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Aneumastus laosica sp. nov. and *A. genkalii* sp. nov. – two new diatom species from Laos (Southeast Asia) with comments on the biogeography of the genus

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Abstract – Two new diatom species of the genus *Aneumastus* are described, *A. laosica* sp. nov. and *A. genkalii* sp. nov., based on light and scanning electron microscope observations. These species were found in Laos, and this is first finding of this genus from Southeast Asia. Almost all species from this genus were described and exist in Holarctic. Many species have restricted distributions and are known from fossil deposits, North America, Mongolia or local endemics of ancient lakes such as Baikal, Prespa and Ohrid. Diversity of *Aneumastus* species from these ancient lakes is higher than from waterbodies of the Holarctic region taken together. A few reports of this genus are from the southern Hemisphere (e.g. New Caledonia, Australia, Lake Tanganyika in Africa), but these taxon records are represented by a single valve only from every region and they require additional taxonomical investigation and verification to understand the full extent of the distribution of the genus.

Diatoms / *Aneumastus* / new species / morphology / biogeography / Laos / Southeast Asia

INTRODUCTION

The genus *Aneumastus* D.G. Mann & Stickle was described in 1990 with the generitype proposed as *Aneumastus tusculus* (Ehrenberg) D.G. Mann & Stickle (= *Navicula tuscula* Ehrenberg). *Aneumastus* is a small freshwater genus, which possesses two fore and aft chloroplasts that are H-shaped in girdle view, open porous girdle bands and complex areolae occluded by cribra, but not hymenes (Round

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et al., 1990; Spaulding *et al.*, 2003, 2009; Cox, 2006; Kulikovskiy *et al.*, 2012; Maltzev *et al.*, 2017). It was suggested that the genus *Aneumastus* is closely related to *Mastogloia* Thwaites, with the latter genus bearing partectal chambers on the valvocopulae (Hajos, 1973; Round *et al.*, 1990). This was represented in the classification system of diatoms, where genus *Aneumastus* together with *Mastogloia* were included within the family Mastogloiaceae, in the order Mastogloiales (Round *et al.*, 1990). The phylogenetic position of the genus *Aneumastus* was determined using molecular approaches (Maltzev *et al.*, 2017). These authors confirmed the idea that *Aneumastus* is closely related to *Mastogloia* (Round *et al.*, 1990).

Three species were transferred to the genus *Aneumastus* in Round *et al.* (1990). These are *A. tusculus* (Ehrenberg) D.G. Mann & Stickle and *A. stroesei* (Østrup) D.G. Mann, *A. tusculoides* (Cleve-Euler) D.G. Mann. Lange-Bertalot (2001) prepared taxonomic revision of species belonging to this genus, providing some clarity of species synonymies and distinctions. After Round *et al.* (1990) and Lange-Bertalot (2001), other authors described new species of *Aneumastus* (Spaulding *et al.*, 2003; Levkov *et al.*, 2007; Kulikovskiy *et al.*, 2012b). The genus *Aneumastus* now includes more than 30 species (Fourtanier & Kociolek, 2011; Guiry & Guiry, 2017), that are known from oligotrophic to eutrophic water ecosystems with slightly acidic or mesotrophic conditions (Lange-Bertalot, 2001; Kulikovskiy, 2009).

Investigations of diatoms from Southeast Asia, including countries such as Myanmar, Thailand, Laos, Vietnam, Cambodia and Malaysia extend over 85 years but are generally poor (Meister, 1932; Patrick, 1936; Prowse, 1962; Shirota, 1966; Amossé, 1969; Voigt, 1969; Foged, 1971; Ohno *et al.*, 1972; Zyong Dyk Tien, 1982; Salleh & Rahim, 1995; Fujita & Ohtsuka, 2005; Duong *et al.*, 2006; Leelahakriengkrai *et al.*, 2009; Yana & Peerapornpisal, 2009; Suphan & Peerapornpisal, 2009, 2010; Inthasotti & Peerapornpisal, 2011; Leelahakriengrai & Peerapornpisal, 2011; Blanco *et al.*, 2012; Moonsin *et al.*, 2013; Yana *et al,* 2013; Yana & Mayama, 2015). Findings of species from the genus *Aneumastus* are absent in these publications. Our comprehensive investigation of diatoms from Laos, Vietnam, Cambodia and Indonesia allows us to find many rare endemic or new for science species (Gusev & Kulikovskiy, 2014; Glushchenko & Kulikovskiy, 2015, 2017a-c; Kociolek *et al.*, 2015; Kulikovskiy *et al.*, 2015a, 2017; Glushchenko *et al.*, 2016, 2017; Kapustin *et al.*, 2017). Our recent investigations reveal species of the genus *Aneumastus* that appear to be new to science.

The aim of this publication is to present LM and SEM observations of two new species of the genus *Aneumastus* from South East Asia (Laos) with comments in biogeography of the genus.

MATERIAL AND METHODS

Species of *Aneumastus* were found in two samples. The first sample was collected from Laos by E.L. Konstantinov on 06.12.2014, and designated N° 01612, from the Nam Hinboun River near Kong Lor Cave, Vientiane Province, periphyton (N17°53.783' E104°45.667'). Water temperature was about 20ºC and conductivity 111 μS/cm. The second sample was collected from Laos by A.M. Glushchenko on 23.11.2011, and designated N° 00996 from the Nam Lik River flowing into village Nam Lik, Vientiane Province, periphyton (N18°36.808' E102°24.605').

The samples were treated with 10% hydrochloric acid to remove carbonates and washed several times with deionized water for 12 h. Afterwards, samples were boiled in concentrated hydrogen peroxide (≈ 37%) to dissolve organic matter. They were washed again with deionized water four times at 12 h intervals. After decanting and

filling with deionized water up to 100 ml, the suspension was spread onto coverslips and left to dry at room temperature. Permanent diatom preparations were mounted in Naphrax®. Light microscopic (LM) observations were performed with a Zeiss Axio Scope A1 microscope equipped with an oil immersion objective (× 100, n.a. 1.4, differential interference contrast [DIC]) and Axio Cam ERc 5s camera (Zeiss). Valve ultrastructure was examined by means of a JSM-6510LV scanning electron microscope (IBIW, Institute for Biology of Inland Waters RAS, Borok, Russia). For scanning electron microscopy (SEM), parts of the suspensions were fixed on aluminum stubs after air-drying. The stubs were sputter coated with 50 nm of Au by means of a Eiko IB 3.

All samples and slides are deposited in the collection of Maxim Kulikovskiy at the Herbarium of the Institute for Biology of Inland Waters, Russian Academy of Science (IBIW), Borok, Russia.

RESULTS

Aneumastus laosica Glushchenko, Kulikovskiy & Kociolek sp. nov. Figs 1-16

Description. Valves linear-elliptic, ends protracted, abruptly short-rostrate. Length 35-50 μm, breadth 14-16 μm. Raphe weakly lateral with two slight undulations of the outer fissure of both raphe branches. Central raphe ending slightly enarged. Axial area narrow, almost linear and start to be narrower to distal ends. Central area a small, staurose, tapering towards each margin, irregularly rectangular by three or six shortened striae at each side. Striae radiate throughout, 13-14 in 10 μm.

SEM, external view (Figs 9-13): The raphe is undulate, the proximal raphe ends are slightly dilated while the distal raphe ends are hooked and extend onto the valve mantle. Striae composed of variously-shaped areolae, from X- or hourglass-shaped to elongate to rounded. Areolae lining the margin of the axial are straight (near the apices) to lunate. Striae appear to be biseriate at or near the margins, and uniseriate towards the axial area and onto the mantle. Within each areola on the valve face is a medially-positioned occlusion. Such occlusions are absent from the mantle. The occlusions appear to have rounded areas.

SEM, internal view (Figs 14-16): The valve has a distinct axial area, with narrow central stauros extending perpendicular to the axial area. Virgae are crossed, especially near the poles, and connected longitudinally, giving the impression of longitudinal lines. The central nodule is indistinct, and bears the straight proximal raphe ends. Striae openings are elongate near the margin, and become round to ellipsoidal towards the axial area. Within the striae are dome-like structures that are finely perforated. Distal raphe ends terminate as helictoglossae. At each pole there is a short pseudoseptum and septum.

Holotype. slide no. N° 01612 (Holotype here designated, illustrated in Fig. 2) in collection Maxim Kulikovskiy, I.D. Papanin Institute for Biology of Inland Waters, Russian Academy of Sciences (IBIW), 06.12.2014, leg. E.L. Konstantinov.

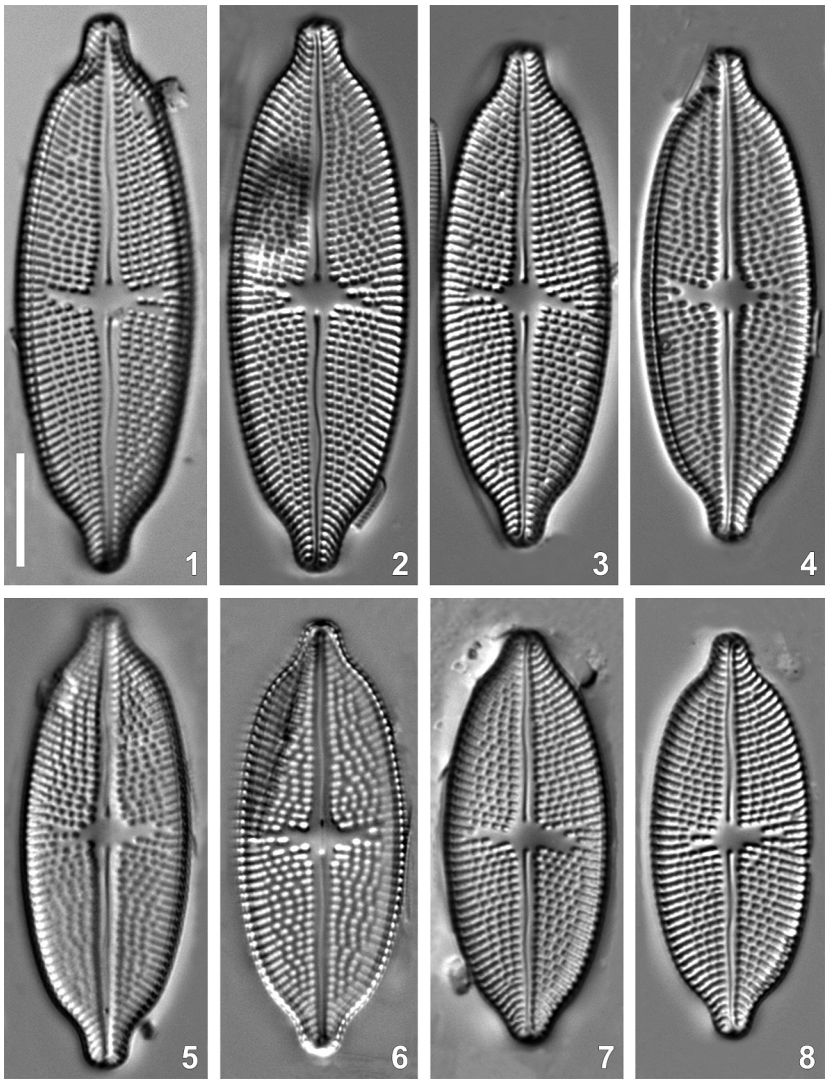
Type locality. Nam Hinboun River near Kong Lor Cave, Vientiane province, Laos, (N17°53.783' E104°45.667').

Etymology. The specific epithet "*laosica*" refers to the name of the country where this species was observed.

Distribution. As yet known only from the type locality.

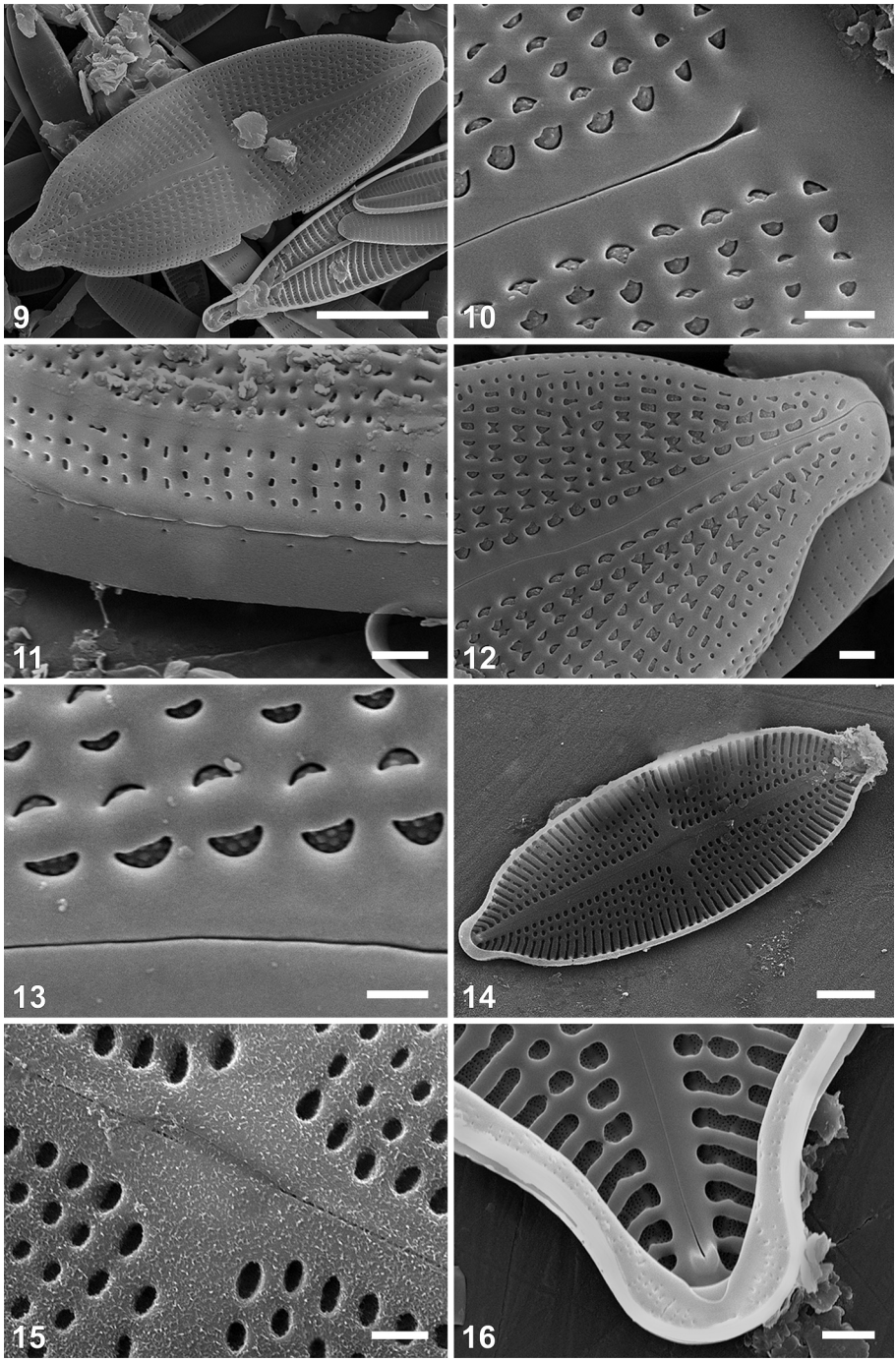
Aneumastus genkalii Glushchenko, Kulikovskiy & Kociolek sp. nov. Figs 17-33

Description. Valves linear-elliptic, ends protracted, abruptly short-rostrate. Length 36-48 μm, breadth 12-14 μm. Raphe almost filiform to weakly lateral with



Figs 1-8. *Aneumastus laosica* sp. nov., LM, DIC. Valves views. Size diminution series. Type population (slide No 01612). Figure 2 represents the holotype. Scale bar: 10 µm.

Figs 9-16. *Aneumastus laosica* sp. nov., SEM. Figs 9-13, external views. **9.** Whole valve showing shape, undulate raphe and distinct areolar pattern. **10.** View of the center of the valve face showing dilated proximal raphe end, areolae occlusions and longitudinal siliceous bands crossing virgae. **11.** Valve mantle showing uniseriate striae interrupted from those on the valve face. An irregular row of round poroids on the valvocopula. **12.** Valve face with variously shaped areolae. The undulate raphe is evident, and the distal end curves onto the mantle. **13.** High-resolution image of the areolae next to the raphe slit, with characteristic lunate shape with small, round structures on the medially-placed occlusions. Figs 14-16, internal views. **14.** Valve view. Striae are elongate at the margins, more rounded to ellipsoidal wards the central sternum. Pseudoseptum and helictoglossa are present at the apex. Longitudinal connections between virgae are also evident. The central nodule is indistinct. **15.** Central nodule with small, straight proximal ends evident. **16.** Apex, with fine perforations in the striae. A helictoglossa is evident. Pseudoseptum present. Scale bars: 5 µm (9, 14); 1 µm (Figs 10-12; 15, 16); 0.5 µm (Fig. 13). ▶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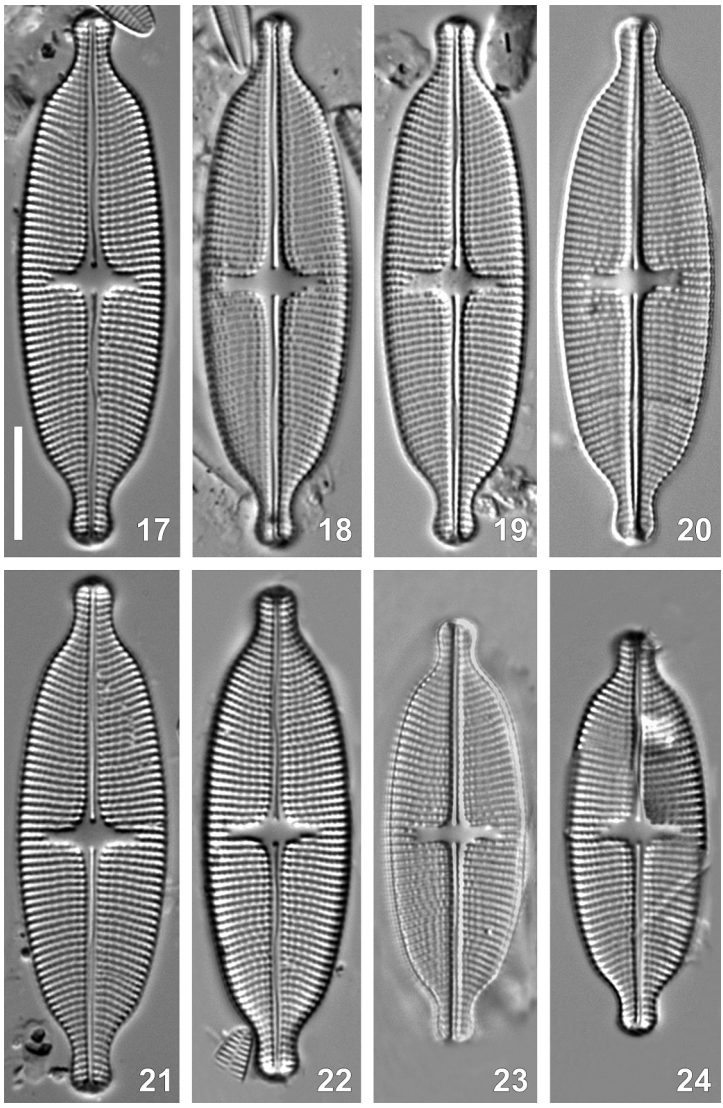
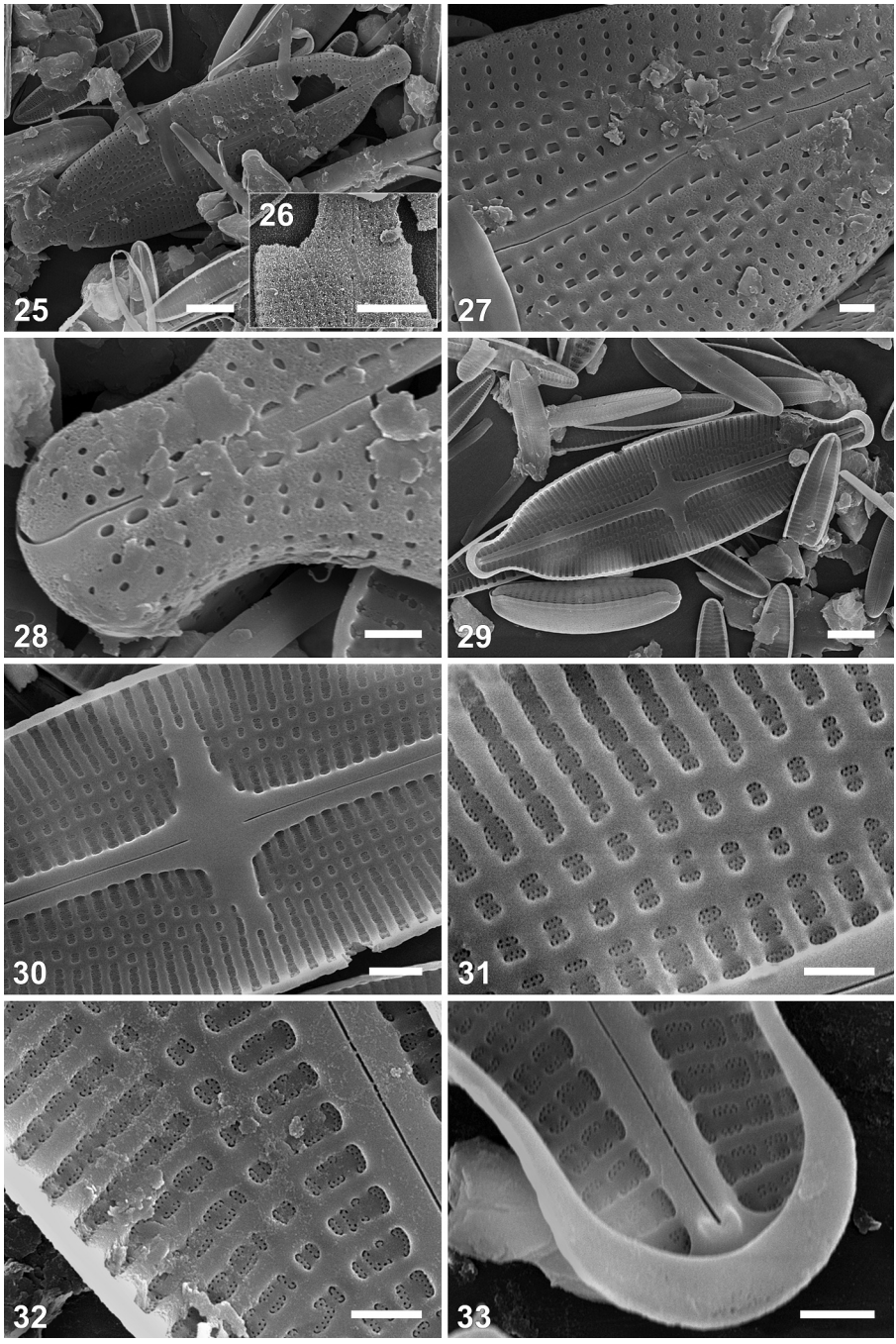


Figs 17-24. *Aneumastus genkalii* sp. nov., LM, DIC. Valves views. Size diminution series. Figure 17 represents the holotype. Fig. 23- Nam Lik River, Laos (slide No 00996), others – type population (slide No 01612). Scale bar: 10 µm.

Figs 25-33. *Aneumastus genkalii* sp. nov., SEM. Figs 25-28, external views. **25.** Valve view. Striae are uniseriate. Distal raphe end extends onto the mantle at the apex. **26.** Central area. Proximal raphe ends are slightly dilated. **27.** Valve face, undulate raphe, smaller areolae closer to the margin and onto the mantle, larger towards the axial area. Areolae have medially-placed occlusions. **28.** Valve apex. Figs 29-33, internal views. **29.** Valve view, with prominent central sternum, expanded, staurose-like central area, and pseudosepta and helictoglossae at the apices. **30.** Central area with fine, straight proximal raphe ends. **31, 32.** Views of the striae with finely perforate occlusions between virgae. Siliceous connections between the virgae running longitudinally. **33.** Valve apex, helictoglossae and pseudoseptum. The raised central sternum with small arch-like buttresses, formed between the virgae. Small perforations are evident. Scale bars: 5 µm (Figs 25, 26, 29); 2 µm (Fig 30); 1 µm (Figs 27, 28, 31-33).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two slight undulations of the outer fissure of both raphe branches. Central raphe ending slightly enarged. Axial area very narrow, weakly widens toward the valve center. Central area a narrow staurose, surrounded by two or three irregularly shortened striae. Striae radiate and almost parallel at the poles, 15-16 in 10 μm.

SEM, external view (Figs 25-28): Valves with striae that are composed of areolae, small and rounded on the mantle and near the margin, becoming larger near the middle of the valve. Areolae are narrowly ellipsoid to lunate along and parallel to the axial area, with sunken, medially-placed occlusions. Proximal raphe ends are small, straight, distal raphe ends are curved to the same direction onto the valve mantle, not hooked.

SEM, internal view (Figs 29-33): A thickened, raised central sternum, with thickened, staurose-like extensions in the center, dominate the valve interior. Proximal raphe ends are straight and small. Elongate striae are evident near the margin, become smaller, in pairs, then larger towards the central sternum. The striae have small openings at the base of the robust virgae. Arch-like structures are positioned along the base of the central sternum, and striae extend beneath them. Distal raphe ends terminate as helictoglossae. At each pole there is a short pseudoseptum.

Holotype. Slide no. N° 01612 (holotypus here designated; illustrated in Fig. 17) in collection of Maxim Kulikovskiy, I.D. Papanin Institute for Biology of Inland Waters (IBIW), Russian Academy of Sciences, 06.12.2014, leg. E.L. Konstantinov.

Type locality. Nam Hinboun River near Kong Lor Cave, Vientiane province, Laos (N17°53.783' E104°45.667').

Etymology. Species epithet honors the famous Russian diatomist Dr. Sergei Ivanovich Genkal from Institute for Biology of Inland Waters Russian Academy of Science, Borok, Yaroslavl Region.

Distribution. New species known from the type locality and from Nam Lik River flowing into village Nam Lik, Vientiane Province, periphyton, illustrated in Fig. 23.

DISCUSSION

Two new species of *Aneumastus* species described here share the same morphological features with other taxa that are typical for the genus. These features include an undulate raphe and combined areolae covered by cribrum and absence of hymenes, as well as a lack of partecta (Round *et al.*, 1990). It is possible to divide the previously-described species of the genus (including those presented here) into three morphological groups according to striae composition: 1) those species with singly punctate striae (e.g. *Aneumastus albanicus* Lange-Bertalot & Miho, *A. irishkin* Kulikovskiy, Lange-Bertalot & Metzeltin), 2) species with doubly punctate (*A. aksaraiensis* Spaulding, Akbulut & Kociolek and *A. juriljii* Levkov & Metzeltin) and, 3) species with singly punctate striae becoming doubly punctate near mantle (*A. balticus* Lange-Bertalot, *A. macedonicus* Levkov, *A. minor* (Hustedt) Lange-Bertalot, *A. rostratus* (Hustedt) Lange-Bertalot and *A. tusculus*). *Aneumastus laosica* sp. nov. belongs to the group of species with single punctate striae becoming doubly punctate near mantle. *Aneumastus genkalii* sp. nov. is characterized by having uniseriate striae throughout. These species are similar to each other having a stauros-like central area with irregularly rectangular, few shortened striae at each side.

However, the central area of *A. genkalii* sp. nov. is strongly elevated internally. Presence of a stauros forming the central area internally is a unique character of *A. genkalii* sp. nov.

Round *et al.* (1990) did not discuss the biogeography of *Aneumastus* in general or of the species. In addition, Lange-Bertalot (2001) did not give information about the distribution of the genus as a whole even though he included 13 species in his treatise of *Navicula* and genera separated from that large, unnatural group. And many other species were described after the original description in Round *et al.* (1990) and subsequent summary in Lange-Bertalot (2001). Up to now, the genus includes 35 species and infraspecific taxa, and they are reported especially from the Holarctic region (see Table). Reports of the genus from other areas is less well documented. For example, Moser *et al.* (1998) showed one valve of *Aneumastus stroesei* (Østrup) D.G. Mann from New Caledonia (Pl. 58: 3). However, this valve differs distinctly from type of *A. stroesei* (see Lange-Bertalot, 2001, Pl. 119: 1) by shape of valves being more elliptical and more narrower ends. Identification of this valve is erroneous in our opinion and it need further investigation. John (2012, Pl. 102: H) showed one valve of a similar *Aneumastus* taxon from Australia. John (2012) identified this species as *A. tusculus*, however this valve is morphologically more similar to the valve shown from New Caledonia. We are not able to exclude the possibility that two valves shown from New Caledonia and Australia belong to new, undescribed species that exists in the Australian region; additional observations are necessary before we can reach this conclusion. Cocquyt (1998, Pl. 24: 12) showed one valve of *Aneumastus tusculus* (as "*Navicula tuscula* Ehrenberg") from the Northern Basin of Lake Tanganyika. This valve is clearly morphologically similar to valves from the type material of *A. tusculus* (see Lange-Bertalot, 2001: Pl. 113: 1-4) by the more elliptical valve shape, central area being nearly absent, and the arrangement of the striae. Some findings of *Aneumastus tusculus* sensu lato and its infraspecific taxa were shown from Africa additionally (Foged, 1966, 1980, Starmach, 1975). So, these data suggest the diversity of *Aneumastus* species can be high in the South Hemisphere and that the some known species from this area are not identical with the common Holarctic species. To a great degree, species diversity of previously-known taxa are from the Northern Hemisphere and especially from the Holarctic (see Table).

Species such as *A. minor*, *A. rosettae*, *A. rostratus*, *A. stroesei*, *A. tusculus*, *A. apiculatus*, *A. maculosus, A. albanicus* are widespread in the Holarctic region (see Table). However, many species described and known from Holarctic individually have restricted distributions. *A. aratus* are known just from fossil deposits (Lange-Bertalot, 2001). *Aneumastus aksaraiensis* was described from Turkey, but it's position in *Aneumastus* may need clarification (Spaulding *et al.* 2003). *Aneumastus carolinianus* has a distribution restricted to North America and *A. mongolicus* is known from some waterbodies in Mongolia (see Table).

Many endemic species were described from ancient lakes Baikal, Ohrid and Prespa. Ten species were described from Lake Baikal and the Lake also includes previously-described species (Kulikovskiy *et al.*, 2012). In total, the species composition of *Aneumastus* taxa in Lake Baikal includes about 20 described and undescribed taxa. This number is larger than known diversity from the rest of the Holarctic, except for the species present in ancient lakes Ohrid and Prespa. Presently, nine species are known just from these lakes and the possibility exists they are local endemics (see Table). The large diversity of *Aneumastus* species in ancient lakes is a good example that genera that are not diverse in the Holarctic region in general may have a large diversity in ancient lakes. These may be explained by long history

Table. Species and infraspecific taxa of the genus *Aneumastus* and its distribution

Species	*Distribution*	*References*
A. aksaraiensis Spaulding, Akbulut & Kociolek 2003	Central Turkey: Esmekaya Lake	Spaulding *et al.*, 2003
A. albanicus Lange-Bertalot & Miho in Lange-Bertalot 2001	Albania: Lake Ohrid	Lange-Bertalot, 2001
	Russia: Lake Baikal	Kulikovskiy *et al.*, 2012
	Mongolia: Lake Hag	Dorofeyuk & Kulikovskiy, 2012
A. apiculatus (Østrup) Lange-Bertalot in Lange-Bertalot & Genkal 1999 **Basionym:** *Navicula lacustris* var. *apiculata* Østrup 1910 **Synonyms:** *Navicula pseudotuscula* auct. non Hustedt; ?*Navicula stroesei* var. *lanceolata* Haworth 1974; *Cavinula lacustris* var. *apiculata* (Østrup) Bukhtiyarova 1995	Russia: NW Siberia	Lange-Bertalot & Genkal, 1999; Lange-Bertalot, 2001
	Russia: Eastern Siberia	Genkal *et al.*, 2011
	Switzerland	Lange-Bertalot, 2001
	Alaska	Lange-Bertalot & Genkal, 1999
	Mongolia: Lake Khokh	Metzeltin *et al.*, 2009
	Mongolia: Lake Bayan	Dorofeyuk & Kulikovskiy, 2012
	Russia: Northern coast of the Okhotsk Sea	Kharitonov, 2010
	Russia: Kolyma region	Kharitonov, 2014
A. aratus (Grunow) Lange-Bertalot 2001 **Basionym:** *Navicula arata* Grunow 1882	Rumania, fossil	Lange-Bertalot, 2001
A. baicalensis (Jasnitsky) Kulikovskiy, Metzeltin & Lange-Bertalot 2012 **Basionym:** *Stauroneis* anceps var. *baicalensis* Jasnitsky 1936	Russia: Lake Baikal	Kulikovskiy *et al.*, 2012
A. balticus Lange-Bertalot 2001	Baltic Sea region, fossil	Lange-Bertalot, 2001
	Southern Siberia: Mongolia	Own unpublished data
	Germany: Lake Stechlin	Lange-Bertalot, 2001
A. beleg Metzeltin, Kulikovskiy & Lange-Bertalot 2012	Russia: Lake Baikal	Kulikovskiy *et al.*, 2012
A. carolinianus (Patrick) Lange-Bertalot 2001 **Basionym:** *Navicula caroliniana* Patrick 1959	North America: Great Lakes	Stoermer *et al.*, 1999; Lange-Bertalot, 2001
	North America: South Carolina	Patrick, 1959; Patrick & Reimer, 1966
A. gerelma Kulikovskiy, Lange-Bertalot & Metzeltin 2012	Russia: Lake Baikal	Kulikovskiy *et al.*, 2012
A. gracilis Kulikovskiy & Lange-Bertalot 2012	Russia: Lake Baikal	Kulikovskiy *et al.*, 2012
A. humboldtianus Lange-Bertalot & Miho in Lange-Bertalot 2001	Europe: Lake Ohrid	Lange-Bertalot, 2001; Levkov *et al.*, 2007
A. huruud Metzeltin, Kulikovskiy & Lange-Bertalot 2012	Russia: Lake Baikal	Kulikovskiy *et al.*, 2012
A. irishkin Kulikovskiy, Lange-Bertalot & Metzeltin 2012	Russia: Lake Baikal	Kulikovskiy *et al.*, 2012
A. juriljii Levkov in Levkov, Krstic, Metzeltin & Nakov 2007	Macedonia: Lake Ohrid	Levkov *et al.*, 2007
A. macedonicus Levkov in Levkov, Krstic, Metzeltin & Nakov 2007	Macedonia: Lake Prespa	Levkov *et al.*, 2007

A. maculosus (Donkin) Lange-Bertalot 2001 **Basionym:** *Navicula maculosa* Donkin 1871 **Synonym:** *Navicula albinensis* Grunow 1884	Nord Sea coast	Lange-Bertalot, 2001
	UK: coast of Island Jersey	Donkin, 1871
A. metzeltinii Levkov in Levkov, Krstic, Metzeltin & Nakov 2007	Macedonia: Lake Ohrid	Levkov *et al*., 2007
A. minor Lange-Bertalot 1993 **Basionym:** *Navicula tuscula* f. *minor* Hustedt 1930 **Synonyms:** *Navicula tuscula* var. *obtusa* Hustedt 1922; *Navicula tuscula* var. *minor* Hustedt 1930	Europe: Balkan Peninsula	Hustedt, 1945
	North America: Great Lakes	Stoermer *et al*., 1999
	Iceland	Foged, 1974
	Sweden: Öland	Foged, 1980b
	Alaska	Foged, 1981
	Sweden: Northern Bothnian Bay	Snoeijs & Potapova, 1995
	Northern Germany	Lange-Bertalot, 2001
	Russia: Northern coast of the Okhotsk Sea	Kharitonov, 2010
	Russia: Kolyma region	Kharitonov, 2014
	Russia: Far East	Medvedeva & Nikulina, 2014
	Kazakhstan: Eastern Balkhash	Own unpublished data
	North Africa: Egypt	Foged, 1980a
	Mongolia: Lake Har and Lake Bayan	Dorofeyuk & Kulikovskiy, 2012
A. minor var. *densistriatus* Levkov & Krstic in Levkov, Krstic, Metzeltin & Nakov 2007	Macedonia: Lake Ohrid	Levkov *et al*., 2007
A. mongolicus Metzeltin, Lange-Bertalot & Nergui 2009	Mongolia: Ynegtin River	Metzeltin *et al*., 2009
A. munge Metzeltin, Kulikovskiy & Lange-Bertalot 2012	Russia: Lake Baikal	Kulikovskiy *et al*., 2012
A. odon Kulikovskiy, Metzeltin & Lange-Bertalot 2012	Russia: Lake Baikal	Kulikovskiy *et al*., 2012
A. ohridanus Levkov & Metzeltin in Levkov, Krstic, Metzeltin & Nakov 2007	Macedonia: Lake Ohrid	Levkov *et al*., 2007
A. perodon Kulikovskiy, Metzeltin, Lange-Bertalot 2012	Russia: Lake Baikal	Kulikovskiy *et al*., 2012
A. pseudoapiculatus Levkov & Metzeltin in Levkov, Krstic, Metzeltin & Nakov 2007	Macedonia: Lake Prespa	Levkov *et al*., 2007
A. rhombicus Levkov & Metzeltin Levkov, Krstic, Metzeltin & Nakov 2007	Macedonia: Lake Ohrid	Levkov *et al*., 2007
A. rosettae Lange-Bertalot & Miho in Lange-Bertalot 2001	Albania: Lake Ohrid; Southern Europe: some lakes	Lange-Bertalot, 2001
	Russia: Lake Baikal	Kulikovskiy *et al*., 2012
A. rostratus (Hustedt) Lange-Bertalot 2001 **Basionym:** *Navicula tuscula* var. *rostrata* Hustedt 1911 **Synonyms:** *Navicula tuscula* f. *rostrata* (Hustedt) Hustedt 1930; *Aneumastus tusculus* f. *rostrata* (Hustedt) Bukhtiyarova 1995; *Aneumastus tuscula* var. *rostratus* (Hustedt) Andresen, Stoermer & Kreis 2000	North America: Great Lakes	Stoermer *et al*., 1999
	Macedonia: Kozhuf Mountain	Levkov *et al*., 2007
	China: fossil	Li *et al*., 2005
	Belarus: Svisloch River	Genkal *et al*., 2013
	Mongolia: Lake Har Us & Lake Buyr	Dorofeyuk & Kulikovskiy, 2012
	Russia: NE Europe	Loseva *et al*., 2004
	India: Katta	Sarode & Kamat, 1984
	Sweden: Öland	Foged, 1980b

Species	*Distribution*	*References*
A. sharbin Kulikovskiy, Metzeltin & Lange-Bertalot 2012	Russia: Lake Baikal	Kulikovskiy *et al*., 2012
A. stroesei (Østrup) Mann in Round, Crawford & Mann 1990 **Basionym:** *Navicula tuscula* var. *stroesei* Østrup 1910 **Synonym:** *Navicula pseudotuscula* Hustedt 1943	Europe: Swiss Alps, Davos	Hustedt, 1943
	Europe: Lake Ohrid	Petrovska & Stojanov, 2007
	France: Rhône-Alpes	Bey & Ector, 2013
	Belarus: River Svisloch	Genkal *et al*., 2013
	Russia: Northern coast of the Okhotsk Sea	Kharitonov, 2010
	Russia: NW Siberia	Lange-Bertalot & Genkal, 1999
	Iceland	Foged, 1974
	New Caledonia	Moser *et al*., 1995
A. subapiculatus Levkov & Nakov in Levkov, Krstic, Metzeltin & Nakov 2007	Macedonia: Lake Ohrid	Levkov *et al*., 2007
A. tusculus (Ehrenberg) Mann & Stickle in Round, Crawford & Mann 1990 **Basionym:** *Navicula tuscula* Ehrenberg 1840	Italy: fossil	Lange-Bertalot, 2001
	Japan: Lake Akan	Kawashima & Mayama, 1997
	Mongolia	Metzeltin *et al*., 2009
	Eastern Canada	Lavoie *et al*., 2008
	Estonia: Moonsund; Germany: Greifwalder Bodden; Russia: island of Malyi, Gulf of Finland	Snoeijs & Kasperoviciene, 1996
	South Korea: Kwang River	Lee *et al*., 1994
	Russia: Northern coast of the Okhotsk Sea	Kharitonov, 2010
	Russia: Kolyma	Kharitonov, 2014
	Russia: NE Europe	Loseva *et al*., 2004
	Russia: Eastern Siberia	Genkal *et al*., 2011
	Russia: Sakha Republic	Komarenko & Vasil'eva, 1975
	Russia: River Yenisei	Levadnaja, 1986
	Russia: Euro-Arctic region, fossil and recent	Kagan, 2012
	Russia: Far East	Medvedeva & Nikulina, 2014
	Russia: Lake Glubokoe	Chudaev & Gololobova, 2016
	Ukraine	Bukhtiyarova, 1999
	India: Bombay	Gonzalves & Gandhi, 1954
	India: Alibag, Thane	Sarode & Kamat, 1984
	West Africa: Ghana	Foged, 1966
	West Africa: West Sudan	Starmach, 1975
	Central Africa: Lake Tanganyika	Cocquyt, 1998
A. tusculus f. *angulatus* (Hustedt) Lange-Bertalot in Lange-Bertalot & Genkal 1999 **Basionym:** *Navicula tuscula* f. *angulata* Hustedt 1942	Sweden: Swedish Lapland	Hustedt, 1942
	Russia: NW Siberia	Lange-Bertalot & Genkal, 1999

of these ecosystems and presence of species flocks (Pavlov *et al.*, 2013; Kulikovskiy *et al.*, 2015e; Kociolek *et al.*, 2017). Such species from Lake Baikal as *A. odon*, *A. perodon*, *A. sharbin* and *A. munge* share the same morphological features in shape, areolae, striae structure and raphe, suggesting they are monophyletic and can be described as species flock from Lake Baikal. The species flock phenomenon according to diatoms is comprehensively discussed in publication of Kociolek *et al.* 2017. Diversity of different genera, like the genus *Aneumastus*, in ancient lakes is a reason of high diatom species diversity as compared to those in surrounding areas (Levkov *et al.*, 2007; Metzeltin *et al.*, 2008; Kulikovskiy *et al.*, 2011, 2012a,b,c, 2013, 2014a,b, 2015b–f, 2016a,b; Kociolek *et al.*, 2013; Kulikovskiy & Kociolek, 2014; Kulikovskiy & Kuznetsova, 2014, 2016; Vishnyakov *et al.*, 2014, 2015). Alternatively, it is possible that the ancient lakes mentioned above have served as refugia for more widely-distributed species now extinct elsewhere as these ancient ecosystems.

Aneumastus minor var. *densistriatus* was described by Levkov & Krstic from Lake Prespa. This infraspecific taxon clearly differs from the nominate variety of *A. minor* by the smaller and circular central area, more elliptical valves, denser striation, as well as smaller areolae in stria. Below we effect the transfer this taxon to species level.

Our data show that many species of *Aneumastus* have restricted distributions and are known mainly from the Holarctic region. *Aneumastus laosica* sp. nov. and *A. genkalii* sp. nov. represent the first discovery and formal description of new species from Tropics. Our comprehensive investigation of diatoms from Southeast Asia show that this genus is almost unknown for this region and other tropic areas and Southern Hemisphere.

New taxonomic combination:

Aneumastus densistriatus (Levkov & Krstic) Kulikovskiy, Glushchenko & Kociolek stat. nov.

Basionym: *Aneumastus minor* var. *densistriatus* Levkov & Krstic 2007. Iconographia Diatomologica. V. 16. Diatoms of Lakes Prespa and Ohrid. About 500 taxa from ancient lake system. P. 26-27. Pl. 80: 1-16, fig. 2 represent the holotype.

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