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Achnanthidium delmontii sp. nov., a new species from French rivers

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Abstract: A new freshwater diatom species belonging to the genus *Achnanthidium* KÜTZ. has been found in several French rivers namely those of the Languedoc–Roussillon and Rhin–Meuse regions. *Achnanthidium delmontii* sp. nov. is described based on light and scanning electron microscopy observations. This species belongs to the group of *Achnanthidium* taxa with terminal raphe fissures curved to the same side of the valve and it is compared to the most related species: *Achnanthidium pfisteri* LANGE–BERTALOT, *Achnanthidium pyrenaicum* (HUSDEDT) KOBAYASI, *Achnanthidium temniskovae* IVANOV et ECTOR and *Achnanthidium zhakovschikovii* POTAPOVA. The assessment of its ecology was made through physicochemical analysis carried out on sites which are part of water quality monitoring networks. Some comments are made on the rapid expansion of *Achnanthidium delmontii* sp. nov.

Key words: Diatoms, *Achnanthidium*, *Achnanthidium delmontii* sp. nov., Languedoc–Roussillon region, France

Introduction

ROUND et al. (1990) provided a renewed interest in diatom systematic with the creation of many new genera and also the reappraisal of older genera such as *Achnanthidium* KÜTZING (1844), later more precisely defined by ROUND & BUKHTIYAROVA (1996). Presently, the *Achnanthidium* genus includes two groups: a first group with straight terminal raphe endings and a second group with the terminal fissures deflected to the same side of the valve. Following the survey of samples taken from rivers of the Languedoc–Roussillon region in the framework of water quality monitoring, five species with curved raphe fissures were found. One of them, *Achnanthidium delmontii*, is proposed as a new species. The four others species, *Achnanthidium druartii* RIMET et COUTÉ, *Achnanthidium latecephalum* KOBAYASI, *Achnanthidium pyrenaicum* (HUSDEDT) KOBAYASI and *Achnanthidium rivulare* POTAPOVA et PONADER, present a high percentage of relative abundance in some rivers of this region. *A. delmontii* sp. nov. was recorded in several rivers of the river water monitoring networks in France. Till now, in routine inventories which aim to monitor biological quality of rivers, *A. delmontii* sp. nov. was misidentified under the name *A. pyrenaicum*, *A. pfisteri* LANGE–BERTALOT or non identified to the species level.

This paper presents the morphology, the ecology and the distribution in French rivers of *A. delmontii* sp. nov., compares it to the most closely related species and give information on its rapid expansion.

Material and Methods

The samples were collected using the French standard NF T 90–354 (AFNOR 2000) in many different rivers of the Languedoc–Roussillon and Rhin–Meuse regions by different public and private organisations in charge of monitoring water quality control networks (ASCONIT Consultants, AQUASCOP, DREAL Languedoc–Roussillon). The diatoms were sampled on natural substrates such as pebbles and stones. The samples were treated by oxidation with hot 40% hydrogen peroxide and hydrochloric acid and rinsed three times with deionised water. For LM observations, cleaned diatoms were mounted in Naphrax. LM and morphometric measurements were performed with

a LEICA DM5000B microscope using a 100x oil immersion objective and a LEICA DFC 420 camera. Measurements on *Achnanthidium delmontii* sp. nov. were taken from 127 individuals. Relative abundance data were based on LM counts of at least 400 valves. For SEM examinations, cleaned diatoms were dried onto glass coverslips attached to aluminium stubs using nail varnish and sputter–coated with a thin layer of platinum. The observations were performed using a JEOL–JSM 6700F and a JEOL JMS 6490. The temporal changes in abundance of *A. delmontii* sp. nov. was analysed using existing collection slides (from consultancy firm F. PERES, ASCONIT Consultants, AQUASCOP and DREAL Languedoc–Roussillon) dating back to 2005 for some of the stations. Physico–chemical data were provided by DREAL Languedoc–Roussillon and by the water agency Rhin–Meuse (Table 1).

Results

Achnanthidium delmontii Pérès, Le Cohu et Barthès sp. nov.

Diagnosis

In microscopio photonico: valvae lineari–ellipticae apicibus non protractis late rotundatis, lineares quoad specimina maxima sed ellipticae quoad specimina minima. Longitudo: 7.3–21.4 µm. Latitudo: 3.3–5.1 µm. Longitudo/Latitudo ratio: 2.0–4.7. Raphovalva: area axialis angusta lanceolata; area centralis circiter rectangulata margines valvae attingens, interdum una curta stria ab altero latere. Raphe filiformis recta cum poris centralibus distinctis. Striae subparallelae ad parallelas, 20–26 in 10 µm usque ad 35 in 10 µm ad apices. Araphovalva: valvae plerumque ellipticae. Area axialis acicularis. Striae subparallelae ad parallelas, 18–22 in 10µm usque 25 in 10 µm ad apices; plerumque duae striae remotiores in media parte.

In microscopio electronico: raphovalva leviter concava. Rami raphis cum centralibus extremis forma guttulae in externo visu et curvatis ad opposita latera in interno visu. Fissurae terminales raphis curvatae ad idem latus valvae. Area centralis formans fasciam sed interdum una stria composita una ad tres areolas ad alteram marginem frontis valvae. Striae compositae 5–7 quadrangularibus areolis (45–57 in 10 µm) in valvae fronte et una elongata areola in valvae limbo. Araphovalva: area axialis cum angusto sterno leviter dilatata in media parte. Striae compositae 6–7 areolis (55–61 in 10 µm) in fronte valvae et una elongata areola in valvae limbo.

In duabus valvis: in interno visu, areolarum hymenes ex parte perforati poris tenuiter elongatis; striae interruptae sunt duabus polis in limbo et hyalina aream circum frontem valvae adest.

Holotype: PC0142667, Laboratoire de Cryptogamie, Muséum d'Histoire Naturelle. Paris, France.

Isotype: BR–4247, Departement de Bryophyta, Thallophyta, Jardin Botanique National de Belgique, Meise, Belgique.

Type locality: Chusclan – Rivière Cèze – France (alt : 34 m ; longitude : 1 907 854; latitude: 787811,198 – Lambert II)

Etymology: the specific epithet refers to Dominique Delmont, diatomist at DREAL Languedoc–Roussillon.

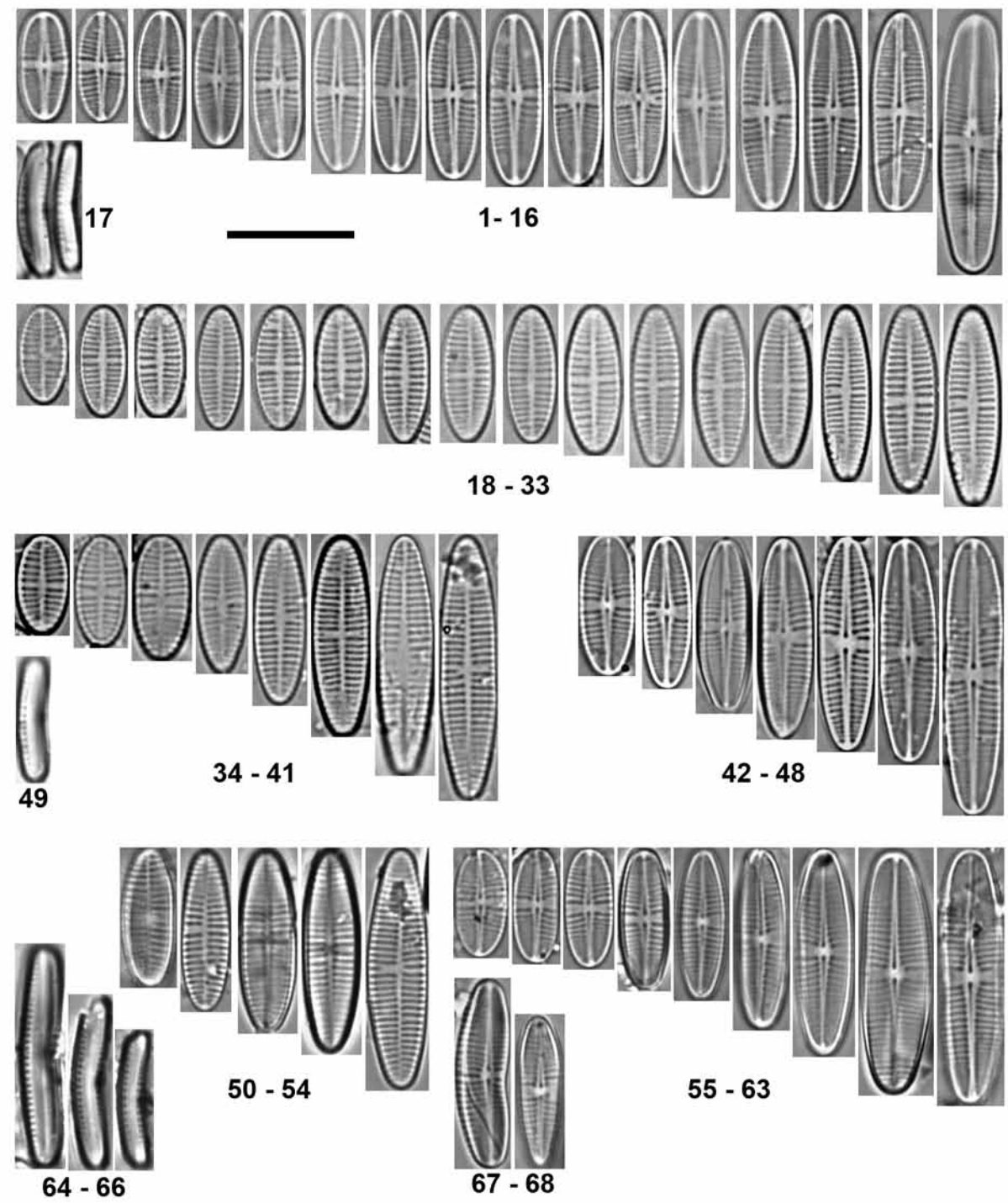
Description

LM: valves are linear with rounded apices becoming elliptical in small individuals (Figs 1–68). On the raphe valve, the axial area is narrow. The central area is irregular forming generally a rectangular fascia but a shortened stria can be present on one of the margin side. The raphe is filiform, straight, with distinct central pores. Striae are slightly radial.

On the rapheless valve the axial area is acicular. Striae are parallel to slightly radiate at the apices; in most cases, two striae are slightly more spaced apart in the middle part of the valve.

SEM: externally, the raphe is straight with droplet–like proximal endings spaced about 0.8 µm apart (Figs 69, 70, 74). The terminal fissures are deflected to the same side and terminate on the valve face near the valve margin (Figs 69, 70, 75). Internally, the proximal raphe endings are curved in opposite directions (Figs 71, 72, 77) and the distal raphe endings terminate in small helictoglossae (Figs 71, 72). The central area is generally irregular, forming a more or less rectangular fascia but can present on one side a shortened stria consisting of one to three areolae (Figs 74, 77). On the raphe valve, the striae are composed of 5 to 7 (45 to 57 in 10 µm) more or less rounded areolae (Figs 69, 70). Sometimes, the Voigt fault is marked by a shortened stria (Figs 70, 72). On the rapheless valve, the striae consist of 6 to 7 areolae (55 à 61 in 10 µm) and two of them are more distant in the middle part of the valve (Figs 73, 76).

On both valves, the areolae are internally occluded by hymenes perforated by delicate slits (Figs 79–82). There is a row of elongated areolae on the mantle (Figs 75, 76, 78) with a gap at the poles; the mantle elongated areolae are separated from those of the valve face by a hyaline area



Figs 1–68. *Achnanthidium delmontii* sp. nov. LM: (1–16) raphe valves; (17) girdle view; (18–33) and rapheless valves from type population (river Cèze at Chusclan); (34–41) rapheless valves; (42–48) raphe valves; (49) and girdle view (river Rhône at Roquemaure); (50 – 54) rapheless valves; (55–63) raphe valves; (64–66) girdle view; (67–68) and abnormal forms (67–68) (river Vieux Rhin at Kembs). Scale bar 10 µm.

Table 1. Minimum and Maximum values of physico–chemical parameters in 2009 and/or 2010 of stations (CZ = Cèze à Chusclan; RO1 = Rhône à Roquemaure; RO2 = Rhône à Arles; CH = Canal de Huningue à Kembs; CI = Canal de la Hardt à Bantzenheim; GC = Grand Canal d'Alsace à Roseneau; IL1 = Ill à Ruelisheim; RH1 = Rhin à Rhinau; RH2 = Rhin à Gambsheim; RH3 = Rhin à Lautebourg–Karlsruhe; VR = Vieux Rhin à Kembs) where *A. delmontii* is found in abundance (– : no data).

	CZ	RO1	RO2	CH	CI	GC	IL1	RH1	RH2	RH3	VR
$N–NH_4$ ($mg.l^{-1}$)	<0.04–0.09	<0.04–0.09	<0.04–0.10	0.064–0.115	0.064–0.15	0.064–0.42	0.064–2.57	0.06–0.206	0.064–0.23	0.039–0.12	0.064–0.23
HCO_3^{2-} ($mg.l^{-1}$)	211–234	167–189	166–195	141–217	137–206	138–206	162–198	139–199	143–195	–	155–191
Ca^{2+} ($mg.l^{-1}$)	67–84	64–75	63–75	45–69	44–64	44–63	54–69	43–67	44–67	46–61.8	47–64
CO_3^{2-} ($mg.l^{-1}$)	–	–	–	2	2	2	2	2	2	–	2
DOC ($mg.l^{-1}$)	0.9–2.0	1.1–2.5	1.1–2.9	1.5–2.4	1.5–2.5	1.4–4	1.8–4.4	1.3–3.6	1.4–4.7	1.3–3.4	1.3–3.1
Chl *a* ($\mu g.l^{-1}$)	<1–4	<1–2	<1–4	0.4–2.8	0.5–5.7	0.1–5.4	0.4–11	0.1–5.3	0.1–5.9	0.336–7.11	0.1–5.3
Cl^- ($mg.l^{-1}$)	8.0–13.2	13–19.5	13.7–21.3	7.7–20	8.4–21	7.3–29	14–27	7.5–36	8.6–34	10.4–33.7	8.3–23
Cond ($\mu S.cm^{-1}$)	364–481	351–491	359–535	310–460	300–440	275–465	275–520	270–470	275–480	305–463	290–430
BOD_5 ($mg.l^{-1}$)	<0.5–1.5	<0.5–2.4	<0.5–2.9	3–5	3–5	3–5	3–10	3–4	3–5	0.8–1.8	3–5
COD ($mg.l^{-1}$)	<5.0–7.5	<5–14	<5–38	5–11	5–9	5–23	5–47	5–25	5–19	5–7	5–12
Mg^{2+} ($mg.l^{-1}$)	6.3–7.3	5.7–6.9	5.4–7.5	6.3–8.4	6.2–8.7	6.1–8.7	5.9–8	5.8–9.3	6–9	6.8–9	6.1–8.4
MeS ($mg.l^{-1}$)	<1–7	<2–48	<1–1122	3.2–33	2–13	2–40	2–130	2–78	2.3–100	2.6–12.5	2–56
$N–NO_3$ ($mg.l^{-1}$)	0.54–1.4	0.74–2.05	0.88–2.05	0.95–2.19	0.90–2.26	0.88–2.12	0.97–3.61	0.93–2.48	0.95–2.15	0.77–2.19	0.90–2.03
$N–NO_2$ ($mg.l^{-1}$)	<0.006–0.015	<0.006–0.027	<0.006–0.033	0.009–0.021	0.009–0.018	0.006–0.027	0.006–0.049	0.012–0.046	0.009–0.040	0.005–0.025	0.012–0.027
DO ($mg.l^{-1}$)	6.75–11.96	8.04–13.1	7.62–12.4	8.2–13.7	8.6–14.9	7–13.8	5.5–14	7.2–14.7	6.8–14.6	7.6–12.9	7–13.8
pH	7.83–8.25	7.86–8.37	7.88–8.55	7.6–8.2	7.85–8.65	7.65–8.4	7.9–8.5	7.85–8.45	7.7–8.4	7.7–8.5	7.55–8.35
Pheo ($\mu g.l^{-1}$)	<1–3	<1–3	<1–5	0.5–3.2	0.1–2.7	0.1–6.9	0.1–10	0.1–6	0.1–6	0.001–2.25	0.1–8.3
$P–PO_4$ ($mg.l^{-1}$)	<0.003–0.075	0.029–0.077	0.026–0.068	0.032–0.042	0.032–0.039	0.032–0.039	0.032–0.202	0.033–0.075	0.033–0.042	0.014–0.034	0.033–0.039
TP ($mg.l^{-1}$)	<0.02–0.08	0.04–0.1	0.04–0.25	0.03–0.2	0.03–0.58	0.03–0.15	0.04–0.53	0.03–0.19	0.03–0.25	0.029–0.055	0.03–0.13
K^+ ($mg.l^{-1}$)	1.2–2.0	1.6–1.9	1.6–2.1	1.6–2.6	1.5–2.3	1.2–2.2	1.9–2.5	1.1–3.4	1.3–3.7	1.7–2.7	1.7–2.1
SiO_2 ($mg.l^{-1}$)	2.92–9.27	4.064–8.382	3.937–8.128	–	–	–	–	–	–	–	–

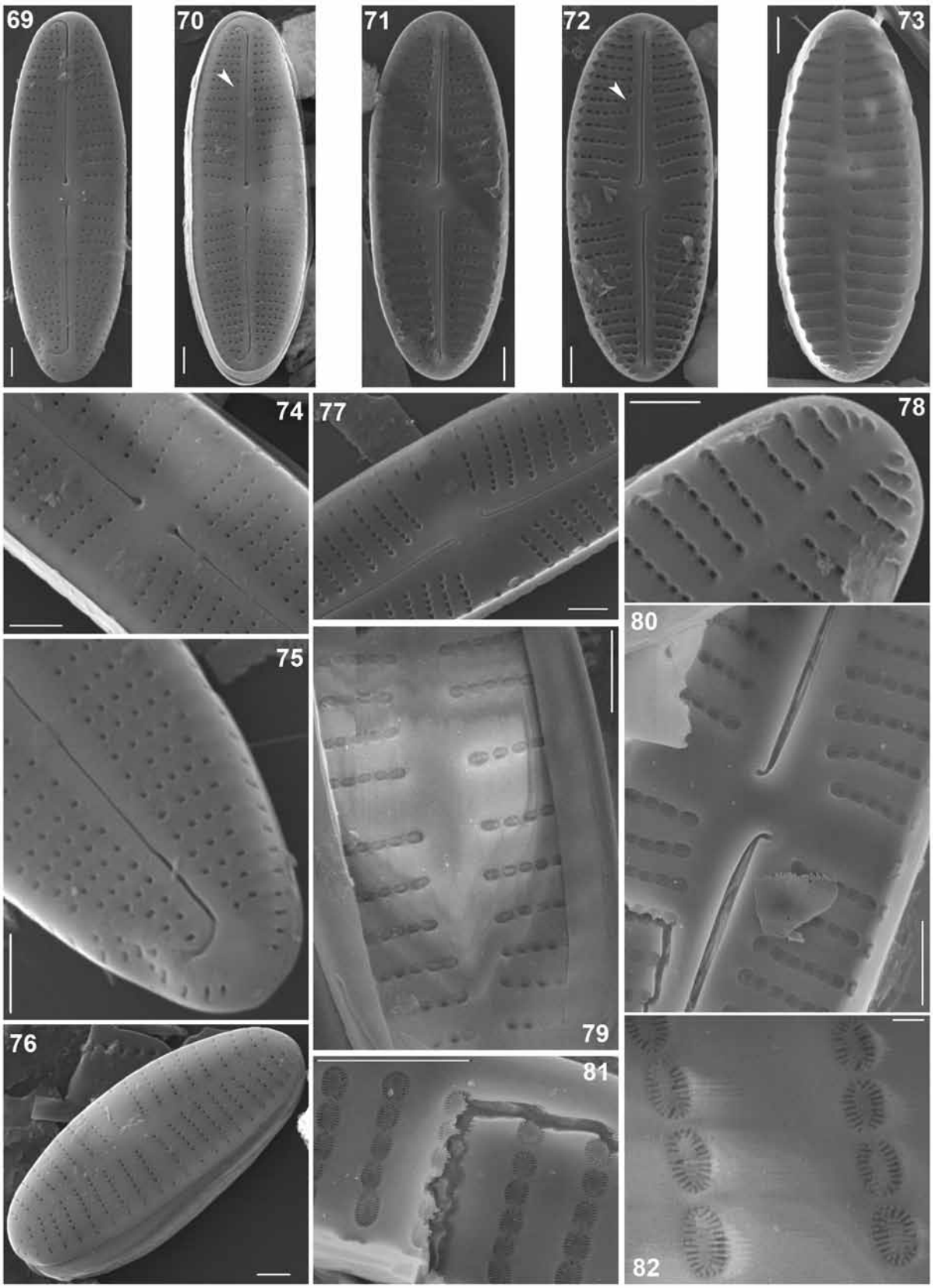
Table 1 Cont.

Na^+ (mg.l^{-1})	8.3–10.5	9.7–13.8	10.4–14.6	6–13	5.9–13	5.8–19	8.5–14	6.4–22	7.2–23	9–20	6.2–15
SO_4^{2-} (mg.l^{-1})	35.0–41.4	39.5–48.2	29–56.1	23–30	23–30	20–30	14–24	20–33	19–33	22–32	22–29
T (°C)	7.6–25.5	6.07–23.8	4.81–25.6	2.4–21.3	2.2–21.9	2.8–21.8	0.8–21	3.6–24.6	3.6–24.7	2.5–24	2.3–21.5
Turb (NTU)	0.7–5.7	6.8–28	3.4–280	1.7–24	1.4–13	1.3–47	1.8–86	0.31–66	0.32–120	–	1.3–49

BOD_5 = Biological Oxygen Demand; Chl *a* = Chlorophyll *a*; COD = Chemical Oxygen Demand; Cond = Conductivity at 25°C; DO = Dissolved Oxygen; DOC = Dissolved Organic Carbon; MeS = Suspended Matter; Pheo = Pheopigments; TP = Total Phosphorus; Turb = Turbidity.

Table 2. Comparison of morphological characteristics of 5 *Achnanthidium* morphologically close.* striae in the middle part of the valve; NC = Not communicated; L – length, W - width

Taxa	Valve characteristics				Raphe valve				Rapheless valve			
	L (µm)	W(µm)	Valve shape	Apex	Striae density.10^{-1}µm Centre	Striae density.10^{-1}µm Apex	Layout of striae	Number of areolae .10^{-1} µm)*	Striae density.10^{-1}µm Centre	Striae density.10^{-1}µm Apex	Layout of striae	Number of areolae .10^{-1} µm)*
Achnanthidium delmontii (Pérès et al.)	7.3–21.4	3.3–5.1	Linear to elliptical	Rounded	20–26	35	Parallel to slightly radial at the endings and more strongly radial towards the centre	5 (45)–7 (57)	18–22	25	Parallel to slightly radial along the whole valve	6 (55)–7 (61)
Achnanthidium pyrenaicum (Pérès et al.)	8.2–16.4	2.9–4.9	linear-lanceolate to slightly elliptical	Rostral	20–28	35–45	Parallel	3 (31)–4 (42)	18–28	35-40	Parallel	3 (25)–5 (42)
Achnanthidium pfisteri (Werum & Lange-Bertalot 2004)	7–12	2.8–5	Linear to elliptical	Rounded	18–20	30	Slightly radial to parallel	4–5 (about 60)	18–25	30	Parallel	6–7 (about 60)
Achnanthidium zhakovschikovii (Potapova 2006)	9.1–14.8	2.9–4.0	Linear to elliptical	Rounded	23–30	>48	Slightly radial	4–5 (about 40)	21–28	30-40	Parallel	4–5 (about 45)
Achnanthidium temniskovae (Ivanov & Ector 2006)	8.8–13.2	2.8–3.8	Linear to elliptical	Slighlty subrostral	22–27	NC	Radial close to the centre and strongly radial at the endings	3–5 (about 46)	19–26	NC	Parallel to slightly radial at the endings	3–4 (about 41)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along the edge of the valve face (Figs 75, 76).

Ecology, associated diatom flora and distribution of *Achnanthidium delmontii* sp. nov.

The type population of *Achnanthidium delmontii* sp. nov., was found in the river Cèze at Chusclan (Gard, Languedoc–Roussillon). It is a Mediterranean river (20 to 50 m wide), which takes its source in the mountain Cévennes and emerges in the downstream part of the Rhône. The type population of *A. delmontii* has its maximum relative abundance in a location characterized by an alkaline pH, high conductivity and calcium concentration (Table 1). The PO_4–P^- level in the river Cèze at Chusclan (maximum PO_4–P^- concentration: 75 µg.l^{-1}) indicates an eutrophic environment (Table 1). These values are quite close to those observed for *A. latecephalum* and *A. druartii* (see also Rimet et al. 2010) in the Languedoc–Roussillon region. When *A. delmontii* was strongly dominant (75%) in the river Cèze at Chusclan, it was associated with *Amphora pediculus* (Kützing) Grunow (4%), *Achnanthidium minutissimum* (Kützing) Czarnecki (4%), *Navicula cryptotenella* Lange–Bertalot (3%) and *Gomphonema minutum* Agardh (2%). The associated taxa are mainly β–mesosaprobe and eutrophic, according to Van Dam et al. (1994), which tend to corroborate the chemical data (Table 1).

A. delmontii appears for the first time in 2007 (Fig. 85) with a low percentage of relative abundance (<1%) in one sample: Vieux Rhin in Kembs (VR). In the type location (CZ), *A. delmontii* appears from 2008 and after that, proliferates (>60%). At the same period, *A. delmontii* is observed for the first time in rivers such as Canal de Huningue (CH), Canal de Neuf–Brisach (CN), Canal de la Hardt (CH), Ehn (EH), Grand Canal d'Alsace (GC), Ill (IL1 and IL2), Lertzbach (LB), Muhlbach de la Hardt (MH), Quatelbach (QB), Rhin (RH1, RH2, RH3), Vieux Rhin (VR), Rhône (RO1 and RO2) and Cèze (CZ) (Fig. 85). The rapid expansion of *A. delmontii* seems to correspond to the criteria defined by Coste & Ector (2000) which qualify invasive species (recent appearance, proliferation, rapid expansion).

Figs 69–82. *Achnanthidium delmontii* sp. nov. SEM: (69) external view of the raphe valve; (70) external view of the raphe valve showing a voigt fault (arrowhead); (71) internal view of the raphe valve; (72) internal view of the raphe valve showing a voigt fault (arrowhead); (73) internal view of the rapheless valve; (74) external view of central area with shortened stria composed of one areola; (75) external view of raphe valve showing the terminal fissures and the interruption of the striae at the end; (76) external view of the rapheless valve showing the hyaline area; (77) internal view of central area showing the raphe proximal endings deflected in opposite side; (78) internal view of the rapheless valve showing the interruption of the striae at the end; (79) internal view of the rapheless valve showing the areolae structure; (80) internal view of the raphe valve showing the areolae structure; (81) internal view of the raphe valve areolae; (82) internal view of the rapheless valve areolae. Scale bars 1 µm (69–81), 100 nm (82).

Discussion

In LM, *Achnanthidium delmontii* sp. nov. most closely resembles *A. pfisteri* Lange–Bertalot (Werum & Lange–Bertalot 2004). *A. delmontii* is much longer than *A. pfisteri* (7.3–21.4 µm vs 7–12 µm, Table 2) and has a higher number of striae on the raphe valve (20–26 vs 18–20 in 10 µm, Table 2). Moreover, in *A. delmontii*, the raphe valve has a well delimited fascia in most cases whereas in *A. pfisteri*, the central area is asymmetrical (Werum & Lange–Bertalot 2004, plate 20, figs 12–13). In SEM, the terminal fissures, deflected to the same side, are much shorter in *A. pfisteri* than in *A. delmontii*. On the raphe valve, the number of areolae per stria is higher in *A. delmontii* (Table 2). The mantle areolae row is interrupted at the apices on the rapheless valve in *A. delmontii* whereas it is continuous in *A. pfisteri* (Werum & Lange–Bertalot 2004, plate 20, fig. 14).

In LM, the presence of a fascia on the raphe valve is a feature shared by *A. delmontii* and *A. temniskovae* Ivanov et Ector (Ivanov & Ector 2006, figs 1–11). Otherwise, both species can be distinguished by the valve morphology and the dimensions (Table 2). In SEM, the raphe valve striae are subparallel towards the apices in *A. delmontii* whereas they are strongly radiate in *A. temniskovae* (Ivanov & Ector 2006, figs 27, 28). The number of areolae per stria is very different in both species and, moreover, the row of mantle areolae does not stop at the apices in *A. temniskovae*, unlike *A. delmontii*.

In LM, *A. delmontii* and *A. zhakovschikovii* Potapova (Potapova 2006) have linear to elliptic valves with rounded apices. Both species differ in

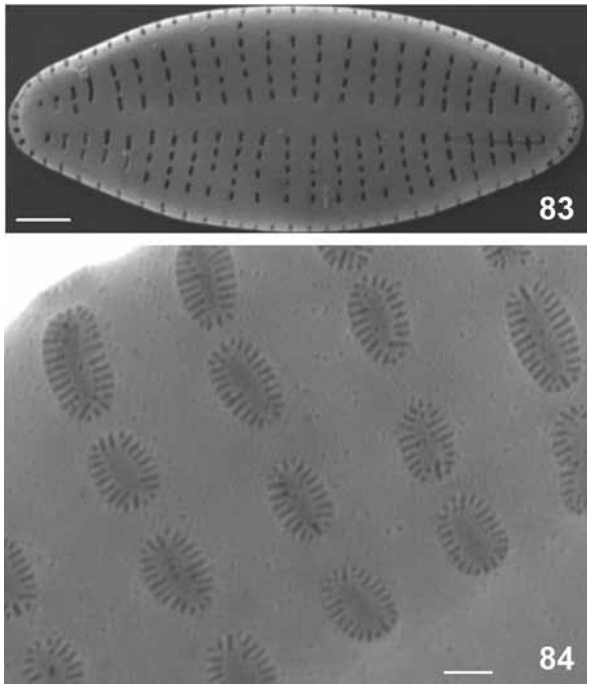

Fig. 83. External view of rapheless valve of *Achnanthidium pyrenaicum*, SEM, Rebenty river, France. Scale bar 1 µm.
Fig. 84. Internal view of the rapheless valve areolae of *Achnanthidium latecephalum*, SEM, river Hérault, France. Scale bar 100 nm.

the dimensions (Table 2) and the number of striae on the raphe valve, ranging from 20 to 26 in *A. delmontii* and from 23 to 30 in *A. zhakovschikovii* (Table 2). Moreover, on the rapheless valve the configuration of the axial area is different (Potapova 2006, figs 2, 6, 7). In SEM, the curved terminal fissures are shorter in *A. zhakovschikovii* (Potapova 2006, fig. 30) than in *A. delmontii*. The number of areolae per stria is lower in *A. zhakovschikovi* on both valves (Table 2). The raphe valve areolae are more or less rounded in *A. delmontii* whereas they are transapically elongated slits in *A. zhakovschikovii* (Potapova 2006, fig. 30). Moreover, *A. delmontii* differs from *A. zhakovschikovii* (Potapova 2006, fig. 31) by the interruption of the mantle striae row on the rapheless valve at the apices.

In LM, *A. delmontii* can be differentiated from *A. pyrenaicum* by the valve morphology (Kobayasi 1997, figs 3–6; Potapova & Ponader 2004, fig. 105). *A. delmontii* is characterized by a fascia on the raphe valve (Figs 1–16) whereas the central area is asymmetrical with often a stria on one side in *A. pyrenaicum* (Kobayasi 1997, fig. 11; Potapova & Ponader 2004, fig. 105). Length being equal, *A. delmontii* is wider than *A. pyrenaicum* (personal observation). In SEM, in external view, the areolae are rounded on both valves in *A. delmontii* whereas they are transapically elongated in *A. pyrenaicum* (Fig. 83, Kobayasi 1997, fig. 15; Potapova & Ponader 2004, figs 105, 106). On both valves, *A. delmontii* has 5 to 7 areolae per stria against only 3 to 4 in *A. pyrenaicum* (Table 2). On the rapheless valve, the row of mantle striae is interrupted at the apices in *A. delmontii* (Fig. 78) whereas it is continuous in *A. pyrenaicum* (Fig. 83).

To conclude, we draw attention to the detailed ultrastructure of the areolae. *A. delmontii* (Fig. 82), *A. latecephalum* (Fig. 84), *A. convergens* (Kobayasi) Kobayasi (Kobayasi 1997, fig. 68) and *A. japonicum* (Kobayasi) Kobayasi (Kobayasi et al. 2006, pl. 155, figs 17, 18) have areolae internally occluded by hymenes perforated by delicate slits. A few *Achnanthidium* species with straight raphe such as *A. saprophilum* (Kobayasi et Mayama) Round et Bukhtiyarova (Lange–bertalot & Krammer 1989, plate 56, fig. 8; Kobayasi et al. 2006, pl. 161, fig. 17) and *A. pusillum* (Grunow) Czarnecki (Kobayasi et al. 2006, pl. 158, fig. 14) show areolae with a cribrum. This feature is possibly another criterion to separate both *Achnanthidium* groups. Nevertheless, further investigation is required.

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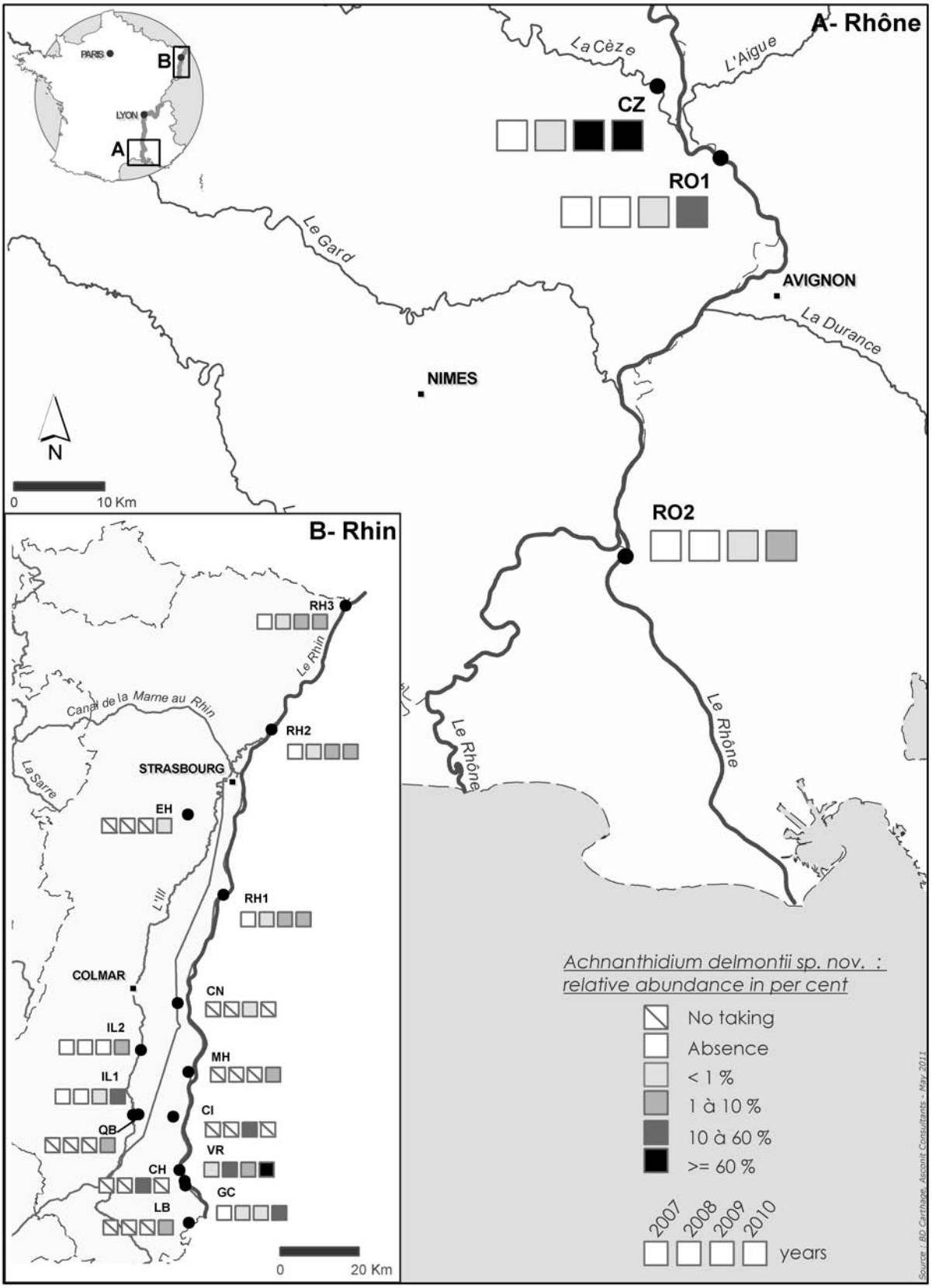

Fig. 85. Distribution and temporal changes of *Achnanthidium delmontii* sp. nov. in percentage of relative abundance from 2007 to 2010.

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