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Morphology and Molecular Phylogeny of Some Tibetan Ground Beetles Belonging to the Subgenera *Neoplesius* and *Eocechenus* (Coleoptera, Carabidae)

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Abstract Morphological characters are described in detail for some Tibetan carabine beetles belonging to the subgenera *Neoplesius* and *Eocechenus* of the genus *Carabus* (s. lat.). *Carabus* (*Neoplesius*) *markamensis* is raised from the subspecies of *C.* (*N.*) *wagae* to an independent species, with descriptions of two new subspecies under the names *wangdanus* nov. and *rawuensis* nov. A molecular phylogenetic analysis of these beetles indicates that they form a single (monophyletic) cluster that shares a common ancestry with *Damaster* (s. lat.). No phylogenetic evidence is obtained to regard *Eocechenus* as a discrete subgenus from *Neoplesius*.

Neoplesius REITTER (sensu IMURA, 1996, and IMURA & MIZUSAWA, 1996) and *Eocechenus* SEMENOV-TIAN-SHANSKIJ et ZNOJKO are rather poorly known elements of the Chinese Carabina. These subgenera occur mainly on the high mountains of the so-called Tibet regions (western part of China represented by such areas as Xizang, western Sichuan, southeastern Qinghai and northwestern Yunnan). They are treated as two allied subgenera belonging to the subdivision Procrustimorphi in the division Multistriati of the grand genus *Carabus* (IMURA, 1996). Though not a few members of these groups have been newly introduced to science and though our knowledge on their distribution and local variation has been rapidly increasing in recent years, their morphological and phylogenetic evaluations have not been satisfactorily made; even detailed structure of the male genital organ has never been described nor figured for most species, so that the taxonomy of these and allied subgenera is still incomplete.

We had recently an opportunity to examine a series of specimens belonging to these two subgenera collected from several localities in the eastern part of Xizang Zizhiqu. Since morphology and molecular phylogeny are complements each of the other, a phylogenetic tree has been constructed based on the mitochondrial ND5 gene

sequences, and the results have been discussed in conjunction with their morphology, especially of male genital organ.

Materials and Methods

Abbreviations used in the text are the same as those in previous papers by SU *et al.* (1996 c), and the length of the specimens was measured from the apical tip of mandibles to the apices of elytra. The subtribe Carabina is regarded herein as to contain a single genus *Carabus* (s. lat.), as is now adopted by most authors, though it was treated as an assemblage of many distinct genera in the previous papers by SU *et al.* (1996 a–c).

In the present study, total twenty-nine specimens belonging to the genus *Carabus* (s. lat.) obtained from six different localities of the eastern part of Xizang Zizhiqu of West China (Fig. 1) were used. A few examples from each locality were killed and

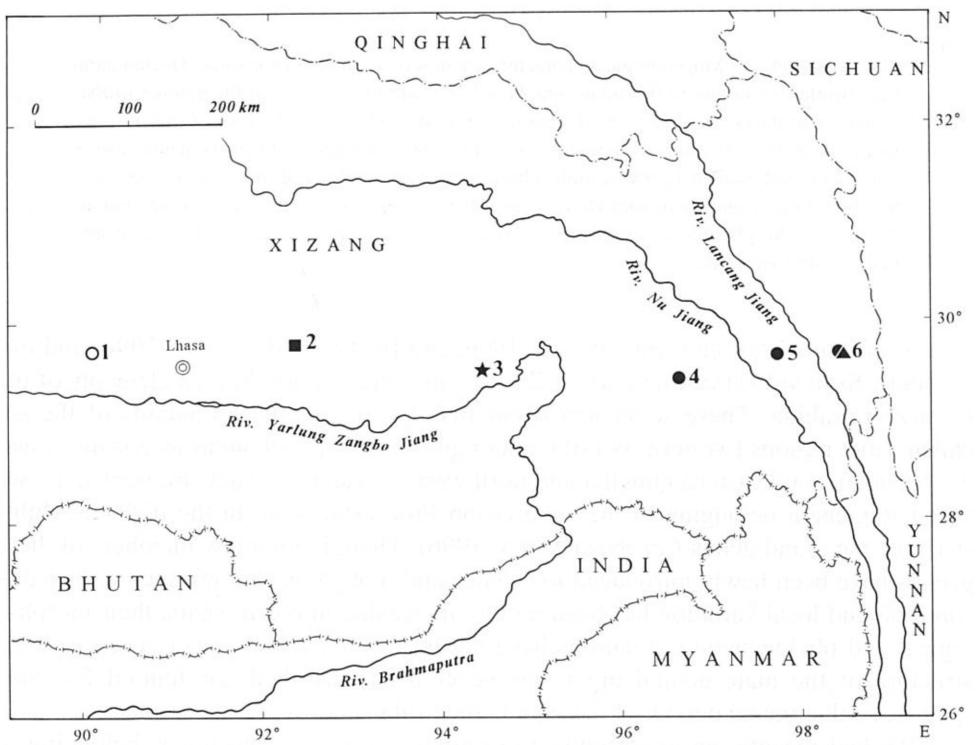


Fig. 1. Map showing the collecting sites of the materials in East Xizang Zizhiqu of West China; ○ – *Carabus* (*Neoplesius*) *wagae*; ● – *C.* (*N.*) *markamensis* subspp.; ■ – *C.* (*N.*) *borodini*; ★ – *C.* (*N.*) *ludmilae*; ▲ – *C.* (*Eocechenus*) *leptoplesioides*. —— 1, Near Margyang (*C. wagae*); 2, Pass Mi La (*C. borodini*); 3, Nyingchi (*C. ludmilae*); 4, Rawu (*C. markamensis rawuensis* nov.); 5, Wangda, SE of Zogang (*C. m. wangdanus* nov.); 6, Markam (*C. m. markamensis* & *C. leptoplesioides*).

stored in 95% ethanol until use to prevent DNA degradation, and the remaining samples were preserved as dried specimens for morphological examination.

A 1083 bp fragment of mitochondrial ND5 gene was amplified, and directly sequenced using a PRISM cycle sequencing kit and an ABI model 377 automated sequencer (Perkin-Elmer, USA). For the details of DNA extraction, PCR amplification and sequencing, see SU *et al.* (1996 a). The DNA sequences were aligned using CLUSTAL V (HIGGINS *et al.*, 1992) for phylogenetic analyses. The resulting data alignment was subsequently used to construct a phylogenetic tree by the neighbour-joining (NJ) method (SAITOU & NEI, 1987). Evolutionary distances were computed by KIMURA's two-parameter method (KIMURA, 1980). The tree was evaluated using bootstrap test based on 1,000 resamplings (FELSENSTEIN, 1985). All these procedures for phylogenetic analyses were carried out using a computer software program, BIORESEARCH/SINCA ver. 3.0 (Fujitsu System Engineering, Japan). For these analyses, the following reported ND5 gene sequences from nineteen Japanese (SU *et al.*,1996 a, b) and two French (SU *et al.*,1996 c) species were used: *Carabus* (*Limnocarabus*) *clathratus* (or *maacki*) *aquatilis* (from Aomori), D50358 (Gen Bank accession number); *C.* (*Euleptocarabus*) *porrecticollis porrecticollis* (from Chino-shi, Nagano) D50352; *C.* (*Hemicarabus*) *tuberculosus* (from Fukui), D50353; *C.* (*Homoeocarabus*) *maeander paludis* (from Hokkaido), D50354; *C.* (*Coptolabrus*) *fruhstorferi*, D50346; *C.* (*Damaster*) *blaptoides rugipennis* (from Hokkaido), D50351; *C.* (*D.*) *b. viridipennis* (from Aomori), D50428; *C.* (*D.*) *b. babaianus* (from Miyagi), D50348; *C.* (*D.*) *b. fortunei* (from Is. Awa-shima), D50426; *C.* (*D.*) *b. capito* (from Is. Sado), D50424; *C.* (*D.*) *b. oxuroides* (so-called "*cyanostola*", from Nagano) D50425; ditto (from Kawasaki), D50350; ditto (from Gifu), D50427; *C.* (*D.*) *b. blaptoides* (from Hiroshima), D50349; ditto (from Is. Shikoku), D50423; ditto (from Is. Fukué-jima of the Goto Islands., off western Kyushu), D50422; *C.* (*Acoptolabrus*) *gehinii radiatocostatus*, D50429; *C.* (*A.*) *g. aereicollis*, D50339; *C.* (*Megodontus*) *kolbei aino* (from the Daisetsu-zan Mts.), D50365; *C.* (*Megodontus*) *violaceus purpurascens* (from Burgundy, France), D86211; *C.* (*Procrustes*) *coriaceus coriaceus* (from Burgundy, France), D86206.

Results

Morphological Identification and Description of the Materials Used

Morphological examinations revealed that the specimens used were classified into seven taxa, i.e., four species consisting of six subspecies belonging to the subgenus *Neoplesius* and a single species to the subgenus *Eocechenus*.

1) ***Carabus* (*Neoplesius*) *wagae*** FAIRMAIRE, 1882

(Figs. 2, 8)

Length: 19.8–20.8 mm. Male genital organ as shown in Fig. 8; aedeagus rather

abruptly curved ventrad at basal third, with the median portion subcylindrical and almost parallel-sided, evidently tumid right laterad at about apical third, then rather deeply concave latero-ventrad at the right side; apical lobe short and subtriangular in shape, with the tip gently rounded and obviously compressed latero-dorsad; ostium lobe short and robust, with the apical part widely bilobate; endophallus with the basic structure almost identical with that of some Chinese subgenera belonging to the subdivision Procrustimorphi; neither paraligula nor basal lateral lobe is recognised, and median lobe also absent; prepraeputial lobes moderately inflated, both the lobes almost symmetrical; parapraeputial lobes well-developed, with the right one larger than the left; praeputial pad rather strongly protrudent dorsad in fully everted condition; podian lobe (cf. IMURA, 1997, p. 225) medium-sized and moderately produced ventrad; aggonoporius not sclerotized but characterized by a mere membraneous peripheral rim of gonopore.

Specimens examined. 5 ♂♂ (1 ♂ for DNA analysis), near Margyang between Pass Shogu La and Pass Dongu La, 4,500 m alt., ca. 100 km distant to the west from Lhasa, central eastern Xizang, China, 15–VII–1996, in coll. Y. IMURA.

Notes. The population from near Margyang almost agrees in morphology with the nominotypical subspecies described from the northern part of India (without indication of exact locality), though the head and pronotum of our specimens seem to be a little more strongly punctate and the elytral costae a little more strongly crenulate.

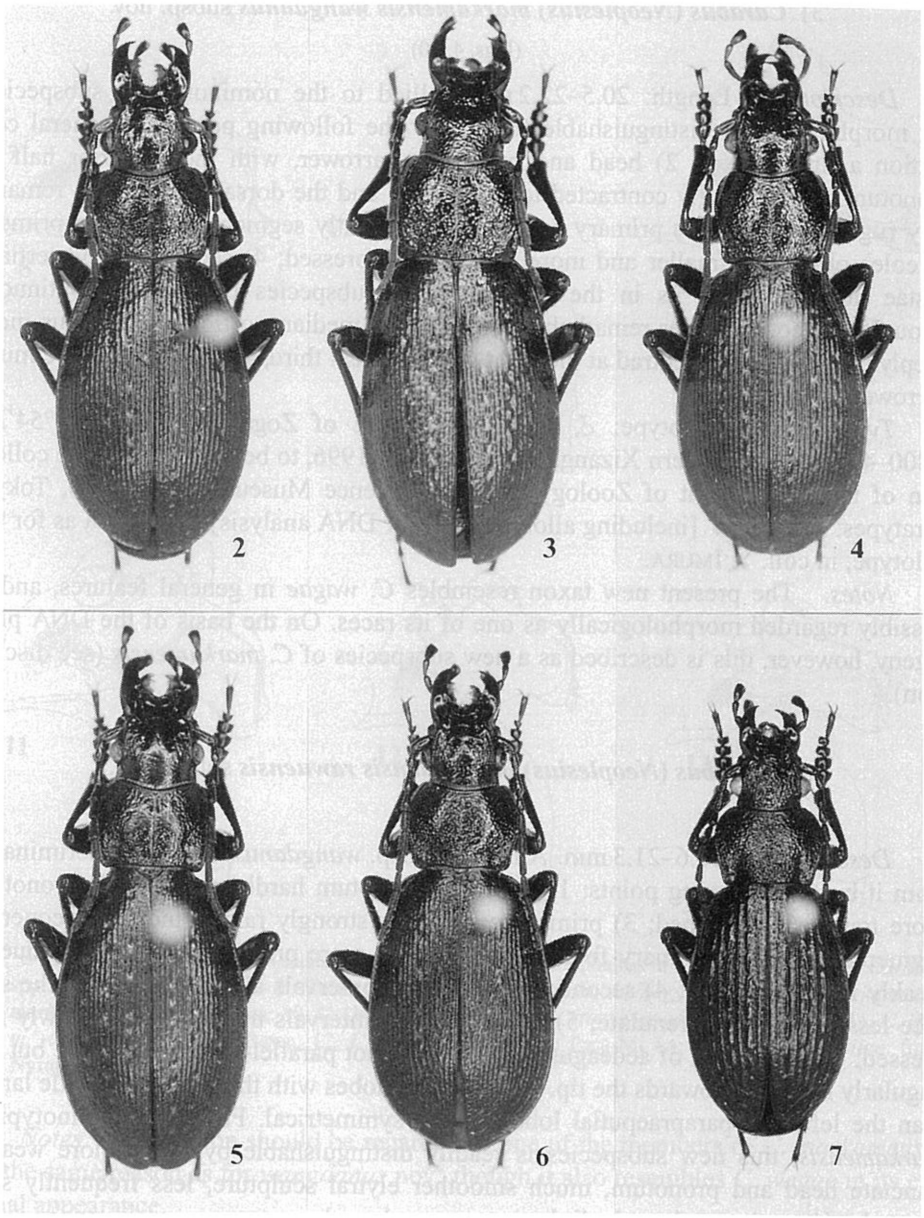
2) ***Carabus (Neoplesius) markamensis*** DEUVE, 1992, stat. nov.

(Figs. 3, 9)

Length: 22.0–22.7 mm. External morphology as described by DEUVE (1992, p. 270, fig. 12). Male genitalia as shown in Fig. 9; basic structure of aedeagus almost identical with that of *C. wagae*, though the apical portion is less strongly arcuate ventrad, its basal part being much more shallowly concave latero-ventrad at the right side, and the apical lobe robuster.

Specimens examined. 2 ♂♂, 1 ♀ (1 ♀ for DNA analysis), Markam env. (29°40′–42′N/98°35′E), 3,800–3,900 m alt., near the eastern end of Xizang, China, 17–VII–1996, in coll. Y. IMURA.

Notes. Since the two male examples examined are more or less teneral, we were unable to show the endophallus of this taxon in a fully everted condition. However, the dissection of its membraneous wall revealed near-identity of the basic structure of endophallus with that of the preceding species. Though originally described as a subspecies of *Carabus wagae*, we regarded it as a distinct species based on the mitochondrial DNA phylogeny (see discussion).



Figs. 2–7. *Carabus* (*Neoplesius*) spp. from East Xizang. —— 2, *Carabus* (*Neoplesius*) *wagae*, ♂, from Margyang; 3, *C.* (*N.*) *markamensis markamensis*, ♂, from Markam; 4, *C.* (*N.*) *m. wangdanus* nov., ♂, holotype, from Wangda, SE of Zogang; 5, *C.* (*N.*) *m. rawuensis* nov., ♂, holotype, from Rawu; 6, *C.* (*N.*) *borodini*, ♂, from Pass Mi La; 7, *C.* (*N.*) *ludmilae*, ♂, from Nyingchi. Scale: 2 mm.

3) *Carabus* (*Neoplesius*) *markamensis wangdanus* subsp. nov.

(Figs. 4, 10)

Description. Length: 20.5–22.2 mm. Allied to the nominotypical subspecies, but morphologically distinguishable from it by the following points: 1) general coloration a little darker; 2) head and pronotum narrower, with the posterior half of pronotum more strongly contracted towards base, and the dorsal surface less remarkably rugoso-punctate; 3) primary costae less frequently segmented, with the primary foveoles obviously smaller and more shallowly impressed; 4) secondary and tertiary costae not segmented as in the nominotypical subspecies but almost continuous throughout, though rather remarkably crenulate; 5) median portion of aedeagus more deeply concave latero-ventrad at the right side in apical third, and the apical lobe much narrower.

Type series. Holotype: ♂, Wangda env., SE of Zogang (29°38′N/97°54′E), 3,800–4,400 m alt., eastern Xizang, China, 14–VII–1996, to be preserved in the collection of the Department of Zoology, National Science Museum (Nat. Hist.), Tokyo. Paratypes: 2 ♂♂, 3 ♀♀ [including allotype (1 ♀) for DNA analysis], same data as for the holotype, in coll. Y. IMURA.

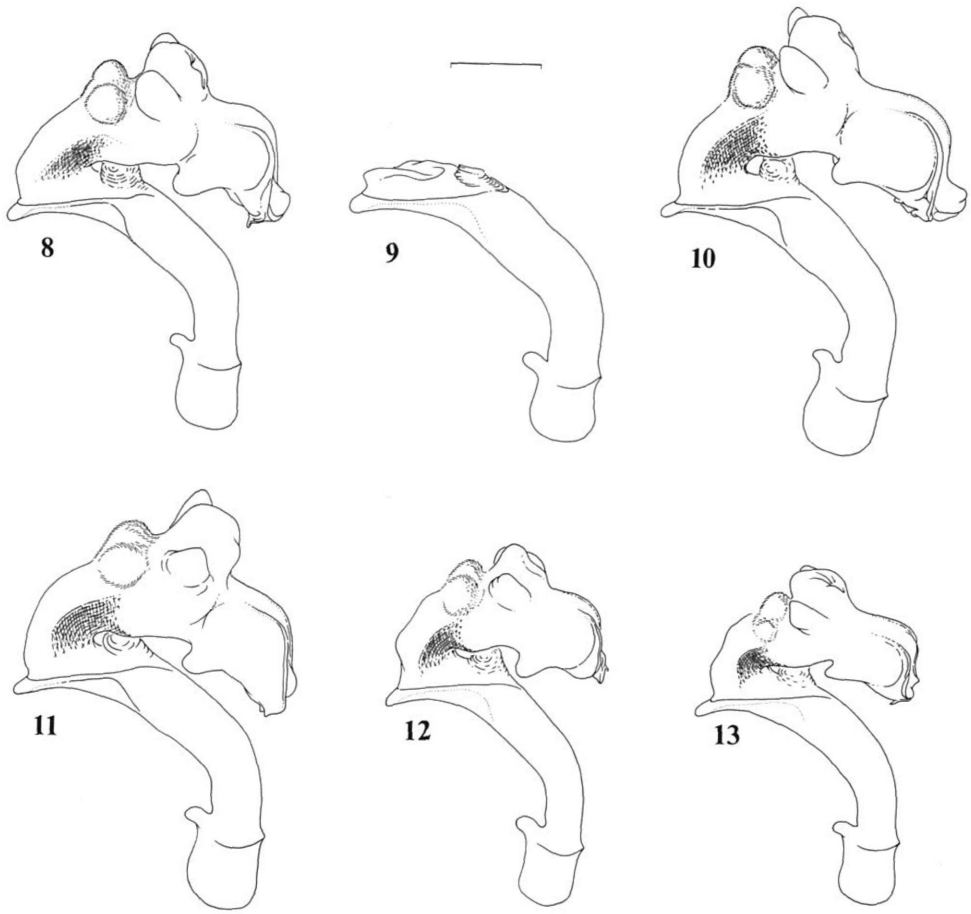
Notes. The present new taxon resembles *C. wagae* in general features, and is possibly regarded morphologically as one of its races. On the basis of the DNA phylogeny, however, this is described as a new subspecies of *C. markamensis* (see discussion).

4) *Carabus* (*Neoplesius*) *markamensis rawuensis* subsp. nov.

(Figs. 5, 11)

Description. 20.6–21.3 mm. Allied to subsp. *wangdanus* nov., but discriminated from it by the following points: 1) head and pronotum hardly punctate; 2) pronotum more transversely shaped; 3) primary costae less strongly raised and less frequently segmented, with the primary foveoles smaller but more prominently observed due to weakly raised intervals; 4) secondary and tertiary intervals also weaker, with the surface less remarkably crenulate; 5) striae between intervals much more shallowly impressed; 6) apical lobe of aedeagus a little wider, not parallel-sided at the base but triangularly narrowed towards the tip. Prepraeputial lobes with the right one a little larger than the left, and parapraeputial lobes almost symmetrical. From the nominotypical *markamensis*, this new subspecies is readily distinguishable by much more weakly punctate head and pronotum, much smoother elytral sculpture, less frequently segmented primary intervals and a little narrower aedeagal apex.

Type series. Holotype: ♂, N of Rawu, (29°33′N/96°47′E), 4,200 m alt., eastern Xizang, China, 11–VII–1996, to be preserved in the collection of the Department of Zoology, National Science Museum (Nat. Hist.), Tokyo. Paratypes: 2 ♂♂, 3 ♀♀ [including allotype (1 ♀) for DNA analysis], same data as for the holotype, in coll. Y. IMURA.



Figs. 8–13. Male genital organ (aedeagus with fully everted endophallus in right lateral view) of *Carabus* spp. from East Xizang. —— 8, *Carabus* (*Neoplesius*) *wagae*, from Margyang; 9, *C.* (*N.*) *markamensis markamensis*, from Markam; 10, *C.* (*N.*) *m. wangdanus* nov., from Wangda, SE of Zogang; 11, *C.* (*N.*) *m. rawuensis* nov., from Rawu; 12, *C.* (*N.*) *borodini*, from Pass Mi La; 13, *C.* (*N.*) *ludmilae*, from Nyingchi. Scale: 2 mm.

Notes. This taxon should be regarded as one of the members of *C. markamensis* for the same reason as for *wangdanus* nov., though it also resembles *C. wagae* in its external appearance.

5) ***Carabus* (*Neoplesius*) *borodini*** HEINZ, 1996

(Figs. 6, 12)

Length: 19.2–21.0 mm. External morphology as fully described and figured by

HEINZ (1996, pp. 20–22, figs. 1–2). Male genitalia as shown in Fig. 12; aedeagus a little smaller in size than in the above two species, hardly concave latero-ventrad at the right side in apical third, with the apex more sharply pointed; basic structure of endophallus not remarkably different from that of *C. wagae* and *C. markamensis*, though somewhat robuster in fully everted condition; apical part of ostium lobe widely bilobed, prepraeputial lobes with the right one a little more strongly produced than the left, and parapraeputial lobes almost symmetrical.

Specimens examined. 3 ♂♂, 2 ♀♀ (1 ♀ for DNA analysis), Pass Mi La (=Pa La) (29°46′N/92°19′E), 4,600 m alt., eastern Xizang, China, 15–VI–1996, in coll. Y. IMURA.

Notes. This is a monotypical species recently described from the Pass Mi La. Our materials came from the type locality, and agree in morphology with the type series.

6) *Carabus* (*Neoplesius*) *ludmilae* DEUVE, 1992

(Figs. 7, 13)

Length: 17.2–20.4 mm. External morphology as described and figured by DEUVE (1992, pp. 268–269, fig. 5) and IMURA & MIZUSAWA (1996, pl. 61, fig. 515). Male genitalia as shown in Fig. 13; aedeagus similar in shape to that of *C. borodini*, but a little robuster as a whole, less acutely curved ventrad at the base, with the median portion gradually divergent towards apex which is slenderer and subparallel-sided in lateral view; endophallus somewhat different in details — prepraeputial lobes with the right one smaller than the left though more strongly produced dorsad, podian lobe triangularly pointed and directed towards base, and apical portion more narrowly elongated.

Specimens examined. 1 ♂, 2 ♀♀ (1 ♀ for DNA analysis), N to NE Nyingchi (29°38′–42′N/94°29′–36′E), 4,300–4,500 m alt., eastern Xizang, China, 21~28–VI–1996, in coll. Y. IMURA.

Notes. This species is also monotypical and is known only from the northern hills of Nyingchi. Our materials were collected in the NNE part of the same village, a few hundreds meters away from the type locality, and are somewhat different in the external features, i.e., a narrower pronotum and a simpler elytral sculpture, etc., which may be regarded as a mere individual variation within the same species.

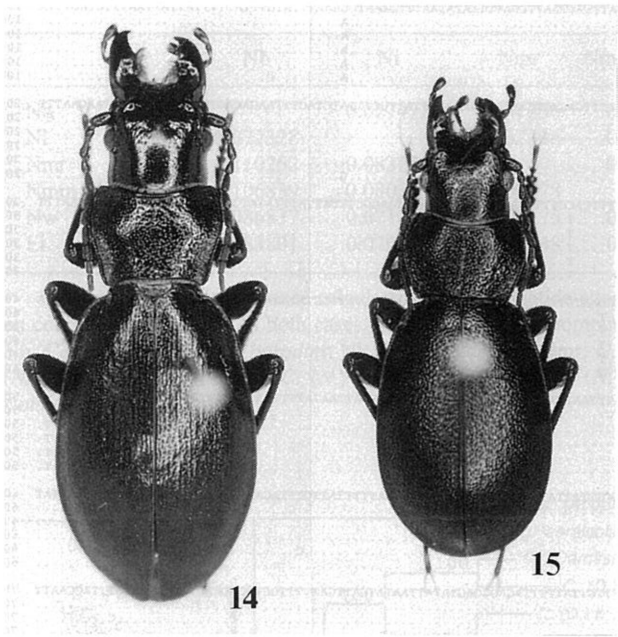
7) *Carabus* (*Eocechenus*) *leptoplesioides* DEUVE, 1992

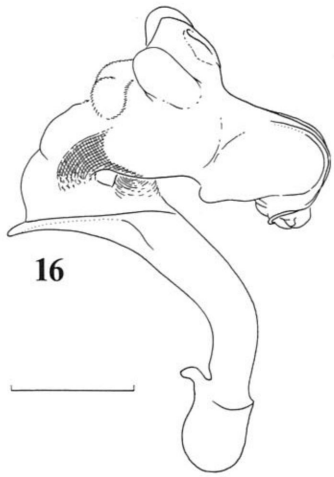
(Fig. 14)

Length: 23.6 mm. External morphology as described and figured by DEUVE, (1992, pp. 270–271, fig. 14) and IMURA & MIZUSAWA (1996, pl. 60, fig. 499).

Specimen examined. 1 ♀ (for DNA analysis), Markam env. (29°40′–42′N/98°35′E), 3,900 m alt., near the eastern end of Xizang, China, 16~17–VII–1996.

Notes. Only two female type specimens of the nominotypical *leptoplesioides* have hitherto been known. Our material becomes the third sample.





Figs. 14–16. *Carabus* (*Eocechenus*) spp. from China. —— 14, *Carabus* (*Eocechenus*) *leptoplesioides*, ♀, holotype, from Markam; 15–16, *C.* (*E.*) *kaznakovi*, ♂, from N of Nangqên, SE Qinghai, 15, imago, 16, aedeagus with fully everted endophallus in right lateral view. Scale: 2 mm.

Molecular Phylogeny Based on the DNA Sequences of the Mitochondrial ND5 Gene

The DNA sequence of a 1083-bp PCR product was determined for the listed 7 Tibetan carabids. The sequence contained a 1069 bp 3′-region of the ND5 gene, 7 bp of noncoding sequence, and a 7 bp 5′-terminus of the Phe-tRNA gene (Fig. 17). No length variations were found in all DNA sequences determined. The G+C contents of the sequenced region were nearly constant (21±1%) throughout the samples. The genetic distances between the Tibetan carabids estimated by KIMURA's two-parameter model of base substitution (KIMURA, 1980) ranged from 0.2 to 4.1% in all sites, and 0.3 to 11.0% in codon third positions (Table 1). A specimen of *Carabus* (*Neoplesius*) *markamensis markamensis* was identical in the DNA sequence with that of *C.* (*N.*) *m. wangdanus*, though they were collected at two different localities and could be morphologically discriminated from each other (see discussion). Of 1,069 sites in the ND5 gene, 80 were variable sites, and most substitutions (mainly transitions) occurred at the codon third positions (Fig. 17). The sequences of the ND5 gene region were used to analyze the phylogenetic relationships of the Tibetan carabids. To know their phylogenetic positions in the Carabina, 21 reported ND5 gene sequences of some carabine

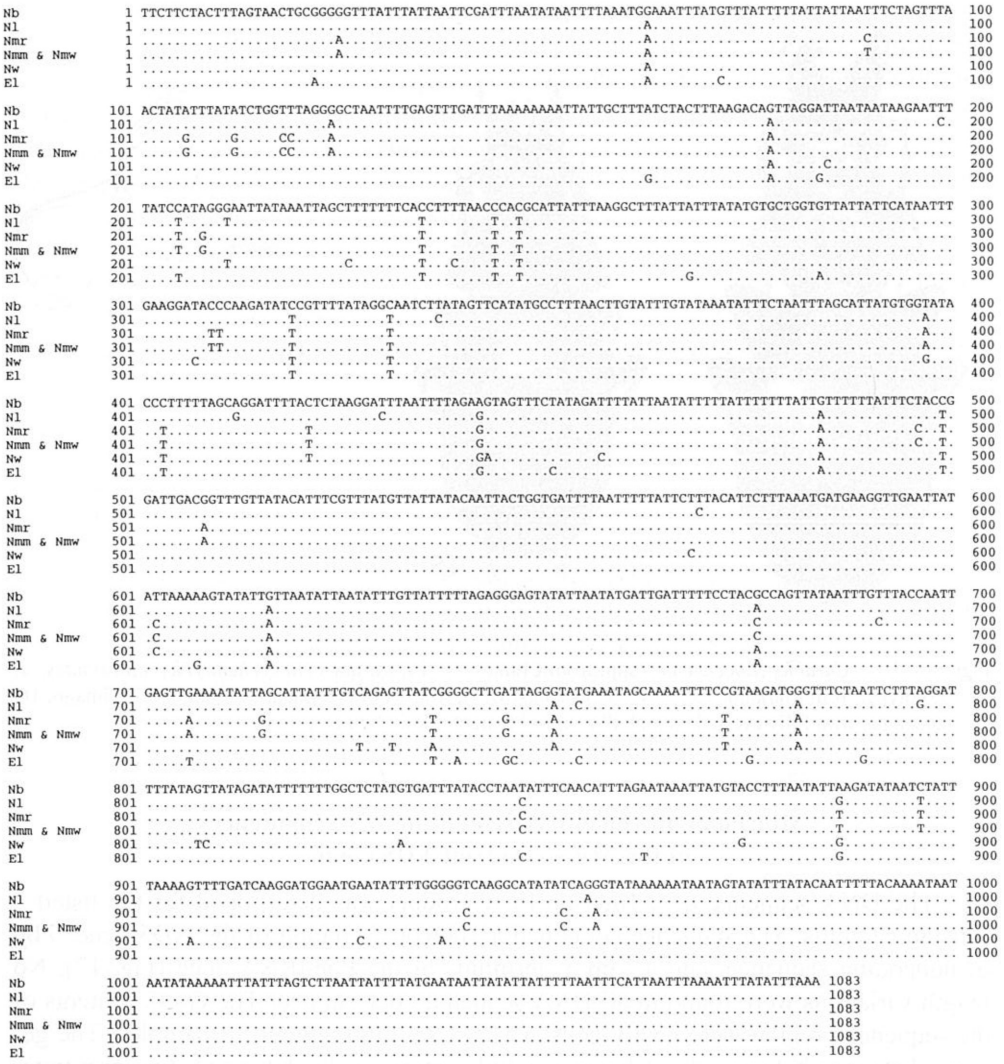


Fig. 17. Nucleotide sequence alignment of 1083 bp amplified from mitochondrial genome of 7 specimens of Tibetan carabids. Position 1–1069: 3′-region of ND5 gene; 1070–1076: flanking sequence; 1077–1083: 5′-terminal sequence of Phe-tRNA gene. For abbreviations of scientific names, see Table 1. Dot indicates identical nucleotide.

species from Japan and France were added to this analysis. A phylogenetic tree constructed by NJ method is shown in Fig. 18. All Tibetan beetles analyzed formed a single cluster, and were clearly monophyletic as supported by a high bootstrap value (100%) (see discussion). This cluster constituted a large group together with the *Coptolabrus–Damaster–Acoptolabrus* lineage that is composed of *C.* (*Coptolabrus*) *fruh-*

Table 1. Distance Matrix.

	Nb	Nl	Nmr	Nmm & Nmw	Nw	El
Nb		0.027472	0.041256	0.039260	0.036195	0.031355
Nl	0.077328		0.032336	0.030376	0.032310	0.028379
Nmr	0.110252	0.083579		0.001850	0.041124	0.039165
Nmm & Nmw	0.106839	0.080343	0.002778		0.039136	0.037183
Nw	0.086837	0.071131	0.093175	0.089886		0.040073
El	0.083801	0.070591	0.092948	0.089669	0.089669	

Upper-right matrix: distance calculated on all nucleotide sites. Lower-left matrix: distance calculated on codon third positions. In both cases, the distances were computed by KIMURA's two-parameter method. Nb: *Carabus* (*Neoplesius*) *borodini*; Nl: *C.* (*N.*) *ludmilae*; Nmr: *C.* (*N.*) *markamensis rawuensis*; Nmm: *C.* (*N.*) *m. markamensis*; Nmw: *C.* (*N.*) *m. wangdanus*; Nw: *C.* (*N.*) *wagae*; El: *C.* (*Eocechenus*) *leptoplesioides*.

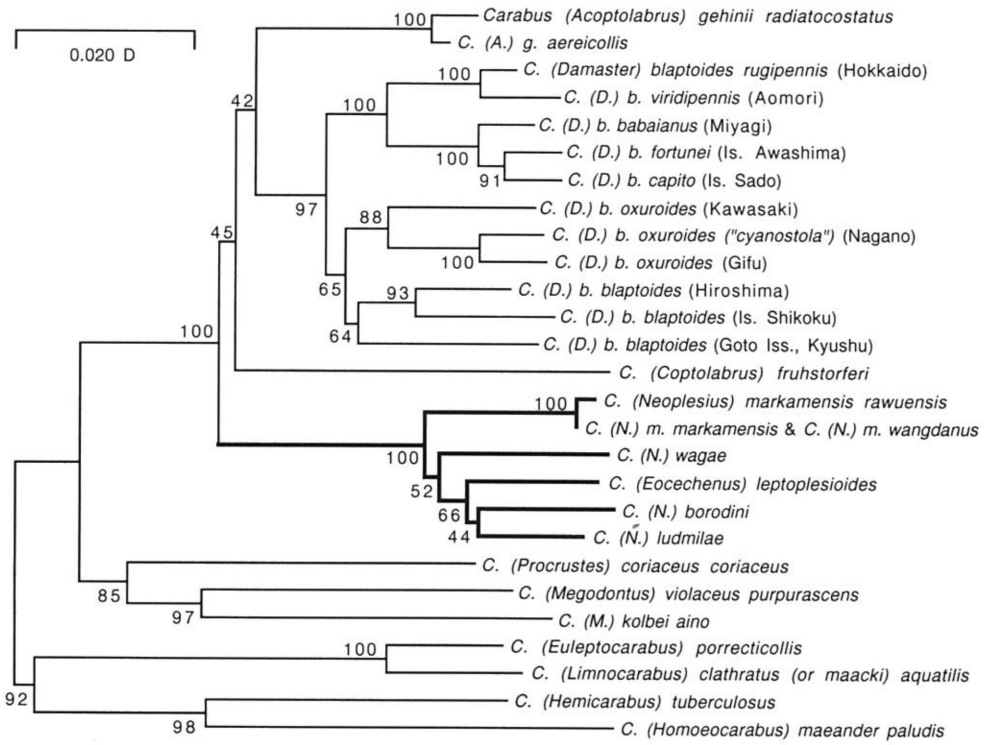


Fig. 18. Neighbour-joining tree based on ND5 gene sequence of 7 Tibetan carabids together with 21 other carabine species from Japan and France. Localities of some specimens are indicated in parentheses following the scientific name. D denotes the evolutionary distance corrected by KIMURA's two-parameter model. The bootstrap confidence level (based on 1,000 resamplings) is shown at each branching point.

storferi, *C.* (*Acoptolabrus*) *gehinii* and all the subspecies of *C.* (*Damaster*) *blaptoides*. The *Procrustes–Megodontus* lineage was the out-group of them. Four Japanese carabine species, *C.* (*Limnocarabus*) *clathratus* (or *maacki*) *aquatilis*, *C.* (*Euleptocarabus*) *porrecticollis*, *C.* (*Hemicarabus*) *tuberculosus* and *C.* (*Homoeocarabus*) *maeander paludis* [as well as other subgenera in the Carabina examined (not shown; see SU *et al.*, 1996 c)] constituted the out-group of all the members mentioned above.

Discussion

One of the remarkable findings is the relationship between the morphology and molecular phylogeny of the species-complex composed of *Carabus* (*Neoplesius*) *wagae* and *C.* (*N.*) *markamensis*. Morphologically, specimens in all the populations from the sites 1, 4, 5 and 6 (Fig. 1) can be unified into a single species, but showing apparent subspecific differentiation linked to their locality. The population from the site 1 and those from the sites 4 to 6 are separated with a rather deep branching point in the ND5 tree. They may be regarded as two distinct groups, and are conventionally treated as two species. On the other hand, though the samples from the sites 4, 5 and 6 (the *markamensis* complex) revealed the locality-dependent morphological variations, their DNA sequences are almost identical. This suggests recent radiation of this complex with a rapid morphological differentiation.

Whether the morphological differences recognized above can be taken as species or subspecies criteria will be debated. In this connection, it should be noted that there are no "objective" morphological criteria to define the species, subspecies or else, and one must sometimes settle the taxonomic rank on the "subjective" criteria. Molecular phylogeny does not give a solution to this problem, because the extent of morphological differentiation does not always run parallel with the time elapsed after emergence (as estimated by the branch length in the phylogenetic tree). The species- or subspecies-rankings in this paper should be considered conventional simply to distinguish the locality-linked and morphologically separable populations.

Also remarkable is the relationship between *Neoplesius* and *Eocechenus* deduced from ND5 phylogenetic tree. Generally, these two higher taxa are treated as two independent subgenera, consisting of some twenty species and nine species, respectively, and they are discriminated from each other by the following morphological key.

Key to the Subgenera

1 (2) Head normal or at most moderately hypertrophic; retinaculum of the right mandible bidentate; submentum asetose; elytral sculpture usually conspicuously carved with heterodynamic intervals; male foretarsus with hair pads in the basal four segments . . . . . *Neoplesius*.

2 (1) Head unusually hypertrophic with smaller eyes; retinaculum of the right mandible unidentate; submentum setiferous in some species; elytral sculp-

ture very weakly carved with homodynamic intervals; male foretarsus with hair pads only in the basal three segments. . . . . *Eocechenus*.

From the ND5 tree, however, *Carabus* (*Eocechenus*) *leptoplesioides* clearly belongs to a monophyletic cluster to which all the *Neoplesius* species analyzed also belong. Thus, the DNA phylogeny provides no evidence to support the taxonomical independence of *Eocechenus* from *Neoplesius*. This is consistent with the fact that both the subgenera are identical in the basic structure of the male genital organ, especially in the construction of endophallus observed under a fully everted condition.[1)] In other words, endophallic structure of the male genital organ would reflect the reasonable phylogenetic relationship among the higher taxa of the Carabina more than the external features would, at least in the groups treated in this study. The synonymization of *Eocechenus* with *Neoplesius* is awaited until more subgenera related to the above two are examined.

In the previous papers, Su *et al.* (1996 a, b) noted that such three subgenera as *Coptolabrus*, *Damaster* (s. str.) and *Acoptolabrus* form a large cluster, namely, the *Damaster* (s. lat.) cluster, are associated with the *Procrustes–Megodontus* lineage as its sister group. The present results indicate that the *Neoplesius–Eocechenus* complex is more closely related to *Damaster* (s. lat.) than to *Procrustes–Megodontus*. This suggests that these Tibetan carabids may be the direct sister group of *Damaster* (s. lat.), though this relation is supported only by a low bootstrap value (45%), and is therefore unstable. Basic structure of the endophallus is strikingly uniform in all the species belonging to the *Neoplesius–Eocechenus* complex as mentioned above, and the same character is shared also with *Damaster* (s. lat.). To know more detailed phylogenetic relationships of the above mentioned groups, it is highly desirable to examine the DNA sequences of the following Chinese subgenera: *Pseudocranion* (Sichuan, Gansu, Qinghai and Shaanxi), *Shunichiocarabus* (Sichuan), *Pseudocoptolabrus* (Yunnan and Sichuan), *Aristocarabus* (Sichuan and Hubei), *Eccoptolabrus* (Sichuan, Gansu and Shaanxi), *Lasiocoptolabrus* (Shaanxi), *Coptolabrodes* (Shaanxi), *Pagocarabus* (sensu Imura, 1996) (North China), *Megodontoides* (sensu Imura, 1996) (Sichuan), *Imaibiodes* (Yunnan), *Calocarabus* (Sichuan, Qinghai and Gansu), *Cathaicus* (North China), *Acathaicus* (Gansu), *Eupachys* (Nei Menggŭ, Hebei and Shanxi), *Cychrostomus* (Qinghai and Gansu), *Cephalornis* (Gansu and Sichuan), and *Teratocarabus* (Heilongjiang and Liaoning).

Considering the phylogenetic topography and the distributional ranges of the Tibetan and other carabid beetles, we tentatively propose a possible sequence of event:— The common ancestry of the *Neoplesius–Eocechenus* complex and *Damaster* (s. lat.) had inhabited the mountainous regions of West China. The ancestral form of *Damaster* (s. lat.) branched off from that of the *Neoplesius–Eocechenus* complex some 24 Myr

1) Since the male of *C.* (*Eocechenus*) *leptoplesioides* is unknown, we show here the genital organ of *C.* (*E.*) *kaznakovi* Semenov-Tian-Shanskij et Znojko, the type species of the subgenus (Figs. 15, 16).

ago (see below), subsequently evolved to *Coptolabrus*, and then to *Acoptolabrus* along with the expansion of distribution to the eastern periphery of the Eurasian Continent. The proto-Japanese Archipelago was a part of the continent, where the proto-*Damaster* (s. str.) emerged presumably from *Acoptolabrus* (cf. SU *et al.*,1996 b).

Radiation of *C.* (*N.*) *markamensis*, *C.* (*N.*) *wagae*, *C.* (*N.*) *borodini*, *C.* (*N.*) *ludmilae* and *C.* (*E.*) *leptoplesioides* may be estimated to have occurred some 13 Myr ago, provided that the *Damaster* (s. str.) diversification started about 15 Myr ago (SU *et al.*, 1996 b).

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要　約

井村有希，蘇　智慧，大澤省三：チベット産オサムシ（チベットオサムシ亜属とタカネオオズオサムシ亜属）の形態と分子系統．—— 中国西蔵自治区東部から得られたチベットオサムシ亜属*Neoplesius*に属する4種6亜種とタカネオオズオサムシ亜属*Eocechenus*に属するマルカムタカネオオズオサムシ*C.* (*E.*) *leptoplesioides*について，ミトコンドリアND5遺伝子のDNA塩基配列を決定し，分子系統樹を作成して，日本とフランスに産するいくつかのオサムシとともに比較検討したところ，以下のような結論が得られた：1）これまでの形態種の概念からは1種とみなされるもののなかに，DNAでみると複数の系統が含まれており，逆に，形態上は亜種分化を示しているとみなしうる集団同志が，DNAの塩基配列においてほぼ同じである場合があって，形態と分子系統の間にかならずしも相関関係はみられなかった；2）形態学的にこれまで独立した亜属とみなされてきた*Neoplesius*と*Eocechenus*は，DNA解析を行うと同一のクラスターに納まり，分子系統上，これらを区分することはできなかった；3）この*Neoplesius–Eocechenus*群は，広義の*Damaster*としてまとめられるカブリモドキ，狭義のマイマイカブリ，クビナガオサムシの3亜属と祖先を共有する直接の姉妹関係にあることが示唆された．なお，本論文において，*C.* (*N.*) *wagae*の亜種として記載された*markamensis*を独立種に昇格し，同種の名のもとに*wangdanus*と*rawuensis*の2新亜種を記載した．

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A New Subspecies of *Carabus* (*Eucarabus*) *nitididorsus* from South Korea

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Carabus nitididorsus ISHIKAWA et KIM, 1983 has been known monotypical, distributed only on the Chiri-san Mountains near the southern end of the Korean Peninsula. Early in the summer of 1994, I made a short collecting trip to Mt. Paegun-san, about 20 km distant to the south from the centre of the Chiri-san Mountains, and succeeded in obtaining the same species. The Paegun-san population is definitely different from the nominotypical *nitididorsus* in detailed structure of the male genital organ, and I will describe it as a new subspecies.