Diversity of the genus *Genkalia* (Bacillariophyta) in boreal and mountain lakes – taxonomy, distribution and ecology

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Abstract: *Genkalia digitulus* (Hustedt) Lange–Bert. et Kulikovskiy (≡ *Navicula digitulus* Hustedt) is recorded from several lakes in the northern Europe and from high altitude lakes all over Europe. Wide distribution and characteristic autecology of this species allow its use as a bioindicator of pH changes and reliable marker of environmental reconstruction. A comparison of specimens found in lakes from the Carpathian and Balkan Mountains with the type material of *Genkalia digitulus* (Hustedt) Lange–Bert. et Kulikovskiy resulted in description of a new species – *G. boreoalpina* Wojtal, C.E. Wetzel, Ector, Ognjanova–Rumenova et Buczkó. *Genkalia boreoalpina* is characterised by valve outline, size and parallel striae throughout most of the valve. The separation was based on light and scanning electron microscopy. *Genkalia boreoalpina* was the most common diatom of the genus from high mountain lakes of slightly acidic waters with a very low mineral content. The third species, *Navicula subprocera* Hustedt, was found in the Lake Câlcescu (Parâng Mountains) in alkaline waters and in a mountain fen in Herzegovina. The nomenclatorial combination to *Genkalia* was made for this species. Previously published data suggest there is a much larger diversity of species in the genus *Genkalia* and the cosmopolitan distribution of this group is mostly presented at the genus level.

Key words: Balkans, Carpathians, morphological variability, *Navicula*, oligotrophy, pH change, type material

Introduction

The genus *Genkalia* Kulikovskiy, Lange–Bert. et Metzeltin was recently described from the lake Baikal (Kulikovskiy et al. 2012). It presently comprises eleven species, most of them of unknown distribution and observed only at their type localities. This genus is characterized by a naviculoid outline and shares several characteristics with the genera *Adlafia* Lange–Bert. and *Boreozonacola* Lange–Bert., Kulikovskiy et Witkowski. Among the taxa actually placed in the genus, *Navicula digitulus* Hustedt (1943: 162) [≡ *Genkalia digitulus* (Hustedt) Lange–Bert. et Kulikovskiy in Kulikovskiy et al. (2012: 142)] is a common taxon in high latitude and high altitude lakes of the Holarctic region (Sabater & Roca 1992; Bigler & Hall 2002, 2003; Catalan et al. 2009; Krstić et al. 2012). According to the literature, *G. digitulus* (as *Navicula digitulus*) is a common species, abundant in lakes of the northern Europe (Bigler & Hall 2002, 2003; Larsen et al. 2006) though it is also known from several mountain lakes (Kawecka & Galas 2003; Štefková 2006; Buczkó et al. 2009, 2013a; Ognjanova–Rumenova et al. 2009, 2011; Gąsiorowski & Sienkiewicz 2010). This diatom is widespread in boreoalpine climate and especially abundant in cold, oligotrophic and weakly acidic waters, poor in electrolytes (Bigler et al. 2000; Fallu et al. 2000; Rosén et al. 2000; Bigler & Hall 2002; Larsen et al. 2006). Despite importance and commonness of *G. digitulus* in dilute waters problems with its identification have been found (e.g. Marciniak & Cieśla 1983; Fallu et al. 2000). Moreover, the European Diatom Database (EDDI) (Juggins 2001) which encompasses a large dataset for *G. digitulus* (as *Navicula digitulus*) has only one vouchered (illustrated) specimen that differs from *N. digitulus* sensu Hustedt 1943.

Genkalia digitulus was described as *Navicula digitulus* by Hustedt (1943: 162) from a high mountain lake – Schwarzsee near Davos in Switzerland. The species was found in only one sample; though another locality (spring near Benneckenstein in Harz, Germany) was also mentioned by Hustedt (1943). The species diagnosis is supported by five drawings showing finely striated valves of linear–lanceolate outline with protracted, rounded apices and round central area of different size. A similar species – *Navicula subprocera* Hustedt (1945: 920, plate 41, fig. 1) – found in the Buna spring (Bosnia and Herzegovina) was established two years later. The original drawing of *N. subprocera* supports the description showing lanceolate valve with bluntly rounded ends and a large, rhombic central area (Hustedt 1945, plate 41, fig. 1). In 1986 Krammer & Lange Bertalot extended the morphological range of *Navicula digitulus* (1986: 204, plate 77, figs 19–24 & "evtl. 25–28") including *N. subprocera* (plate 77, fig. 25) and even the specimens with linear valves having 28–40 striae in 10 µm. Then the detailed study of diatom flora of oligotrophic Lake Julma Ölkky in Finland resulted in description of *Naviculadicta digituloides* Lange–Bert. in Lange–Bertalot & Metzeltin (1996: 83, plate 109, figs 8–9, plate 115, fig. 4). Among characteristics which distinguish *Naviculadicta digituloides* from *Navicula digitulus* there were stated – more linear than lanceolate outline, obtusely rounded ends, large and rhombic central area. According to Lange–Bertalot (in Lange–Bertalot & Metzeltin 1996) both species can co–occur in oligo–dystrophic waters. They were included to the genus *Genkalia* Kulikovskiy, Lange–Bert. et Metzeltin (Kulikovskiy et al. 2012: 134) along with nine newly described species from Lake Baikal.

The aim of this study was to reexamine the morphology of *Navicula digitulus* from the Hustedt type material and make comparisons with *Genkalia* specimens from Carpathian and Balkan localities from eastern and southern Europe.

Materials and Methods

The following material was examined:

1. Type material of *Navicula digitulus* Hustedt 1943: material number BRM E1319 bottom of Schwarzsee, Davos 126, Switzerland, 24th July 1930, located over timberline (2388 m a.s.l.).
2. Type material of *Navicula subprocera* Hustedt 1945 (page 920): material number BRM E4191, from a puddle at the source of the river Buna at Blagaj, about 12 km from Mostar (Herzegovina), collected on 2nd May 1909.
3. Mountain fen in Bijambare, Dinaric Mts near Sarajevo (Bosnia and Herzegovina) collected in May and October 2007 and May 2008 from the marginal part of the fen.
4. Nine lakes, situated in three different cirques in Rila Mountains (Table 1). Sediment cores from the deepest part of the nine lakes were retrieved in July and August 2000; the samples were considered older than c. 1800 AD. Epilithon from the Lake Bubreka was additionally collected in 26th September 2013.
5. Lake Avrig (Făgăraş Mountains) on 7th August 2012, Lake Calcescu (Parâng Mountains) on 14th August 2012, Lake Caprelor (Retezat Mountains) in July 2013, all collected by Csilla Kövér and core samples from Lake Lia, Lake Brazi and Lake Gales (Retezat Mountains), drilled in 2007 and 2008, collected by Mihály Braun and Enikő Magyari.
6. Near–shore sediments from Lake Zadni Staw Gąsienicowy. The recent samples were collected on 23rd September 2005 by Jolanta Piątek.

All studied habitats were characterized by slightly acidic to circumneutral water of very low water conductivity (12–36 $\mu S.cm^{-1}$). The only exception in terms of ion concentration was the fen in Bijambare protected area (Kapetanović et al. 2011). Higher pH values were recorded only in the Lake Okoto, Lake Avrig and Lake Calcescu (Table 1). Schwarzsee near Davos (Switzerland) is a small lake in the western Swiss Alps. Spring Buna (Bosnia and Herzegovina) is a cold–water, karstic spring with a large discharge, south–east from Mostar. The mountain fen in the Bijambare protected area (Bosnia and Herzegovina) is of postglacial origin and the climate of this region is moderately continental, with strong impacts of mountain climate. All the lakes of Rila Mountains (Bulgaria) are located above the timberline and are of glacial origin. Glacial lakes in Retezat Mountains (Southern Carpathians) were formed mainly during the Late Glacial (Buczkó et al. 2009). The Lake Zadni Staw Gąsienicowy remains frozen for most of the year.

The samples were digested using concentrated H_2O_2 and heating. Air–dried material was mounted in Naphrax®. The diatoms were identified with a Nikon Eclipse 80i light microscope with phase contrast (DIC), Leica DM LB2 with 100 HCX PLAN APO objective, Leica® DMRX bright–field microscope with 100× oil immersion objective. Light micrographs were taken with a Nikon DS–Fi, VSI–3. OM(H)a Leica® and DC500 cameras, respectively. The cleaned samples were sputter–coated with gold–palladium or palladium and a Hitachi S–26000N scanning electron microscope (Figs 36, 93–96, 116, 117) and an ultra–high–resolution analytical field emission (FE) scanning electron microscope Hitachi SU–70 (Hitachi High–Technologies, Europe, GmbH) (Figs 37–48, 97–109) were used for the analysis. Micrographs were digitally manipulated and plates containing light and scanning electron microscopy images were created using CorelDraw X5®.

Table 1. List of localities with geographical and chemical characteristics.

Locality	**Altitude (m a.s.l.)**	**pH**	**Conductivity (µS.cm^{-1})**	**Source**
Lake Schwarzsee (Alps, Switzerland)	2388	nd	nd	Hustedt 1943
Buna spring (Bosnia and Herzegovina)	nd	nd	nd	Hustedt 1945
Mountain fen (Dinaric Alps, Bosnia and Herzegovina)	930	6.6	125	Kapetanović et al. 2011
Lake Ledeno (Rila Mountains, Bulgaria)	2709	5.9	12	Ognjanova-Rumenova et al. 2009
Lake Okoto (Rila Mountains, Bulgaria)	2440	7.2	26	Ognjanova-Rumenova et al. 2009
Lake Bubreka (Rila Mountains, Bulgaria)	2282	6.6	26	Ognjanova-Rumenova et al. 2009 and unpublished data
Lake Bliznaka (Rila Mountains, Bulgaria)	2243	6.8	32	Ognjanova-Rumenova et al. 2009
Lake Alekovo (Rila Mountains, Bulgaria)	2545	6.0	13	Ognjanova-Rumenova et al. 2009
Lake Karakashevo (Rila Mountains, Bulgaria)	2391	6.2	17	Ognjanova-Rumenova et al. 2009
Lake Gorno Marichino (Rila Mountains, Bulgaria)	2378	6.5	20	Ognjanova-Rumenova et al. 2009
Lake Dolno Marichino (Rila Mountains, Bulgaria)	2368	6.6	22	Ognjanova-Rumenova et al. 2009
Lake Sulzata (Rila Mountains, Bulgaria)	2535	7.0	35	Ognjanova-Rumenova et al. 2009
Lake Caprelor (Retezat Mountains, Romania)	2135	6.9	21	unpublished data
Lake Lia (Retezat Mountains, Romania)	1910	6.4–6.7	13	Buczkó et al. 2013b
Lake Brazi (Retezat Mountains, Romania)	1740	6.2–6.7	14–17	Buczkó et al. 2013b
Lake Calcescu (Parâng Mountains, Romania)	1934	8.1	10	unpublished data
Lake Avrig (Făgăras Mountains, Romania)	2007	7.7	36	unpublished data
Lake Zadni Staw Gąsienicowy (Tatra Mountains, Poland)	1852	nd	13	unpublished data

Results

Three species of the genus *Genkalia* were found in lake habitats of the Carpathian and Balkan mountains. The most common was here described as *Genkalia boreoalpina* sp. nov. The highest abundance of *G. digitulus* was observed in weakly acidic lakes. The third species, *G. subprocera* (= *Navicula subprocera*) was found in the Lake Calcescu and in a mountain fen in the Bijambare protected area, where sparse individuals co–occurred with less abundant and smaller specimens of *G. digitulus*.

Genkalia digitulus **(Hustedt) Lange–Bert. et Kulikovskiy in Kulikovskiy et al. (2012) [(Figs 1–35, LM; 36–48 (SEM)]**

Basionym: *Navicula digitulus* Hustedt (1943: 162, figs 26–30)

≡ *Naviculadicta digitulus* (Hustedt) Lange–Bert. in Lange–Bertalot & Metzeltin 1996: 84, figs 109: 30–33; 115: 3.

Type: Switzerland, Schwarzsee; sample E1319, 24th July 1930.

Description of type material (Figs 2–14, 37–48): Valves are lanceolate with gradually tapered and rounded ends or are protracted into short subrostrate apices. They are 8.0–20.0 µm long (N = 22) and 3.0–5.0 µm wide (N = 22). Axial area is narrow, linear. Central area is variable in size, from very small up to moderate corresponding with Hustedt drawings (here Fig. 1a–e). Striae are uniseriate, slightly radiate in the mid–valve (27–34 in 10 µm) becoming parallel towards the apices (32–40 in 10 µm). Areolae are discernible in LM. Length and width ratio varies between 2.0 and 4.5.

In SEM terminal fissures are externally unilaterally curved to the secondary side. Proximal raphe terminals are not expanded and very slightly bent to the primary side. Raphe is straight, or weakly sigmoid, lying in the middle of axial area. Striae are composed of round areolae, running continuously over valve face across the margin and down onto the mantle. A Voigt discordance is present. Internally raphe branches are straight with abruptly bent to the primary side proximal terminals. Distal terminals are in small, simple helictoglossa. Central nodule is elevated; striae are composed of transapically elongated or round areolae.

Description of material from lakes Lia, Gales, Calcescu, Avril, Caprelor and Brazi (Figs 15–36): Valves are lanceolate or linear–lanceolate with short protracted apices. They are 9.0–17.0 µm long (N = 32) and 3.5–5.5 µm wide (N = 29). Axial area is narrow and linear. The central area is small. Striae are slightly radiate in the mid–valve becoming parallel towards apices. Areolae are discernible in LM. Foramina are closed by hymenes on the outside and can be observed in SEM when valves are not corroded (Fig. 36).

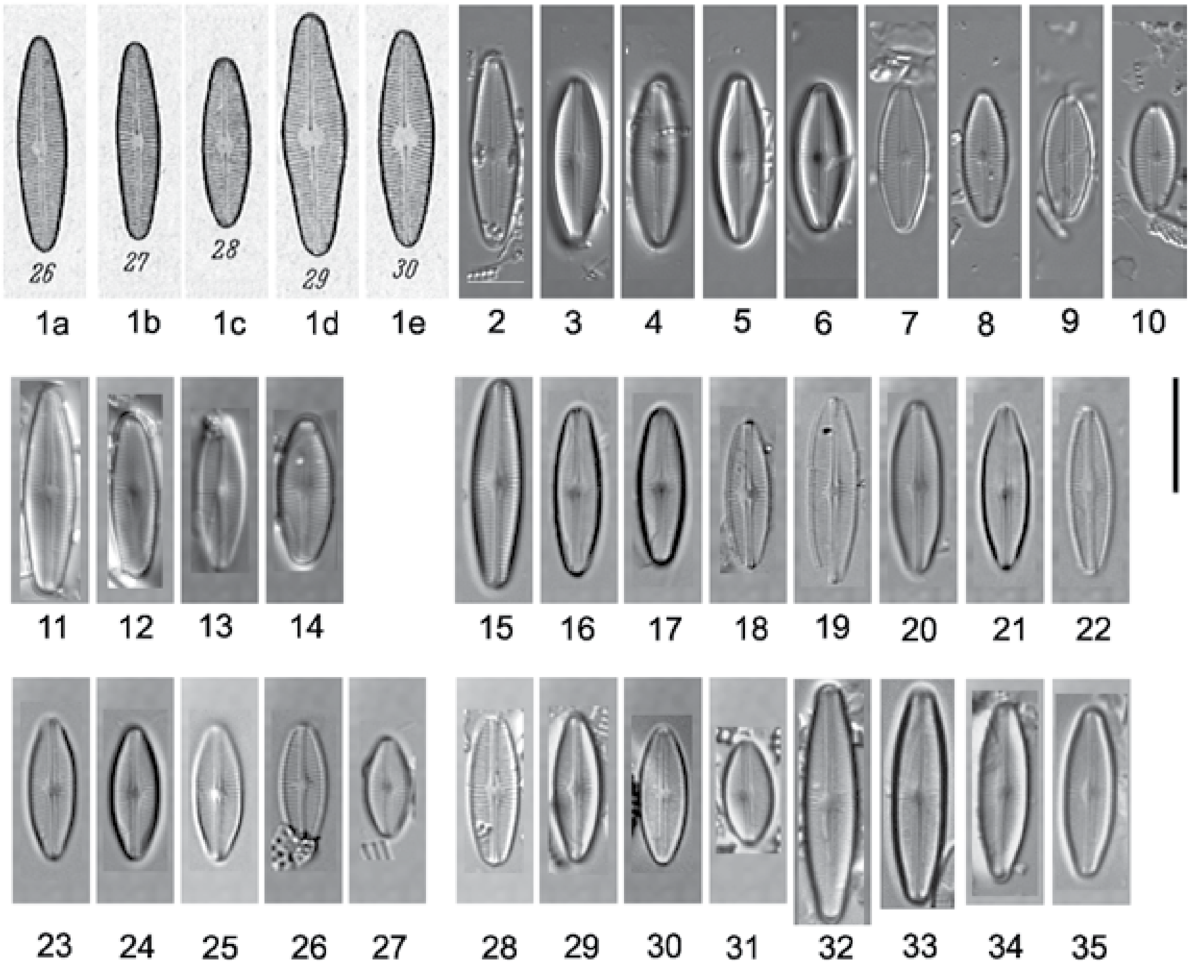
Ecology and Distribution: *Genkalia digitulus* was found in recent and fossil samples from Dinaric Mountains and Carpathians. The majority of habitats were of very low mineral content and slightly acidic water (Table 1). The only exceptions were: margins of the lakes Calcescu and Avril, sediment samples of Lake Okoto (the deepest lake), Lake Sulzata (the only lake without inflow) and the mountain fen in Bijambare area (Table 1). Wide distribution and conspicuous morphology suggest a high indicative value of this diatom, especially for pH changes reconstruction and ionic concentration.

Remarks: Analysis of the original description and type material suggests that the Hustedt' concept of *Genkalia digitulus* (as *Navicula digitulus*) was based on morphologically heterogeneous assemblage of diatoms. The original Hustedt' description (1943) is supported by five drawings showing finely striated valves of linear–lanceolate outline with rounded apices, round central area of different size and narrow, linear axial area (Fig. 1a–e). The description of *Navicula digitulus* repeated by Hustedt in 1961–1966 is illustrated by six drawings of which five specimens have large and round central area and rounded ends, although in the diagnosis (Hustedt 1943) valves with broadly rounded or sometimes very weakly protracted ends and small central area are stated. The specimens selected by Simonsen (1987) originate from the slide originally labeled by Hustedt, but they are not the same as those used by Hustedt for line–drawings (the references to iconotypes are lacking). Four lanceolate valves/frustules of *Navicula digitulus* (Simonsen 1987, plate 470, figs 4–9) cover the morphological range indicated by Hustedt (1943). First specimen (Simonsen 1987, p. 311, plate 470, figs 4, 5) has bluntly rounded ends and a relatively large central area. Three other specimens have a small central area and short protracted ends.

The specimens with short protracted apices and small central area represent a morphological range in terms of length and width (Figs 3–10, 13, 14, 18–24, 29, 30, 34, 35). The cells with gradually tapered valves co–occurred with them in the type material of *Navicula digitulus* and in populations from lakes in the Retezat Mountains (Figs 2, 11, 12, 15–17, 25, 26, 28–33). Comparison of both morphologies in SEM has not shown distinct differences between these valve forms. Only one valve, the smallest specimen found in the type material has an elliptic outline with a very small central area and very weak radial striae in the middle portion of valve. Areolae on the valve face are smaller and transapically elongated along the axial area (Fig. 41). The specimens with and without short protracted ends were most abundant in the Retezat Mountains. Among them some specimens found in the Lake Lia show almost parallel valve margins in the middle portion of valve (Figs 19–21). Each stria on the mantle around the

Table 2. Morphometric characteristics of *Genkalia digitulus* and related taxa.

	Genkalia digitulus			***Genkalia subprocera***		***Genkalia digituloides***	***Genkalia boreoalpina***	
Data source	Hustedt (1943)	Type population (present study)	Other localities	Hustedt (1943)	Other localities	Lange-Bertalot in Lange-Bertalot & Metzeltin (1996)	Type population	Other localities
valve length (µm)	14.0–22.0	8.0–20.0	9.0–17.0	22.0	13.0–22.0	16.0–27.0	10.0–20.0	12.0–20.0
valve width (µm)	3.5–5.5	3.0–5.0	3.5–5.5	5.0	4.0–5.5	4.5–5.5	3.0–4.5	3.5–4.5
valve outline	lanceolate	lanceolate	linear or linear-lanceolate	lanceolate	lanceolate	linear or linear-lanceolate	linear with or without a weak central constriction	linear with or without a weak central constriction, the smallest - elliptical
valve apices	gradually tapered and rounded ends or protracted into short subrostrate apices	gradually tapered and rounded ends or protracted into short subrostrate apices	gradually tapered and rounded ends or protracted into short subrostrate apices	gradually tapered, rounded ends	gradually tapered, rounded ends	bluntly rounded	bluntly rounded	bluntly rounded
number of striae in the middle of valve (in 10 µm)		27–34	25–32	28	25–28	26	29–36	29–36
number of striae near valve ends (in 10 µm)		32–40	26–35	29–30	26–30	28	29–36	29–36
striation pattern in the middle of valve	radiate	radiate	radiate	radiate	radiate	radiate	near parallel or weakly radiate	near parallel or weakly radiate
striation pattern near valve ends	parallel	parallel	parallel	parallel	parallel	near parallel or weakly radiate	parallel	parallel
distribution	Alps	Alps	Mountain lakes	Herzegowina	Herzegowina Retezat	Finland	Balkan	Balkan, Carpathinas



Figs 1–35. *Genkalia digitulus* (HUSTEDT) LANGE–BERT. et KULIKOVSKIY: (1a–1e) original drawings of Navicula digitulus after HUSTEDT 1943; (2–14) *Genkalia digitulus* from the type material – Schwarzsee, sample E1319, LM; (15–35) *G. digitulus* from other localities: Carpathians, Balkans, LM. Scale bar 10 µm.

apex is composed of only one elongated areola.

Genkalia boreoalpina **WOJTAL, C.E. WETZEL, ECTOR, OGNJANOVA–RUMENOVA et BUCZKÓ sp. nov. (Figs 49–109)**

Description: Valves are linear with or without a weak central constriction, 10–20 µm long (N=66) and 3.0–4.5 µm wide (N=56), with bluntly rounded apices. The smallest valves have linear–elliptic outline. Axial area is narrow, linear. Central area is distinct and round or rhombic. Raphe branches are filiform and straight. Striae are uniseriate, composed of fine but distinct areolae, parallel throughout most of the valve only about the central area – parallel or slightly radiate.

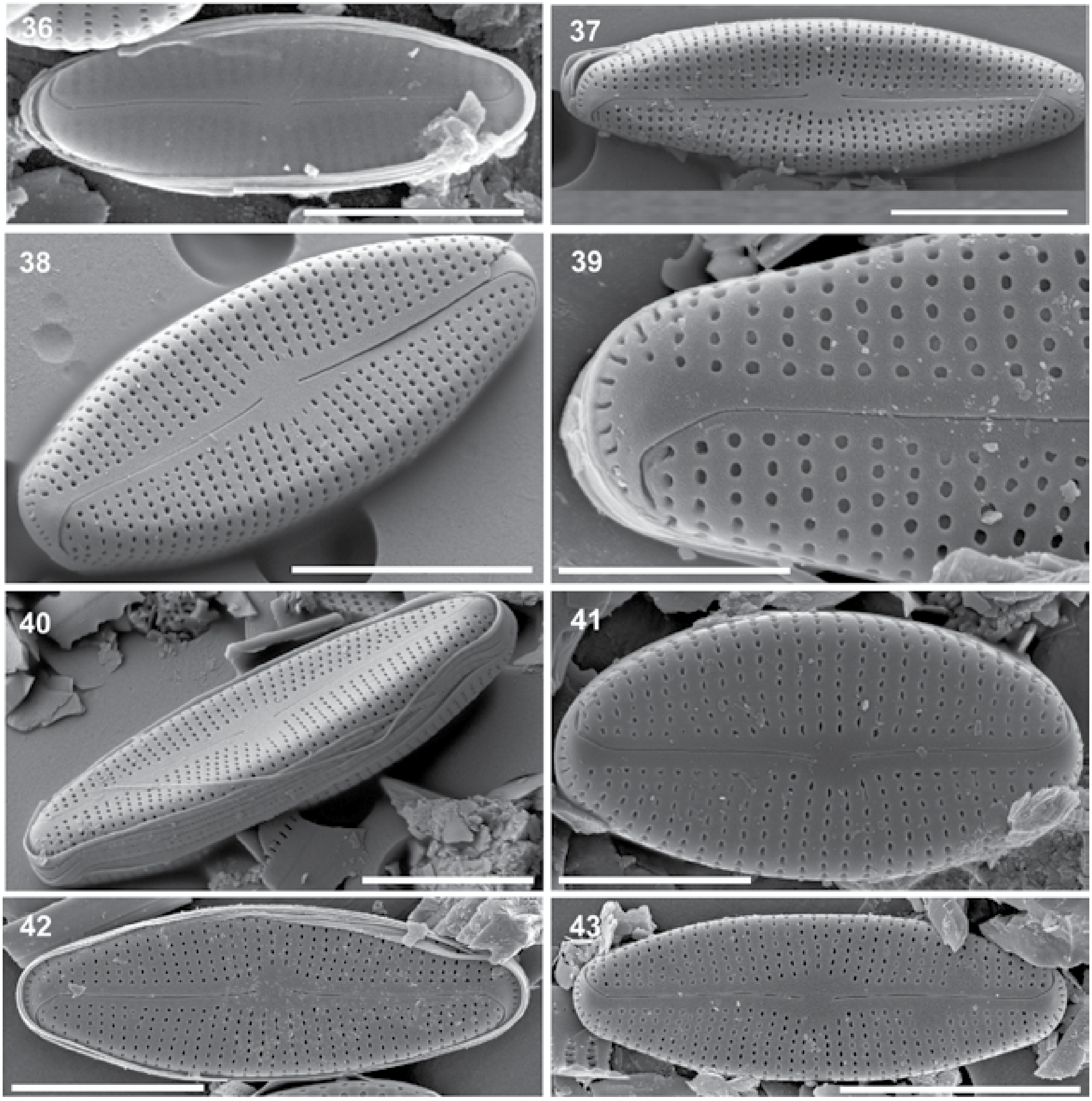
SEM: Externally the valve face is flat. The raphe is slightly sigmoid and lateral, lying closer to secondary side. Distal raphe terminals are turned to the secondary side. Proximal raphe ends are simple, straight or slightly bent to the primary side. Axial area is narrow and linear. The shape of central area is usually rounded, though it varies according to the number of short striae near the central area. Sometimes, one short stria is unilaterally intercalated. Striae are parallel, only near the central area they can be slightly radiate and are more distant from each other than in the other part of the valve. A Voigt discordance is present (Figs 93, 94, 96). Striae are uniseriate, 29–36 in 10 µm. Areolae are round or transapically elongated, especially on the valve mantle. The striae run continuously over the valve surface, across the margin and down onto the valve mantle. At the base of the mantle at each apex each stria is composed of two areolae. The narrow girdle of each theca consists of copulae. Internally proximal raphe ends are abruptly bent towards primary side. Distal raphe terminals are in simple, small helictoglossa. Areolae are round or transapically elongated, occluded. Central nodule is slightly elevated.

Type locality: Switzerland, Schwarzsee

Holotype (designated here)**:** Fig. 49, Schwarzsee Friedrich Hustedt Collection Alfred Wegener Institut, Bremerhaven, Germany, access number – ZU9/74.

Isotype: Institute of Nature Conservation, Kraków, Poland, access number – DW 14.

Etymology: The species epithet is related to the

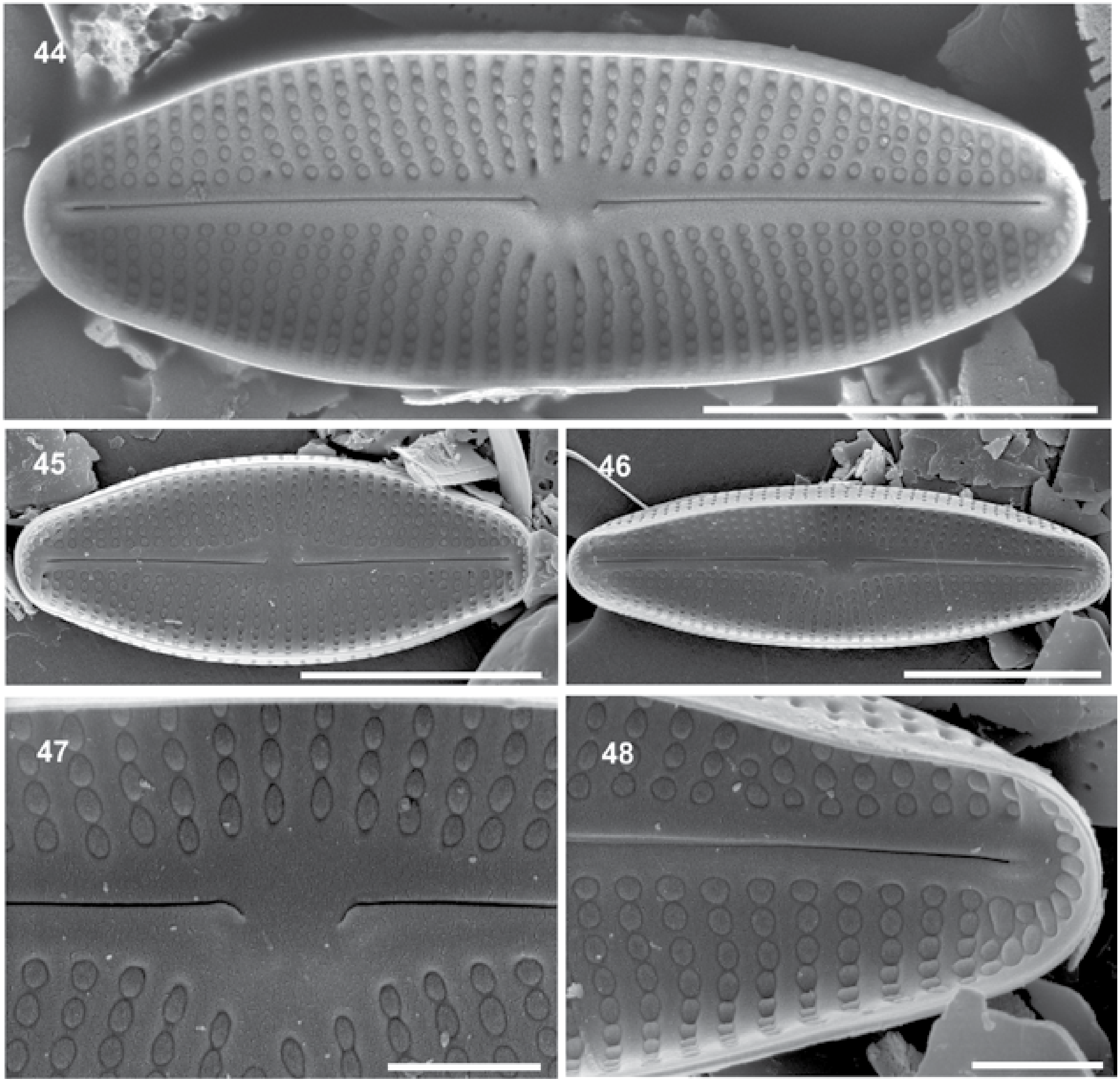


Figs 36–43. *Genkalia digitulus* (Hustedt) Lange–Bert. et Kulikovskiy: (36) from Carpathians, external view of not corroded valve; (37–43) *G. digitulus* from the type material – Schwarzsee, sample E1319. External view of valves, SEM. Scale bars 5 μm (36–38, 40, 42, 43), 2 μm (39), 3 μm (41).

distribution of this taxon in the northern hemisphere and mountain areas.

Ecology and Distribution: *Genkalia boreoalpina* was found in several recent and fossil samples from lakes in Rila Mountains (Balkans), Făgăras Mountains, Retezat Mountains (Southern Carpathians), Tatra Mountains (Western Carpathians) and in the type material of *Navicula digitulus* from the Alps. A stable suite including weakly acidic to alkaline water of very low mineral content, wide distribution (in the Holarctic region), and conspicuous valve morphology suggest that this taxon has a high indicative value for environmental assessments.

Remarks: *Genkalia boreoalpina* is similar to *G. digitulus* in size, raphe morphology and presence of uniseriate striae. However, it differs in outline, morphologies of transapical striae, ornamentation of the mantle around apices, the position of central raphe fissure, and central area size. The most similar species *G. digituloides* (Lange–Bert.) Lange–Bert. et Kulikovskiy in Kulikovskiy et al. (2012) is described from Julma Ölkky Lake in Finland (Lange–Bertalot & Metzeltin 1996). *Genkalia digituloides* has curving striae in the middle part of the valve whereas the transapical striae of *G. boreoalpina* are straight. The larger size, distinct radial striae through a large part of the valve suggest the separation of *G. boreoalpina*



Figs 44–48. *Genkalia digitulus* from the type material – Schwarzsee, sample E1319. Internal view of valve, SEM. Scale bars 5 µm (44–46), 1 µm (47, 48).

from *G. digituloides*, though their conspecifity cannot be definitely excluded. *Navicula lange–bertalotii* E. REICHARDT (1985: 176) [= *Fallacia lange–bertalotii* (E. REICHARDT) E. REICHARDT] is another diatom similar in LM. *Navicula lange–bertalotii* and *Genkalia boreoalpina* are similar in outline, dimensions, round central area, striae organization but differ in genus specific features, e.g. *F. lange–bertalotii* has multi–seriate striae. Some specimens found in the Rila lakes (Figs 91, 92) are slender and have a linear–lanceolate outline (13.0–15.0 µm long and 3.5–3.6 µm wide).

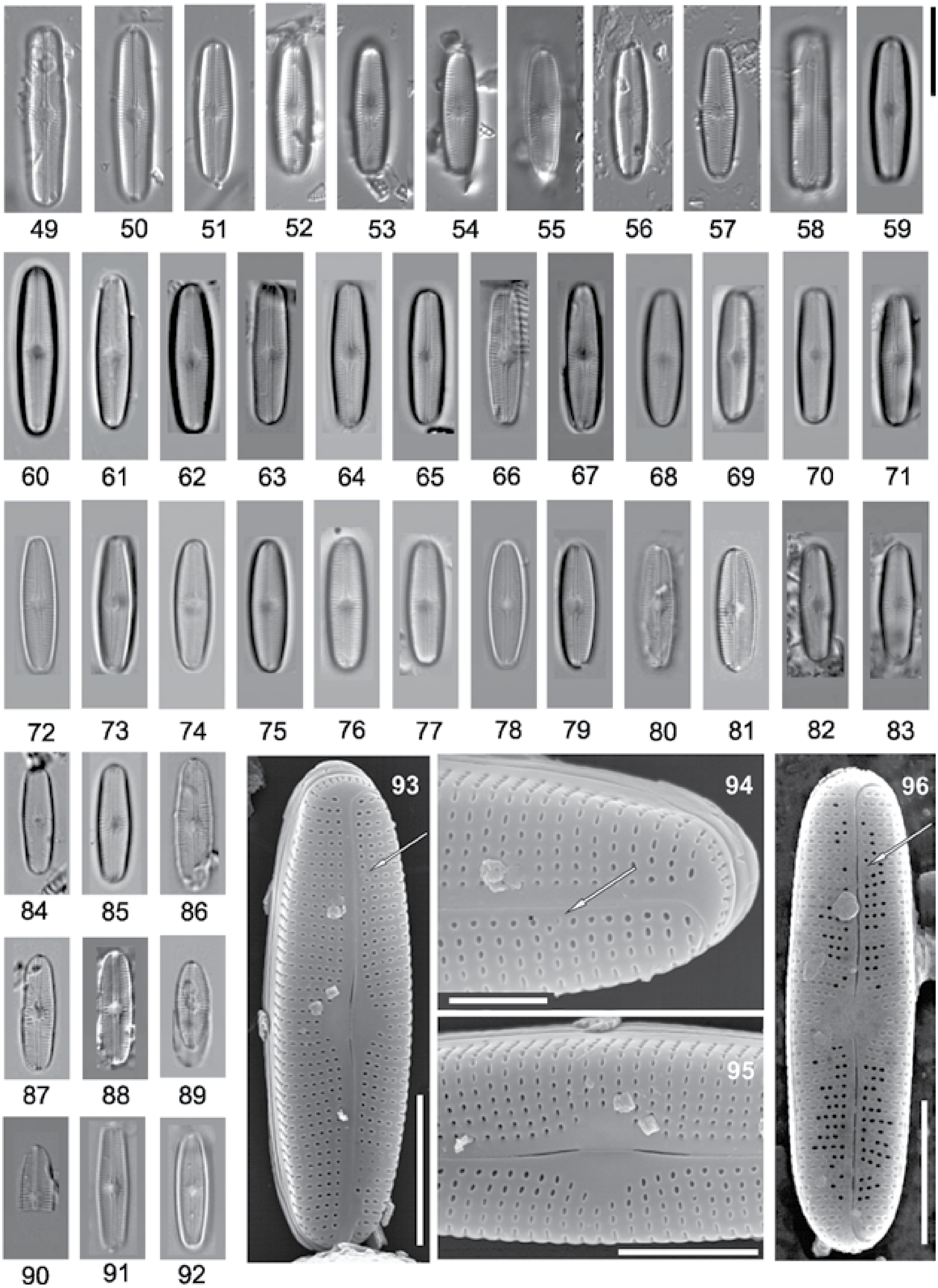
After examining previously published illustrations, *Genkalia boreoalpina* appears to have a broad northern hemisphere distribution. Beginning from the type material of *Navicula digitulus* (KRAMMER & LANGE–BERTALOT 1986, fig. 77: 20 – "Typenpräp. Coll. HUSTEDT N2/69" and from other, unknown locality (KRAMMER & LANGE–BERTALOT 1986, fig. 77: 23), Canada (FALLU et al. 2000, plate 14, fig. 19 as *Navicula digitulus* and as *Navicula* sp. 37 Quebec), Siberia (GENKAL & KHARITONOV 2010, plate 1, figs 1–5, 7, 8 as *Naviculadicta digituloides*) and a small alpine lake (GÜTTINGER 1999).

***Genkalia subprocera* (HUSTEDT) WOJTAL, ECTOR, C.E. WETZEL, OGNJANOVA–RUMENOVA et BUCZKÓ comb. nov. (Figs 110–117)**

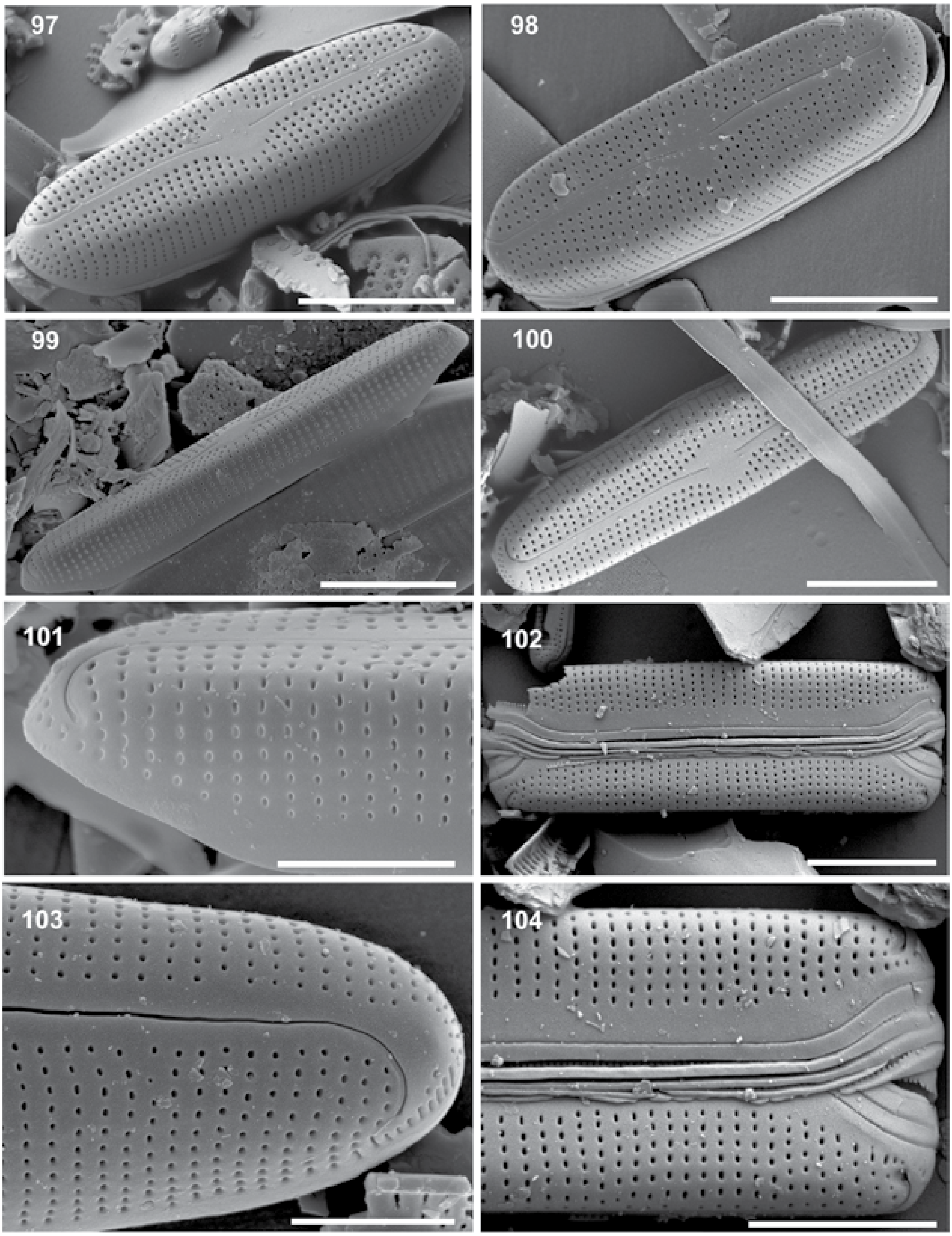
Basionym: *Navicula subprocera* HUSTEDT (Archiv für Hydrobiologie 1945: 920, plate XLI, fig. 1).

Type: Herzegovina, Buna spring; sample E 4191, 2nd May 1909; holotype slide N17/47.

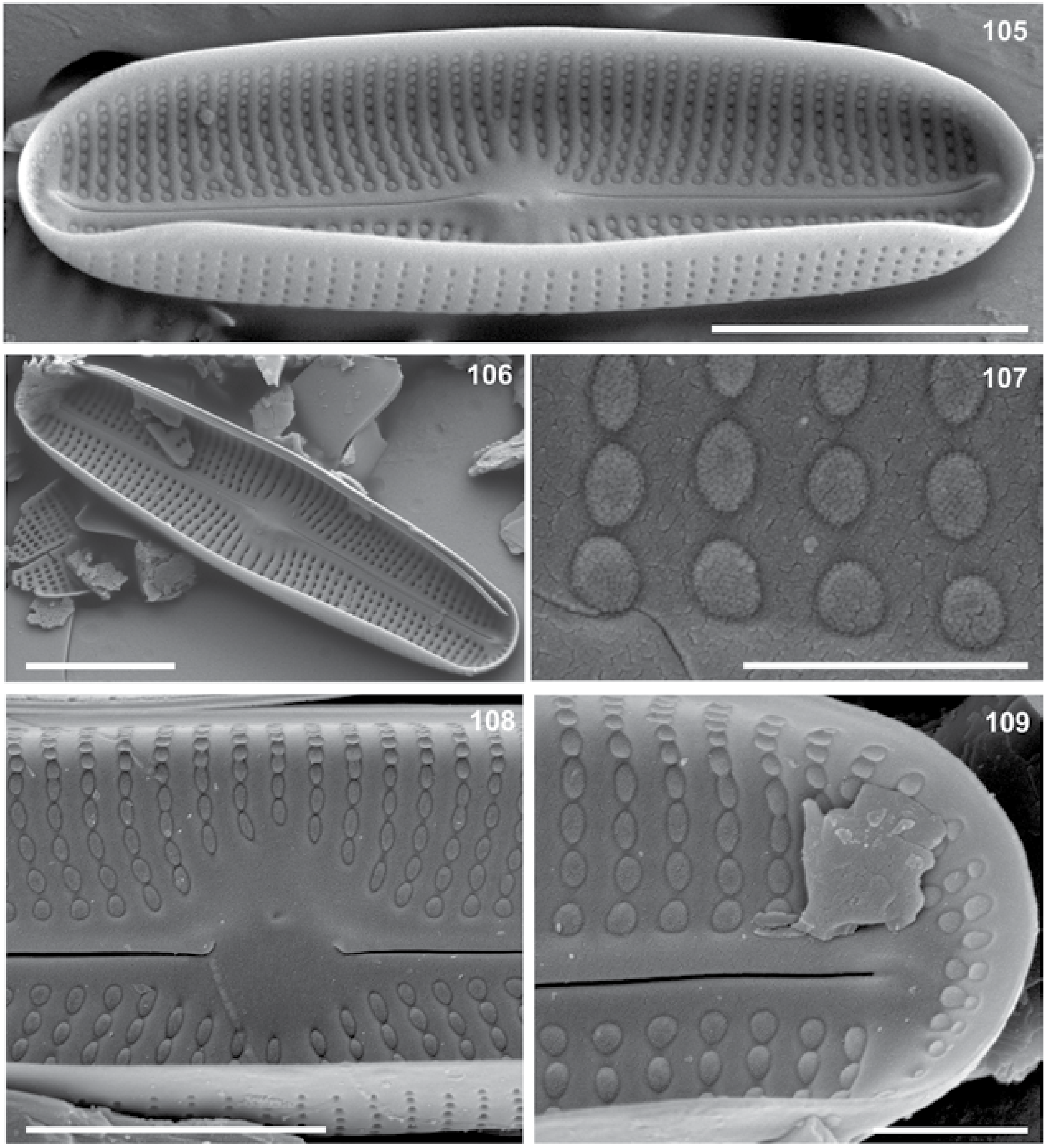
The description of *Genkalia subprocera* is based on the HUSTEDT drawing (1945, fig. 1): the valve is lanceolate



Figs 49–96. *Genkalia boreoalpina* Wojtal, C.E. Wetzel, Ector, Ognjanova–Rumenova et Buczkó sp. nov.: (49–58) type material of *G. boreoalpina* – Schwarzsee, sample E1319, (51) holotype of *G. boreoalpina*; (59–96) *G. boreoalpina* from Rila Mountains lakes, Retezat Mountains and Tatra Mountains, LM; external view of valve from Ledeno lake (93–95) and lateglacial part of Lake Brazi (96), Voigt discordance arrowed, SEM. Scale bars 10 µm (49–92); 5 µm (93, 96), 2 µm (94), 3 µm (95).



Figs 97–104. *Genkalia boreoalpina* WOJTAL, C.E. WETZEL, ECTOR, OGNJANOVA–RUMENOVA et BUCZKÓ sp. nov., external view of valve, type material, Schwarzsee, SEM. Scale bars 5 µm (97–100, 102), 2 µm (101, 103), 4 µm (104).

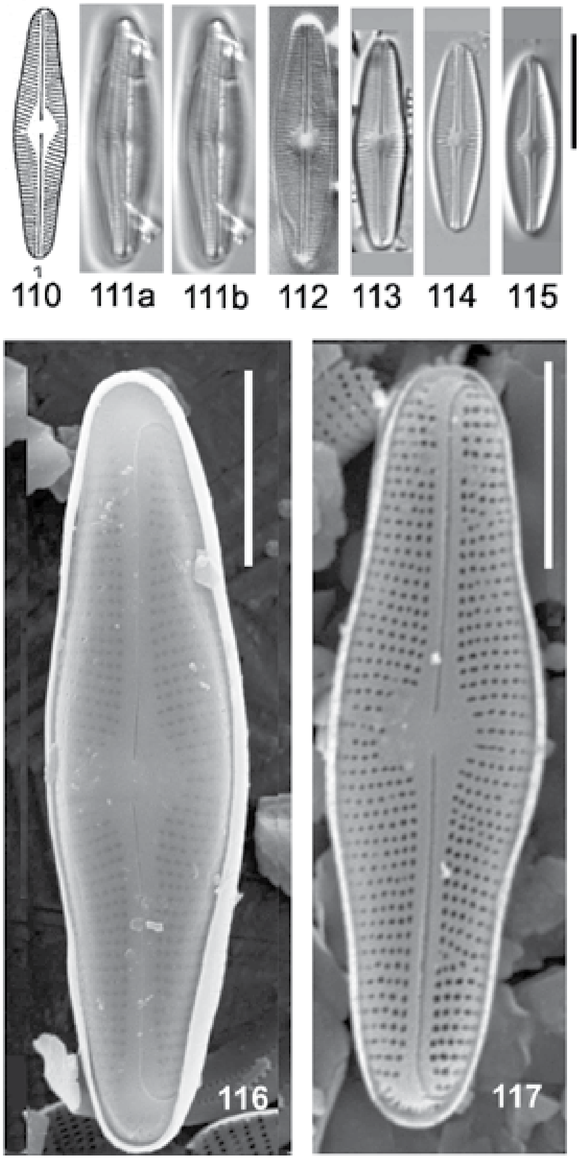


Figs 105–109. *Genkalia boreoalpina* Wojtal, C.E. Wetzel, Ector, Ognjanova–Rumenova et Buczkó sp. nov., internal view of valve, type material, Schwarzsee, SEM. Scale bars 5 µm (105,106), 1 µm (107, 109), 3 µm (108).

with gradually tapering, rounded ends; 22 µm long and 5 µm wide. Axial area is linear; central area is large and rhombic, sometimes it is asymmetric but does not reach the valve margins. Striae are uniseriate, radial in the middle position of the valve becoming near–parallel towards the apices, 28–30 in 10 µm. Striae are more distant from each other around the central area.

Description of material from Lake Câlcescu and Bijambare fen (Figs 112–117): Valves are lanceolate with gradually tapered, rounded apices, 13.0–22.0 µm long (N=8) and 4.0–5.5 µm wide (N=8). Raphe is filiform but distinct with small central pores. Axial area is linear and narrow. Central area is large, rhombic, and sometimes asymmetric but does not reach the valve margin. Striae are radial in the middle of the valve becoming parallel towards apices, 25–30 in 10 µm. In the middle portion of the valve the striae are more distant from each other than in the other.

Remarks: The original diagnosis was illustrated only by one specimen. In the diagnosis Hustedt stated that *Genkalia subprocera* (as *Navicula subprocera*) was very rare in the type material and probably only



Figs 110–117. *Genkalia subprocera* (HUSTEDT) WOJTAL, ECTOR, C.E. WETZEL, OGNJANOVA–RUMENOVA et BUCZKÓ comb. nov.: (110) original drawing of *Navicula subprocera* after HUSTEDT 1945; (111a–b) frustule at two different focuses, holotype slide, Hustedt Collection, Bremerhaven; (112–115) *Genkalia subprocera* from fen in the Bijambare area and Lake Câlescu; (116, 117) *G. subprocera* from Lake Calescu, SEM external view. Scale bars 10 µm (110–115), 5 µm (116, 117).

one specimen was known from the type material. The dimensions provided by HUSTEDT in 1945 overlap with the length and width attributed to *G. digitulus* by him (as *Navicula digitulus*) (Table 2). Probably the same specimen was documented by SIMONSEN (1987, plate 508, figs 28, 29) and then showed by KRAMMER & LANGE–BERTALOT (1986, plate 77, fig. 25). No other specimen was found in the type material to our knowledge and the main morphological characters are known from HUSTEDT's drawing, as the only specimen lies in oblique position (Fig. 111a, b). The uncertainty around the identity of *G. subprocera* (as *N. subprocera*) was enhanced later in HUSTEDT (1961–1966: 252, fig. 1378), where a description of *N. digitulus* was illustrated by six drawings of which five show specimens having lanceolate, gradually tapered valves with rounded apices and a large, round central area.

Genkalia subprocera co–occurred with *G. digitulus* in Bijambare fen area. The range of morphological variability of *G. digitulus* partly overlaps valve morphology of *G. subprocera*. However, they can be distinguished by outline of central area and ornamentation of the mantle near apices. *Genkalia subprocera* has a large and asymmetric central area with irregularly shortened striae – alternating long and short. Moreover, *G. digitulus* has on the mantle striae composed of one areola per stria, whereas the striae of *G. subprocera* are composed of at least two striae.

DISCUSSION

Genkalia digitulus differs from most genus members by sometimes having short protracted apices and near parallel sides in the mid–valve portion, whereas, the rest possesses valves with obtusely or broadly rounded apices.

The near parallel sides observed in some populations (Figs 19–21) are at first sight similar to *Adlafia* species, but the valves differ in organization of the striae (*Genkalia* – radial becoming to parallel; *Adlafia* – radial becoming abruptly to divergent towards apices). *Adlafia* taxa have a narrow girdle, each theca consists of two copulae with a biseriate row of areolae (LANGE–BERTALOT 2001). Comparing habitats of *Genkalia* and *Adlafia* species, the pH and ionic tolerance ranges of *Genkalia* are narrower. Moreover, a large part of the known records of *Genkalia digitulus* (as *Navicula digitulus*) are from lakes, whereas species of the genus *Adlafia* are usually aerophilous diatoms, which can be washed into a lake from surrounding area.

The similarities in morphology between *Genkalia* taxa and *Boreozonacola* LANGE–BERT., KULIKOVSKIY et WITKOWSKI in KULIKOVSKIY et al. (2010) taxa are also quite remarkable and should be taken into consideration.

According to KULIKOVSKIY et al. (2012), these genera differ in the position of hymenes "lying in a middle between external and internal apertures of areola" or not. In fact very little information on the position of the hymen in *Boreozonacola* is available. Images of the external and internal valve of *Navicula(dicta) pseudosilicula* HUSTEDT are only provided by LANGE–BERTALOT & GENKAL (1999, pl. 23, figs 1, 2) and solely an external view was published by KULIKOVSKIY et al. (2010).

The observed increase in abundance of *Genkalia digitulus* during periods of large inorganic, allochtonous inputs into lakes (LOTTER & HÖLZER

1994; Bigler & Hall 2003) raises a question about the preference of this species for unstable habitat conditions.

Genkalia subprocera was described in 1945 from calcium–rich water in Buna spring. However, the description was based probably on one specimen (Hustedt 1945). Unfortunately within the time the only one frustule in the slide designated by Hustedt has probably changed its location being at present in oblique position. But it cannot be excluded that the original Hustedt drawing (1945, plate XLI, fig. 1) was his interpretation of a specimen lying in oblique position. This species was then neglected in the later elaborations (e.g. Hustedt 1961–1966; Krammer & Lange–Bertalot 1986). It remains uncertain if Hustedt omitted *Navicula subprocera* due to its rarity or range of the recognized morphological variability or any other reason but the species was not synonimized with *N. digitulus* by Hustedt (1961–1966) nor Krammer & Lange–Bertalot (1986).

Buna spring water is calcium rich and of high mineral content. The localities where *Genkalia subprocera* was found characterize water pH from 6.2 to 6.8 in mountain fen (Kapetanović et al. 2011) and 8.1 in Lake Câlcescu. *Genkalia digitulus* and *G. subprocera* have similar valves dimensions, partly overlapping, but they differ in size and shape of central area and striae organization in the middle valve portion (Figs 2–35, 112–115) and on the mantle near apices.

The most common species in this study from high mountain lakes seems to be *G. boreoalpina*. In the Schwarzsee type material of *G. digitulus* and in the sample from Retezat Mountains typical specimens of *G. digitulus* co–occurred with sparse specimens of *G. boreoalpina*. The valves morphology of *G. digitulus* clearly differs from *G. boreoalpina* in light microscope (Figs 2–35 vs. 49–92). A similar morphologically species is *G. digituloides* (Table 2), known from Finland and reported from the Balkans (Lotter & Hofman 2003), Scandinavia (Wilander 2003), Carpathian high altitude lakes (Buczkó et al. 2013a, b), Canadian Arctic (Bouchard et al. 2004), and Siberia (Genkal et al. 2012). Despite the fact that *Genkalia digitulus* (as *Navicula digitulus*) is the most cited species in the genus *Genkalia* but in fact *G. digituloides* or *G. boreoalpina* are probably more common. The co–existence of *Genkalia digitulus* with *G. boreoalpina* or *G. digituloides* suggests similar autecology for these diatoms.

High abundances of *Genkalia digitulus* and *G. boreoalpina* indicate slightly acidic, cold waters of a very low mineral content, though sparse, small cells of the first species were found in the Bijambare fen water of moderate mineral content. Another "mineral–rich *Sphagnum* fen", from which the species was reported (without illustration) is located in the Western Carpathians (Fránková et al. 2009).

According to the literature environmental changes and especially hydrogen potential (pH) changes are characteristic feature for European *Genkalia*. According to Buczkó et al. (2013a) taxa belonging to *Genkalia* were observed during abrupt pH changes in lakes from the Retezat Mountains. According to Bigler & Hall (2003) a large increase in abundance of *Genkalia digitulus* (as *Navicula digitulus*) coincided with the onset of rapid sedimentation rates. *Genkalia digitulus* was found most often in slightly acidic waters e.g. in Tatra Mountains (Gąsiorowski & Sienkiewicz 2010). Increase in abundance of this diatom coincided with a change of pH up to a range of 6–7 in Pyrenees (Catalan et al. 2009). Additionally pH optimum for *G. digitulus* from northern Sweden lakes was determinated as 6.49 (Bigler et al. 2000) or 6.6 by Rosén et al. (2000). A narrow pH tolerance range for *G. digitulus* and probably also for *G. boreoalpina* allows the use of these taxa in paleoreconstructions for environment or climate change. The probable preference of *Genkalia subprocera* for circumneutral–alkaline water makes an exception among *Genkalia* species.

Published illustrations may suggest that morphological variability and diversity of the genus *Genkalia* is much larger, including northern Europe (e.g. Metzeltin & Witkowski 1996), Africa (Cocquyt 2007), Australia (Chessman et al. 2007) and South America (Servant–Vildary 1986).

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