautian *et al.,* 2006), *Cistugo* was recognized as a valid genus in the species lists of Bronner *et al.* (2003), Simmons (2005), Monadjem *et al.* (2010), and the ACR (2011).

The recorded differences of *Cistugo* relative to other vespertilionids are that they have an ancestral diploid number ($2n = 50$) with an all-acrocentric autosomal complement, and a fundamental chromosome number of FN = 48 (Rautenbach *et al.,* 1993). Based on mitochondrial cytochrome-*b* data, Stadelmann *et al.* (2004: 185) found *Cistugo* genetically distinct from other *Myotis* at >20% uncorrected genetic distance, while Bickham *et al.* (2004: 335) found an average *p*-distance between *Cistugo* and *Myotis* of 21%. Using nuclear genes Eick *et al.* (2005) identified two distinctions between *Cistugo* and all the other vespertilionids they examined; *Cistugo* has a unique insertion in PRKC1 and lacks a unique deletion in the SPTBN intron, neither of which were found in the other vespertilionids they examined. Lack *et al.* (2010) also found a single 18-bp indel in exon 6 of the nuclear exon, DMP1, gene in *Cistugo* that was not found in any other Vespertilionidae sequenced.

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Observed morphological differences of *Cistugo* relative to *Myotis* include their usually having glands present on the wings (Meester *et al.,* 1986; Simmons, 2005). Roberts (1919: 112), however, did not observe glands on the holotype of *C. lesueuri,* and since the skin of the holotype is dried with the wings folded against the body it has subsequently not been possible to re-assess this character from the specimen. Shortridge (1942: 79) also mentioned in the account of individuals of *C. lesueuri* caught at the Hex River Valley Estate near Citrusdal that 'In dry skins these glands become absorbed and lost to sight'. In *Cistugo* the anterior pair of premolars is smaller than in other *Myotis,* and the large upper premolar has a better developed antero-internal cusp than in other *Myotis* (Thomas, 1912). Stadelmann *et al.* (2004: 178), however, incorrectly reported this character as relating to the lower fourth premolar, rather than the upper fourth premolar.

Two species are described within the genus *Cistugo: C. seabrae* Thomas, 1912 and *C. lesueuri* Roberts, 1919. When Roberts described *C. lesueuri* no other records of *C. seabrae,* other than the holotype, were known. Roberts (1919: 112) described *C. lesueuri* as differing 'most markedly from *Cistugo seabrae* in size, the cranial and external characters seemingly differing but little; there appear to be no glands on the wing, however, and the colouration is somewhat different, both of which may eventually prove to be of little importance'. *Cistugo seabrae* was described from Mossamedes in Angola (Thomas, 1912), whereas *C. lesueuri* was described from Lormarins [= L'Ormarins, Franschhoek Valley], Paarl District, in the Western Cape of South Africa. Corbet and Hill (1980) synonymized *C. lesueuri* with *C. seabrae,* given the limited distinction between the two taxa other than size and colour. Hayman and Hill (1971) and subsequent authors (Honacki *et al.,* 1982; Meester *et al.,* 1986; Taylor, 2000; Simmons, 2005; Monadjem *et al.,* 2010), however, retained *C. lesueuri* as a separate species from *C. seabrae,* although they usually indicated some reservation about the validity of *C. lesueuri,* pointing out the taxonomic relationship between these two species requires revision.

The classification proposed to distinguish the two species was initially based on measurements of each of the holotype individuals, thus according to Roberts (1954) the holotype of *C. seabrae,* which was known at the time from Mossamedes in Angola to Goodhouse on the Orange River was smaller, with a forearm and greatest skull length of 32.5 mm and 13.2 mm, respectively; by contrast, the holotype of the larger *C. lesueuri* measured 34.5 mm and 14.4 mm. As new specimens of these relatively rare species have been collected and reported (Smithers, 1983; Herselman and Norton, 1985; Lynch, 1994; Watson, 1998; Taylor, 2000; Seamark and Kearney, 2006; Monadjem *et al.,* 2010) the known distributions of the species have expanded, and ranges of measurements have been introduced to the key characters to encompass the variation introduced by additional specimens. Thus Hayman and Hill (1971) identified *C. seabrae* as being distributed from Angola, Namibia and northwestern Cape Province, and having a smaller forearm (32.5–32.9 mm); while *C. lesueuri* was recorded as being distributed in the southwestern Cape Province (also from Ceres), with a larger forearm of 34.5 mm. The aspects presented in the classification of Meester *et al.* (1986) were the same as those in Hayman and Hill (1971), with the distribution for *C. seabrae* given as the northwestern Cape Province, western Namibia and southwestern Angola, while that of *C. lesueuri* was recorded as the southwestern Cape Province, from the Franschhoek area north to Citrusdal, and northeast to Beaufort West and Mazelsfontein. Taylor (2000) repeated the same forearm range for *C. seabrae* as previous authors (Hayman and Hill, 1971; Meester *et al.,* 1986), but reduced the minimum size for *C. lesueuri* to 34 mm, and introduced colour differences as indicated in the original species descriptions, the dorsal colour of *C. seabrae* being described as a dull greyish-yellow, relative to the honey-yellow dorsal colour in *C. lesueuri.* The distribution reported by Taylor (2000) for *C. seabrae* was similar to that given by Hayman and Hill (1971) and Meester *et al.* (1986), i.e., Northern Cape, Namibia, southwestern Angola, but was more extensive for *C. lesueuri,* i.e., besides the distribution in the Western and Northern Cape, the species was also reported to extend into the Free State in South Africa and Lesotho.

Shortridge (1942: 37, 79) and Herselman and Norton (1985: 94, 96) comment on differences between *C. lesueuri* and *C. seabrae* in the number, shape, size and position of the glands, those in *C. lesueuri* being narrower, less thick and a shorter distance to the middle of the radius than those in *C. seabrae,* and in *C. lesueuri* they may only occur on one wing, whereas in *C. seabrae* they are present on both wings and in some cases there are two glands rather than one gland present on each side. However, these characters associated with the wing glands have not been extensively evaluated across all specimens, possibly in part because the glands are not always visible on the specimens.

Based on the complete mitochondrial cytochrome-*b* sequence Stadelmann *et al.* (2004: 185) reported the 'two closely related species differed by less than 30 substitutions from each other', and that within each species two individuals of *C. lesueuri* showed no substitutions, while two individuals of *C. seabrae* differed by two substitutions. Bickham

et al. (2004: 335) also sequenced the complete cytochrome-*b* gene and showed inter-species *p*-distances between *C. lesueuri* and *C. seabrae* averaging 2.15%, with intra-species *p*-distances averaging 0.18% and 0.29%, respectively. Bickham *et al.* (*loc. cit.*) also reported that inter-species *p*-distances between *Cistugo* and *Myotis* averaged 21% , and those within *Myotis* (excluding *Cistugo*) averaged 15%. Within other *Myotis* species sequence divergences between individuals appear to vary depending on the geographic distance between individuals, hence Ruedi and Mayer (2001: 440) and Stadelmann *et al.* (2004: 181) reported complete cytochrome-*b* gene sequences between individuals that were identical or had less than 1% variation, as well as instances of eight substitutions between individuals of *M. formosus* from Laos and South Korea, 15 substitutions (1.3% sequence divergence) between *M. welwitschii* from Uganda and Guinea, and 50–51 substitutions (4.5% sequence divergence) between *M. welwitschii* from Uganda and Guinea and individuals from South Africa, respectively. According to Ruedi and Mayer (2001: 440) interspecific divergence between sibling species of *Myotis* varies from 2.5% sequence divergence between *M. myotis* and *M. blythii,* and 16% between *M. mystacinus* and *M. brandtii.* The results for *C. lesueuri* and *C. seabrae* that are based on a few individuals would suggest various possibilities: that the level of sequence divergence does not warrant recognition of two species, or that the species have very similar cytochrome-*b* sequences, or that the molecular studies did not in fact include individuals from two species.

Some of the tissues on which the published molecular work was based appear to have been taken at a time when the value of having tissues linked to a voucher specimen was not recognized, and therefore the specimens from which the tissues were taken, are not accessible for further study (Ruedas *et al.,* 2000). The sequences reported by Stadelmann *et al.* (2004) for *C. lesueuri* (GenBank accession number AJ841961) are based on tissue from an individual captured at the Algeria Forest, Western Cape, South Africa, with no associated voucher specimen. Eick *et al.* (2005) reported they used tissue for *C. lesueuri* (8.10.02Ml1 (A)) obtained from David S. Jacobs (University of Cape Town), but gave no information as to where the individual was from, and whether there was an associated voucher specimen. Miller-Butterworth *et al.* (2005: 1135) used tissue from 8.10.02Ml1 (A) as an outgroup in a study on Miniopterinae, and indicated it came from the Algeria Forestry Station (which is also referred to as Algeria Forest). David Jacobs (pers. comm.) confirmed the tissue, 8.10.02Ml1 (A), came from an individual caught at the Algeria Forestry Station, but that there is no associated voucher specimen. Bickham *et al.* (2004), however, reported sequences for *C. lesueuri* based on tissues of individuals from the Algeria Forest, which are associated with museum vouchers (TMSA 38422, TMSA 38424) in the Ditsong National Museum of Natural History (formerly Transvaal Museum).

The sequences reported for *C. seabrae* by Stadelmann *et al.* (2004), Eick *et al.* (2005) and Lack *et al.* (2010) were all based on tissue from an individual from Goodhouse, Northern Cape, South Africa, which was associated with an uncatalogued specimen (MR-M977) collected by Manuel Ruedi; the latter (pers. comm.) has subsequently indicated MR-M977 has been accessioned in the Natural History Museum, Geneva, Switzerland as MHNG 1972.093. Bickham *et al.* (2004) reported sequences for *C. seabrae* based on tissues of individuals from two localities in Namibia; Klein Aus 8 in the Lüderitz District that is associated with a voucher specimen (TMSA 37563), and Zwartmodder 101 in the Maltahöhe District that is associated with a voucher specimen (CGC-6785) deposited in the State Museum in Windhoek. However, CGC-6785 is in fact referable to *Neoromicia zuluensis*, based on notes from a visit by one of the authors (T.K. from 16 to 18 September 2008) to the collection, and Seth Eiseb on 17 August 2011 (pers. comm.), and to date has not yet been traced. Stadelmann *et al.* (2004: 181) also mentioned 'Two individuals of *M.* [= *Cistugo*] *lesueuri* and two of *M. seabrae*, were almost identical with zero and two substitutions, respectively, even for animals captured in very distinct geographic areas', however, they gave no details as to where the two additional tissues came from, or whether they have associated voucher specimens.

In a break from the more traditional dichotomous classification keys, Monadjem *et al.* (2010: 391) provide identification matrixes. The matrix comparing *C. seabrae* and *C. lesueuri* considers forearm and condylo-incisor lengths, pelage colour, peak frequency of the echolocation call, roost type and distribution, and indicates considerable similarity between the two species:

1) Forearm length of *C. seabrae* was given as 29–35 mm, and that of *C. lesueuri* as 33–38 mm.
2) The condylo-incisor length of *C. seabrae* being less than 13.4 mm, and that of *C. lesueuri* being more than 13.1 mm.
3) Pelage colour remains slightly different between the species, with *C. lesueuri* reportedly being paler, yellower, and less brown in colour than *C. seabrae* (Thomas, 1912; Roberts, 1919; Monadjem *et al.,* 2010). However, while the matrix follows Taylor (2000) and indicates pelage colour differences between the species as indicated in

the original species descriptions, the information in the species account (p. 400) for *C. seabrae* adds an additional dorsal colour of 'yellow-orange' to the 'dull yellow to yellow-beige' previously mentioned in the matrix (p. 391).

4) The distribution information given in the identification matrix for *C. seabrae* as 'endemic to the west coast of southern Africa' with no indication of a southward cut-off in their distribution, suggests an overlap between the species. However, the information in the species account for *C. seabrae* (p. 403) does remedy this, suggesting that the species extends northwards from the extreme northwest of South Africa.
5) The roost difference between the two species in the identification matrix suggests that *C. seabrae* can be found in buildings, while *C. lesueuri* roosts in rock crevices. It appears from the species account of *C. seabrae* that the evidence for *C. seabrae* roosting in buildings is based on the observation by Shortridge (1934) of individuals at Berseba in Namibia which, however, was not conclusive about the roost: 'A few of these bats (two of which were shot) were observed on the wing in Berseba Village; they were flying round the church, in the steeple of which they apparently roosted by day.' It is not clear if any other sources confirmed the roost site information in Monadjem *et al.* (2010), since previously the roost of *C. seabrae* had not been reported as specimens have been observed, shot or netted away from a roost (Shortridge, 1934; Herselman and Norton, 1985; Seamark and Kearney, 2006). Although in the course of enquiry regarding the individual of *C. seabrae,* MHNG 1972.093, from Goodhouse in the Western Cape, Manuel Ruedi (pers. comm.) mentioned that he caught two males of this species at night from where he observed them roosting in a disused house.

Against this background we present an updated distribution map for the genus that includes new localities for each species, and assess how two measurements that are similar to those used by Roberts (1954) to separate *C. lesueuri* from *C. seabrae* are able to separate a larger number of *Cistugo* specimens.

METHODS

Forearm length was plotted against condylo-incisor length for 34 specimens of *Cistugo,* which included the holotypes of each species, as well as 10 specimens identified as *C. lesueuri* and 22 specimens identified as *C. seabrae* (see Appendix 1 for specimen and locality details). Species identifications followed those in the museum records, with the exception of TMSA 29310, which followed Friedman and Daly (2004) and Monadjem *et al.* (2010). Eight of the most recently collected specimens (MMK: JW-168-170, SAMC 41458, TMSA 47581–47582, TMSA 47584, TMSA 48190) were identified by one of the authors (T.K.) following the classification in Meester *et al.* (1986), which separates the two species on forearm length. For the other specimens it is presumed forearm length, and/or geographic position (assuming the species have allopatric distributions) were used for their identification.

Condylo-incisor length was recorded from cleaned skulls, and forearm length was recorded from dried museum skins, or from fresh specimens in the case of SAMC 41458, TMSA 47581, TMSA 47582, and TMSA 47584. Most measurements were recorded to the nearest 0.01 mm using Mitutoyo® digital callipers (Mitutoyo American Corporation, Aurora, Illinois, U.S.A.) by one of the authors (T.K.), the exceptions being the forearm measurement of the holotype of *C. seabrae* (BNMH 1906.1.3.3), which was taken from published information (Thomas, 1912: 206), and the forearm and condylo-incisor lengths of MHNG 1972.093 that were provided by Manuel Ruedi (pers. comm.).

RESULTS AND DISCUSSION

Figure 1 shows a plot of forearm length against condylo-incisor length of the holotypes of *C. lesueuri* and *C. seabrae* and 32 other specimens of both species. The plot separates the holotypes and the other specimens into two groups. However, the group that includes the holotype of *C. lesueuri* also includes specimens from localities in the south of Namibia (Farm Zwartmodder 101 and Klein Aus Farm 8) that appear to have been identified as *C. seabrae* by virtue of their locality, and not forearm length. The recent classification in Monadjem *et al.* (2010) increased the forearm range for *C. seabrae* from that given in previous classifications, possibly to include these specimens in *C. seabrae.*

These particular measurements show no evidence of a clinal pattern in size increases or decreases associated with changing latitude, either considering *Cistugo* comprising a single species or two species. The holotype of *C. seabrae* from Angola and the specimen from the most northerly locality in Namibia (TMSA 31277) are smaller than other individuals from more southerly localities in Namibia (Klein Aus and Zwartmodder), and closer in size to individuals identified as *C. seabrae* from the Northern Cape in South Africa (Henkries and Khamkirri). Hence, within *C. seabrae,* at localities along a north-to-south axis, individuals show notable variation. An assessment of the pattern of distribution of specimens in Fig. 1 relative to the sex and degree of

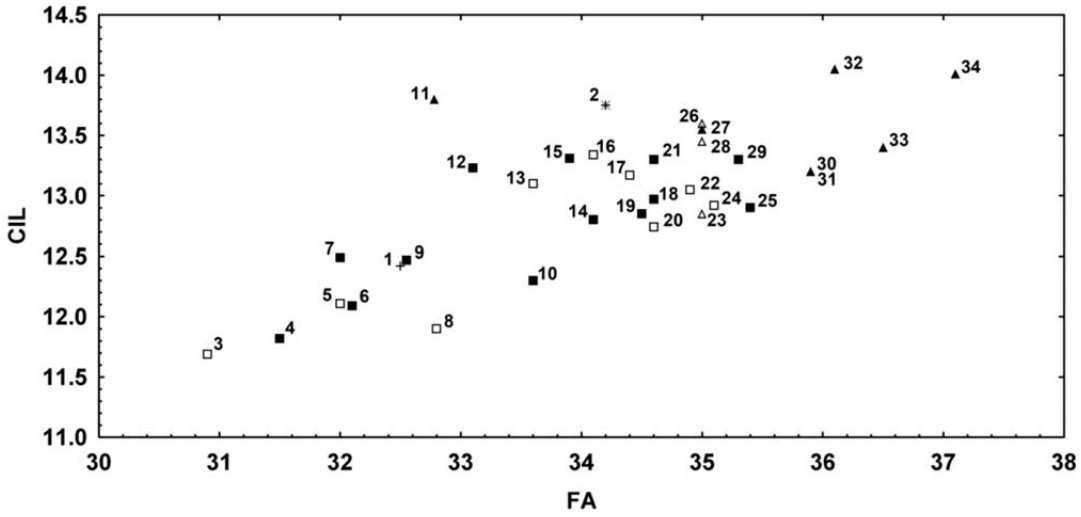

Fig. 1

Plot of forearm (FA) against condylo-incisor (CIL) lengths for 23 individuals of *Cistugo seabrae* Thomas, 1912 (squares, and a cross for the holotype of unknown sex) and 11 individuals of *Cistugo lesueuri* Roberts, 1919 (triangles, and a star for the male holotype). Open symbols indicate males, closed symbols females. See Appendix 1 for details of each individual.

toothwear (as a proxy for age), indicates that while some specimens with little toothwear are sometimes smaller than others of the same sex and from nearby localities, neither the sex nor the degree of toothwear can explain the patterns of variation.

The separation of specimens into two groups in Fig. 1 may be an artifact of sampling, or it may indicate that forearm and condylo-incisor lengths are able to identify the two species, i.e., *C. seabrae* has a condylo-incisor length of less than 12.6 mm and a forearm length of less than 33.7 mm, while both these measurements are larger in *C. lesueuri.* If this were the case, it would mean the distribution ranges of the species overlap, and that individuals from southerly localities in Namibia (Farm Zwartmodder 101 and Klein Aus Farm 8) currently identified as *C. seabrae* are 'misidentified'. This classification of the species would mean the dorsal pelage colour described for each of the species was no longer a distinguishing character, since the range of colour differences would be found within each of the species. However, as seems to be the case with species in the family Vespertilionidae, pelage colour may not entirely be an indicator of species distinction, but may instead reflect the environment of a particular locality. Hence, individuals located in the drier northwestern parts of the species range are paler than those from wetter southeastern localities (Seamark and Kearney, 2008). This alternative suggestion for separating the species that allows their distributions to overlap, rather than their forearm and condylo-incisor lengths, still needs to be more thoroughly tested with measurements from additional specimens in other collections to see whether the distinction identified here will hold, and a more extensive analysis of variation in the dorsal pelage colour.

TMSA 29310, an adult female, which was collected next to an indigenous forest at Saasveld, near George on the east coast of the Western Cape in South Africa in July 1978, was identified in the museum collection, probably on the basis of its small forearm length, as *C. seabrae.* The collection locality of TMSA 29310 was recently confirmed by the collector, Johann Breytenbach (pers. comm.). In the distribution maps in Friedman and Daly (2004) and Monadjem *et al.* (2010) this individual has been 're-identified' as *C. lesueuri,* probably on the basis of its locality and reasoning that the two species do not have overlapping distributions, but do have overlapping sizes. TMSA 29310 does, however, have a relatively long condylo-incisor length, hence in the plot of forearm length against condylo-incisor length it plots (No. 11 in Fig. 1) above the gap separating the two holotypes, within the group of the *C. lesueuri* holotype, although closest to specimens from southerly localities in Namibia currently identified as *C. seabrae.*

Figure 2 gives an updated distribution map based on known voucher specimen records for *C. lesueuri* and *C. seabrae* (see Appendix 1 for specimen and locality details). Since the suggestion that *C. lesueuri* and *C. seabrae* might have overlapping distributions still needs to be more extensively tested, the species designations in the distribution map still follow those in Friedman and Daly (2004)

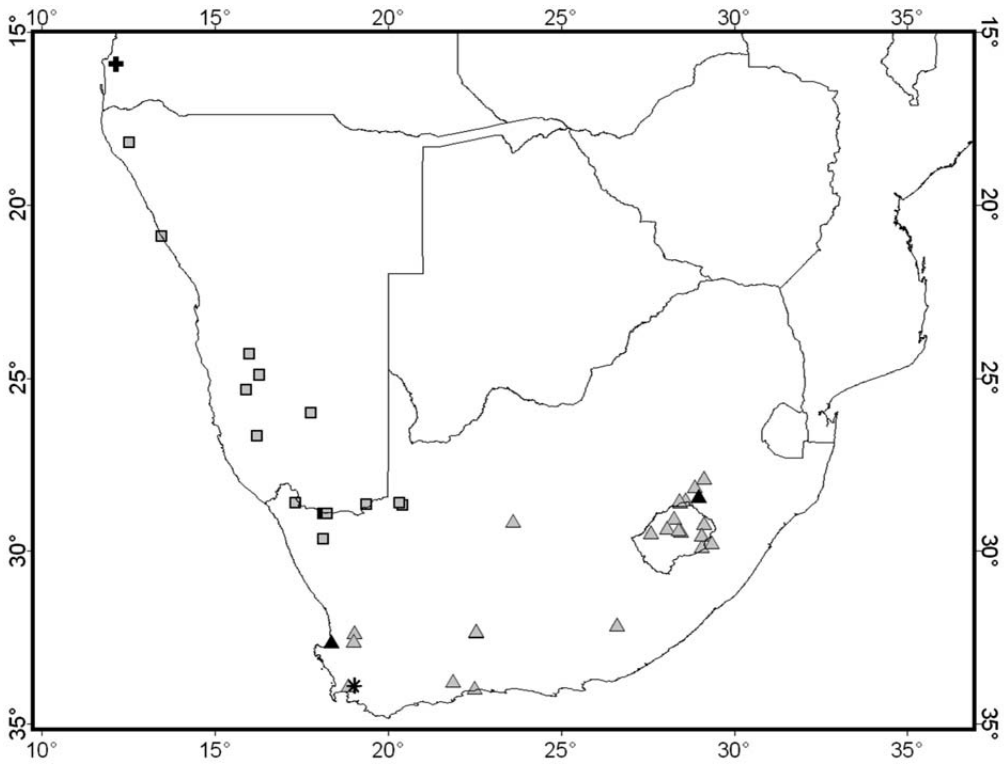

Fig. 2

Updated distribution map for *Cistugo seabrae* Thomas, 1912 (squares, and a cross for the holotype) and *C. lesueuri* Roberts, 1919 (triangles, and a star for the holotype). Black symbols represent localities that have not been plotted in previous publications; grey symbols indicate previously plotted localities. See Appendix 1 for details of each locality.

and Monadjem *et al.* (2010), i.e., assuming the species distributions are allopatric. Relative to the most recent distribution maps in Monadjem *et al.* (2010), an additional ten localities are plotted; six for *C. lesueuri* and four for *C. seabrae.* For *C. lesueuri* these include localities; for the holotype from (1) Lormarins, three localities for specimens collected by Herselman and Norton (1985), at (2) Hex River Estate, (3) Mazelsfontein, and the specimen re-identified in Kearney and Van Schalkwyk (2009) from (4) Rocherpan, a new specimen collected by the authors from another locality (5) close to Clarens, and an unaccessioned specimen in the Iziko South African Museum identified by one of us (T.K.) from (6) Tierkloof. For *C. seabrae* the additional localities are for specimens collected by Shortridge at (1) Berseba in Namibia, two localities for specimens collected by Herselman and Norton (1985) at (2) Goegab and (3) Steyerskraal, and new specimens collected by one of the authors (J.W.) from (4) Henkries. Of these ten localities, three have not been published before in distribution maps as specific records of existing voucher specimens: (1) the specimen collected by the authors from an additional locality close to Clarens in the Free State, (2) the specimen collected by Herselman and Norton (1985) that was re-identified in Kearney and Van Schalkwyk (2009) from Rocherpan, and (3) the specimens collected by J.W. from Henkries, the other seven have previously been published in distribution maps in Herselman and Norton (1985), Friedman and Daly (2004), and the ACR (2011). It should be noted that the most northwesterly locality plotted in Friedman and Daly (2004) in the distribution map for *C. lesueuri* is erroneous; the quarter degree square, 2821 Ba, was incorrectly assigned on the index card for the holotype of *C. lesueuri* (TMSA 2286) from L'Ormarins in the Franschhoek Valley.

CONCLUSION

It is evident the information accumulated to date for *C. lesueuri* and *C. seabrae* supports even less morphological distinction between the two species than was originally described. This, together with the limited molecular information available, clearly shows that further work is required to resolve the relationship between these two taxa. It is suggested

that further consideration also be given to the possibility that the species ranges may overlap, contrary to previous assumptions that the distributions are allopatric. Finally, expanded molecular genetic studies, including possible sequence data from holotypes or topotypes, could shed some important light on the question of the validity of the two taxa and their geographical distribution.

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REFERENCES

AFRICAN CHIROPTERA REPORT, 2011. *African Chiroptera report 2011.* African Chiroptera Project, Pretoria. http://www.Africanbats.org

BICKHAM, J. W., PATTON, J. C., SCHLITTER, D. A., RAUTENBACH, I. L. and HONEYCUTT, R. L., 2004. Molecular phylogenetics, karyotypic diversity, and partition of the genus *Myotis* (Chiroptera: Vespertilionidae). *Molecular Phylogenetics and Evolution* **33**(2): 333–338.

BRONNER, G. N., HOFFMANN, M., TAYLOR, P. J., CHIMIMBA, C. T., BEST, P. B., MATTHEE, C. A. and ROBINSON, T. J., 2003. A revised systematic checklist of the extant mammals of the southern African subregion. *Durban Museum Novitates* **28:** 56–106.

CORBET, G. B. and HILL, J. E., 1980. *A world list of mammalian species.* Cornstock Publishing Associates, Cornell University Press, Ithaca.

CORBET, G. B. and HILL, J. E., 1991. *A world list of mammalian species,* 3rd edn. Natural History Museum Publications and Oxford University Press, Oxford.

CRAWFORD-CABRAL, J., 1989. A list of Angolan Chiroptera with notes on their distribution. *Garcia de Orta, Sérias Zoologias, Lisboa* **13**(1–2): 7–48.

DE GRAAF, G. and RAUTENBACH, I. L., 1983. A survey of mammals in the newly proclaimed Karoo National Park, South Africa. *In:* VAN DER STRAETEN, E., VERHEYEN, W. N. and DE VREE, F., eds, Proceedings of the Third International Colloquium on Ecology and Taxonomy of African Small Mammals. Antwerpen, July 20–24, 1981. *Annalen van het Koninklijk Museum voor Midden-Afrika, Zoologische Wetenschappen* **237:** 89–99.

EICK, G. N., JACOBS, D. S. and MATTHEE, C. A., 2005. A nuclear DNA Phylogenetic perspective on the evolution of echolocation and historical biogeography of extant bats (Chiroptera). *Molecular Biology and Evolution* **22** (9): 1869–1886.

ELLERMAN, J. R., 1954. Die taksonomie van die soogdiere van die Unie van Suid-Afrika. *Annale van die Universiteit van Stellenbosch* **30A**(1): 14–33.

ELLERMAN, J. R. and MORRISON-SCOTT, T. C. S., 1951. *Checklist of Palaeartic and Indian mammals 1758 to 1946.* Trustees of the British Museum (Natural History), London.

ELLERMAN, J. R. and MORRISON-SCOTT, T. C. S., 1966. *Checklist of Palaearctic and Indian mammals 1758 to 1946,* 2nd edn. Trustees of the British Museum (Natural History), London.

ELLERMAN, J. R., MORRISON-SCOTT, T. C. S. and HAYMAN, R. W., 1953. *Southern African mammals 1758 to 1951: a reclassification.* Trustees of the British Museum (Natural History), London.

FRIEDMANN, Y. and DALY, B., eds, 2004. *Red data book of the mammals of South Africa: a conservation assessment.* CBSG Southern Africa, Conservation Breeding Specialist Group (SSC/IUCN), Endangered Wildlife Trust, South Africa.

GODAWA-STORMARK, J., 1998. Phenetic analysis of Old World *Myotis* (Chiroptera: Vespertilionidae) based on dental characters. *Acta Theriologica* **43**(1): 1–11.

HAYMAN, R. W. and HILL, J. E., 1971. Part 2: Order Chiroptera. *In:* MEESTER, J. and SETZER, H. W., eds, *The mammals of Africa: an identification manual,* pp. 1–73. Smithsonian Institution Press, Washington, D.C.

HERSELMAN, J. C. and NORTON, P. M., 1985. The distribution and status of bats (Mammalia: Chiroptera) in the Cape Province. *Annals of the Cape Provincial Museums* (*Natural History*) **16**(4): 73–126.

HONACKI, J. H., KINMAN, K. E. and KOEPPL, J. W., eds, 1978. *Mammal species of the world: a taxonomic and geographic reference.* Allen Press and The Association of Systematics Collections, Lawrence, Kansas, U.S.A.

HONACKI, J. H., KINMAN, K. E., and KOEPPL, J. W., eds, 1982. *Mammal species of the world. A taxonomic and geographic reference.* Allen Press and The Association of Systematics Collections, Lawrence, Kansas, U.S.A.

KEARNEY, T. C. and VAN SCHALKWYK, E., 2009. Variation in the position within the tooth row of the minute premolars of *Cistugo lesueuri* and *C. seabrae* (Chiroptera: Vespertilionidae), and re-identification of some museum voucher specimens. *Annals of the Transvaal Museum* **46:** 117–120.

KOOPMAN, K. F., 1993. Order Chiroptera. *In:* WILSON, D. E. and REEDER, D. M., eds, *Mammal species of the world: a taxonomic and geographic reference,* 2nd edn, pp. 137–241. Smithsonian Institution Press, Washington D.C. and London.

KOOPMAN, K. F., 1994. Chiroptera: Systematics. *In:* NIETHAMMER, J., SCHLIEMANN, H. and STARCK, D., eds, *Handbook of zoology, volume VIII (Mammalia) (60),* pp. i–vii, 1–117. W. De Gruyter, Berlin–New York.

LACK, J. B., ROEHRS, Z. P., STANLEY Jr, C. E., RUEDI, M. and VAN DEN BUSSCHE, R. A., 2010. Molecular phylogenetics of *Myotis* indicate familial-level divergence for the genus *Cistugo* (Chiroptera). *Journal of Mammalogy* **91**(4): 976–992.

LYNCH, C. D., 1994. Mammals of Lesotho. *Navorsinge van die Nasionale Museum Bloemfontein* **10**(4): 177–241.

LYNCH, C. D. and WATSON, J. P., 1990. The mammals of

Sehlabathebe National Park, Lesotho. *Navorsinge van die Nasionale Museum Bloemfontein* **6**(12): 523–554.
MEESTER, J. A. J., RAUTENBACH, I. L., DIPPENAAR, N. J. and BAKER, C. M., 1986. *Classification of Southern African mammals.* Transvaal Museum Monograph No. **5**, Transvaal Museum, Pretoria.
MILLER-BUTTERWORTH, C. M., EICK, G., JACOBS, D. S., SCHOEMAN, M. C. and HARLEY, E. H., 2005. Genetic and phenotypic differences between South African long-fingered bats, with a global miniopterine phylogeny. *Journal of Mammalogy* **86**(6): 1121–1135.
MONADJEM, A., TAYLOR, P. J., COTTERILL, F. P. D. and SCHOEMAN, M. C., 2010. *Bats of southern and central Africa: a biogeographic and taxonomic synthesis.* Wits University Press, Johannesburg.
RAUTENBACH, I. L., BRONNER, G. N. and SCHLITTER, D. A., 1993. Karyotypic data and attendant systematic implications for the bats of southern Africa. *Koedoe* **36**(2): 87–104.
RAUTIAN, G. S., ROSSINA, V. V. and RAUTIAN, A. S., 2006. Approaches to the resolution of contradictions between phylogenetic reconstructions based on morpho-functional and genetic data. *Paleontological Journal* **40** (suppl. 4): 508–523.
ROBERTS, A., 1919. Descriptions of some new mammals. *Annals of the Transvaal Museum* **6**(3): 112–115.
ROBERTS, A., 1954. *The mammals of South Africa,* 2nd edn. The Mammals of South Africa Book Fund, Johannesburg.
RUEDAS, L. A., SALAZAR-BRAVO, J., DRAGOO, J. W. and YATES, T. L., 2000. The importance of being earnest: what, if anything, constitutes a 'specimen examined?'. *Molecular Phylogenetics and Evolution* **17**(1): 129–132.
RUEDI, M. and MAYER, F., 2001. Molecular systematics of bats of the genus *Myotis* (Vespertilionidae) suggests deterministic ecomorphological convergences. *Molecular Phylogenetics and Evolution* **21**(3): 436–448.
SEAMARK, E. C. J. and BRAND, M., 2005. Bat survey in the Cederberg Wilderness Area, Western Cape, South Africa (28 January – 3 February 1999). *African Bat Conservation News* **3:** 7–9.
SEAMARK, E. C. J. and KEARNEY, T. C., 2006. New distribution of the Angolan wing-gland bat (*Cistugo seabrae* Thomas, 1912). *African Bat Conservation News* **7:** 2–4.
SEAMARK, E. C. J. and KEARNEY, T., 2008. Bats of Messina Nature Reserve, Limpopo Province, South Africa. *African Bat Conservation News* **18:** 5–7.
SHORTRIDGE, G. C., 1934. *The mammals of South West Africa. A biological account of the forms occurring in that region.* William Heinemann Ltd, London.
SHORTRIDGE, G. C., 1942. 2. Field notes on the first and second expeditions of the Cape Museum's Mammal Survey of the Cape Province; and descriptions of some new subgenera and subspecies. *Annals of the South African Museum* **36:** 27–100.
SIMMONS, N. B., 2005. Order Chiroptera. *In:* WILSON, D. E. and REEDER, D. M., eds, *Mammal species of the world: a taxonomic and geographical reference,* 3rd edn, pp. 312–529. Johns Hopkins University Press, Baltimore, Maryland.
SMITHERS, R. H. N., 1983. *The mammals of the southern African subregion.* University of Pretoria, Pretoria, South Africa.
STADELMANN, B., JACOBS, D. S., SCHOEMAN, C. and RUEDI, M., 2004. Phylogeny of African *Myotis* bats (Chiroptera, Vespertilionidae) inferred from cytochrome *b* sequences. *Acta Chiropterologica* **6**(2): 177–192.
TAYLOR, P. J., 2000. *Bats of southern Africa: guide to biology, identification, and conservation.* University of Natal Press, Pietermaritzburg.
THOMAS, O., 1912. A new vespertilionine bat from Angola. *Annals and Magazine of Natural History* (8)**10**(56): 204–206.
THOMAS, O. and HINTON, M. A. C., 1925. On mammals collected in 1923 by Captain G. C. Shortridge during the Percy Sladen and Kaffrarian Museum Expedition to South-West Africa. With field notes by the collector. *Proceedings of the Zoological Society of London* **1925**(1): 221–246.
WATSON, J. P., 1998. New distributional records for three microchiropteran bats (Vespertilionidae, Rhinolophidae) from the Free State Province, South Africa. *South African Journal of Wildlife Research* **28**(4): 127–131.

APPENDIX 1

Details of known voucher specimens of *Cistugo seabrae* and *C. lesueuri.* Underlined, bold face numbers from 1 to 34 relate to the numbers indicated in Fig. 1. + = new records introduced in this study. * = specimen originally identified as *Eptesicus melckorum.* # = Monadjem *et al.* (2010) recorded JW-142 as possibly being in the National Museum, Bloemfontein, whereas it has been lodged in the MacGregor Museum, Kimberley. Accession numbers in bold indicate the specimen was examined and its identity confirmed by the authors.

Museum accession abbreviations: BMNH, The Natural History Museum, London (formerly British Museum of Natural History); DNSM, Durban Natural Science Museum, Durban; KM, Amathole Museum, King William's Town (formerly the Kaffrarian Museum); LACM, Los Angeles County Museum, Los Angeles; MMK, MacGregor Museum, Kimberley; MHNG, Natural History Museum, Geneva; NMB, National Museum of Bloemfontein; SAMC, Iziko South African Museum (formerly South African Museum), Cape Town; SMW (specimens prefixed by SMM), State Museum Windhoek; SNPL, Sehlabathebe National Park reference collection, Lesotho; TMSA, Ditsong National Museum of Natural History, Pretoria (formerly Transvaal Museum); USNM, National Museum of Natural History, Smithsonian Institution, Washington D.C.

Publications in which a given specimen was previously cited: [1]Thomas (1912), [2]Roberts (1919), [3]Thomas and Hinton (1925), [4]Shortridge (1934), [5]Shortridge (1942), [6]De Graaf and Rautenbach (1983), [7]Herselman and Norton (1985), [8]Crawford-Cabral (1989), [9]Lynch and Watson (1990), [10]Rautenbach *et al.* (1993), [11]Godawa-Stormark (1998), [12]Lynch (1994); [13]Bickham *et al.* (2004), [14]Stadelmann *et al.* (2004), [15]Eick *et al.* (2005), [16]Seamark and Brand (2005), [17]Seamark and Kearney (2006), [18]Kearney and Van Schalkwyk (2009), [19]Lack *et al.* (2010), [20]Monadjem *et al.* (2010), [21]African Chiroptera Report (2011). If no direct link is provided to a specimen but the species is identified by specimens from a particular locality, the number associated with the publication is listed directly after the locality information.

***Cistugo lesueuri* Roberts, 1919**

LESOTHO: Lepaqoa (29°02′24″S 28°16′12″E): NMB: **7036**[12,20,21]. Mahlanapeng (29°21′36″S 28°24′00″E): NMB: **8158**[12,20,21]. Marakabei (29°19′12″S 28°04′48″E): NMB: **7265**[12,21], **7268**[12,20,21], **7269**[12,21], **7271**[12,21], **7319**[12,21], **7320**[12,21], **7321**[12,21], **7323**[12,21].

Mateanong (29°12′00″S 29°07′48″E): NMB: **6887**[12,20,21], **6888**[12,21], **6898**[12,21], **6899**[12,21], **6900**[12,21], **6901**[12,21], **6902**[12,21], **6903**[12,21], **6908**[12,21], **6910**[12,21], **6911**[12,21], **6912**[12,21], **6913**[12,21], 6988[12,21], 6989[12,21], 6992[12,21]. Moqotoane (29°25′12″S 28°28′48″E): NMB: **8483**[12,20,21], **8488**[12,21]. Phallang (29°27′36″S 27°36′00″E): NMB: **8289**[12,20,21], **8290**[12,21], **8291**[12,21], **8292**[12,21], **8293**[12,21], **8294**[12,21], **8295**[12,21], **8296**[12,21], **8297**[12,21], **8298**[12,21], **8299**[12,21], **8301**[12,21], **8302**[12,21], **8303**[12,21], **8304**[12,21], **8305**[12,21], **8306**[12,21], **8307**[12,21], **8308**[12,21]. Sehlabathebe National Park (29°31′48″S 29°04′12″E): SNPL: (unnumbered)[9,12]. Sehlabathebe National Park (29°52′S 29°05′E): KM: **24805**[9,12,18,20,21]. Sehlabathebe National Park, dam wall near stables (29°31′48″S 29°04′12″E): NMB: **6685**[9,12,21], **6696**[9,12,20,21].

SOUTH AFRICA: BMNH–27.4.13A4. Cederberg, Algeria State Forest, campsite (32°21′S 19°03′E)[10]: MMK: 2496[20], SAMC: **41458**[16,21,**26**]; TMSA: **35121**[18,20,21,**27**], **35152**[18,21], **35172**[18,21], **38422**[13,18,21,**23**], **38423**[18,21,**31**], **38424**[13,18,21,**30**], **38428**[18,21,**33**], **39429**[18,21]. Clarens, near (28°31′12″S 28°25′12″E): DNSM 88[20]. Farm Schaapplaats 930 (2728Dd): NMB: 7394[14,21]. Farm Schaapplaats 930 (S28.573590 E28.452050): TMSA: **48190**[+,18,**34**]. Farm Waterval 747 (27°52′48″S 29°07′48″E): NMB: **7700**[14,20,21], **7703**[14,21], **7705**[14,21], **7706**[14,21], **7708**[14,21], **7710**[14,21], **7711**[14,21], **7712**[14,21], **7713**[14,21], **7714**[14,21], **7715**[14,21], **7731**[14,21], **7732**[14,21]. Golden Gate National Park, Glen Reenen (28°30′18″S 28°36′36″E): NMB: **7902**[14,20,21]. Hex River Estate, Citrusdal (S32.36 E19.01): KM**1880**[5,7], **1881**[5,7]. Jonkershoek (3318Dd): KM: **MYO-001J**[7,18]. Kaffraria: KM: **2070**[18]. Kaggasmoudt (32°07′S 26°37′E): KM: **2022**[18], **2024**[18], **2025**[18], **2026**[18], **2028**[18], **2029**[18], **2031**[18,20], **2032**[18], **2041**[18], **2042**[18], **2051**[18], **2052**[18]. Kamberg Valley (29°45′00″S 29°21′36E): DNSM: 7504[20]. Karoo-NP (32°20′S 22°33′E): TMSA: **29511**[6,7,10,18,20,21,**32**]. Karoo-NP, Bulkraal, dry river bed near old pump station (32°18′00″S 22°33′36″E): MMK: **JW-139**, **JW-140**, **JW-141**, **JW-142**[17,#]. L'Ormarins [= Lormarins] (33°53′S 19°03′E): TMSA: **2286**[2,18,21,**2**] (Holotype – *Cistugo lesueuri* Roberts, 1919). Mazelsfontein (2923Ba): KM: **MYO-002J**[7,18].Rocherpan (3218Cb): KM: **26055**[*,7,18]. Saasveld Forestry College (33°57′S 22°31′E): TMSA: **29310**[7,18,20,21,**11**]. Sterkfontein Dam Nature Reserve, Farm Ararat 791 (28°07′48″S 28°52′48″E): NMB: **7847**[14,20,21]. Sterkfontein Dam-NR (28°24′57″S 28°59′26″E): NMB: **JW-122**, **JW-123**, **JW-127** – Tierkloof, Bushcamp (33°44′30″S 21°53′30″E): SAMC: **GB-16/00**[21].

***Cistugo seabrae* Thomas, 1912**

ANGOLA: Namibe [= syn Mossamedes, Mocamedes] (15°56′S 12°09′E): BMNH: **1906.1.3.3.**[1,2,8,17,20,21,**1**] (Holotype – *Cistugo seabræ* Thomas, 1912).

NAMIBIA: Berseba (25°59′S 17°47′E, 3067 ft)[17]: BMNH: CN510[3,4,21], CN591[3,4,21]. Farm Zwartmodder 101 (24°54′S 16°17′E): TMSA: **37587**[13,17,20,21,**20**], **37610**[17,18,21,**14**], **37611**[17,18,21,**12**], **37612**[17,18,21,**25**], **37613**[17,18,21,**18**]. Gorrasis 99 (25°19′27″S 15°54′07″E): SMM: **4392**[20]. Klein Aus Farm 8 (26°39′S 16°13′E)[10]: SMM: **11160**, **11197**[20], SMM: CGC-6785[11]; TMSA: **37541**[17,18,20,21,**29**], **37542**[17,18,21,**17**], **37543**[17,18,21,**22**], **37544**[17,18,21,**24**], **37549**[17,18,21,**21**], **37550**[17,18,21,**15**], **37561**[17,18,21,**19**], **37562**[17,18,21,**16**], **37563**[13,17,18,21,**13**]. Naukluft-NP, De Valle (24°18′S 16°00′E): SMM: **10538**[20], **10539**, **10540**, **10541**, **10542**, **10543**. Orumpembe (18°10′48″S 12°31′12″E): LACM: 59088[20]. Skeleton Coast Park, Huab River, Jack Scott Bridge (20°54′S 13°28′E)[10]: SMM: **9757**[20]; TMSA: **31277**[10,17,18,20,21,**10**]. Skeleton Coast Park, Hoanib R. floodplain (20°54′S 13°28′E): SMM: **12488**. Zwartmodder 101 (24°54′S 16°17′E): SMM: **11263**[20], **11264**, **11265**, **11267**, **11268**, **11269**.

SOUTH AFRICA: BMNH–25.1.2.6[11]. Augrabies Falls NP (28°36′00″S 20°19′48″E): MMK: 7051[20]. Farm Northern Kakamas, behind cottages at Khamkirri (28°39′S 20°26′E): TMSA: **47581**[17,18,20,21,**3**], **47582**[17,18,21,**5**], **47584**[17,18,21,**4**]. Goegab (2918Ca)[17]: KM: **MYO-003J**[7,18], **MYO-004J**[7,18].Goodhouse (28°54′S 18°15′E): MHNG: 1972.093[14,15,19,**8**]. Goodhouse, Orange River (28°54′S 18°15′E, 650 ft): KM: 1882[17,18], 1883[5,7,17,18,21], **1884**[5,7,17,18,21], **1885**[5,7,17,18,21], **1886**[5,7,17,18,21], **1887**[5,7,17,18,21], **1888**[5,7,17,18,21], **1889**[5,7,17,18,20,21]. Henkries, Mango orchard (28°53′56″S 18°08′16″E): MMK: **JW-168**[+,**7**], **JW-169**[+,**9**], **JW-170**[+,**6**]. Springbok, 10 km (29°40′12″S 17°52′12″E): USNM: 342632[18]. Steyerskraal (2819CB)[17]: KM: **MYO-049J**[7,18], **MYO-050J**[7,18], **MYO-051J**[7,18].