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Instructions for use

Intraspecific phylogeny of the house shrews, *Suncus murinus-S. montanus* species complex, based on the mitochondrial cytochrome *b* gene

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Abstract. A phylogenetic tree was reconstructed based on the mitochondrial cytochrome *b* gene nucleotide sequences of 169 individuals of house shrews (*Suncus murinus* and *S. montanus*) from 44 localities in East Asia, Southeast Asia, West Asia, and islands in the western Indian Ocean. Shrews from China (Zhejiang), Japan (Okinawa), Vietnam, and Indonesia (Java) formed a monophyletic group with less genetic variation. Therefore, the shrews of these regions appeared to have originated from one or a few localities. Contrary to this, shrews from Sri Lanka, Myanmar, and Pakistan consisted of several haplogroups. This finding suggests immigration movements to these areas. Fascinating findings were also obtained concerning the islands in the western Indian Ocean. First, shrews on Zanzibar Island (Tanzania) had almost the same haplotype as those in southwestern Iran. Therefore, the house shrew in Zanzibar may have immigrated from Iran (or vice versa). Second, shrews from Madagascar and Grande Comore Island shared the same haplotype, whereas the shrews on Réunion Island were clearly different from those of Madagascar and Comoros. Thus, there appears to have been several immigration routes to the islands of the western Indian Ocean.

Key words: human introduction, immigrations, Indian Ocean, Suncus montanus, Suncus murinus.

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The house shrew or musk shrew, Suncus murinus (Linnaeus, 1766) is widely distributed across southern Japan, southern China, Southeast Asia, the Indian subcontinent including Sri Lanka, the coastal region of the Arabic Peninsula and Iran, Madagascar, the coastal region of eastern Africa, and across many small islands in the Indian Ocean (Hutterer 2005; Motokawa 2015). Suncus murinus has been found on the island of Guam in the Pacific Ocean (Peterson 1956; Wiewel et al. 2009), but it now seems to be nearly or completely extinct there (Richard Yanagihara, personal communication). In some regions, it is estimated that house shrews were unintentionally introduced by humans, transported by trade ships (Hutterer and Trainier 1990; Yamagata et al. 1995; Kurachi et al. 2007a, 2007b). It is reported that S. montanus (Kelaart, 1850), which is regarded as a distinct species from S. murinus, is distributed in Sri Lanka (Phillips 1980; Meegaskumbura et al. 2010). However, because the phylogenetic relationships between S. montanus and S. murinus have not been fully investigated, we have treated them as a Suncus murinus-S. montanus species complex and treated them conventionally as one taxon in the present paper.

Intraspecific phylogeny of *S. murinus* has been partially reported based on the mitochondrial DNA or protein polymorphisms (Yamagata et al. 2004; Yamagata and

Kurachi 2005; Kurachi et al. 2007a, 2007b) and on karyotype (Yosida 1982). However, these previous studies mainly focused on shrews from the eastern part of the range. In the present paper, we briefly report the phylogenetic relationship of the *S. murinus-S. montanus* complex, including shrews from the western parts of the range, based on mitochondrial cytochrome *b* gene (*cytb*). The main purpose is to gain a basic information of the phylogenetic relationship based on the *cytb* among house shrews to infer the routes of immigration of the shrews.

Materials and methods

Sampling

Nucleotide sequences of the mitochondrial cytochrome b gene for 149 individuals of the S. murinus-S. montanus complex from 33 localities ranging from East Asia, via Southeast Asia, to islands in the western Indian Ocean were determined by ourselves. In addition to these, we cited the sequence data for 20 individuals of the S. murinus-S. montanus complex from 11 localities (including S. murinus murinus, S. murinus kandianus, S. murinus caerulescens, and S. montanus in Sri Lanka) from DNA databases to reconstruct a phylogenetic tree. In total, 169 individuals from 44 localities were analyzed (Appendix 1, Fig. 1). As outgroups, Suncus etruscus (Savi, 1822),

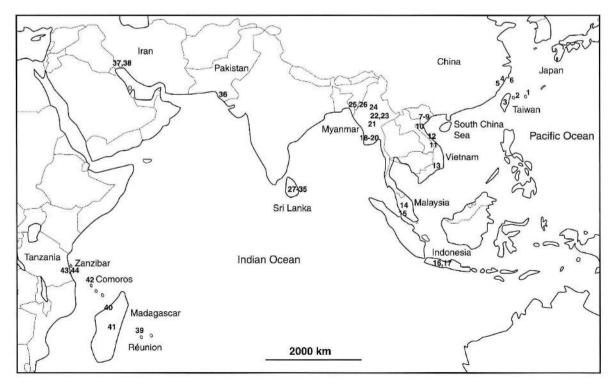


Fig. 1. Sample localities of the Suncus murinus-S. montanus complex. The numbers correspond with those of Appendix 1.

S. madagascariensis (Coquerel, 1848) (probably, a synonym of S. etruscus; Meegaskumbura et al., 2012a), S. stoliczkanus (Anderson, 1877), S. fellowesgordoni Phillips, 1932, Crocidura rapax G. Allen, 1923, and C. dsinezumi (Temminck, 1842) were used (Appendix 1).

DNA sequencing and phylogenetic analysis

The nucleotide sequence of the mitochondrial cytochrome *b* gene (1,140 bp) was determined following the method and procedure (primer set, chemicals, PCR conditions, etc.) of the previous investigations (Ohdachi et al. 2004; Dubey et al. 2007; Arai et al. 2012).

To infer the phylogenetic tree, a Bayesian phylogenetic tree was calculated, using BEAST version 2.3.0 (Bouckaert et al. 2014) with an exponential population coalescent as the tree prior. The dataset was analyzed in TN93+G+I model of nucleotide substitution, which was selected with the lowest Bayesian information criterion scores using a best fit model selection as implemented in MEGA version 5.05 (Tamura et al. 2011). Although the main purpose of the present paper is to gain a topology of the phylogenetic relationship, we show the phylogenetic tree of a dating analysis under a strict clock model with a conventional clock rate of a 3.1%/site/million years (Suzuki et al. 2015) to roughly estimate divergence time among lineages.

Results and discussion

All of 149 samples of the *S. murinus-S. montanus* complex and one specimen of *S. etruscus* were successfully sequenced and no indels were found. The monophyly of the *S. murinus-S. montanus* complex was strongly supported based on the *cytb* sequences, and *S. stoliczkanus* was a sister species to them (Fig. 2). In the phylogenetic tree (Fig. 2), there are 14 main clades in the *S. murinus-S. montanus* complex.

Shrews from Japan (Okinawa Prefecture, Loc. # 1–2), China (Zhejiang Province, Loc. # 4–6), Vietnam (Loc. # 7–13), and Indonesia (Java Island, Loc. # 16–17) formed a monophyletic group with less genetic variation (Fig. 2). Okinawa (the Ryukyu Arc) and Java are islands which finally separated from the Asian continent approximately 200,000 years ago (Kimura 2002) and approximately 11,000 years ago (Biswas 1973; Voris 2000; Bintanja et al. 2005), respectively. If the shrews in Okinawa and Java were originally distributed there, they would have more genetic differentiation from Vietnam and southern China. Therefore, it seems most natural

that the shrews of these islands immigrated from the Asian continent (Vietnam, southern China, or the nearby area) after the geologic separation. However, the result of the present investigation cannot determine the direction of immigration, and the immigration from Okinawa (the Ryukyu Arc) and Java to the continent might be possible. To determine the direction of immigration, remain (or semi-fossil) records of the house shrew are necessary. However to date, so far, only a single report has been published discussing the remain record of house shrews in the Ryukyu Arc (Nishioka et al. 2016). After an intensive survey of fossils in these areas (Japan, China, and Southeast Asia) has been done, we will conclude the direction of the immigration in this region.

Contrary to this, shrews from Sri Lanka and Myanmar consisted of at least four different haplogroups (including S. montanus) each (Fig. 2). Especially in Sri Lanka, the genetic diversity was very high. For instance, KDSL2/ KDLS3 and KDSL1/KDSL4/KDSK5 were captured in the same location in Kandy, Sri Lanka (Loc. # 29), but their haplotypes of cytb differed significantly (Fig. 2). Suncus montanus, S. murinus kandianus, and some house shrews from Sri Lanka were grouped into one clade (S. montanus-S. mu. kandianus complex = [Sri Lanka 3 + Sri Lanka 4]). According to the divergence time analysis under a strict clock model with 3.1%/site/million years, estimation of the divergence time between the S. montanus-S. mu. kandianus complex and the other haplogroups of house shrews (including Sri Lanka 1 & Taiwan and Sri Lanka 2) was approximately 1.36 million years ago (MYA) (Fig. 2). This divergence degree could be regarded as a distinct species level. The divergence time between Sri Lanka 1 & Taiwan and Sri Lanka 2 was 0.42 MYA and that between Sri Lanka 3 and Sri Lanka 4 was 0.10 MYA. For Myanmar shrews, the divergence time was estimated to be 0.28 MYA between Myanmar 2 and Myanmar 3 and 0.88 MYA between Myanmar 4 and Myanmar 2 + Myanmar 3 (Fig. 2). Moreover, the divergence time between Myanmar 1 & Malaysia and Myanmar Main Clade (Myanmar 2-4) was 0.93 MYA. Thus, Myanmar 1 (& Malaysia) and Sri Lanka 1 (& Taiwan) were phylogenetically quite distant from other shrews in Myanmar and Sri Lanka, respectively. This finding suggests the populations in Sri Lanka and Myanmar consist of several species/or lineages of house shrews.

In addition, it is also found that hybridizations among distinct lineages occurred in Myanmar and Sri Lanka, based on karyological investigation (Yosida 1982),

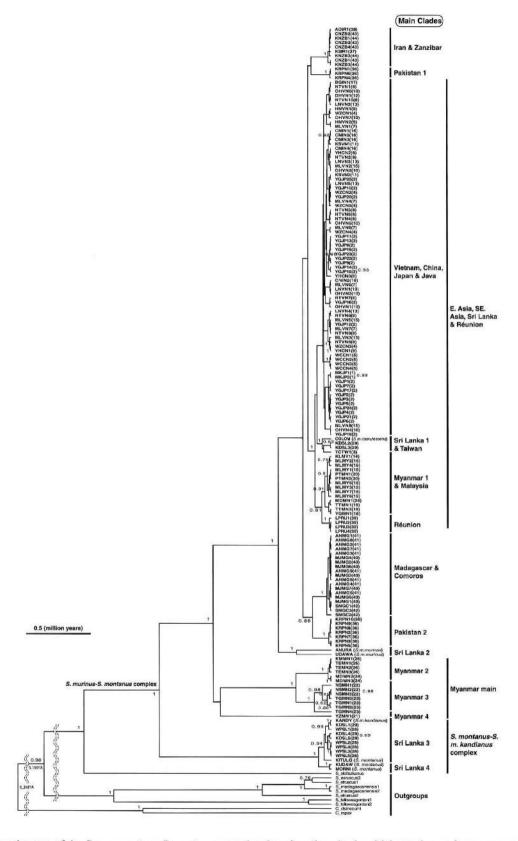


Fig. 2. Bayesian tree of the Suncus murinus-S. montanus complex, based on the mitochondrial cytochrome b gene sequences. The scale is the divergence time under a strict clock model with the assumption of the evolutionary rate of 3.1%/site/million years. See Appendix 1 for the code of OTUs. Numbers near nodes denote posterior probability and those less than 0.6 are omitted. Numbers in parentheses are locality numbers corresponding with those in Appendix 1 and Fig. 1.

suggesting immigration from other regions occurred in Sri Lanka and Myanmar. Furthermore, our phylogenetic analysis based on mitochondrial DNA demonstrated that *S. montanus* and *S. mu. kandianus* were included in a monophyletic group (Fig. 2), although they have different morphotypes (Meegaskumbura et al. 2010) and habitats (Phillips 1980; Yapa and Ratnavira 2013) from each other. The discrepancy between phylogeny and morphology/ecology in the *S. montanus-S. mu. kandianus* complex could be caused by hybridization of several species or lineages. Further investigation of the hybridization of the house shrews in Sri Lanka might resolve this problem.

Haplotypes of the shrews from Karachi, Pakistan (Loc. #36) were divided into two clades which are significantly differentiated from each other (Fig. 2). In the present study, we cannot conclude whether or not the shrews in Pakistan immigrated from another region.

For the islands in the western Indian Ocean, some interesting results were found. First, shrews on Zanzibar Island, Tanzania (Loc. # 43-44) had almost the same haplotype as those in southwestern Iran (Loc # 37–38) (Fig. 2), despite the two regions being far from each other (Fig. 1). Therefore, the house shrews in Zanzibar probably immigrated from Iran (or vice versa). Second, shrews from Madagascar and Grande Comore (Ngazidja) Island had almost the same haplotype (Fig. 2). However, the phylogenetic position of the shrews on Réunion Island (an overseas department of France, Loc. #39) was clearly different from those of Madagascar and Comoros (Fig. 2) although these regions are close to each other (Fig. 1). Since the shrews of Réunion are phylogenetically close to those from Sri Lanka, East Asia, and Southeast Asia, they might have immigrated from these regions.

Further collection of shrew samples, especially from the Arabic Peninsula and India, are greatly needed to complete our understanding of the emigration-immigration process and hybridization among the *Suncus murinus-S. montanus* complex, and further genetic information, such as nuclear genes, microsatellite DNA, and karyotype, and morphological relationship should be investigated in the future.

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References

- Arai, S., Gu, S. H., Back, L. J., Tabara, K., Bennett, S. N., Oh, H. S., Takada, N., Kang, H. J., Tanaka-Taya, K., Morikawa, S., Okabe, N., Yanagihara, R. and Song, J. W. 2012. Divergent ancestral lineages of newfound hantaviruses harbored by phylogenetically related crocidurine shrew species in Korea. Virology 424: 99–105.
- Bintanja, R., Van de Wal, R. S. and Oerlemans, J. 2005. Modelled atmospheric temperatures and global sea levels over the past million years. Nature 437: 125–128.
- Biswas, B. 1973. Quaternary changes in sea-level in the South China Sea. Bulletin of the Geoloical Society of Malyaysia 6: 229–256.
- Bouckaert, R., Heled, J., Kuhnert, D., Vaughan, T., Wu, C. H., Xie, D.,
 Suchard, M. A., Rambaut, A. and Drummond, A. J. 2014. BEAST
 2: a software platform for Bayesian evolutionary analysis. PLoS
 Computational Biolology 10: c1003537.
- Dubey, S., Salamin, N., Ohdachi, S. D., Barrière, P. and Vogel, P. 2007. Molecular phylogenetics of Soricidae (Mammalia, Soricidae) reveals timing of transcontinental colonisations. Molecular Phylogenetics and Evolution 44: 126–137.
- Hutterer, R. 2005. Order Soricomorpha. In (Wilson, D. E. and Reeder, D. M., eds.) Mammal Species of the World. A Taxonomic and Geographic Reference. 3rd edition. Vol. 1, pp. 220–311. The Johns Hopkins University Press, Baltimore.
- Hutterer, R. and Trainier, M. 1990. The immigration of the Asian house shrew (Suncus murinus) into Africa and Madagascar. In (Peters, G. and Hutterer, R., eds.) Proceedings of the International Symposium on Vertebrate Biogeography and Systematics in the Tropics, pp. 309–319. Zoologisches Forschungsinstitut und Museum Alexander Koenig, Bonn.
- Kimura, M. 2002. The fomation and the paleogeology of the Ryuku Arc. In (Kimura, M., ed.) The Formation of the Ryukyu Arch and Migration of Biota to the Arc, pp. 19–54. The Okinawa Times Inc., Naha (in Japanese).
- Kurachi, M., Chau, B.-L., Dang, V.-B., Dorji, T., Yamamoto, Y., Nyunt, M. M., Maeda, Y., Chhum-Phith, L., Namikawa, T. and Yamagata, T. 2007a. Population structure of wild musk shrew (Suncus murinus) in Asia based on mitochondrial DNA variation, with research in Cambodia and Bhutan. Biochemical Genetics 45: 165–183.
- Kurachi, M., Kawamoto, Y., Tsubota, Y., Chau, B.-L., Dang, V. B., Dorji, T., Yamamoto, Y., Maung, M. N., Maeda, Y., Chhum-Phith, L., Namikawa, T. and Yamagata, T. 2007b. Phylogeography of wild musk shrew (*Suncus murinus*) populations in Asia based on blood protein/enzyme variation. Biochemical Genetics 45: 543–563.

Lin, X. D., Zhou, R. H., Fan, F. N., Ying, X. H., Sun, X. Y., Wang, W., Holmes, E. C. and Zhang, Y. Z. 2014. Biodiversity and evolution of Imjin virus and Thottapalayam virus in Crocidurinae shrews in Zheijang Province, China. Virus Research 189: 114–120.

- Meegaskumbura, S., Meegaskumbura, M. and Schneider, C. J. 2010. Systematic relationships and taxonomy of *Suncus montanus* and S. murinus from Sri Lanka. Molecular Phylogenetics and Evolution 55: 473–487.
- Meegaskumbura, S., Meegaskumbura, M. and Schneider, C. J. 2012a.
 Phylogenetic posistion of Suncus fellowesgordoni with pigmy shrews from Madagascar and Southeast Asia inferred from cytochrome-b. Ceylon Journal of Science (Biological Science) 41: 83–87.
- Meegaskumbura, S., Meegaskumbura, M. and Schneider, C. J. 2012b.
 Re-evaluation of the taxonomy of the Sri Lankan pigmy shrew
 Suncus fellowesgordoni (Soricidae: Crocidurinae) and its phylogenetic relationship with S. etruscus. Zootaxta 3187: 57–68.
- Meegaskumbura, S. H., Meegaskumbura, M. P. B., Pethiyagoda, R., Manamendra-Arachchi, K. and Schneider, C. J. 2007. Crocidura hikmiya, a new shrew (Mammalia: Soricomorpha: Soricidae) from Sri Lanka. Zootaxa 1665: 19–30.
- Motokawa, M. 2015. Suncus murinus (Linnaeus, 1766). In (Ohdachi, S. D., Ishibashi, Y., Iwasa, M. A., Fukui, D. and Saitoh, T., eds.) The Wild Mammals of Japan. 2nd edition, pp. 26–27. Shoukadoh, Kyoto.
- Nishioka, Y., Nakagawa, R., Nunami, S. and Hirasawa, S. 2016. Small mammalian remains from the late Holocene deposits on Ishigaki and Yonaguni Islands, southwestern Japan. Zoological Studies 55: 1–21.
- Ohdachi, S. D., Hasegawa, M., Iwasa, M. A., Abe, H., Vogel, P., Oshida, T. and Lin, L.-K. 2006. Molecular phylogenetics of soricid shrews (Mammalia) based on mitochondrial cytochrome b gene sequences: with special reference to the Soricinae. Journal of Zoology, London 270: 177-191.
- Ohdachi, S. D., Iwasa, M. A., Nesterenko, V. A., Abe, H., Masuda, R. and Haberl, W. 2004. Molecular phylogenetics of *Crocidura* shrews (Insectivora) in East and Central Asia. Journal of Mammalogy 85: 396–403.
- Peterson, G. D. J. 1956. Suncus murinus, a recent introduction to Guam. Journal of Mammalogy 37: 278–279.
- Phillips, W. W. A. 1980. Manual of the Mammals of Sri Lanka. Part 1.

- 2nd edition. Wildlife and Nature Protection Society of Sri Lanka, Unknown Binding, 116 pp.
- Suzuki, Y., Tomozawa, M., Koizumi, Y., Tsuchiya, K.and Suzuki, H. 2015. Estimating the molecular evolutionary rates of mitochondrial genes referring to Quaternary ice age events with inferred population expansions and dispersals in Japanese *Apodemus*. BMC Evolutionary Biology 15: 187
- Tamura, K., Peterson, D., Peterson, N., Stecher, G., Nei, M. and Kumar, S. 2011. MEGA5: Molecular Evolutionary Genetics Analysis Using Maximum Likelihood, Evolutionary Distance, and Maximum Parsimony Methods. Molecular Biology and Evolution 28: 2731–2739.
- Voris, H. K. 2000. Map of Pleistocene sea levels in Southeast Asia: shorelines, river systems and time durations. Journal of Biogeography 27: 1153–1167.
- Wiewel, A. S., Yackel-Adams, A. A. and Rodda, G. H. 2009. Distribution, density, and biomass of introduced small mammals in the southern Mariana Islands. Pacific Science 63: 205–222.
- Yamagata, T. and Kurachi, M. 2005. Phylogenetic evolution on wild populations of the musk shrew in Asia based on mitochondrial DNA. The Journal of Animal Genetics 32: 47–54 (in Japanese).
- Yamagata, T., Kurachi, M., Tanaka, K., Maeda, Y., Win, N., Hla, T., Daing, T. and Nyunt, M. M. 2004. Genetic differentiation of wild musk shrews in Myanmar based on morphology and mitochondrial DNA variation. Report of the Society for Researches on Native Livestock 21: 257–266.
- Yamagata, T., Ohishi, K., Faruque, M. O., Masangkay, J. S., Ba-Loc, C., Vu-Binh, D., Mansjoer, S. S., Ikeda, H. and Namikawa, T. 1995. Genetic variation and geographic distribution on the mitochondrial DNA in local population of the musk shrew, Suncus murinus. Japanese Journal of Genetics 70: 321–337.
- Yapa, A. and Ratnavira, G. 2013. The Mammal of Sri Lanka. Field Ornithology Group of Sri Lanka, Colomobo, 1012 pp.
- Yosida, T. H. 1982. Cytogenetical studies on Insectivora. II. Geographical variation of chromosomes in the house shrew, Suncus murinus (Soricidae), in East, Southeast and Southwest Asia, with a note on the karyotype evolution and distribution. The Japanese Journal of Genetics 57: 101–111.

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Appendix 1. Localities and accession number of DNA database of the Suncus murinus complex and outgroups of crocidurine shrews

Loc. #	OTU code	Taxonomic treatment	Accession #	Source	Locality
1	MKJP1	S. murinus	LC126448	PS	Ikema Island, Okinawa Pref., Japan
1	MKJP2	S. murinus	LC126449	PS	ditto
2	YGJP1	S. murinus	LC126450	PS	Yonaguni Island, Okinawa Pref., Japan
2	YGJP2	S. murinus	LC126451	PS	ditto
2	YGJP3	S. murinus	LC126452	PS	ditto
2	YGJP4	S. murinus	LC126453	PS	ditto
2	YGJP5	S. murinus	LC126454	PS	ditto
2	YGJP6	S. murinus	LC126455	PS	ditto
2	YGJP7	S. murinus	LC126456	PS	ditto
2	YGJP8	S. murinus	LC126457	PS	ditto
2	YGJP9	S. murinus	LC126458	PS	ditto
2	YGJP10	S. murinus	LC126459	PS	ditto
2	YGJP11	S. murinus	LC126460	PS	ditto
2	YGJP12	S. murinus	LC126461	PS	ditto
2	YGJP13	S. murinus	LC126462	PS	ditto
2	YGJP14	S. murinus	LC126463	PS	ditto
2	YGJP15	S. murinus	LC126464	PS	ditto
2	YGJP16	S. murinus	LC126465	PS	ditto
2	YGJP17	S. murinus	LC126466	PS	ditto
2	YGJP18	S. murinus	LC126467	PS	ditto
2	YGJP19	S. murinus	LC126468	PS	ditto
2	YGJP20	S. murinus	LC126469	PS	ditto
2	YGJP21	S. murinus	LC126470	PS	ditto
2	YGJP22	S. murinus	LC126471	PS	ditto
2	YGJP23	S. murinus	LC126472	PS	ditto
2	YGJP24	S. murinus	LC126473	PS	ditto
2	YGJP25	S. murinus	LC126474	PS	ditto
3	TCTW1	S. murinus	AB175075	1	Taichung, Taiwan
4	WZCN1	S. murinus	JF784169	DB	Longwan District, Wenzhou City, Zhejiang Province, Chi
4	WZCN2	S. murinus	JF784170	DB	ditto
4	WZCN3	S. murinus	JF784171	DB	ditto
4	WZCN4	S. murinus	KJ420556	2	ditto
4	WZCN5	S. murinus	KJ420557	2	ditto
5	WCCN1	S. murinus	KJ420549	2	Wencheng, Zhejiang Province, China
5	WCCN2	S. murinus	KJ420550	2	ditto
5	WCCN3	S. murinus	KJ420551	2	ditto
5	WCCN4	S. murinus	KJ420552	2	ditto
6	YHCN1	S. murinus	KJ420553	2	Yuhuan, Zhejiang Province, China
6	YHCN2	S. murinus	KJ420554	2	ditto
6	YHCN3	S. murinus	KJ420555	2	ditto
7	MLVN1	S. murinus	LC126475	PS	Mê Linh District, Vĩnh Phúc Province, Vietnam
7	MLVN2	S. murinus	LC126476	PS	ditto
7	MLVN3	S. murinus	LC126477	PS	ditto
7	MLVN4	S. murinus	LC126478	PS	ditto
7	MLVN5	S. murinus	LC126479	PS	ditto
7	MLVN6	S. murinus	LC126480	PS	ditto
7	MLVN7	S. murinus	LC126481	PS	ditto
7	MLVN8	S. murinus	LC126482	PS	ditto
7	MLVN9	S. murinus	LC126483	PS	ditto

Appendix 1. (continued)

Loc. #	OTU code	Taxonomic treatment	Accession #	Source	Locality
8	NTVN1	S. murinus	LC126484	PS	Ngọc Thanh, Vĩnh Phúc Province, Vietnam
8	NTVN2	S. murinus	LC126485	PS	ditto
8	NTVN3	S. murinus	LC126486	PS	ditto
8	NTVN4	S. murinus	LC126487	PS	ditto
8	NTVN5	S. murinus	LC126488	PS	ditto
8	NTVN6	S. murinus	LC126489	PS	ditto
8	NTVN7	S. murinus	LC126490	PS	ditto
8	NTVN8	S. murinus	LC126491	PS	ditto
8	NTVN9	S. murinus	LC126492	PS	ditto
8	NTVN10	S. murinus	LC126493	PS	ditto
9	HNVN1	S. murinus	LC126494	PS	Dong Anh District, Ha Noi Province, Vietnam
9	HNVN2	S. murinus	LC126495	PS	ditto
10	OHVN1	S. murinus	LC126496	PS	Quỳ Hợp District, Nghe An Province, Vietnam
10	OHVN2	S. murinus	LC126497	PS	ditto
10	OHVN3	S. murinus	LC126498	PS	ditto
10	OHVN4	S. murinus	LC126499	PS	ditto
10	OHVN5	S. murinus	LC126500	PS	ditto
10	OHVN6	S. murinus	LC126501	PS	ditto
10	OHVN7	S. murinus	LC126502	PS	ditto
11	KSVN1	S. murinus	LC126503	PS	Khe Sanh, Hướng Hóa District, Quảng Trị Province, Vietna
11	KSVN2	S. murinus	LC126504	PS	ditto
12	DHVN1	S. murinus	LC126505	PS	Đồng Hới, Quảng Bình, Province, Vietnam
13	LNVN1	S. murinus	LC126506	PS	Liên Nghĩa Town, Lâm Đồng Province, Vietnam
13	LNVN2	S. murinus	LC126507	PS	ditto
13	LNVN3	S. murinus	LC126508	PS	ditto
13	LNVN4	S. murinus	LC126509	PS	ditto
13	LNVN5	S. murinus	LC126510	PS	ditto
14	KLMY1	S. murinus	LC126511	PS	Ulu Gombak, Kuala Lumpur, Malaysia
15	MLMY1	S. murinus	LC126512	PS	Taman Bukit Katil Indah, Melaka, Malaysia
15	MLMY2	S. murinus	LC126513	PS	ditto
15	MLMY3	S. murinus	LC126514	PS	ditto
15	MLMY4	S. murinus	LC126515	PS	ditto
15	MLMY5	S. murinus	LC126516	PS	ditto
15	MLMY6	S. murinus	LC126517	PS	ditto
15	MLMY7	S. murinus	LC126518	PS	ditto
16	CNIN1	S. murinus	LC126519	PS	Cibinong, Bogor Regancy, West Java, Indonesia
16	CNIN2	S. murinus	LC126520	PS	ditto
16	CNIN3	S. murinus	LC126521	PS	ditto
16	CNIN4	S. murinus	LC126522	PS	ditto
16	CNIN5	S. murinus	LC126523	PS	ditto
17	BGIN1	S. murinus	LC126524	PS	Kec. Tamansari, Bogor, West Java, Indonesia
18	YGMN1	S. murinus	LC126525	PS	Kamayut Township, Yangon, Myanmar
19	TTMN1	S. murinus	LC126526	PS	Twantay (Tontay), Yangon Region, Myanmar
19	TTMN2	S. murinus	LC126527	PS	ditto
20	PTMN1	S. murinus	LC126528	PS	Pathein, Ayeyarwady Region, Myamnar
20	PTMN2	S. murinus	LC126529	PS	ditto
21	YZMN1	S. murinus	LC126530	PS	Yezin, Nay Pyi Taw, Myanmar
22	NSMN1	S. murinus	LC126531	PS	Nyaung Shwe, Shan State, Myanmar
22	NSMN2	S. murinus	LC126532	PS	ditto

Appendix 1. (continued)

Loc. #	OTU code	Taxonomic treatment	Accession #	Source	Locality
22	NSMN3	S. murinus	LC126533	PS	ditto
23	TGMN1	S. murinus	LC126534	PS	Taunggyi City, Shan State, Myanmar
23	TGMN2	S. murinus	LC126535	PS	ditto
23	TGMN3	S. murinus	LC126536	PS	ditto
23	TGMN4	S. murinus	LC126537	PS	ditto
24	MDMN1	S. murinus	LC126538	PS	Mandalay, Mandalay Region, Myanmar
24	MDMN2	S. murinus	LC126539	PS	ditto
24	MDMN3	S. murinus	LC126540	PS	ditto
25	KMMN1	S. murinus	LC126541	PS	Kalaymyo, Sagaing Province, Myanmar
26	TEMN1	S. murinus	LC126542	PS	Teetain, Chin State, Myanmar
26	TEMN2	S. murinus	LC126543	PS	ditto
26	TEMN3	S. murinus	LC126544	PS	ditto
27	Anura	S. m. murinus	EU122224	3	Anuradhapura, Sri Lanka
28	WPSL1	S. murinus	LC126545	PS	Watapuluwa, Kandy, Sri Lanka
28	WPSL2	S. murinus	LC126546	PS	ditto
28	WPSL3	S. murinus	LC126547	PS	ditto
28	WPSL4	S. murinus	LC126548	PS	ditto
28	WPSL5	S. murinus	LC126549	PS	ditto
29	KDSL1	S. murinus	LC126550	PS	Center of Kandy, Sri Lanka
29	KDSL2	S. murinus	LC126551	PS	ditto
29	KDSL3	S. murinus	LC126552	PS	ditto
29	KDSL4	S. murinus	LC126553	PS	ditto
29	KDSL5	S. murinus	LC126554	PS	ditto
30	Kandy	S. m. kandianus	GQ290366	4	Peradeniya, Kandy, Sri Lanka
31	Kitulg	S. montanus	GQ290373	4	Kitulgala, Sri Lanka
32	Colom	S. m. caerulescens	GQ290365	4	Colombo, Sri Lanka
33	Udawa	S. m. murinus	GQ290380	4	Pokunuthanne, Udawalawe, Sri Lanka
34	Kudaw	S. montanus	GQ290378	4	Kudawa, Sinharaja, Sri Lanka
35	Morni	S. montanus	GQ290375	4	Morningside, Sri Lanka
36	KRPNI	S. murinus	LC126555	PS	Karachi, Pakistan
36	KRPN2	S. murinus	LC126556	PS	ditto
36	KRPN3	S. murinus	LC126557	PS	ditto
36	KRPN4	S. murinus	LC126558	PS	ditto
36	KRPN5	S. murinus	LC126559	PS	ditto
36	KRPN6	S. murinus	LC126560	PS	ditto
36	KRPN7	S. murinus	LC126561	PS	ditto
36	KRPN8	S. murinus	LC126562	PS	ditto
36	KRPN9	S. murinus	LC126563	PS	ditto
36	KRPN10	S. murinus	LC126564	PS	ditto
37	KSIR1	S. murinus	LC126565	PS	Khorramshahr, Khuzestan State, Iran
38	ADIR1	S. murinus	LC126566	PS	Fayyeh, Abadan, Khuzestan State, Iran
39	LPRU1	S. murinus	LC126567	PS	Le Port, La Réunion Island
39	LPRU2	S. murinus	LC126568	PS	ditto
39		S. murinus	LC126569	PS	ditto
	LPRU3			PS	ditto
39	LPRU4	S. murinus	LC126570		
40	MJMG1	S. murinus	LC126571	PS	Mahajanga, Madagascar ditto
40	MJMG2	S. murinus	LC126572	PS	
40	MJMG3 MJMG4	S. murinus S. murinus	LC126573 LC126574	PS PS	ditto ditto

Appendix 1. (continued)

Loc. #	OTU code	Taxonomic treatment	Accession #	Source	Locality
40	MJMG5	S. murinus	LC126575	PS	ditto
40	MJMG6	S. murinus	LC126576	PS	ditto
40	MJMG7	S. murinus	LC126577	PS	ditto
41	ANMG1	S. murinus	LC126578	PS	Antananarivo, Madagascar
41	ANMG2	S. murinus	LC126579	PS	ditto
41	ANMG3	S. murinus	LC126580	PS	ditto
41	ANMG4	S. murinus	LC126581	PS	ditto
41	ANMG5	S. murinus	LC126582	PS	ditto
41	ANMG6	S. murinus	LC126583	PS	ditto
41	ANMG7	S. murinus	LC126584	PS	ditto
41	ANMG8	S. murinus	LC126585	PS	ditto
41	ANMG9	S. murinus	LC126586	PS	ditto
42	SMGC1	S. murinus	LC126587	PS	Salimani, Grande Comore Island
42	SMGC2	S. murinus	LC126588	PS	ditto
42	SMGC3	S. murimis	LC126589	PS	ditto
43	CNZB1	S. murinus	LC126590	PS	Chaani Vill., Zanzibar, Tanzania
43	CNZB2	S. murinus	LC126591	PS	ditto
43	CNZB3	S. murinus	LC126592	PS	ditto
43	CNZB4	S. murinus	LC126593	PS	ditto
44	KNZB1	S. murinus	LC126594	PS	Kibeni Vill., Zanzibar, Tanzania
44	KNZB2	S. murinus	LC126595	PS	ditto
44	KNZB3	S. murinus	LC126596	PS	ditto
	S_etruscus1	S. etruscus	JN556043	5	Chennai, South India
	S_etruscus2	S. etruscus	LC126597	PS	Haffār-e Shargī, Khūzestān state, Iran
	S_etruscus3	S. etruscus	FJ716836	5	Sri Lanka
	S_madagascariensis1	S. madagascariensis	JF817394	5	Madagascar
	S_madagascariensis2	S. madagascariensis	JF817395	5	Madagascar
	S_fellowesgordoni1	S. fellowesgordoni	JF914980	6	Agarapathana, Sri Lanka
	S_fellowesgordoni2	S. fellowesgordoni	JF914981	6	Agarapathana, Sri Lanka
	S_stoliczkanus	S. stoliczkanus	AB175076	1	Terai, Adhabar, Nepal
	C_rapax	Crocidura rapax	AB115557	DB	Taichung Hsien, Taiwan
	C_dsinezumi	Crocidura dsinezumi	HQ663926	DB	Hokkaido, Japan

PS, present study. DB, direct submission to the DNA database. 1, Ohdachi et al. (2006). 2, Lin et al. (2014). 3, Meegaskumbura et al. (2007). 4, Meegaskumbura et al. (2012a). 6, Meegaskumbura et al. (2012b).