SEDIMENTARY DIATOMS AS INDICATORS OF WATER QUALITY AND ECOSYSTEM CHANGE IN LAKES OF RIDING MOUNTAIN NATIONAL PARK OF CANADA

by

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BSc. (Hon), Mount Allison University, 2010

A THESIS SUBMITTED IN PARTIAL FULFILLMENT OF

THE REQUIREMENTS FOR THE DEGREE OF

Master of Science

in

The College of Graduate Studies

(Environmental Science)

THE UNIVERSITY OF BRITISH COLUMBIA
(Okanagan)

August, 2012

Abstract

The relationship between diatoms and water quality variables was examined in Riding Mountain National Park, Manitoba. In addition, fossil diatom assemblages in Clear Lake and Lake Katherine were assessed in relation to lake trophic status. Through the use of multivariate statistical methods, including Canonical Correspondence Analysis ordination, total phosphorus was determined to be the most significant environmental variable accounting for the greatest proportion of variation among modern diatom communities. Weighted Averaging and Partial Least Squares models were developed as transfer functions to be applied to fossil diatom assemblages, facilitating inferences of historical and pre-historical total phosphorus and lake trophic state. Short core diatom assemblages provided information regarding the most recent changes in lake trophic state, including those driven by human influence, whereas long core assemblages revealed pre-historical conditions indicative of the natural lake state. Paleolimnological assessments of Clear Lake and Lake Katherine revealed a natural borderline oligo-mesotrophic state. Marked changes in the diatom communities coincide with human settlement of the area and, more recently, expanded activities, cottage and golf course development, and sewage system failure. In the most recent decades, changes meant to reduce total phosphorus input have been implemented but the diatom communities have not returned to their pre-settlement composition.

Preface

This research project was undertaken as a collaboration between The University of British Columbia Okanagan and Parks Canada as part of the Clear Lake Action on the Ground Project within Riding Mountain National Park of Canada. All written works and the majority of diagrams presented in this document are of my own making. Several maps (Figure 4.2, 4.3, and 5.2) were produced by Sean Frey of Parks Canada and are reproduced with permission. The Lake Katherine sediment lithology was produced by Terri Lacourse (Figure 5.4), and the Lake Katherine bathymetry was produced by Burt Kooyman (Figure 5.3), both collaborators with Parks Canada; both figures are reproduced with permission.

Ian Walker and Marlow Pellatt contributed to study design, sampling procedures, and manuscript editing. Jeff Curtis also provided guidance with overall project design and implementation. Heather Gray aided in fieldwork and sample collection. Alison Henry participated in processing and preparation of materials for microscope analysis.

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List of Abbreviations

BP – before present (present = 1950)

C2 – program used for model execution

CANOCO – program used for ordination analysis

CCA – Canonical Correspondence Analysis

Chl a – Chlorophyll *a*

Spec. Cond. – specific conductance

CONISS - Constrained Incremental Sums of Squares cluster analysis

DCA – Detrended Correspondence Analysis

DIC – Differential Interference Contrast microscopy

DI-TP – Diatom Inferred Total Phosphorus

DO – Dissolved Oxygen

ML – Maximum Likelihood

PCA – Principal Components Analysis

PLS – Partial Least Squares

r^2_{boot} – bootstrapped coefficient of variation

RMNP – Riding Mountain National Park of Canada

RMSEP – Root Mean Square Error of Prediction

SD – Standard Deviation

Secchi/Depth – Secchi depth divided by depth of water at sample point

Temp. – Temperature

TN – Total Nitrogen

TP – Total Phosphorus

WA – Weighted Averaging

WA-PLS – Weighted Averaging Partial Least Squares

Acknowledgements

I would like to sincerely thank all members of my committee for helping me through this process, from their advice on sampling procedures down to the last edits of my manuscript. I would like to especially thank my supervisor Ian Walker for answering my never-ending stream of questions with few but reassuring words. Also a big thank you to Marlow Pellatt for providing guidance, even from afar, and Jeff Curtis for asking me those questions that needed asking.

All members of the Paleolab provided me with a sense of place and community and acted as an exceptional sounding board. Erin Fleming acted as both a friend and example throughout all stages of my research. Thank you to Travis Dickson for your assistance in the steep learning curve that is the statistics, and for being a calming influence. I could not have asked for a better laboratory assistant than Alison Henry, who exceeded all my expectations by handling samples with all the care and attention as if they were her own. A special thank you to Heather Gray, with whom I found out how entertaining fieldwork, and grad school in general, can be. Thank you to all other members of the Paleolab, if for nothing else than putting up with the counter sounds associated with the enumeration of over 100,000 diatom valves.

The successful collection of field data, sediment samples and historical records could not have been completed without the generous support and assistance provided by the Parks Canada staff in Riding Mountain National Park (RMNP). I thank Bob Reside for all his efforts in providing everything needed for a successful field season, from safety vests to a place to live. I also thank Ross Robinson for all his skills and patience during the entire field season; all of his help was priceless. A thank you to Roxy Grzela, without whom I am sure I would be at least five water samples and one role model poorer. All of the assistance and friendship from the summer students was greatly appreciated, including Karlee McLaughlin, Eric Anderson, Peter Tarleton, Will McCutcheon and Jayson Bennett. Thank you to Paul Tarleton, Tim Sallows and Sean Frey as well, for their assistance in Manitoba and beyond. I would like to thank all other members of the RMNP staff who are not mentioned here for accommodating the needs of my research throughout the summer months and beyond.

My research was financed primarily through funds provided by Parks Canada as well as various graduate scholarships and grants including the University of British Columbia Okanagan (UBCO) Graduate Travel Grant, UBCO University Graduate Fellowship, UBCO Graduate Entrance Scholarship, Hannah T. Croasdale Fellowship, and Friends of the Iowa Lakeside Lab Merit Scholarship.

The expansion of my phycological education could not have been accomplished without both Sarah Spaulding and Mark Edlund, my professors at the Iowa Lakeside Lab facility. With their patience and guidance, my effort to use diatom frustules as paleoecological tools seemed much more manageable.

Finally, I would like to thank my family, including my parents, Mary and John White, as well as my sisters Ashley and Allison White, who listened with open ears to all the ups and downs and always provided me with love and support. I appreciate all that my nephew Hayden helped me with, including my homework; he had a special way of making all problems seem manageable. To the rest of my family, I thank you for your support in all of my endeavours as a student. To my best friends, from all parts of the country, without which I could not have reached this goal, many thanks for providing affirmation that I can accomplish all goals, in my research and elsewhere.

Dedication

To my family and friends, thank you for being my distraction

Chapter 1. Introduction

Determining appropriate goals for lake system management requires knowledge of both historical and prehistoric environmental conditions. Thus, to maintain the ecological integrity of aquatic ecosystems (one of the missions of Parks Canada) in Riding Mountain National Park, an understanding of the natural ecology should first be established. Paleoecology, and specifically paleolimnology, provides tools that can be used in this regard (Smol 1992). Here I focus on one of these tools, diatom microfossils as paleoindicators of lake water quality, trophic state, and ecosystem integrity.

One main goal of this thesis is to establish a regionally-based diatom transfer function (i.e. a model for inferring water quality variables from diatom microfossils) that has potential to be used with fossil diatom assemblages to reconstruct a nutrient of importance in RMNP lakes. In order to accomplish this goal, the nutrient(s) of importance to the RMNP diatoms must first be identified. An additional goal of this thesis is to establish the trophic state changes that have occurred in both Clear Lake and Lake Katherine in RMNP, through the analysis of diatom microfossils present in these lakes' sediments. This quantitative analysis allows reconstruction of the nutrient of importance (e.g. total phosphorus) values extending from the present to pre-historic times. In addition to this, a qualitative assessment of trophic changes is achieved by examining the ecological preferences of dominant diatom taxa and changes in the diatom assemblages through time.

Chapter 2. Literature Review

2.1 Division Bacillariophyta – The Diatoms

The first scientific record of diatoms can be found in the proceedings of the Royal Society of London Philosophical Transactions in 1703 as presented by a Mr. C. (Round et al. 1990). Diatoms were not incorporated into the Latin binomial nomenclature until the late 18th century (Round et al. 1990). After their discovery, diatoms were classified into both the animal and plant kingdoms respectively due to the motility of some species in combination with the presence of a protoplast, and in contrast, other non-motile species that grew in colonial form (Round et al. 1990). Today diatoms are classified as the Division Bacillariophyta (Krammer et al. 1997-2000). There is some controversy regarding classification and taxonomy of some diatom species. With respect to this project, the classification as presented in Krammer et al. (1997-2000) will be followed.

Diatoms are unicellular, microscopic, eukaryotic organisms (Round et al. 1990). In living form, the protoplasts present within the cell aid in identification along with the intricate structure of the cell wall (Round et al. 1990). It is the unique structure of the cell wall and its glass-like composition that has intrigued scientists and sparked research in this group over many decades (Round et al. 1990). The frustule (cell wall) is composed of three main parts: two valves (the epivalve and hypovalve) and a girdle, a band-like structure that acts as a connector between the two valves (Ross et al. 1979). It is the face of the valve that is most commonly used in the identification of microfossils, as it well preserved and intact in the majority of cases, and has sufficiently detailed morphology for identification purposes (Round et al. 1990).

2.1.1 Diatom morphology

There are many structures located on the face and mantle of the diatom valve. These are the structures that allow for its identification under the light microscope. The overall valve shape is usually the initial feature observed, which differentiates the bilaterally symmetric pennate diatoms from the radially symmetric centric diatoms. The presence or absence of a raphe further delineates diatom groups. A raphe is a complex or simple slit that runs along the apical axis of some diatoms that functions in motility (Barber & Haworth 1981). The shape of the raphe is considered to be an important feature in species identification within the Order Pennales. Perforations and processes, including alveoli, setae and spines, are key morphological features used in diatom taxonomy (Krammer et al. 1997-2000). Other surface structures such as costae and furrows can also aid in diatom identification (Krammer et al. 1997-2000). In many cases a

Scanning Electron Microscope (SEM) is required to see diatom frustules at the level of detail required in order to view these individual structures and processes on the surface of the frustule. However, positive identification is possible with the use of a light microscope and Differential Interference Contrast (DIC). Full detail regarding the structures and morphology of diatoms is summarized by Barber and Haworth (1981).

2.1.2 Diatoms and paleoecology

Diatoms have been used so prolifically in the examination of past paleolimnological variables because of their generally well known environmental tolerances and their wide distribution in almost every environment (ter Braak & van Dam 1989; Smol & Cumming 2000). It is due to these factors that many studies have used diatoms as a paleoindicator of multiple limnological variables, including salinity (Battarbee et al. 1984; Laird et al. 1996), pH (Birks et al. 1990a), macrophyte populations (Sayer et al. 2010), and primary production (Whiteside 1983). They also have strong relationships to such macronutrients as phosphorus (Smol & Cummings 2000).

Diatoms were included in 34% of the paleolimnological papers reviewed by Davis (1989). Davis (1989) notes that 23.85% of studies cite water chemistry as a factor affecting these biological communities and lake state. Examination of diatoms, and their relationship to water quality variables and macrophytes, provides an informed mechanism to assess factors affecting lake states, including those in Riding Mountain National Park of Canada (RMNP).

The ability of the microfossils to provide an accurate record of environmental change relies heavily upon many factors including the rate of sediment accumulation, degree of mixing within the lake and the exactness of the chronology (Battarbee et al. 2002a). It is important when detailing the composition of the diatom community within a lake sediment core to take into account the effect of local events and sedimentation rates (Battarbee et al. 2002b). As noted by Battarbee et al. (2002a) diatoms have shown little response to climate change at remote locations in recent decades. However, diatoms may be influenced indirectly through the impact of climatic change on other environmental factors such as salinity, ice cover, and growing season (Battarbee et al. 2002a).

Many sediment cores are dated using radiometric techniques including ^{210}Pb, ^{137}Cs, and ^{241}Am (Appleby 2002; Anderson 1990; Battarbee et al. 2002a; Bennion et al. 2001; Smol & Cumming 2000). These methods provide useful chronologies for sediments younger than 150 years. In the case of RMNP, we are most concerned with evaluating the impact of human

populations and management; thus, these radiometric dating techniques can provide an adequate chronology with which to work (Smol & Cumming 2000). In the context of my research, this chronology will provide a framework with which to compare dates of interest to diatom community changes, such as the changes accompanying the initiation of Government management of the RMNP lands, which began as early as 1895 when the Federal Government declared the area a Dominion Timber reserve, and the official establishment of RMNP in 1933 (Parks Canada 2010).

2.1.3 Diatoms and salinity

The relationship between air temperature and diatoms may not be so easily inferred, but the relationship between increased air temperature and salinity is well established, and salinity changes correspond well with changes in diatom community assemblages (Fritz 1990). Indirectly, as inferred from paleorecords, salinity, temperature, periods of drought, and site hydrology can be determined for lakes located in semi-arid to arid regions (Fritz 1990; Laird et al. 2007). An increase in benthic diatoms often accompanies increased salinities (Fritz 1990). The reconstruction of salinity through the use of diatom transfer functions has been very successful at fresh to moderate salinities (Battarbee et al. 1984; Fritz 1990; Laird et al. 2007). This relationship with salinity can aid in the detection of drought frequency (Fritz 1990). The relationship between diatoms and salinity levels has been well established (Battarbee et al. 1984).

2.1.4 Diatoms and pH

The pH of a lake can vary over time with changes in hydrology and inputs, both natural and anthropogenic. Diatoms have been identified as a group of organisms that respond rapidly, displayed as a change in the diatom community, to changes in pH and record this in the microfossil record (Birks et al. 1990a). The reconstruction of pH from diatoms involves modelling the response of contemporary diatoms to modern pH levels (the development of a training set), followed by the inference of past pH from fossil diatom assemblages by applying this model (Birks et al. 1990a).

2.1.5 Diatoms and other chemical variables

Total phosphorous (TP) is a commonly measured indicator of eutrophication. A transfer function can be developed which relates modern TP values and diatom assemblages in order to reconstruct changes in TP from microfossil records (Smol & Cummings 2000). The utility of

this transfer function can depend greatly on the amount of variance and the range in TP levels in the lakes studied (Bennion et al. 2001). In some cases the TP changes may be too small for accurate reconstructions causing discrepancies between diatom-inferred TP and other eutrophication indicators (Bennion et al. 2001). There are many studies, from diverse regions, however where a diatom TP transfer function has provided significant evidence of a shift in lakes towards a more eutrophic state (Reavie & Smol 1998; Reavie & Smol 2001; Bennion et al. 2004). The use of diatoms to develop a TP transfer function is particularly valuable in those lakes that are in an oligotrophic or mesotrophic state, but can be more difficult to establish within lakes already of a more eutrophic state (Reavie & Smol 2001). It should be noted that the relationship between planktonic taxa and nutrient concentration is much easier to establish than a relationship with those diatoms that grow on the surface of submerged macrophytes, as the epiphytes may derive their nutrients mostly from their host rather than from the open aquatic environment (Bennion et al. 2001).

2.1.6 Diatoms and macrophytes

The change in lake state that accompanies eutrophication, particularly in shallow lakes, is most often detected by the shift in abundance and dominance between macrophytes and diatoms (McGowan et al. 2005; Vermaire & Gregory-Eaves 2008). The movement of a lake from a clear, oligotrophic state can be detected in the paleolimnological record through the change from a more macrophyte-dominated community to a more turbid and eutrophic condition where planktonic diatom species take over the role of primary producers. Macrophyte abundance has been found to be a significant factor controlling algal community structure (McGowan et al. 2005). A steady decline in macrophyte abundance often coincides with a gradual increase in pelagic diatom species and may indicate eutrophication or a change in overall lake depth (Sayer et al. 2010). Although not the goal of this study, the pollution history of a lake can also be inferred through diatom and macrophyte assemblages (Reavie & Smol 1998).

2.2 Regression and calibration statistical background

Several statistical methods are used to identify the relationships between environmental variables and diatom communities. This relationship can then be hindcasted through time. Percent abundances or relative abundances of diatom populations in communities are commonly used in summarizing diatom count data (Reavie & Smol 1998; Bennion et al. 2001). Ordinations and cluster analysis methods that are used in the development of transfer functions are important when working with paleo-data and can be powerful tools in the identification of lake type,

ecological condition and chemical reference conditions (Bennion et al. 2004). These statistical approaches are valuable in confirming reference conditions of lake state and assessing impact on lake state (Bennion et al. 2004).

Cluster analysis is often used for the identification of zones in diatom stratigraphies (Bennion et al. 2001). Principal Components Analysis (PCA) and Redundancy Analysis (RDA) are commonly used to determine those environmental variables of importance or to group and summarize groups of diatom species. Correspondence Analysis (CA) and Canonical Correspondence Analysis (CCA) are often more appropriate techniques for the assessment of variables, as they do not assume linearity, unlike PCA and RDA (ter Braak & van Dam 1989). CCA is most appropriate in the assessment of aquatic ecological data, such as paleolimnological microfossils, as the unit of analysis is the site, not the individual (ter Braak & Verdonschot 1995). CCA is only recommended to be used where the relationships are unimodal, the data has positive values but may also contain many zeros, and/or the data have relative values that can be used in the analysis of community composition (ter Braak & Verdonschot 1995).

The most widely-used statistical method for the development of paleo-inference models is a Weighted Average (WA), as described by ter Braak & Juggins (1993). Maximum Likelihood (ML) is another statistical technique that could be used in the determination of this relationship, however the WA method has been shown to perform better in terms of root mean square error, over the ML technique (ter Braak & van Dam 1989; Birks et al. 1990a). The WA technique is preferred over other possible methods within this field of paleoreconstruction due to its combination of simplicity, good performance and its possible application to limnological variables apart from pH (ter Braak & van Dam 1989). The WA method requires three steps as described by ter Braak & Juggins (1993): 1) the WA regression where species' optima are estimated by averaging the values of environmental variables of interest at each site, 2) the WA calibration where the environmental variable of interest is estimated at each site through the weighted averaging of species' optima, and 3) a deshrinking regression used to reduce the impact of multiple averages on reconstruction estimates (ter Braak & Juggins 1989). There are two methods of deshrinking: classical and inverse. In classical deshrinking, the inferred values are regressed with respect to observed values in such a way that residuals are orthogonal to predicted values (ter Braak & Juggins 1993). Inverse deshrinking methods provide errors that are orthogonal to the originally measured values (ter Braak & Juggins 1993). Another method of model development merges use of WA and Partial Least Squares (PLS) techniques into one

model, WA-PLS, which in comparison to WA has the potential to remove bias within the sample set and improve the estimates of taxa optima (Bennion et al. 2001; ter Braak & Juggins 1993).

CCA ordination techniques as well as WA methods operate on the assumption that diatom species respond to environmental gradients in a unimodal fashion (Hall & Smol 1992). The WA method is an approximation of the ML method, as expressed over a Gaussian (unimodal) response model (ter Braak & Juggins 1993).

The assumptions associated with building a paleoenvironmental reconstruction (taken from Imbrie & Webb (1981) in Birks et al. (1990a)) include: that the taxa included in the training set are systematically related to their current environment, that the variable of interest is important or linearly related to the variable to be reconstructed, the ecological responses of taxa in the training set are not significantly different from those in the fossil data, the methods used in regression adequately model the variable of interest, and finally, that environmental variables not of primary interest have negligible influence.

2.3 Eutrophication and total phosphorus

Diatoms are widely used to assess the extent of eutrophication in lakes and TP is widely regarded as the principal nutrient relating lake productivity and trophic state (Anderson 1997). Natural sources of total phosphorus provide background levels of input into aquatic systems, but human activities have often caused a dramatic increase in TP loading over the course of the 20th century.

The oligotrophic, mesotrophic, and eutrophic ranges for total phosphorus values are defined in the Canadian water quality guidelines (Table 2.1) (Canadian Council of Ministers of the Environment 2004). In this thesis, I use this Canadian standard when referring to trophic state.

It is important to interpret results of TP water quality testing in terms of the natural state of the lake. If the lake, for example, is naturally eutrophic, then concerns regarding the current eutrophic state can be abated (Hall & Leavitt 1999).

The delineation of trophic ranges based on TP values gives both context and clarification when discussing TP reconstructions in comparison to both modern and natural levels.

Table 2.1: Lake trophic status and corresponding TP levels (μg L^{-1}) as adapted from the 'Canadian water quality guidelines for the protection of aquatic life: Phosphorus: Canadian Guidance Framework for the Management of Freshwater Systems'. (Canadian Council of Ministers of the Environment 2004).

Trophic status	TP levels (μg L^{-1})
Ultra-oligotrophic	<4
Oligotrophic	4-10
Mesotrophic	10-20
Meso-eutrophic	20-35
Eutrophic	35-100
Hyper-eutrophic	>100

2.4 Geological history of Riding Mountain National Park

The south-western portion of Manitoba, where RMNP is located, was inundated with seawater until approximately 50 -100 million years ago (Saunders 1979); thus, the Park is underlain by a thick sequence of marine sedimentary rocks. The general bedrock geology of RMNP is Cretaceous, consisting mainly of shale, siltstone and sandstone, most of which were deposited when shallow seas were present (Klassen 1989). Following the recession of this sea, the Manitoba escarpment was formed through erosional processes (Saunders 1979). At an elevation of approximately 366-520 metres above sea level today, the Manitoba Escarpment is one of the most prominent features of the south-western Manitoba landscape; it is a 'Prairie step', formed in a region where the bedrock was previously overlain with more resistant beds in preglacial times (Klassen 1989; Saunders 1979). The uplands in the Riding Mountain region of Manitoba, including Riding Mountain, Porcupine Hills and Duck Mountain, are rare remnant high points in this landscape that lie between former pre-glacial valleys (Klassen 1989). The escarpment and other topographic features in RMNP were subjected to additional erosion with glaciations that began approximately 800,000 years ago (Saunders 1979). Glaciation ended with the retreat of the Laurentide ice-sheet approximately 12,000 years ago, with many of the freshwater lakes in RMNP forming in the wake of the retreating ice-sheet (Saunders 1979). The depressions in the landscape that form the lakes of today are most likely derived from the glacial activity that has shaped much of the Manitoban landscape (Klassen 1989).

2.5 Anthropogenic history of Riding Mountain National Park

The region within which RMNP is established has been subjected to many human activities. It is thought that First Nations people had settled the area 6,000 years ago, following the glacier retreated approximately 10,000 years ago (Parks Canada 2011). In the 18th century the Assiniboine and Cree settled and inhabited the area, but by the early 19th century the Riding Mountain region was dominated by the Anishnabe people (Parks Canada and Wilderness Society 2004).

Fur trading was established within the Riding Mountain region and settlers from non-aboriginal populations began moving into the area in the 1880s, shortly after the Canadian Pacific Rail line reached Brandon in 1881 (Parks Canada 2010). Also at this time there were approximately 10 large logging and milling operations present on the Riding Mountain plateau, with much of the milling activity concentrated near settlements along the escarpment (Parks Canada and Wilderness Society 2004). This harvesting activity peaked at the end of the 19th century, coinciding with two major forest fires that occurred in 1890 (Parks Canada and Wilderness Society 2004). In 1895, the area that was to become RMNP was established as a Timber Reserve, and was designated as a Forest Reserve in 1906 (Parks Canada and Wilderness Society 2004). Many farm animals were allowed to graze and haying activities were also permitted within the Forest Reserve (Parks Canada and Wilderness Society 2004).

The area was under consideration for designation as a National Park by 1919 with the original suggestion being that it encompass an area extending 180 km outward from Clear Lake (Parks Canada 2010). This reduced area was rejected and the entire Forest Reserve was given National Park status in 1930, opening officially in 1933, with one mill operation continuing to log within the Park until 1949 (Parks Canada 2010; Parks Canada and Wilderness Society 2004). Permits for the leasing of land and building of cottages within the park commenced in 1925; prior to this time, small hunting cabins were the only homes and were found primarily along the shores of Clear Lake (Parks Canada 2010).

Throughout WWII (1939-1945) the development of recreational facilities within RMNP was reduced, and due to fuel demands of the war, RMNP was again used as a mainstay in wood supply (Parks Canada 2010). German prisoners of war were kept at Whitewater Lake, from 1943-1945, where they served as labourers in wood collection (Parks Canada 2010). Due to the remoteness of the war camp from both the boundary of the Park and surrounding communities, coupled with its location in the centre of Canada, escape from the camp was considered impossible, making it the ideal location to house prisoners (Parks Canada 2010).

In 1941 the population in RMNP and the surrounding 15 municipalities directly adjacent to RMNP was approximately 47,000; population within Wasagaming continued to increase up until 1981, and then subsequently declined (Parks Canada and Wilderness Society 2004). In 1986, RMNP was designated, along with 15 surrounding rural municipalities, as a United Nations Educational, Scientific and Cultural Organization (UNESCO) Biosphere Reserve, aimed at the conservation of biodiversity and sustainable resource use (Parks Canada 2011; Riding Mountain Biosphere Reserve 2012).

2.6 Earlier paleoecological research in RMNP

Several earlier paleoecological studies have examined aspects of RMNP.

Pollen analyses were conducted by Ritchie (1980). Ritchie (1980 in Ritchie, [1989]) interpreted pollen assemblages in the park as indicating a tundra landscape from 18 to 10 thousand years ago (Ritchie 1989). Boreal forests were established at the beginning of the Holocene. Since that time forests of the RMNP region have changed little in terms of species composition, consisting principally of jack pine (*Pinus banksiana),* with white (*Picea glauca)* and black spruce (*Picea mariana*) as well as poplars (*Populus* spp.), white birch (*Betula papyrifera*) and tamarack (*Larix laricina*) (Ritchie 1989).

Diatom analysis was performed on sediments from South Lake, the lake adjacent to, and directly south, of Clear Lake, by Scott & Kling (2006). Scott & Kling (2006) noted significant changes in the biota present in the sediments over the last 40 years, resulting from increased nutrient supply to the South Lake system. Specifically, the decrease in the benthic diatom species *Denticula kuetzingii* and *Mastogloia smithii*, coupled with an increase in the pollution tolerant species *Nitzschia paleae* and *N. palaceae* (Scott & Kling 2006).

Gray (2009) used fossil midges to assess recent changes in the benthic fauna and hypolimnetic oxygen in Clear Lake. Her results indicated little change in Clear Lake over the past 150 years.

These studies have helped elucidate the history of RMNP. My research will expand on this history, via construction of a transfer function, based on lakes in and around RMNP and its down-core application to two RMNP lakes of particular interest. These two lakes are of particular management interest because of their multiple stakeholder status and high public value.

Chapter 3. Modern diatom assemblages and their relationship to water quality variables in the Little Saskatchewan watershed

3.1 Background information

A mission central to a National Park's mandate is the maintenance and restoration of ecological integrity; this includes both biotic and abiotic conditions that are deemed to be "characteristic of its natural region" (Parks Canada 2009). Riding Mountain National Park of Canada (RMNP) is located in the south-western temperate forest region of Manitoba, Canada, and as such has been relatively open to influences from external change, including diverse human activities. Although the Park has enjoyed protected status for over 50 years, this alone does not guarantee that the national park exists in its natural state. Park managers are often faced with a difficult task, trying to preserve the ecological integrity of park ecosystems without solid historical information to define what the original natural state of the park was. Fortunately, paleoecological methods, employing diatoms and other indicators, provide sound methods for reconstructing past conditions of both terrestrial and aquatic systems (Smol 1992).

This chapter focuses exclusively on the first step in quantitative paleoenvironmental reconstruction, transfer function development, establishing the relationship between water quality predictor variables and diatom species assemblages. The objective of this chapter is to identify those limnological variables that can potentially be reconstructed through the use of diatom transfer functions. The relationship between diatoms and water quality variables varies on a regional basis, thus requiring a separate training set to be constructed for each region to ensure optimal model performance (Kauppila & Valpola 2003; Reavie & Smol 1998).

3.2 Study Sites

RMNP covers approximately 2,973 km^2 (Parks Canada 2011). It is comprised mostly of Boreal Forest and grasslands with 96 km^2 occupied by aquatic habitat, including wetlands, streams, etc. (Parks Canada 2011; Saunders 1979). The majority of RMNP is surrounded by agricultural and other rural landscapes (Parks Canada 2011). The forests, due to fires and logging, are stands of mixed ages (Saunders 1979). Freshwater bodies include marshes, bogs, swamps, as well as both shallow and deep lakes (Saunders 1979). The lakes tend to be circumneutral to alkaline (pH range of 7.7 to 9.4) and vary from oligotrophic to eutrophic states (TP range of 7 to 780 $\mu g\ L^{-1}$).

The majority of water bodies within the park are accessible to Park visitors, however restrictions on boating and other recreational use, as well as accessibility issues, limit the extent to which present day human activities impact these sites. Within these lakes phytoplankton is dominated by diatoms, dinoflagellates and both green and blue-green algae (Saunders 1979).

3.3 Methods

3.3.1 Site selection

For my study a sample set of 52 lakes was established within the Little Saskatchewan watershed (Figure 3.1). Lake selection was performed using Parks Canada GIS data and the program ArcGIS® (ESRI 2011). Four lakes of management concern (Clear Lake, Lake Katherine, Moon Lake and South Lake) were included. Other freshwater bodies were selected both within and outside the Park boundary. Selection criteria included that the lakes be located north of HWY 45 (south of RMNP) and within 500 meters of an accessible road or trail. Eighteen of the lakes were selected using these criteria in conjunction with previous macrophyte sampling undertaken by R.D. Saunders (1979). The remaining water bodies were classified into 5 size groups and the number of lakes chosen from each size group was based on the percentage of the total number of water bodies, using stratified random sampling. Of the 52 lakes selected for sampling, 47 were successfully sampled for surface sediments. A list of those lakes sampled and their respective water quality variables is listed in Table 3.1.

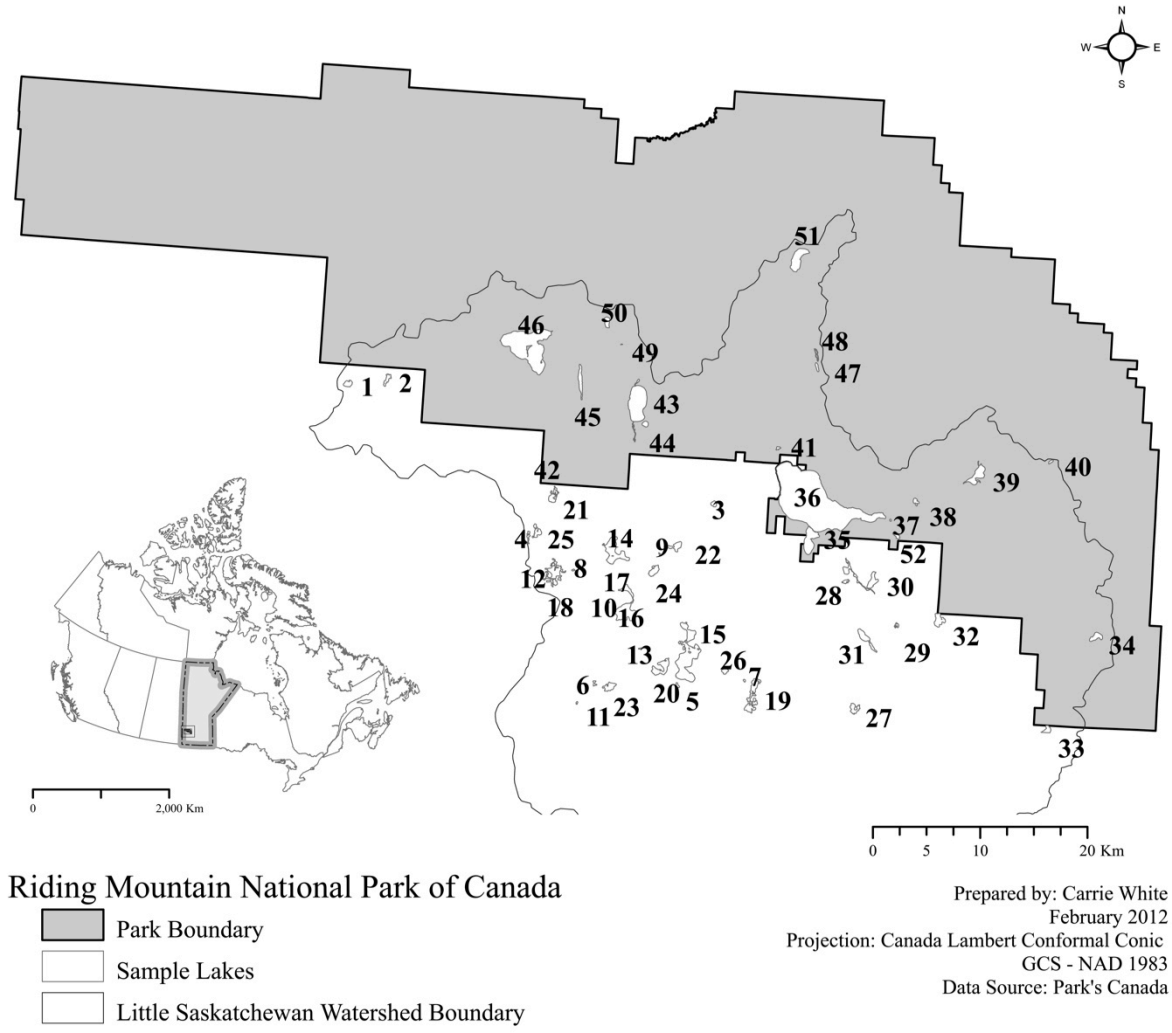


Figure 3.1: Lakes in the Little Saskatchewan watershed from which surface sediment and water samples were collected to be used in transfer function development

Table 3.1: Surface sediment sample lakes and their associated lake characteristics (* indicates those lakes identified as outliers based on DCA and PCA analysis).

Lake Number	Official or Local Name	Latitude	Longitude	Sample Depth (m)	# Diatom species	TP (µg/L)	TN (µg/L)	pH	Specific conductance (µS/cm)
1	n/a	50° 45' 19"	100° 37' 35"	4.1	25	46	830	8.17	363
2	Herchak Lake	50° 45' 22"	100° 34' 56"	1.7	32	131	1880	8.27	399
3	n/a	50° 40' 32"	100° 08' 22"	1.9	21	18	670	8.34	272
4	n/a	50° 35' 40"	100° 20' 31"	3.7	12	111	1920	8.16	1082
5	n/a	50° 31' 16"	100° 10' 19"	3.4	37	31	1190	8.24	1865
6	n/a	50° 31' 06"	100° 16' 52"	3.0	15	148	1450	8.25	1053
7	n/a	50° 31' 44"	100° 05' 02"	0.5	13	40	1300	8.38	637
8	n/a	50° 35' 42"	100° 19' 03"	1.7	25	58	1800	7.95	1368
9	n/a	50° 37' 46"	100° 12' 23"	1.2	26	31	1540	8.58	554
10	n/a	50° 35' 14"	100° 16' 12"	2.2	21	57	1710	8.71	1114
11	n/a			2.2	30	222	1330	8.20	1098
13*	n/a	50° 32' 59"	100° 13' 11"	0.5	22	323	2700	8.88	671
14	n/a	50° 34' 23"	100° 11' 22"	2.5	20	780	3420	8.51	1012
15	Sandy Lake	50° 32' 60"	100° 13' 11"	6.0	16	57	1390	8.18	1273
16	Thomas Lake	50° 34' 44"	100° 13' 52"	11.0	17	52	1240	8.75	547
17*	Stuart Lake	50° 38' 03"	100° 16' 44"	4.3	16	44	880	8.50	1383
18	Wargatie Lake	50° 36' 38"	100° 20' 46"	14.8	20	27	1700	8.64	828
19	n/a	50° 30' 58"	100° 04' 24"	2.7	34	67	1480	8.28	1349
20	n/a	50° 32' 27"	100° 11' 04"	5.9	18	47	1390	8.00	426
21	Czornyj Lake	50° 40' 13"	100° 21' 06"	5.3	25	18	800	8.20	495
22	Battle Lake	50° 38' 13"	100° 11' 02"	3.5	16	273	1160	8.65	1626
23*	Crawford Lake	50° 31' 02"	100° 15' 37"	12	15	83	1670	8.43	2037
24*	Corstorphine Lake	50° 39' 22"	100° 12' 38"	4.6	8	55	2100	9.06	492
25	n/a	50° 36' 58"	100° 22' 13"	1.4	23	38	1390	8.42	484

Lake Number	Official or Local Name	Latitude	Longitude	Sample Depth (m)	# Diatom species	TP (µg/L)	TN (µg/L)	pH	Specific conductance (µS/cm)
26	n/a	50° 32' 11"	100° 06' 52"	2.0	13	44	960	8.23	1061
27*	Leda Lake	50° 30' 18"	99° 55' 10"	2.0	20	343	1650	8.12	374
28*	n/a	50° 36' 57"	99° 57' 49"	2.5	19	10	700	8.98	629
29	n/a	50° 32' 05"	99° 54' 29"	0.4	14	18	1530	8.01	320
30*	Octopus Lake	50° 36' 41"	99° 55' 42"	1.5	25	15	670	8.42	432
31	Ditch Lake	50° 34' 39"	99° 56' 07"	9.6	31	17	1000	8.70	236
33	Kerr Lake	50° 30' 19"	99° 41' 00"	4.0	14	50	590	8.18	197
34	Muskrat Lake	50° 35' 07"	99° 37' 18"	3.03	17	19	630	8.32	361
35	South Lake	50° 38' 44"	100° 01' 01"	1.5	23	28	1150	8.60	374
36	Clear Lake	50° 42' 29"	100° 00' 46"	23	21	8	670	8.31	404
37*	Pudge Lake	50° 40' 16"	99° 54' 19"	6.0	22	12	410	7.75	363
38	Lake Kinosao	50° 41' 18"	99° 52' 27"	6.1	17	17	530	8.09	200
39*	Whirlpool Lake	50° 42' 24"	99° 48' 28"	1.7	28	15	520	8.17	241
40*	East Deep Lake	50° 43' 43"	99° 41' 49"	4.0	36	13	1130	8.18	128
43	Lake Audy	50° 44' 46"	100° 14' 21"	4.5	23	34	870	8.32	391
44	Lake Audy Runoff	50° 44' 18"	100° 14' 14"	1.0	20	13	1170	8.07	952
45	Long Lake	50° 46' 31"	100° 19' 37"	7.3	32	13	670	8.42	361
46	Whitewater Lake	50° 48' 30"	100° 22' 22"	1.1	25	35	860	9.40	317
48	4th Bead Lake	50° 47' 47"	100° 00' 50"	7.0	30	14	610	8.67	288
49	n/a	50° 48' 13"	100° 16' 29"	0.7	34	7	560	7.70	333
50	Cripple Lake	50° 49' 19"	100° 17' 42"	1.0	22	17	900	8.51	226
51*	Moon Lake	50° 52' 52"	100° 03' 12"	5.0	12	47	660	9.28	197
52	Lake Katherine	50° 39' 36"	99° 53' 47"	10.5	23	18	1850	8.19	357

3.3.2 Sediment coring, diatom analysis

To ensure capture of the most recently deposited sediments, the uppermost sediments were collected with a Pylonex HTH corer. Samples were collected at approximately the centre of each lake, as this likely approximates the deepest point. The deepest location is theoretically not disturbed to the same extent as the shores and has a smaller amount of mixing and re-suspension of algal remains (Anderson 1990). Although the cores were typically 30 to 40 cm long only the top three centimetres were sectioned off at 1 cm intervals into Whirlpaks® on shore. For the purposes of this study, only the top 1 cm interval for each lake was processed and included in diatom analysis. The uppermost sediments will contain the most recent diatom populations and as such, will be best related to the water quality measurements derived from each lake site (Smol & Cumming 2000; Wilson et al. 1996).

Sediment samples were stored in a refrigerator at RMNP and were packed in ice and shipped to UBC Okanagan campus where they were stored at approximately 4°C until ready to be processed. Water samples were taken and processed at the Maxxam Analytics laboratory facility in Winnipeg, MB. Water quality variables measured included: total nitrogen (TN), total phosphorus (TP), and chlorophyll a (Chl. a) concentrations. Other water quality variables: temperature, dissolved oxygen (DO), Secchi depth, pH and Specific Conductance (Spec. Cond.), were measured on site, at surface, using a YSI water quality system (Model: YSI 650 MDS with sonde model 600QS-O-M) and a Secchi disk.

Diatom sample preparation approximated the standard method described by Battarbee (1986). Much care was taken to avoid cross contamination. In order to remove calcareous material, as well as many metal salts, the sediment samples were mixed with 10% hydrochloric acid (HCl). When the carbonate had dissolved completely, indicated by a lack of effervescence when additional HCl is added, the samples were then centrifuged, decanted and washed thoroughly with de-ionized water.

The removal of organic matter within the sample can be accomplished with the use of 30% hydrogen peroxide (H_2O_2), as suggested by Renberg (1990), or through the use of a 50:50 solution of concentrated nitric (HNO_3) and sulphuric acids (H_2SO_4) (Battarbee 1986), both of which were used in sample processing. Most samples were processed using the hydrogen peroxide method as described by Renberg (1990). The samples were subsequently washed and centrifuged several times in de-ionized water. This removes any excess acid or peroxide residue still present in the sample (Wilson et al. 1996). As the mineral matter content of most samples

was low, no means of physical separation such as sieving was employed. The samples were mounted on glass slides using Naphrax mounting medium.

Diatom frustules were identified and enumerated at 1000× magnification under light microsopy, using oil immersion and differential interference contrast (DIC) (Wilson et al. 1996). A minimum of 500 diatom valves was counted in transects at 1000× on each slide (Birks et al. 1990a). There is a great range in the number of valves counted in paleolimnological studies; the enumeration of 500 valves lies in the middle of this range and should be sufficient for the development of a transfer function, as it is well above the common count of 300 valves (Bennion et al. 2001; Birks et al. 1990a).

Diatom terminology follows Ross et al. (1979) and descriptions of genera by Round et al. (1990). Several taxonomic references were used in species identification. Mainly the Bacillariophyceae keys, as set out by Krammer & Lange-Bertalot (1997-2000), were used, with supplemental guides including: Camburn & Charles (2000), Cumming et al. (1995), Fallu et al. (2000), Foged (1981), Lange-Bertalot & Krammer (1987), Moser et al. (2004), and Patrick & Reimer (1966, 1975). All species identifications and sample enumerations were performed by the same analyst, providing consistency in taxonomy and sample counts (Birks 1998). As a further assurance of data quality, a taxonomic guide to the diatoms of the RMNP region was prepared and included as Appendix 1 (Figures A1.1 to A1.143) of this thesis. This guide will also facilitate future use of the model by other analysts.

3.3.3 Numerical and statistical analysis

Diatom data were collated into a spreadsheet and relative abundances calculated (percentage of total) for statistical analysis and data screening processes. Only species that occurred in at least two sample lakes and at a relative abundance >1% in at least one of those samples were included in this study. This removed rare species that can have extreme influence on the transfer function (Birks et al. 1990a).

Diatom data were assessed using unimodal multivariate statistical techniques implemented in CANOCO for Windows v. 4.56, a software program used for ordination analysis (ter Braak & Šmilauer 2011). Unimodal multivariate analysis techniques are most appropriate where gradient lengths exceed 3 SD units (ter Braak & Šmilauer 2002). A preliminary detrended correspondence analysis (DCA) was performed to assess the floristic gradient length.

Canonical correspondence analysis (CCA) was subsequently used to interpret the relationship between diatom species assemblages and environmental data. This is appropriate for

unimodal data that have positive values yet contain many zeros (ter Braak & Verdonschot 1995). A CCA is often used as a method of preliminary analysis to determine whether environmental variables have potential for paleolimnological reconstruction (Walker et al. 1991).

Environmental variables were then assessed through a correlation matrix and assessment of normality based on frequency diagrams (Reavie & Smol 1998). Data was log (x+1) or log (x) transformed, where appropriate, and those environmental variables that were very highly correlated were assessed through a series of partial CCAs to determine independent influence. It is often difficult to establish which individual variables of a group of highly correlated variables are affecting the diatom communities (Hall & Smol 1992; Smol & Cumming 2000).

Following this, a principal components analysis (PCA) was performed on the transformed environmental variables. This, in conjunction with the DCA of species data, was used to assess the presence of any outlier lakes within the sample set (Hall & Smol 1992). Lakes were considered outliers if the calculated sample score for the PCA and DCA analyses fell outside the 95% confidence interval about the sample score mean for both the first and second axes for both tests (Hall & Smol 1992).

To account for the greatest proportion of variance within the diatom species data (Walker et al 1991), a full CCA, with manual forward selection and 999 Monte Carlo permutation tests in CANOCO (v. 4.56), was then used to assess the influence and significance of the environmental variables. Only statistically significant variables were included in the final analysis (Birks et al. 1990b; ter Braak & Šmilauer 2011). Ordination diagrams were created using CanoDraw™ for Windows v. 4.14 to show the spread of both sites and species around the two ordination axes in relation to the significant environmental variables (Šmilauer 2011).

Modelling exercises were performed using several techniques including weighted averaging (WA), partial least squares (PLS) and WA-PLS using C2 v. 1.6.8, a software program used for model development and execution (Juggins 2005; Juggins 2010). The goal was to select the minimum adequate model through the inclusion of the smallest number of relevant parameters (Birks 1998). Both the CCA and models were run with varying transformations to the data, including the inclusion or exclusion of those lakes originally identified as outliers and square-root transformation of species data, as well as untransformed species data.

3.4 Results

3.4.1 Relationship of surface sediment assemblages to environmental variables

Over 180 diatom species were identified in the 47 surface samples. After removal of rare taxa, 114 taxa were included in the final analysis. The species included and their associated codes and characteristics for the model are listed in Table 3.2. The taxon with the greatest number of occurrences was *Amphora copulata*, present within 34 of the 47 sample lakes. *Fragilaria brevistriata* had the greatest abundance in a single lake at 81.19%, followed closely by *Stephanodiscus hantzschii* at 81.03%. The WA calculated TP optima ranged from 189.8 to 10.5 (µg L^{-1}) with *Aulacoseira crenulata* and *Nitzschia linearis* having the highest and lowest calculated optima, respectively.

The N2 values (Table 3.2) are values representative of the effective number of occurrences of a taxon, that is, the number of occurrences from which its optimum will be calculated. The N2 values are one of Hill's diversity measures that are calculated through the inverse of the Simpson's Diversity Index (ter Braak & Šmilauer 2002). Following the example put forth by ter Braak & Šmilauer (2002), if, for example, a diatom taxon occurred in four samples, with abundances of 500, 1, 1, and 1, the species WA calculated TP optimum would effectively be calculated from the sample in which it had an abundance of 500, bringing its N2 value close to 1. The N2 values within this dataset range from 1.55, for *Tryblionella angustata*, to 23.62 for *Amphora copulata*.

The relative (percent) abundances of the most abundant and important diatom taxa and their occurrence in the 47 sample lakes are depicted in Figure 3.2. Diatom taxa are arranged from highest (left) to lowest (right) TP optima, and sample lakes are also ranked from highest (top) to lowest (bottom) measured TP values from the 2011 field season.

A preliminary DCA was performed to determine the appropriateness of unimodal versus linear multivariate analysis techniques. Based on the gradient length of 4.493 SD for untransformed, and 3.696 SD for square root transformed species data, it was appropriate to proceed with unimodal techniques, as gradient lengths greater than 4 SD are considered strongly unimodal, and those greater than 3 SD are moderately unimodal (ter Braak & Šmilauer 2002). The abundance of zeros within the data set also suggests that unimodal methods are preferred over linear analysis (ter Braak & Prentice 1988).

Table 3.2: Diatom taxa included in CCA ordination, their associated species codes, the number of lakes in which the taxa were found, their maximum relative abundance, and weighted averaging calculated TP bootstrapped optima in μg L^{-1}.

Species Code	Species Name	# of occurrences	Max Abundance	N2	TP Boot Optima
1	*Achnanthes exigua*	7	8.52	4.14	22.00
2	*Achnanthes lanceolata*	5	5.37	3.44	28.80
3	*Amphipleura pellucida*	7	1.11	6.35	35.41
4	*Amphora copulata*	34	47.20	23.62	41.36
5	*Achnanthes minutissima*	7	7.41	5.01	18.47
6	*Amphora affinis*	4	1.20	3.25	36.89
7	*Amphora pediculus*	13	9.05	9.39	31.89
8	*Anomoeneis sphaerophora*	4	1.01	3.87	34.72
9	*Anom. sphaerophora f. costata*	2	1.21	1.96	32.78
10	*Asterionella formosa*	3	12.39	2.89	44.47
11	*Aulacoseira ambigua*	11	66.80	7.46	37.60
12	*Aulacoseira crenulata*	2	5.96	1.63	189.81
13	*Aulacoseira granulata*	10	42.50	7.54	54.24
14	*Aulacoseira lacustris*	2	1.79	2.00	47.72
15	*Caloneis trochus* var.*trinodis*	4	2.91	2.92	47.30
16	*Caloneis ventricosa*	3	1.55	2.79	129.12
17	*Cocconeis placentula*	20	45.78	10.76	60.85
18	*Cocconeis placentula* var. *lineata*	4	12.50	2.88	112.07
19	*Craticula ambigua*	6	3.20	5.06	73.48
20	*Craticula cuspidata*	15	2.13	13.16	74.48
21	*Craticula halophiloides*	3	1.92	2.72	75.11
22	*Cyclotella bodanica*	3	8.95	2.94	14.63
23	*Cyclotella bodanica* var. *lemanica*	2	42.45	1.64	16.50
24	*Cyclotella comensis*	4	19.15	2.90	23.23
25	*Cyclotella gamma*	7	4.41	5.21	82.86
26	*Cyclotella meneghiniana*	12	15.02	8.31	123.86
27	*Cyclotella michiganiana*	9	5.16	7.60	19.72
28	*Cymbella cesatii*	4	13.01	3.07	15.01
29	*Cymbella subcistula*	6	1.71	5.28	26.60
30	*Cymbella cymbiformis* var. *nonpunctata*	2	2.23	1.68	13.41
31	*Cymbella delicatula*	3	47.59	1.99	12.65
32	*Cymbella descripta*	8	3.52	5.94	14.77
33	*Cymbella ehrenbergii*	5	3.50	3.99	19.32
34	*Cymbella microcephala*	4	12.48	2.84	20.43
35	*Cymbella minuta*	22	7.38	15.88	28.20
36	*Cymbella muelleri*	3	1.94	2.59	22.29
37	*Cymbella neocistula*	13	6.45	10.20	25.58
38	*Cymbella proxima*	11	1.03	10.09	19.31

Species Code	Species Name	# of occurrences	Max Abundance	N2	TP Boot Optima
39	*Cymbellafalsa diluviana*	3	11.85	2.37	13.86
40	*Denticula kuetzingii*	28	49.91	20.35	38.37
41	*Denticula valida*	5	2.69	4.28	18.14
42	*Diatoma tenuis*	5	4.45	3.78	56.88
43	*Diploneis marginestriata*	3	1.99	2.53	18.61
44	*Navicula oblongella*	4	1.99	3.35	12.87
45	*Epithemia adnata*	6	6.48	4.29	23.39
46	*Epithemia turgida*	16	5.89	13.00	37.19
47	*Eunotia arcus*	2	1.03	1.89	13.09
48	*Eunotia bilinearis*	6	2.77	5.21	53.64
49	*Fragilaria brevistriata*	32	81.19	19.88	25.06
50	*Fragilaria capucina*	26	34.15	17.74	50.40
51	*Fragilaria capucina* var. *mesolepta*	9	3.90	7.63	43.14
52	*Fragilaria capucina* var. *gracilis*	2	2.21	1.92	56.14
53	*Fragilaria construens*	17	13.50	13.19	22.73
54	*Fragilaria construens* var. *venter*	2	2.65	1.98	44.87
55	*Fragilaria crotonensis*	23	58.82	17.17	28.05
56	*Fragilaria cyclopum*	6	2.45	5.50	48.27
57	*Fragilaria delecatissima*	11	13.90	8.76	43.29
58	*Staurosirella pinnata*	20	36.85	14.57	18.90
59	*Fragilaria tenera*	3	1.73	2.98	116.01
60	*Fragilaria vaucheriae*	6	2.64	5.47	35.56
61	*Gomphonema acuminatum*	14	17.90	8.32	29.34
62	*Gomphonema angustum*	17	13.85	13.09	46.70
63	*Gomphonema gracile*	2	1.15	1.71	173.95
64	*Gomphonema intricatum*	15	10.39	10.64	28.59
65	*Gomphonema parvulum*	9	7.31	6.92	43.15
66	*Gomphonema truncatum*	9	1.37	8.29	21.06
67	*Gyrosigma acuminatum*	7	1.41	6.56	17.01
68	*Mastogloia grevillei*	2	1.78	1.89	20.25
69	*Mastogloia smithii*	20	17.77	12.71	23.25
70	*Martyana martyii*	2	1.37	1.66	27.35
71	*Hippodonta capitata*	8	23.45	4.83	42.62
72	*Hippodonta hungarica*	8	19.40	4.68	73.78
73	*Navicula cari*	3	1.67	2.84	14.81
74	*Navicula cryptocephala*	7	3.82	5.69	27.14
75	*Navicula cryptotenella*	4	39.62	2.34	54.16
76	*Navicula disjuncta*	13	2.99	11.02	20.40
77	*Navicula explanata*	9	2.39	8.15	26.78
78	*Navicula laevissima*	4	3.11	3.42	56.97
79	*Navicula minima*	2	1.28	1.99	42.43
80	*Navicula oblonga*	19	25.94	12.17	23.80

Species Code	Species Name	# of occurrences	Max Abundance	N2	TP Boot Optima
81	*Navicula pupula*	28	30.10	17.59	30.51
82	*Navicula radiosa*	28	21.94	20.07	24.67
83	*Navicula rhynchocephala*	4	21.53	1.82	24.72
84	*Navicula trivialis*	25	34.40	17.00	29.84
85	*Navicula tuscula*	9	6.95	5.54	15.40
86	*Navicula veneta*	20	29.27	12.05	50.61
87	*Neidium hankensis*	2	2.27	1.92	55.26
88	*Nitzschia dissipata*	4	2.04	3.66	36.39
89	*Nitzschia frustulium*	10	6.69	7.15	67.15
90	*Nitzschia gracilis*	5	3.38	4.29	54.70
91	*Nitzschia linearis*	2	1.59	1.78	10.54
92	*Nitzschia obtusa*	2	1.54	1.62	86.46
93	*Nitzschia palea*	22	8.95	16.29	47.27
94	*Nitzschia sigmoidea*	5	1.55	4.58	21.26
95	*Pinnularia divergens*	4	5.98	3.28	14.24
96	*Pinnularia interupta*	7	1.75	6.14	17.61
97	*Pinnularia nodosa*	2	3.11	1.95	15.04
98	*Pseudostaurosira parasitica*	10	8.13	8.23	28.75
99	*Rhopalodia gibba*	17	10.29	12.16	35.49
100	*Stauroneis anceps*	4	38.06	2.14	15.73
101	*Stauroneis phoenicenteron*	4	1.52	3.57	16.90
102	*Stephanodiscus hantzschii*	21	81.03	16.47	102.90
103	*Stephanodiscus medius*	5	11.40	4.45	146.20
104	*Stephanodiscus minutulus*	14	30.13	9.61	88.57
105	*Stephanodiscus niagarae*	12	4.64	10.68	41.06
106	*Stephanodiscus parvus*	4	15.74	3.42	90.59
107	*Surirella angusta*	4	2.77	2.66	106.94
108	*Surirella linearis*	7	1.94	5.74	66.88
109	*Surirella ovalis*	3	15.82	2.00	174.52
110	*Synedra acus*	2	1.03	1.97	22.65
111	*Synedra incisa*	3	1.81	2.57	41.76
112	*Synedra ulna*	10	14.20	6.92	39.50
113	*Tabellaria flocculosa*	11	9.14	8.42	29.66
114	*Tryblionella angustata*	2	3.54	1.55	23.99

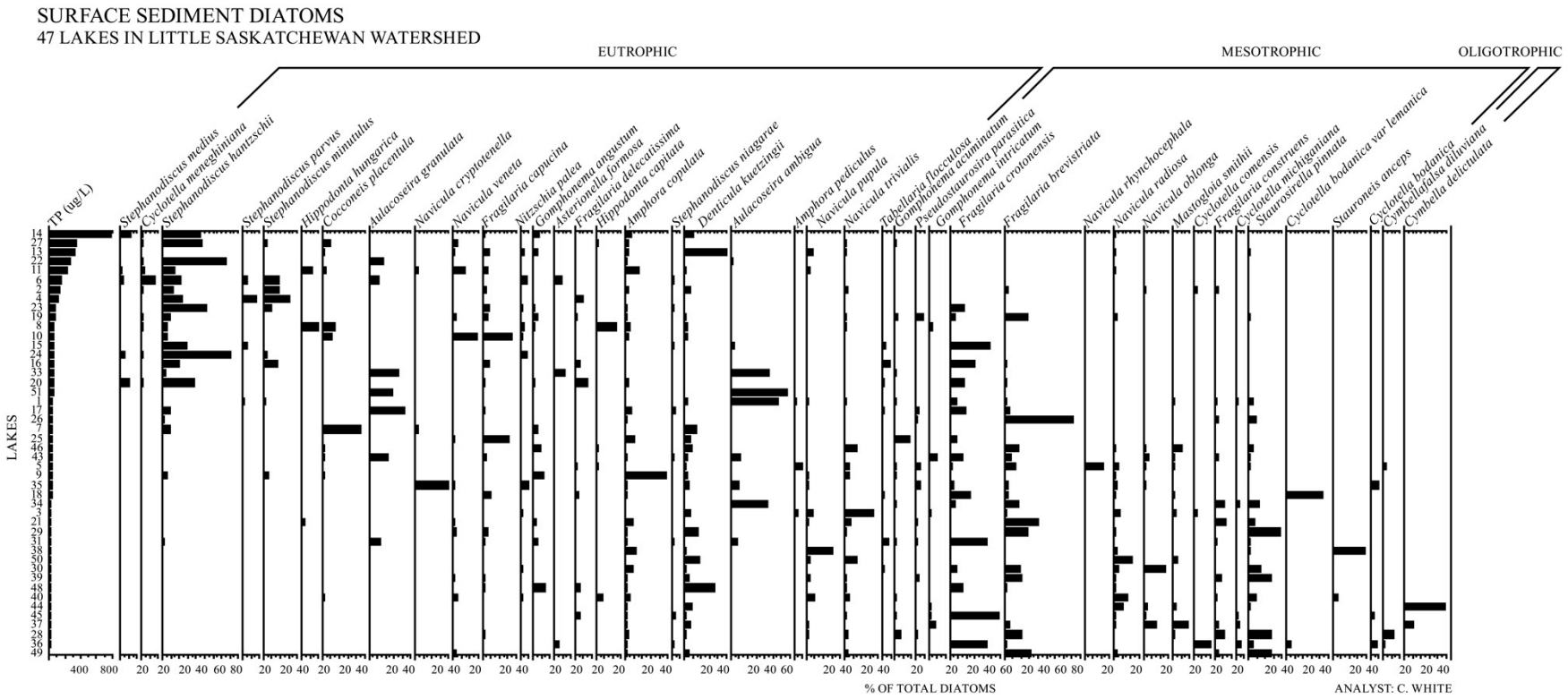


Figure 3.2: Distribution of diatom taxa within 47 surface sediment samples included in model; sample sites arranged by measured TP from highest (top) to lowest (bottom) and diatom taxa arranged from highest (left) to lowest (right) weighted averaging calculated TP bootstrapped optima in µg L^{-1}

The presence of outlier lakes within the dataset was assessed using a preliminary DCA and PCA of species and environmental data (respectively) as presented by Hall & Smol (1992). The 95% confidence intervals were calculated for each of the four axes (PCA Axis 1 and 2, DCA Axis 1 and 2). Those sample sites where all four axes scores fell outside the calculated confidence intervals are considered, from a statistical standpoint, outlier sample sites in the dataset. Lakes 13, 17, 23, 24, 27, 28, 30, 37, 39, 40, and 51, fall into this category. Full descriptions of these lake sites and a summary of the outlier lake analysis is presented in Appendix 2: Field Notes and Additional Data. Statistical methods proceeded under two dataset scenarios, one with the inclusion of outlier lakes and another excluding outlier lake samples.

Correlation analysis (Table 3.3) revealed that several environmental variables were significantly correlated ($p \leq 0.05$). There are a strong correlations among Specific Conductance, Chlorophyll *a*, Total Nitrogen, Total Phosphorus and Secchi/Depth (measured Secchi depth divided by depth of lake at sampling location) (all log transformed). Secchi/Depth was also found to be intercorrelated with DO, and Chl. *a* in addition to TP. A significant correlation between two variables indicates that the variables are not independent statistically (Reavie & Smol 1998).

Following this, the variables were also assessed to determine whether each had an independent correlation with the diatom assemblages. Independence was assessed using a sequence of partial CCAs, (Table 3.4). In each partial CCA run, one environmental variable was declared "the variable", while the other seven environmental variables were included as co-variables. Secchi/Depth (log transformed) was statistically independent from the other environmental variables ($p \leq 0.05$) under all variations in the dataset. TP was the only other variable found to be significantly independent, but only where outlier lakes were included and species data were square root transformed. This relationship was also only significant to the $p \leq 0.10$ level (Table 3.4).

Table 3.3: Pearson correlation matrix depicting the correlation coefficients among environmental variables (*indicates t-test between variables significant at $p \leq 0.05$, with a t-critical value of 2.0141).

	Temp.	Log (Spec. Cond.)	DO	pH	Log(Chl a)	Log (TN)	Log10(TP)
Log(Spec. Cond.)	0.20155						
DO	0.29696	-0.26044					
pH	-0.12038	-0.02952	0.21231				
Log(Chl a)	0.15674	0.31952*	0.22102	0.19348			
Log (TN)	0.00575	0.51626*	-0.14694	0.13185	0.32956*		
Log10(TP)	0.12716	0.49316*	0.02625	0.15192	0.70818*	0.69329*	
Log (Secchi/Depth)	-0.08745	-0.22967	-0.32077*	-0.13344	-0.5467*	-0.19477	-0.40964*

Table 3.4: Analysis of independence of environmental variables, as performed through a series of partial Canonical Correspondence Analyses. Canonical coefficients of the eight environmental variables included in the analysis (their approximate significance) (**indicates t-test significant at $p \leq 0.05$, * at $p \leq 0.10$).

	Outlier Lakes Excluded		Outlier Lakes Included	
	Transformed (Square Root)	Untransformed	Transformed (Square Root)	Untransformed
Variable	P-Value	P-Value	P-Value	P-Value
DO	0.688	0.730	0.505	0.812
pH	0.727	0.613	0.770	0.930
Temp	0.877	0.837	0.810	0.730
LogChla	0.687	0.728	0.516	0.466
LogTN	0.266	0.295	0.083	0.199
LogCOND	0.320	0.386	0.141	0.258
LogTP	0.132	0.183	0.100*	0.201
LogSecchiDepth Ratio	0.006**	0.017**	0.001**	0.002**

All eight environmental variables were included for forward selection in CCA ordination. Table 3.5 outlines those variables that were significant given the four trial variations employed. Log (TP) was the initial variable selected in all trials, as determined by forward selection, and thus was chosen for transfer function development. Log (Secchi/Depth) was also significant under all variations in the data. Log (TN) was significant where outlier lakes were included and species data were square root transformed. Log (TN) was also significant in both trials when the

species data were not square root transformed. Log (Spec. Cond.) was significant in both ordinations where species data were square root transformed.

Table 3.5: Results of CCA using manual forward selection. *Ranking* indicates the order in which the environmental variables were selected throughout this analysis. 'x' indicates the inclusion of the selected variable. 'Proportion explained' is the amount of variation explained by the variable upon selection. Four variations of the CCA were performed, including and excluding outlier lake samples, using untransformed as well as square-root transformed species data. Shaded cells indicate significant ($p \leq 0.05$) environmental variables

Transformed (Square Root)	Outlier Lakes Excluded						Outlier Lakes Included					
Variable	P-Value	Proportion Explained	Ranking				P-Value	Proportion Explained	Ranking			
			Round a	b	c	d			Round a	b	c	d
DO	0.435	0.136	6	5	4	3	0.174	0.125	6	5	3	2
pH	0.673	0.120	8	7	6	4	0.731	0.093	8	7	6	5
Temp	0.739	0.117	7	6	5	5	0.276	0.101	7	6	5	4
LogChla	0.347	0.142	5	4	3	2	0.596	0.113	5	4	4	3
LogTN	0.126	0.161	2	3	2	1	0.002	0.172	2	2	1	x
LogCOND	0.005	0.198	4	2	1	x	0.020	0.146	4	3	2	1
LogTP	0.001	0.267	1	x	x	x	0.001	0.281	1	x	x	x
LogSecchi/Depth	0.003	0.240	3	1	x	x	0.001	0.225	3	1	x	x
Untransformed	Outlier Lakes Excluded						Outlier Lakes Included					
Variable	P-Value	Proportion Explained	Ranking				P-Value	Proportion Explained	Ranking			
			Round a	b	c	d			Round a	b	c	d
DO	0.597	0.196	7	6	4	5	0.656	0.161	7	6	5	4
pH	0.535	0.202	8	7	6	4	0.876	0.132	8	7	6	5
Temp	0.541	0.203	6	5	5	3	0.259	0.193	6	5	4	2
LogChla	0.487	0.210	5	4	3	2	0.364	0.187	4	3	3	3
LogTN	0.011	0.322	3	2	1	x	0.003	0.293	2	2	1	x
LogCOND	0.167	0.253	4	3	2	1	0.077	0.220	5	4	2	1
LogTP	0.001	0.415	1	x	x	x	0.001	0.408	1	x	x	x
LogSecchi/Depth	0.003	0.382	2	1	x	x	0.001	0.354	3	1	x	x

The CCA presented (Figure 3.3) includes outlier lakes and square root transformed species data. Four variables were chosen using forward selection in this CCA with Monte Carlo permutation tests (999 permutations), including: Log (TP), Log (Secchi/Depth), Log (TN) and Log (Spec. Cond.), as they were significant to the $p \leq 0.05$ level. The eigenvalues for the first and second axes were 0.309 and 0.322, respectively.

The first axis, which accounts for the majority of the variance within the data, was highly significant ($p=0.001$), as was the second axis ($p= 0.001$) (Ramstack et al. 1993). TP, TN, and Spec. Cond. were closely associated with the first (horizontal) axis in the dataset. The Secchi/Depth environmental variable follows closely with the second (vertical) axis. The TP arrow length and strong association with the first axis of the ordination diagram indicates that TP is one of the driving environmental variables within this system (Fig. 3.3 & 3.4).

The diatom taxa were well spread around the ordination axes with several species positively associated with the TP vector including species: *Stephanodiscus medius*, *Surirella ovalis, Fragilaria tenera, Cyclotella gamma,* and *C. meneghiniana* (corresponding species codes can be found in Table 3.2).

The association of TP and TN with the first axis suggests a strong role of trophic status within the system, as these macronutrient levels are a strong determining factor in lake trophic status (Ramstack et al. 1993). The Secchi/Depth environmental variable also has potential to be related to trophic state, as with increased nutrient levels there is potential for a dominance of planktonic algal populations, leading to a decrease in Secchi depth due to increased turbidity (McGowan et al. 2005). Those species and samples of higher trophic state (i.e., more eutrophic) are plotted to the top-right of the diagram, and those plotting to the bottom-left are more closely associated with a more oligotrophic state.

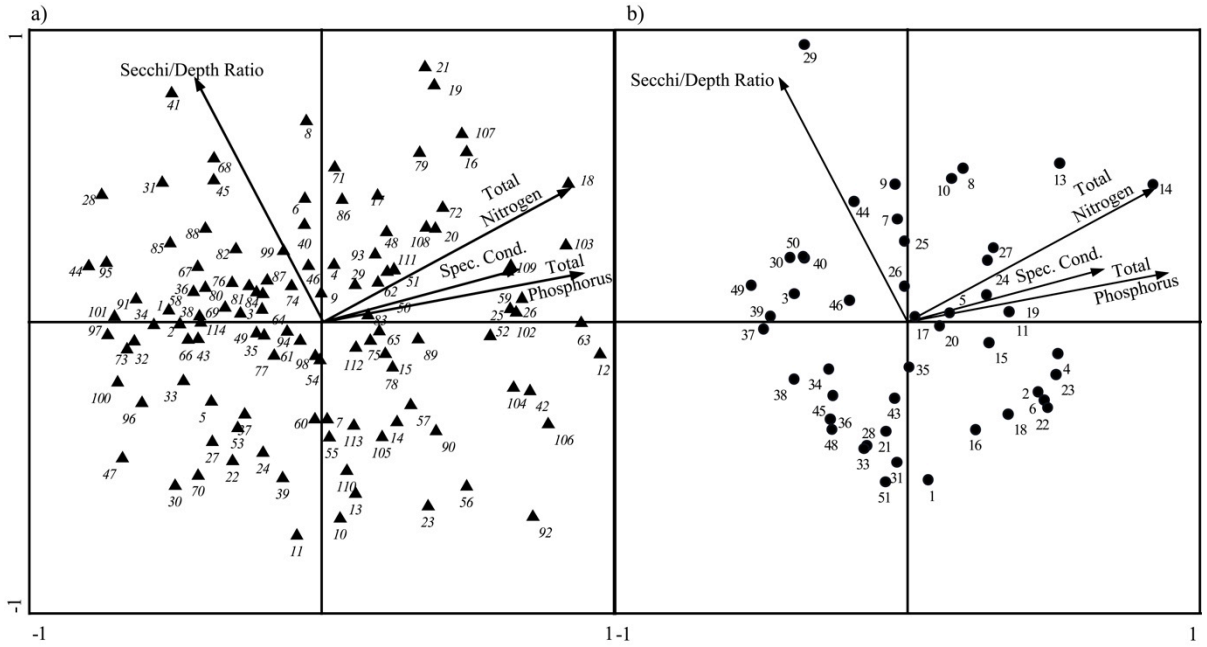


Figure 3.3: CCA ordination diagram of (a) species and significant environmental variables and (b) sample lakes and significant environmental variables (Axis 1 $\lambda = 0.309$, p=0.001; Axis 2 $\lambda = 0.322$, p= 0.001). Secchi/Depth ratio, TN and Spec. Cond. were all $Log_{10}(x+1)$ transformed, and TP was $Log_{10}(x)$ transformed. Species codes listed in Table 3.2. Species data including outlier lakes and square-root transformed.

3.4.2 Development of statistical models and Total Phosphorous transfer functions

Trials used to assess various TP reconstruction models are summarized in Tables 3.6 and Table 3.7. The various modeling scenarios include trials with (Table 3.7) and without (Table 3.6) outlier lakes included, as well as using either square root transformed or untransformed species data. Modelling techniques employed included: 1) Weighted averaging (WA), 2) Weighted Averaging – Partial Least Squares (WA-PLS), and 3) Partial Least Squares (PLS).

All models were assessed via their bootstrapped coefficient of determination (r^2_{boot}) and root mean square error of prediction (RMSEP). Through this analysis it was determined that the two component PLS model had a slightly higher $r^2_{boot.}$ in comparison to the one component model, WA (classical deshrinking) had slightly lower values and WA-PLS had the lowest. A comparison of RMSEP values reveals the same pattern of performance. Based on the minute differences between both the $r^2_{boot.}$ and RMSEP values of the WA and PLS models, both models were chosen to be applied to down core sediment samples for TP reconstruction.

Table 3.6: Comparison of WA, WA-PLS, and PLS models obtained with inverse vs. classical deshrinking and multiple components, with and without tolerance down-weighting, and with and without square root transformation of the species data, with all models excluding outlier lakes.

Comparison of WA models		r^2	$r^2_{boot.}$	RMSE	RMSEP	Avg. bias	Max. bias	Avg. $bias_{boot.}$	Max. $bias_{boot.}$
Untransformed percentage species data:									
Classical deshrinking:	WA	0.78148	0.5034	0.22573	0.33799	-6.846e-016	0.40116	-6.352e-004	-0.3355
	Watol	0.76832	0.4603	0.23441	0.37259	5.921e-0.16	0.45698	0.009649	0.5109
Inverse deshrinking:	WA	0.78148	0.4938	0.19955	0.33067	-3.516e-016	0.21329	1.0759e-004	0.4278
	Watol	0.76832	0.4496	0.20547	0.36613	5.058e-016	0.2566	0.0099352	0.5708
Square root transformed percentage species data:									
Classical deshrinking:	WA	0.74448	0.4558	0.25008	0.34266	-6.908e-016	0.48878	0.0050409	0.5114
	Watol	0.67887	0.4343	0.29360	0.39005	7.401e-017	0.50732	0.0105660	0.5852
Inverse deshrinking:	WA	0.74448	0.4459	0.21578	0.33938	-4.688e-016	0.30262	0.0028147	0.5731
	Watol	0.67887	0.4248	0.24190	0.37778	-1.234e-017	0.39025	0.0088209	0.6401
Comparison of WA-PLS models		r^2	$r^2_{boot.}$	RMSE	RMSEP	Avg. bias	Max. bias	Avg. $bias_{boot.}$	Max. $bias_{boot.}$
Untransformed percentage species data:									
Component 1		0.78148	0.4821	0.19955	0.33504	-3.361e-016	0.213290	-0.0038273	0.4259
Component 2		0.89134	0.4515	0.14072	0.36413	7.833e-016	0.048768	0.0089279	0.3646
Component 3		0.93775	0.4055	0.10650	0.39537	-1.904e-015	0.103510	0.003849	0.3714
Square root transformed percentage species data:									
Component 1		0.74448	0.4418	0.21596	0.34158	-8.711e-004	0.2899	0.0043479	0.5672
Component 2		0.88397	0.3950	0.14544	0.36445	0.0028434	0.11014	0.017109	0.5527
Component 3		0.94278	0.3635	0.10214	0.38266	-2.286e-004	0.081672	0.02058	0.5743
Comparison of PLS models		r^2	$r^2_{boot.}$	RMSE	RMSEP	Avg. bias	Max. bias	Avg. $bias_{boot.}$	Max. $bias_{boot.}$
Untransformed percentage species data:									
Component 1		0.68916	0.5380	0.238	0.31664	-3.084e-016	0.23004	-0.017525	0.3008
Component 2		0.75191	0.5312	0.21262	0.31982	-3.022e-016	0.19894	-0.013753	0.2625
Component 3		0.080284	0.5057	0.18955	0.3439	-3.038e-016	0.16411	-0.030943	0.2415
Square root transformed percentage species data:									
Component 1		0.72817	0.5543	0.22256	0.30855	-3.176e-016	0.19325	-0.0066245	0.4085
Component 2		0.83533	0.5620	0.17323	0.30337	-2.868e-016	0.096596	-0.0043489	0.3217
Component 3		0.89952	0.5452	0.13532	0.31249	-2.729e-016	0.11003	-0.0058279	0.3156

Table 3.7: Comparison of WA, WA-PLS, and PLS models obtained with inverse *v* classical deshrinking and multiple components, with and without tolerance down-weighting, and with and without square root transformation of the species data, with models including outlier lakes.

Comparison of WA models		r^2	$r^2_{boot.}$	RMSE	RMSEP	Avg. bias	Max. bias	Avg. $bias_{boot.}$	Max. $bias_{boot.}$
Untransformed percentage species data:									
Classical deshrinking:	WA	0.7069	0.47312	0.29439	0.37908	7.5785e-016	0.38233	0.0070322	0.66438
	Watol	0.7227	0.42115	0.28319	0.40474	-7.241e-017	0.34543	0.042524	0.87013
Inverse deshrinking:	WA	0.7069	0.45983	0.24751	0.35559	5.792e-016	0.50124	0.0045985	0.73739
	Watol	0.7227	0.40933	0.24075	0.3864	3.379e-017	0.47145	0.034049	0.89031
Square root transformed percentage species data:									
Classical deshrinking:	WA	0.67023	0.45374	0.32069	0.37572	-1.878e-015	0.39558	0.010347	0.70504
	Watol	0.68175	0.3919	0.31236	0.42637	5.020e-016	0.3493	0.062298	0.9703
Inverse deshrinking:	WA	0.67023	0.43681	0.26254	0.35825	-1.067e-015	0.54143	0.0047697	0.77221
	Watol	0.68175	0.37958	0.25791	0.40187	3.017e-016	0.51966	0.047412	0.96933

Comparison of WA-PLS models	r^2	$r^2_{boot.}$	RMSE	RMSEP	Avg. bias	Max. bias	Avg. $bias_{boot.}$	Max. $bias_{boot.}$
Untransformed percentage species data:								
Component 1	0.7069	0.45173	0.24751	0.35882	3.669e-016	0.50124	-0.0010094	0.73973
Component 2	0.82793	0.39569	0.18964	0.40445	-9.340e-016	0.26694	0.0068952	0.73494
Component 3	0.88837	0.34901	0.15275	0.43812	2.529e-015	0.21632	-0.0015834	0.77857
Square root transformed percentage species data:								
Component 1	0.67023	0.44089	0.26388	0.35710	-0.006856	0.50763	0.0013654	0.76185
Component 2	0.80250	0.37488	0.20347	0.39696	0.001266	0.26764	0.010971	0.76677
Component 3	0.87636	0.31839	0.16100	0.43119	-0.006085	0.20451	0.0052138	0.81748

Comparison of PLS models	r^2	$r^2_{boot.}$	RMSE	RMSEP	Avg. bias	Max. bias	Avg. $bias_{boot.}$	Max. $bias_{boot.}$
Untransformed percentage species data:								
Component 1	0.50732	0.35734	0.3209	0.38548	-9.171e-017	0.64461	-0.011305	0.75313
Component 2	0.64262	0.36583	0.27331	0.40084	-4.827e-017	0.46841	0.0031365	0.76807
Component 3	0.72943	0.37695	0.23781	0.40690	-6.758e-017	0.35574	-0.0051911	0.76615
Square root transformed percentage species data:								
Component 1	0.63350	0.49416	0.27677	0.33728	-2.896e-017	0.60366	-0.0091059	0.74015
Component 2	0.74468	0.48403	0.23101	0.35019	-7.241e-018	0.42397	-0.0046465	0.72810
Component 3	0.80372	0.44919	0.20255	0.36892	2.172e-017	0.42306	-0.0070296	0.78389

Plots for assessment of model performance, through comparison of observed and inferred TP values, are represented as residual diagrams in Figure 3.4 for WA and Figure 3.5 for PLS (outlier lakes included and square-root transformed species data). In Figure 3.4, the predicted TP values are well correlated with observed TP values in both WA classical and inverse deshrinking scenarios, as represented by their approximation and spread along the 1:1 line. The first and

second components of the PLS model show little variation with respect to their spread along the 1:1 in Figure 3.5, comparing observed to inferred TP values.

An assessment of residual (observed-inferred) values to observed values for WA models indicates no significant outliers present within the data and the spread of the data across the horizontal axis shows a greater range in the inverse deshrinking scenario. For the PLS model there appears to be no substantial difference in the spread of residuals across the horizontal axis. There is, however, a prominent trend in the residuals, suggesting an overestimation and underestimation of values at the lower and upper limits, respectively. This trend is common in diatom transfer functions (ter Braak & Juggins 1993).

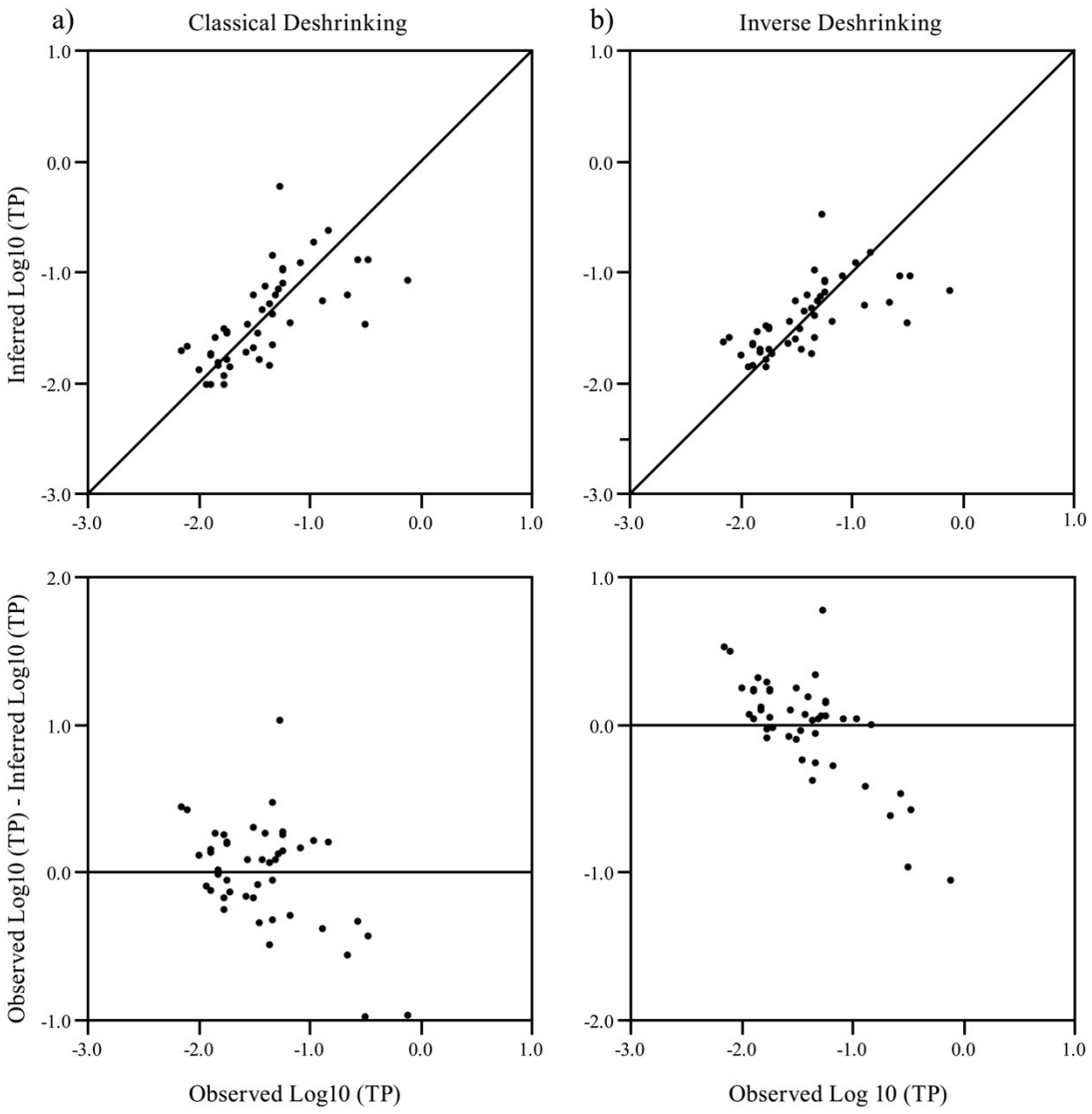


Figure 3.4: Plots of observed *v* diatom- inferred total phosphorus concentration (DI-TP), and observed *v* residual TP (observed TP - DI-TP), based on weighted-averaging regression and calibration models using (a) classical deshrinking and (b) inverse deshrinking. Outlier lakes included and species data square-root transformed.

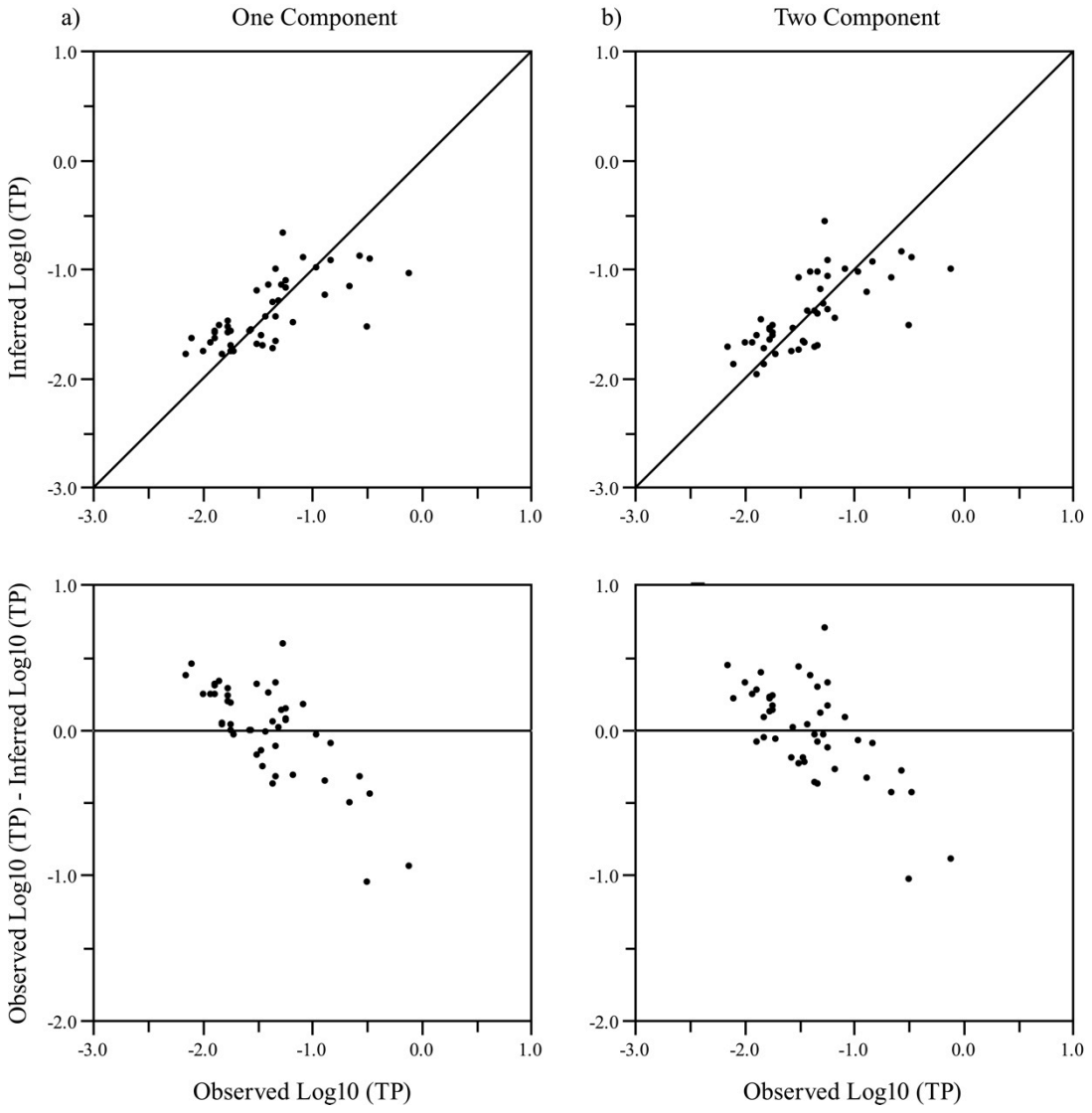


Figure 3.5: Plots of observed *v* diatom- inferred total phosphorus concentration (DI-TP), and observed *v* residual TP (observed TP - DI-TP), based on partial least squares regression and calibration models using (a) one component and (b) two component. Outlier lakes included and species data square-root transformed.

3.5 Discussion

3.5.1 Surface sediment assemblage in relation to trophic state

Although the water quality of the lakes varied substantially, with TP differing by as much as two orders of magnitude, there were very few lakes sampled within the oligotrophic range. Diatom species assemblages reflected this bias, with few strictly oligotrophic planktonic taxa present. Benthic/epiphytic species prevailed in the shallower lakes, as seen in the appearance of the eutrophic benthic species *Amphora copulata* in 34 of the lakes sampled (van Dam et al.

1994). It is appropriate that *A. copulata* has the highest calculated N2, as it has the greatest number of occurrences within the dataset.

Where planktonic species were prevalent, together they made up a significant portion of the assemblage, anywhere from 5 to 81%. In past studies much weight was placed on the ratio of planktonic to non-planktonic species, as it was assumed that the presence of planktonic species was indicative of a more eutrophied lake state (Battarbee 1986). However, not all planktonic species are indicative of such a lake state. *Cyclotella comensis* was found in 4 of the study lakes, and was dominant within both Clear Lake and Lake Katherine. Both lakes have a more meso-oligotrophic state. This, in combination with the calculated TP optima within the mesotrophic range for *C. comensis*, demonstrates the lack of usefulness in employing solely a planktonic to non-planktonic ratio analysis.

Almost all species of the genus *Stephanodiscus* tend to be associated with increased nutrient levels, including *S. hantzschii, S. medius, S. minutulus,* and *S. parvus* (Bennion et al. 2004; Walker et al. 1993; Yang et al. 1993). This is most evident in Figure 3.2a as these taxa are grouped in the upper right hand quadrant, where TP optima and measured sample lake TP values are highest. *S. niagarae* does not group with these other taxa but it still falls within the eutrophic range (41.06 µg L^{-1}). Other planktonic species that are generally considered typical of more productive lakes fall within the same eutrophic range of calculated TP optima: *Aulacoseira granulata, A. ambigua*, and *Tabellaria flocculosa*, (Bennion et al. 2004; Hall & Smol 1992; Rosén 1981).

Epiphytic species were present in the majority of sample lakes. Due to their association with macrophyte populations it is often assumed that they are more indicative of oligotrophic lake conditions (Bennion et al. 2001). The calculated TP optima for many of these species falls within the mesotrophic range (Table 3.2). However, there is a large number of unknowns associated with the preference of many of these species as there is relatively little known of their ecology and they are found within lakes of varying trophic state. Open water TP levels may not have as great an influence on these taxa as other factors, such as light availability and their association with macrophyte populations (Bennion et al. 2005). This may hinder the determination of the relationship between diatoms and trophic status (Bennion et al. 2005; Chen et al. 2008). The presence of some benthic species, including *Pseudostaurosira parasitica*, *Staurosirella pinnata*, *Fragilaria brevistriata* and *F. construens*, within the dataset may also adversely affect the model development, as these species are considered poor indicators of lake trophic state (Bennion et al. 2005).

Overall, the dominance of either non-planktonic species, which require adequate light penetration in the water column, and planktonic species, which remain suspended in the water column, are useful in determining lake trophic state, provided that the ecological preference of the individual taxa in question are considered within the overall scenario (Battarbee 1986). Where quantitative methods fall short, qualitative interpretations of relationships between diatoms and water quality are both valid and ecologically sound (Anderson 1997).

3.5.2 Species TP optima regional comparison

The calculated TP optima of the diatoms included in this dataset left few species within the oligotrophic range. This is because oligotrophic lakes are rare within the Little Saskatchewan Watershed. The trophic preferences I noted for many taxa differed from those established in other North American regions. As demonstrated in Figure 3.6, the calculated TP optima for the majority of taxa in my dataset are much higher than those calculated for the same species in other regions. The optima provided in Ramstack et al. (2003) are most closely akin to the optima calculated for RMNP. This makes good intuitive sense since their study was completed in Minnesota, USA. Due to proximity, their study lakes are most similar in terms of water chemistry, geographical charateristics and anthropogenic influences. The other studies were mostly completed in regions dominated by more oligotrophic, soft-water lakes.

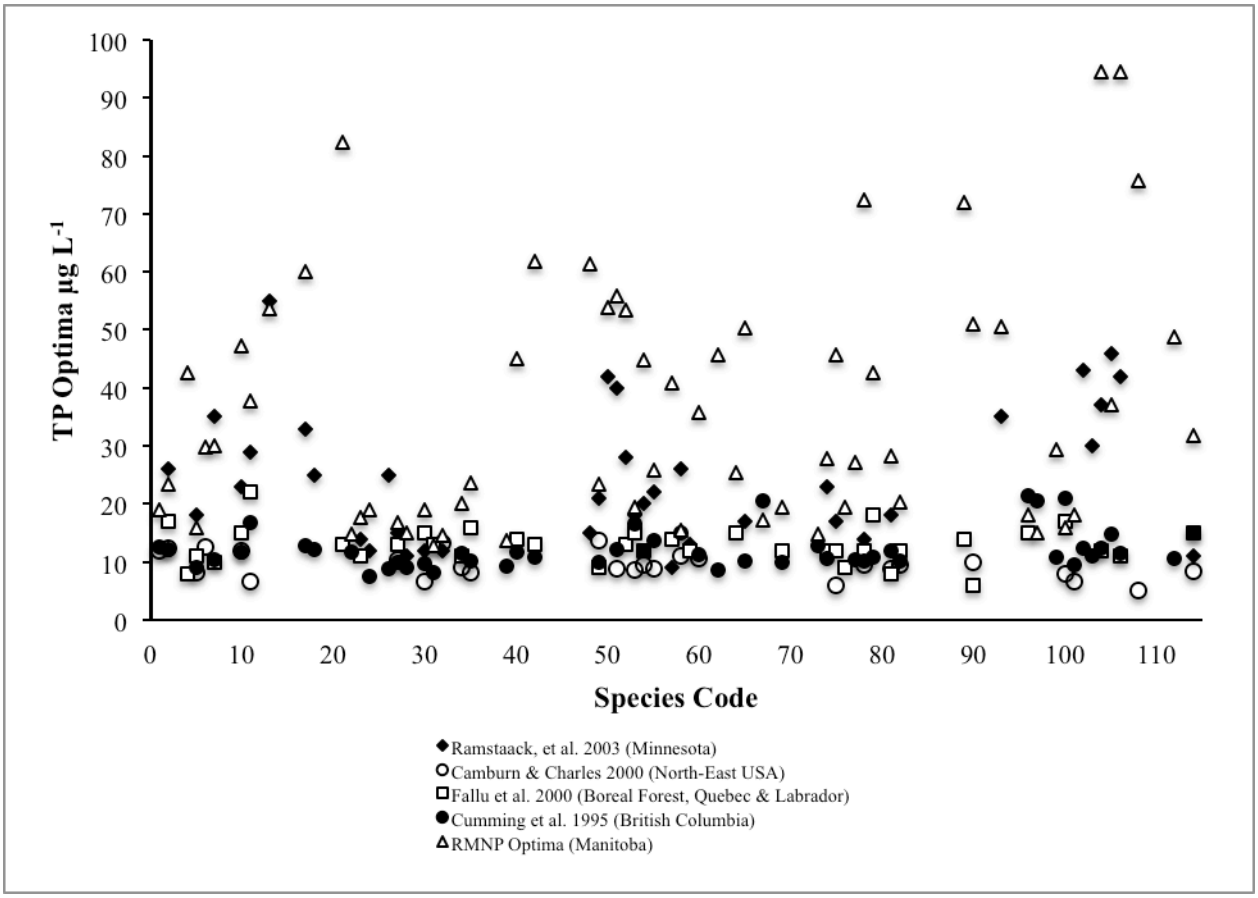


Figure 3.6: Calculated TP optima for diatom taxa occurring in various regions of North America; horizontal axis arranged by species codes provided in Table 3.2 and calculated TP optima on vertical axis in µg L^{-1}.

The principal reason for the higher TP optima calculated for RMNP lakes is therefore, a different sampling bias in my dataset. Lakes in the RMNP region tend to be within the mesotrophic and eutrophic ranges, while oligotrophic lakes are rare. This sampling bias is depicted in Figure 3.7. Time constraints on field methods as well as the inclusion of lakes only occurring within the Little Saskatchewan watershed meant that the range of TP sampled was more heavily weighted on the meso- and eutrophic range of values. The lack of samples within the oligotrophic range, or more specifically, the abundance of samples within the eutrophic range, contributed to the higher calculated TP optima, in comparison with calculated values from previous studies in North America. The presence of this sampling bias should be considered where the calculated TP optima of taxa differs from the general consensus in the literature.

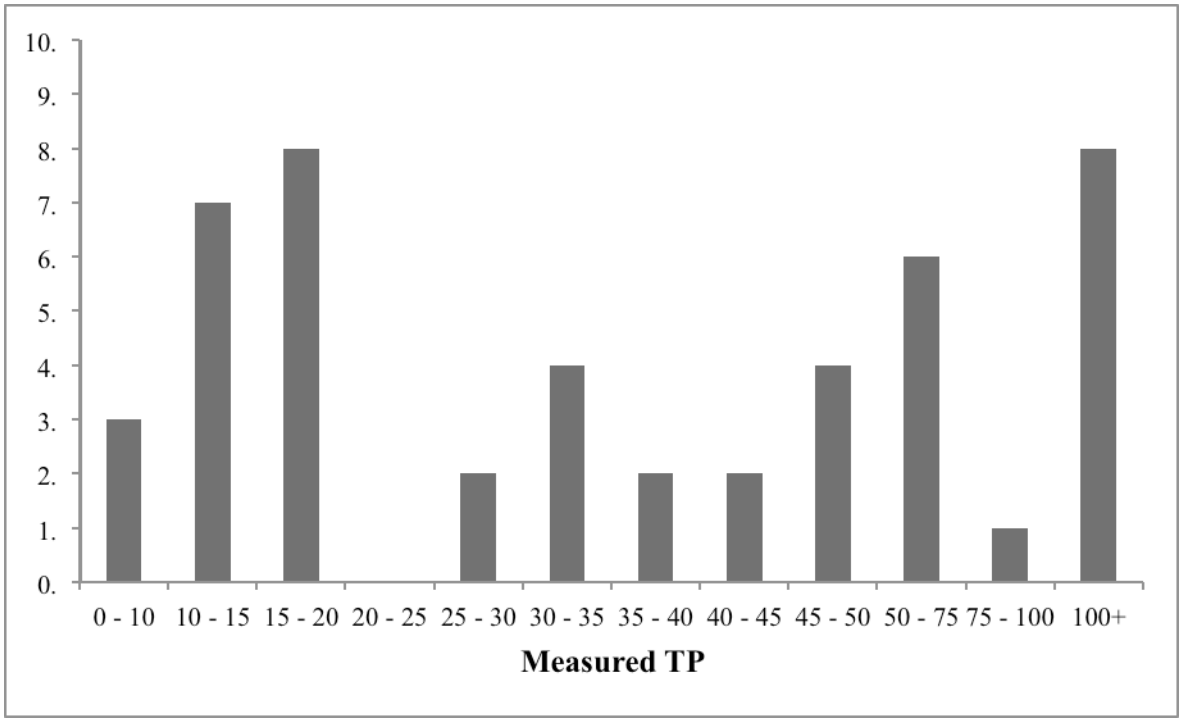


Figure 3.7: Distribution histogram depicting the number of lakes sampled within each range of TP values in in µg L^{-1}.

3.5.3 Total Phosphorus transfer function development

Throughout the data screening process 11 lakes were deemed to be outliers based on preliminary DCA and PCA analysis of diatom species and lake environmental data, respectively. These lakes (see supplementary data available in Appendix 2) were not atypical of the region being examined and although they were considered outliers from a statistical standpoint, from a limnological perspective they were not. The lakes may have been deemed outliers potentially due to the inconsistency between diatom populations, which incorporate limnological variables over a longer period of time, and the relatively instantaneous TP measurement (Hall & Smol 1992).

Subsequent analyses were performed with both the inclusion and the exclusion of the outlier lakes, as those lakes determined to be outliers contribute greatly to the range in TP values (Hall & Smol 1992). It is desirable to maintain the longest gradient of TP possible so as to avoid unnecessary reduction of the gradient length. TP transfer functions that comprise a smaller number of sample lakes can possess weaker predictability and also can have increased error of prediction (Kauppila et al. 2002; Reavie et al. 1995). As you increase the number of lakes included in the dataset there is an expected increase in the number of taxa therefore included in the transfer function (Wilson et al. 1996). The inclusion of as many species as possible within

the predictive models is beneficial in its capacity to improve fit and increase the number of analogues between modern and fossil diatom assemblages (Wilson et al. 1996). As the overall goal is to maintain the longest TP gradient as well as the inclusion of as many taxa as possible, the outlier lakes were included in the analyses and were also included in the dataset for the final model selected for the DI-TP transfer function.

The models developed in this chapter will facilitate the reconstruction of TP and past trophic state for RMNP lakes (Chapters 4 & 5). This will facilitate a better assessment of the lakes' natural state and the extent of any human impact. The models will be key to determining whether eutrophication is occurring and at what rate. In short, these models and their application to down-core fossil analyses will facilitate a detailed objective assessment of the lakes' ecological integrity. These models will allow for the reconstruction of TP within the lake systems of RMNP based on sedimentary diatom populations. This will facilitate the reconstruction of long term lake water quality records, providing a history of lake trophic status. DI-TP values provide better estimates of historical TP than the amount of phosphorus trapped within the sediments themselves (Bennion et al. 1995).

3.6 Summary

- Analyses of diatoms amongst 47 lakes in the RMNP region revealed that species distributions were most strongly correlated with TP.
- Through Canonical Correspondence Analysis, Total Phosphorus was identified as the environmental variable most capable of being reconstructed.
- Modelling trials facilitated the development of diatom-TP transfer functions using WA, PLS, and WA-PLS regression.
- Preliminary modelling to test model performance determined that both WA and PLS models had sufficient performance for the reconstruction of TP based on sedimentary diatom assemblages from lakes within the Little Saskatchewan watershed.
- Contrary to theory and expectation linear PLS models yielded higher r^2_{boot} and lower RMSEP (commonly used measures of model performance) than unimodal WA and WA-PLS models. Nevertheless, differences between the best WA and PLS models were slight and no "best model" clearly emerged.
- The TP optima derived in this study are higher than those reported elsewhere, owing to different sampling biases among study regions.

- The models developed here will facilitate a detailed assessment of ecological integrity in RMNP lakes.

Chapter 4. Diatoms in sediments of Clear Lake, RMNP: A paleoenvironmental assessment of ecological integrity, and natural and anthropogenic change.

4.1 Background information

The assessment of a lake's ecological status is essential as a benchmark for management and provides a comprehensive view of its natural state. Clear Lake, Riding Mountain National Park of Canada (RMNP), is located in south-western Manitoba, and requires such an assessment, in keeping with the mission of Parks Canada (2009) to maintain and restore the ecological integrity of Park Canada's ecosystems.

In such locations where accurate long-term water quality data are lacking, the use of diatom microfossils provides an effective proxy, yielding information regarding past nutrient and ecosystem change (Reavie et al. 2006; Vance 1997). Diatom inferred total phosphorus (DI-TP) values derived from paleolimnological study provides an effective means of establishing pre-industrial or natural limnological conditions (Dixit & Smol 1994).

No such reconstruction has previously been performed on any lake within RMNP. Here, I report the results of the first DI-TP reconstruction for RMNP based on sediment cores for Clear Lake, Manitoba. The DI-TP inferences provide a quantitative look at the past state and an extensive history of Clear Lake.

4.2 Study Site:

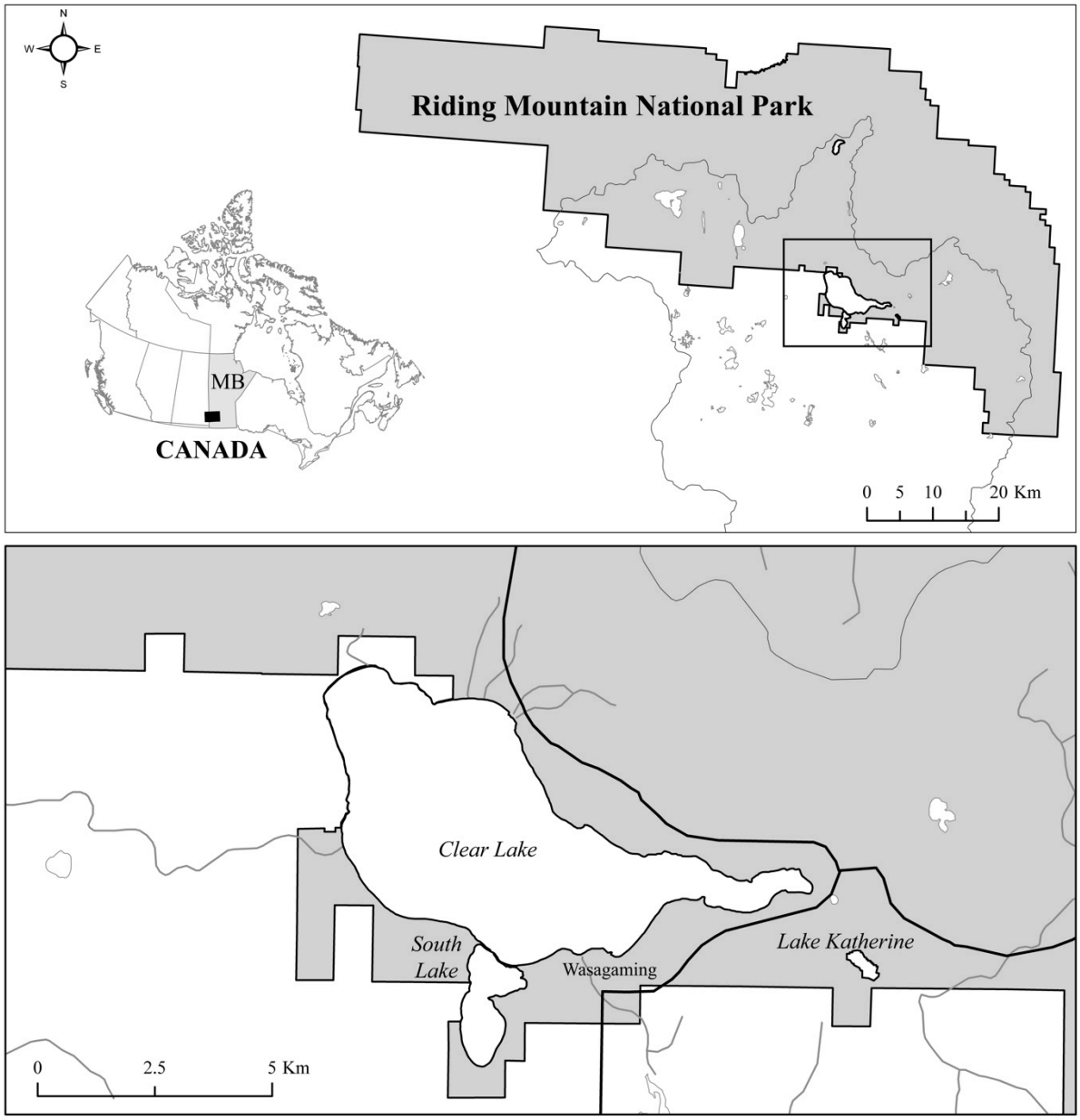
RMNP covers a total area of 2,973 km^2 with 96 km^2 (3.2%) consisting of aquatic habitat (Parks Canada 2011; Saunders 1979). Boreal forest and fescue grasslands dominate the landscape (Parks Canada 2011; Saunders 1979). Freshwater bodies include marshes, bogs, swamps, as well as both shallow and deep lakes (Saunders 1979). Four of these freshwater bodies are of particular management concern: Clear Lake, South Lake, Lake Katherine and Moon Lake. These four lakes are all currently accessible to RMNP visitors and have also been affected by diverse human influences. The location of Clear Lake within RMNP is indicated in Figure 4.1.

Over the years RMNP has been subjected to many human influences, including First Nations habitation in the area following glacier retreat 10,000 YBP (Parks Canada 2011). Settlers from non-aboriginal populations began moving into the area in the 1880s, shortly after the Canadian Pacific Rail line reached Brandon, MB in 1881 (Parks Canada 2010). At this time there were approximately 10 large logging and milling operations within RMNP, an activity which peaked

at the end of the 19th century, coinciding with two major forest fires in 1890 (Parks Canada and Wilderness Society 2004). Due to fire and logging activities RMNP forests consist of mixed age stands (Saunders 1979). In 1895, the area that was to become RMNP was established as a Timber Reserve, and was designated as a Forest Reserve in 1906 (Parks Canada and Wilderness Society 2004). This Forest Reserve was then given National Park status in 1930, opening officially in 1933, with one mill operation continuing to log within the Park until 1949 (Parks Canada 2010; Parks Canada and Wilderness Society 2004). Due to fuel demands during WWII (1939-1945), RMNP was again used as a mainstay in wood supply (Parks Canada 2010). In 1986, RMNP was designated, along with 15 surrounding rural municipalities, as the Riding Mountain Biosphere Reserve, aimed at the conservation of biodiversity and sustainable resource use (Parks Canada 2011; Riding Mountain Biosphere Reserve 2012).

Clear Lake is the largest lake in the park having a maximum depth of 34.2 metres (Figure 4.2) and covering 29.5 km^2 (11.4 $miles^2$) (Saunders 1979). The Wasagaming town site is located on the shore of Clear Lake (Figure 4.3). Common recreation on Clear Lake includes boating, swimming, and shoreline leisure activities. Land use surrounding Clear Lake includes summer camps and a golf course (Figure 4.3). The Keeseekoowenin Ojibway First Nation owns lands along the north and west shores of Clear Lake. Much of the remaining area is protected through Park management (Parks Canada 2011). There are very few macrophytes present within Clear Lake, most likely due to its depth, long fetch and wave action.

Clear Lake is the deepest lake in RMNP and is partly groundwater-fed (Parks Canada 2011). It is connected to the more eutrophic waters of Ominik Marsh and Octopus Creek, and is directly adjacent to the eutrophic South Lake (Scott & Sellers 2003). Relative to other lakes in RMNP Clear Lake is a bit anomolous with regards to its clear waters and oligotrophic lake state, despite high anthropogenic use, and exposure to nutrients from varying sources. It is for these reasons that it is of particular interest from a management perspective.



Clear Lake - Riding Mountain National Park of Canada

Sample Lakes
Rivers and Streams
Little Saskatchewan Watershed Boundary
Park Boundary

Prepared by: Carrie White
May 2012
Projection: Canada Lambert Conformal Conic
GCS - NAD 1983
Data Source: Park's Canada

Figure 4.1: Clear Lake and its location within RMNP, Manitoba, Canada

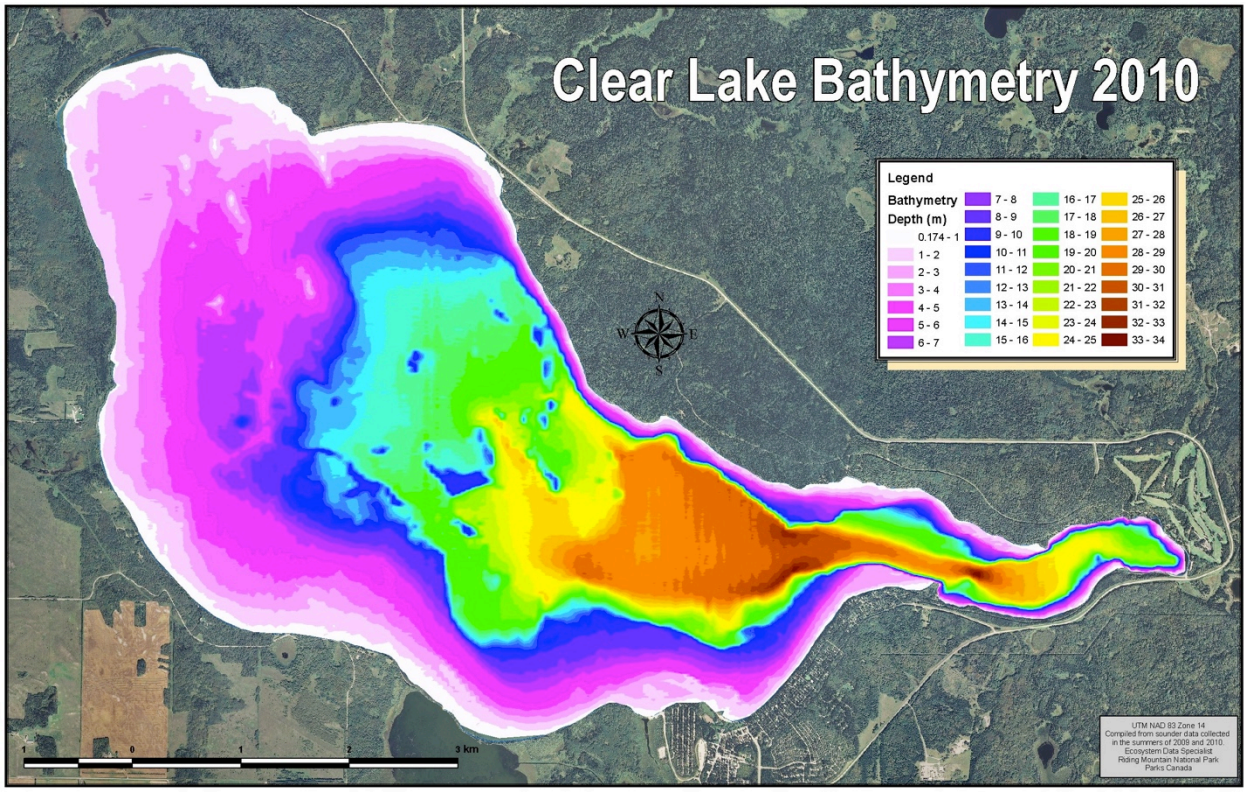


Figure 4.2: Clear Lake bathymetry. Produced by Sean Frey for Parks Canada (2010) using data collected in 2010, reproduced with permission.

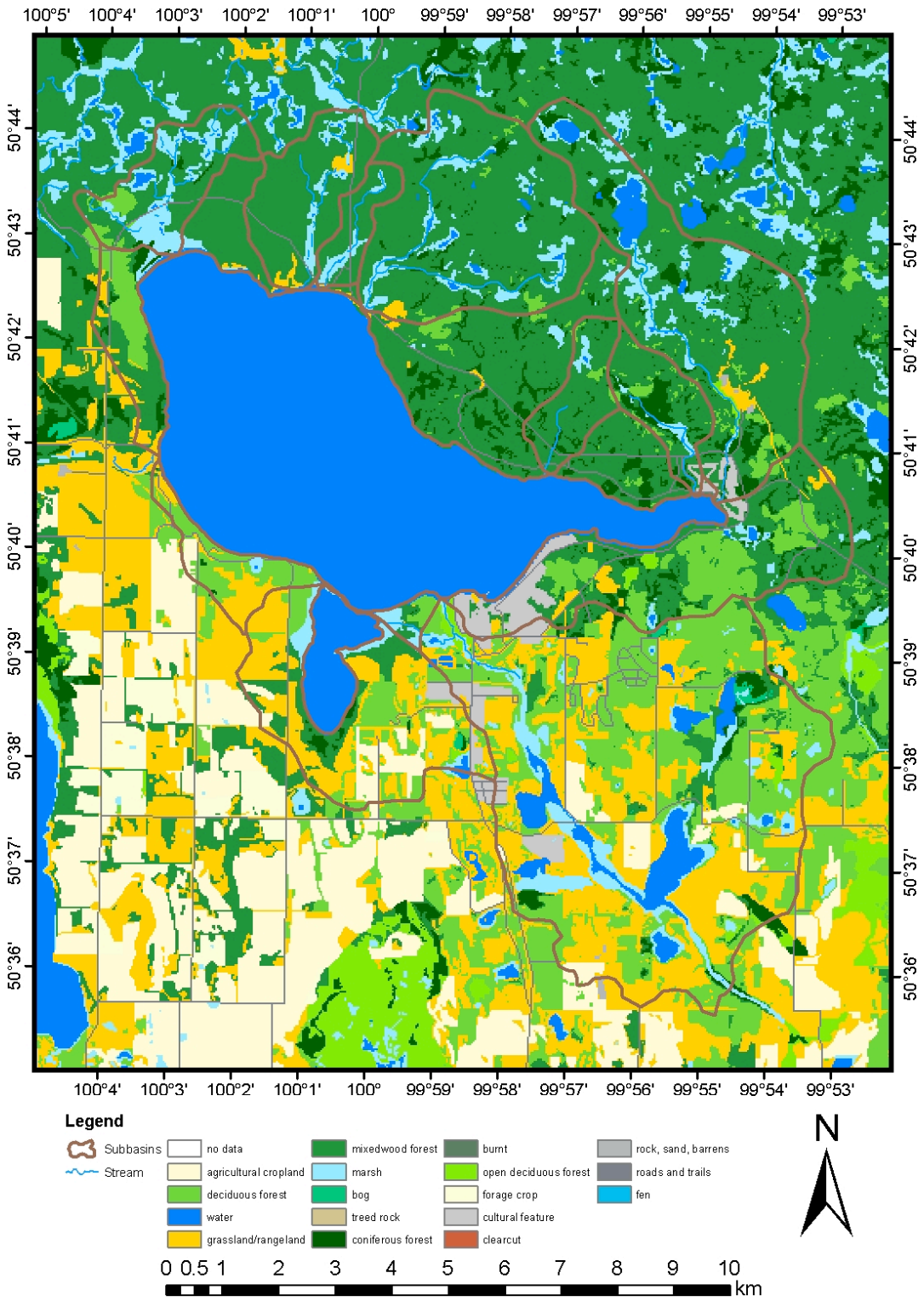


Figure 4.3: Map of land use surrounding Clear Lake. Produced by Sean Frey for Parks Canada (2010), reproduced with permission.

4.3 Methods

4.3.1 Short and long core collection, diatom analysis

A 44 cm long short core was collected from Clear Lake using a Glew gravity corer with a 3-inch Lexan tube. The sediment was then sub-sectioned into one-centimeter intervals and stored in Whirlpaks® and later processed using techniques described in Section 3.3.2. Diatoms were initially enumerated at 4 cm intervals to detect major changes in diatom populations. Samples were later enumerated at a 1 cm sample resolution.

A 2.33 m long core was obtained using a Reasoner percussion coring system in January 2010 (Reasoner 1986). The core was collected at the same site as the Glew core, and close to the site of maximum depth in Clear Lake. The PVC pipe in which the percussion core samples were recovered was sectioned into transportable lengths, capped and transported to the Parks Canada Western Service Centre in Vancouver, BC where samples were stored at 4°C. Long core samples were then sub-sampled at 2 cm intervals and stored in Whirlpaks® and then transported to UBC Okanagan for processing and analysis. Samples were enumerated at 8 cm intervals.

For both cores at least 500 diatom valves were enumerated per sample, following a transect across the microscope slide at 1000× using oil emersion and Differential Interference Constrast (DIC) microscopy.

4.3.2 Core sample dating techniques

The short core was dated using ^{210}Pb standard radiometric techniques (Appleby 2002). A total of 16 sediment sub-samples were analysed for ^{210}Pb at three-centimeter intervals (minus a sample at 24 cm depth) along the length of the 44 cm core. AMS Radiocarbon dates were submitted for sediment samples at 31-32, 141-142, and 232-233 cm from the long core.

4.3.3 Numerical methods

Diatom counts were converted to relative abundance values for each level. Diatom stratigraphies were constructed for each core using the program TILIA v. 1.7.16 (Grimm 2011). Species data was then square root transformed to down-weight the effect of abundant taxa within the model (Vermaire & Gregory-Eaves 2008). Major zones of diatom species composition were delineated based on inspection of the stratigraphically CONstrained Incremental Sums-of-Squares cluster analysis (CONISS) diagram and species distribution patterns throughout the length of the cores (Grimm 1987; Bennion et al. 2001; Kauppila et al. 2002).

Transfer functions developed in Chapter 3 were then applied to down-core diatom species assemblages using the software program C2 v. 1.6.8, to yield a total phosphorus reconstruction (Juggins 2004; Juggins 2010; Smol & Cumming 2000).

4.4 Results and discussion

4.4.1 Diatom analysis of long core sediments

Clear Lake long core sediments were collected to a depth of 243 cm and analysed for diatoms at an 8 cm sample resolution. CONISS cluster analysis was employed to identify major zones within the long core (Grimm 1987). Four zones were identified, as shown in Figure 4.4. The division of the long core species assemblages based on CONISS is potentially weak, as CONISS is only able to divide the diatom stratigraphy into three or more groups where the sample size is large. Zones (L1, L2, L3, L4) have been identified through a combination of CONISS cluster analysis and sample examination.

Diatoms can be assigned to ecological groups in accordance with their general habitat preference. Some species live suspended in the water column; others are periphytic, growing attached to the surface of submerged objects, such as macrophytes or rocks (Lowe 1974). Changes in the relative abundance of these two groups can be used to assess lake state qualitatively.

Zone L1 extends over the longest portion of the core, encompassing 244 to 142 cm depth. At the bottommost portion of the core, 231-232 cm depth, carbon dates yielded an approximate age of 2870 ±30 ^{14}C yr Before Present (BP). At a shallower depth, 141-142 cm, carbon dates were 1990 ±30 ^{14}C yr BP. *Aulacoseira ambigua* is present in large quantities within this zone; together with *Fragilaria brevistriata* they account for as much as 75% of the relative species abundance. *Aulacoseira ambigua* is commonly regarded as an indicator of a more eutrophic lake state prior to European settlement of North America (Rosén 1981; Bennion et al. 2004; Stoermer 1977; Yang et al. 1993). *Staurosirella pinnata* and *Stephanodiscus niagarae* vary in abundance from 10-20% throughout this zone. *Staurosirella pinnata* is usually found in abundance in the pre-settlement period of small lakes in central North America. As a benthic species its presence can often reflect low nutrient levels and clear waters that are associated with pre-settlement lake conditions (Sayer et al. 1999; Stoermer 1977). *Stephanodiscus niagarae* is described as a planktonic taxon typical of eutrophic conditions, but it has also been identified as an oligo-mesotrophic species (Kling 1998; Yang et al. 1993). Other species also present, at smaller percentages (<10%), include *Amphora thumensis*, *Stephanodiscus hantzschii*, *Tabellaria*

flocculosa, *Martyana martyii*, and *Cyclotella bodanica*. In Zone L1, *Fragilaria construens* and *Cymbellafalsa diluviana* both peak in relative abundance. *Fragilaria crotonensis* is scarce in these sediments, save for the sample at 186 cm depth. *Amphora pediculus* peaks at the boundary between Zones L1 and L2. *Fragilaria brevistriata*, also abundant in L1, is an epiphytic species associated with the presence of macrophyte populations, but has been found in waters ranging from oligotrophic to eutrophic (Krammer and Lange-Bertalot 1997). Overall, L1 is considered to be of mesotrophic lake state.

Zone L2 has a short range from 142 to 108 cm depth. *Aulacoseira ambigua* remains abundant within this zone while species such as *Cyclotella bodanica* and *Fragilaria crotonensis* begin to increase in abundance. *C. bodanica* is generally considered an indicator of oligo-mesotrophic lake status (Wunsam et al. 1995; Yang et al. 1993). However, some publications classify it as being strictly oligotrophic, such as Rosén (1981) and Schuette and Bailey (1980). *F. crotonensis* may also be abundant in mesotrophic waters (Patrick & Reimer 1966). In L2 *F. brevistriata* is reduced significantly. *Stephanodiscus niagarae* remain at a constant abundance throughout L2, while *Martyana martyii* increases slightly. *M. martyii* prefers mesotrophic waters. Its abundance, together with the decreased abundance of many epiphytic species may signify a change in trophic state, from mesotrophic to more oligotrophic, and/or a reduction in water clarity (Patrick & Reimer 1966).

Zone L3 ranges from 108 to 62 cm and is defined by the greatly reduced abundance of *Fragilaria crotonensis*, the large variations in *Aulacoseira ambigua* abundance and an increase in the relative abundance of *Cyclotella bodanica* to its peak of 25%. *C. bodanica* is a planktonic and oligo-mesotrophic taxon (Wunsam et al. 1995; Yang et al. 1993). Several non-planktonic are declining in this zone, such as *F. brevistriata*, *F. construens*, *S. pinnata, Amphora pediculus,* and *M. martyii*, many of which possess unclear lake trophic preferences, ranging from oligotrophic to eutrophic conditions. Their reduction in combination with the increase in euplanktonic species, *S. minutulus* and *S. medius,* may be associated with a decline in water quality and a movement towards urbanization (Walker et al. 1993). *Stephanodiscus hantzschii* dissappeared over the course of Zone 3. *Epithemia turgida* is present at its greatest abundance within L3. The shift within the sedimentary diatom assemblages towards a dominance of diatoms that prefer nutrient rich waters and the disappearance of taxa that are epiphytic suggests a movement towards a more eutrophied lake state (Bennion et al. 2001).

Zone L4 encompasses the topmost sediments from 62 cm to a depth of 10 cm. The uppermost 10 cm were compromised during sampling and were not analysed for diatoms. The

approximate carbon date at 31-32 cm is 1010 ±30 ^{14}C yr BP. Many changes occur within these sediments, with most occurring at the very surface sample. *F. crotonensis* dramatically increases (60%) proportionately to the decrease in *A. ambigua*. This marks the change from pre-settlement diatoms to taxa associated with anthropogenic impacts. The non-planktonic taxa that were on the decline in L3 have disappeared in this Zone, including: *A. pediculus, M. martyii*, *F. brevistriata*, and *F. construens*. Several *Stephanodiscus* spp. are present at an abundance between 5 and 10% including: *S. hantzschii*, *S. minutulus*, and *S. niagarae*. Among those species completely absent from L4 is *Fragilaria construens*. *Achnanthes minutissima* and *Cyclotella comensis* only occur within L4. The appearance of *C. comensis*, a species common in oligo-mesotrophic waters, in the topmost sediments, coupled with the reduction in *S. niagarae* and *S. medius* suggests a shift towards a less eutrophic lake state.

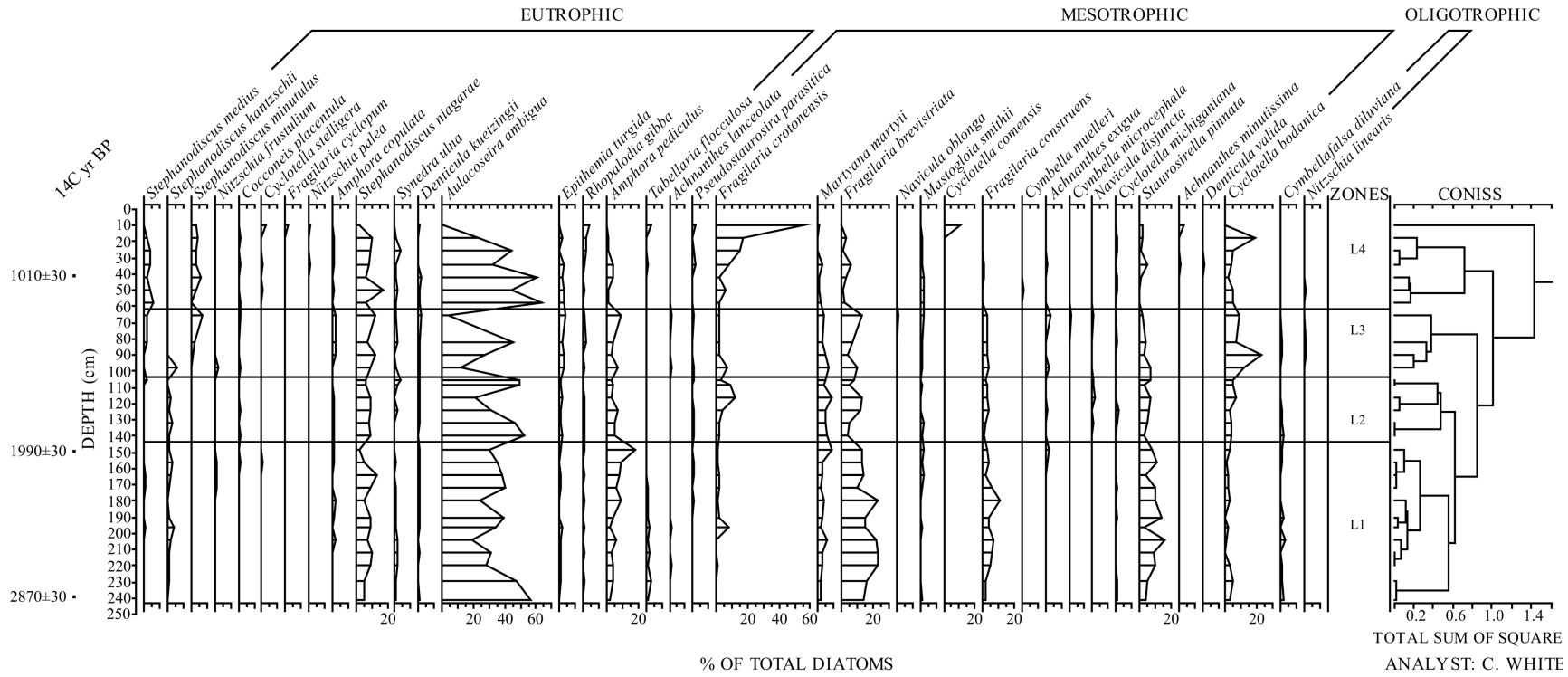


Figure 4.4: Relative abundances of important diatom taxa identified in Clear Lake long core samples; arranged according to calculated weighted averaging bootstrapped TP optima and grouped into corresponding trophic groups. Carbon dates are provided in years BP. The solid black horizontal lines indicate zone boundaries, as determined by CONISS sum of squares analysis (far right).

4.4.2 Diatom analysis of short core sediments

Characteristics of the Clear Lake short core are summarized in Figure 4.5, with respect to ^{210}Pb activity, sediment accumulation rate, and age (as estimated using ^{210}Pb dating). The overall lead activity increases towards the core surface with ^{210}Pb activity peaking at 0.602 Bq/g in the top cm. Sediment accumulation rate increases approximately linearly from 33 to 20 cm depth, from 304 to 824 g/m$^{2/}$year (1868-1955). In the shallower sediments (<20 cm depth) the sediment accumulation rate remains approximately constant, varying from 918 to 1088 g/m$^{2/}$year, with an average accumulation rate of 990 g/m^2/year within this time period (1962-2010). The error surrounding the calculated ^{210}Pb chronology decreases from the core base to the surface. The error from 1955 upward was ≤ ±6 years.

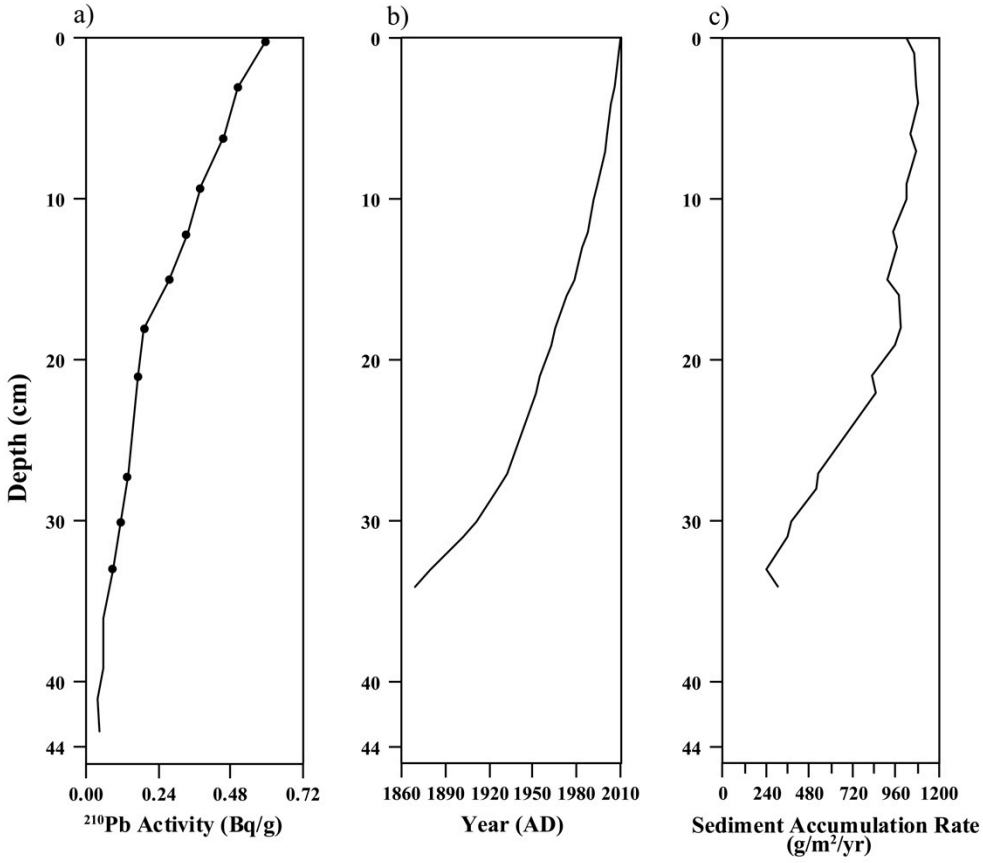


Figure 4.5: Clear Lake core data, (a) Pb 210 activity (Bq/g), (b) Year (AD) and (c) Sediment accumulation rate (g/m^2/year). Depths where samples were submitted for ^{210}Pb analysis are indicated by solid bullets.

It is important to note that there is a discrepancy between the carbon dates obtained for the long core and the ^{210}Pb dates obtained for the short core. The most recent carbon date at 31-32

cm provides a date which is much older, 1010 ±30 ^{14}C yr BP, than the ^{210}Pb date at the same depth, 1901 AD. There are many potential sources of error that may account for this discrepancy, including: reservoir (e.g. hard water) effects, contamination of the carbon used for dating, mixing within the surficial sediments (^{210}Pb), and gravity core shortening (Appleby 2002; Emery & Hülsemann 1964). Core shortening is a problem which accompanies short core sediment extraction using an open tube gravity corer, where the length of the core obtained is less than the depth of core penetration (Emery & Hülsemann 1964). Thus, for example, a corer might penetrate to 40 cm but only 35 cm of sediment is actually retrieved; this depends upon the type of sediment being cored (Emery & Hülsemann 1964). This in turn may cause inaccuracies in both the dates and sedimentation rates calculated from ^{210}Pb.

The Clear Lake short core provides a finer scale examination spanning the same depth range as the top three samples included in the long core analysis. Essentially, the L4 Zone has been examined at a 1 cm resolution in the short core to provide an in depth analysis of recent changes in the diatom species assemblages. Species relative abundances throughout the length of the core are shown in Figure 4.6. CONISS enabled the identification of three Zones in the Clear Lake short core.

Zone S1, which spans the base of the core to approximately 28 cm, is dominated by *Aulacoseira ambigua*, which varies from 10 – 40% of the species abundance. The dominance of this planktonic diatom, along with other eutrophic species, such as *Cyclotella stelligera,* in S1 suggests a eutrophic lake status, (Rosén 1981; Bennion et al. 2004). However, *A. ambigua* is also found in abundance in those lakes of the boreal forest of North America during the period prior to settlement (Stoermer 1977; Yang et al. 1993). *Cyclotella michiganiana*, *Cyclotella bodanica*, and *Amphora pediculus* are more abundant in S1 than elsewhere, indicating a more mesotrophic lake state. Many species have reduced abundances within this zone, including: *Cyclotella comensis*, *Fragilaria crotonensis*, and *Tabellaria flocculosa*. The presence of additional planktonic taxa as well as epiphytic diatoms (i.e., *Stephanodiscus hantzschii* and *Fragilaria brevistriata*) adds additional complexity to assessing lake trophic state during this time. In S1, the low abundance of more eutrophic species, such as *Fragilaria crotonensis* and *Tabellaria flocculosa*, provides an indication that the lake state was not highly eutrophied (Hall & Smol 1992; Smol and Boucherle 1985; Walker et al. 1993). This qualitative assessment suggests the TP levels remained within the mesotrophic range.

The boundary of S1 and S2 is flanked by the establishment of the Timber Reserve (1895) and the establishment of RMNP (1933). Prior to official National Park establishment (1933), in the

1910s, permits for cottages were issued for Clark's Beach on the shore of Clear Lake (Don Huismann, pers. com.). This meant the release of over 250 cottage sites at this time. After the conclusion of WWI the southern boundary of the Timber Reserve changed, removing Proven Marsh, thus reducing the size of what would become RMNP as land was donated to returning veterans (Don Huismann, pers. com.). This meant an increase in population for the region and a reduction in the area protected by the Park, potentially increasing the amount of sewage being introduced to the system.

Within Zone S2 extending from 28 to 14 cm depth (1900-1987), there is a gradual decrease in *A. ambigua* coupled with a substantial increase in *F. crotonensis* and *T. flocculosa.* Similar changes elsewhere have been associated with the onset of cultural eutrophication (Hall & Smol 1992; Smol and Boucherle 1985; Walker et al. 1993). This provides evidence for nutrient enrichment as there is a significant increase in the relative abundance of planktonic diatoms and in particular, an increase in those species generally associated with eutrophic waters (Anderson 1997). The decline in some epiphytic species such as *Fragilaria brevistriata* and *Staurosirella pinnata* implies a decrease in the macrophyte population and an increase in the planktonic taxa. The dominance of planktonic taxa associated with eutrophic lake conditions in conjunction with the elimination of the macrophyte community, as inferred from a reduction in epiphytic diatoms also indicates movement towards a more eutrophic lake state (Bennion et al. 2004). *Cyclotella michiganiana*, which was abundant in S1, disappears almost entirely in S2. This species, as well as *C. bodanica* (which also decreased in S2), is more associated with mesotrophic conditions. Overall this Zone strongly suggests eutrophication in the lake system.

From the 1930s up until the mid 1980s, the golf course located at the east end of Clear Lake was run by RMNP until being contracted out to a private company in the early 1990s. Throughout this entire period fertilizers were employed heavily to maintain golf greens (Don Huismann, pers. com.). Following this, the contract for golf course management was awarded to a new company, one which has since employed a more organic maintenance regimen, eliminating the use of chemical fertilizers and employing alternative methods of management and maintenance (Don Huismann, pers. com.).

Zone S3, the uppermost Zone includes the top 14 cm of the core (1987-2010). There is a great increase in both *Cyclotella comensis* and *Fragilaria crotonensis*, together accounting for almost 70% of the diatoms present in the samples closest to the surface. *F. crotonensis* is absent in areas where pollution is very high, however, its dominance in combination with the presence of *S. minutulus* indicates eutrophic waters (Walker et al. 1993; Stoermer et al. 1978). In contrast

to this, *C. comensis* is commonly present in oligo-mesotrophic waters, but also tends to dominate within lakes that have a depth >10 m (Wunsam et al. 1995; Lotter et al. 2002; Lotter 1998). The presence of *C. comensis,* although indicative of more oligotrophic conditions, may potentially be related to lake depth more so than lake trophic state. *A. ambigua* disappears within the uppermost sediments. It is a more eutrophic species and its abundance is largely limited to the period prior to settlement in lakes of the boreal forest of North America (Stoermer 1977; Yang et al. 1993).

The Zone S2/S3 boundary dates to approximately 1987. At this time there were several changes in Park functions. This provided avenues for alternative protection and interpretation programs to be implemented in RMNP by outside groups, including the Friends of Riding Mountain Biosphere Reserve (Don Huismann, pers. com.). This may have impacted the nutrient input into the Clear Lake system, however it is likely that changes in sewage handling procedures had a more significant impact. Nearby Elkhorn Resort, which is located upstream of Clear Lake, expanded its operation and failed to update its septic system, causing a failure of the system and an increased input of sewage into Clear Lake (Don Huismann, pers. com.). Eventually, as the town site of Wasagaming updated its sewage treatment facility, Elkhorn resort was connected to the updated system, reducing the direct input of sewage into the headwaters feeding Clear Lake (Don Huismann, pers. com.).

In the early 1990s a golf course, employing traditional inorganic fertilizer treatments was constructed at the headwaters of Octopus Creek, which feeds into Clear Lake (Don Huismann, pers. com.). The 33 cottages that were present on the north shore of Clear Lake were forced to update to septic tanks at this time. Prior to this, their sewage was neither contained nor managed (Don Huismann, pers. com.). As park policy changed in the late 1980s the Grey Owl landfill located near Clear Lake was closed (Don Huismann, pers. com.). Prior to its closure, a small creek, which has since dried up, ran through the landfill and fed directly into Clear Lake (Don Huismann, pers. com.). Also at this time beaver dams on Clear Creek were being manipulated to maintain lake levels and prevent flooding of lakeshore trails (Pat Rousseau, pers. com.)

Major sources of phosphorus into aquatic ecosystems include many human sources, comprising industry, urban activity and agriculture. This includes excessive fertilization and manure production (Carpenter et al. 1998). In the course of Clear Lake's recent history there have been many potential human impacts relating to urban activities such as increased wastewater and excess fertilization, driven by recreational and agricultural activities in the region. An important point source of phosphorus within the lake is the Wasagaming Townsite wastewater and sewage treatment facility (Whitehouse, 2010).

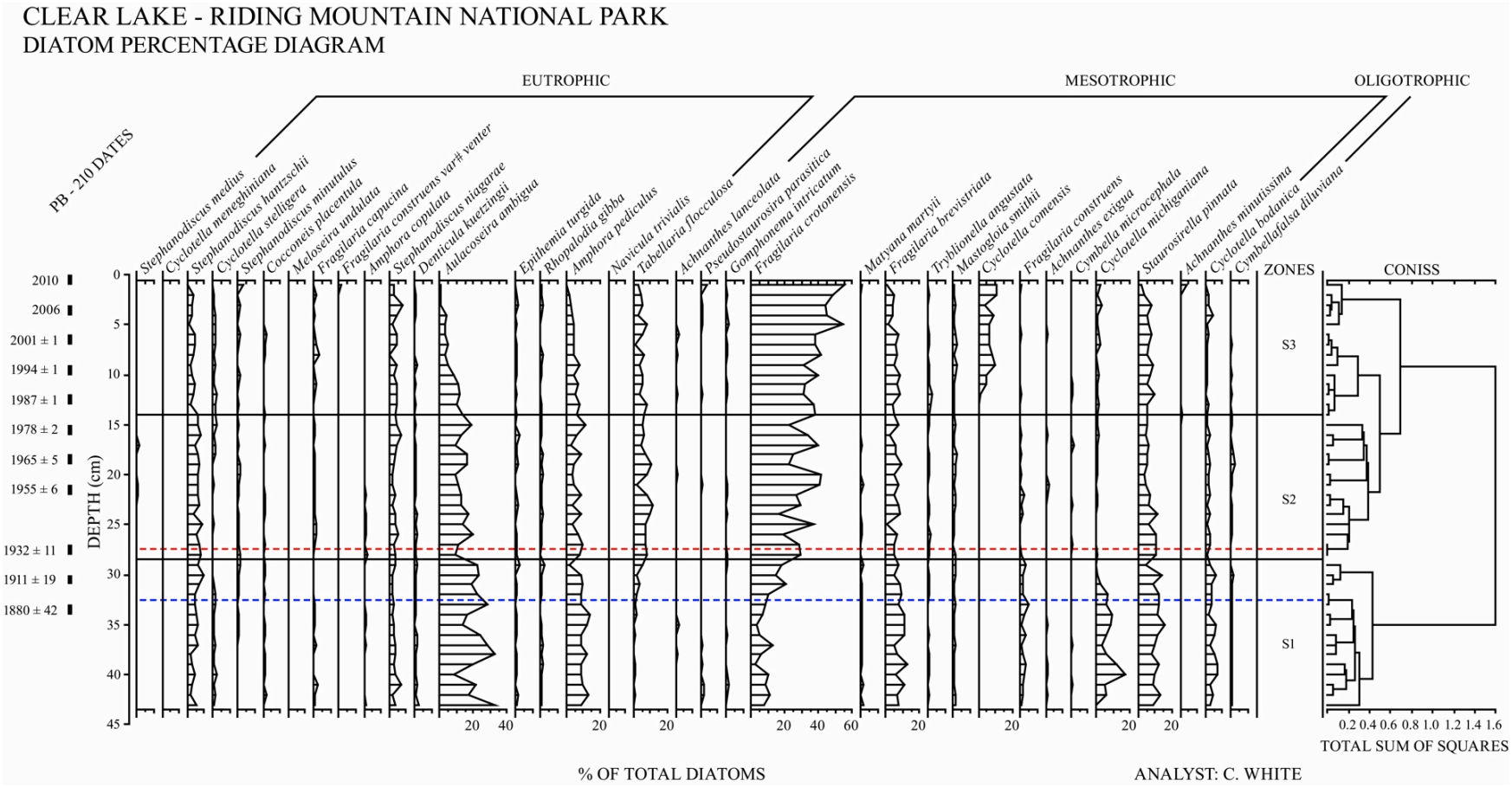


Figure 4.6: Diatom stratigraphy of the most important and abundant taxa from Clear Lake arranged according to weighted averaging bootstrapped TP optima and grouped into corresponding trophic groups. The solid black horizontal lines indicate zone boundaries, as determined by CONISS sum of squares analysis (far right). The blue dashed line represents the approximate time of establishment of the region as a Timber Reserve (1895), and the red dashed line indicates the approximate time of park establishment (1933), as determined by ^{210}Pb dating.

4.4.3 Diatom inferred total phosphorus reconstruction

Diatom inferred TP reconstructions were derived using the transfer functions developed in Chapter 3. Two separate models were employed, a WA (classical deshrinking) and a PLS (one component) model. Each was applied to down core sediment samples for both short and long cores extracted from Clear Lake (Fig. 4.7).

The Clear Lake long core was used to examine the long-term history of DI-TP. Within the long core DI-TP values range from 17.1 to 35.4 µg L^{-1} for the WAclass model and from 19.2 to 35.8 µg L^{-1} for the PLS one component model (Figure 4.7 e,f). The peak in DI-TP levels for both models occurs at a depth of 50 cm. The overall trend is variable throughout the core both prior to and following the peak at 50 cm. In the context of approximate 95% confidence intervals (Figure 4.7 g,h), the peak in DI-TP appears as no more than a small deviation and modern diatom TP values are only slightly higher than presettlement levels.

Within the Clear Lake short cores (dating to 1880 at 33cm depth), the DI-TP values range from 16.9 to 34.3 µg L^{-1} for the WAclass model and from 24.6 to 41.5 µg $L^{-1}$for the PLS one component model (Figure 4.7 a,b). The largest DI-TP values are inferred by both models at a depth of 25 cm. Both models indicate the same general trend of a slight increase towards the peak in DI-TP and then a subsequent gradual reduction in DI-TP moving towards the surface of the core. The approximate 95% confidence intervals are much broader than the DI-TP changes (Figure 4.7 c,d). Any noticeable shifts in DI-TP are reduced to small variations within the context of the error bars, and clearly lack statistical significance.

It should be noted that due to unintentional sampling bias (see Section 3.5.2) there are very significant problems in applying the transfer funtions down-core. In essence, the inferences of a transfer function are artificially constrained to the range of values represented in the training set (Walker et al. 1997). The RMNP region, where the lakes in the training set are located, has a high abundance of both eutrophic and mesotrophic lakes, whereas oligotrophic lakes are rare. In the case of my models, the lowest and highest TP values in the training set are 7 and 780 µg L^{-1}, respectively. Clear Lake TP, measured at the same time, was 8 µg L^{-1}. An unfortunate consequence arises: these models cannot reconstruct TP levels significantly below Clear Lake's current TP levels. Although the transfer function output (Figure 4.7) reliably indicates that Clear Lake TP values were never significantly higher than today, the model is incapable of inferring values significantly lower than Clear Lakes' current TP. Including an increased number of oligotrophic lakes in the model would be a worthwhile exercise to refine the results and improve model predictions in the oligotrophic range.

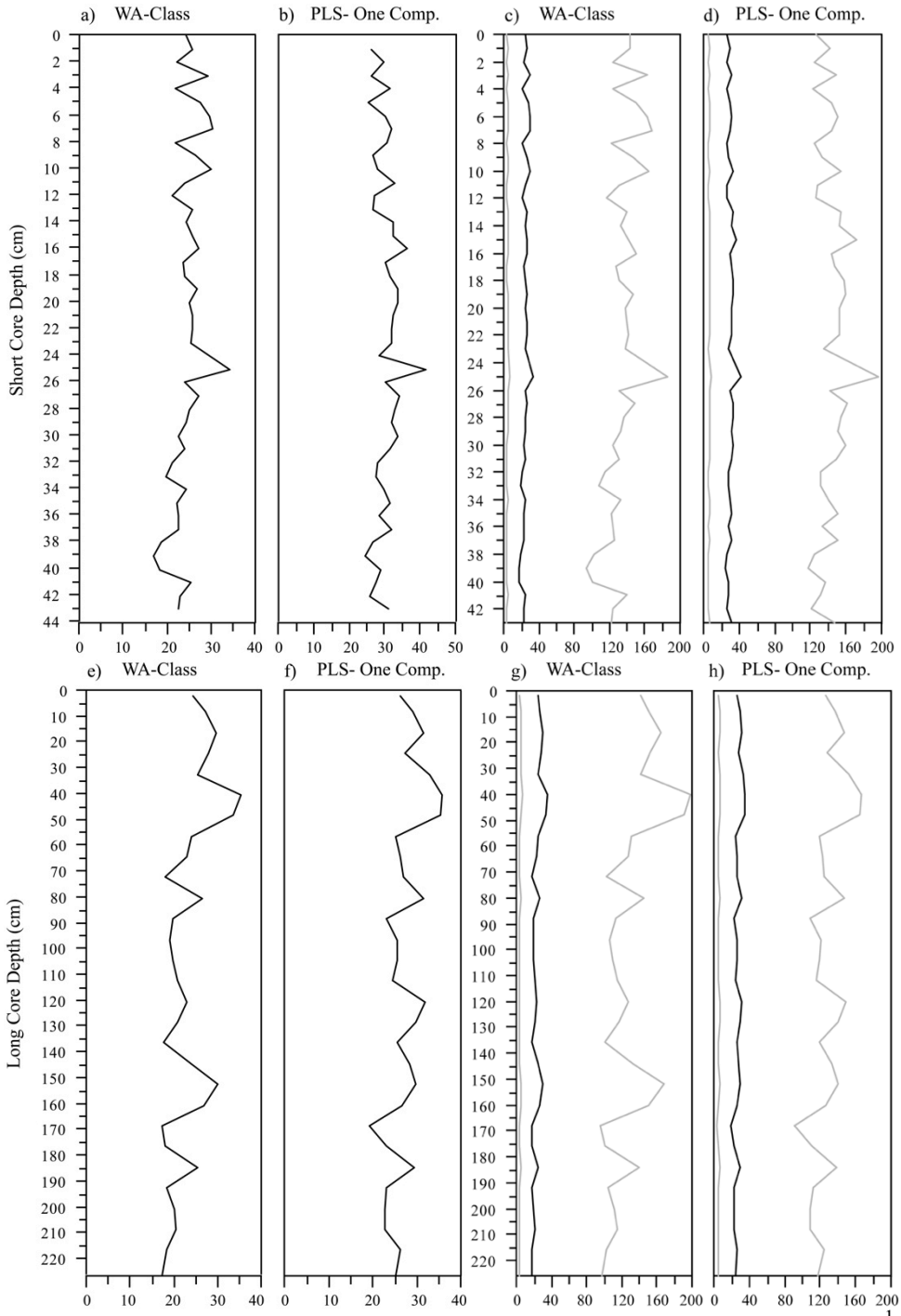


Figure 4.7: A comparison of two models of Diatom Inferred Total Phosphorus (µg L^{-1}) for Clear Lake short core sediments (a-d) and long core sediments (e-h) with (a,b,e,f) no error shown, and (c,d,g,h) two standard errors, indicated by the solid gray line, with Weighted Averaging (classical deshrinking) modelling techniques (a,c,e,g) and Partial Least Squares (one component) modelling techniques (b,d,f,h).

4.4.4 Implications for Park management

This project was initiated by Parks Canada to assess the management of freshwater systems in RMNP. Clear Lake currently is an oligo-mesotrophic lake that is used extensively for recreational activities. Historically, human activities, including those occurring within RMNP, have impacted the lake. These impacts are evident as changes in the lake's diatom flora. Relative to recent changes in diatom floristic composition, pre-settlement changes were minor. Despite the pronounced changes in diatom flora, the diatom-inferred TP record depicts only small, statistically insignificant changes in TP. Similarly, Gray's (2009) analysis of midge remains in Clear Lake sediments revealed no change in bottom fauna or hypolimnetic oxygen. It is possible that the diatoms have been responding to other aspects of ecosystem change (e.g., increased atmospheric N loading, or possibly food web changes induced by deliberate or accidental introductions of fish, aquatic plants, or other organisms, etc.).

The recovery of this system to pre-settlement conditions may rely on improved management of nutrients feeding the system. Although current TP levels are likely close to pre-settlement levels, the diatom assemblage itself is not consistent with the flora found in pre-settlement sediments. This leads me to conclude that while abiotic conditions may have been preserved, biotic conditions have clearly changed. Future monitoring of the diatom flora may be useful, to determine whether natural conditions gradually re-establish, or if the lake drifts further from its natural, pre-settlement condition. In particular, a return to the pre-settlment species, including the dominance of *Aulacoseira ambigua* would indicate a return to the natural floristic composition. Alternately, an increase in the dominance of recently dominant taxa, such as *Cyclotella comensis* would denote a progression away from natural conditions.

4.5 Summary

- Analyses of diatom preserved in Clear Lake's sediments reveal changes in floristic composition and trophic state from prehistoric times to the present day.
- Although diatom floristic changes are evident throughout the record, these changes are especially evident following European settlement of the RMNP region.
- Although diatom floristic changes, based on qualitative interpretation, suggest changes consistent with changes in lake trophic state, there is no clear statistically significant change in DI-TP.

- Unfortunately the DI-TP values are artificially constrained by the dearth of oligotrophic lakes in the training set. Its possible that pre-settlement TP was lower than today, but this cannot be reliably assessed using the DI-TP output.
- Current management practices may successfully be maintaining Clear Lake's trophic state, in terms of TP. Nevertheless, the diatom flora of Clear Lake exists in an altered state, quite different than that existing prior to settlement and park establishment.

Chapter 5. Total phosphorus and trophic state reconstruction from fossil diatoms in Lake Katherine, a lake of special management interest in Riding Mountain National Park of Canada

5.1 Background information

The assessment of a lake's ecological status is essential as a benchmark for its management, and provides a comprehensive view of its natural lake state. Lake Katherine, Riding Mountain National Park of Canada (RMNP), is located in south-western Manitoba, and requires such an assessment, in keeping with the mission of Parks Canada (2009) to maintain and restore the ecological integrity of Park Canada's ecosystems.

Lake sedimentary samples have often been used as a paleolimnological tool to provide long-term lake water quality data that would otherwise be unavailable (Reavie et al. 2006; Vance 1997). The information gathered can be valuable in determining past nutrient and ecosystem change. Diatom inferred total phosphorus (DI-TP) values derived from paleolimnological studies provide effective means of establishing pre-industrial or natural limnological conditions (Dixit & Smol 1994). The establishment of historical total phosphorus (TP) levels and baseline natural lake state will provide a basis for comparison with modern TP levels.

In Chapter 4, I demonstrated the utility of this approach via the first DI-TP reconstruction for a RMNP lake. In this Chapter I extend this approach to a second, smaller lake in the park.

5.2 Study Site

RMNP covers a total area of 2,973 km^2 with 96 km^2 consisting of aquatic habitat (Parks Canada 2011; Saunders 1979). Boreal forest and fescue grasslands dominate the landscape (Parks Canada 2011; Saunders 1979). Freshwater bodies include marshes, bogs, swamps, as well as both shallow and deep lakes (Saunders 1979). Four of these freshwater bodies are of particular management concern: Clear Lake, South Lake, Lake Katherine and Moon Lake. These four lakes are all currently accessible to RMNP visitors and have also been affected by diverse human influences. The location of Lake Katherine within RMNP is indicated in Figure 5.1.

RMNP has been subjected to many human influences. First Nations inhabited the area following glacier retreat 10,000 YBP (Parks Canada 2011). Settlers from non-aboriginal populations began moving into the area in the 1880s, shortly after the Canadian Pacific Rail line reached Brandon, MB in 1881 (Parks Canada 2010). At this time there were approximately 10

large logging and milling operations within RMNP, an activity which peaked at the end of the 19th century, coinciding with two major forest fires in 1890 (Parks Canada and Wilderness Society 2004). Due to fire and logging activities RMNP forests consist of mixed age stands (Saunders 1979). In 1895, the area that was to become RMNP was established as a Timber Reserve, and was designated as a Forest Reserve in 1906 (Parks Canada and Wilderness Society 2004). This Forest Reserve was then given National Park status in 1930, opening officially in 1933, with one mill operation continuing to log within the Park until 1949 (Parks Canada 2010; Parks Canada and Wilderness Society 2004). Due to fuel demands during WWII, RMNP was again used as a mainstay in wood supply (Parks Canada 2010). In 1986, RMNP was designated, along with 15 surrounding rural municipalities, as the Riding Mountain Biosphere Reserve, aimed at the conservation of biodiversity and sustainable resource use (Parks Canada 2011; Riding Mountain Biosphere Reserve 2012).

Lake Katherine is located to the east of Clear Lake. A secondary road runs directly adjacent to the lake (Figure 5.2). It is accessible to visitors, although no motorized watercraft are permitted. Lake Katherine has a dock, making it easily accessible by canoe. A picnic area is located on the northern shore of the lake, and on the opposite side of the lake there are the remnants of a former summer camp facility. There are some macrophytes present within Lake Katherine, however they are mostly emergent or floating-leaved varieties. The lake waters are clear and are of a mesotrophic lake state (18 $\mu g\ L^{-1}$).

Lake Katherine is completely contained within RMNP (Figure 5.1). Lake Katherine has a total area of 0.267 km^2 (26.7 ha) (Kooyman 1980). The maximum depth, as reported by Kooyman (1980) was 8.5 metres (Figure 5.3) with depth averaging around 4.1 metres (Kooyman 1980). The maximum depth, as measured in 2011, was 10.5 metres. This depth discrepancy may be attributed to a difference in water levels since measurements were taken over 30 years apart. The bathymetric profile, as produced by Kooyman (1980), can be seen in Figure 5.3. The lake is surrounded by deciduous and mixed wood forest (See Figure 4.3 in Chapter 4).

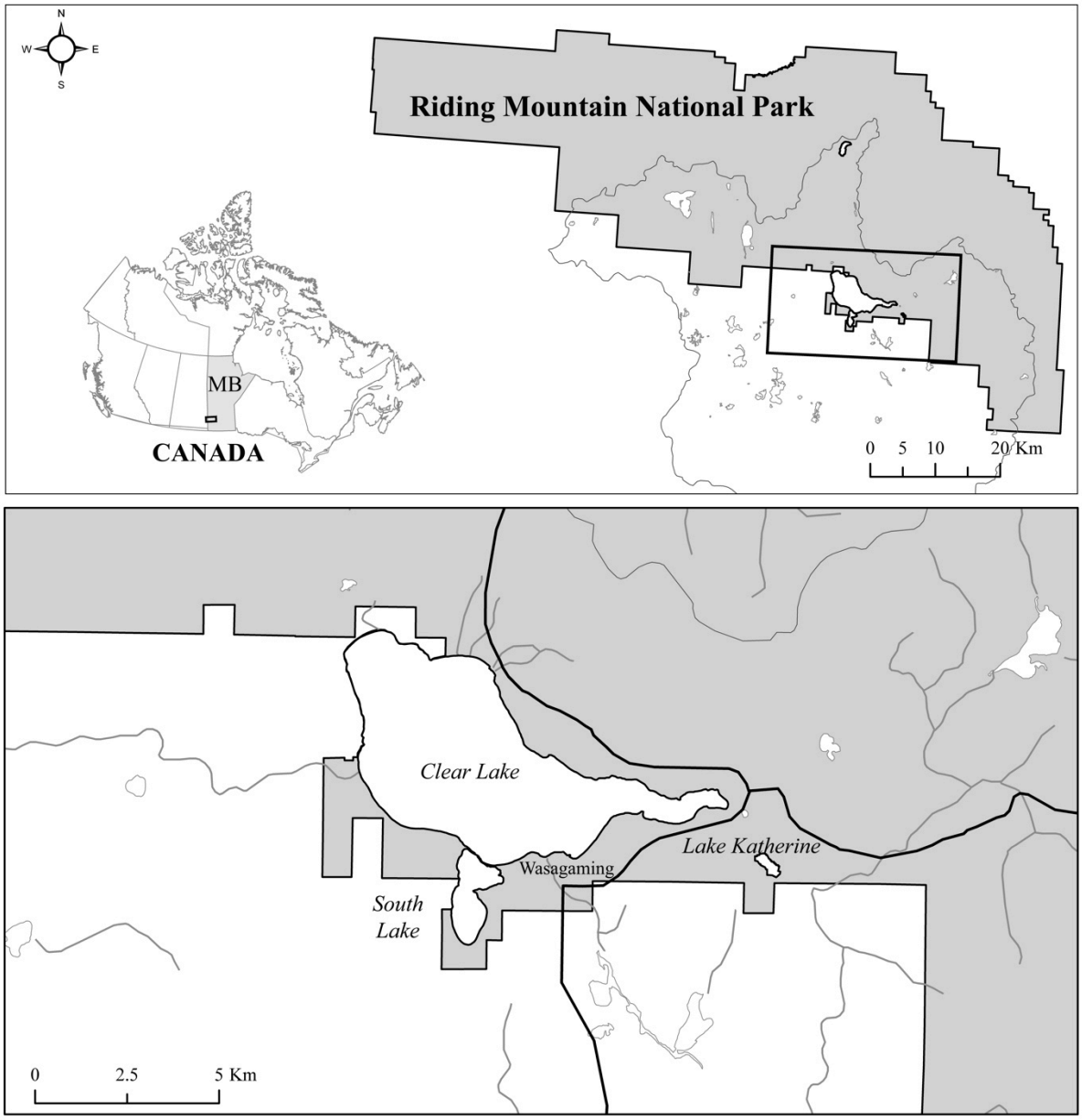


Figure 5.1: Lake Katherine and its location within RMNP, Manitoba, Canada.

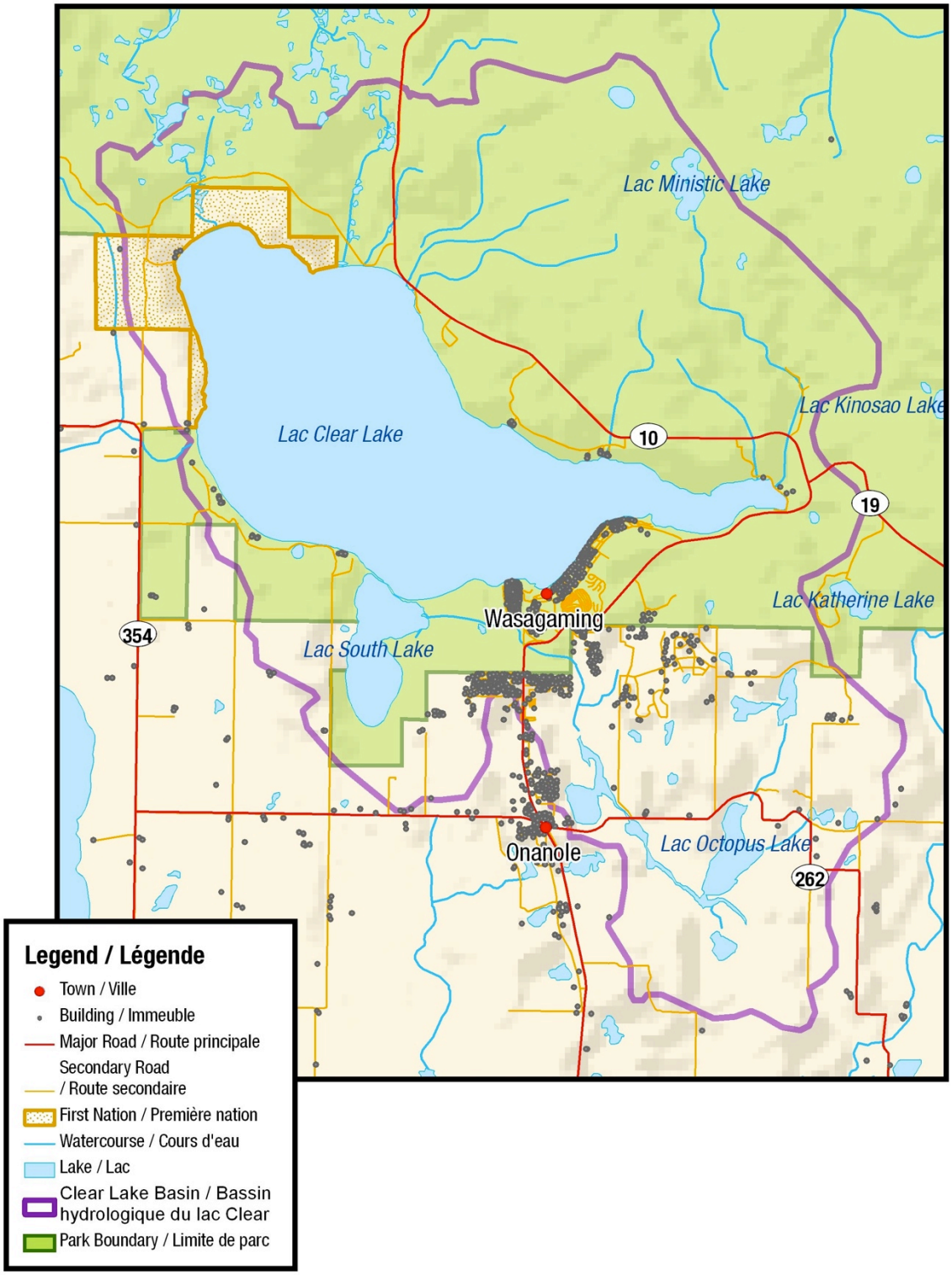


Figure 5.2: Map of Wasagaming, RMNP Townsite, and its proximity to Clear Lake and Lake Katherine. Clear Lake basin, major roads, Park Boundary and other land uses depicted. Produced by Sean Frey for Parks Canada (2010), reproduced with permission.

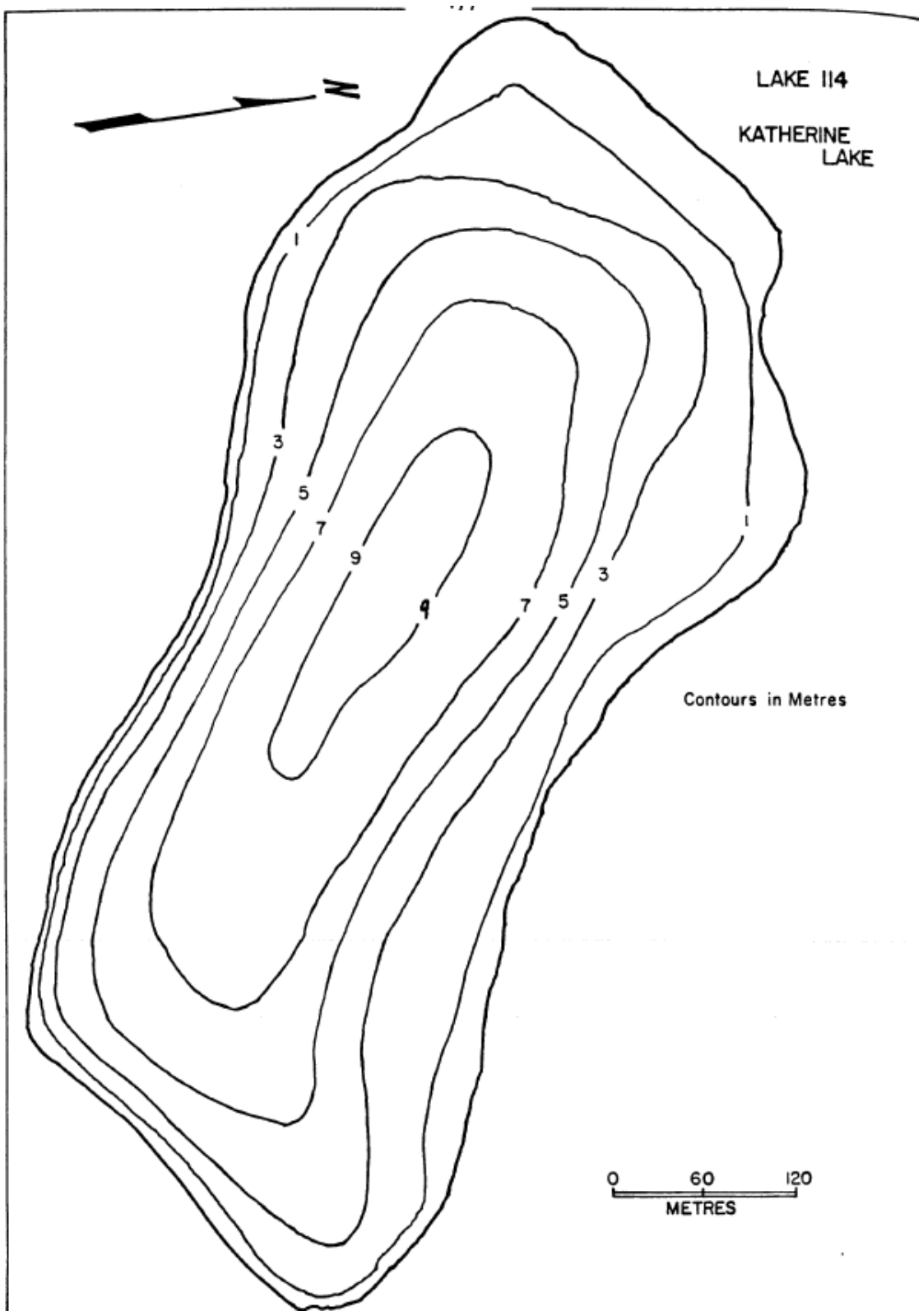


Figure 5.3: Lake Katherine bathymetry, as prepared by Kooyman (1980) using data collected in 1973, reproduced with permission.

5.3 Methods

5.3.1 Short and long core collection, diatom analysis

A short (40 cm) core was collected using a Glew gravity corer with a 3-inch Lexan tube from the centre of Lake Katherine. The sediment was then sectioned into one-centimeter intervals and stored in Whirlpaks®.

A 630 cm long core was extracted close to the short core in Lake Katherine in January 2010 using a Livingstone piston corer. Long core samples were wrapped in plastic wrap and aluminum foil and transported to the Parks Canada Western Service Centre in Vancouver, BC where samples were stored at 4°C. Long core samples were then sub-sampled at 2 cm intervals and stored in Whirlpaks® and then transported to UBC Okanagan for processing and analysis.

All sediments were processed using laboratory techniques described in Section 3.3.2 for the extraction of diatom frustules. Samples from the long core were enumerated at 8 cm intervals. The short core samples were enumerated at a 1 cm sample resolution. At least 500 diatom valves were enumerated per sample, along transects across each slide at 1000× using oil emersion and DIC microscopy.

5.3.2 Core dating techniques

The short core was dated using ^{210}Pb standard radiometric techniques (Appleby 2002). A total of 14 sediment samples were taken for ^{210}Pb analysis at three-centimeter intervals along the length of the 40 cm short core. Radiocarbon dating was used for the long core chronology, with dates derived from samples at 259, 476, 542, and 633 cm.

5.3.3 Numerical methods

Diatom counts were converted to relative abundance values for each level. Diatom stratigraphies were constructed for each core using the program TILIA v. 1.7.16 (Grimm 2011). Species data were then square root transformed to down-weight the effect of abundant taxa within the model (Vermaire & Gregory-Eaves 2008). Major zones of diatom species composition were delineated based on inspection of the stratigraphically constrained incremental sums-of-squares cluster analysis (CONISS) diagram and species distribution patterns throughout the length of the cores (Bennion et al. 2001; Grimm 1987; Kauppila et al. 2002).

Transfer functions developed in Chapter 3 were then applied to down-core diatom species assemblages in the software program C2 v. 1.6.8, yielding a reconstruction of total phosphorus (Juggins 2005; Juggins 2010; Smol & Cumming 2000).

5.4 Results and discussion

5.4.1 Diatom analysis of long core sediments

The Lake Katherine long core reached a depth of 633 centimetres. Below approximately 450 cm, the sediments were coarser and sandier (Figure 5.4). Within these bottom-most sediments diatom populations were diluted to the point where counting 500 valves was no longer feasible. All diatom analyses were performed at 8 cm sample intervals from 450 cm to the surface.

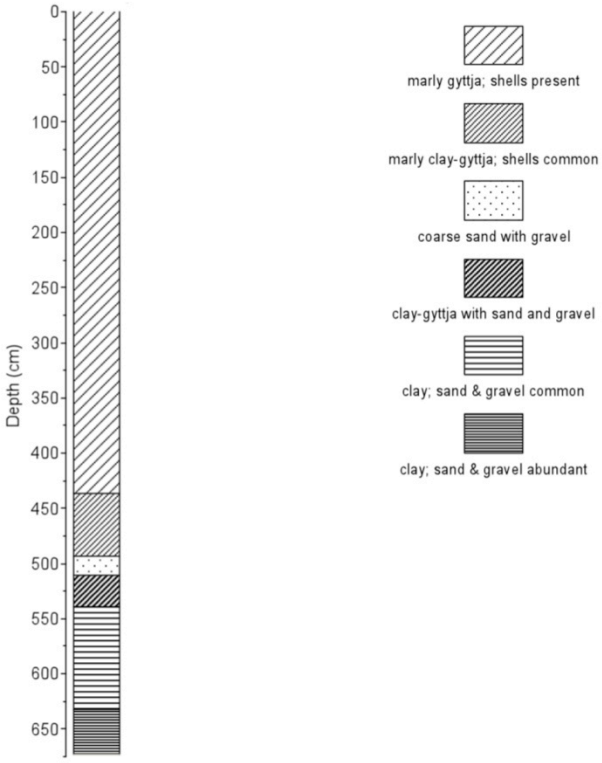


Figure 5.4: Lake Katherine sediment stratigraphy. Lacourse (unpublished data), reproduced with permission.

Figure 5.5 illustrates relative species abundances within the Lake Katherine long core. Cluster analysis aided in division of the core into three major Zones identified based on diatom species assemblages.

Zone L1 encompasses the earliest sediments, of which the majority did not house diatoms. The bottommost diatom-bearing sediments are those extending from 450 to 398 cm. Sediments in L1 date back to the mid to late Holocene: at 476 cm depth 5035 ^{14}C yr BP, at 542 cm depth 8066 ^{14}C yr BP, and at 643 cm depth 11,141 ^{14}C yr BP. The lack of diatoms within certain lake sediments can be the result of a greatly increased sedimentation rate that occurred during

deglaciation, which results in the overall dilution of diatoms within lake sediments (Rawlence 1992). There is also a possibility the lake was completely dry during the early Holocene. *Fragilaria crotonensis* reaches its highest abundance of 70% within this zone thereafter decreasing to levels lower than 5%. *Cyclotella bodanica* and *Amphora pediculus* also reach their highest relative abundance within L1. The majority of *Stephanodiscus* spp. are only present within L1, including: *S. hantzschii*, *S. medius*, *S. minutulus*, and *S. parvus*. Of these, most are indicative of eutrophic lake conditions. *Achnanthes rosenstockii* and *Aulacoseira ambigua* are present at the boundary with L2. Those species present in L1 with small abundances, relative to the other zones, include *Cyclotella michiganiana* and *Staurosirella pinnata*. Both *Navicula oblonga* and *Cymbellafalsa diluviana* are absent from L1. *Martyana martyii* is only present in L1. Both *Cyclotella bodanica* and *Amphora pediculus* are more commonly associated with a mesotrophic lake state, and are found at their maximum abundance within L1. L1 is a period of great change in the core and not considered indicative of established lake conditions.

Zone L2 comprises the longest portion of the core extending from 398 to 60 cm with little overall variation in relative diatom species abundances. The small amount of variance within this zone can denote the natural lake state, prior to anthropogenic influence as it precedes the time of settlement. The planktonic diatoms that were more abundant in L1 are reduced in this Zone and replaced by other species, including *A. ambigua, Cyclotella michiganiana*, and *Stephanodiscus niagarae,* which are present in very small abundances. These planktonic species are classified as indicators of mesotrophic lake status (Yang et al. 1993). *Amphora pediculus* and *A. thumensis* are present in small abudances of <5%, with *A. thumensis* only appearing in L2. *Denticula kuetzingii* appears in L2 and is present at relatively low abundances throughout the remainder of the core. It is an epiphytic species. Other non-planktonic species, including *C. diluviana, N. oblonga, Achnanthes rosenstockii, F. construens, F. brevistriata,* and *S. pinnata,* together make up a large part of the diatom assemblage. These species potentially indicate the presence of macrophytes and/or clear waters that are associated with oligo-mesotrophic lake conditions. In contrast to its dominance in L1 *Fragilaria crotonensis* is severely reduced in L2 to less than 10%, also indicating the less eutrophic lake state.

The uppermost Zone, L3, includes the most recent sediments (60 – 0 cm) wherein there is a substantial increase in the non-planktonic species *C. diluviana, F. construens,* and *S. pinnata.* Species that decrease within this zone include: *Navicula oblonga* and *Cyclotella bodanica*. *Fragilaria crotonensis* disappears from the sediments closest to the surface. *Achnanthes rosenstockii* as well as *Aulacoseira ambigua* are both present at less than 20%. The two

dominant planktonic species in L2, *C. bodanica* and *C. michiganiana*, are reduced in L3. *Cyclotella comensis* appears only within the top-most sediments of L3. *C. comensis* makes up a small portion of the diatom population, however the overall diatom assemblage indicates an oligotrophic lake state.

Throughout this long period there were other factors potentially driving these changes, besides nutrients. This includes climate drivers that have had a regional effect on the lake systems over time through drought and flood cycles, as well as temperature changes (Clark et al. 2002). Changes in the hydrologic budget of a lake system can drive changes in the diatom community, due to variations in both water-level and lakewater chemistry (Clark et al. 2002). During times of low lake levels, associated with periods of drought, an increase in benthic diatoms is commonly expected (Clark et al. 2002). Conversely, the dominance of planktonic diatoms may indicate intervals of higher water levels or more humid climate conditions (Clark et al. 2002). Clark et al. have associated such changes with a 100-130 year drought cycle on the Great Plains during the mid-Holocene. The absence of diatoms in the majority of the Lake Katherine L1 zone, though potentially related to changes in sedimentation, might also indicate a more arid time when the lake was dry. Similarly, the presence of the eutrophic planktonic species in the uppermost portion of L1 might be related to the more humid climate of the Late Holocene (Clark et al. 2002).

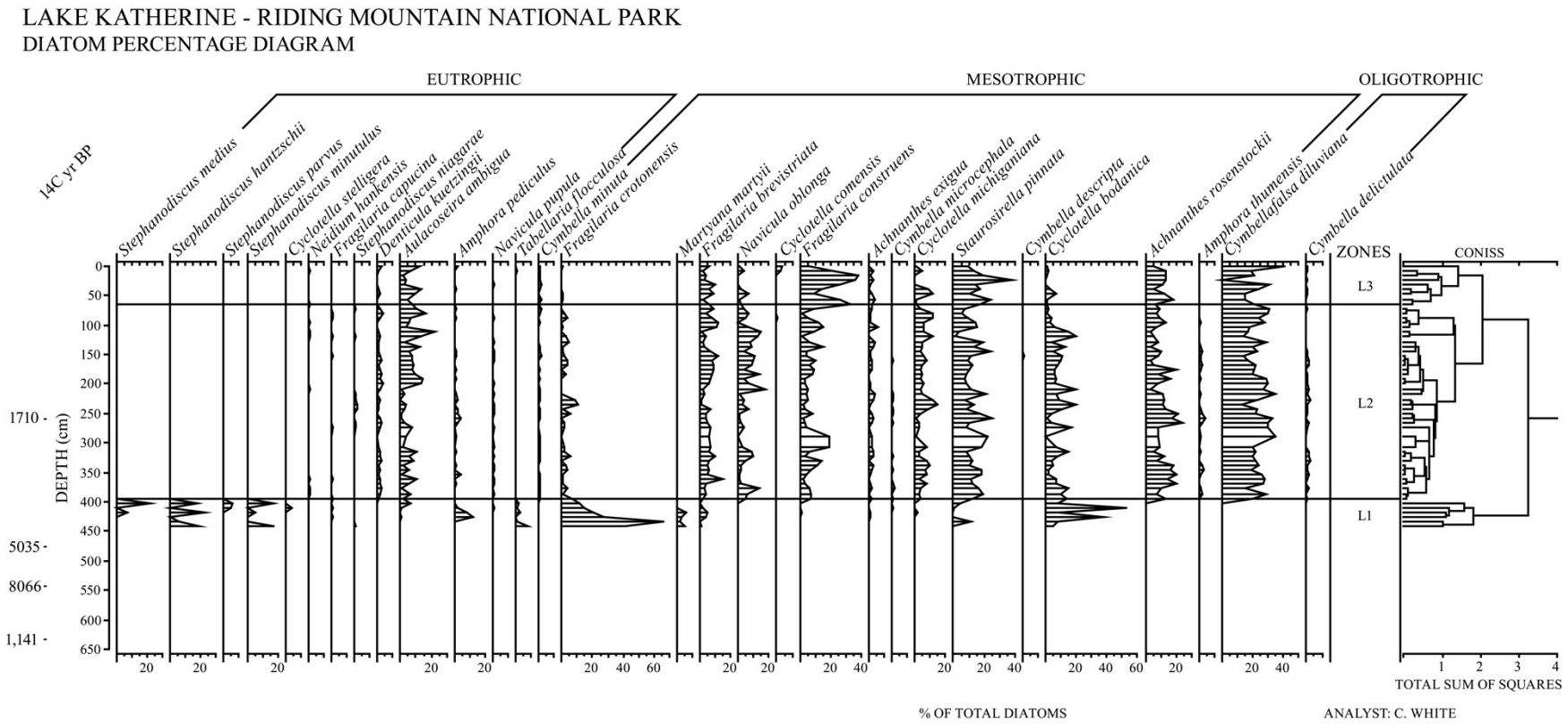


Figure 5.5: Relative abundances of important diatom taxa identified in Lake Katherine long core samples; arranged according to calculated weighted averaging bootstrapped TP optima and grouped into corresponding trophic groups. Carbon dates are provided in years BP. The solid black horizontal lines indicate zone boundaries, as determined by CONISS sum of squares analysis (far right). Total core length was approx. 630 cm, however, due to a steep decline in the abundance of diatom taxa within sediments located deeper than 450 cm, diatoms were not enumerated below this depth.

5.4.2 Diatom analysis of short core sediments

Lake Katherine short core characteristics are summarized in Figure 5.6, with respect to ^{210}Pb activity, sediment accumulation rate, and age (as estimated by ^{210}Pb dating). ^{210}Pb activity peaked at 4 cm depth at 0.949 Bq/g. A sharp break in the ^{210}Pb activity occurs at a depth of 19cm where the activity increases from 0.398 to 0.706 Bq/g. A chronology for the core sediments was obtained using the constant rate of supply model. The sediments at 37 cm depth date back to approximately 1833. The error surrounding the ^{210}Pb dates is greatest at the base of the core (±47 years), but decreases from ±11 years to ±1 year from 1907 (27 cm) upwards. The sediment accumulation rate was greatest in the most recent sediments (0-2 cm) at 229 g/m^2/year. The sediment accumulation rate within the lake has been increasing over the length of the core, with sediment accumulation exceeding 100 g/m^2/year since 1977 (18 cm depth).

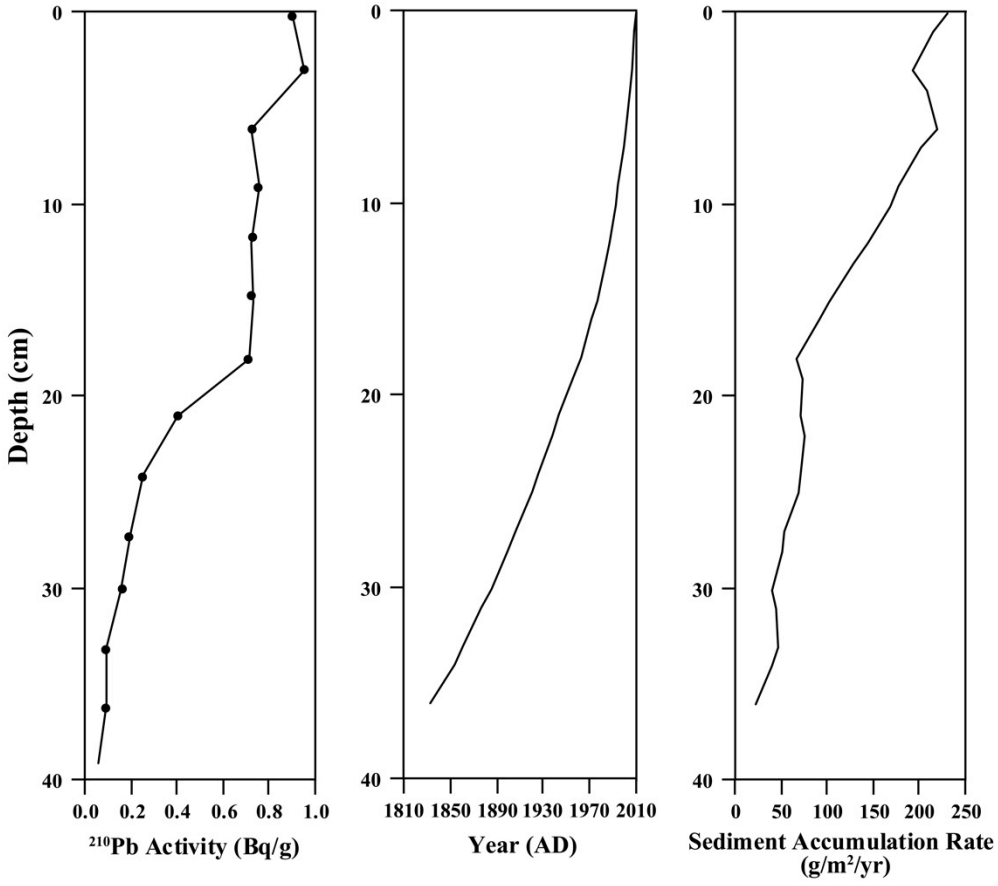


Figure 5.6: Lake Katherine core data, (a) Pb 210 activity (Bq/g), (b) Year (AD) and (c) Sediment accumulation rate (g/m^2/year). Depths where samples were submitted for ^{210}Pb analysis are indicated by solid bullets.

It is important to note that there are many potential sources of error with both the ^{210}Pb and ^{14}C dates, including: reservoir (e.g. hard water) effects, contamination of the carbon used for dating, mixing within the surficial sediments (^{210}Pb), and gravity core shortening (Appleby 2002; Emery & Hülsemann 1964). Core shortening is a problem which accompanies short core sediment extraction using an open tube gravity corer, where the length of the core obtained is less than the depth of core penetration (Emery & Hülsemann 1964). Thus, for example, a corer might penetrate to 40 cm but only 35 cm is actually retrieved; this depends upon the type of sediment being cored (Emery & Hülsemann 1964). This in turn may cause inaccuracies in both the dates and sedimentation rates calculated from ^{210}Pb.

The Lake Katherine short core provides a finer scale examination spanning the same time period as the top three samples included in the long core analysis. Essentially, the L3 Zone has been examined at a 1 cm resolution in the short core to provide a detailed examination of recent changes in the diatom species assemblages. Species relative abundances within the short core are shown in Figure 5.7 with corresponding sums of squares cluster analysis, CONISS (Grimm 1987). CONISS delineated three distinct zones in the core.

Zone S1 ranges from the bottom of the core to 34 cm depth. The ^{210}Pb estimated date at 34 cm is 1860. *Cymbellafalsa diluviana* and *Achnanthes exigua* reach their maximum abundance within this zone. *Cybellafalsa diluviana,* as a benthic taxon, is most common in shallow lakes and would more likely be associated with more oligotrophic conditions as it requires light penetration to the benthos (Hall & Smol 1992). Both *Fragilaria brevistriata* and *F. construens* are present in an abundance of at least 10% and ranging to 20% within S1. Remaining at a relatively constant level of abundance throughout S1 was *Staurosirella pinnata* at approximately 30%. The presence of this species in conjunction with other epiphytes suggests that there were macrophyte populations present. The relatively minor abundance of *Cyclotella comensis* and *Fragilaria crotonensis*, two species more closely associated with the post-settlement time period, affirms the lack of anthropogenic influence within S1 (Lake of the Woods 2006; Smol and Boucherle 1985; Walker et al. 1993).

Zone S2 includes the sediments from approximately the time of Timber Reserve and Park Establishment, as indicated by the blue and red dashed lines, respectively, in Figure 5.7. S2 extends from 34 to 13 cm. Within this zone there is a dramatic increase in the abundance of *Fragilaria construens*, accounting for up to 60% of diatoms. There is a corresponding peak in *F. brevistriata* at the time of Park establishment (1933). This species can be indicative of cultural enrichment of lake waters, following European settlement (Schuette and Bailey 1980).

Staurosirella pinnata and *Fragilaria crotonensis* remain at approximately the same abundance as seen in Zone 1. Both *Achnanthes exigua* and *Cymbellafalsa diluviana* exhibit a decline in abundance over the course of S2. *Navicula oblonga* was present within S2, though in small amounts. The abundance of epiphytes suggests the presence of a large population of macrophytes at this time.

During Zone S2 a multitude of anthropogenic influences potentially impacted on the lake system as it extends from prior to park establishment into the early 1960s. Lake Katherine originally had 125 campsites that were established in the 1930s (Don Huismann, pers. com.). Lake Katherine acted as the overflow from the Clear Lake campsite, providing a more family friendly campground for the park. In the 1950s a lagoon was put in place so as to contain the sewage from the campsites and prevent it from flowing directly into the lake (Don Huismann, pers. com.). Throughout this time period there was extremely high water flow causing the lake to overflow several times. The majority of the input to the lake is derived from groundwater sources (Pat Rousseau, pers. com.).

The lagoon that was adjacent to the Lake Katherine campground was constructed in the 1950s however it was unlined and eventually cracked in the 1970s, leaking sewage into the lake (Don Huismann, pers. com.). Also at this time there was heavy boat usage on the lake (Don Huismann, pers. com.). The crack in the lagoon was left unfixed and both the sewage lagoon and the campground were eventually put out of commission in the 1980s (Don Huismann, pers. com.). The land that was originally the campground was then transferred to the Rolling River Reserve First Nations (Don Huismann, pers. com.).

Zone S3 includes the uppermost sediments (0 to 13 cm) and ranges from the mid 1960s to the present. In S3 there is an increase in the abundance of several planktonic species, including *Cyclotella comensis* and *Fragilaria crotonensis,* as well as an extreme reduction in the previously dominant epiphyte *F. construens*. Throughout S3 *Fragilaria brevistriata* and *Cymbellafalsa diluviana* remain relatively constant between 5 and 10% abundance levels. *Achnanthes exigua* remains at the same reduced abundance as seen in S2, while *Staurosirella pinnata* decreased slightly in the same regard. *Navicula oblonga* is present at its peak abundance. As a species associated with increased levels of dissolved solids and nutrients its appearance is likely related to a progression to a more eutrophic lake state (Bahls 2011). Other epiphytic species also decrease in abundance, such as *S. pinnata*.

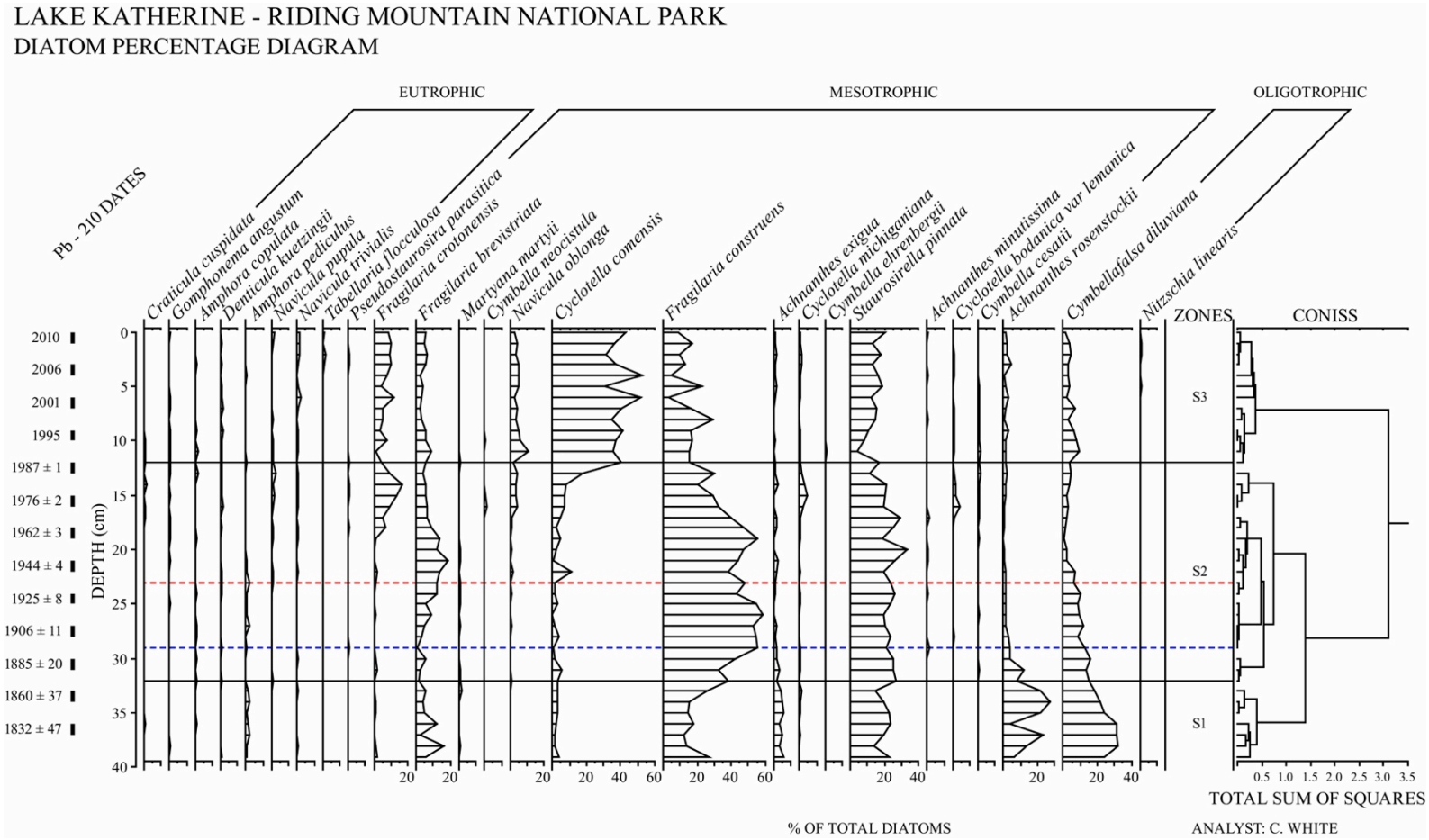


Figure 5.7: Diatom stratigraphy of the most important and abundant taxa from Lake Katherine arranged according to calculated weighted averaging bootstrapped TP optima and grouped into corresponding trophic groups. The solid black horizontal lines indicate zone boundaries, as determined by CONISS sum of squares analysis (far right). The blue dashed line represent the approximate time of establishment of the region as a Timber Reserve (1895), and the red dashed line indicates the approximate time of park establishment (1933), as determined by ^{210}Pb dating techniques.

5.4.3 Diatom inferred total phosphorus reconstruction

The Lake Katherine long core reconstruction of DI-TP is based on WA (classical deshrinking) and PLS (one component) models and depicts long-term inferred changes in nutrient concentration and trophic state (Figure 5.8). In the long core DI-TP values range from 6.3 to 92.2 μg L^{-1} using the WA model and from 15.6 to 74.9 μg $L^{-1}$for the one component PLS model (Figure 5.8 e, f). DI-TP values are highest in the bottommost portion of the core in both model scenarios. The WA and PLS models, show a slight increase in the topmost sediment (Figure 5.4 g, h). Overall reconstructed DI-TP was higher using the PLS versus the WA model. The variations in DI-TP in the upper 390 cm are not obvious. The large variations at the bottom of the core obscure these changes.

The more recent shifts in TP, as inferred from the short core diatom assemblages, in Lake Katherine are illustrated in Figure 5.8. Both the WA classical model and one component PLS model indicate changes over the past 150+ years (1832 at 37 cm depth). The DI-TP range for the WAclass model is 6.7 to 11.3 μg L^{-1} and from 14.1 to 20.6 μg L^{-1} for the one component PLS model (Figure 5.4 a, b). The WA classical and one component PLS models show differing and sometimes opposite trends in recent DI-TP. Inferences using the WAclass model increase to a peak at 12 cm. A slight reduction in DI-TP is evident thereafter. The one component PLS model indicates an overall downward trend in TP levels, with the lowest DI-TP levels occurring at 16 cm depth. Using the one component PLS model there is a minute reduction in DI-TP above 12 cm, with a more pronounced decrease in the top 4 cm of the core. These apparent strong variations and the differences both within and between the models are scarcely apparent when illustrated in the context of two standard error bars (Figure 5.4 c,d).

As noted in Chapter 4, the unintentional sampling bias in the transfer function training set significantly constrains the DI-TP inferences, approximately to the 7 to 780 μg L^{-1} range. The RMNP region, where the lakes in the training set are located, has a high abundance of both eutrophic and mesotrophic lakes, whereas oligotrophic lakes are rare. This range allows the models to reliably infer very high (eutrophic) TP values in the early history of Lake Katherine, but it significantly limits the models' ability to infer TP values below Lake Katherine's current measured TP, 18 μg L^{-1}. This limits my ability, on the basis of the quantitative DI-TP inference, to assess recent changes in DI-TP, and to assess whether current TP values are higher than natural, pre-settlement values.

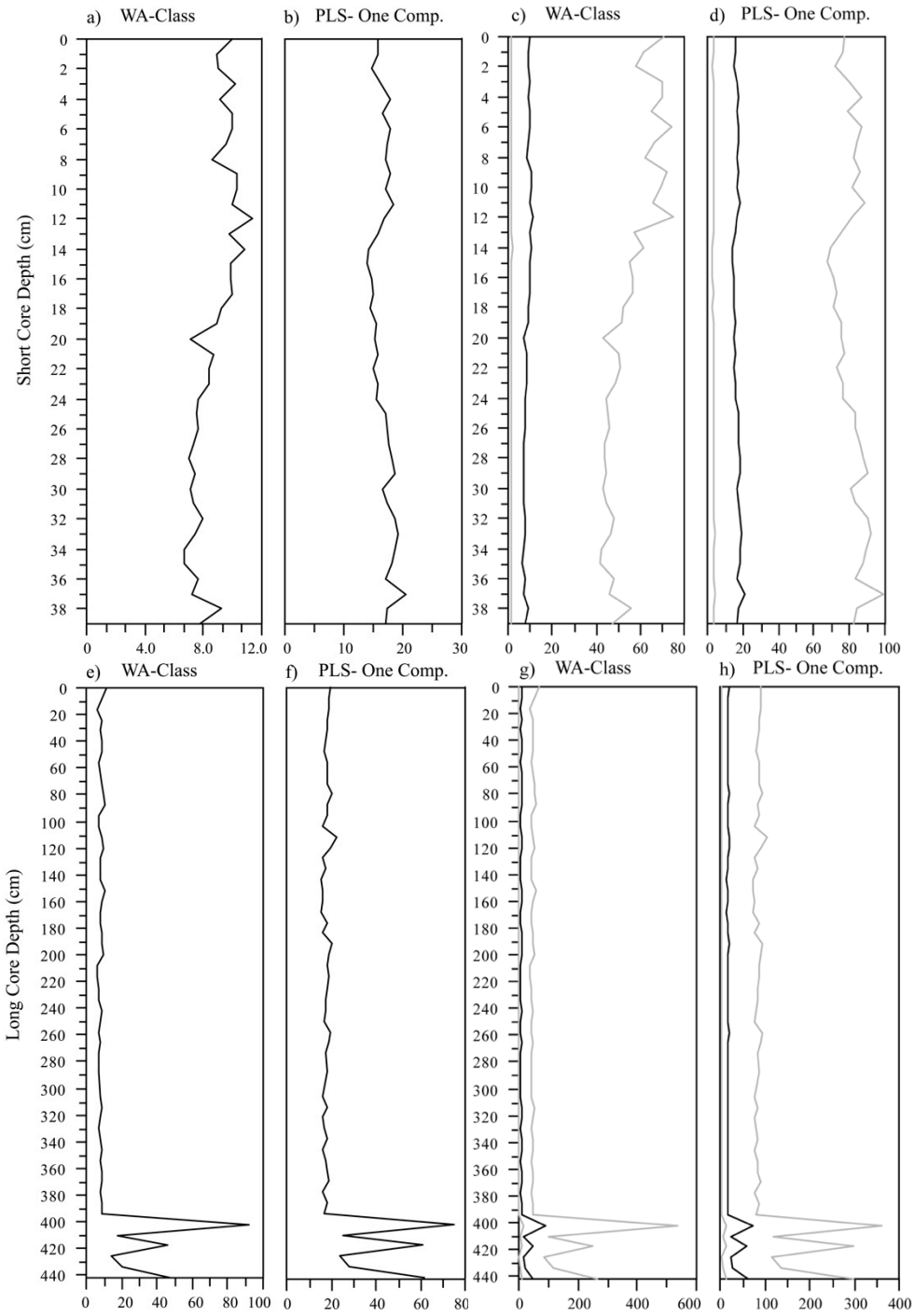


Figure 5.8: A comparison of two models of Diatom Inferred Total Phosphorus (µg L^{-1}) for Lake Katherine short core sediments (a-d) and long core sediments (e-h) with (a,b,e,f) no error shown, and (c,d,g,h) two standard errors, indicated by the solid gray line, with Weighted Averaging (classical deshrinking) modelling techniques (a,c,e,g) and Partial Least Squares (one component) modelling techniques (b,d,f,h).

5.4.4 Implications for Park management

This project was initiated by Parks Canada to assess lake management and ecosystem integrity in RMNP. The Lake Katherine sediment stratigraphy extends well beyond pre-settlement times, perhaps extending to glacial times. Lake Katherine is one of the smaller lakes within the park, though still very accessible to park visitors. The DI-TP inferences do not allow me to confidently state whether, or not, natural TP levels were lower than those measured today. Nevertheless, the current diatom flora is different from that present in the pre-settlement portion of the core. This indicates that Park management has not been adequate to maintain the natural biotic condition. Additional management may be needed to achieve Parks Canada's goal of maintaining ecological integrity. More research is required to determine potential sources of the increased nutrient(s). Although McGowan et al. (2005) suggest a combination of nutrient reduction with biomanipulation measures, such as the introduction of herbivorous fish, from a diatomological perspective the restoration of natural lake state would require a shift towards more natural conditions and allowing time for system recovery, and less so towards an increased amount of interference within the ecosystem.

5.5 Summary

- Analyses of diatoms preserved in Lake Katherine's sediments reveal changes in floristic composition and trophic state from prehistoric time to the present day.
- The earliest diatom asssemblages, dating from the early to mid-Holocene, indicate highly eutrophic conditions with DI-TP values exceeding 92 µg L^{-1} (WA classical deshrinking model).
- Oligo-mesotrophic conditons were established in the Late Holocene (Avg. DI-TP = 8.2 (WA) 17.8 (PLS) µg L^{-1}).
- The DI-TP inferences were artificially constrained to the approximate 7 to 780 µg L^{-1} range; thus, I have not been able to reliably conclude whether, or not, pre-settlement TP was lower than current TP values. Nevertheless, the diatom community of today clearly differs from that extant prior to European settlement.
- Management of Lake Katherine has failed to maintain natural biotic and abiotic conditions; thus, additional attention may be required to re-establish the natural integrity of the Lake Katherine ecosystem.

Chapter 6. Conclusion

Through the use of multivariate statistical methods, including ordination, total phosphorus was determined to be the environmental variable most strongly and significantly influencing diatom communities in RMNP aquatic systems and in the surrounding area. WA and PLS techniques provided effective means for developing total phosphorus transfer functions based on diatoms. Adding more lakes from surrounding regions to the training set could have a great and beneficial impact on model performance and thus on the confidence with which TP levels can be reconstructed. The addition of more lakes to the training set, particularly in the oligotrophic range, would help address the somewhat biased composition of the 47-lake sample set.

The diatom-TP inference models were applied to fossil diatom assmblages in both short and long cores from both Clear Lake and Lake Katherine. The history of trophic change in both lakes was reconstructed. These quantitative data in combination with a qualitative assessment of species assemblages and historical records of human influence on the system provided information to determine natural lake states and to assess the impact of human activities on each system.

In Clear Lake the prehistoric TP-levels were not significantly higher than those reconstructed in the more recent past (i.e. Clear Lake S3 zone). Although the TP level measured on 15 Aug 2011 was lower than that reconstructed for any time in the lake's history, a direct comparison of the TP measured on one summer day with the longer term reconstructed values may not be appropriate given the different methods employed. Also, it is possible that pre-settlement TP was lower than post-settlement levels, but this cannot be confirmed given the limitations of the current diatom–TP inference model. A better model would be required, with smaller errors and incorporating more oligotrophic lakes. Although no significant TP change has been reconstructed, the present diatom flora of Clear Lake clearly differs from that extant prior to European settlement.

Lake Katherine has a much longer sedimentary record that extends down to a depth of 630 cm. However, below 450 cm diatoms were scarce or absent and enumeration was not feasible. Their absence is potentially related to post-glacial processes with rapid sediment accumulation. Although the lake was initially eutrophic, the natural lake state through most of its history was oligo-mesotrophic (DI-TP = WA: 6 – 11 μg L^{-1}, avg. 8, PLS: 15 – 22 μg L^{-1}, avg. 17) with waters clear enough to support a macrophyte community, as indicated by the presence of epiphytic and benthic diatom species. The DI-TP inferences were inadequate to assess whether

pre-settlment TP was lower than TP measured today. Nevertheless, the diatom flora is clearly different, indicating that biotic conditions have changed.

The models developed in this thesis may be applied to other lakes within RMNP for additional assessments of human impacts on the park as a whole. It would be interesting to compare the changes observed in Clear Lake and Lake Katherine to more remote lakes in RMNP, to determine whether the observed changes might be the result of regional vs. local environmental change. Before conducting further analyses however, it would be very helpful to incorporate a suite of oligotrophic lakes into the training set.

Continued biomonitoring requires the examination of diatom assemblages on at least a decadal, but preferably per annum, basis from this point forward. Examination at this interval would provide key data determining if the assemblage is changing, whether it be towards the natural population, or simply a continuation of the current assemblage. An analysis of diatom populations on a seasonal basis within these two lakes would also add another level of understanding between the diatom populations and their relationship to the nutrients available within the lake system.

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Appendices

Appendix 1. Light and Scanning Electron microscope images of diatoms from lakes in Riding Mountain National Park of Canada and surrounding lakes in the Little Saskatchewan Watershed.

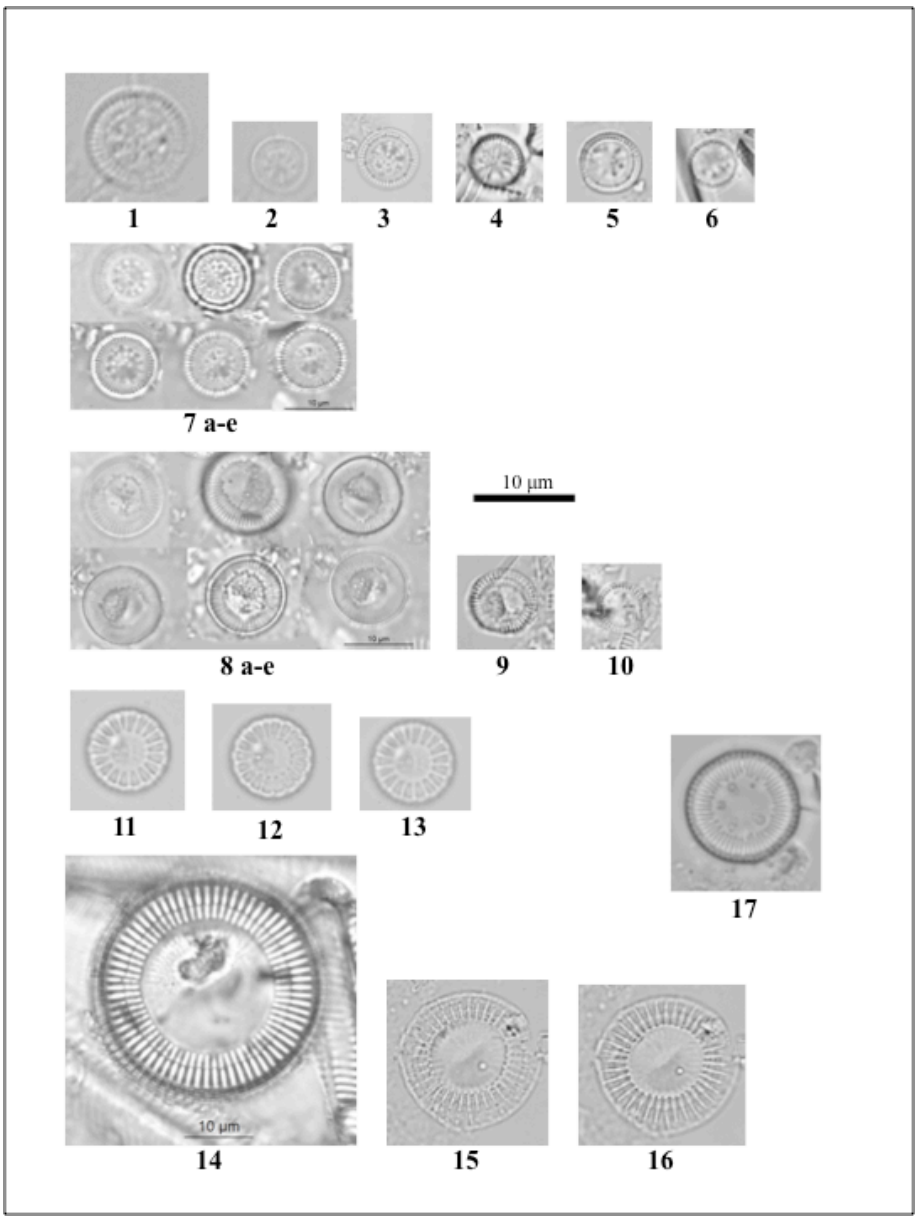


Figure A1.1:Identification plate for *Cyclotella* spp.

1 – 7 a-e: *Cyclotella comensis* Grunow in Van Heurck 1882

8 a-e – 10: *Cyclotella michiganiana* Skvortzow 1937

11-13: *Cyclotella meneghiniana* Kützing 1844

14 – 16: *Cyclotella gamma* Sovereign 1963

17: *Cyclotella rossii* Håkansson 1990

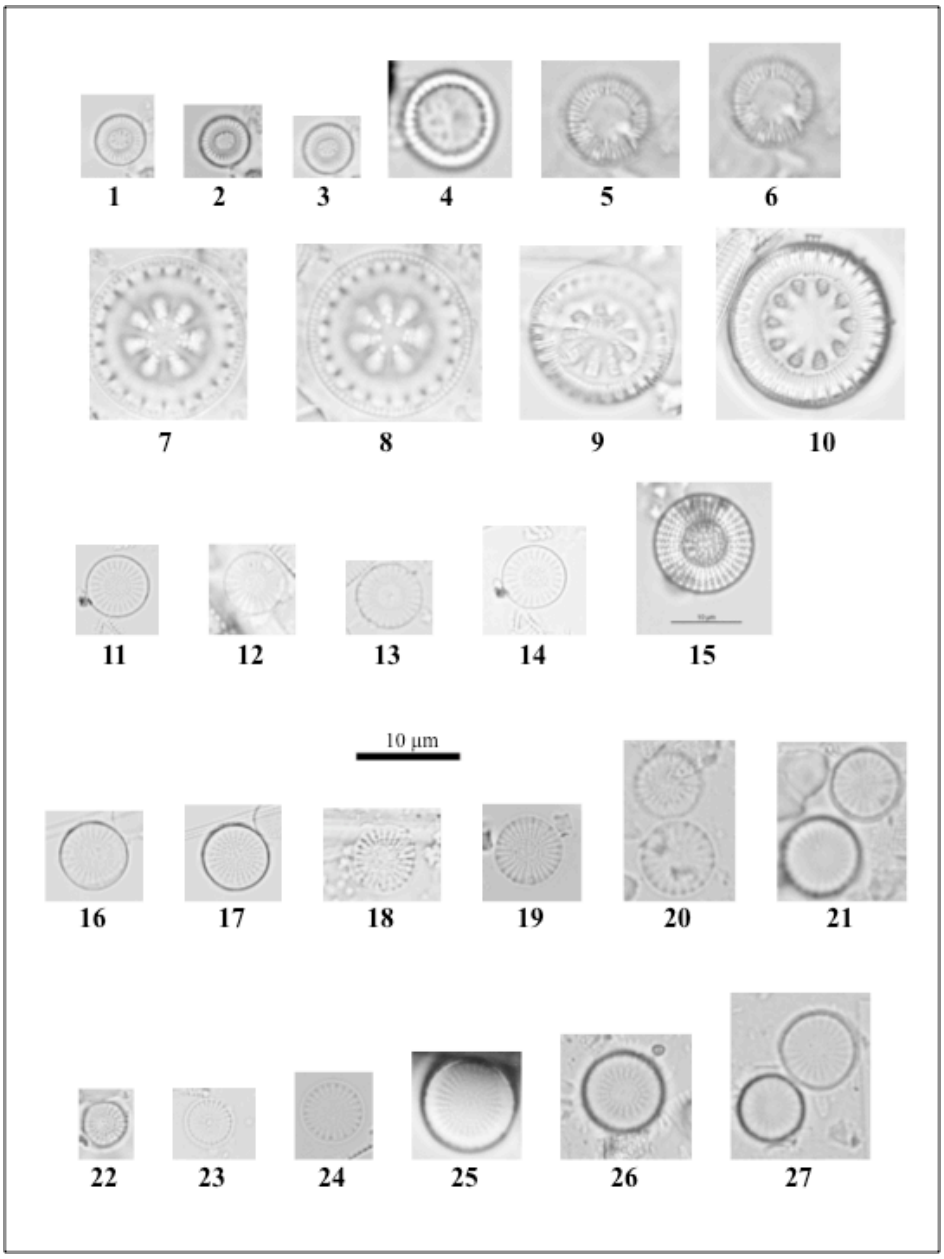


Figure A1.2: Identification plate for *Cyclotella* spp. and *Stephanodiscus* spp.

1 – 3: *Cyclotella stelligera* (Cleve & Grunow in Cleve) Van Heurck 1882

4: *Cyclotella pseudostelligera* Hustedt 1939

5 – 6: *Cyclotella kuetzingiana* var. *schumanni* (*schumannii*) Grunow in Van Heurck 1882

7 – 10: *Cyclotella antiqua* W. Smith 1853

11 – 15: *Stephanodiscus minutulus* (Kützing) Cleve & Möller 1882

16 – 18: *Stephanodiscus parvus* Stoermer & Håkansson 1984

19 – 21: *Stephanodiscus medius* H. Håkansson 1986

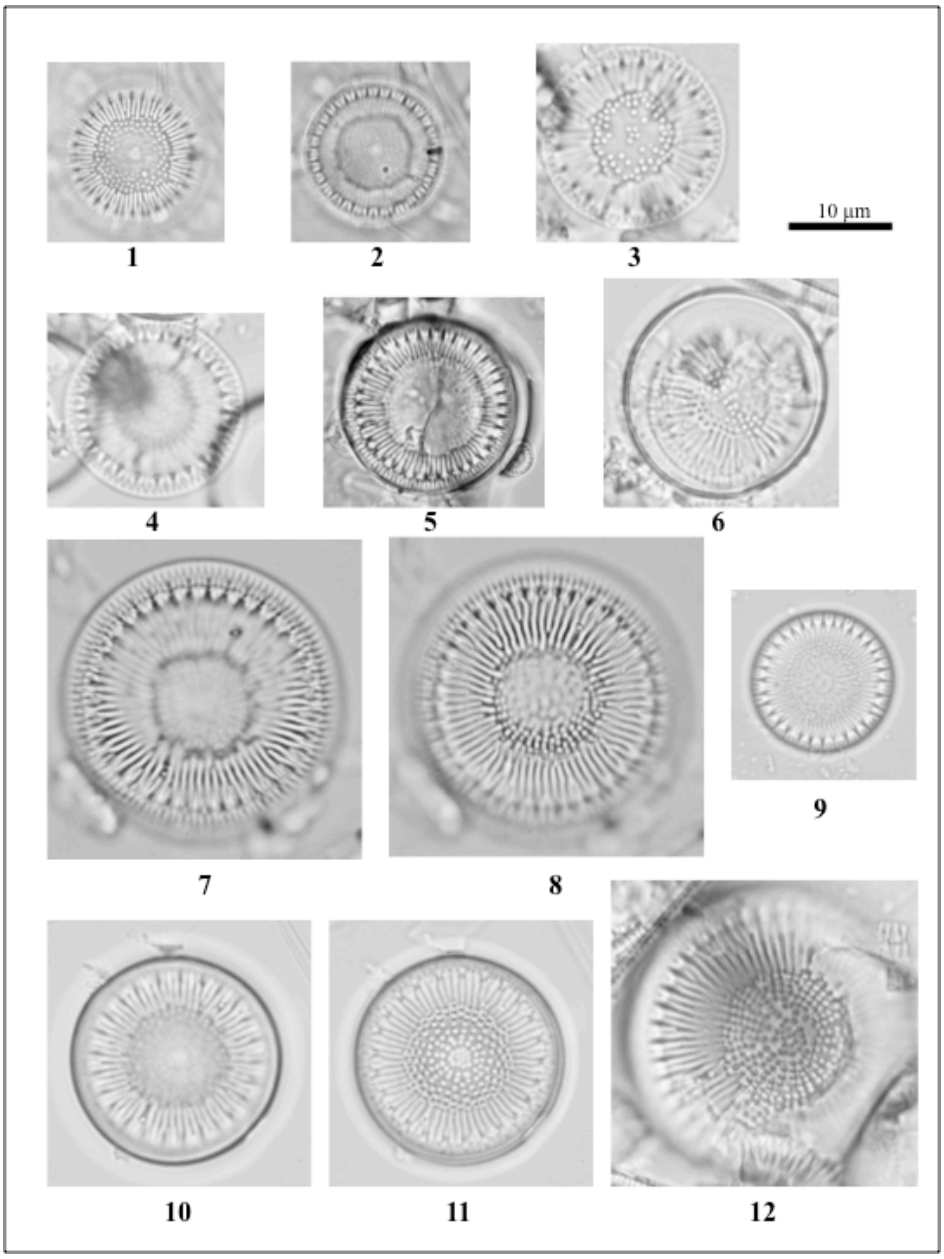


Figure A1.3: Identification plate for *Cyclotella bodanica* spp.

1 – 6, 10 – 12: *Cyclotella bodanica* f. *lemanica* (Otto Muller in Schroter; Otto Muller in Chodat) Bachmann 1903

7 – 9: *Cyclotella bodanica* Grunow in Schneider – Synonym: *Puncticulata bodanica* (Grunow in Schneider) Håkansson 2002

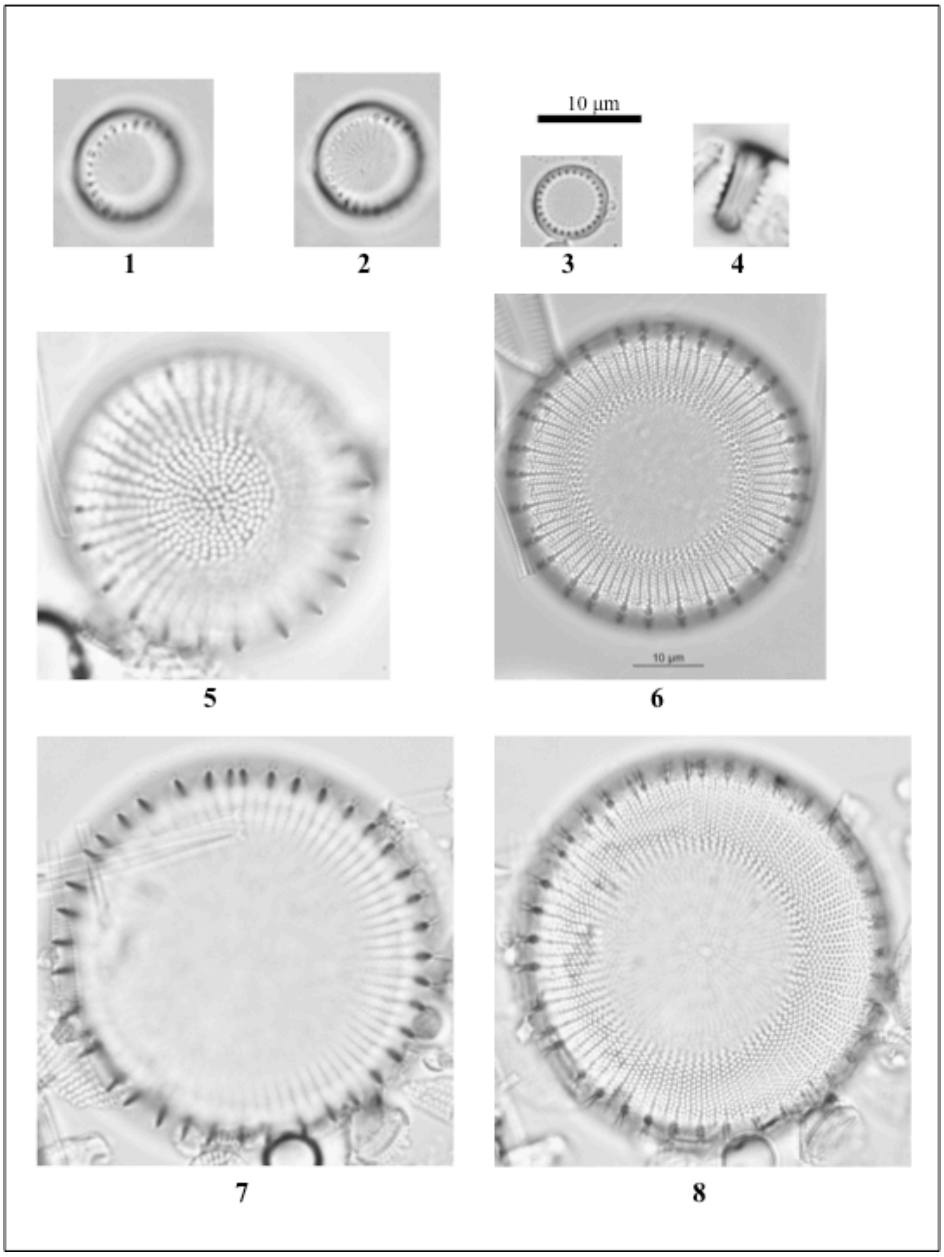


Figure A1.4: Identification plate for *Stephanodiscus* spp.

1 – 4: *Stephanodiscus hantzschii* Grunow in Cleve & Grunow 1880

5 – 8: *Stephanodiscus niagarae* Ehrenberg 1845

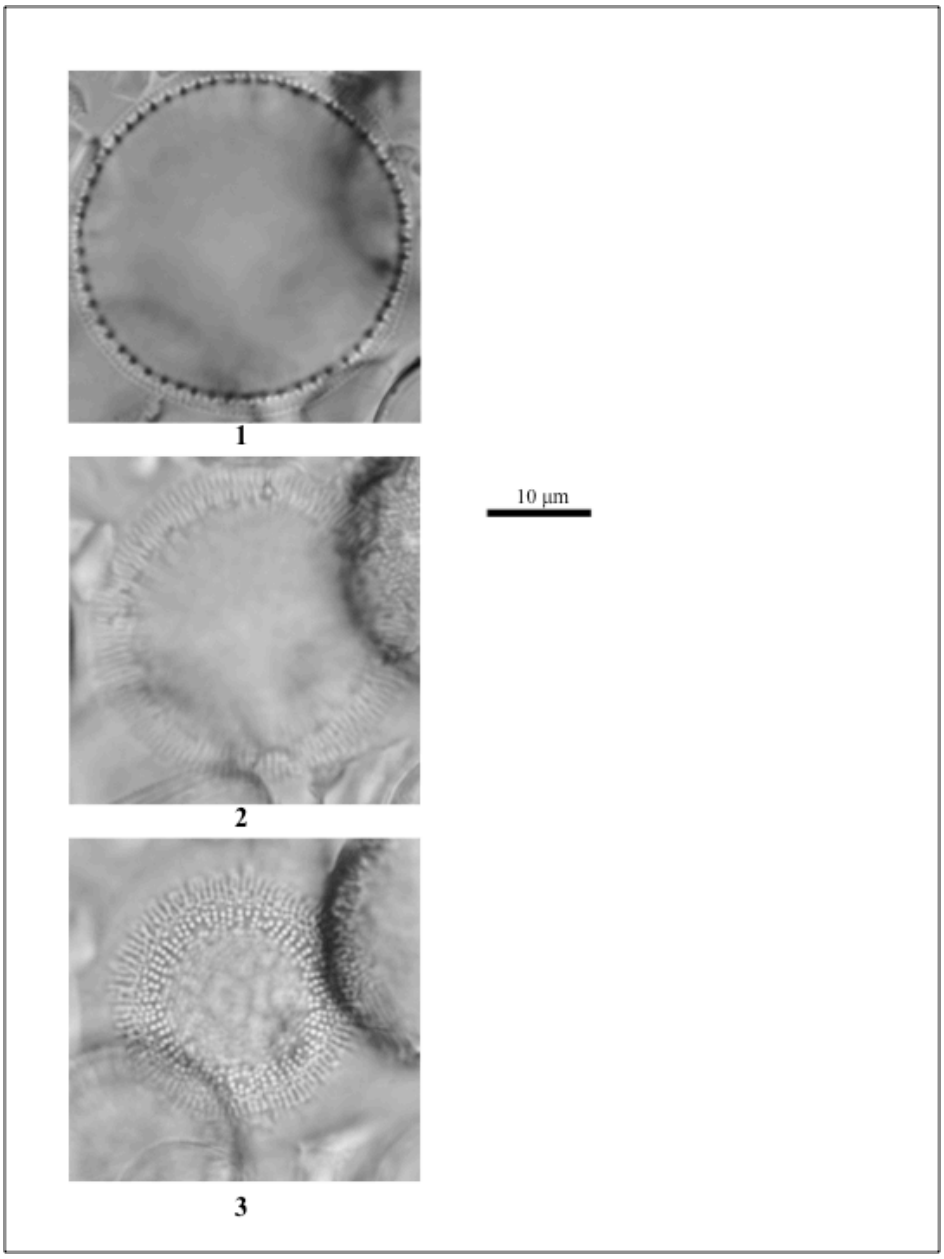


Figure A1.5: Identification plate for *Melosira undulata*

1 – 3: *Melosira undulata* (Ehrenberg) Kützing 1844

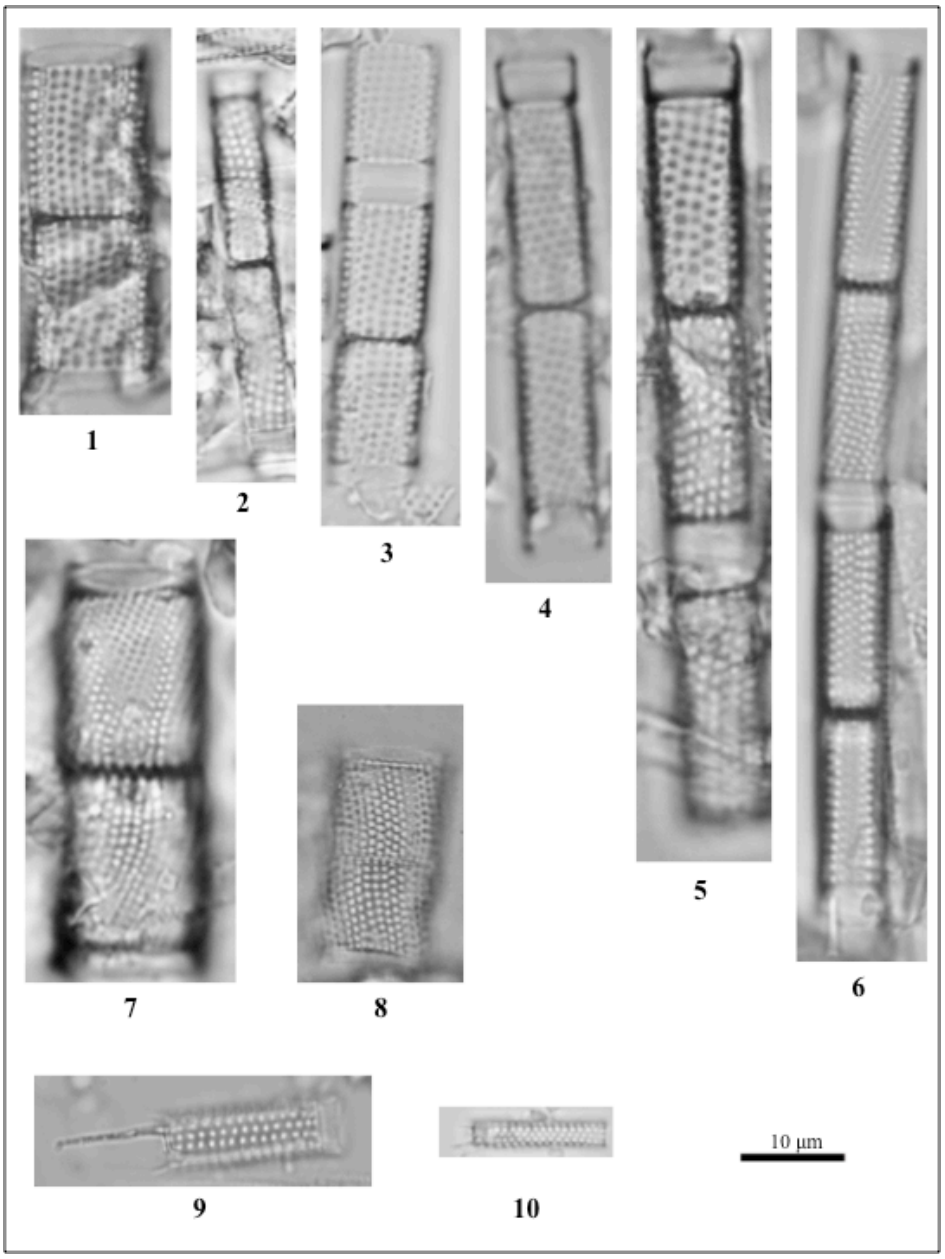


Figure A1.6: Identification plate for *Aulacoseira* spp.

1 – 5: *Aulacoseira granulata* (Ehrenberg) Simonsen 1979

6 – 10: *Aulacoseira ambigua* (Grunow) Simonsen 1979

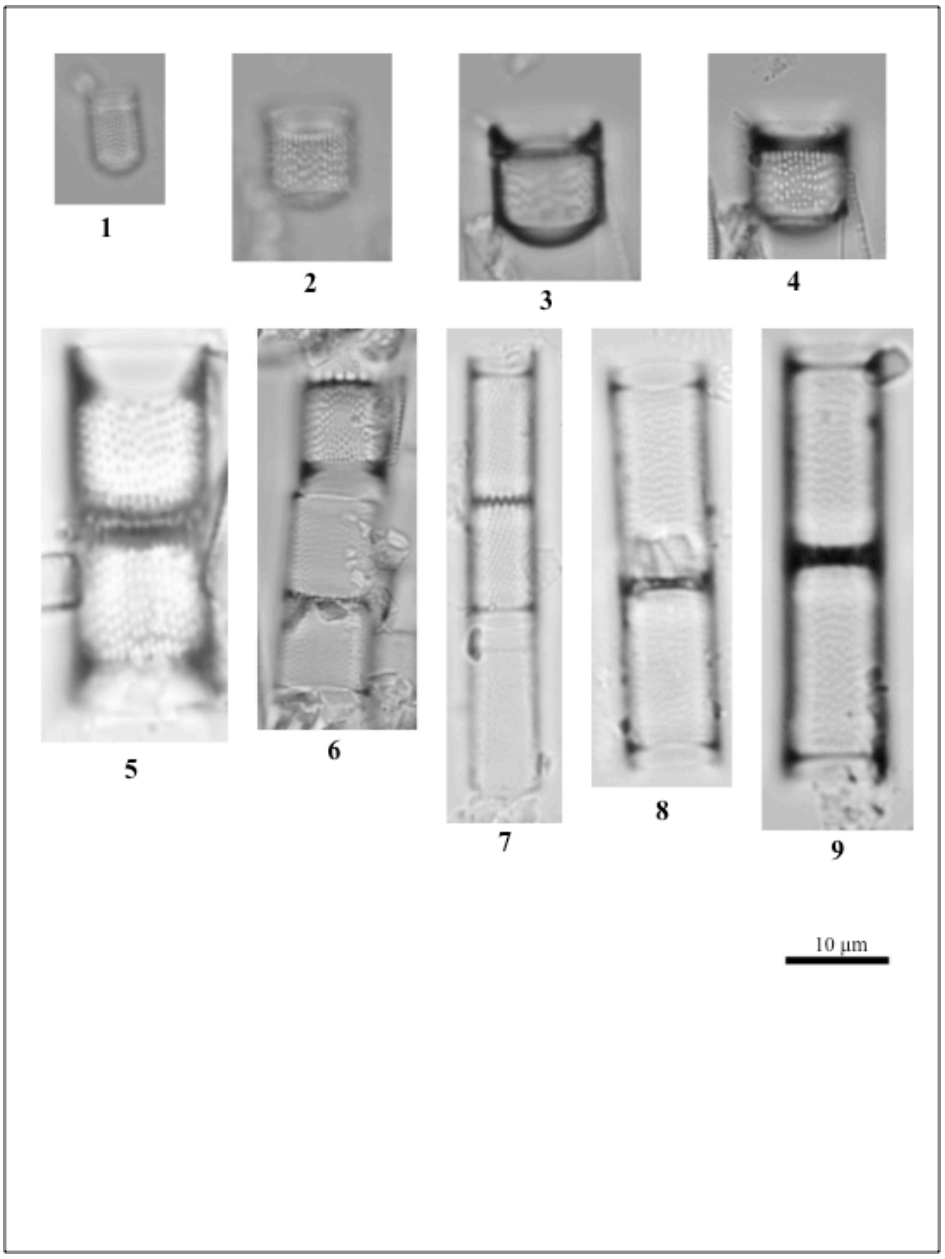


Figure A1.7: Identification plate for *Aulacoseira* spp.

1 – 5: *Aulacoseira crenulata* (Ehrenberg) Thwaites 1848

6 – 7: *Aulacoseira valida* (Grunow in Van Heurck) K. Krammer 1991

8 – 9: *Aulacoseira lacustris* (Grunow in Van Heurck) K. Krammer 1991

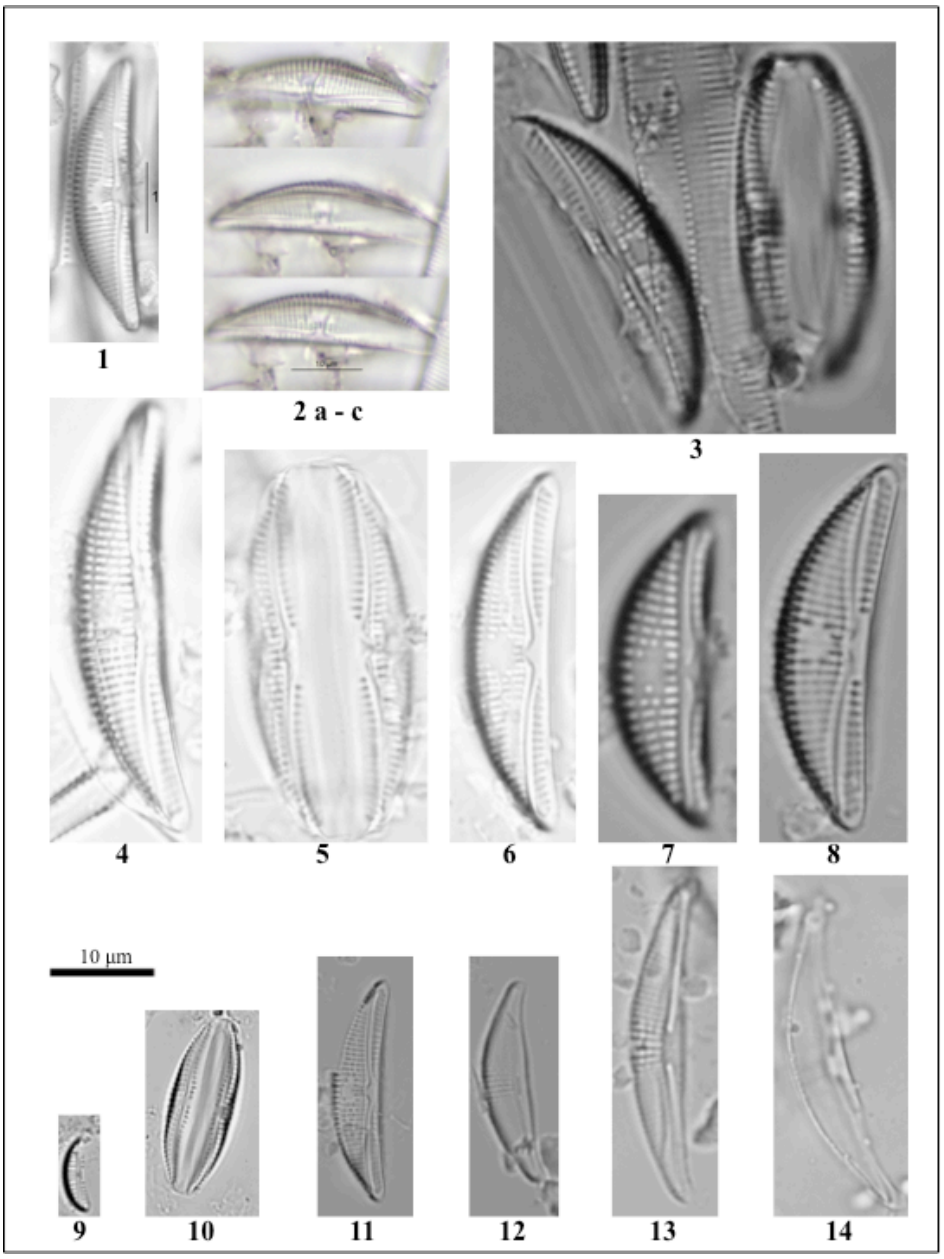


Figure A1.8: Identification plate for *Amphora* spp.

1 – 3: *Amphora copulata* (Kützing) Schoeman & Archibald 1986 – Basionym: *Frustulia copulata* Kützing 1833

4 – 8: *Amphora libyca* Ehrenberg 1840

9: *Amphora pediculus* (Kützing) Grunow in Schmidt et al. 1875

10 – 11: *Amphora affinis* Kützing 1844 – Synonym: *Amphora ovalis* var. *affinis* (Kützing) Van Heurck 1885

12 – 14: *Amphora veneta* Kützing 1844

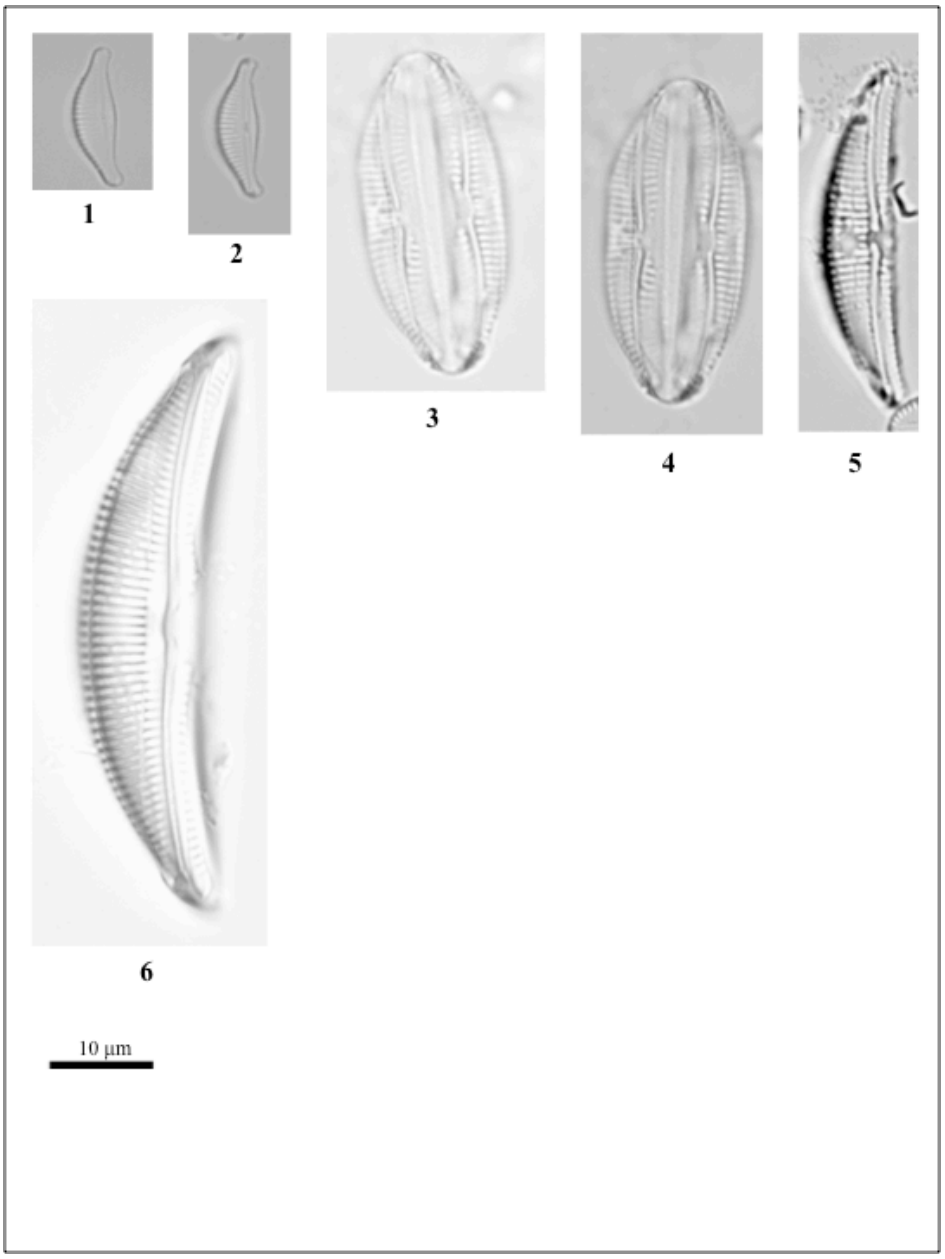


Figure A1.9: Identification plate for *Amphora* spp.

1 – 2: *Amphora thumensis* (A. Mayer) Cleve-Euler 1932

3 – 4: *Amphora affinis* (Girdle view)

5 – 6: *Amphora ovalis* (Kützing) Kützing 1844 – Synonym: *Frustulia ovalis* Kützing 1833

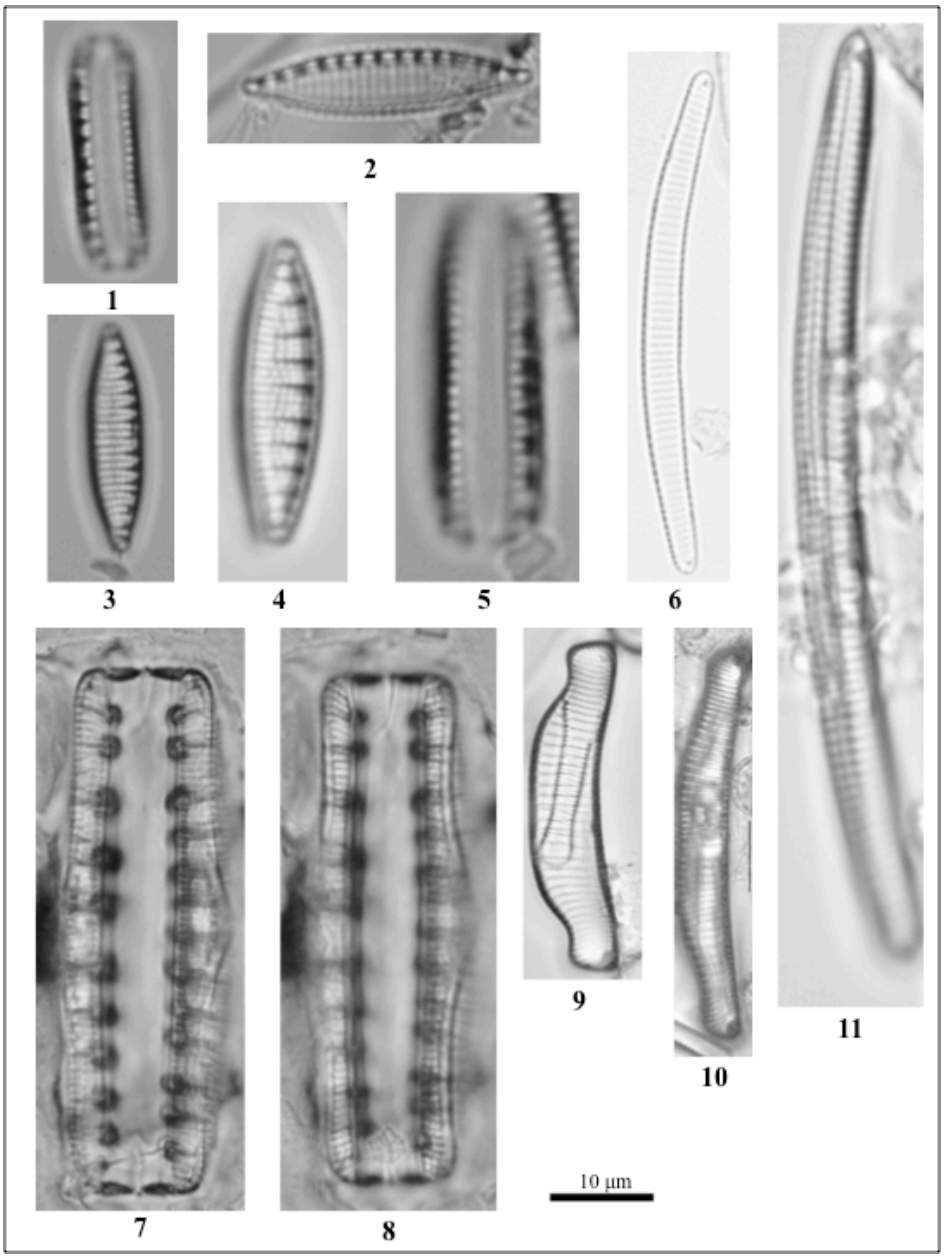


Figure A1.10: Identification plate for *Denticula* spp. and *Eunotia* spp.

1 – 5: *Denticula kuetzingii* Grunow 1862

6, 11: *Eunotia arcus* Ehrenberg 1838

7 – 8: *Denticula valida* (Pedicino) Grunow in Van Heurck 1881

9 – 10: *Eunotia bilunaris* (Ehrenberg) Schaarschmidt 1881

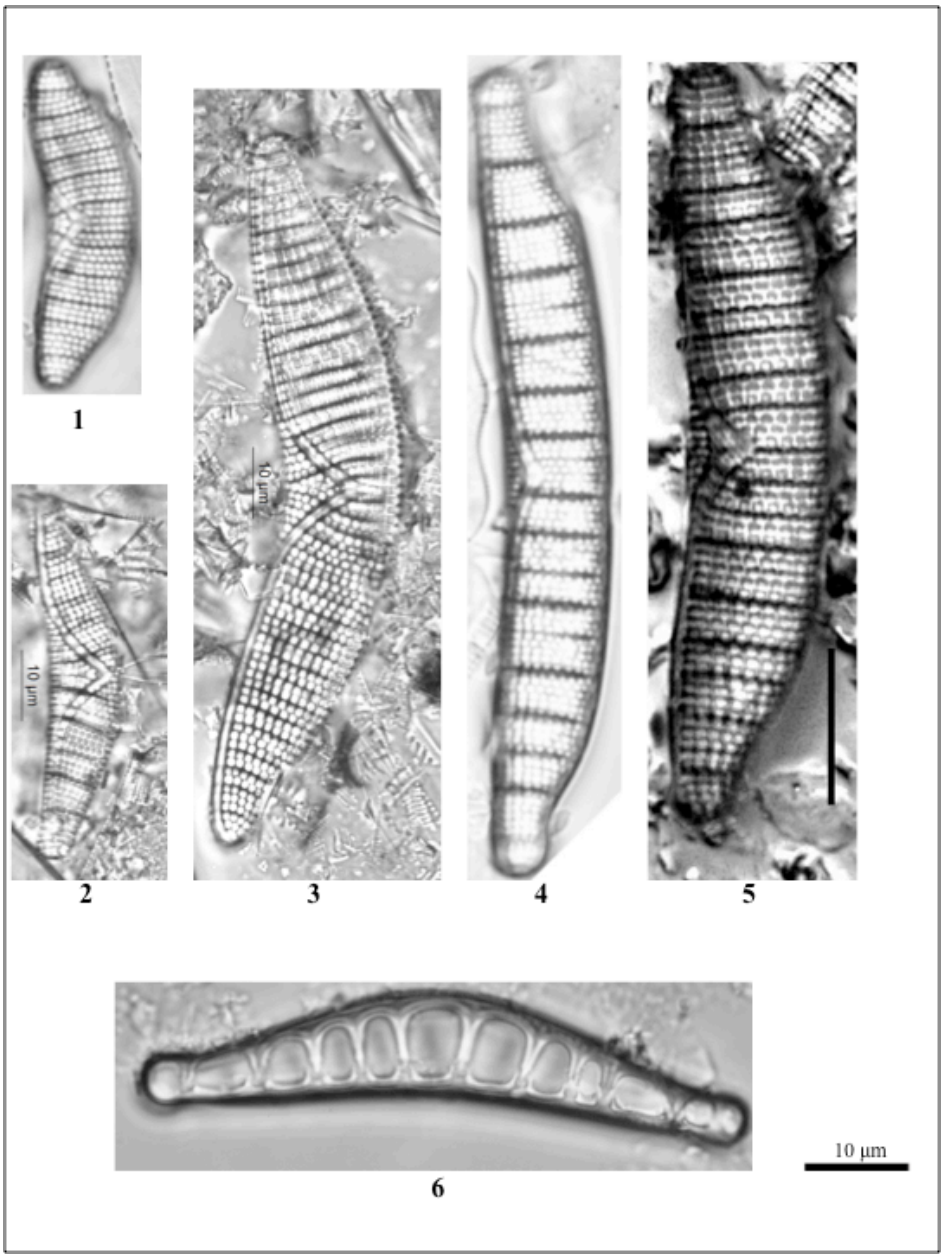


Figure A1.11: Identification plate for *Epithemia* spp.

1 – 2: *Epithemia turgida* (Ehrenberg) Kützing 1844

3: *Epithemia argus* (Ehrenberg) Kützing 1844

4 – 5: *Epithemia adnata* (Kützing) Brébisson 1838

6: *Epithemia* (costae internal view)

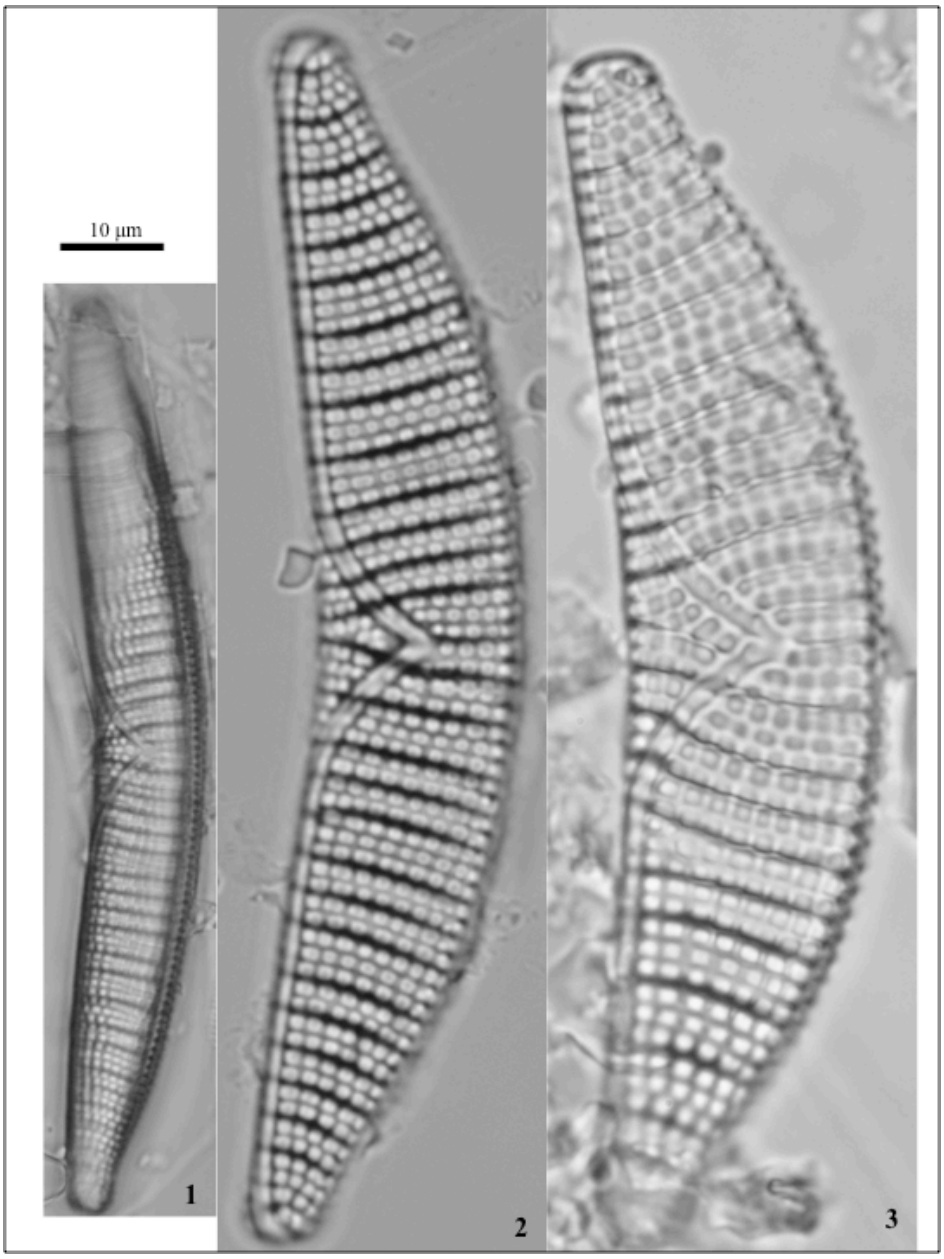


Figure A1.12: Identification plate for *Epithemia* spp.

1: *Epithemia turgida* (Ehrenberg) Kützing 1844

2 – 3: *Epithemia argus* (Ehrenberg) Kützing 1844

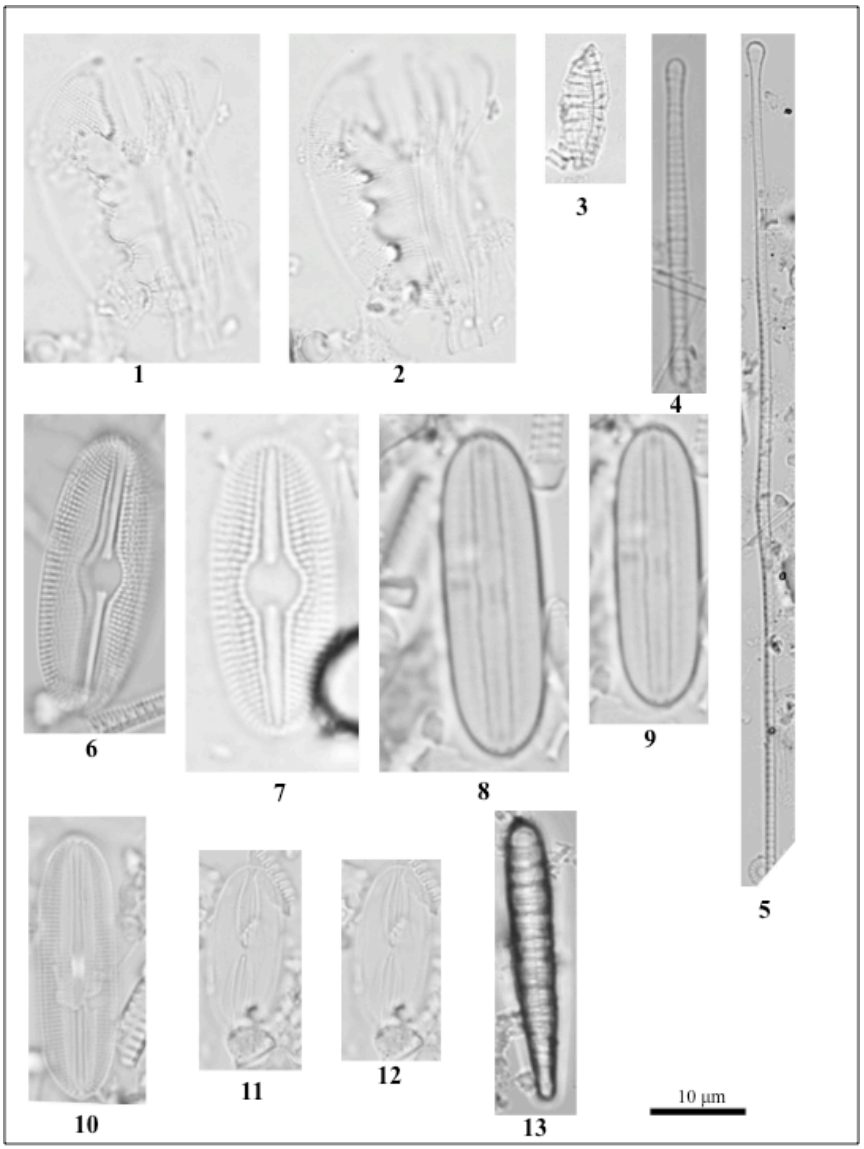


Figure A1.13: Identification plate for *Entomoneis ornata, Diatoma* spp., *Diploneis* spp., and *Meridion circulaire.*

1 – 2: *Entomoneis ornata* (J. W. Bailey) Reimer in Patrick & Reimer 1975 – Synonym: *Amphiprora ornata* J.W. Bailey 1851

3: *Diatoma anceps* (Ehrenberg) Kirchner 1878

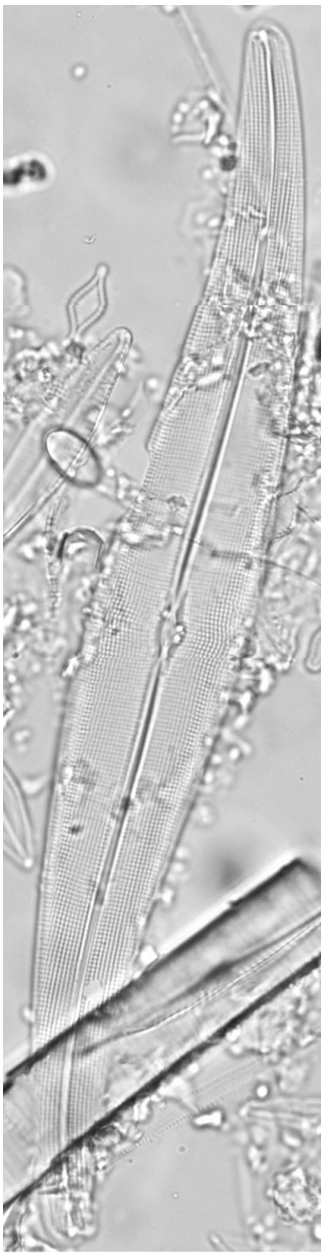
4 – 5: *Diatoma tenue* C. Agardh 1824 – Synonym: *Diatoma elongatum* var. *tenuis* (tenue) (Agardh) Van Heurck 1885

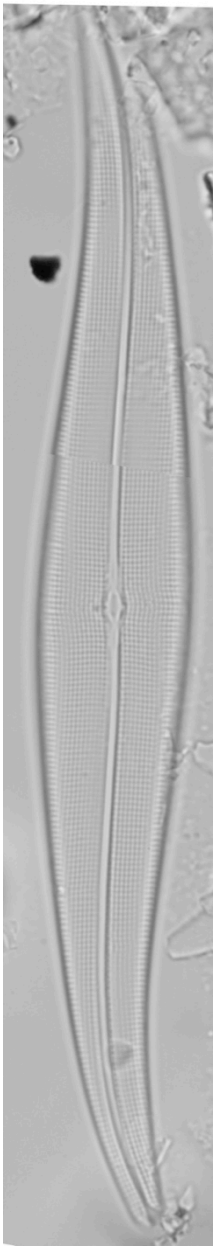
6 – 7: *Diploneis ovalis* (Hilse in Rabenhorst) Cleve 1891

8 – 10: *Diploneis marginestriata* Hustedt 1922

11 – 12: *Diploneis oblongella* (Naegeli ex Kuetzing) Ross 1947 – Synonym: *Navicula oblongella* Naegeli in Kützing 1849

13: *Meridion circulaire* (Greville) Agardh 1831





1 **2**

10 µm

Figure A1.14: Identification plate for *Gyrosigma acuminatum*

1 – 2: *Gyrosigma acuminatum* (Kützing) Rabenhorst 1853

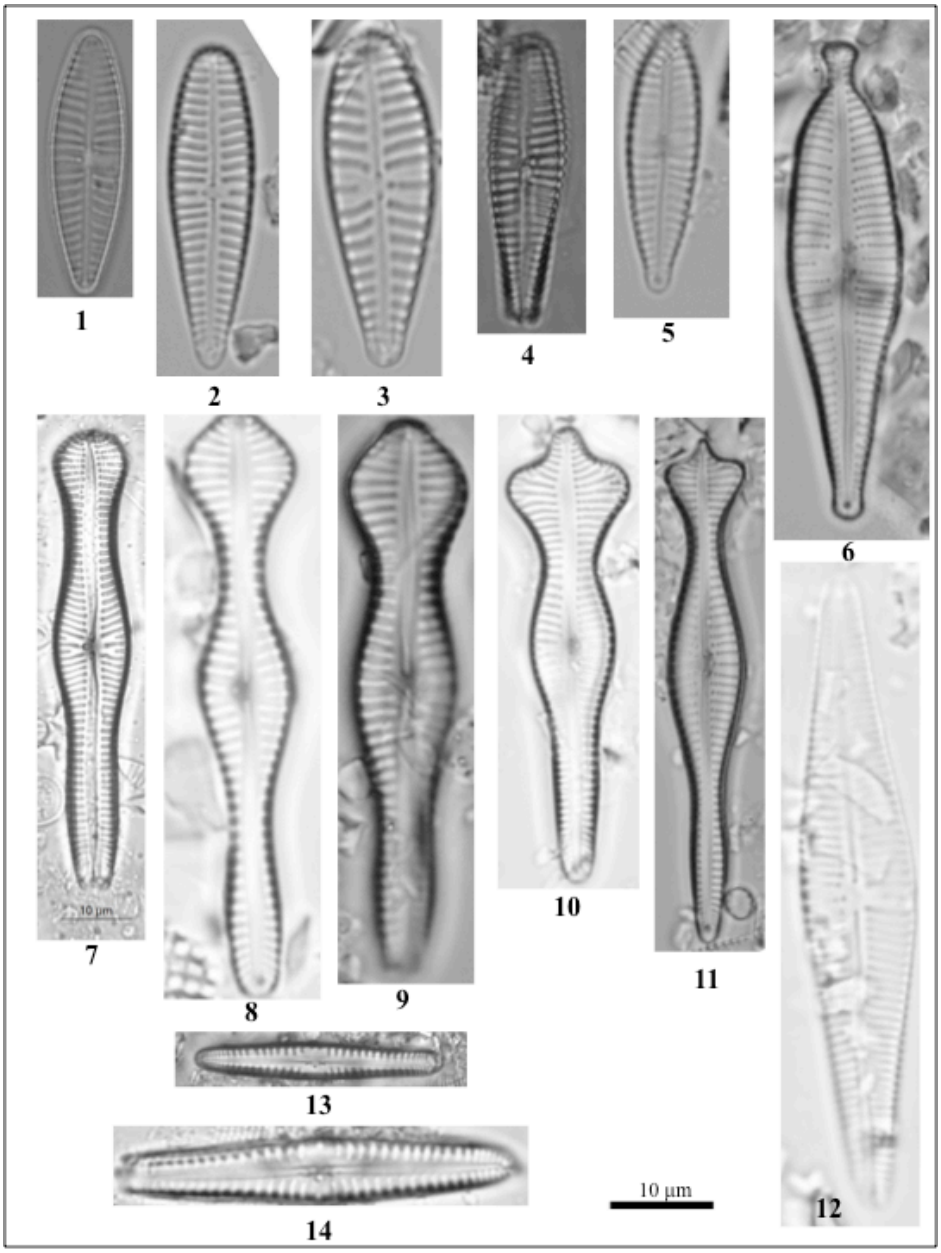


Figure A1.15: Identification plate for *Gomphonema* spp.

1 – 3: *Gomphonema parvula* (*parvulum*) Rabenhorst 1853

4: *Gomphonema angustum* Agardh 1831

5 – 6: *Gomphonema sphaerophorum* Ehrenberg 1845

7 – 9: *Gomphonema truncatum* Ehrenberg 1832

10 – 11: *Gomphonema acuminatum* Ehrenberg 1832

12: *Gomphonema gracile* Ehrenberg 1838

13 – 14: *Gomphonema intricatum* Kützing 1844

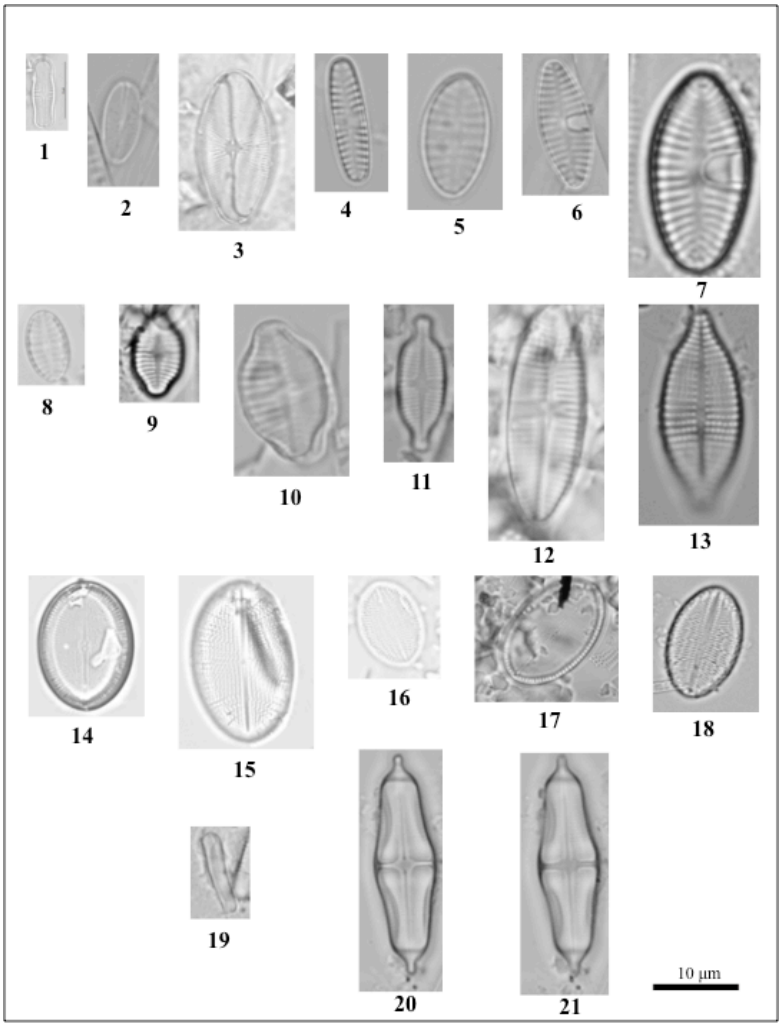


Figure A1.16: Identification plate for *Achnanthes* spp. and *Cocconeis* spp. and *Navicula smithii*

1: *Achnanthes rosenstockii* Lange-Bertalot in Lange-Bertalot & Krammer 1989

2 – 3: *Achnanthes flexella* (Kützing) Brun 1880

4: *Achnanthes lanceolata* (Brébisson ex Kützing) Grunow in Cleve & Grunow 1880

5 – 7: *Achnanthes lanceolata* ssp. *frequentissima* Lange-Bertalot 1993

8: *Achnanthes conspicua* Mayer 1919

9 – 10: *Achnanthes exigua* Grunow in Cleve & Grunow 1880

11: *Achnanthes exigua* var. *heterovalvata* Krasske 1923

12: *Achnanthes helvetica* (Hustedt) Lange-Bertalot in Lange-Bertalot & Krammer 1989

13: *Achnanthes clevei* Grunow in Cleve & Grunow 1880

14: *Cocconeis placentula* var. *lineata* (Ehrenberg) Van Heurck 1885

15 – 18: *Cocconeis placentula* Ehrenberg 1838

19: *Achnanthes minutissima* Kützing 1833

20 - 21: *Navicula smithii* (Agardh) Van Heurck 1896

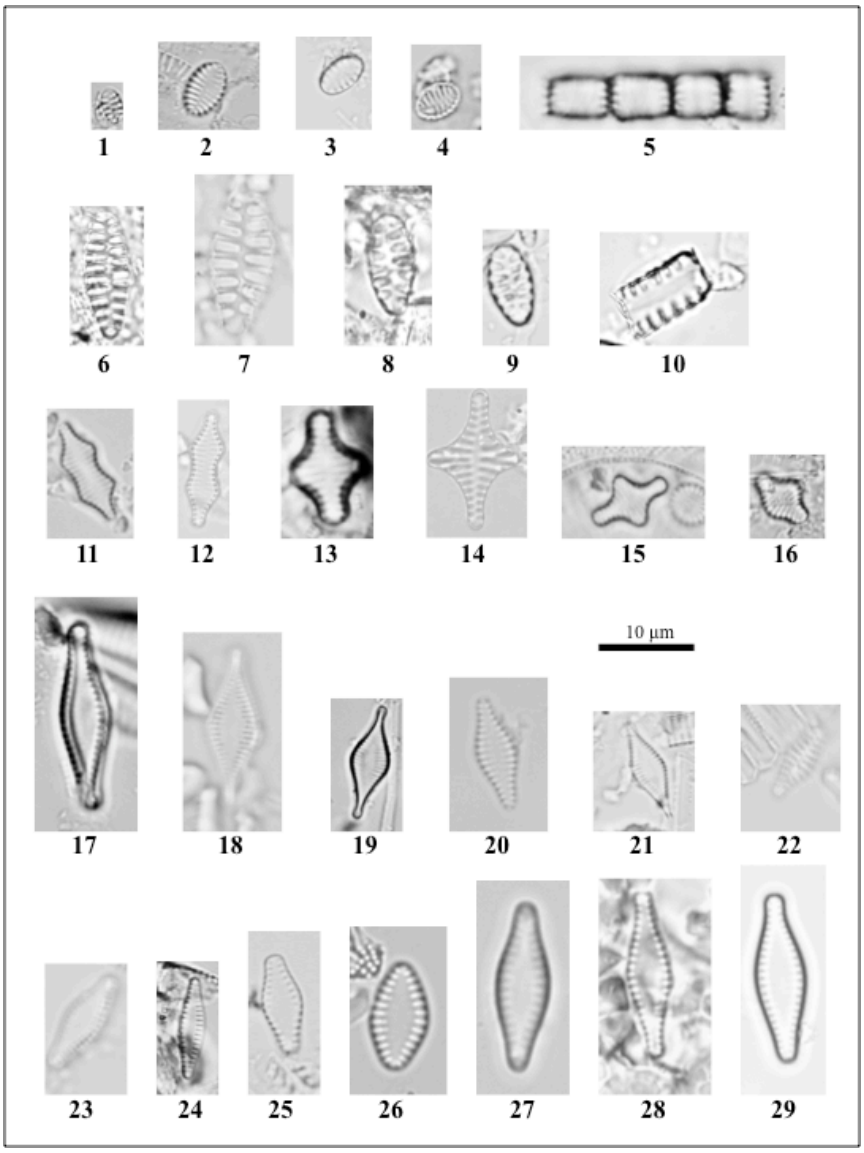


Figure A1.17: Identification plate for *Fragilaria* spp*., Pseudostaurosira parasitica, Staurosirella pinnata*, and *Martyana martyii*

1 – 4: *Staurosirella pinnata* Ehrenberg Williams & Round 1987 – Basionym: *Fragilaria pinnata* Ehrenberg 1843

5: *Staurosirella pinnata* (girdle view)

6 – 10: *Martyana martyii* (Héribaud) Round in Round, Crawford & Mann 1990

11 – 12: *Fragilaria constricta* Ehrenberg 1843

13 – 16: *Fragilaria construens* (Ehrenberg) Grunow 1862

17 – 21: *Pseudostaurosira parasitica* (W. Smith) E.A. Morales 2003 – Synonym: *Fragilaria parasitica* (W. Smith) Heiberg 1863

22: *Fragilaria pinnata* var. *lancettula* (Schumann) Hustedt in Schmidt et al. 1913

23 – 28: *Fragilaria brevistriata* Grunrow in Van Heurck 1885

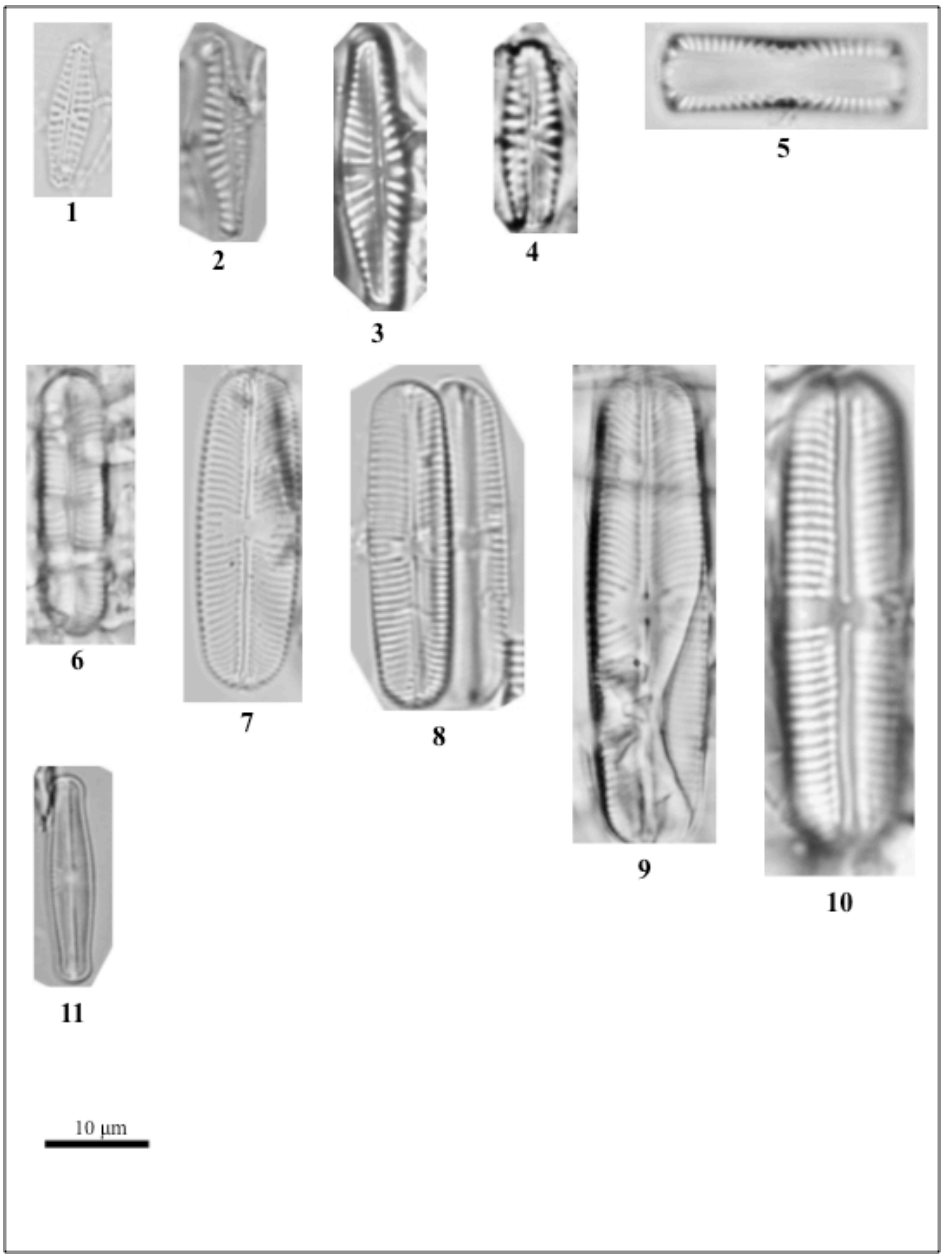


Figure A1.18: Identification plate for *Hippodonta* spp. and *Navicula* spp.

1, 4: *Hippodonta capitata* (Ehrenberg) Lange-Bertalot, Metzeltin & Witkowski 1996 – Synonym: *Navicula capitata* Ehrenberg 1838

2 – 3: *Hippodonta hungarica* (Grunrow) Lange-Bertalot, Metzeltin & Witkowski 1996 – Synonym: *Navicula capitata* var. *hungarica* (Grunrow) Ross 1947

5: *Hippodonta hungarica*- Girdle view

6 – 10: *Navicula laevissima* Kützing 1844

11: *Navicula disjuncta* Hustedt 1930

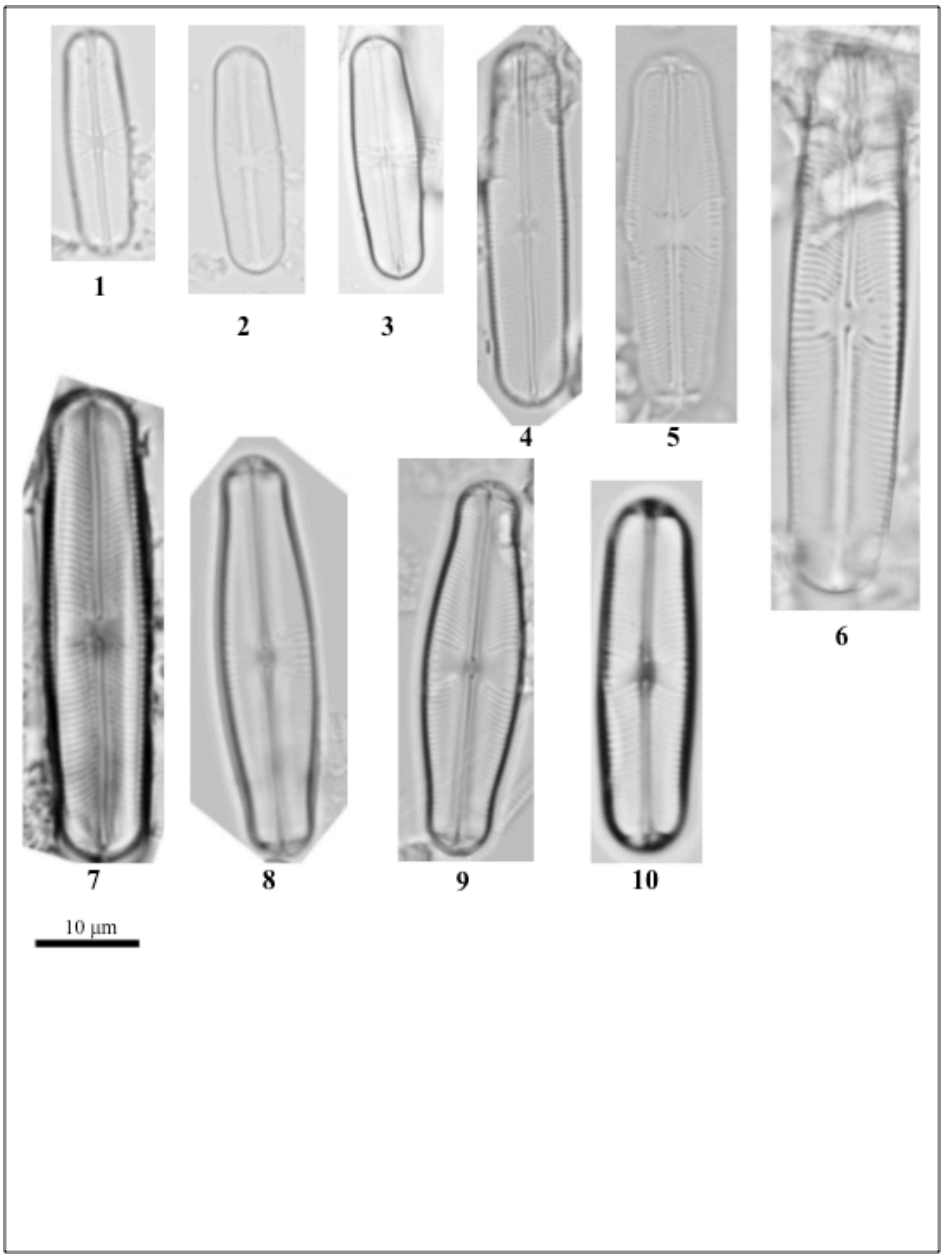


Figure A1.19: Identification plate for *Navicula pupula*

1 – 10: *Navicula pupula* Kützing 1844

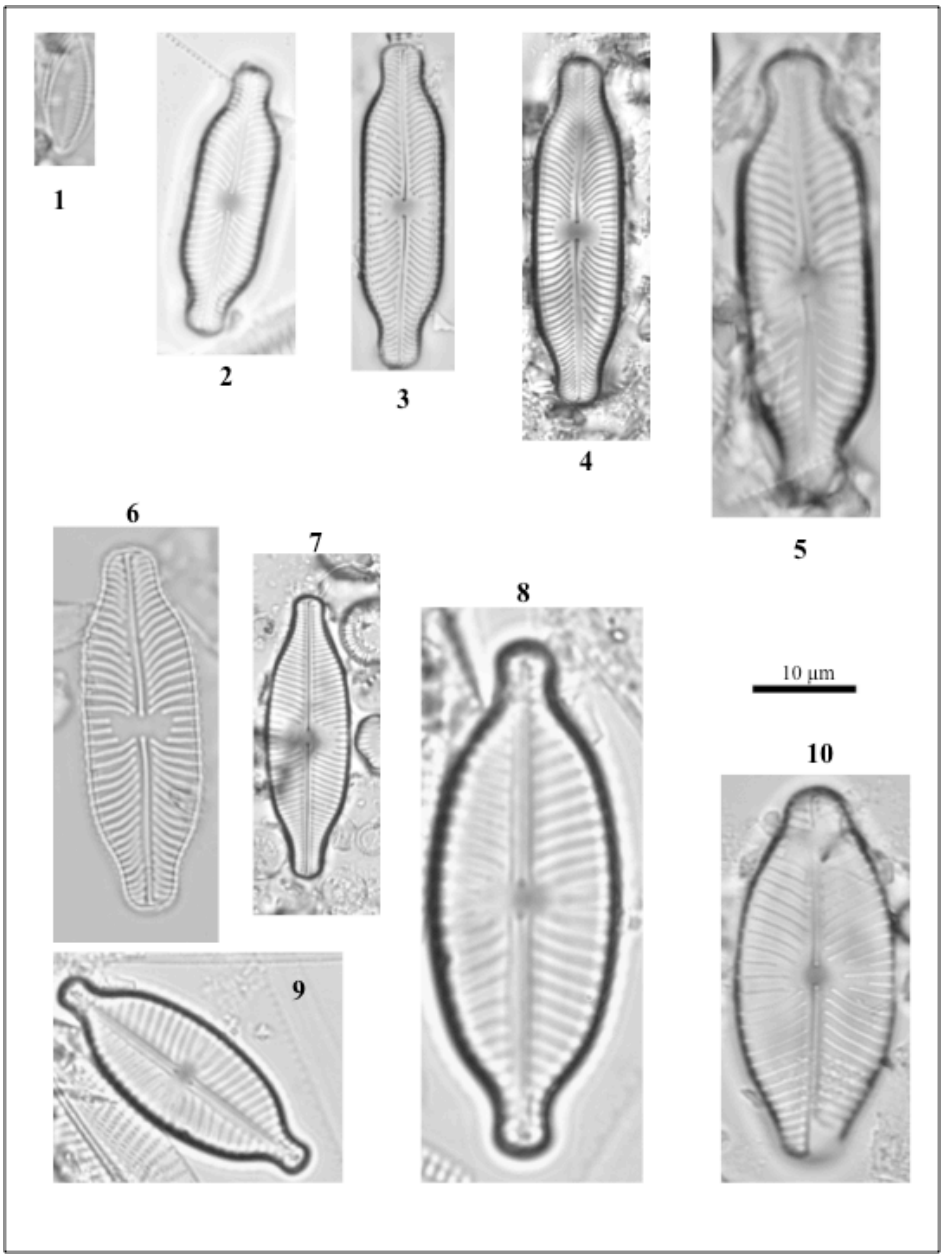


Figure A1.20: Identification plate for *Navicula* spp. and *Pinnularia exigua*

1: *Navicula minima* Grunow in Van Heurck 1880

2 – 5: *Navicula abiskoensis* Hustedt 1942 – Synonym: *Placoneis abiskoensis* (Hustedt) Lange-Bertalot et Metzeltin in Metseltin & Witkowski 1996

6 – 7: *Navicula explanata* Hustedt 1948

8 – 9: *Pinnularia exigua* Gregory 1854 – Synonym: *Navicula exigua* (Gregory) Grunrow in van Heurck 1880

10: *Navicula gastrum* (Ehrenberg) Kützing 1844

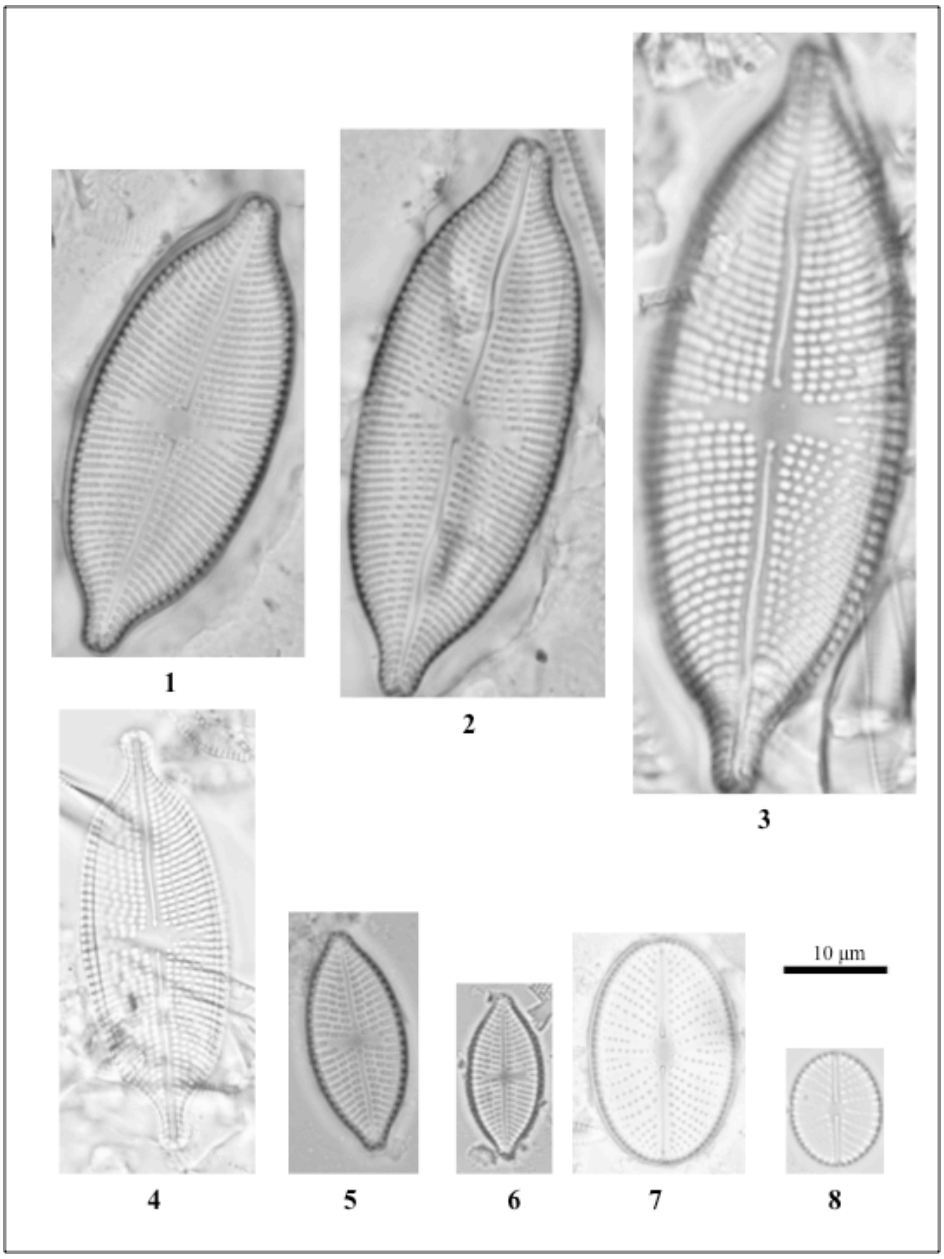


Figure A1.21: Identification plate for *Aneumastus* spp. and *Navicula scutelloides*

1 – 4: *Anuemastus tuscula* (Ehrenberg) Mann & Stickle in Round, Crawford & Mann 1990

– Synonym: *Navicula tuscula* Ehrenberg 1840

5 – 6: *Aneumastus minor* Lange-Bertalot 1993

7 – 8: *Navicula scutelloides* Smith ex Gregory 1856

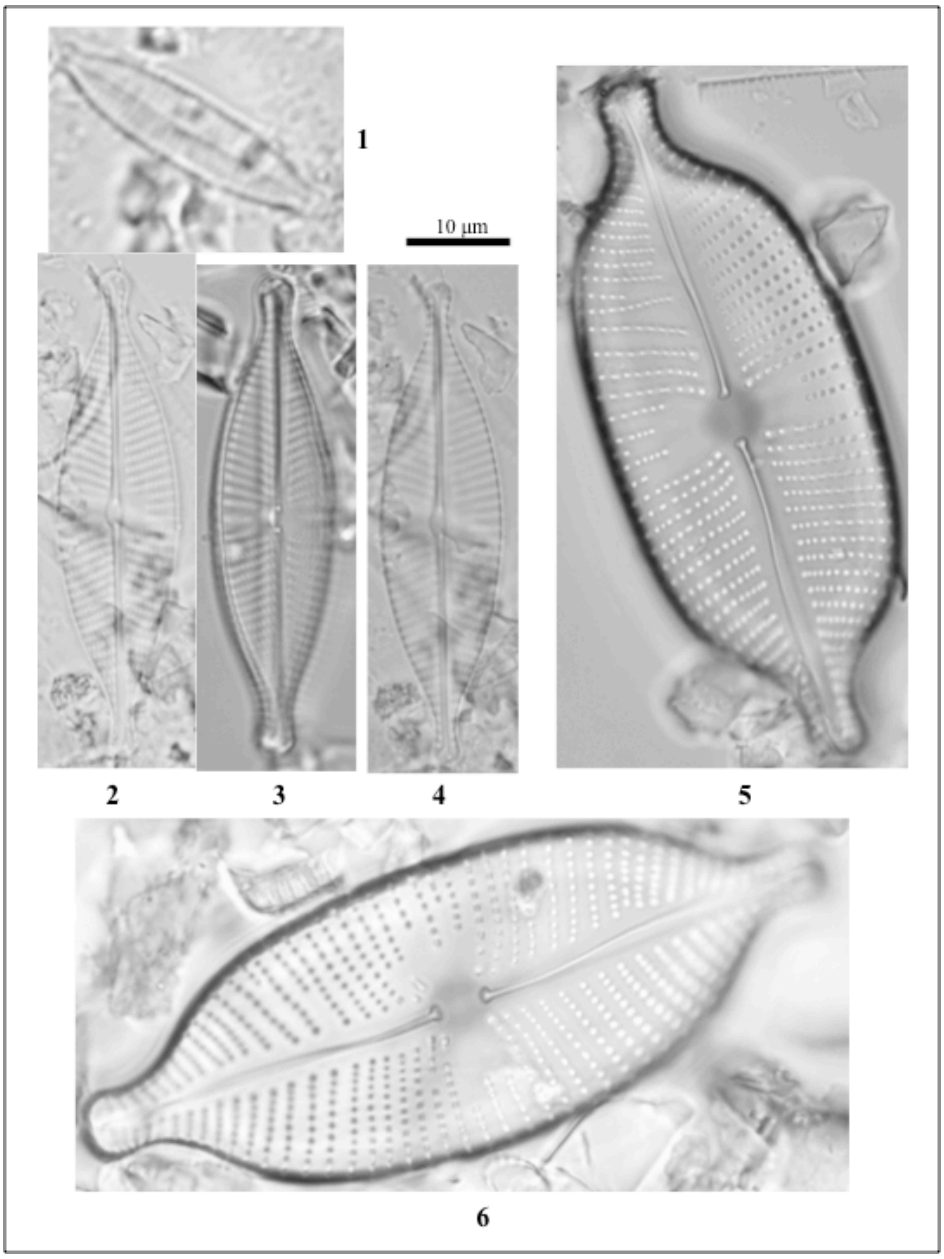


Figure A1.22: Identification plate for *Navicula* spp.

1: *Navicula accommoda (accomoda)* Hustedt 1950

2 – 4: *Navicula rhynchocephala (rhyncocephala)* Kützing 1844 – Synonym: *Navicula rhynchotella* Lange-Bertalot 1993, although not usually separated from *Navicula rhynchocephala*

5 – 6: *Navicula amphibola* Cleve 1891

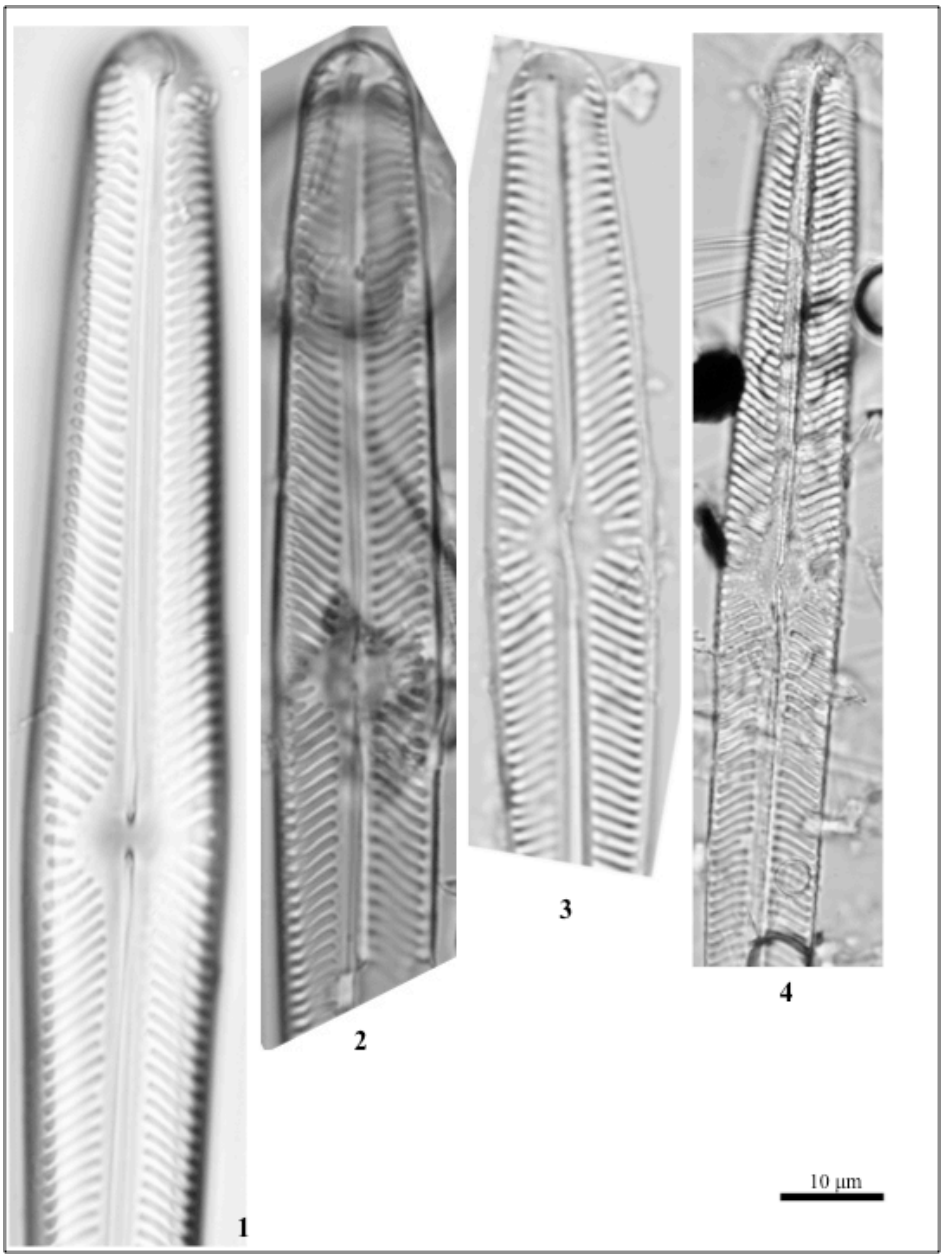


Figure A1.23: Identification plate for *Navicula oblonga*

1 – 4: *Navicula oblonga* (Kützing) Kützing 1844

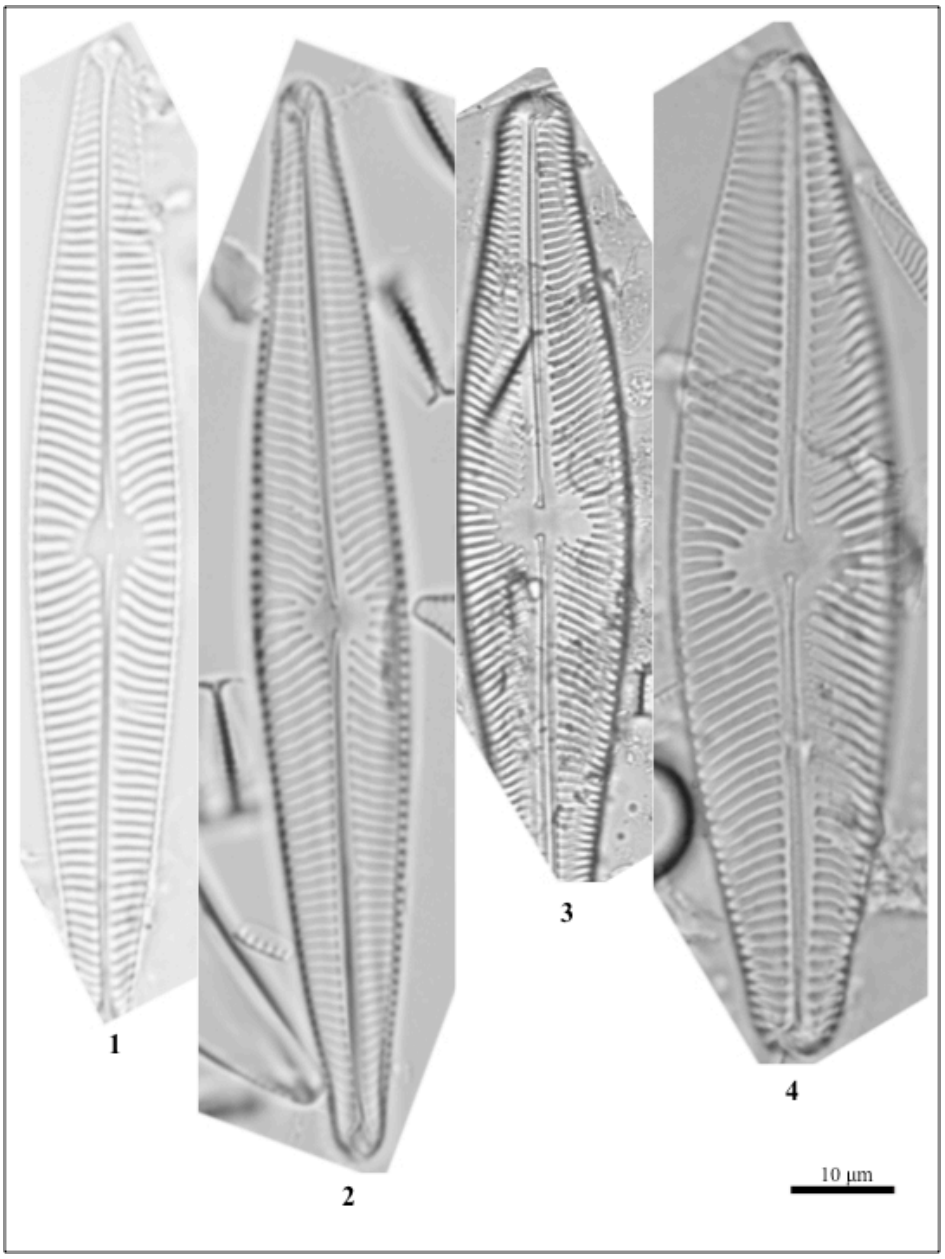


Figure A1.24: Identification plate for *Navicula* spp.

1 – 2: *Navicula radiosa* Kützing 1844

3 – 4: *Navicula aurora* Sovereign 1958

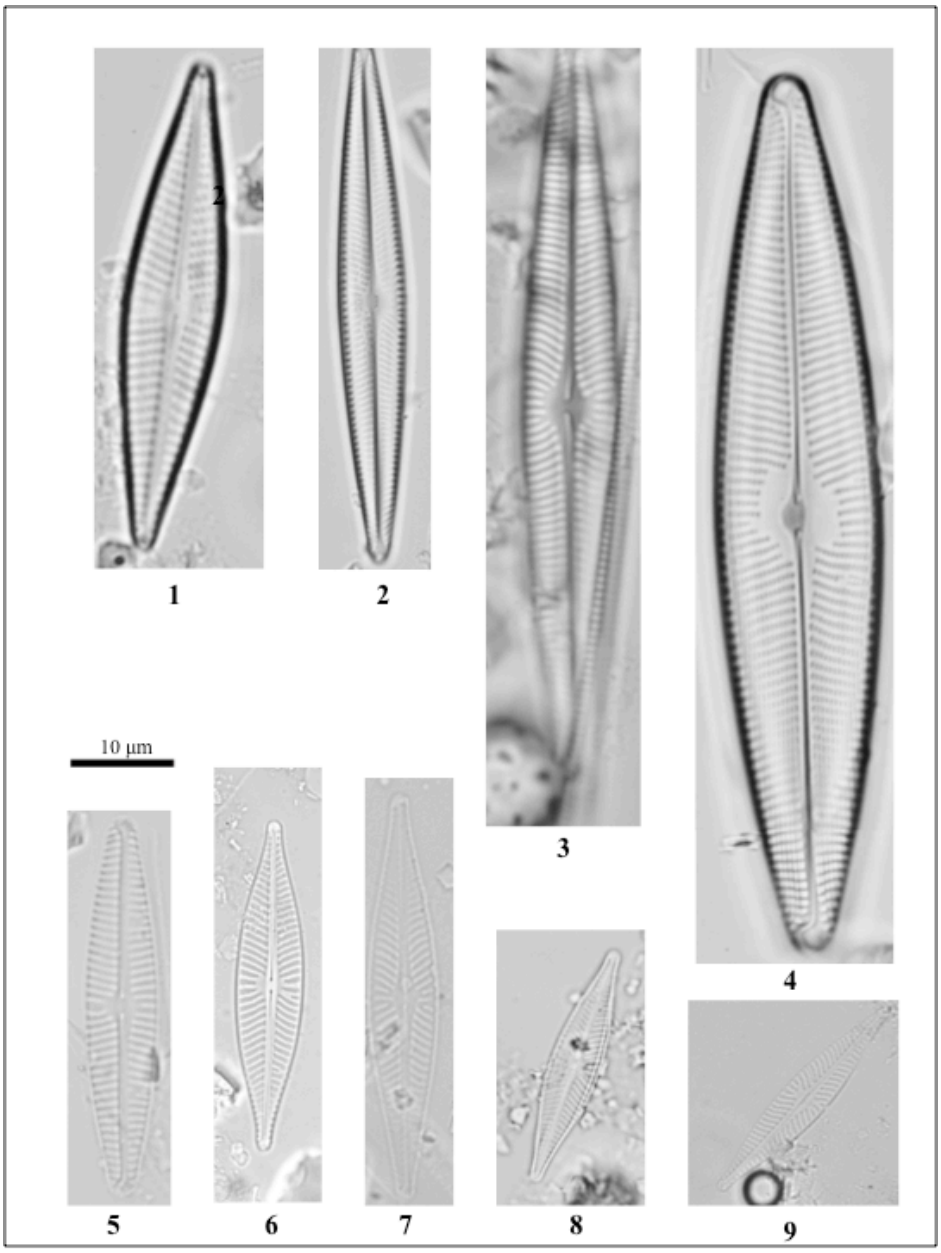


Figure A1.25: Identification plate for *Navicula* spp.

1: *Navicula rhynchocephala (rhyncocephala)* Kützing 1844

2 – 3: *Navicula radiosa* Kützing 1844

4: *Navicula lanceolata* (Agardh) Kützing 1844

5: *Naviculala recens* (Lange-Bertalot) Lange-Bertalot in Krammer & Lange-Bertalot 1985

6 – 9: *Navicula trivialis* Lange-Bertalot 1980

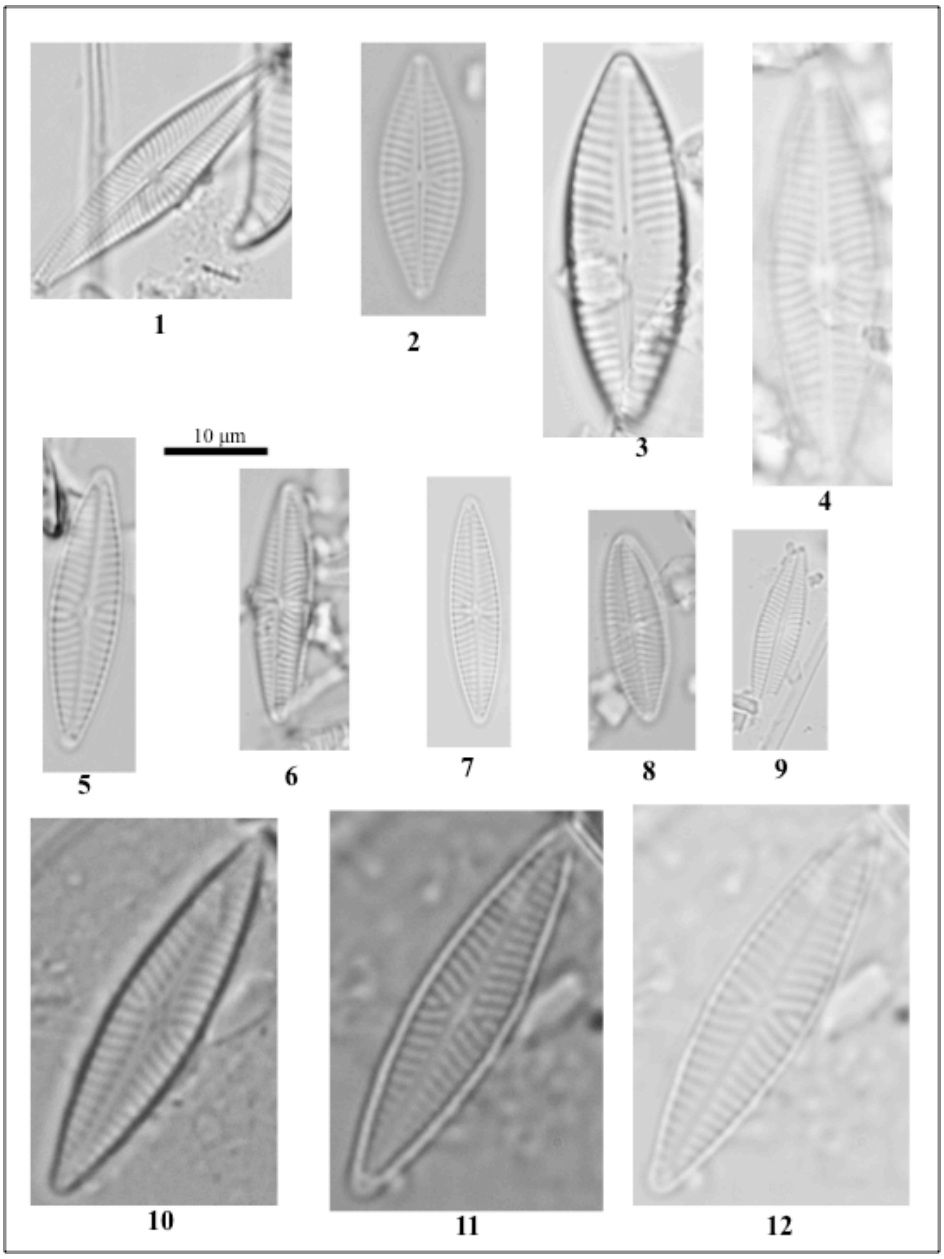


Figure A1.26: Identification plate for *Navicula* spp.

1, 6 – 12: *Navicula cryptotenella* Lange-Bertalot in Krammer & Lange-Bertalot 1985

2 – 5: *Navicula trivialis* Lange-Bertalot 1980

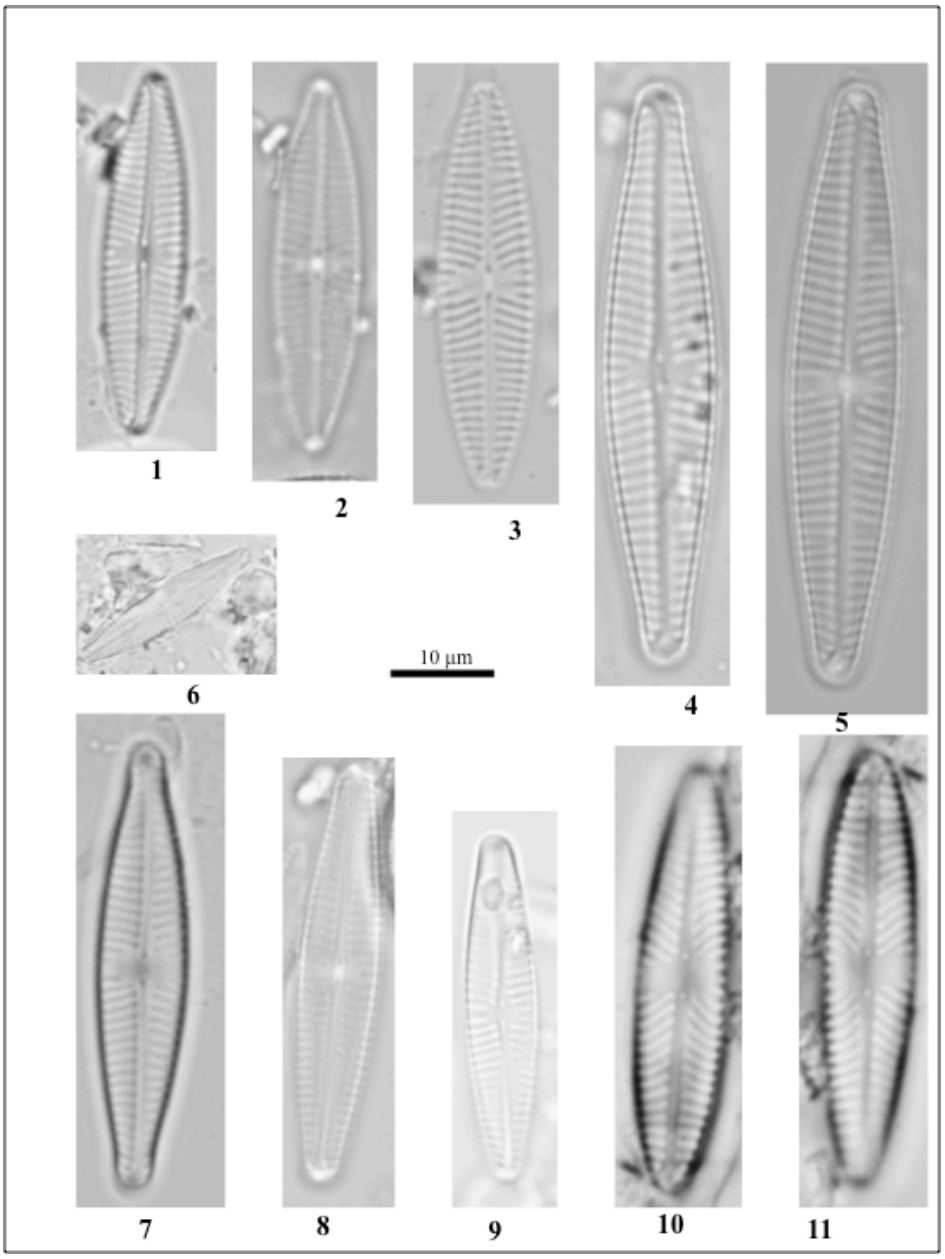


Figure A1.27: Identification plate for *Navicula* spp.

1 – 5: *Navicula veneta* Kützing 1844

6: *Navicula rhynchocephala (rhyncocephala)* Kützing 1844

7: *Navicula cryptocephala* Kützing 1844

8 – 9: *Navicula libonensis* Schoeman *emend* Schoeman 1969

10 – 11: *Navicula cari* Ehrenberg 1836

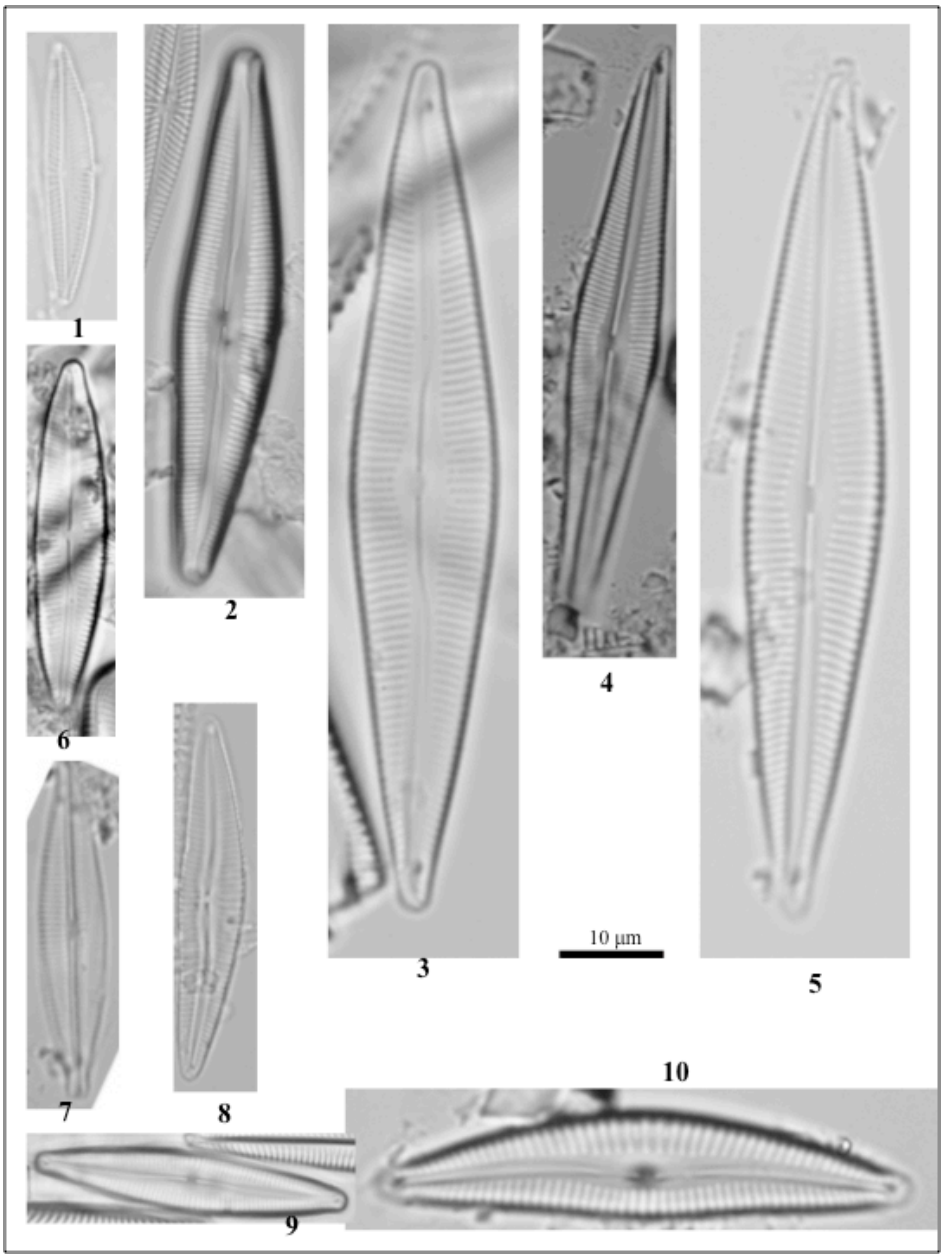


Figure A1.28: Identification plate for *Cymbella* spp.

1 – 5: *Cymbella delicatula* Kützing 1849

6 – 10: *Cymbella cesatii* (Rabenhorst) Grunow in Schmidt et al. 1881

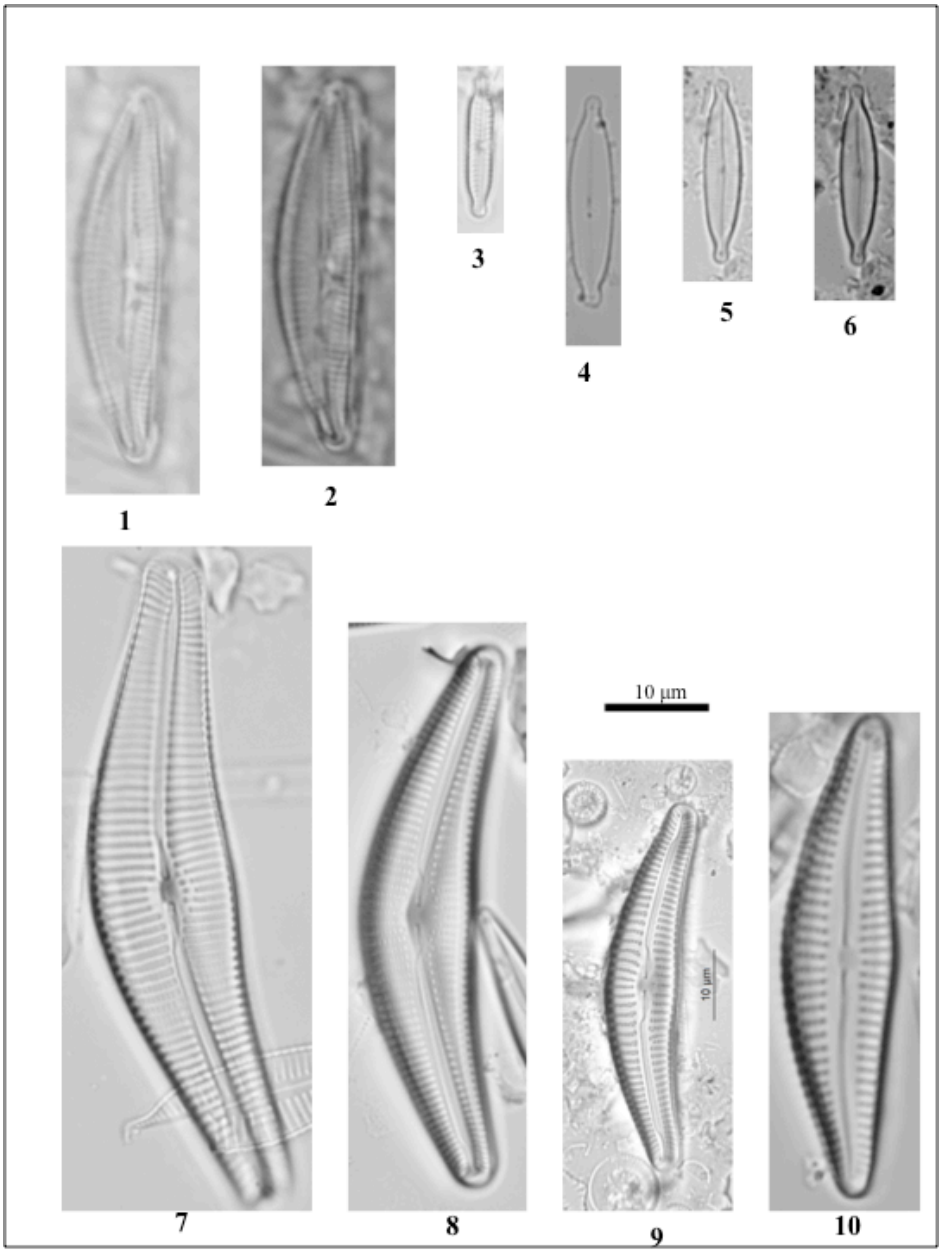


Figure A1.29: Identification plate for *Cymbella* spp.

1 – 2: *Cymbella hantzschiana* K. Krammer 2002

3: *Cymbella microcephala* Grunow in Van Heurck 1885

4 – 6: *Cymbella descripta* (Hustedt) Krammer & Lange-Bertalot 1985

7 – 9: *Cymbella neocistula* K. Krammer 2002

10: *Cymbella leptoceros* (Ehrenberg) Kützing 1844

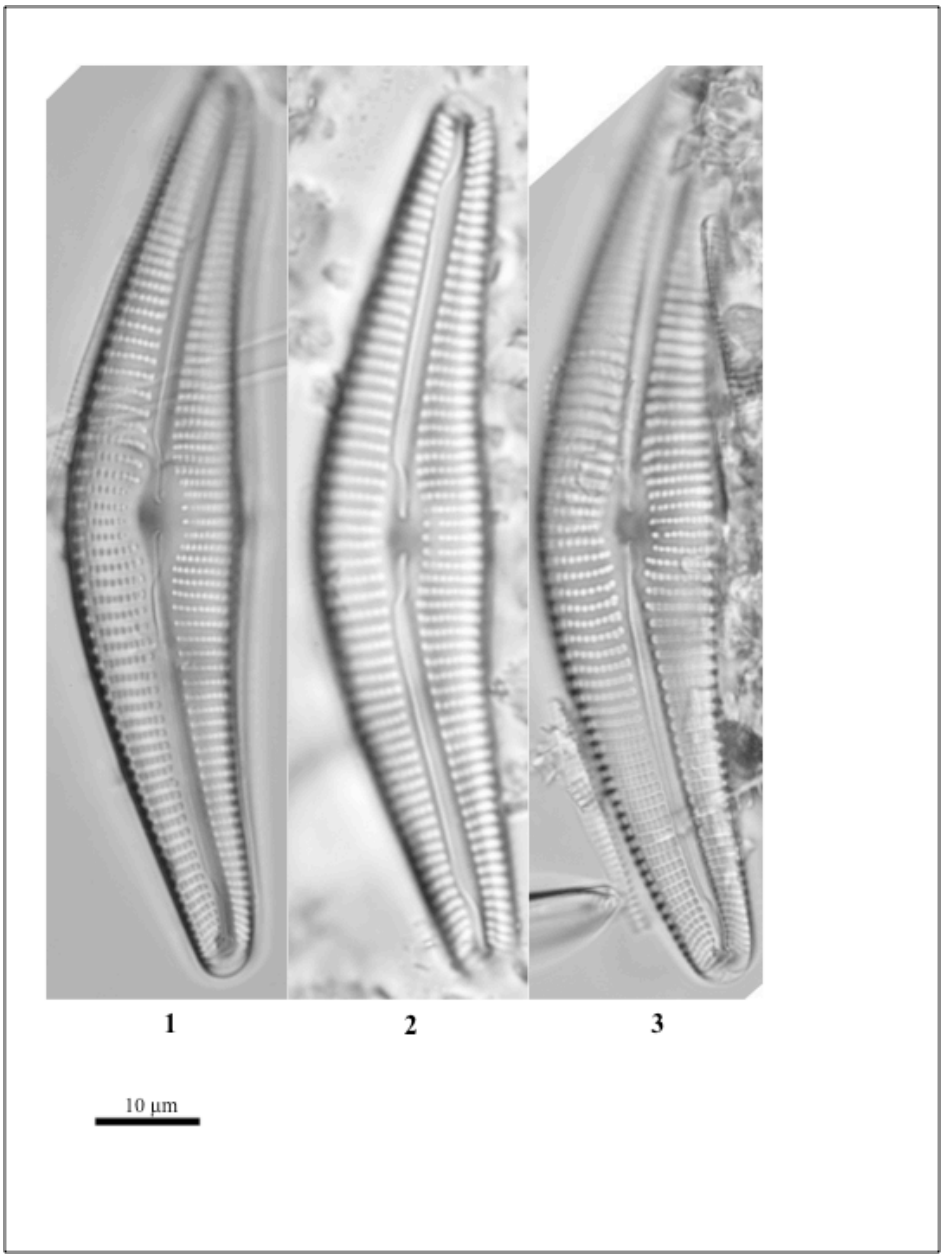


Figure A1.30: Identification plate for *Cymbella* spp.

1: *Cymbella cymbiformis* var. *nonpunctata* Fontell 1917

2 – 3: *Cymbella subcistula* K. Krammer 2002

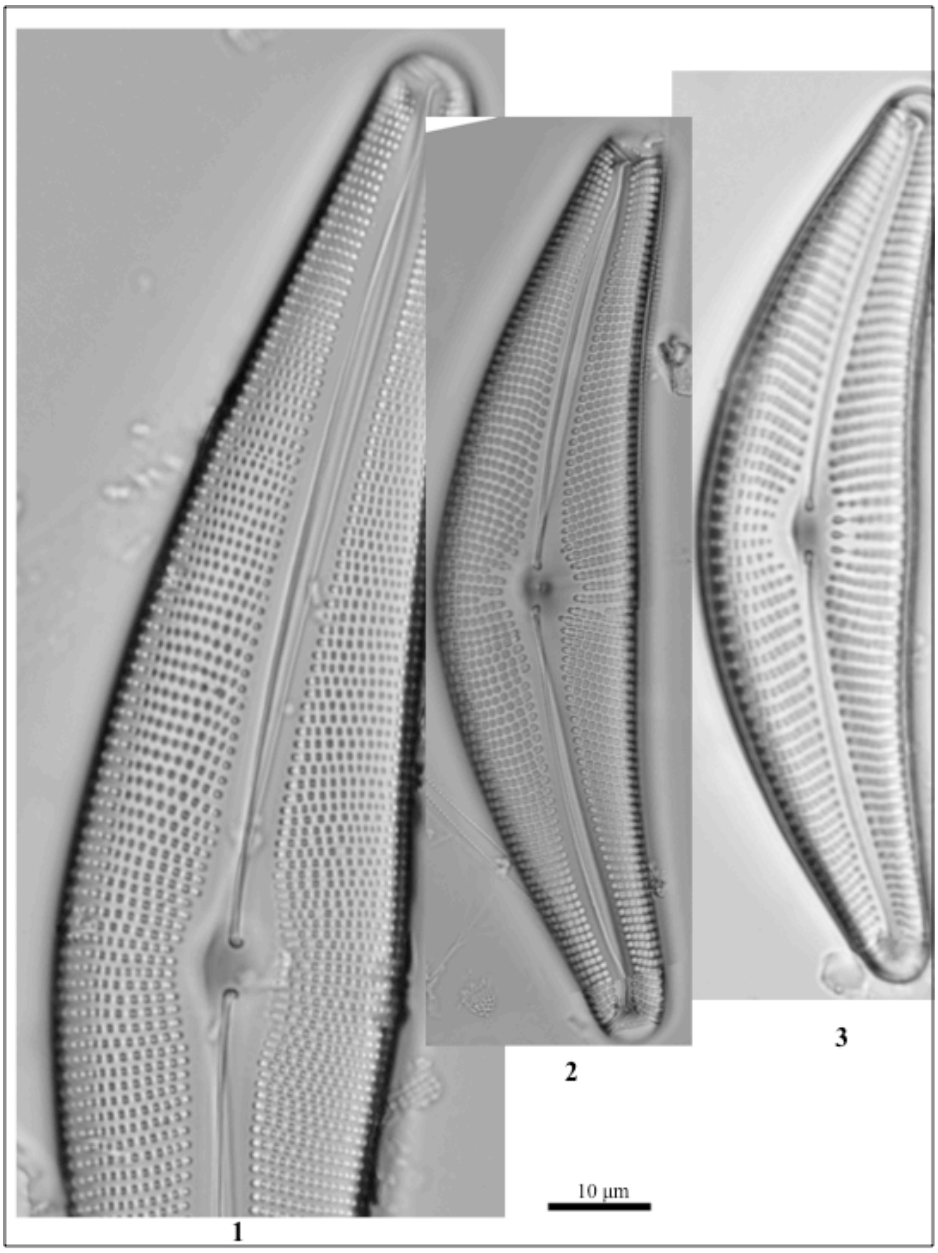


Figure A1.31: Identification plate for *Cymbella* spp.

1: *Cymbella helvetica* Kützing 1844

2 – 3: *Cymbella proxima* Reimer in Patrick & Reimer 1975

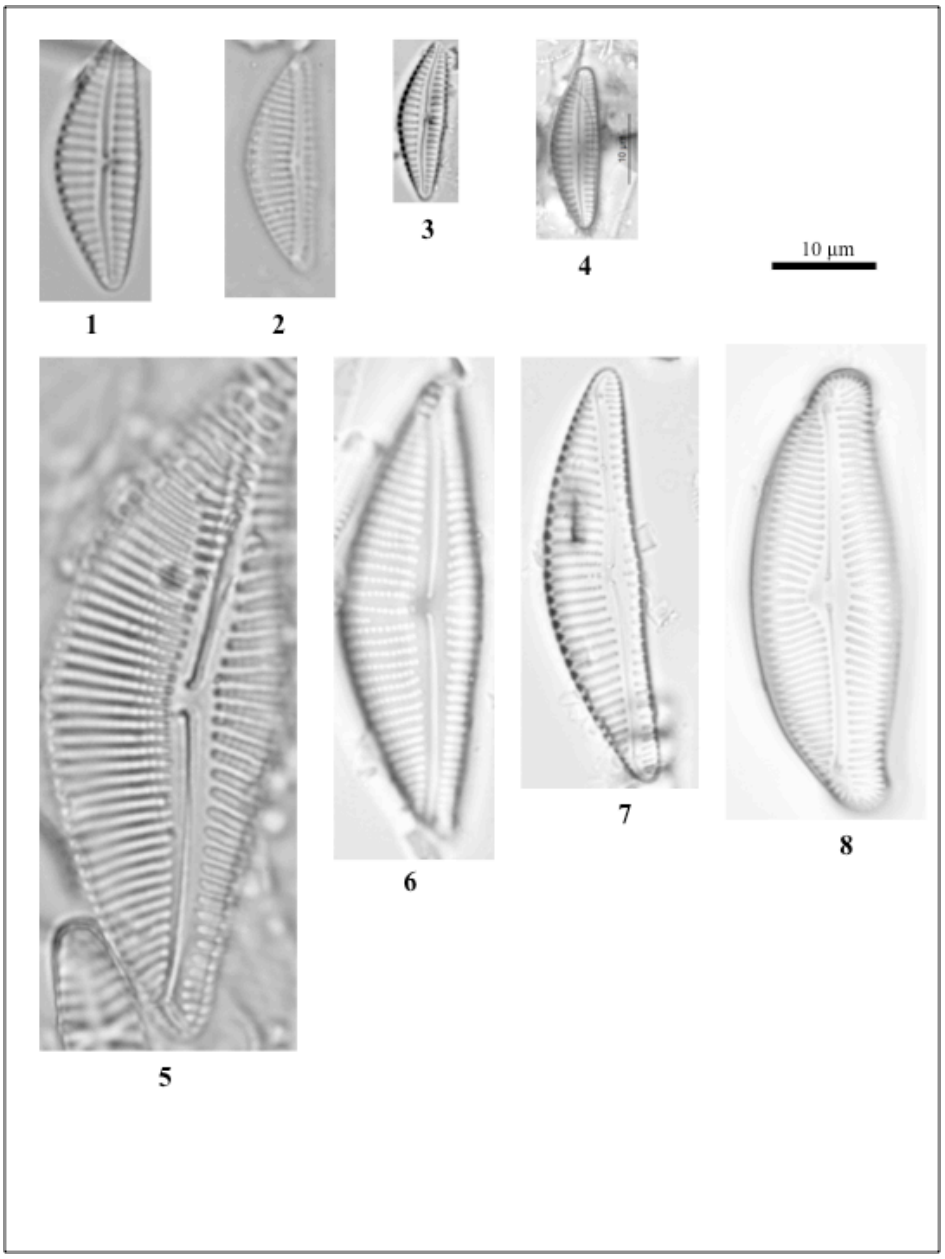


Figure A1.32: Identification plate for *Cymbella* spp.

1 – 2: *Cymbella muelleri (mülleri)* Hustedt 1937

3: *Cymbella minuta* Hilse in Rabenhorst 1862

4: *Cymbopleura hustedtii* (Krasske) E. Novelo, R. Tavera & C. Ibarra 2007 – Synonym: *Cymbella hustedtii* Krasske 1923

5: *Cymbella muelleri* f. *ventricosa* (Tempère & Peragallo) Reimer in Patrick & Reimer 1975

6 – 7: *Cymbella silesiaca* Bleish in Rabenhorst 1861-1882

8: *Cymbella prostrata* (Berkeley) Cleve 1894

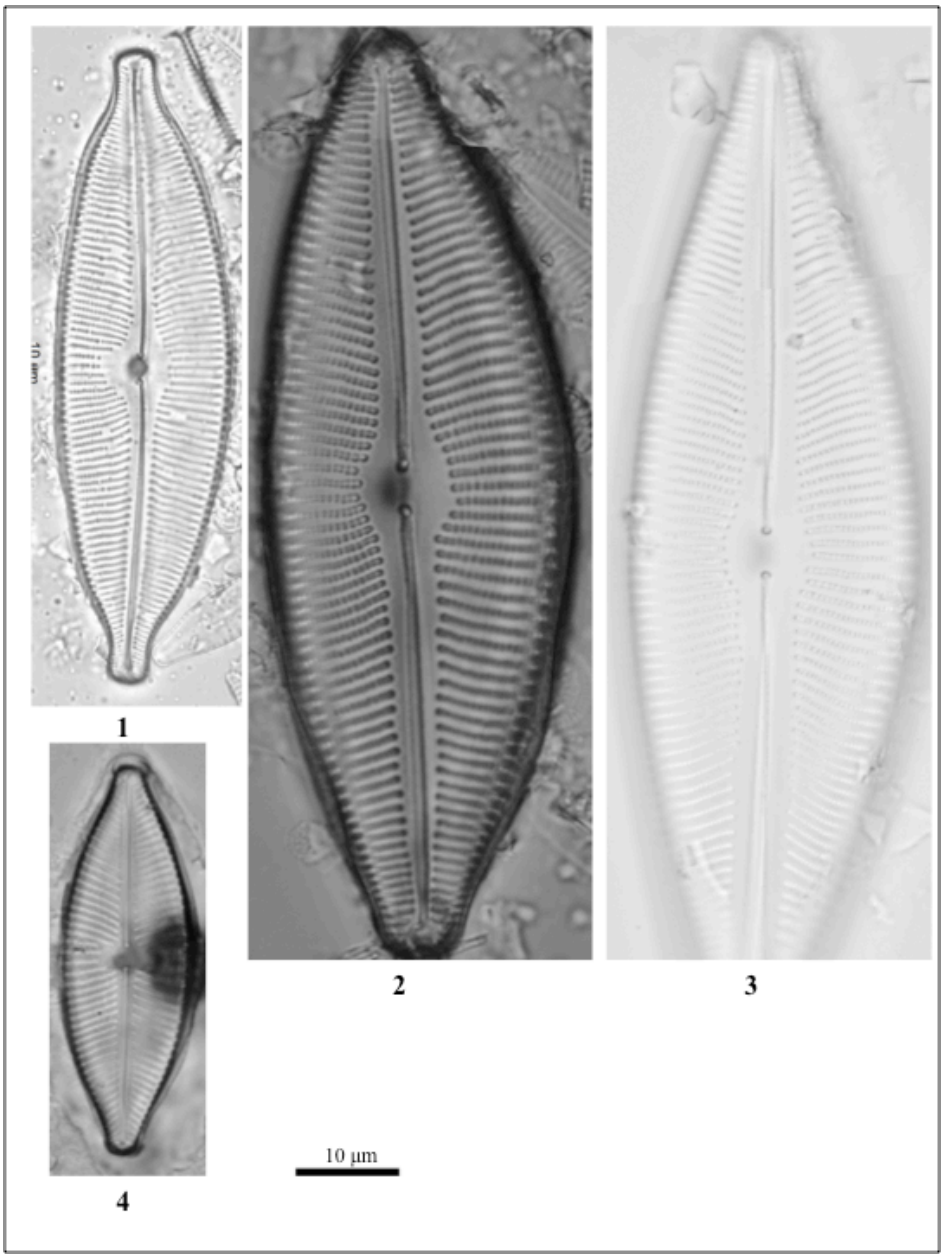


Figure A1.33: Identification plate for *Cymbella* spp.

1 – 2: *Cymbella cuspidata* Kützing 1844 – Synonym: *Cymbopleura cuspidata* (Kützing) K. Krammer 2003

3: *Cymbella ehrenbergii* Kützing 1844

4: *Cymbopleura inaequalis* (Ehrenberg) K. Krammer 2003

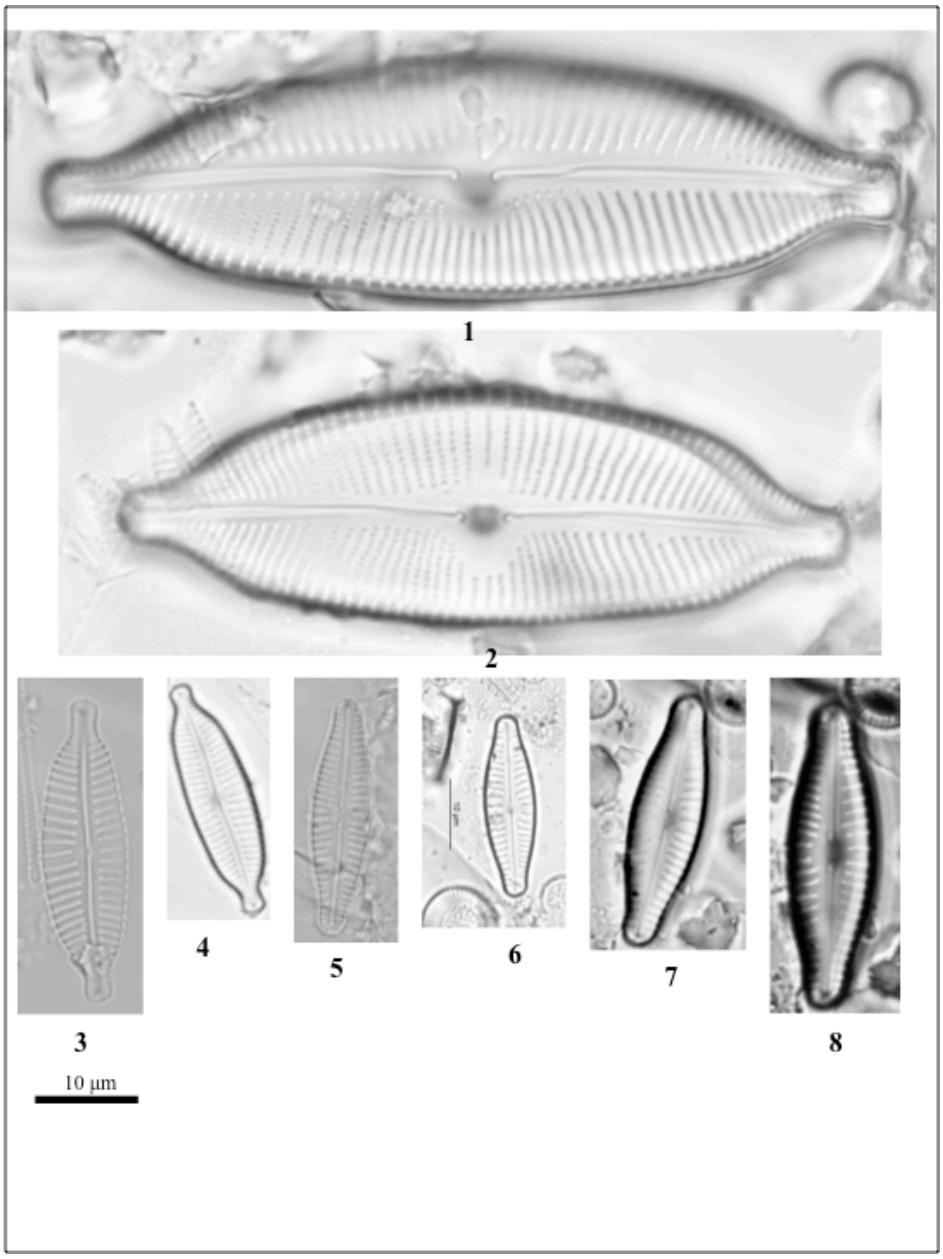


Figure A1.34: Identification plate for *Cymbella* spp.

1 – 2: *Cymbella cuspidata* Kützing 1844 – Synonym: *Cymbopleura cuspidata* (Kützing) K. Krammer 2003

3 – 4: *Cymbella amphicephala* Naegeli ex Kützing 1849

5 – 8: *Cymbellafalsa diluviana* (Krasske) H. Lange-Bertalot & D. Metzeltin in D. Metzeltin, H. Lange-Bertalot & S. Nergui 2009, Synonym: *Cymbella diluviana* (Krasske) Florin 1971

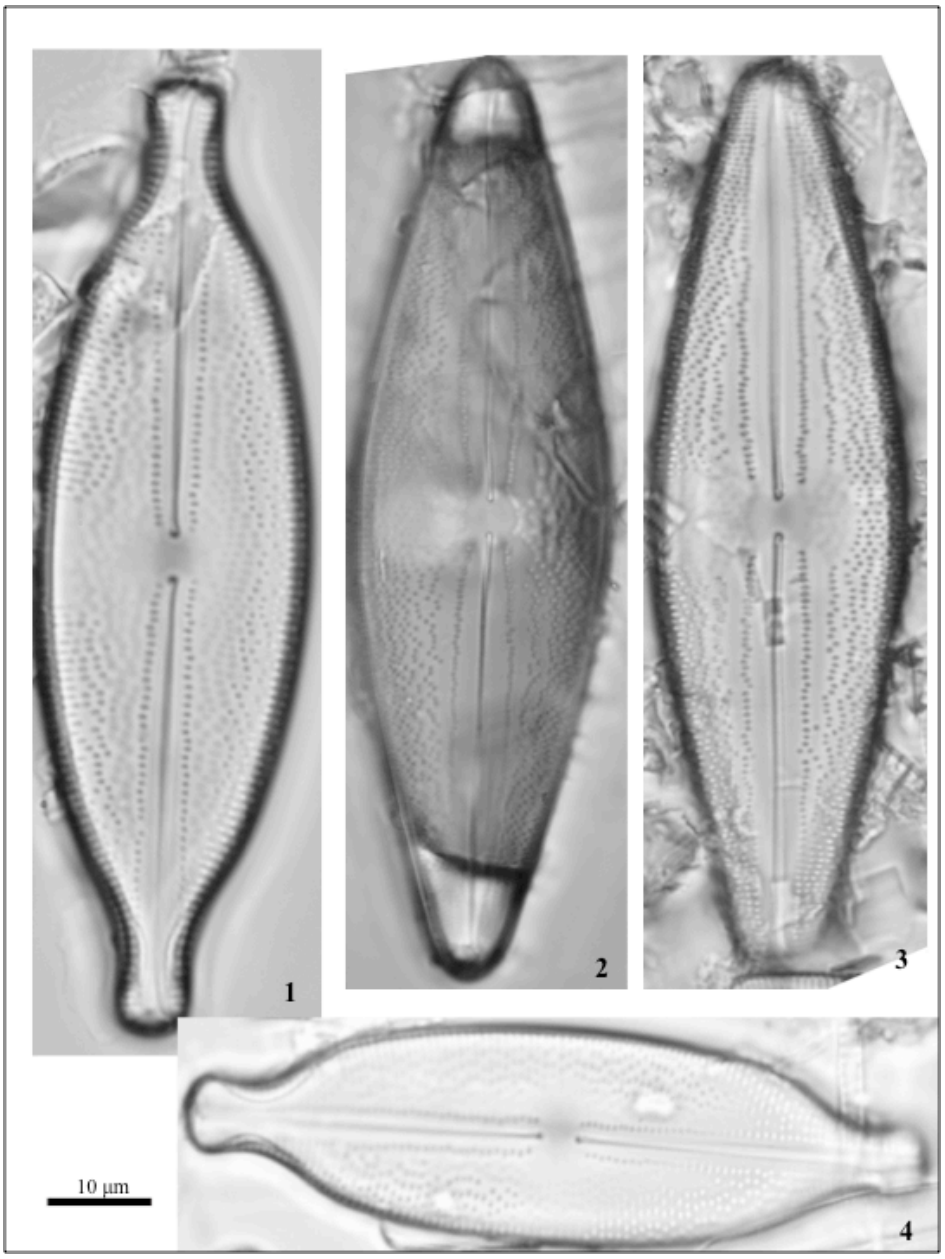


Figure A1.35: Identification plate for *Anomoeoneis* spp.

1, 4: *Anomoeoneis sphaerophora* Pfitzer 1871

2, 3: *Anomoeoneis sphaerophora* f. *costata* (Kützing) A.-M. Schmid 1977

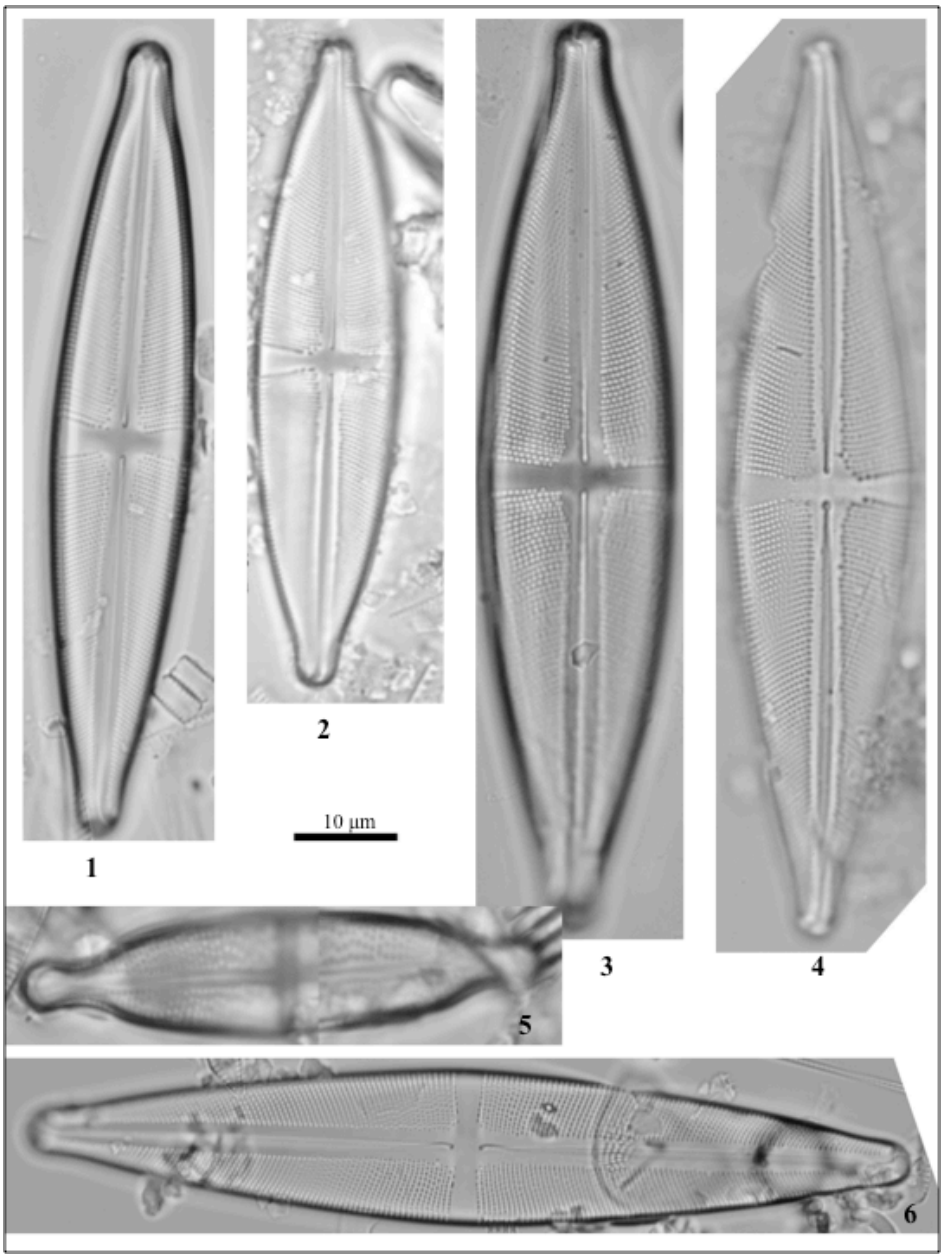


Figure A1.36: Identification plate for *Stauroneis* spp.

1 – 2: *Stauroneis italica* H. Lange-Bertalot, P. Cavacini, N. Tagliaventi & S. Alfinito 2003

5: *Stauroneis anceps* Ehrenberg 1843

3, 4, & 6: *Stauroneis phoenicenteron* (Nitzsch) Ehrenberg 1843

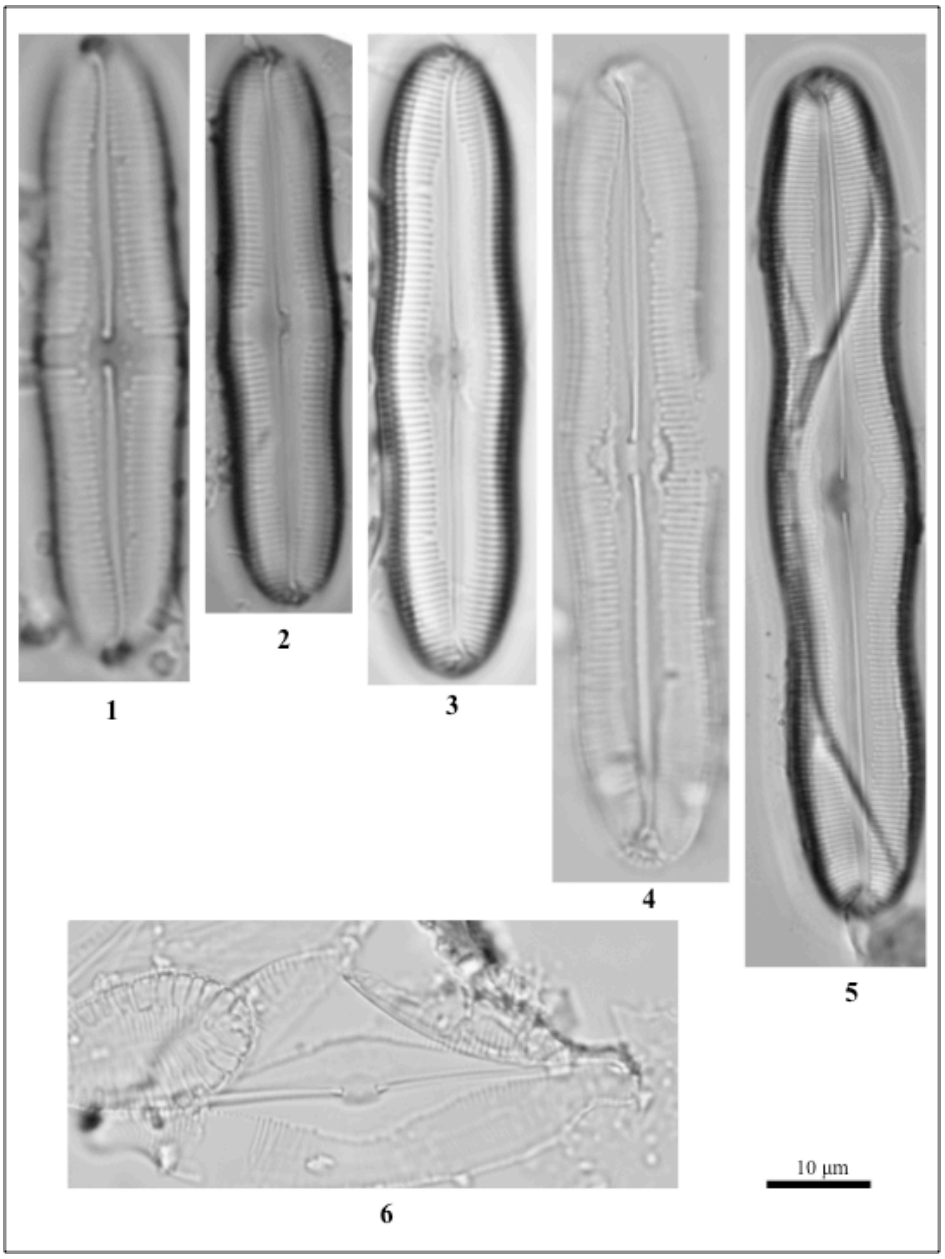


Figure A1.37: Identification plate for *Caloneis* spp.

1, 2: *Caloneis ventricosa* (Ehrenberg) Meister 1912 – Synonym: *Navicula ventricosa* Ehrenberg 1830

3 – 5: *Caloneis silicula* (Ehrenberg) Cleve 1894 – Synonym: *Navicula silicula* Ehrenberg 1843

6: *Caloneis amphisbaena* var. *genuina* Cleve-Euler 1955

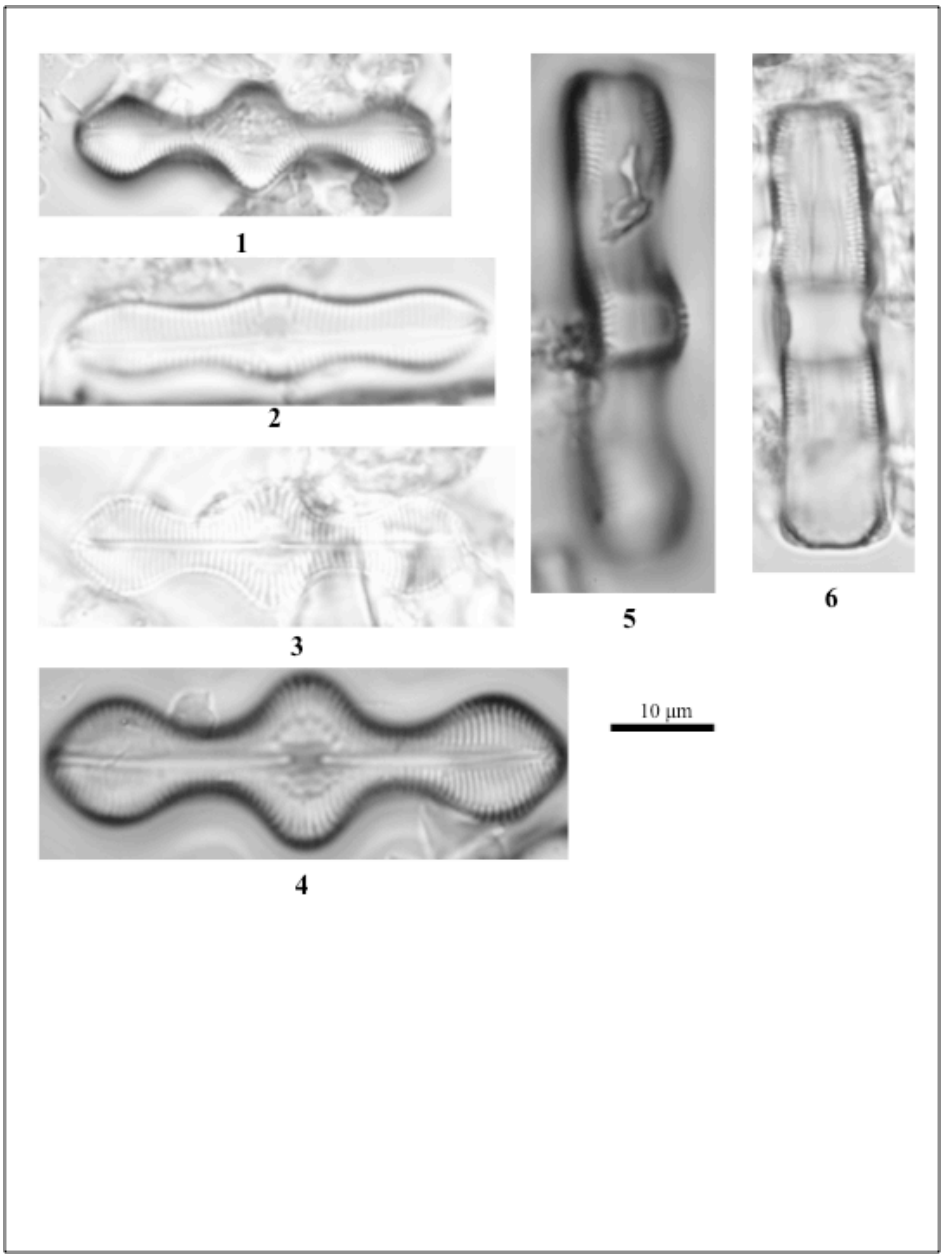


Figure A1.38: Identification plate for *Caloneis trochus* var. *trinodis*

1 – 6: *Caloneis trochus* var. *trinodis* (Cleve) Cleve-Euler 1955

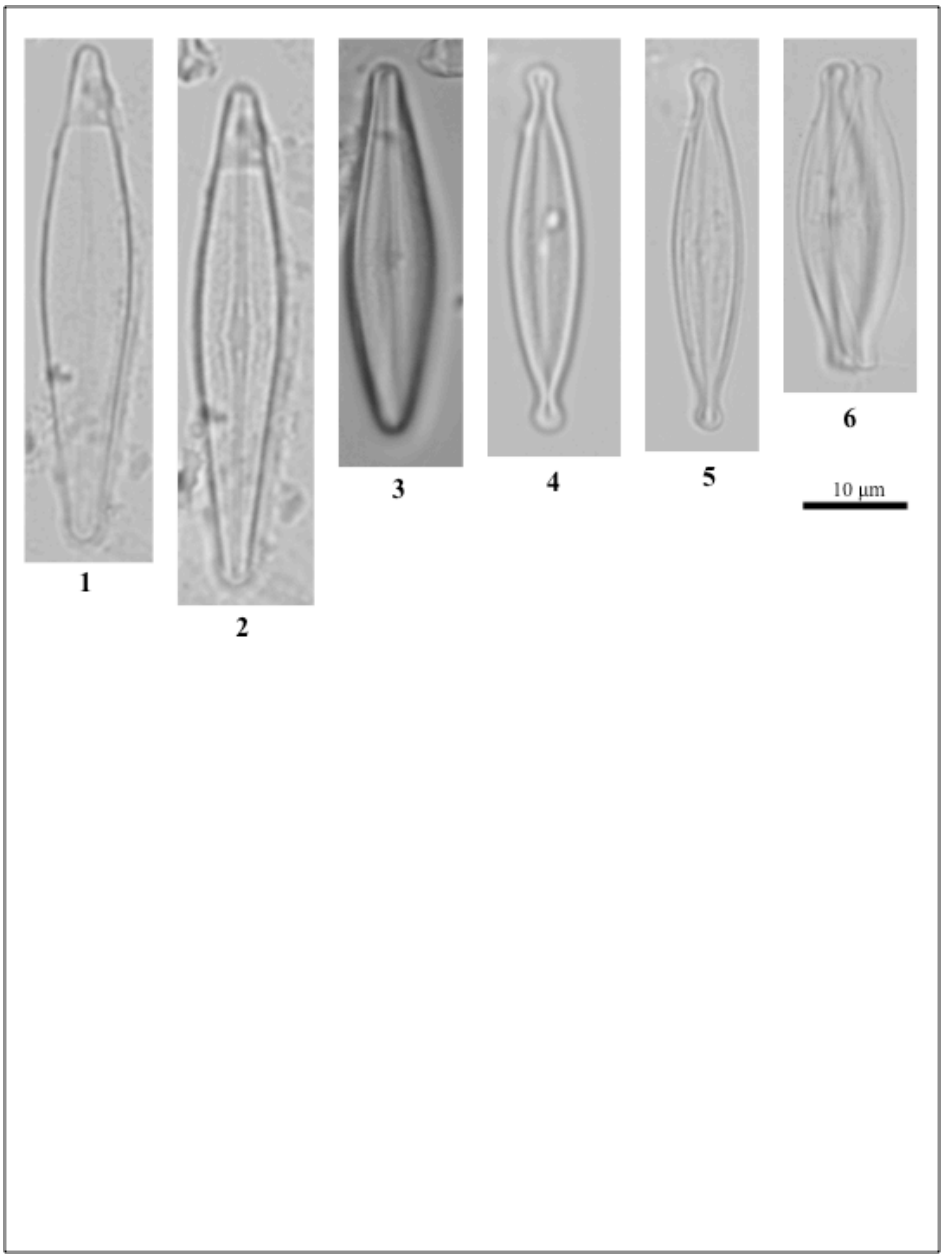


Figure A1.39: Identification plate for *Brachysira* spp.

1 – 3: *Brachysira styriaca* (Grunow in Van Heurck) R. Ross in Hartley 1986 – Basionym: *Navicula styriaca* Grunow in Van Heurck 1880

4 – 5: *Brachysira neoexilis* H. Lange-Bertalot in H. Lange-Bertalot & G. Moser 1994

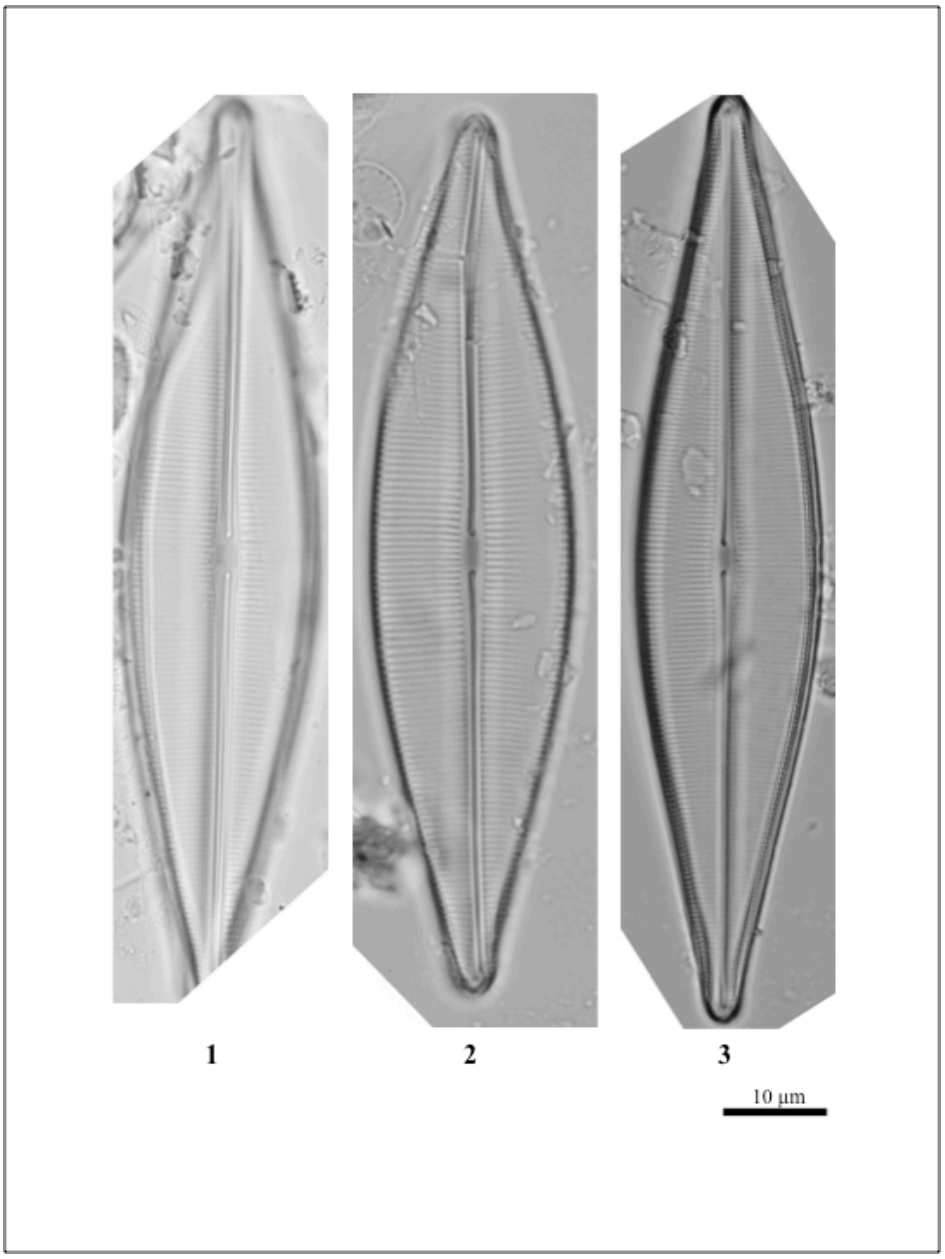


Figure A1.40: Identification plate for *Craticula cuspidata*

1 – 3: *Craticula cuspidata* (Kützing) Mann in Round, Crawford & Mann 1990

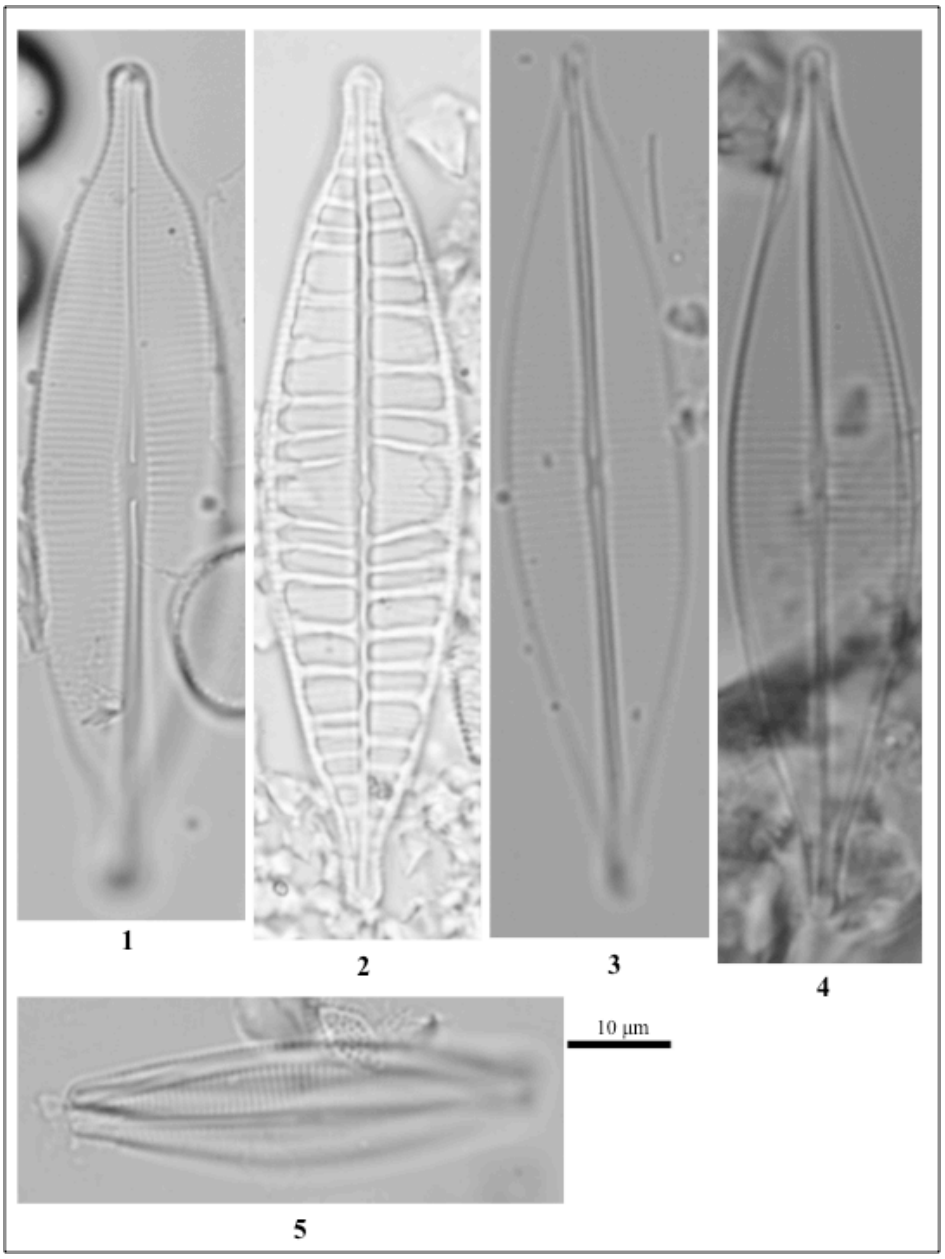


Figure A1.41: Identification plate for *Craticula* spp.

1: *Craticula ambigua* (Ehrenberg) Mann in Round, Crawford & Mann 1990

2: *Craticula ambigua* - Internal craticula structure

3 – 5: *Craticula halophiloides* (Hustedt) H. Lange-Bertalot 2001

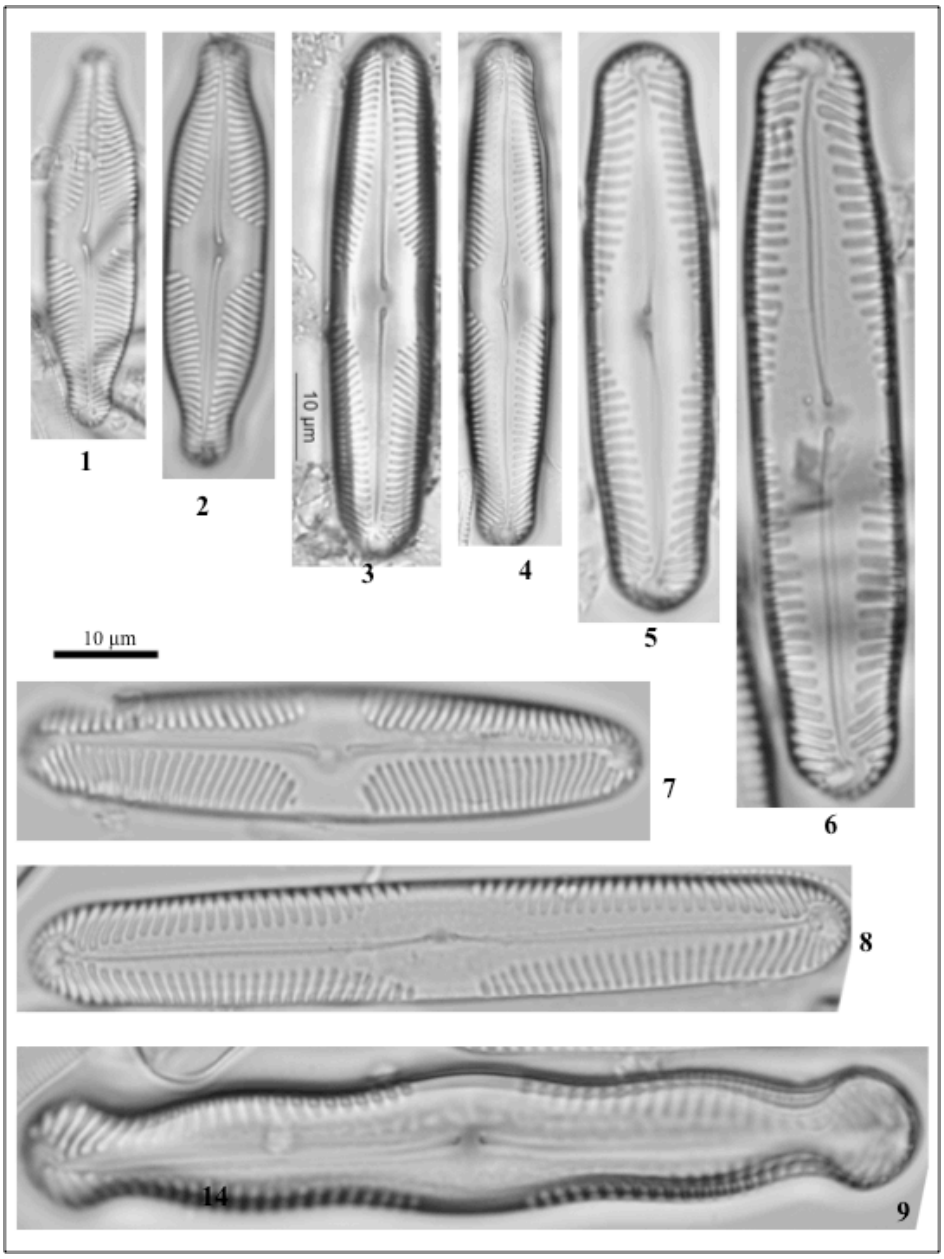


Figure A1.42: Identification plate for *Pinnularia* spp.

1 – 2: *Pinnularia interrupta* W. Smith 1853 – Synonym: *Pinnularia gibba* var. *interrupta* (W. Smith) Woodhead & Tweed 1960

3: *Pinnularia divergens* var. *sublinearis* Cleve 1895

4, 8: *Pinnularia divergens* W. Smith 1853

5 – 6: *Pinnularia braunii* (Grunow in Van Heurck) Cleve 1895

7: *Pinnularia microstauron* (Ehrenberg) Cleve 1891

9: *Pinnularia nodosa* (Ehrenberg) W. Smith 1856 – Basionym: *Navicula nodosa* Ehrenberg 1838

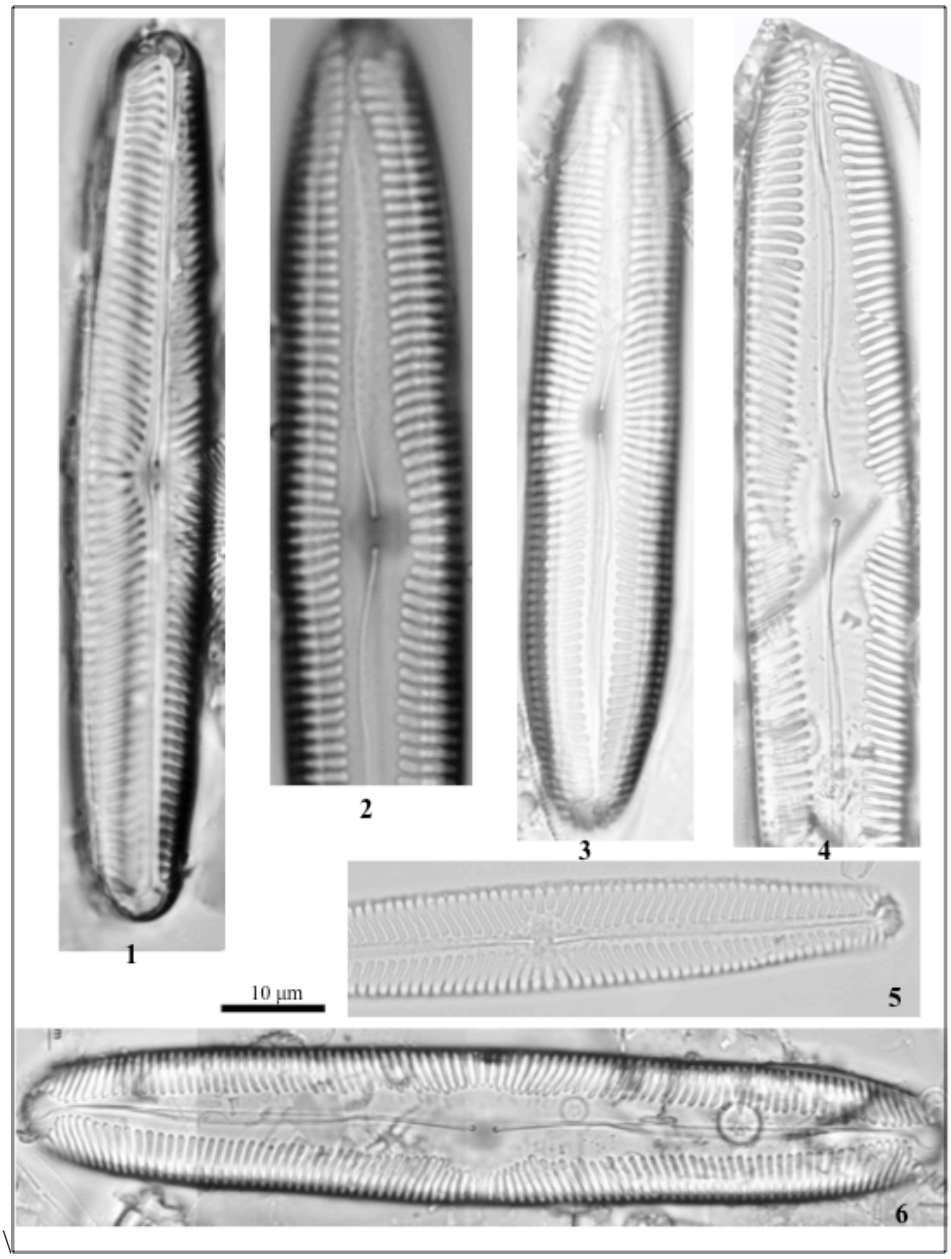


Figure A1.43: Identification plate for *Pinnularia* spp.

1: *Pinnularia subgibba* Krammer 1992

2- 4: *Pinnularia abaujensis* (Pantocsek) Ross 1947 – Basionym: *Navicula gibba* var. *abaujensis* Pantocsek 1889

5: *Pinnularia viridis* (Nitzsch) Ehrenberg 1843

6: *Pinnularia problematica* (Østrup) F.W. Mills 1934 – Basionym: *Navicula problematica* Østrup 1908

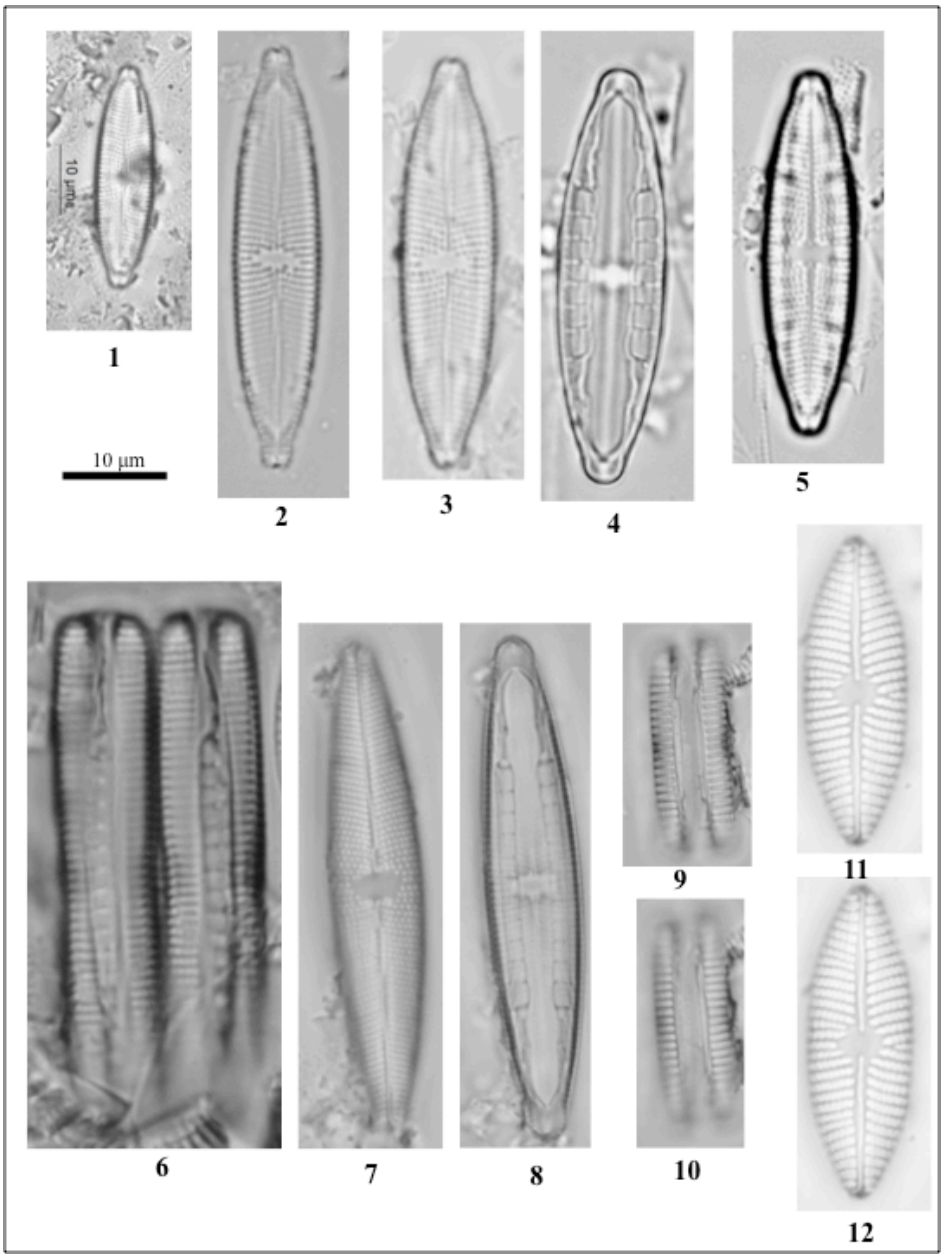


Figure A1.44: Identification plate for *Mastogloia* spp.

1 – 6: *Mastogloia smithii* Thwaites in lit. ex W. Smith 1856

7 – 8: *Mastogloia smithii* var. *lacustris* Grunow 1878

9 – 12: *Mastogloia grevillei* W. Smith in Gregory 1856

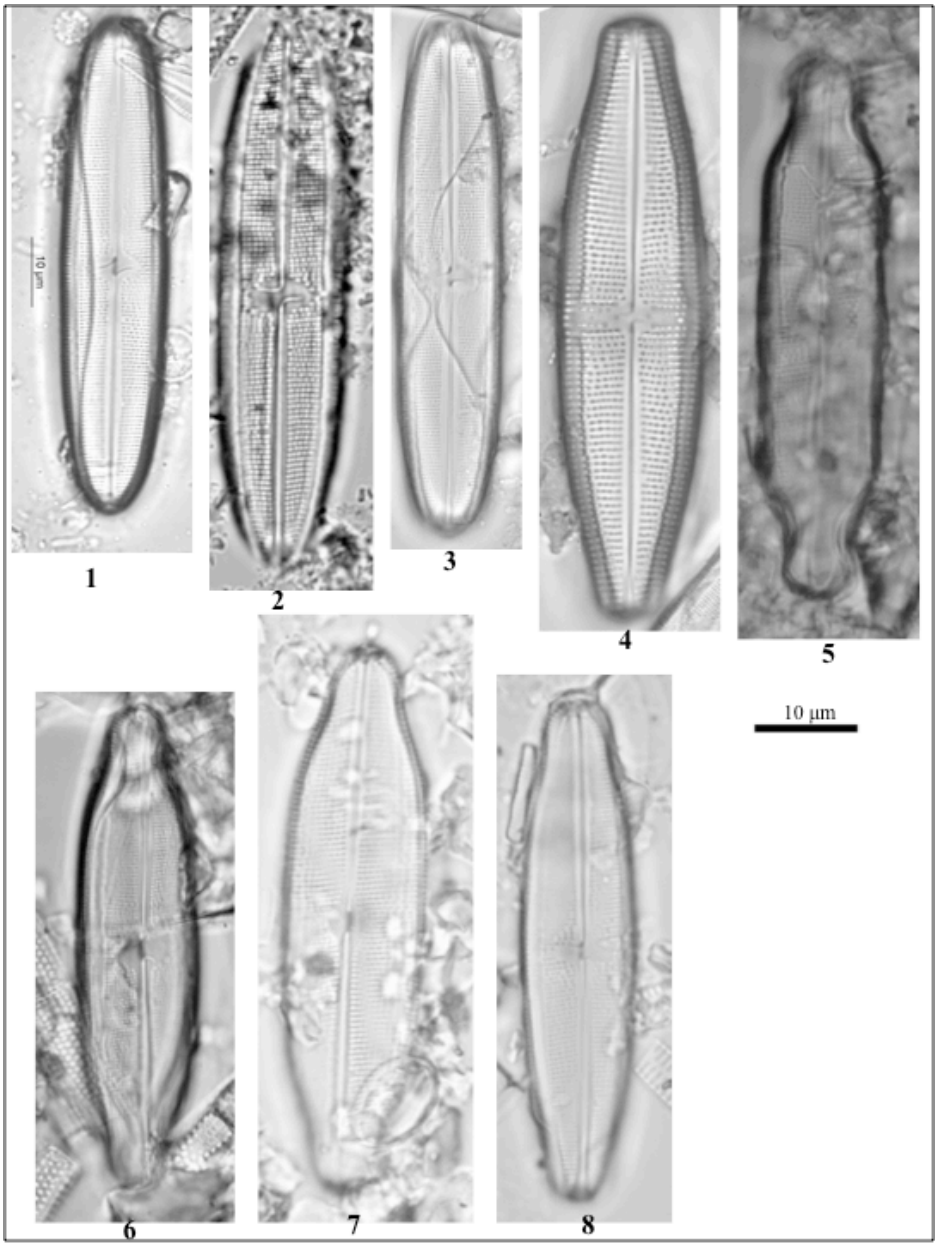


Figure A1.45: Identification plate for *Neidium* spp.

1 – 3: *Neidium hankensis* Skvortzow 1929

4: *Neidium distinctepunctatum* Hustedt 1922

5: *Neidium affine* (Ehrenberg) Pfitzer 1871

6 – 8: *Neidium ampliatum* (Ehrenberg) Krammer in Krammer & Lange-Bertalot 1985

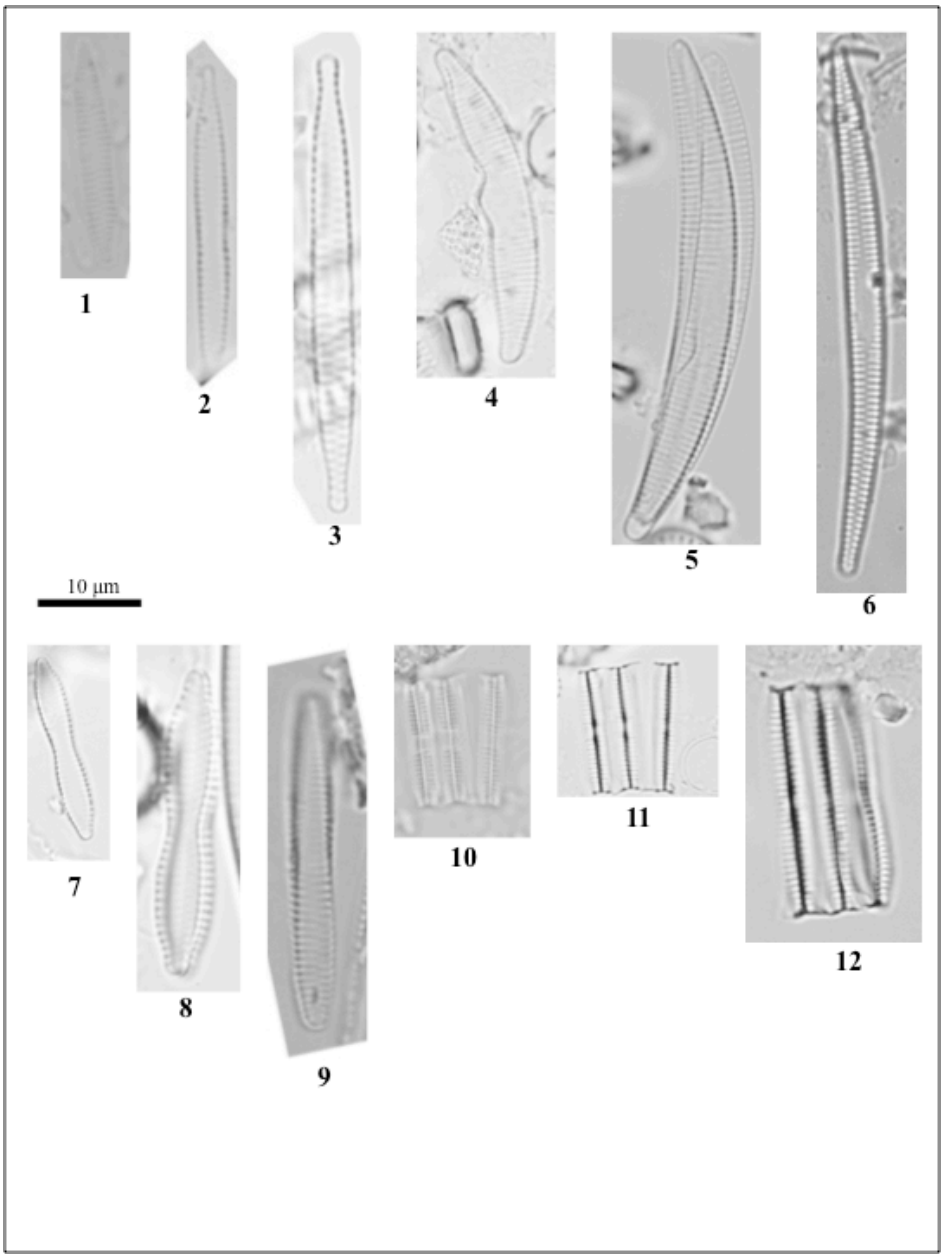


Figure A1.46: Identification plate for *Fragilaria* spp.

1: *Fragilaria vaucheriae* (Kützing) Peterson, 1938

2, 11: *Fragilaria capucina* Desmazieres 1825

3: *Fragilaria capucina* var. *gracilis* (Østrup) Hustedt 1950

4: *Tabularia fasciculata* (Agardh) Williams & Round 1986 – Synonym: *Fragilaria fasiculata* (C. A. Agardh) Lange-Bertalot 1980

5 – 6: *Fragilaria cyclopum* (Brutschy) Lange-Bertalot 1980

7 - 12: *Fragilaria capucina* var. *mesolepta* (Rabenhorst) Rabenhorst 1864

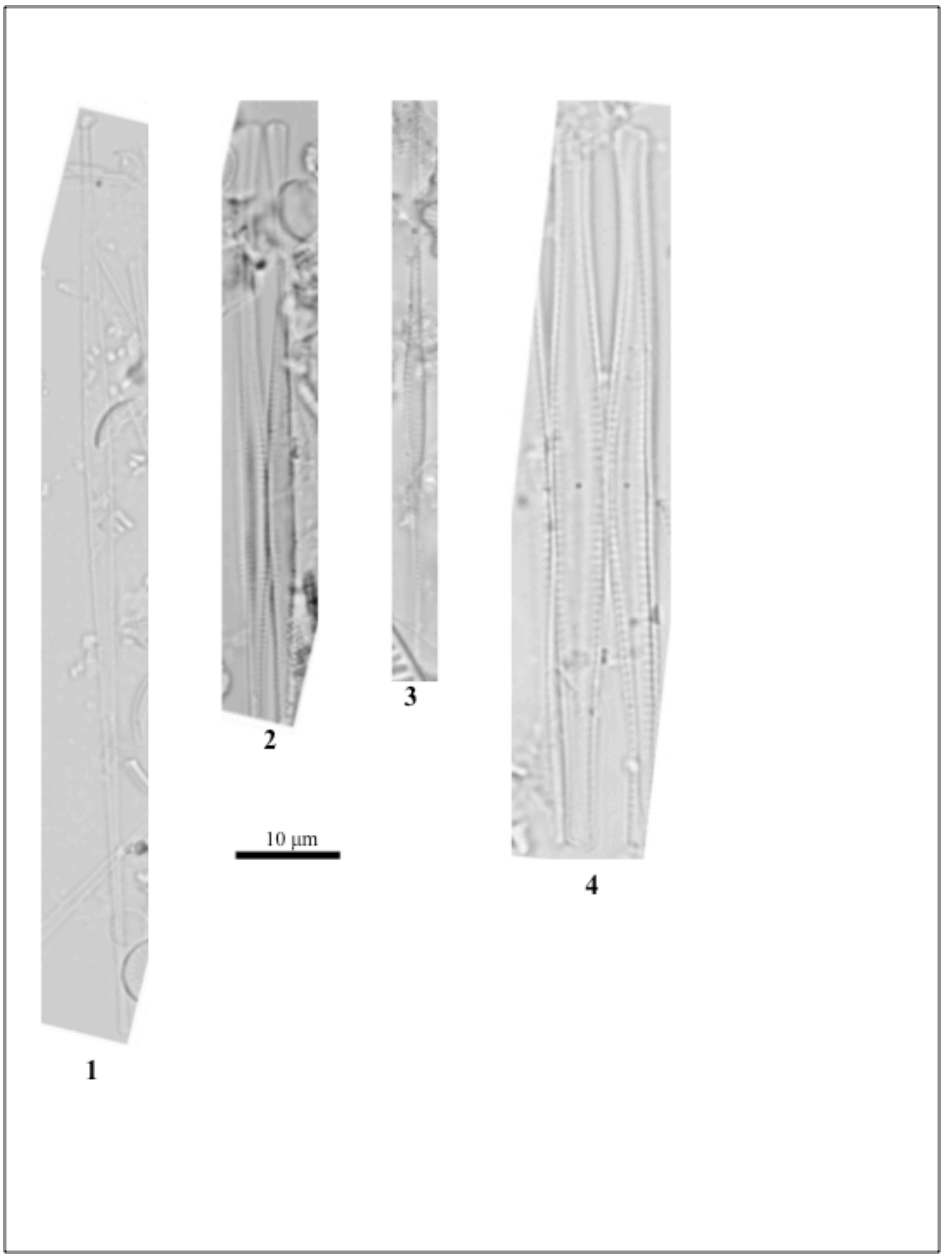


Figure A1.47: Identification plate for *Fragilaria* spp.

1: *Fragilaria delicatissima* (W. Smith) Lange-Bertalot 1980

2 – 4: *Fragilaria crotonensis* Kitton 1869

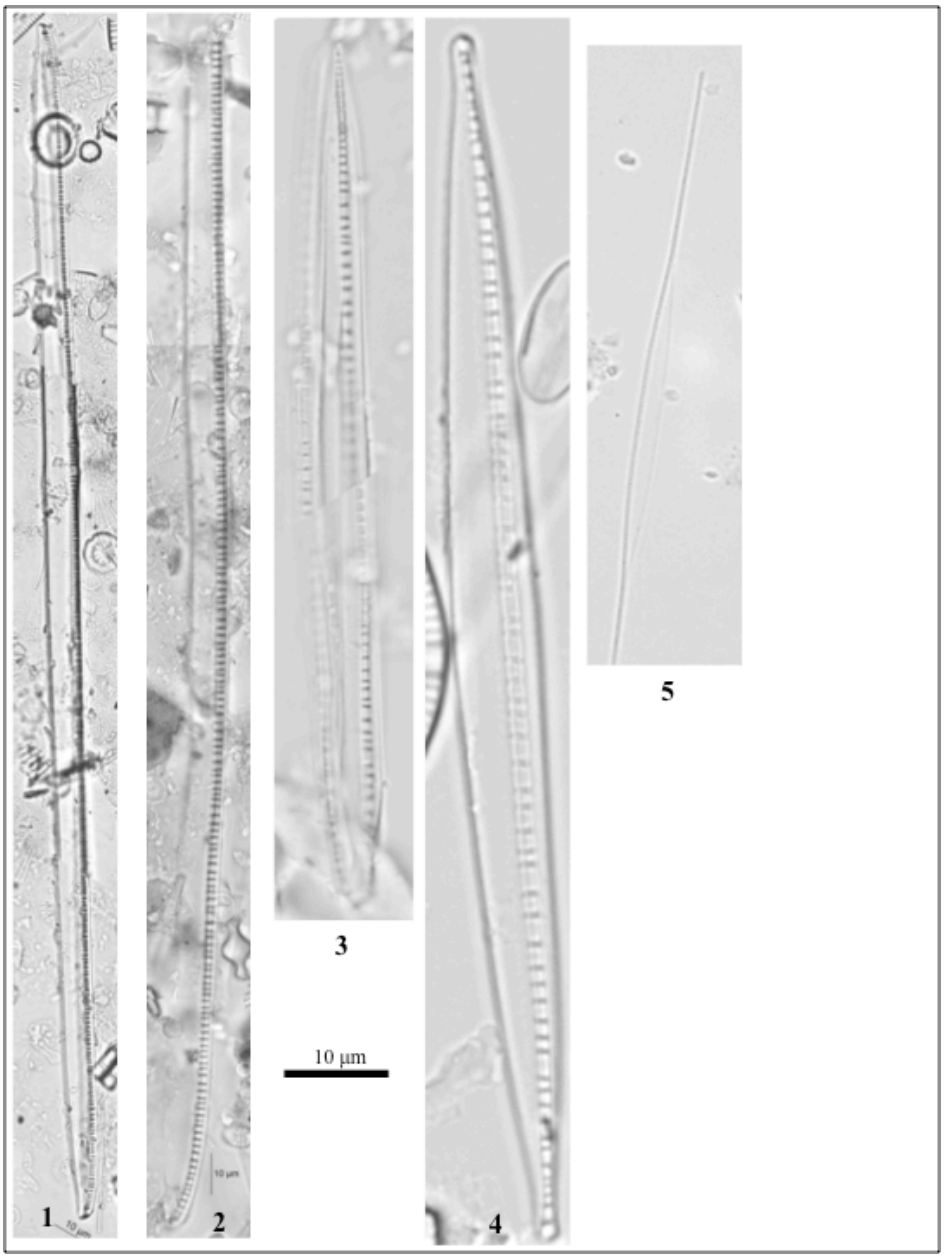


Figure A1.48: Identification plate for *Nitzschia* spp.

1: *Nitzschia linearis* (Agardh) W. Smith 1853

2: *Nitzschia sigmoidea* (Nitzsch) W. Smith 1853

3 – 4: *Nitzschia dissipata* (Kützing) Grunow 1862 – Basionym: *Synedra dissipata* Kützing 1844

5: *Nitzschia acicularis* (Kützing) W. Smith 1853

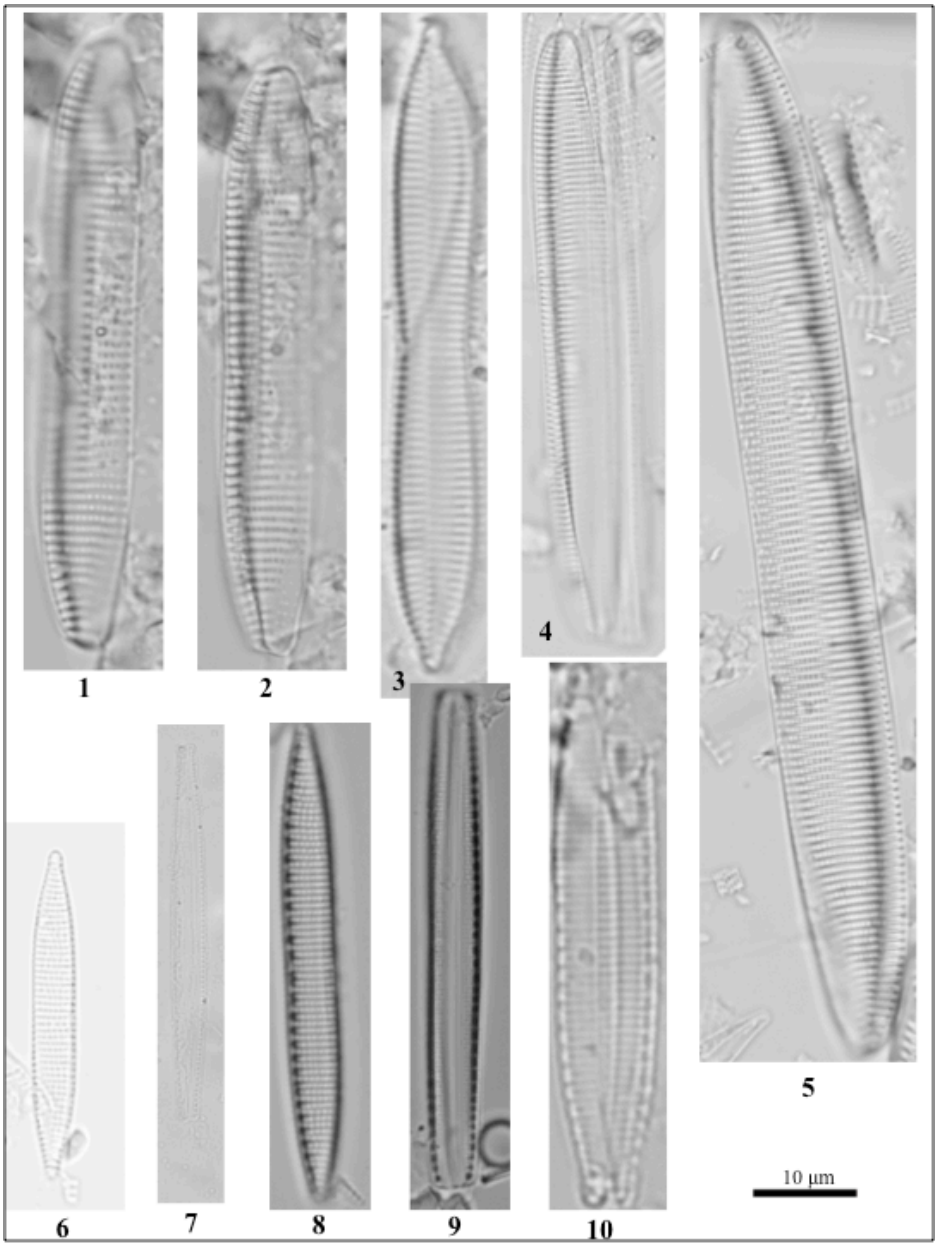


Figure A1.49: Identification plate for *Nitzschia* spp.

1 – 2, 3: *Nitzschia apiculata* (Gregory) Grunow 1878

4 - 5: *Tryblionella angustata* W. Smith 1853

6: *Nitzschia frustulum* (Kützing) Grunow in Cleve & Grunow 1880 – Basionym: *Synedra frustulium* Kützing 1844

7 – 8: *Nitzschia gracilis* Hantzsch 1860

9: *Nitzschia sublinearis* Hustedt in Schmidt et al. 1921

10: *Nitzschia palea* Kützing W. Smith 1856 – Basionym: *Synedra palea* Kützing 1844

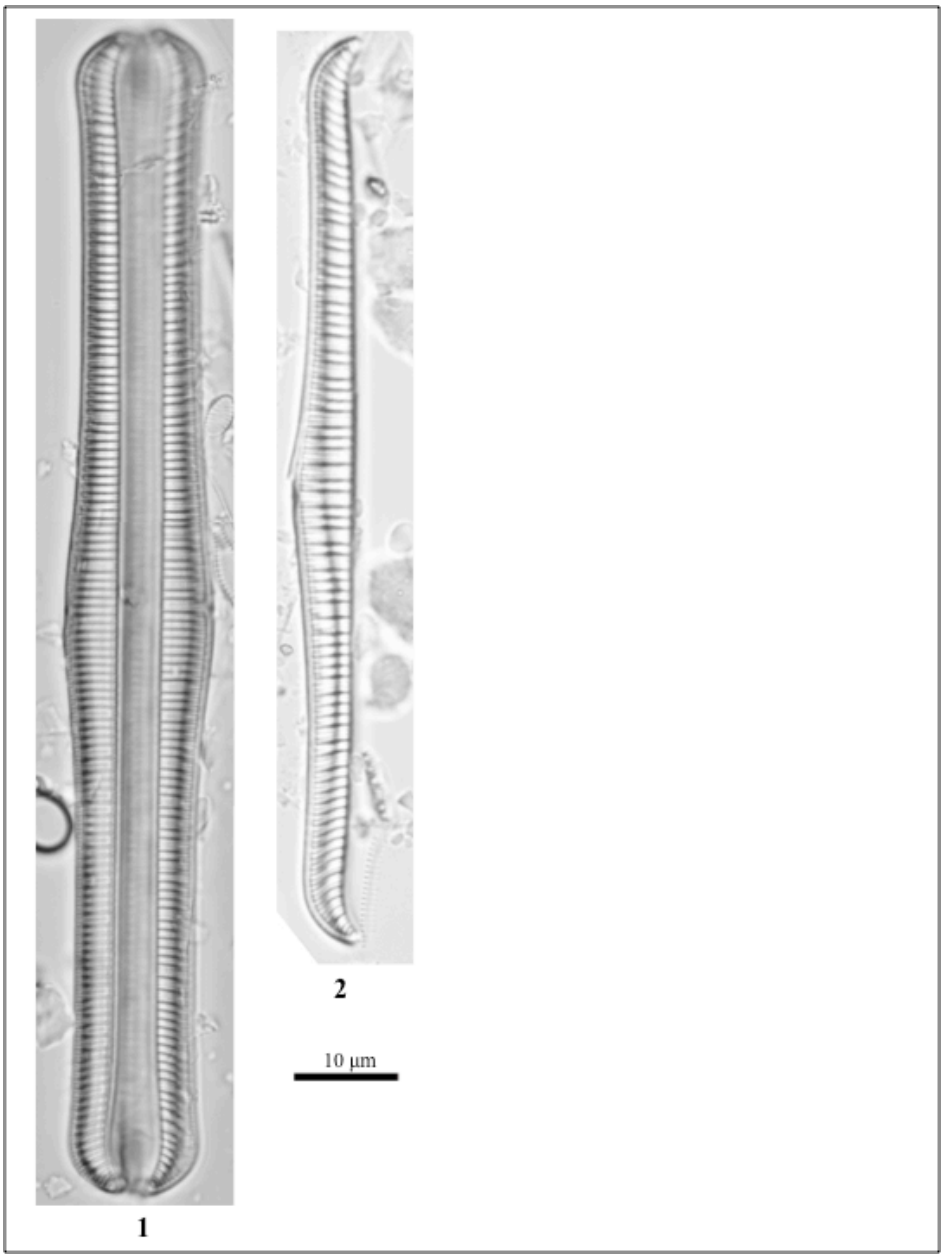


Figure A1.50: Identification plate for *Rhopalodia gibba*

1 – 2: *Rhopalodia gibba* (Ehrenberg) Otto Müller 1895

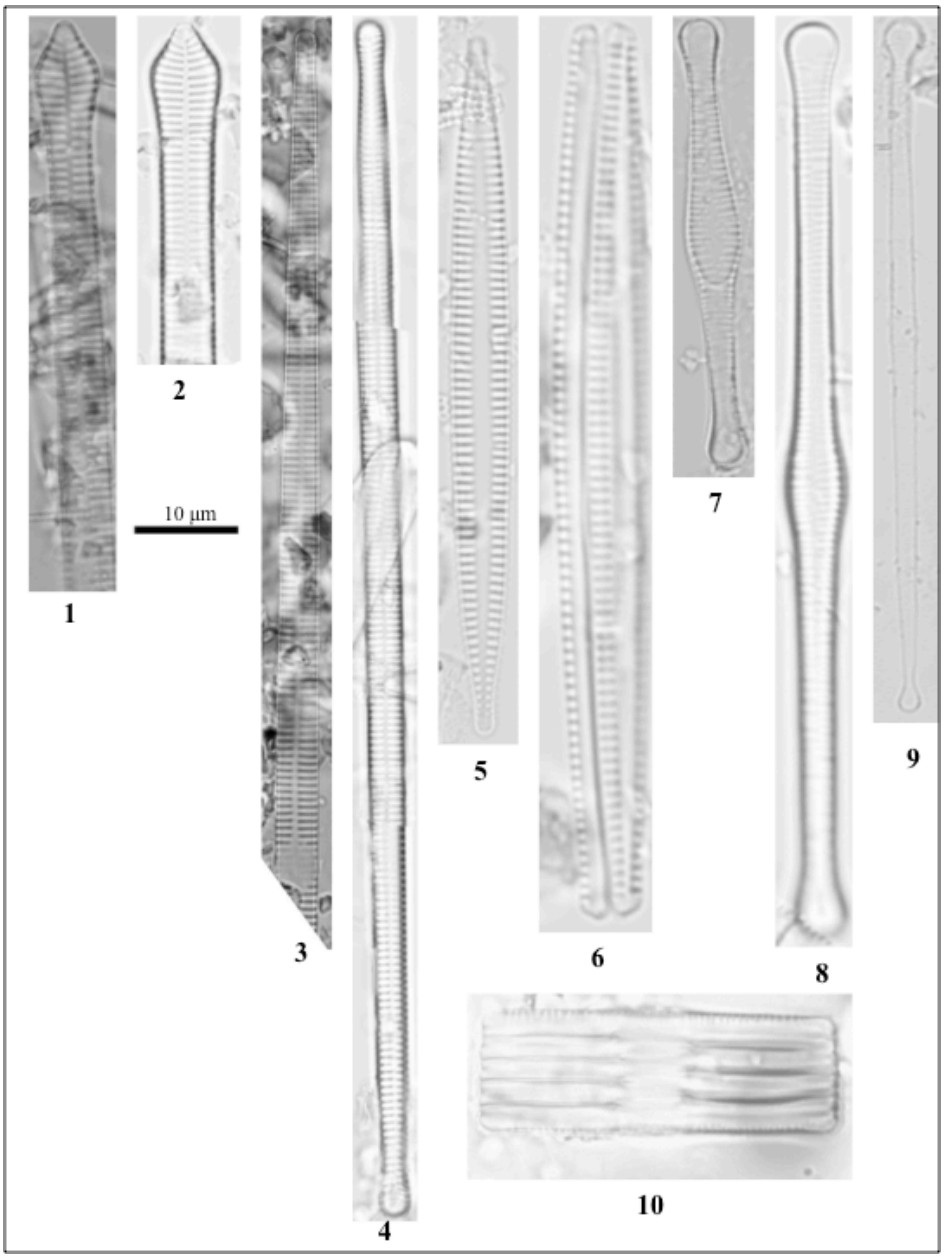


Figure A1.51: Identification plate for *Synedra* spp. and *Tabellaria flocculosa*

1 – 2: *Synedra ulna* var. *claviceps* Hustedt 1937

3 – 4: *Synedra ulna* (Nitzsch) Ehrenberg 1832

5: *Synedra incisa* Boyer 1920

6: *Synedra tabulata* Agardh Kützing 1844

7 – 10: *Tabellaria flocculosa* (Roth) Kützing 1844

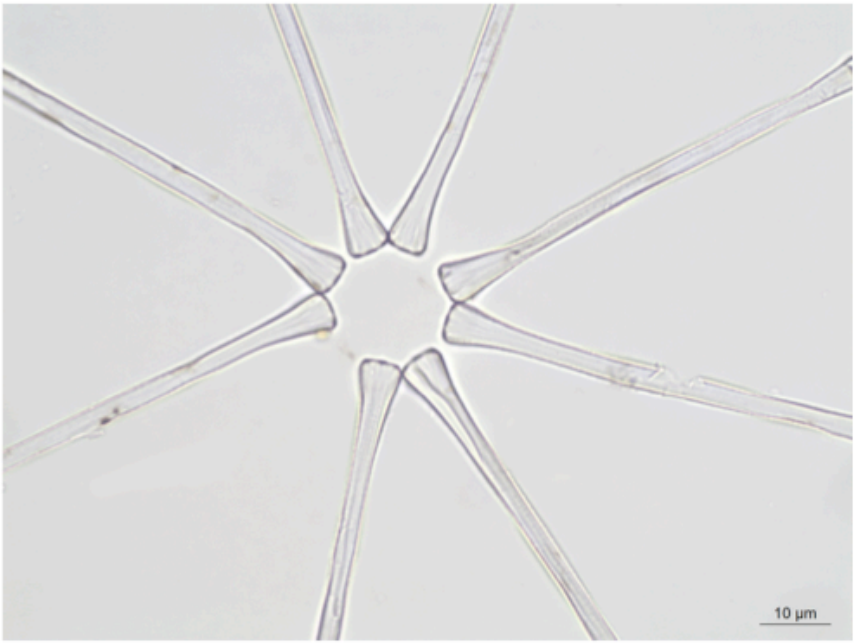


Figure A1.52: Identification plate for *Asterionella formosa*

1: *Asterionella formosa* Hassall 1850

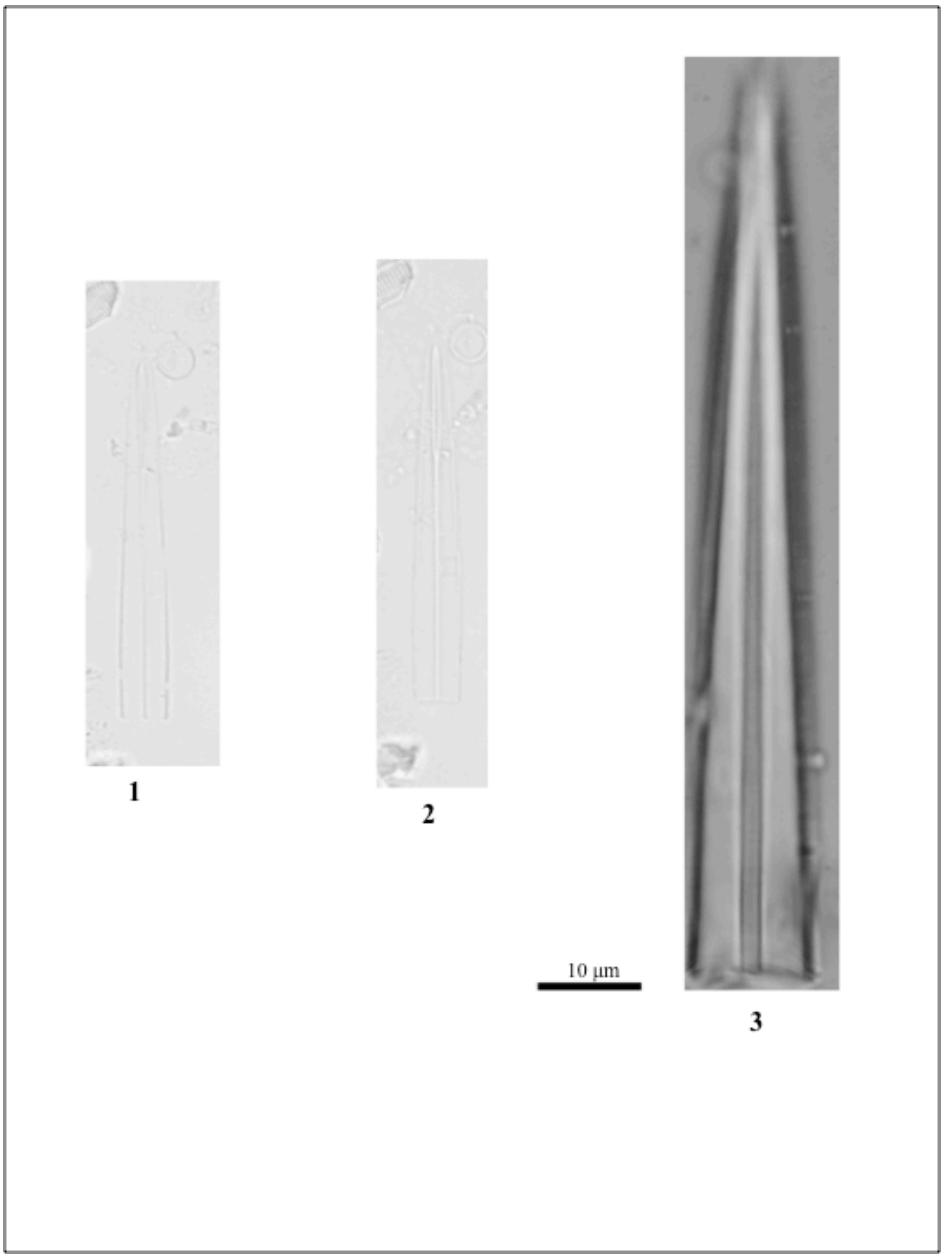


Figure A1.53: Identification plate for *Amphipleura pellucida*

1 – 3: *Amphipleura pellucida* (Kützing) Kützing 1844

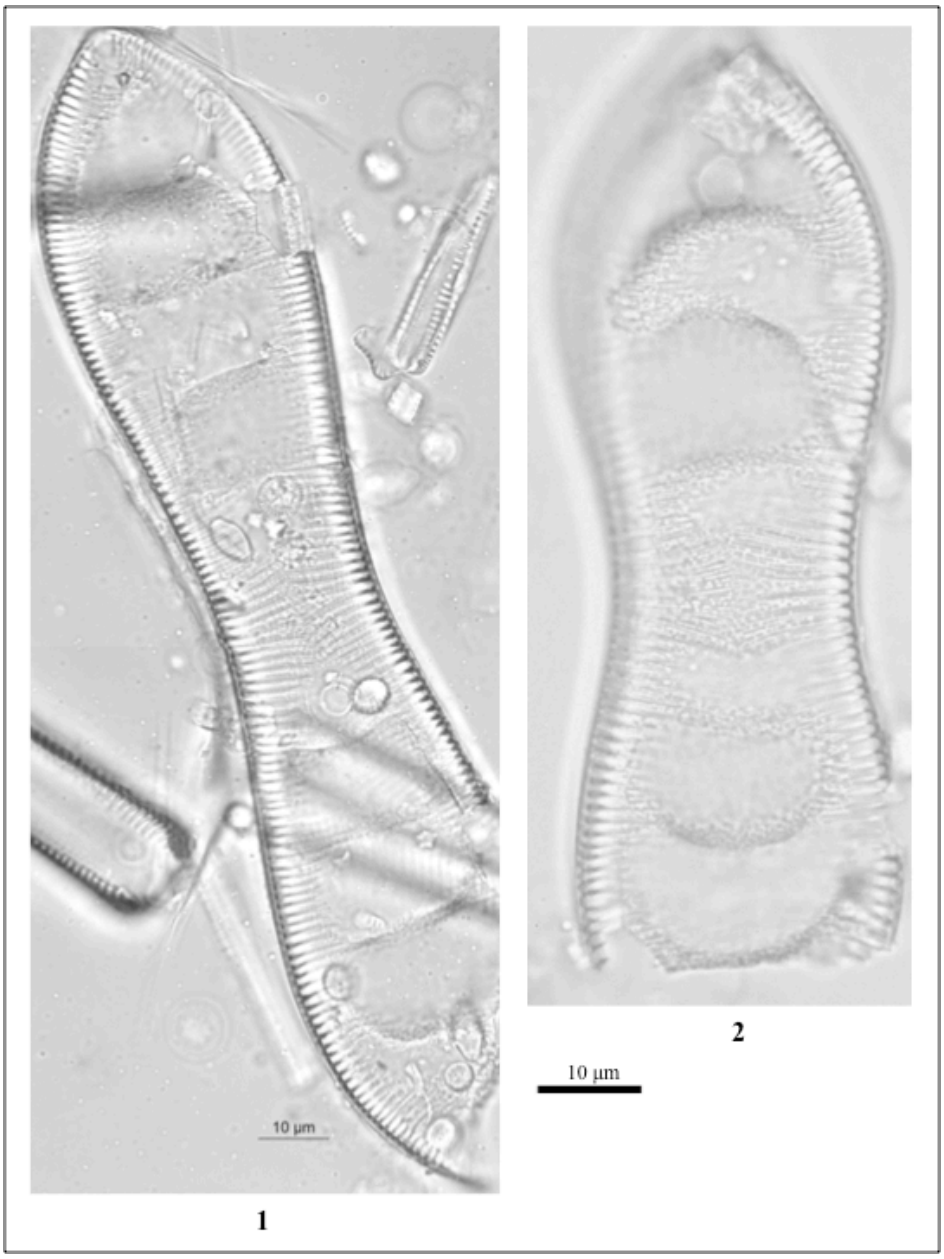


Figure A1.54: Identification plate for *Cymatopleura solea*

1 – 2: *Cymatopleura solea* (Brébisson in Brébisson & Godey) W. Smith 1851

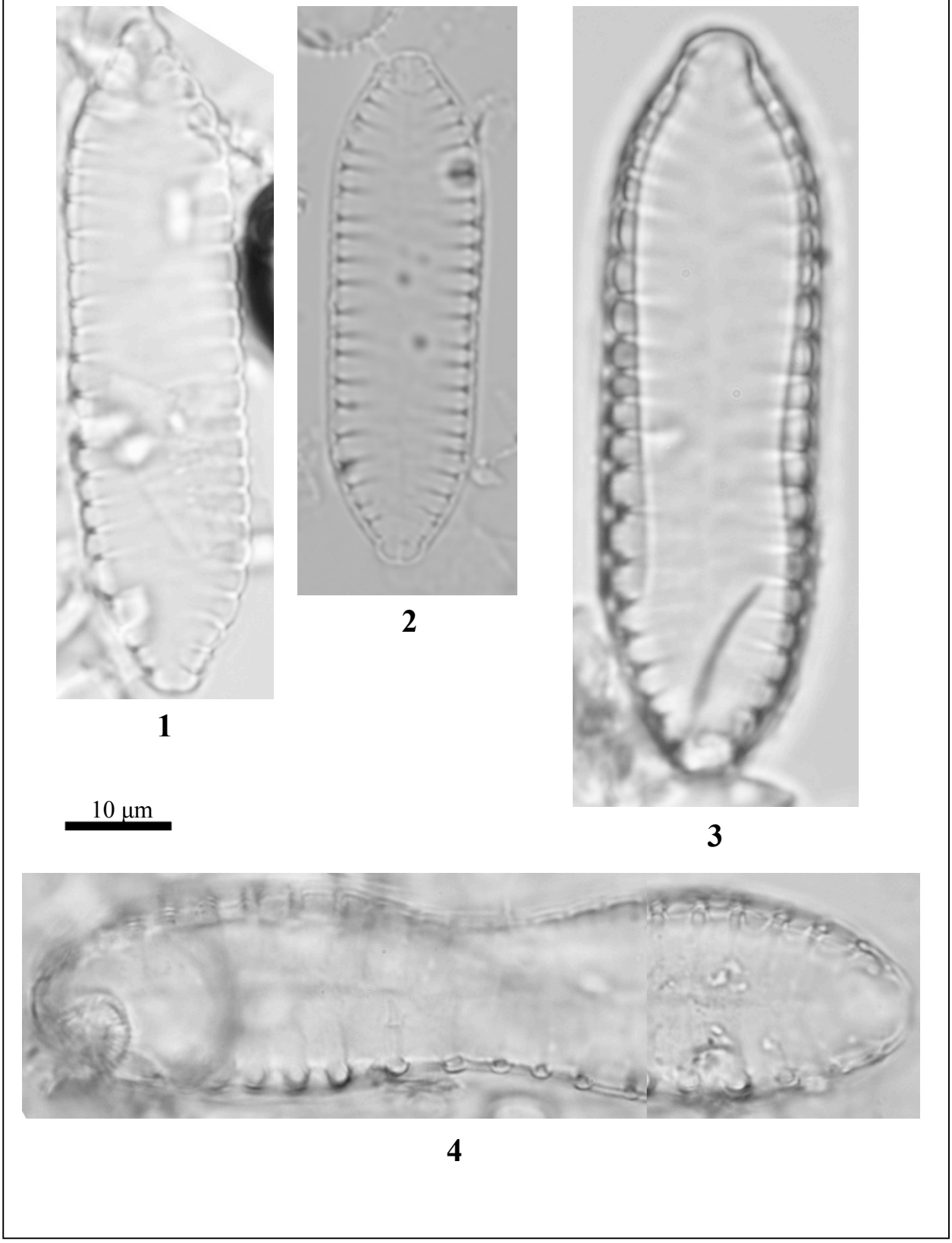


Figure A1.55: Identification plate for *Surirella* spp.

1 – 2: *Surirella angusta* Kützing 1844

3 – 4: *Surirella linearis* W. Smith 1853

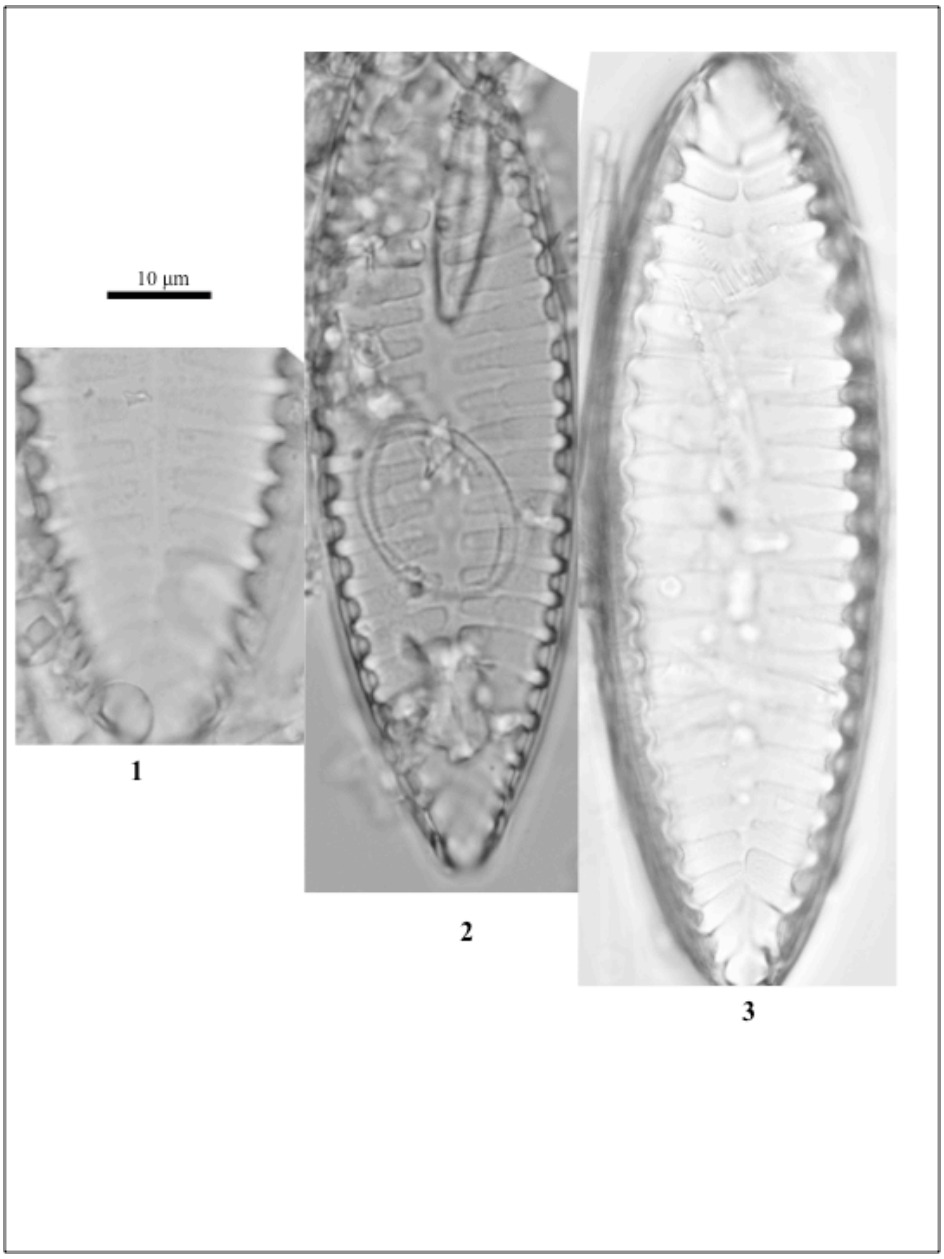


Figure A1.56: Identification plate for *Surirella amphioxys*

1 - 3: *Surirella amphioxys* W. Smith 1856

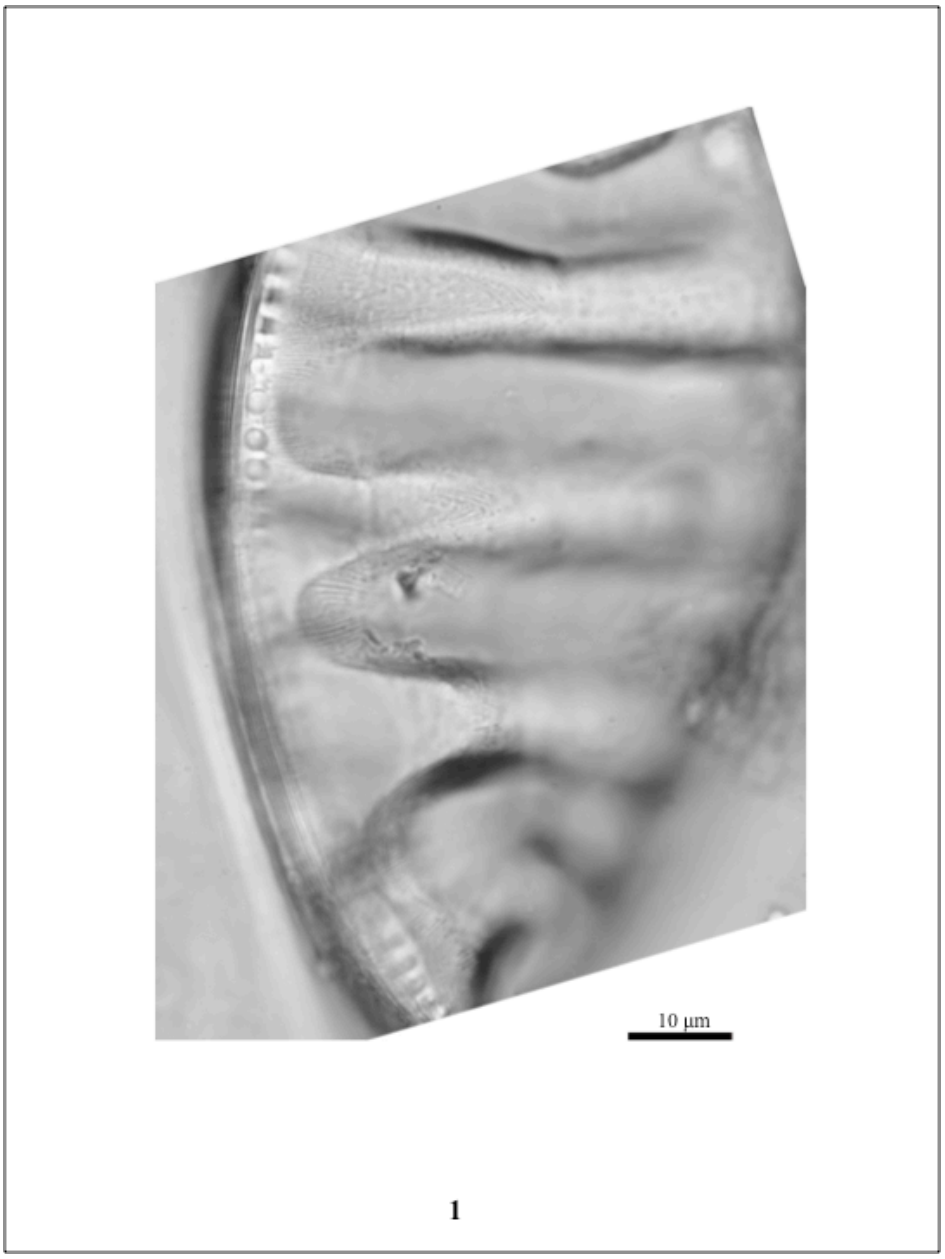


Figure A1.57: Identification plate for *Cymatopleura elliptica*

1: *Cymatopleura elliptica* (Brébisson ex Kutzing) W. Smith 1851

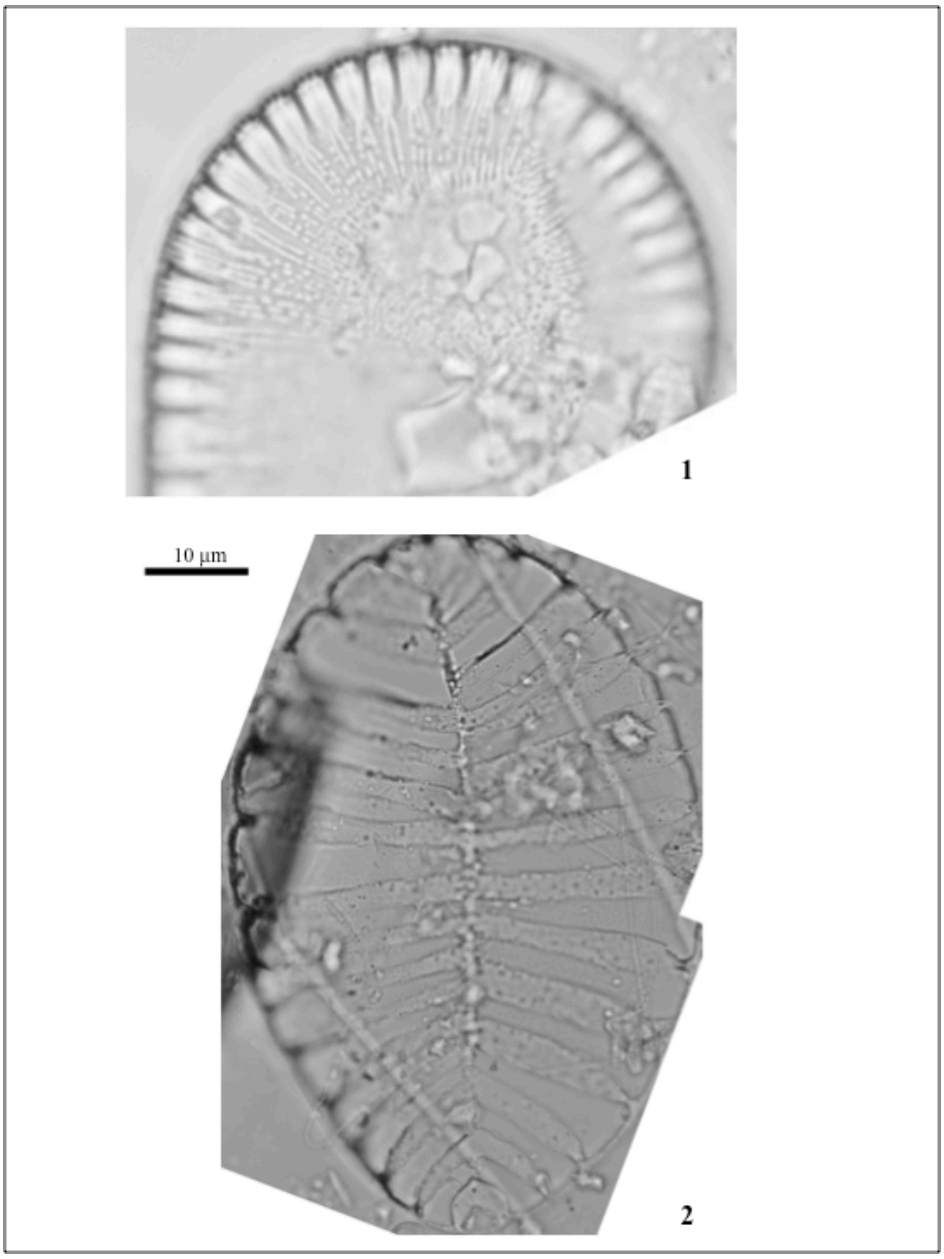


Figure A1.58: Identification plate for *Surirella* spp.

1: *Surirella ovalis* Brébisson 1838

2: *Surirella striatula* Turpin 1816-1829

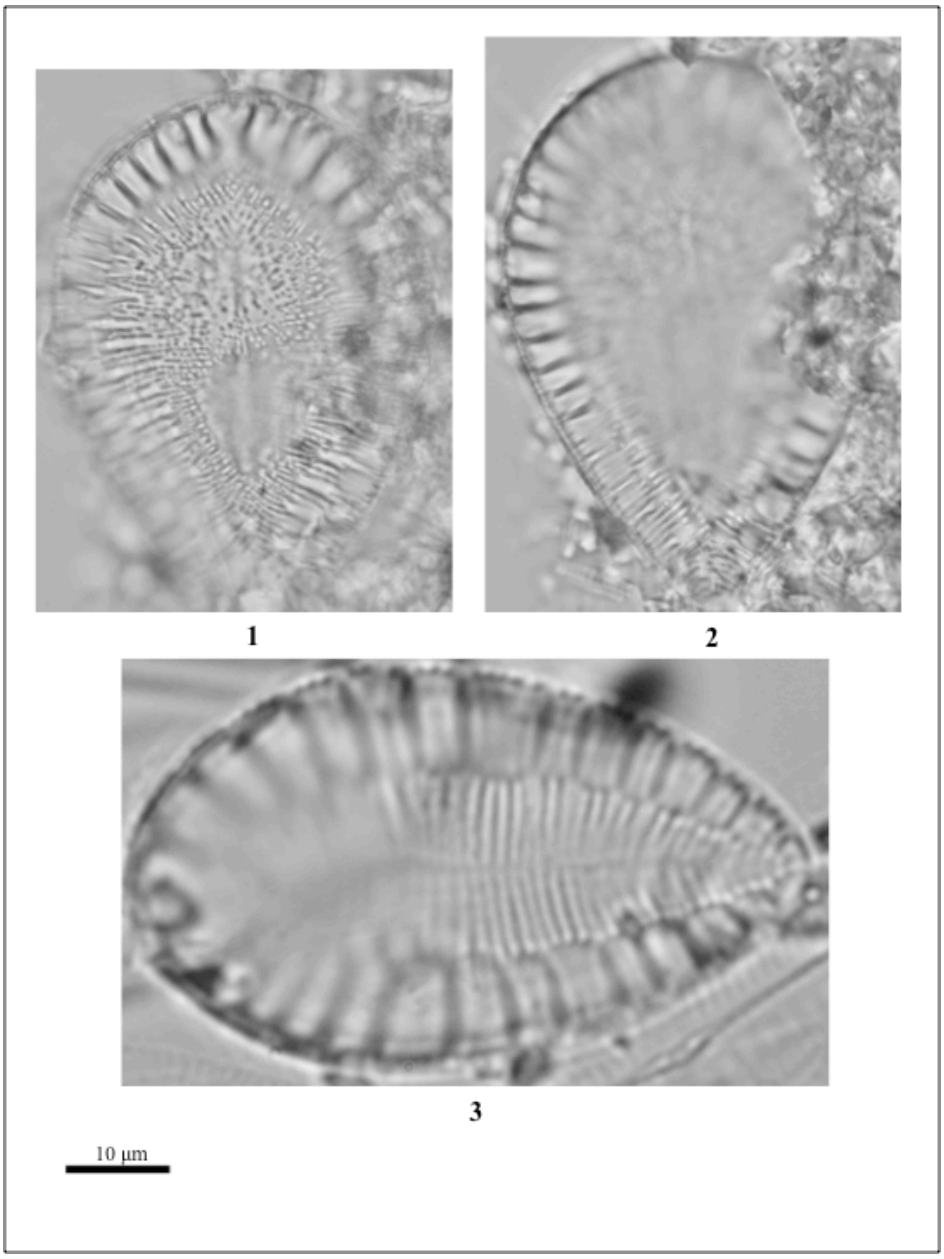


Figure A1.59: Identification plate for *Surirella brebissonii*

1 – 3: *Surirella brebissonii* Krammer & Lange-Bertalot 1987

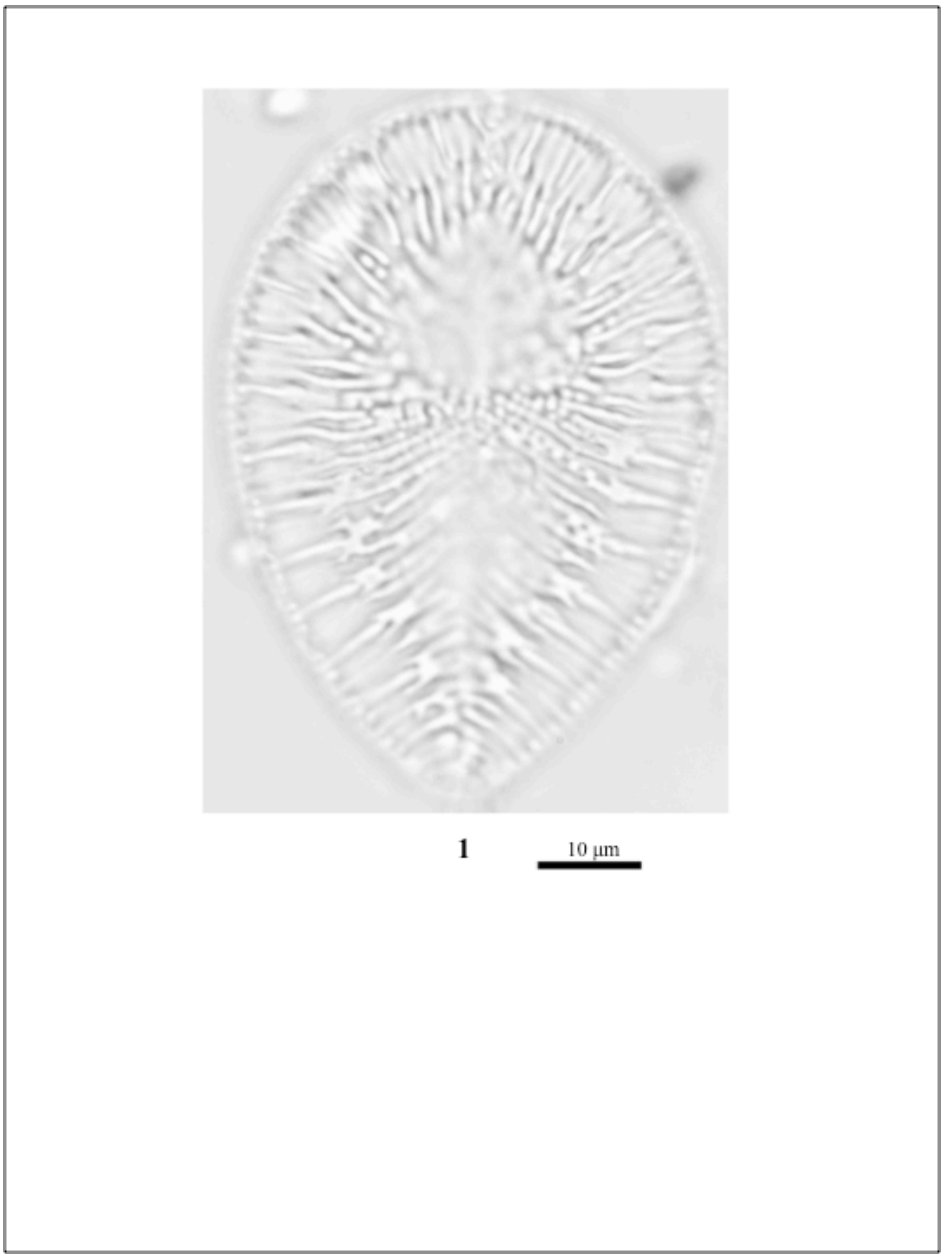


Figure A1.60: Identification plate for *Surirella brebissonii*

1: *Surirella brebissonii* Krammer & Lange-Bertalot 1987

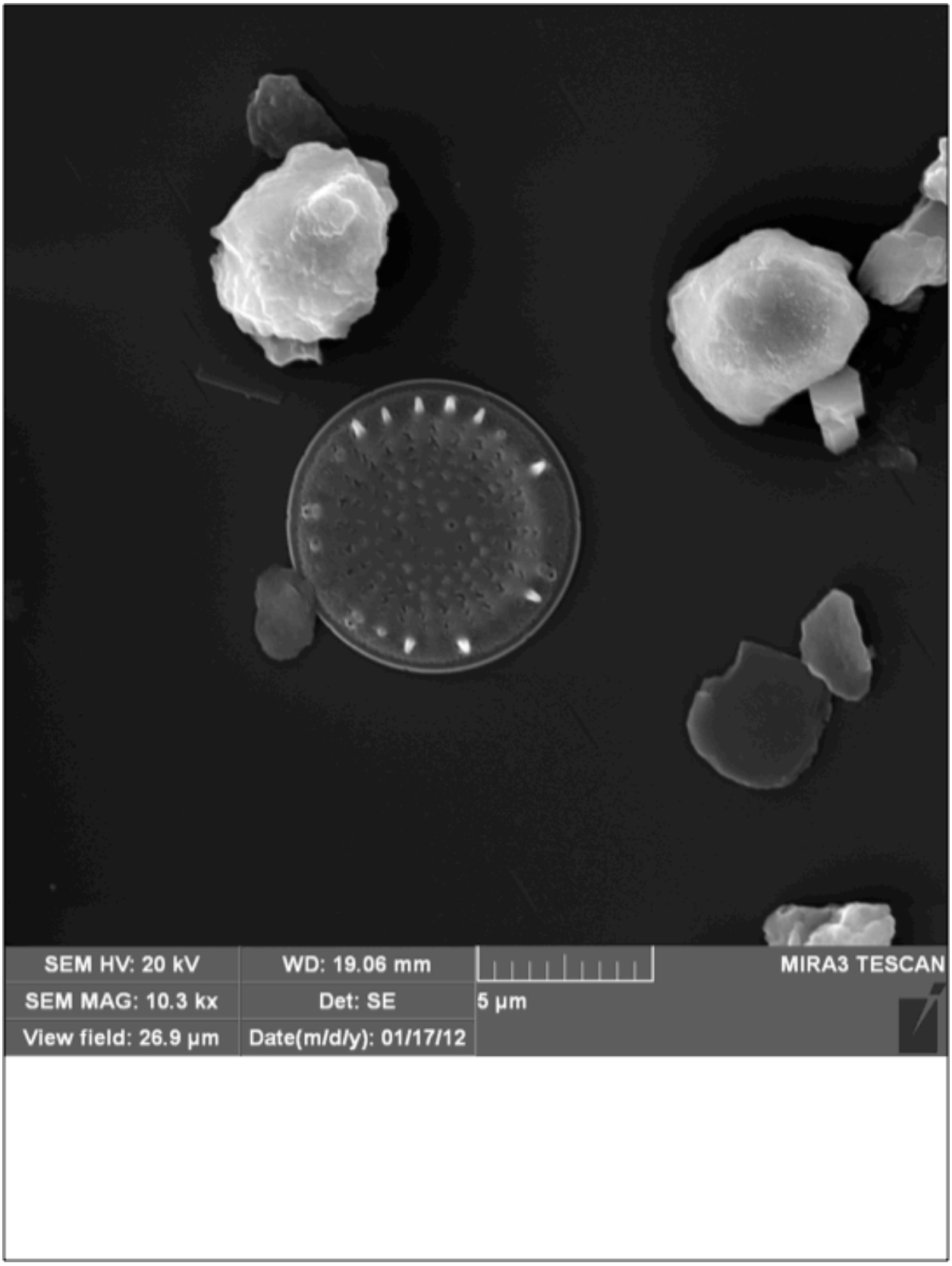


Figure A1.61: SEM identification plate for *Stephanodiscus minutulus*

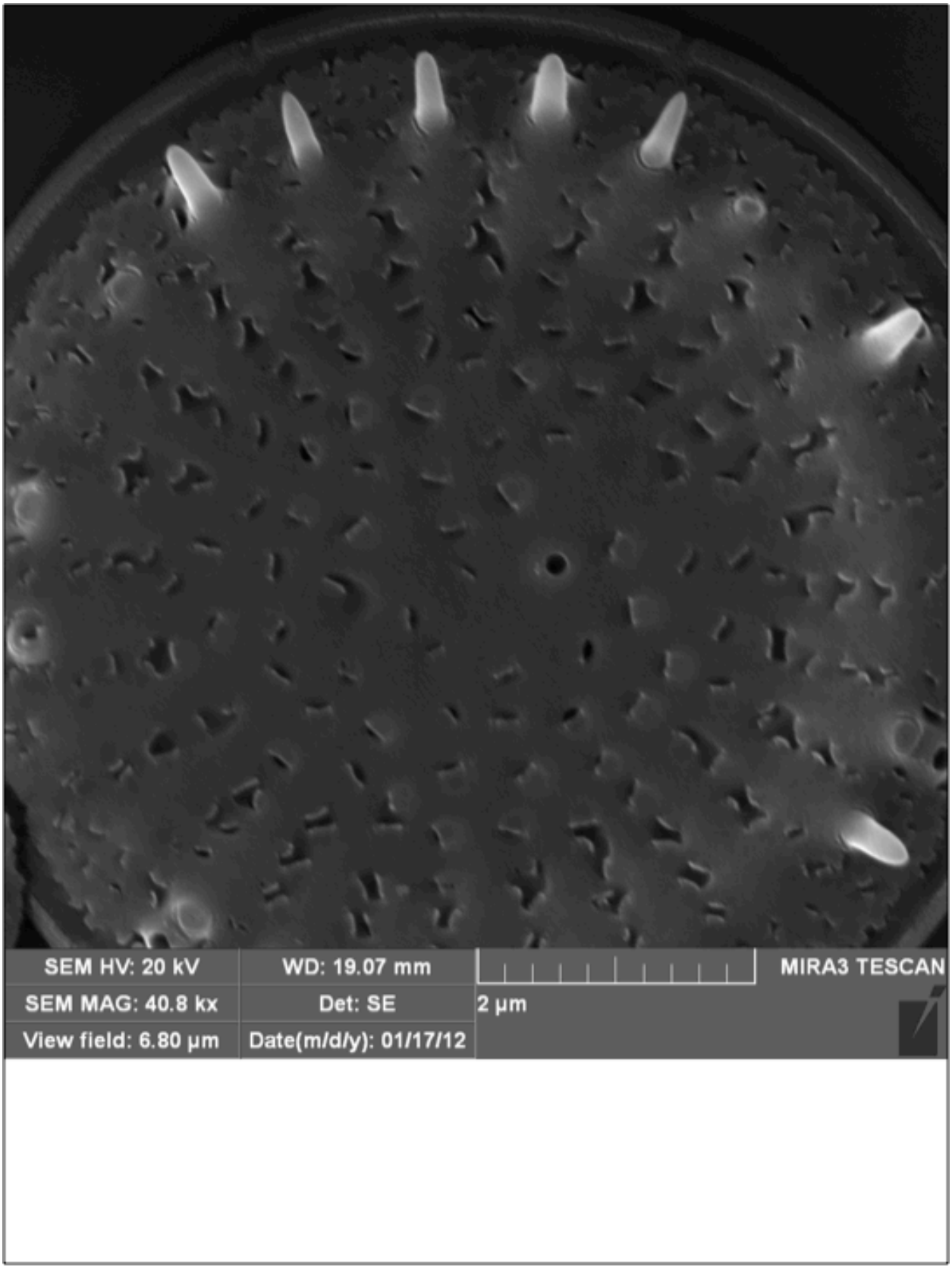


Figure A1.62: SEM identification plate for *Stephanodiscus minutulus*

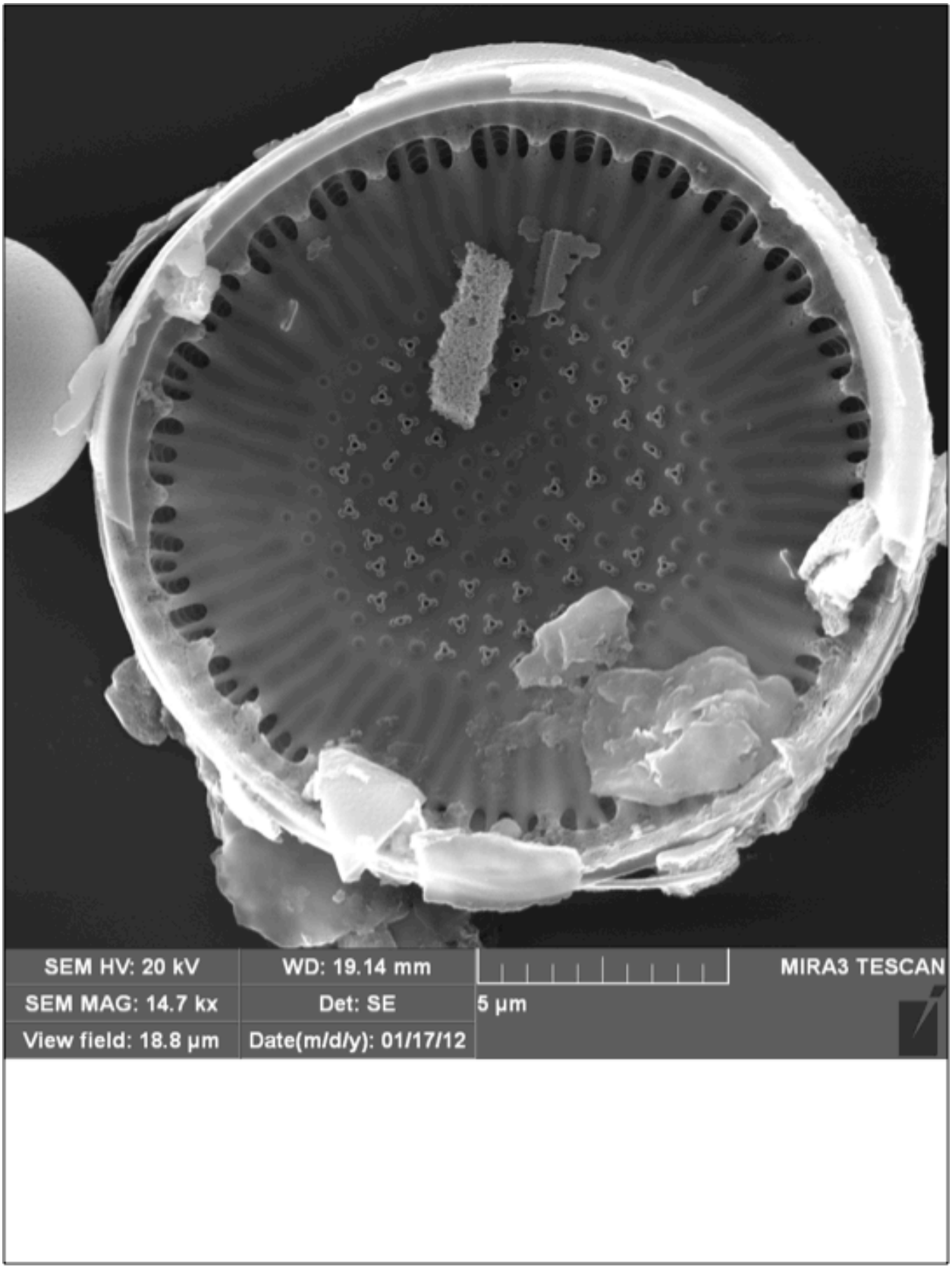


Figure A1.63: SEM identification plate for *Cyclotella bodanica*

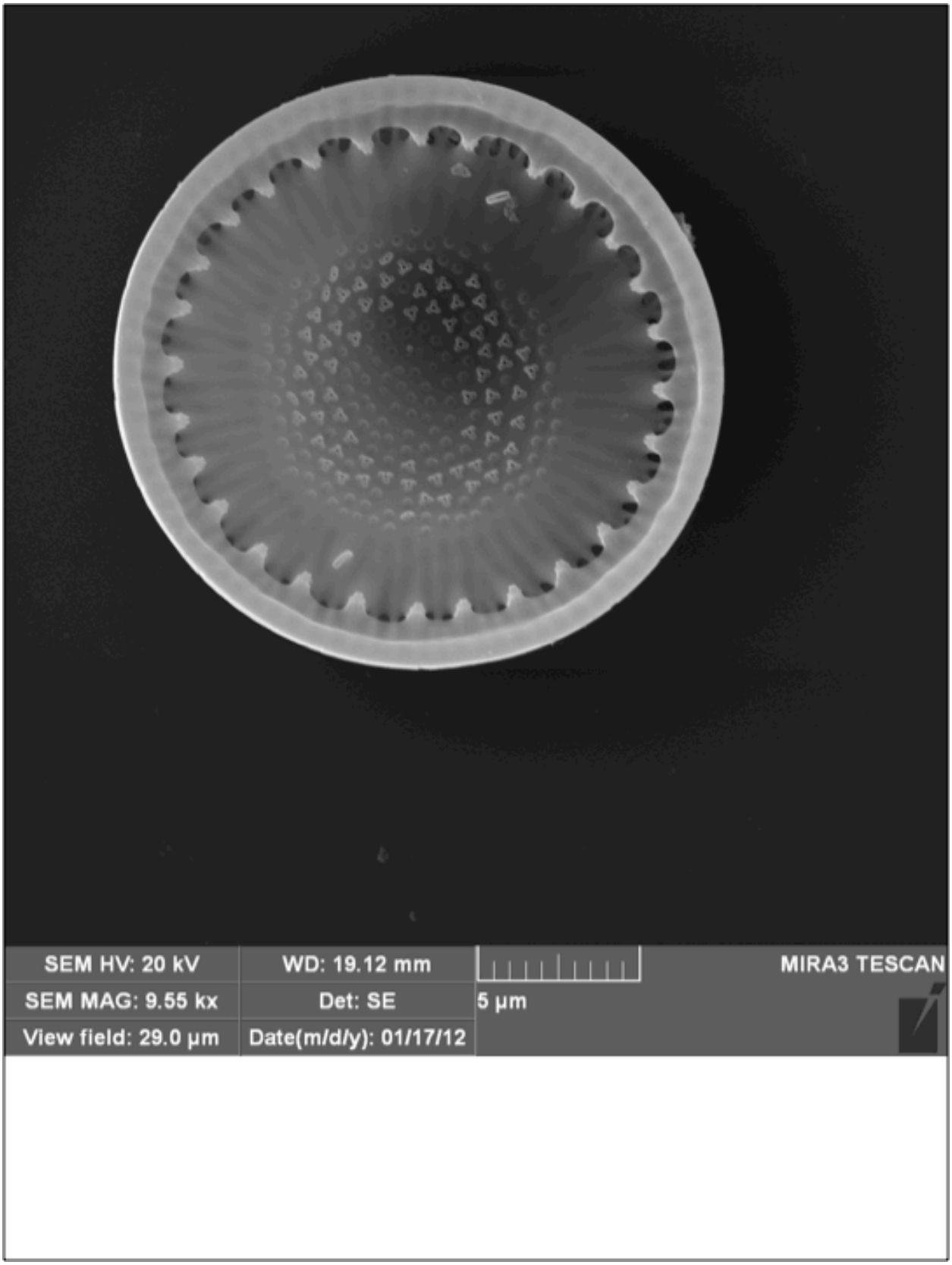


Figure A1.64: SEM identification plate for *Cyclotella bodanica*

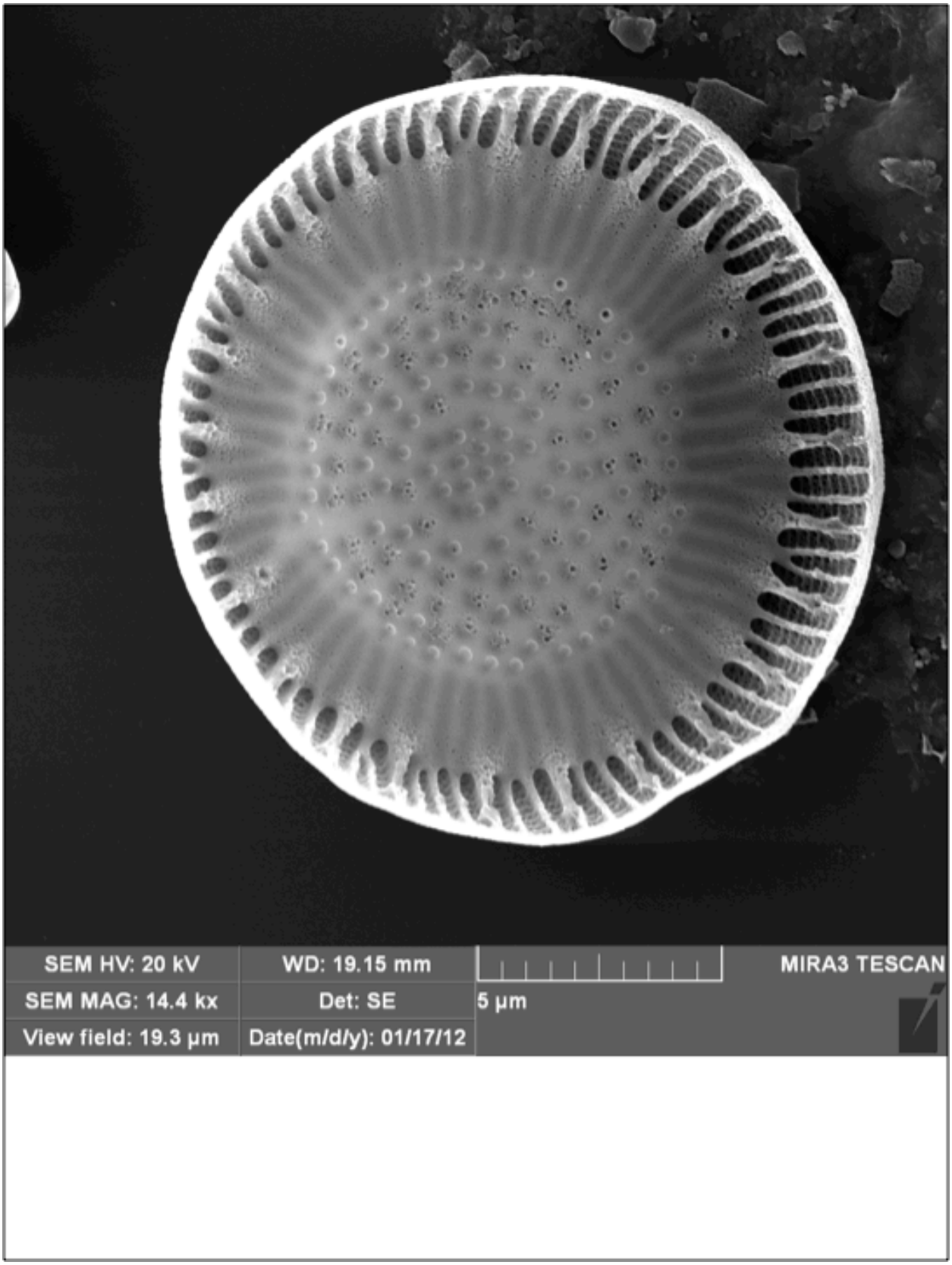


Figure A1.65: SEM identification plate for *Cyclotella bodanica* var. *lemanica*

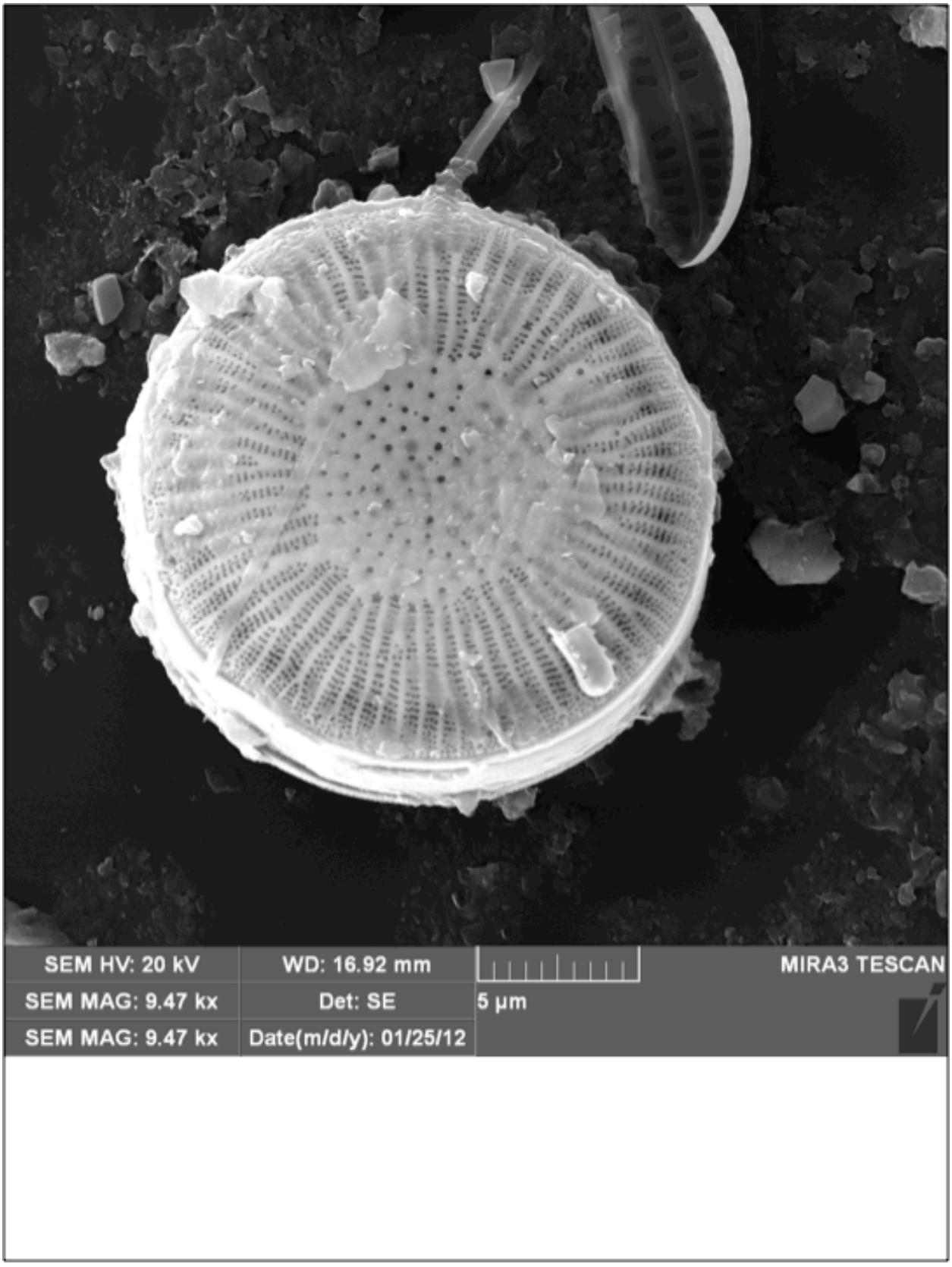


Figure A1.66: SEM identification plate for *Cyclotella bodanica*

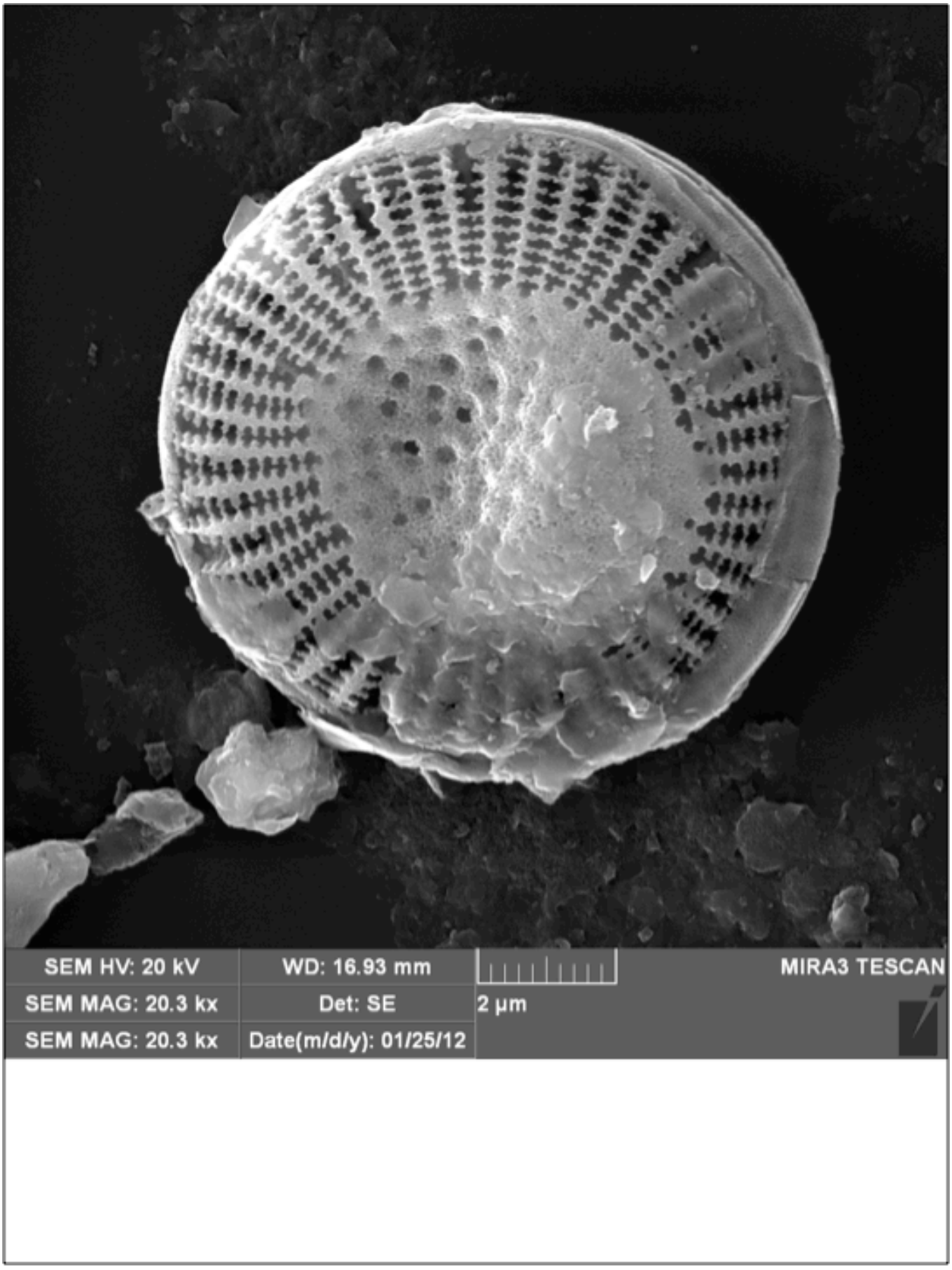


Figure A1.67: SEM identification plate for *Cyclotella michiganiana*

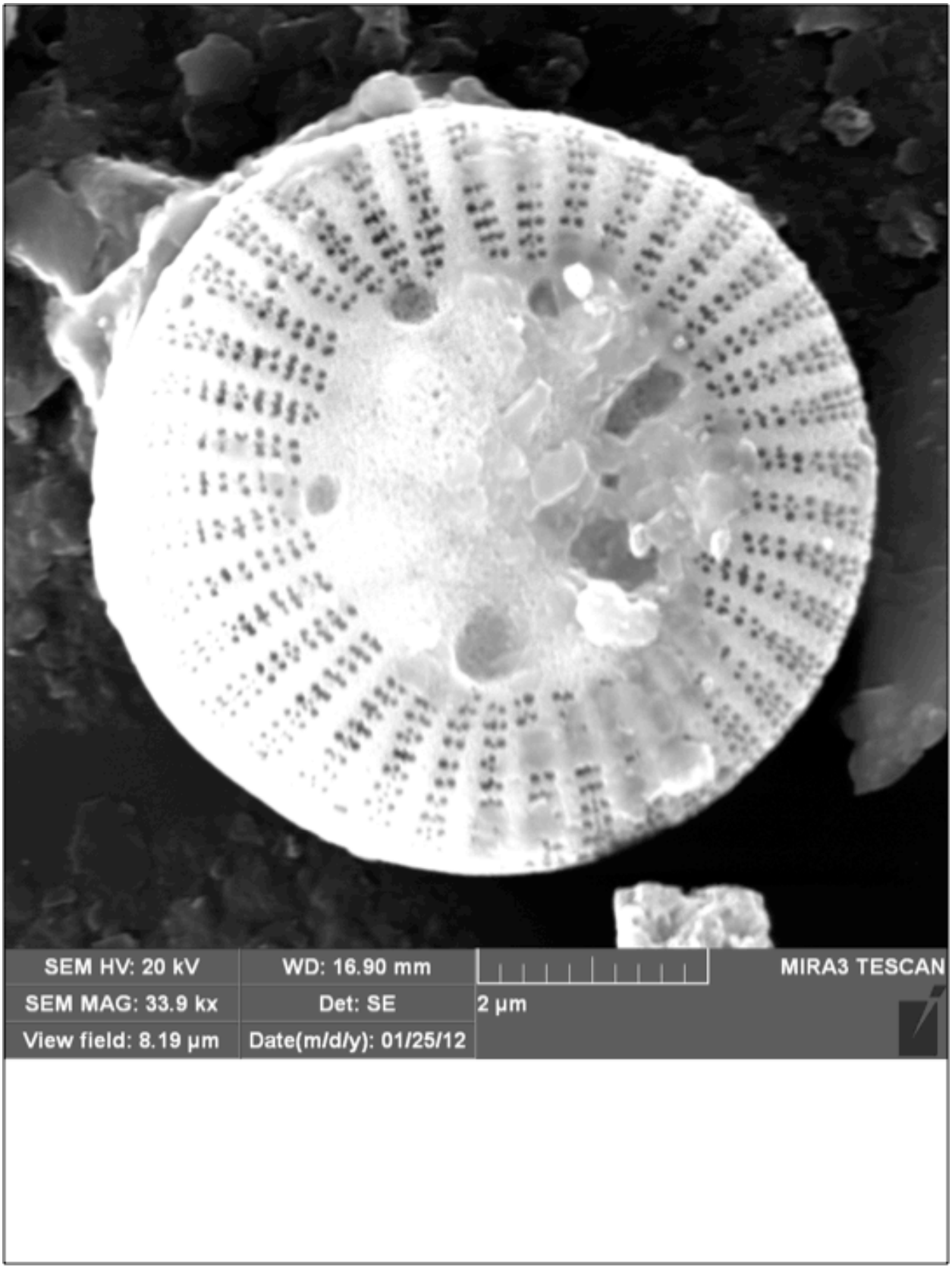


Figure A1.68: SEM identification plate for *Cyclotella comensis*

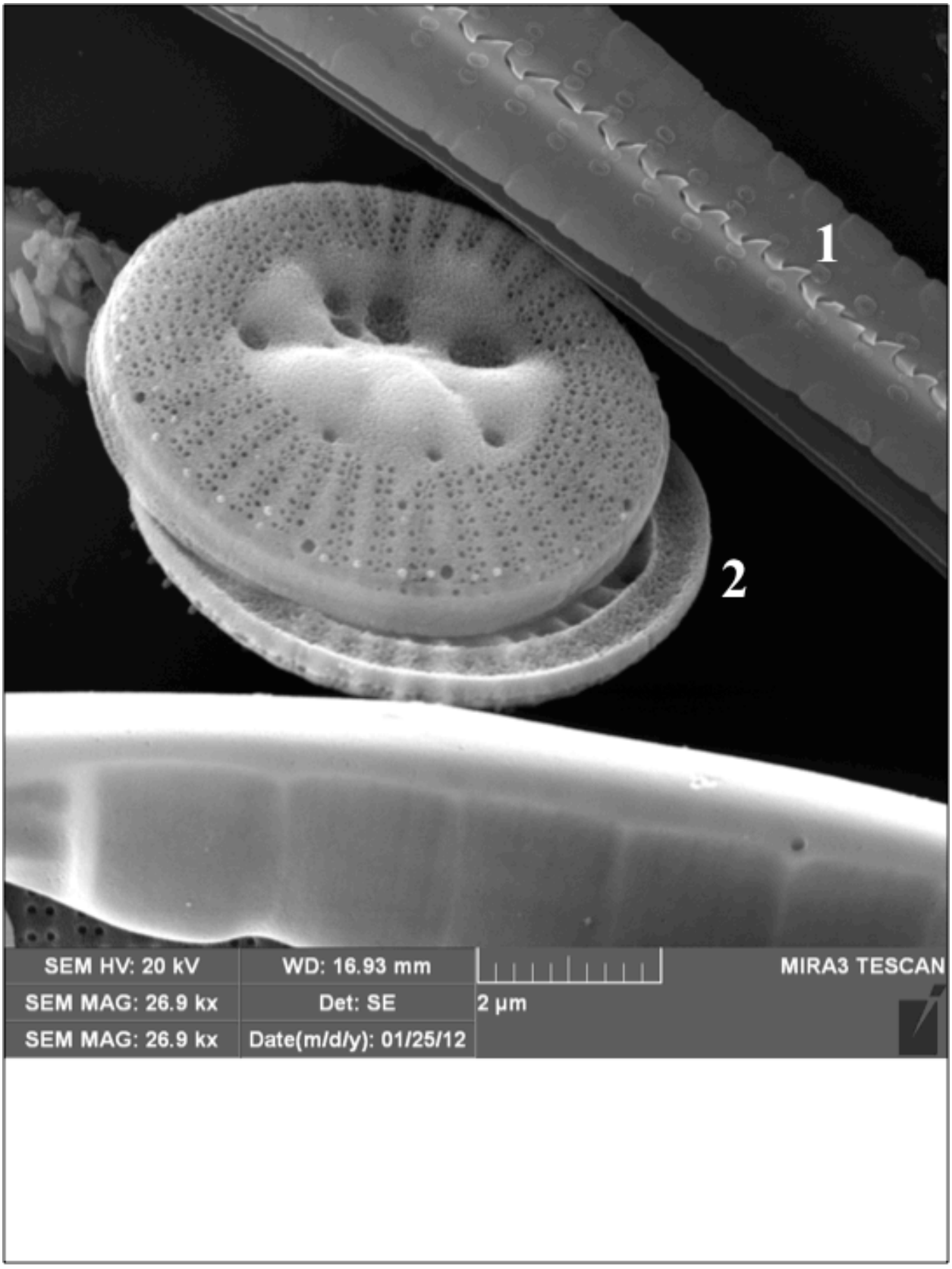


Figure A1.69: SEM identification plate for *Fragilaria crotonensis* and *Cyclotella comensis*

1 – *Fragilaria crotonensis*, 2 – *Cyclotella comensis*

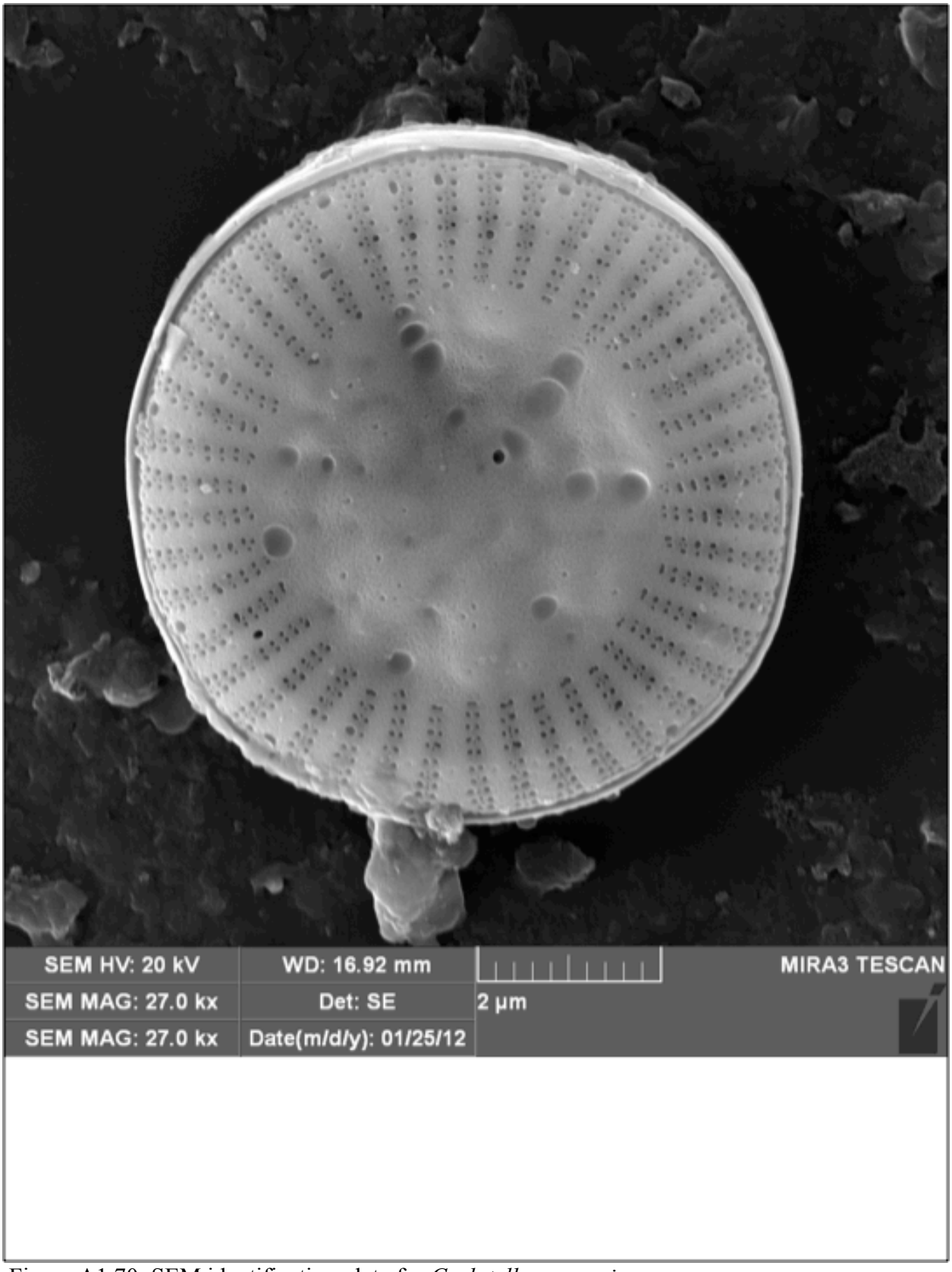


Figure A1.70: SEM identification plate for *Cyclotella comensis*

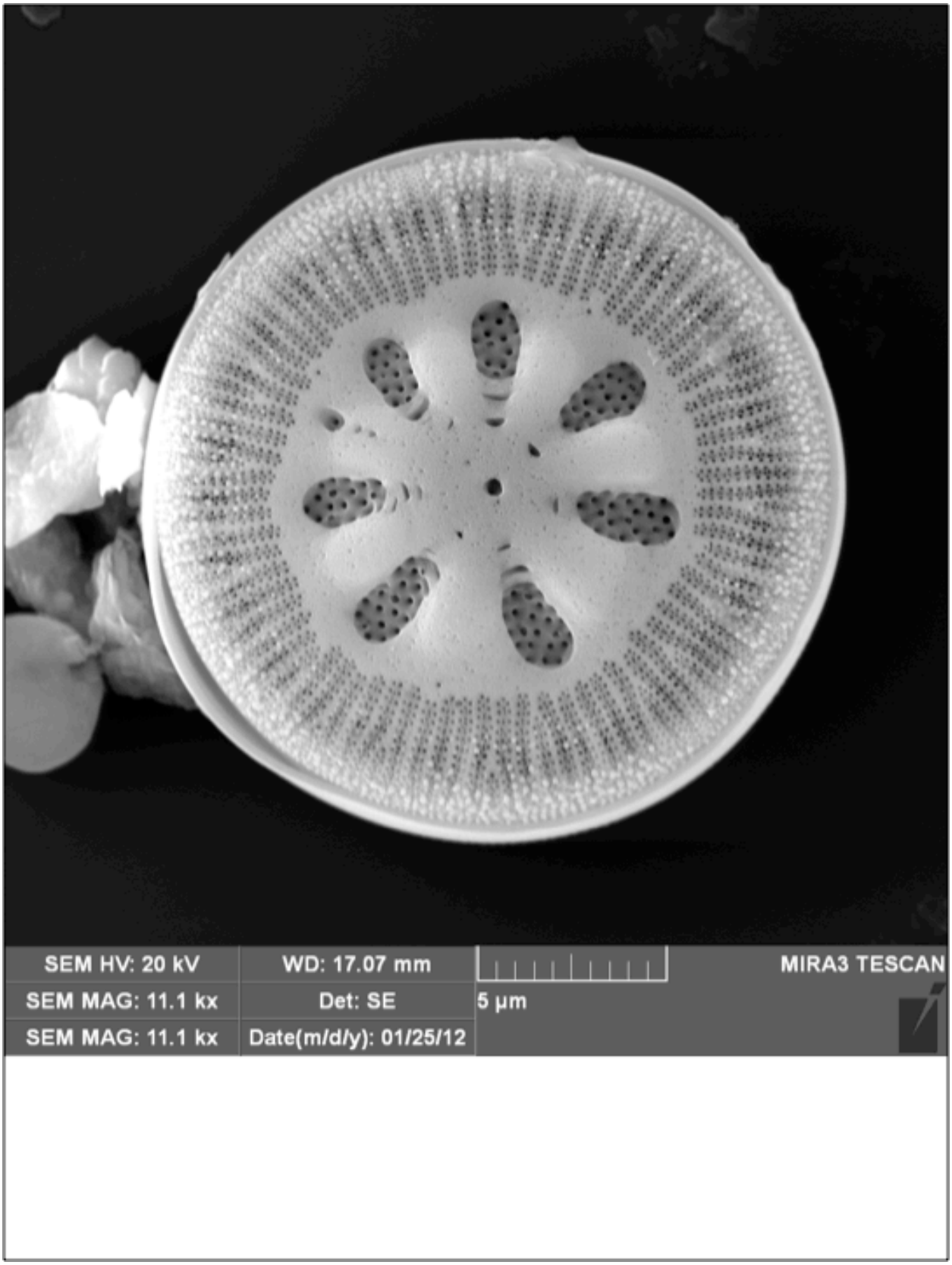


Figure A1.71: SEM identification plate for *Cyclotella antiqua*

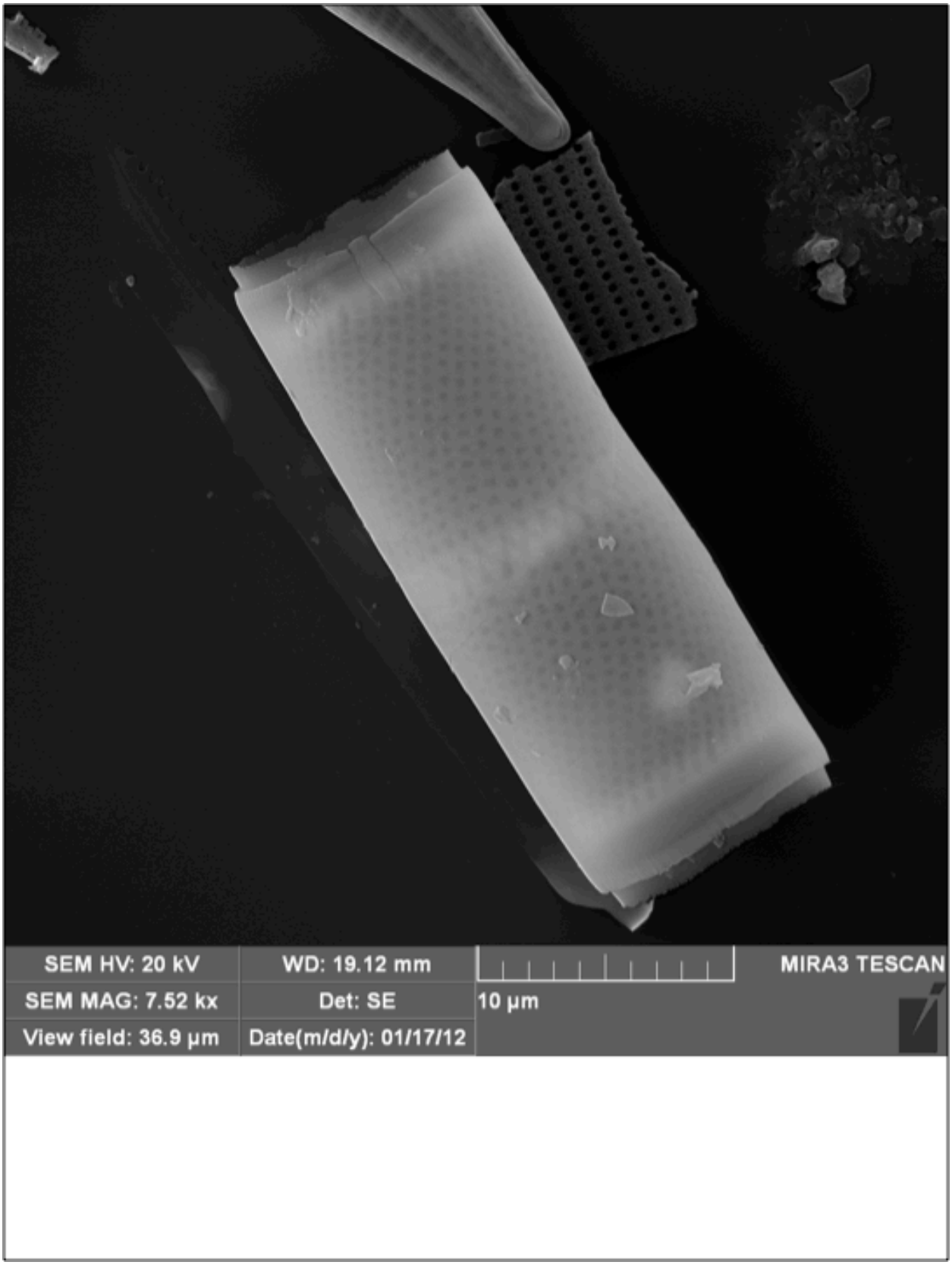


Figure A1.72: SEM identification plate for *Aulacoseira ambigua*

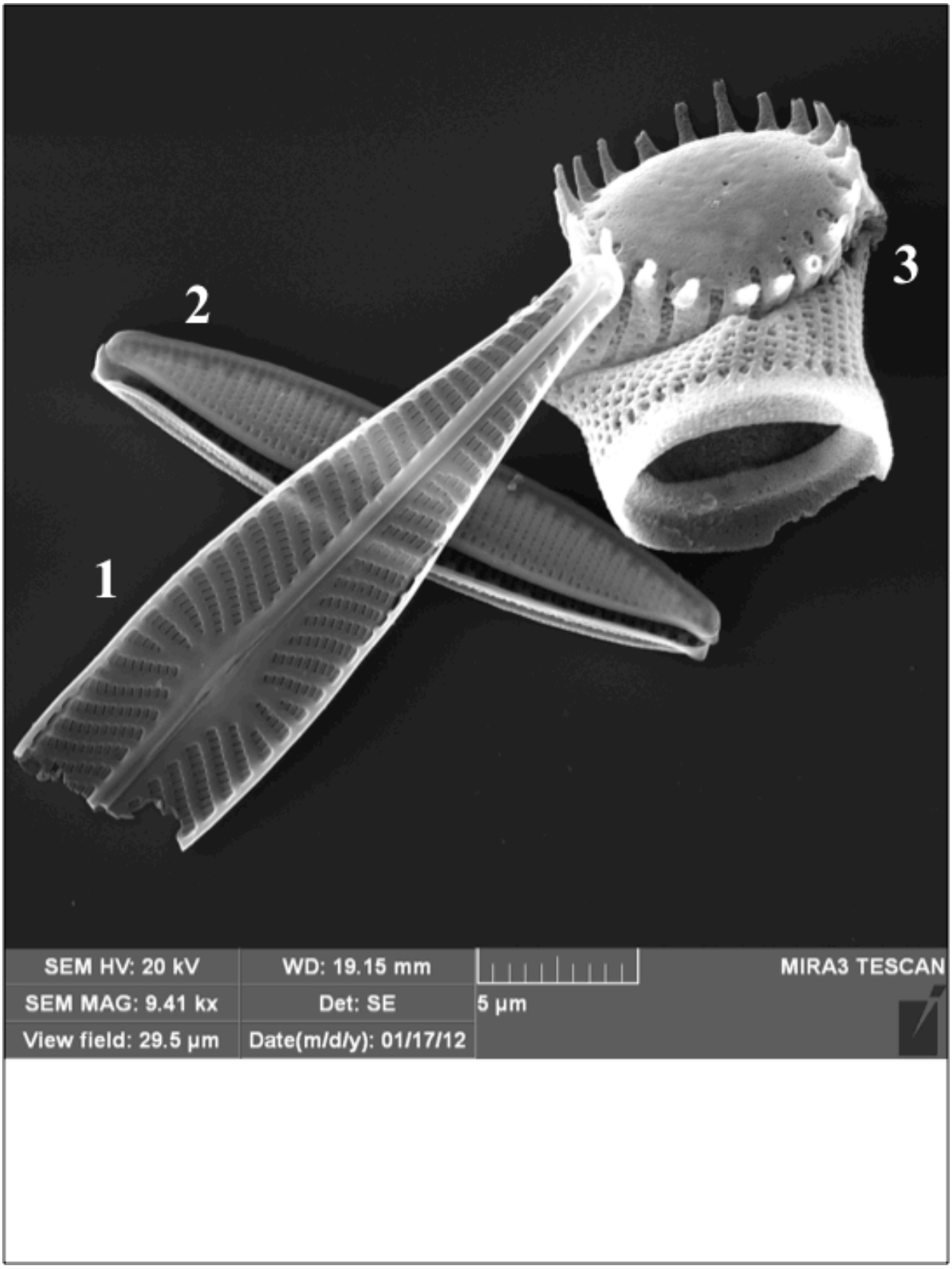


Figure A1.73: SEM identification plate for *Navicula trivialis, Nitzschia frustulum*, and *Aulacoseira ambigua*

1 – *Navicula trivialis*, 2 – *Nitzschia frustulum*, 3 – *Aulacoseira ambigua*

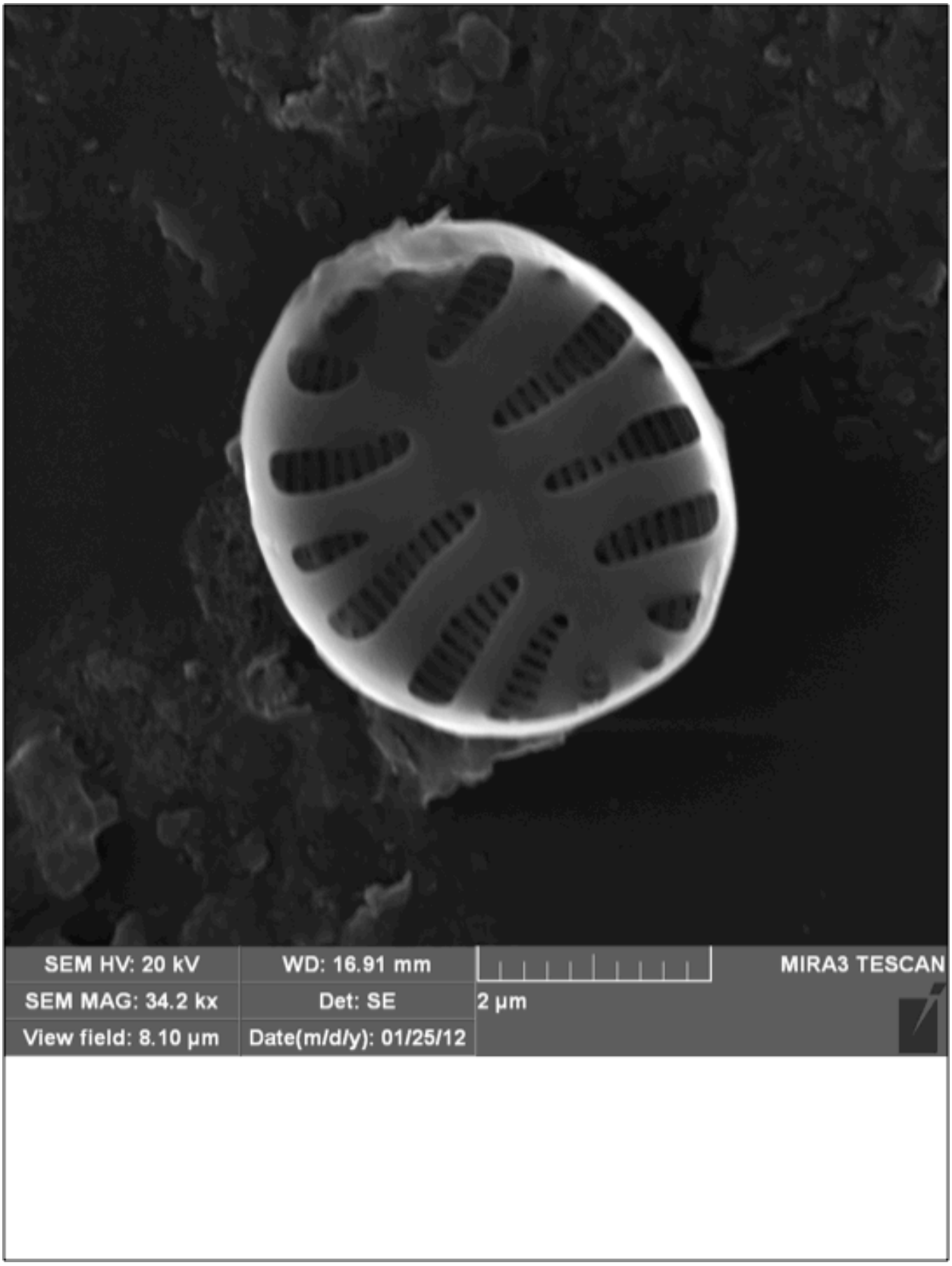


Figure A1.74: SEM identification plate for *Staurosirella pinnata*

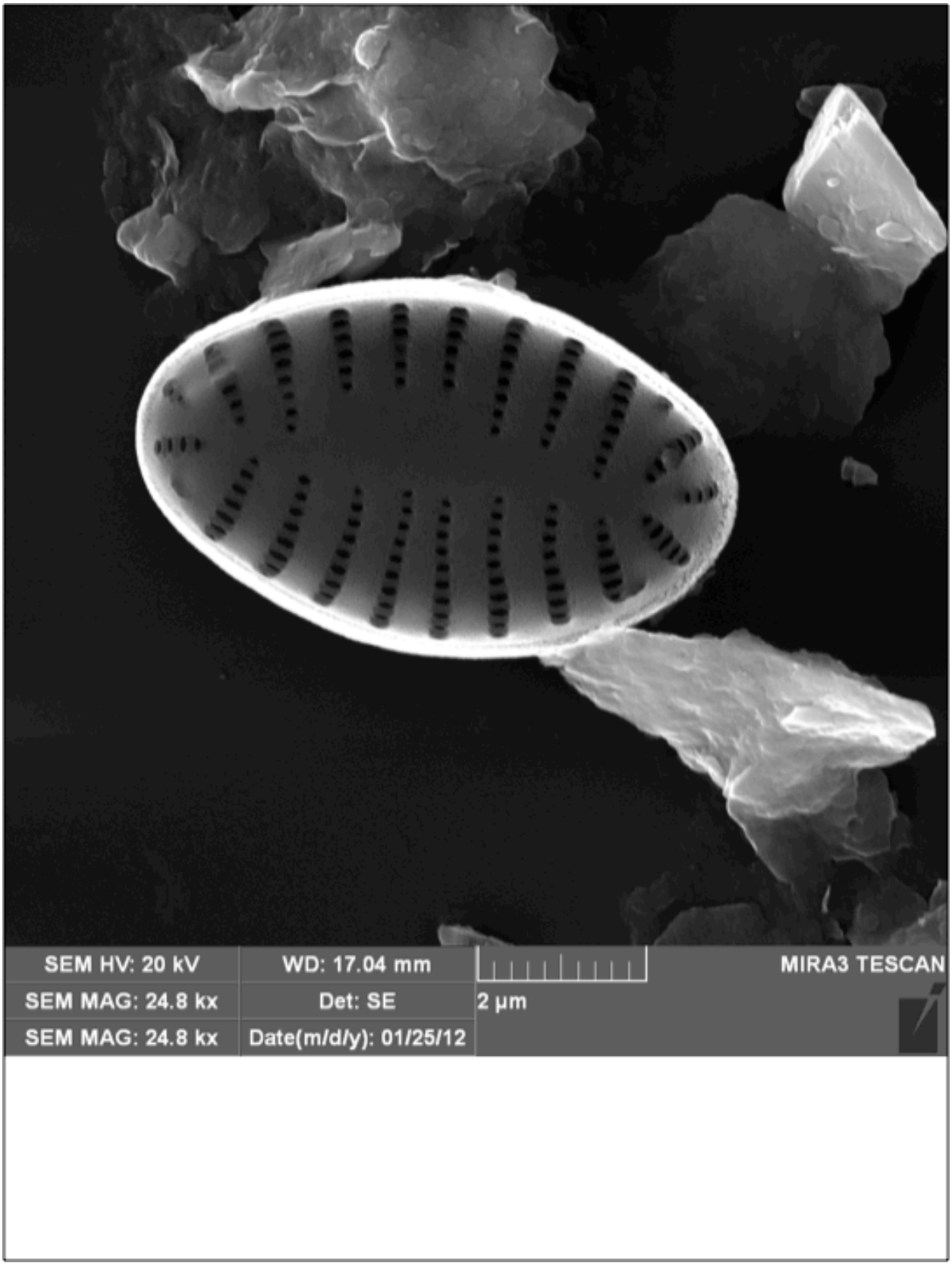


Figure A1.75: SEM identification plate for *Staurosirella pinnata*

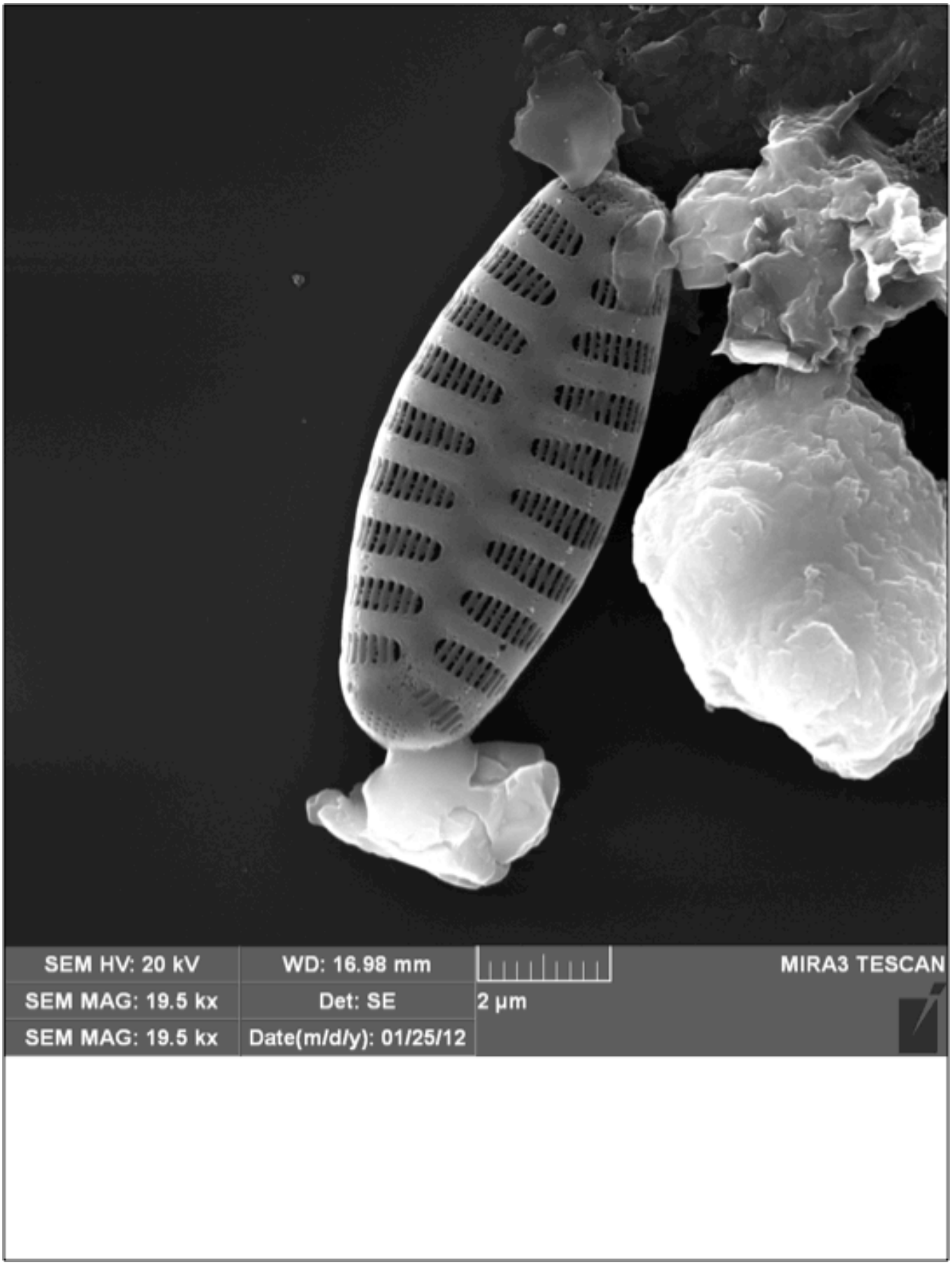


Figure A1.76: SEM identification plate for *Fragilaria brevistriata*

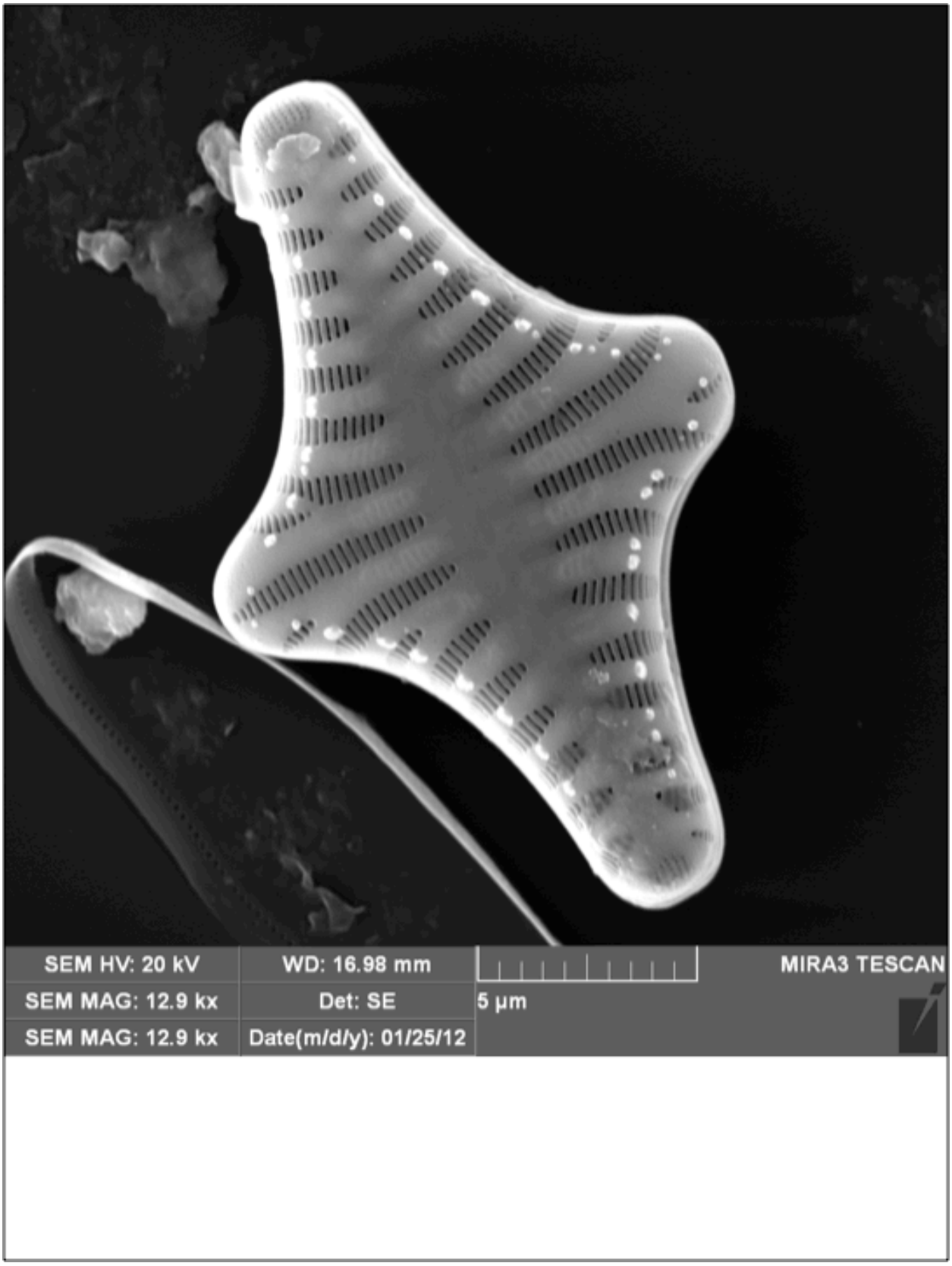


Figure A1.77: SEM identification plate for *Pseudostaurosira parasitica*

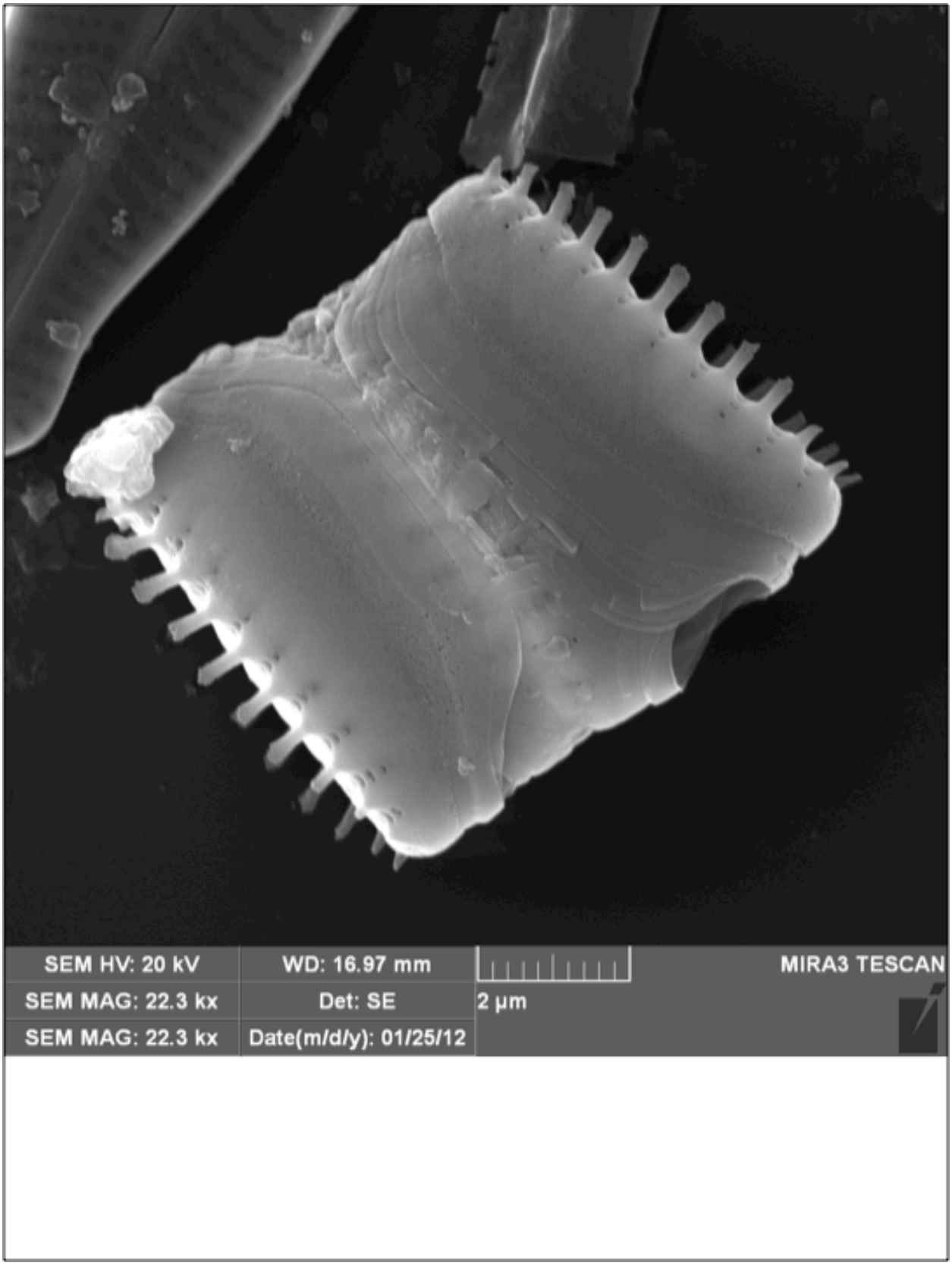


Figure A1.78: SEM identification plate for *Fragilaria construens*

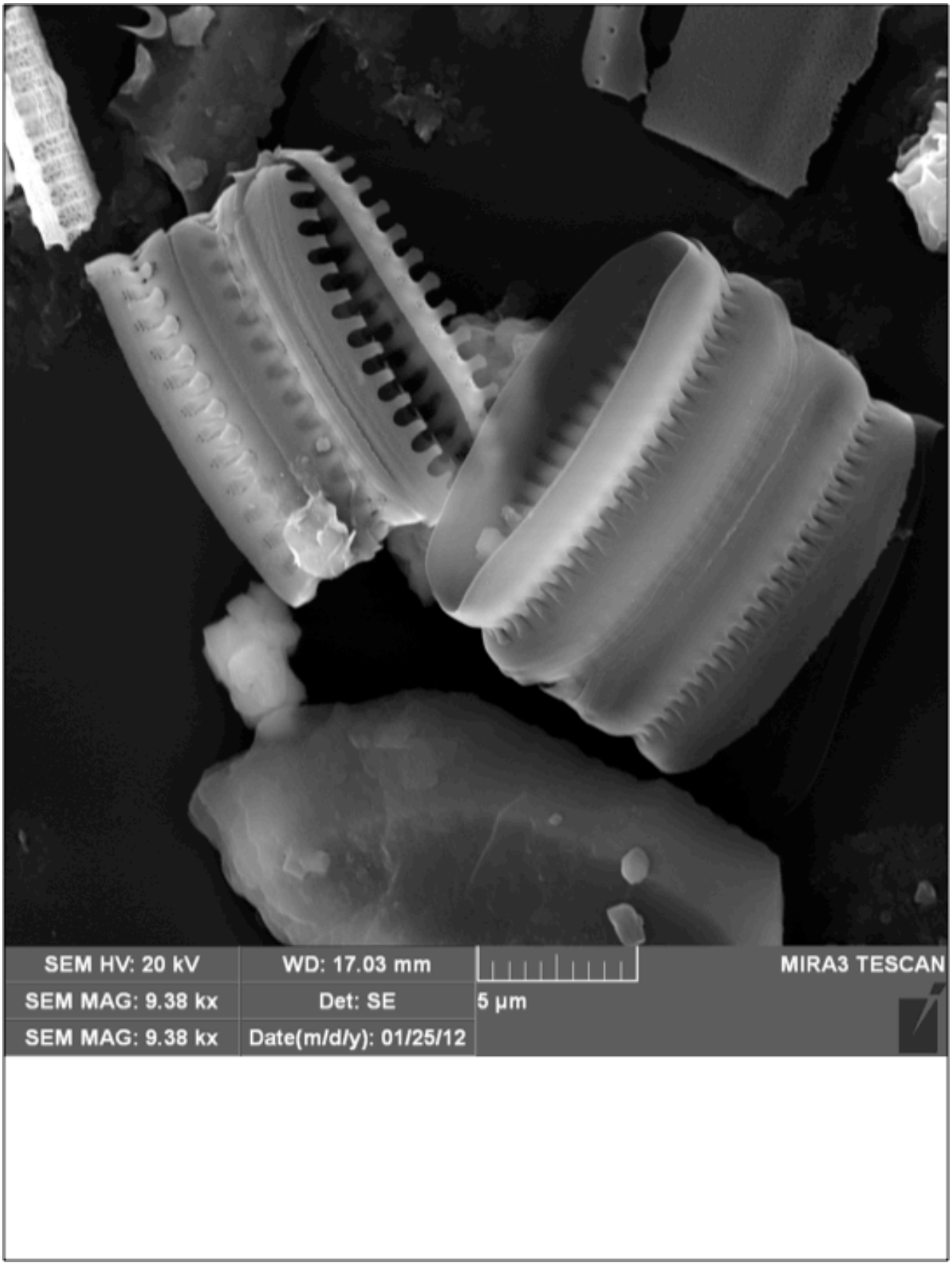


Figure A1.79: SEM identification plate for *Fragilaria construens*

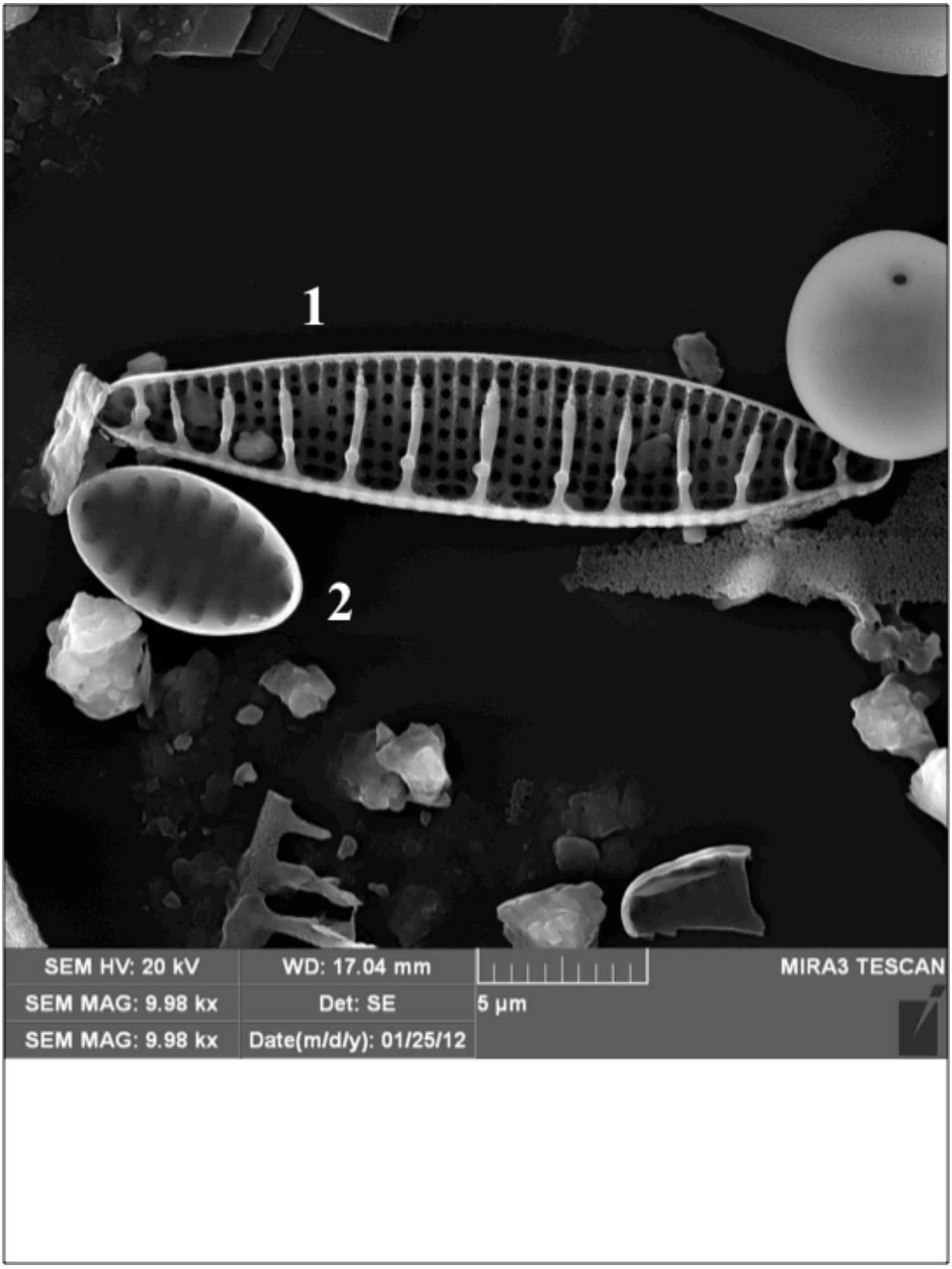


Figure A1.80: SEM identification plate for *Denticula kuetzingii* and *Staurosirella pinnata*
1 – *Denticula kuetzingii* , 2 – *Staurosirella pinnata*

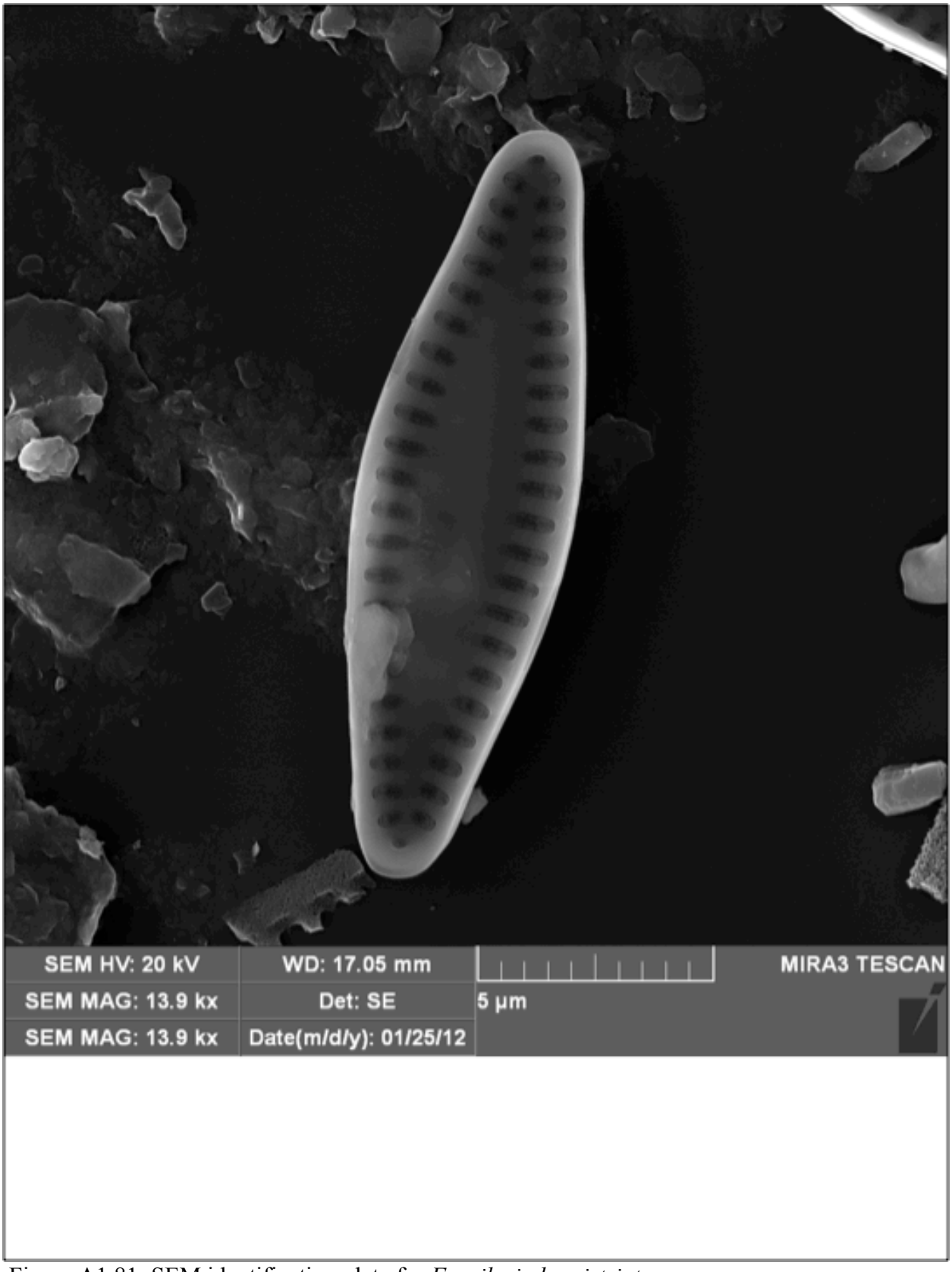


Figure A1.81: SEM identification plate for *Fragilaria brevistriata*

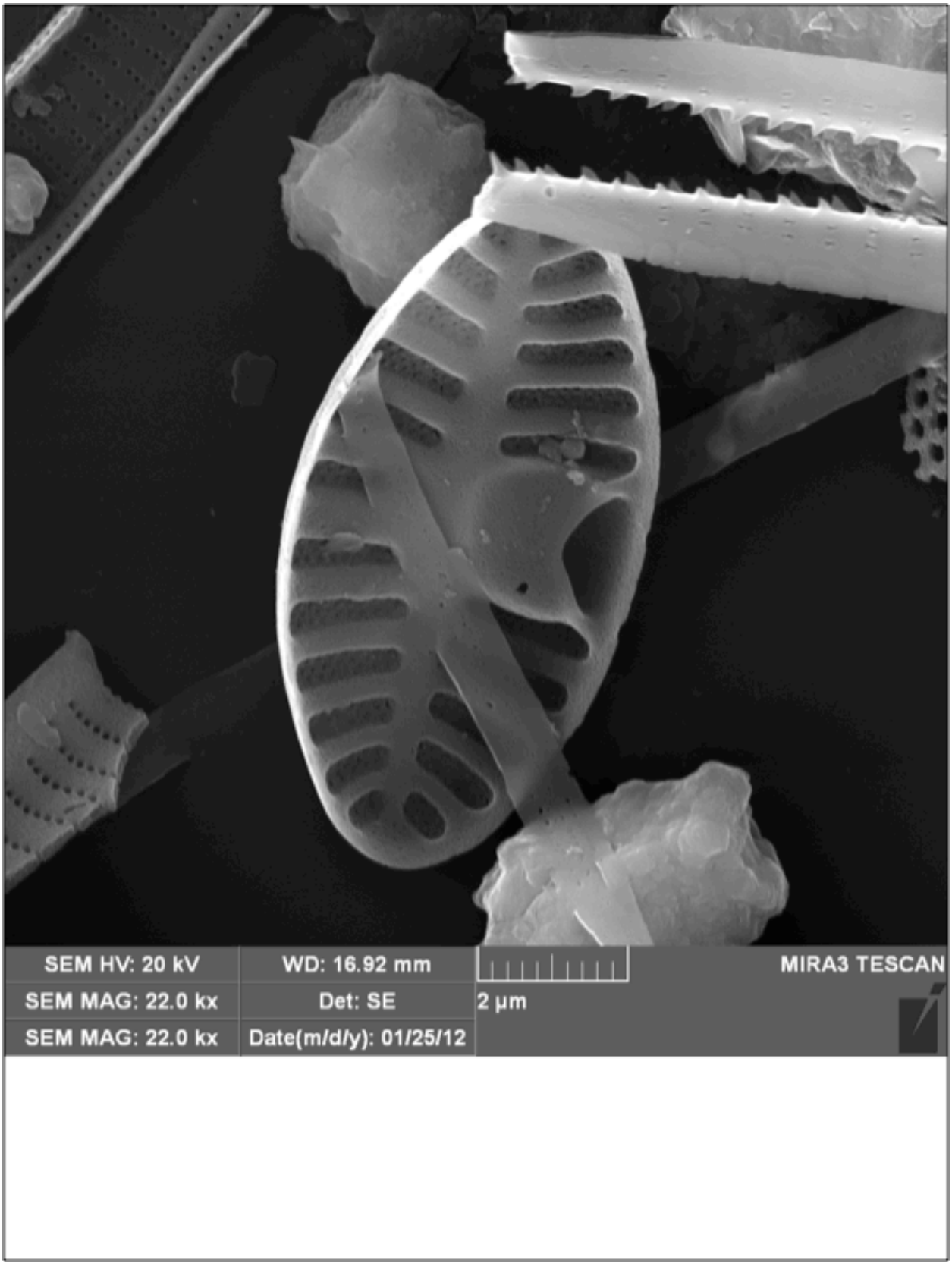


Figure A1.82: SEM identification plate for *Achnanthes lanceolata*

Figure A1.83: SEM identification plate for *Surirella angusta*

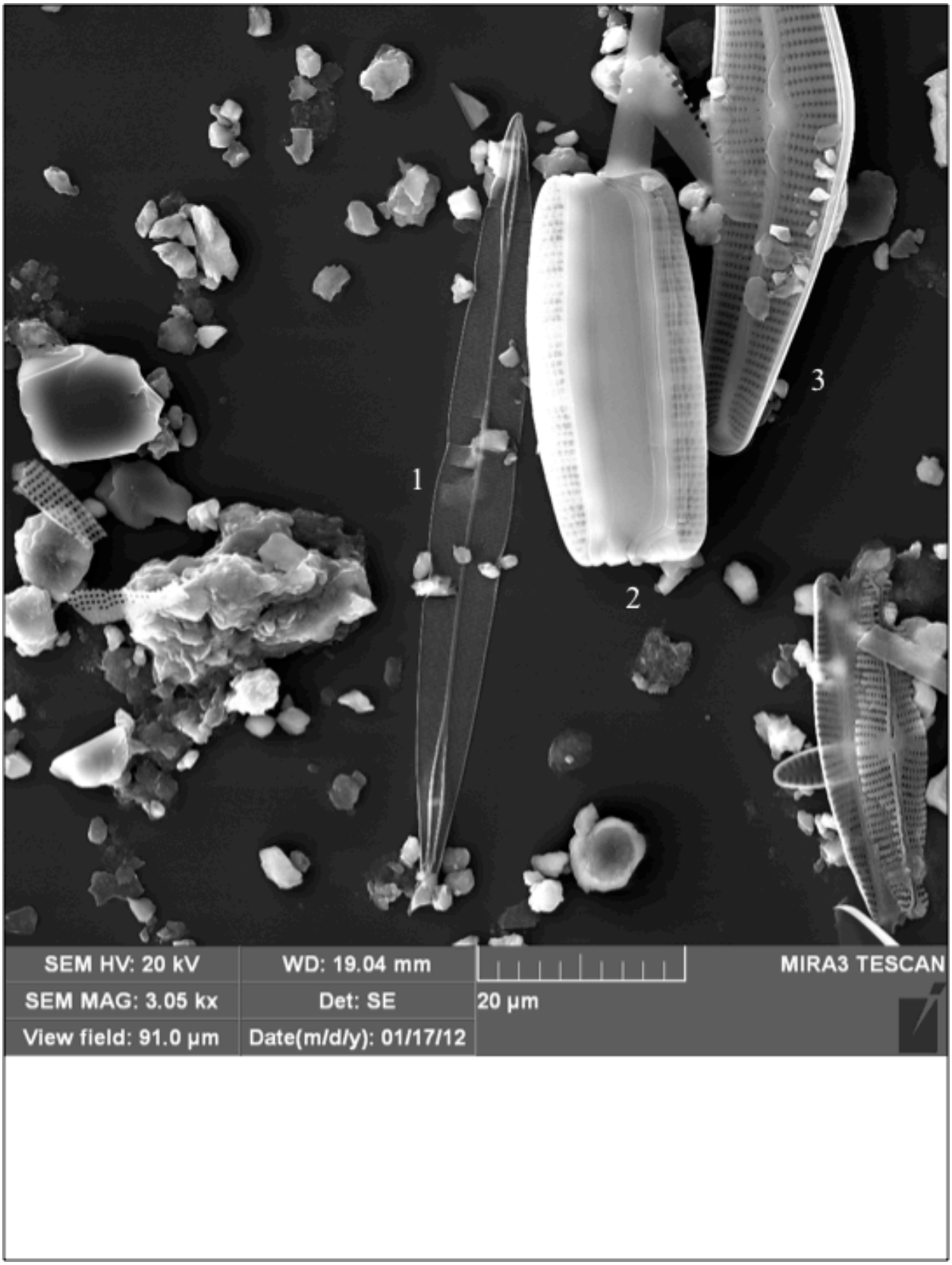


Figure A1.84: SEM identification plate for *Amphipleura pellucida, Epithemia* sp., and *Cymbella neocistula*

1 – *Amphipleura pellucida*, 2 – *Epithemia* sp., 3 – *Cymbella neocistula*

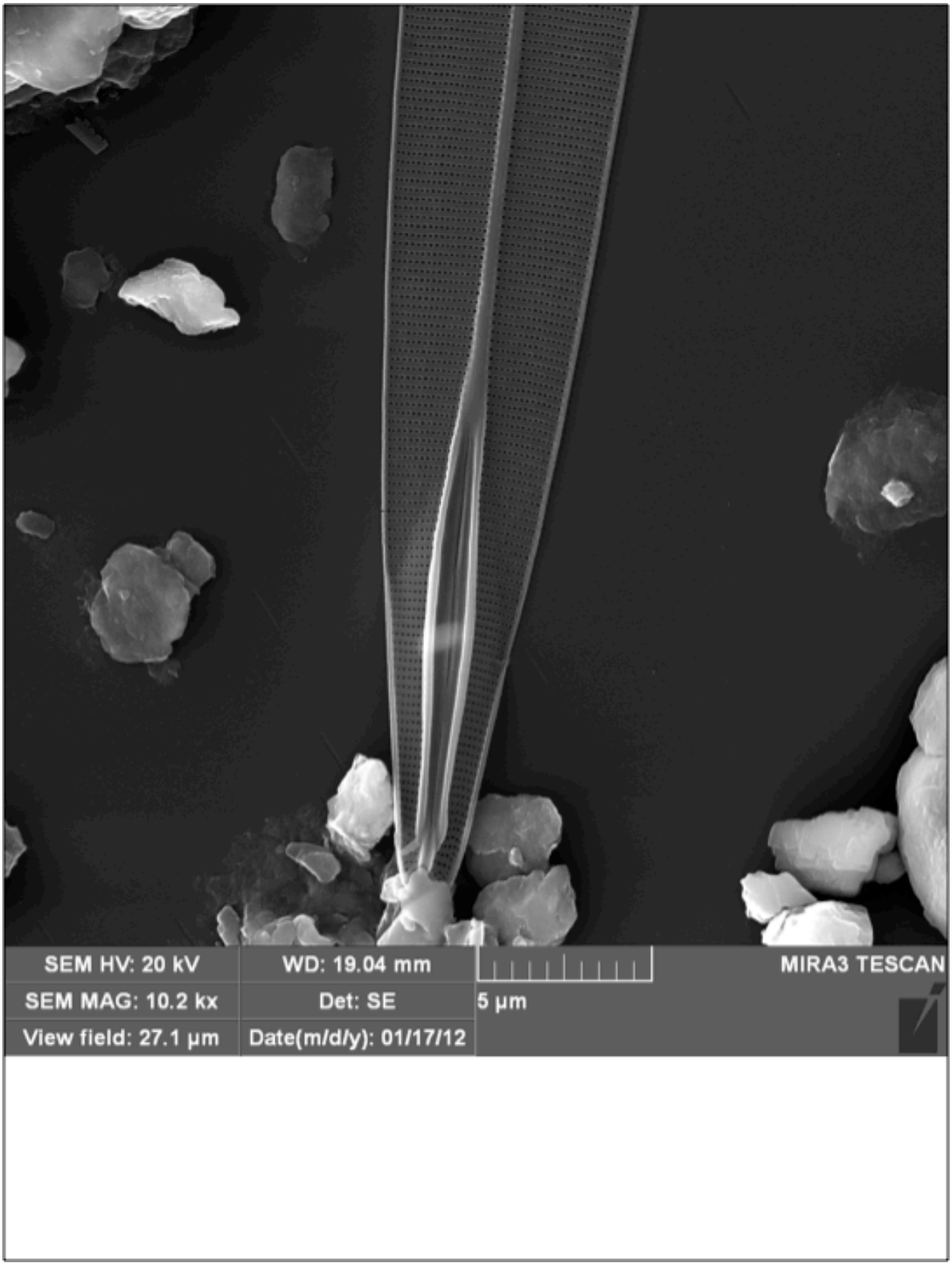


Figure A1.85: SEM identification plate for *Amphipleura pellucida*

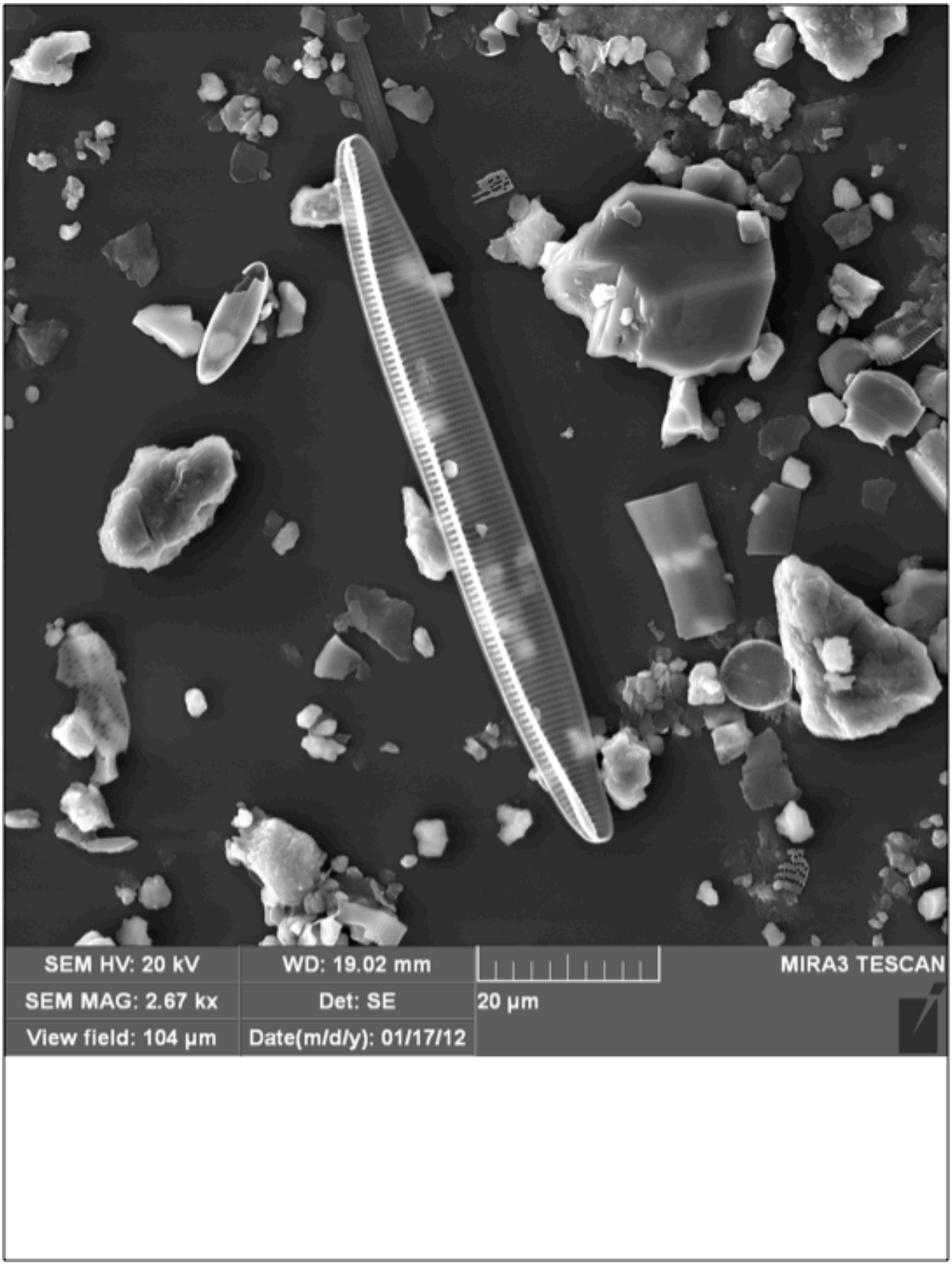


Figure A1.86: SEM identification plate for *Tryblionella angustata*

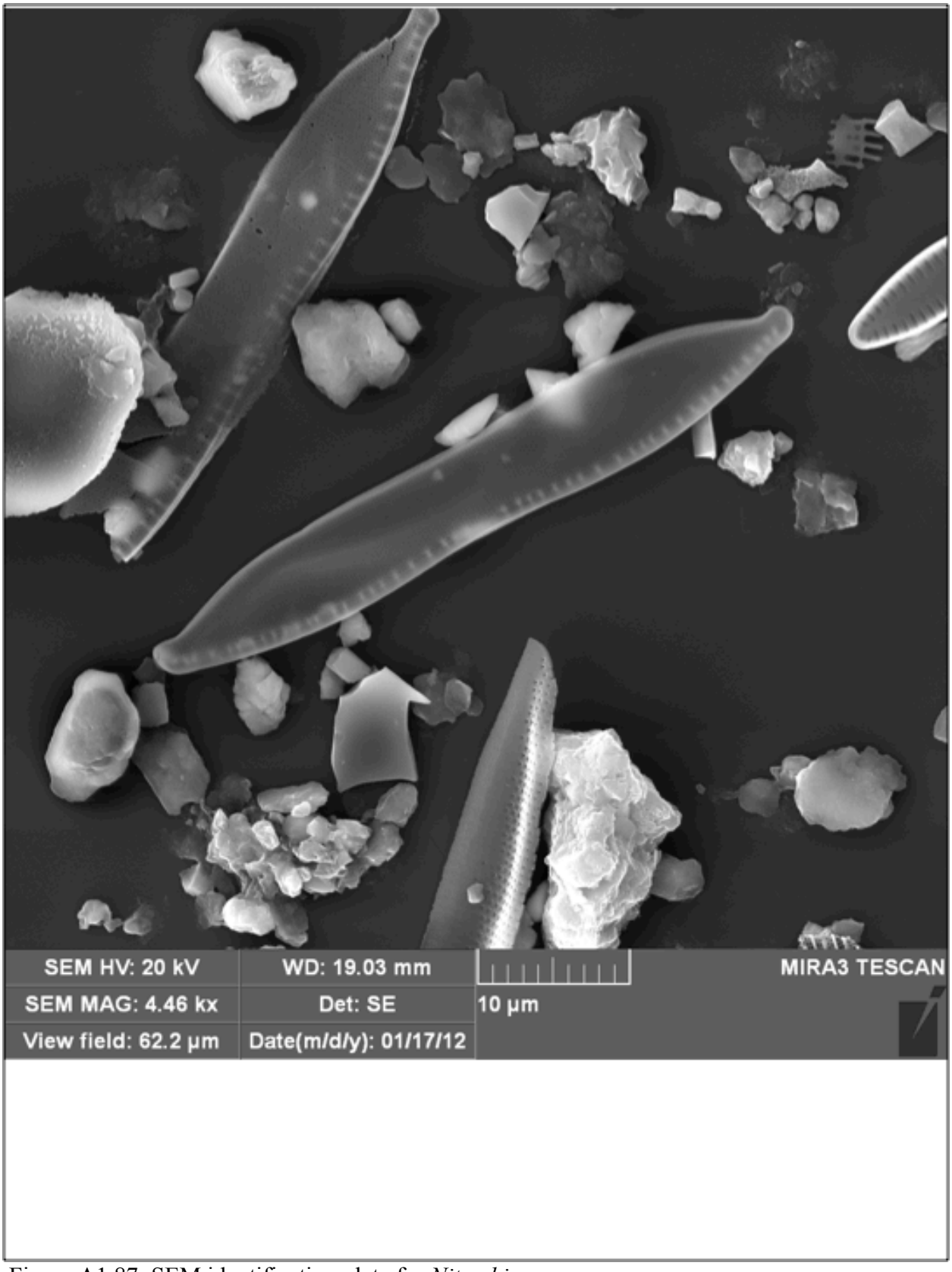


Figure A1.87: SEM identification plate for *Nitzschia* sp.

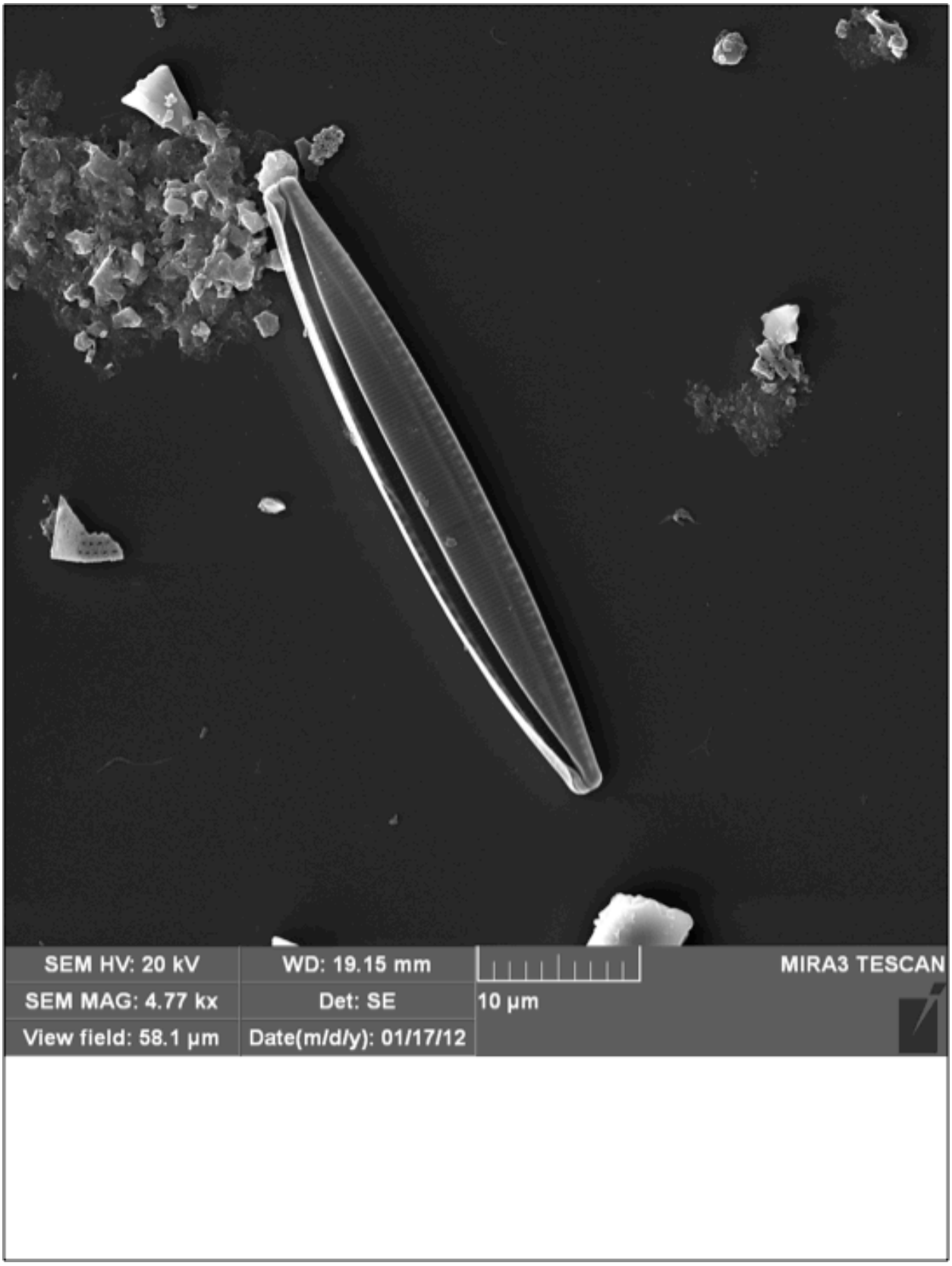


Figure A1.88: SEM identification plate for *Nitzschia* sp.

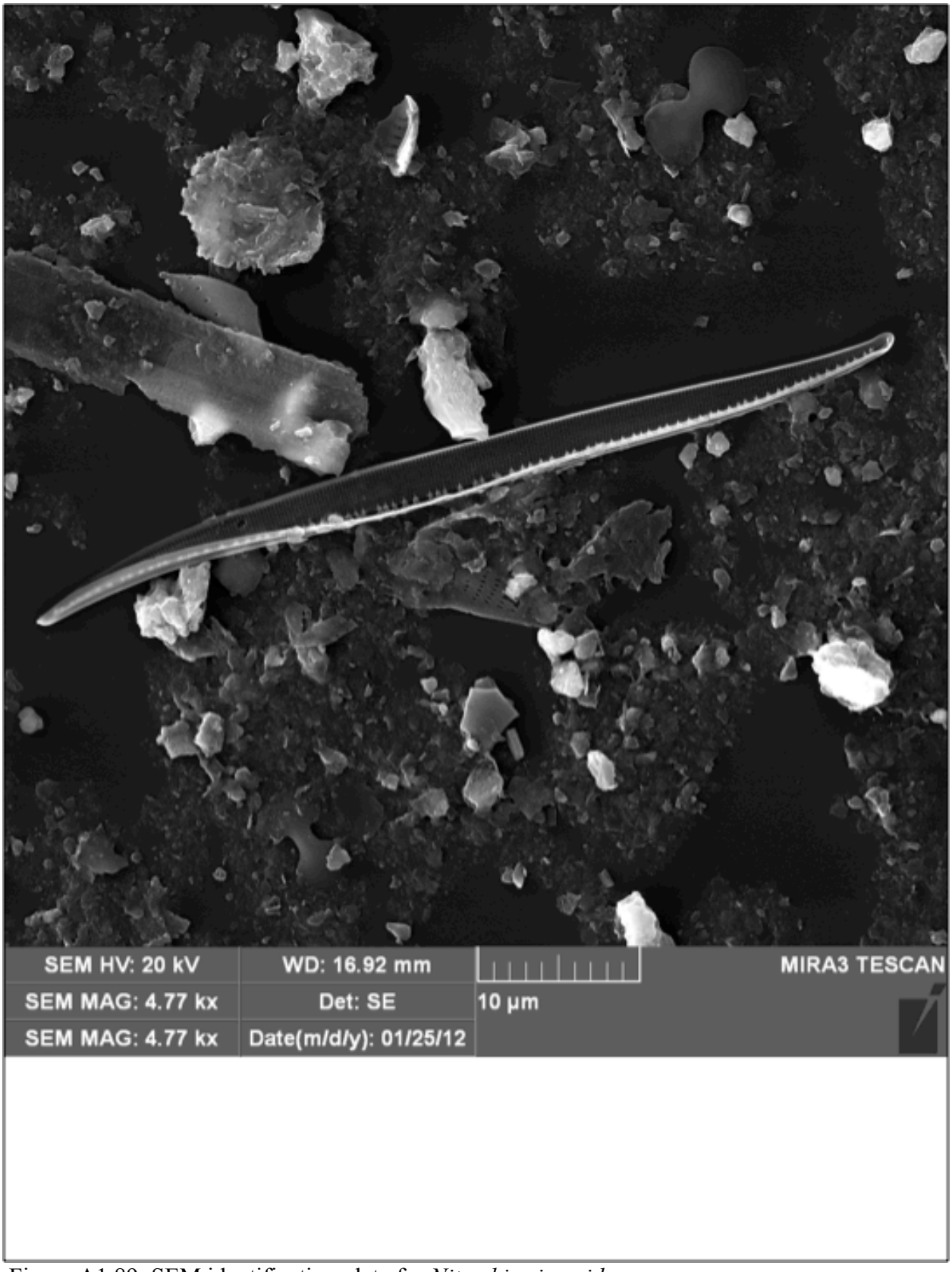


Figure A1.89: SEM identification plate for *Nitzschia sigmoidea*

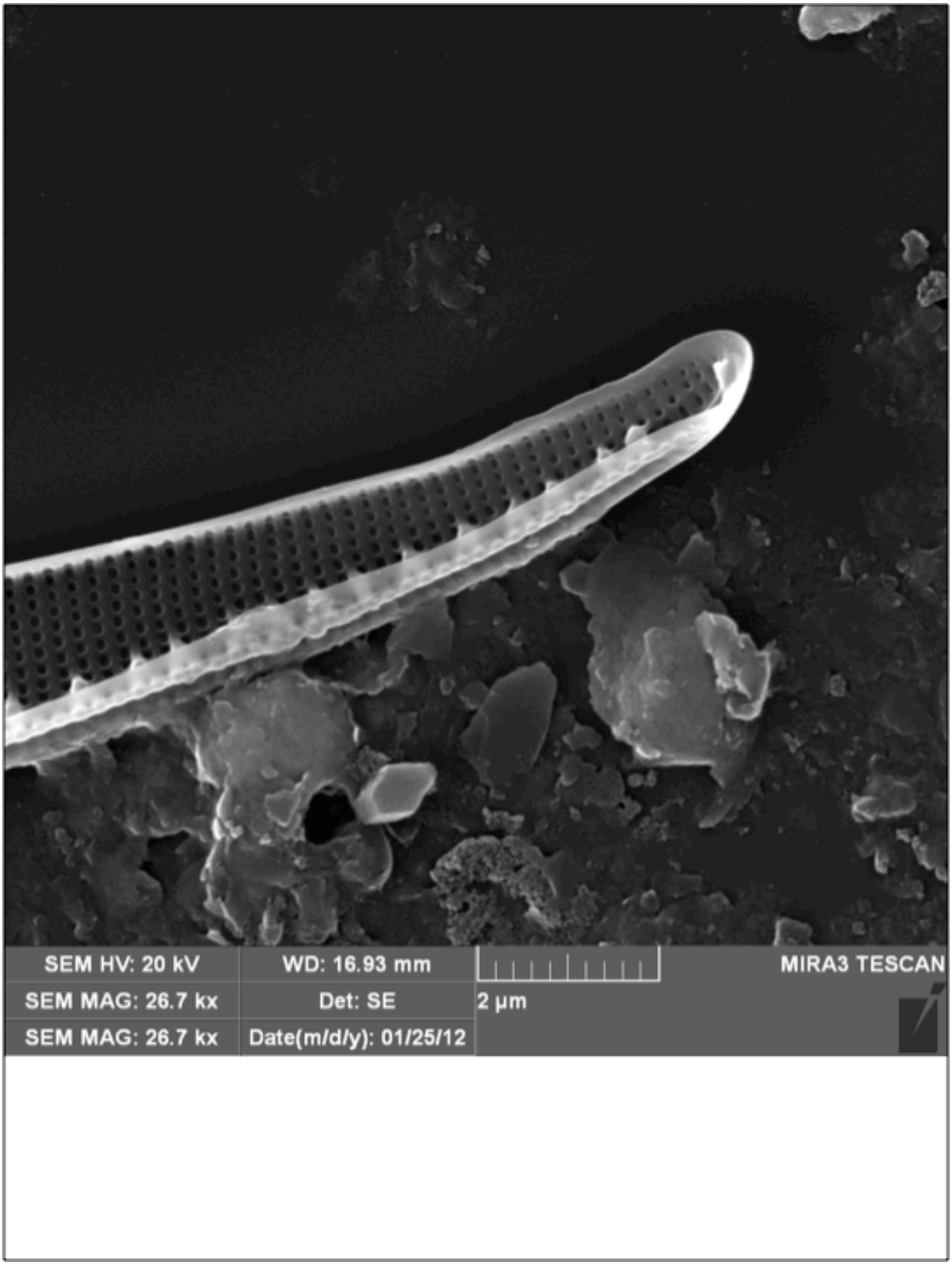


Figure A1.90: SEM identification plate for *Nitzschia sigmoidea*

Figure A1.91: SEM identification plate for *Nitzschia palea*

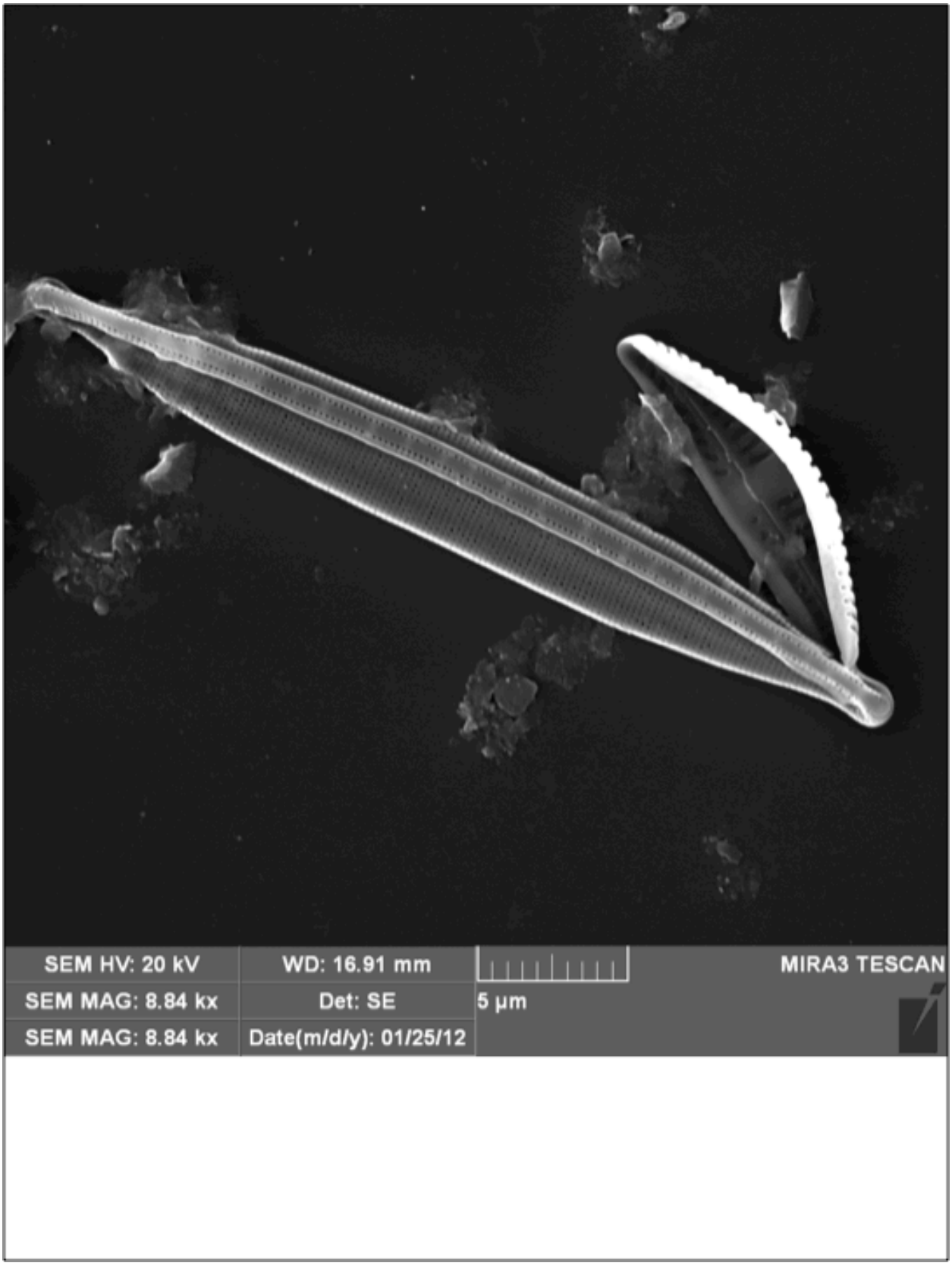


Figure A1.92: SEM identification plate for *Nitzschia dissipata*

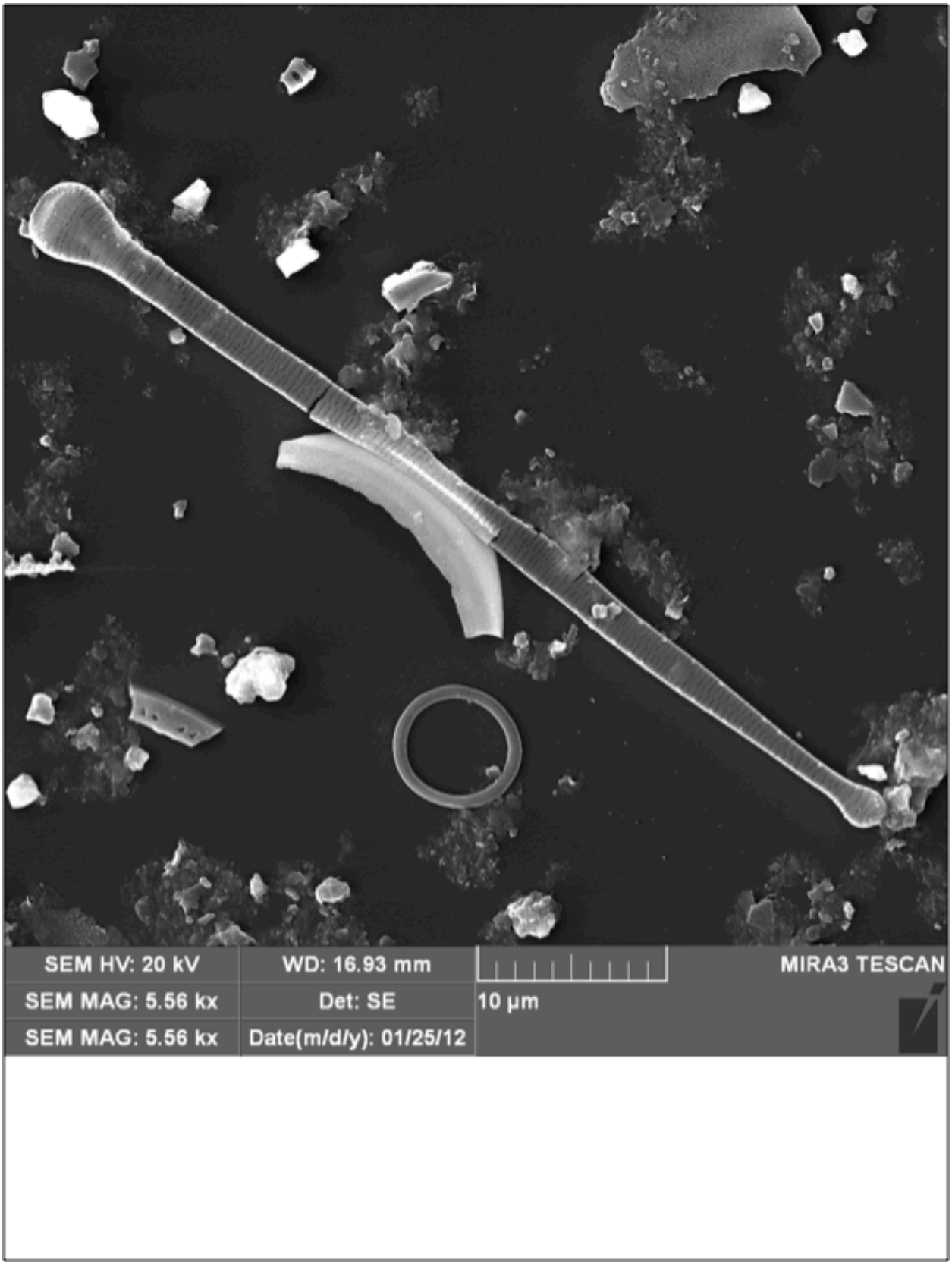


Figure A1.93: SEM identification plate for *Tabellaria flocculosa*

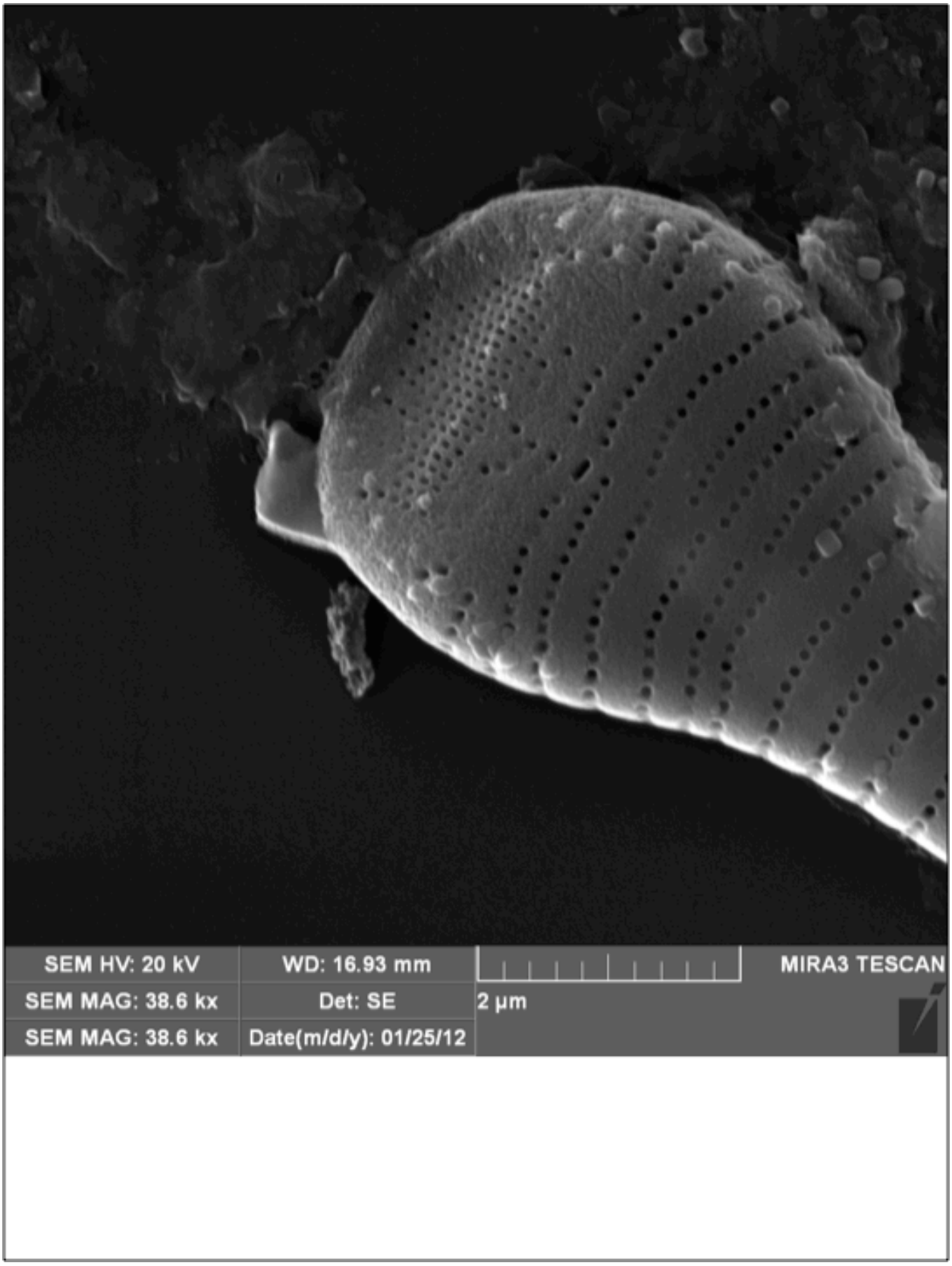


Figure A1.94: SEM identification plate for *Tabellaria flocculosa* (apical pore-field)

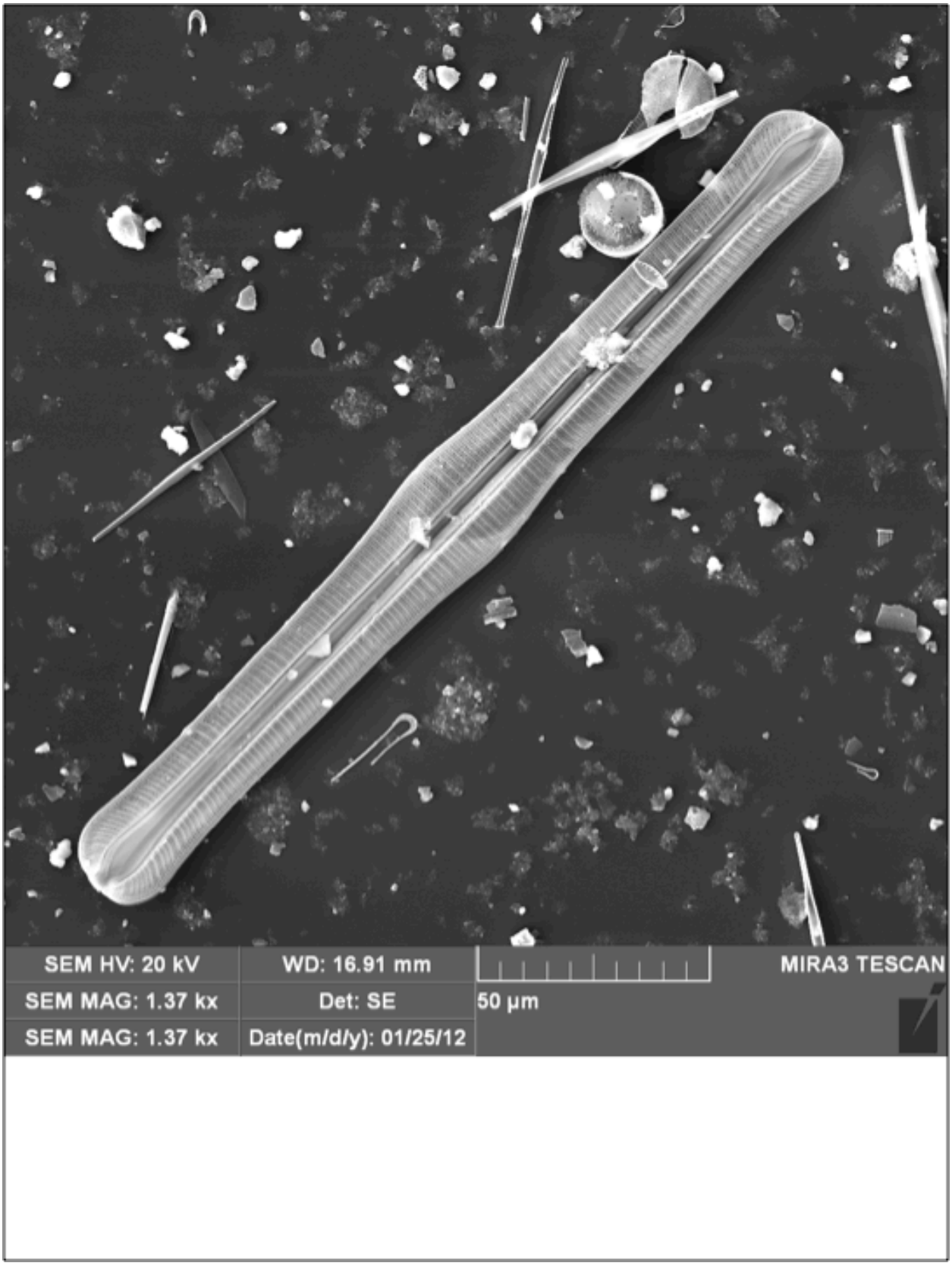


Figure A1.95: SEM identification plate for *Rhopalodia gibba*

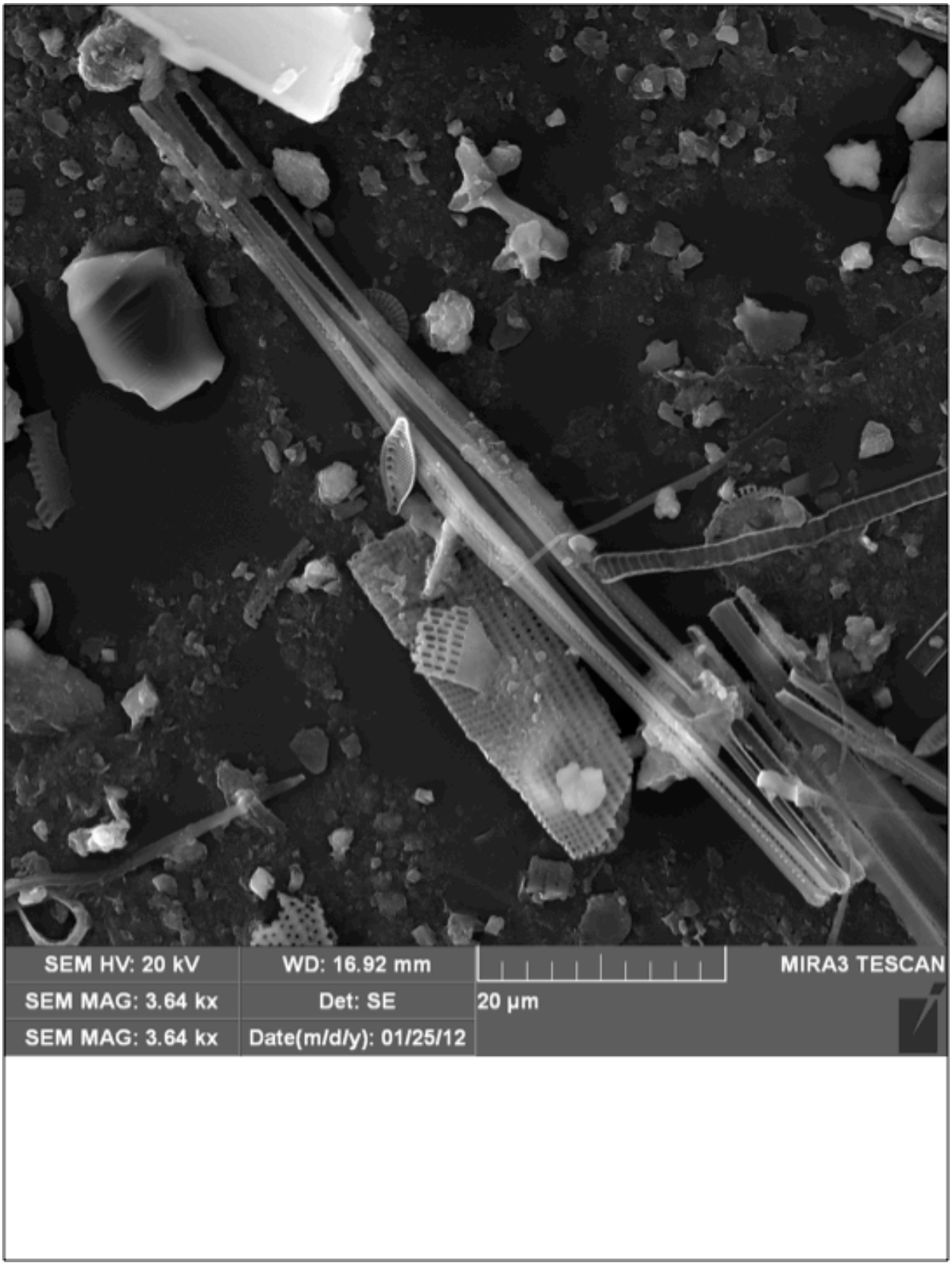


Figure A1.96: SEM identification plate for *Fragilaria crotonensis* (colony)

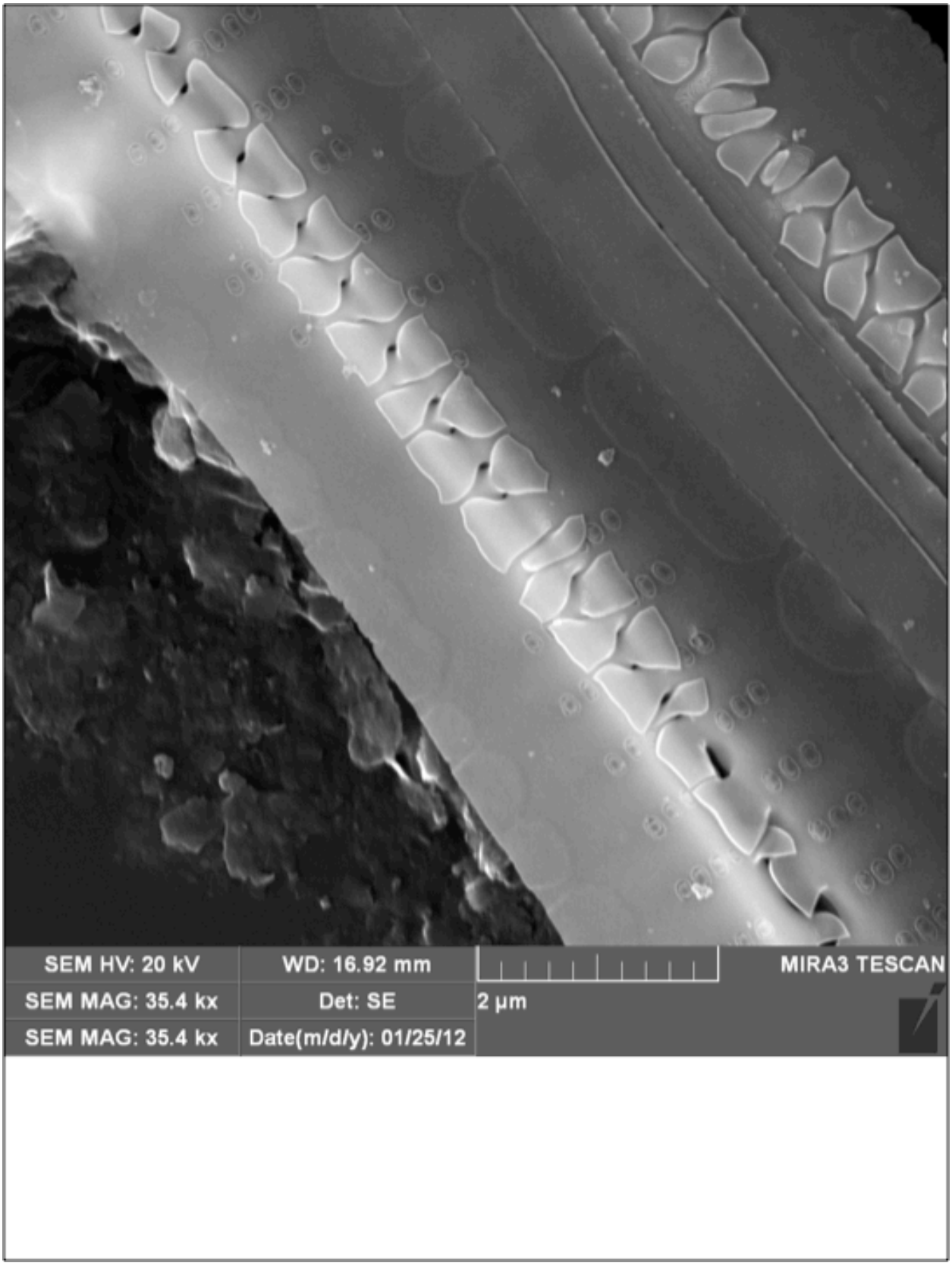


Figure A1.97: SEM identification plate for *Fragilaria crotonensis* (connection of colony)

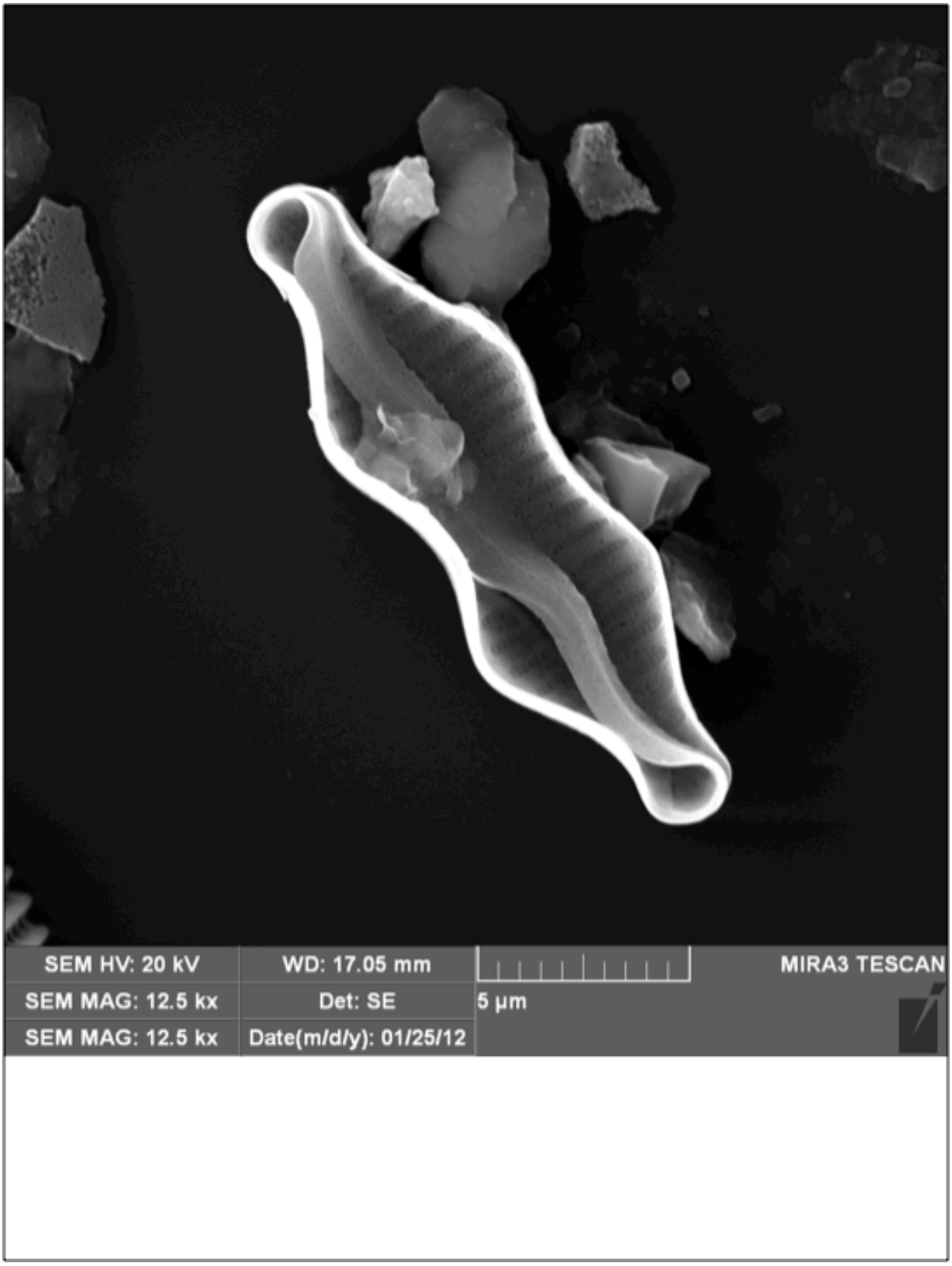


Figure A1.98 : SEM identification plate for *Fragilaria construens* var. *binodis*

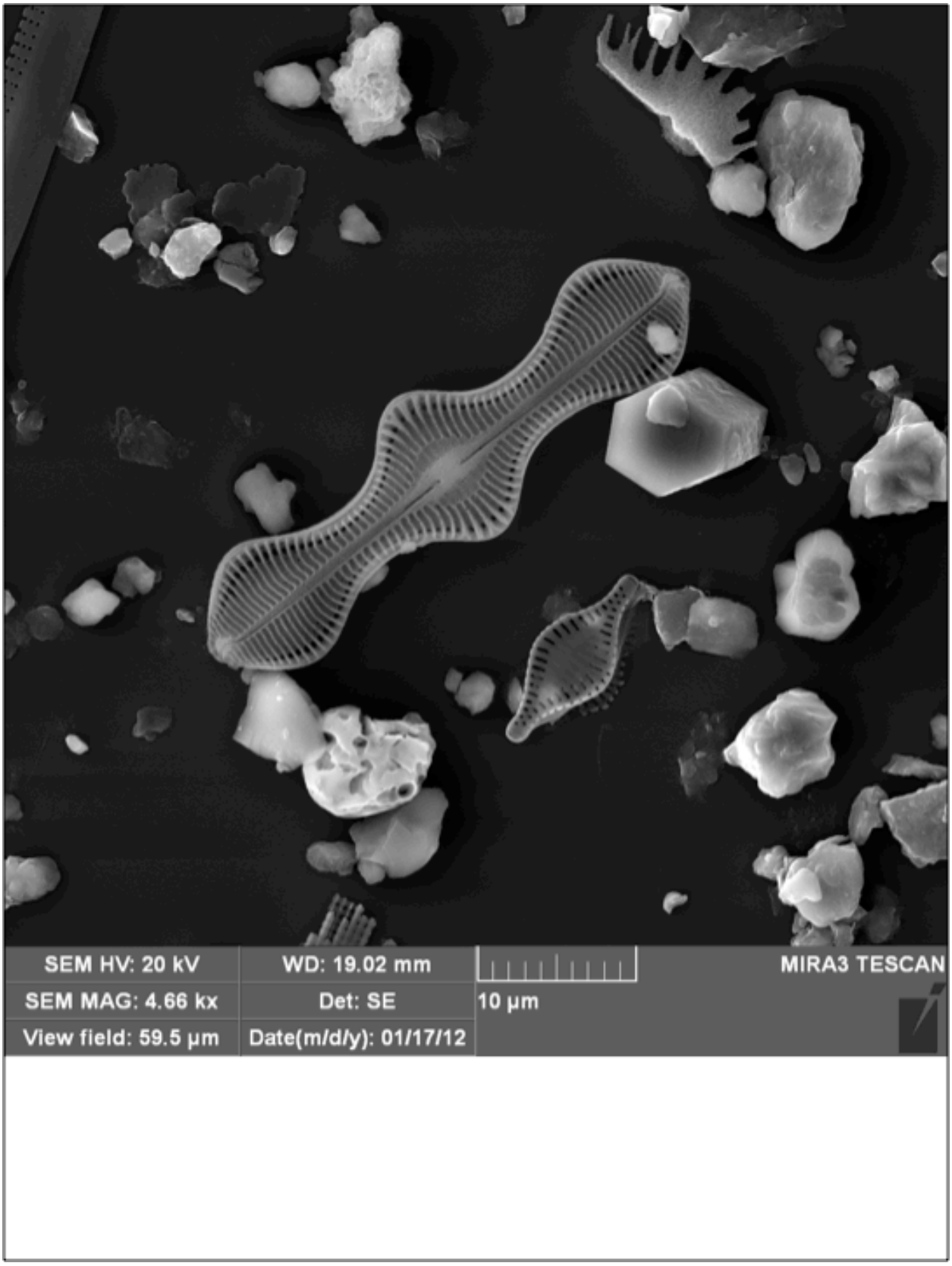


Figure A1.99: SEM identification plate for *Caloneis trochus* var. *trinoidis*

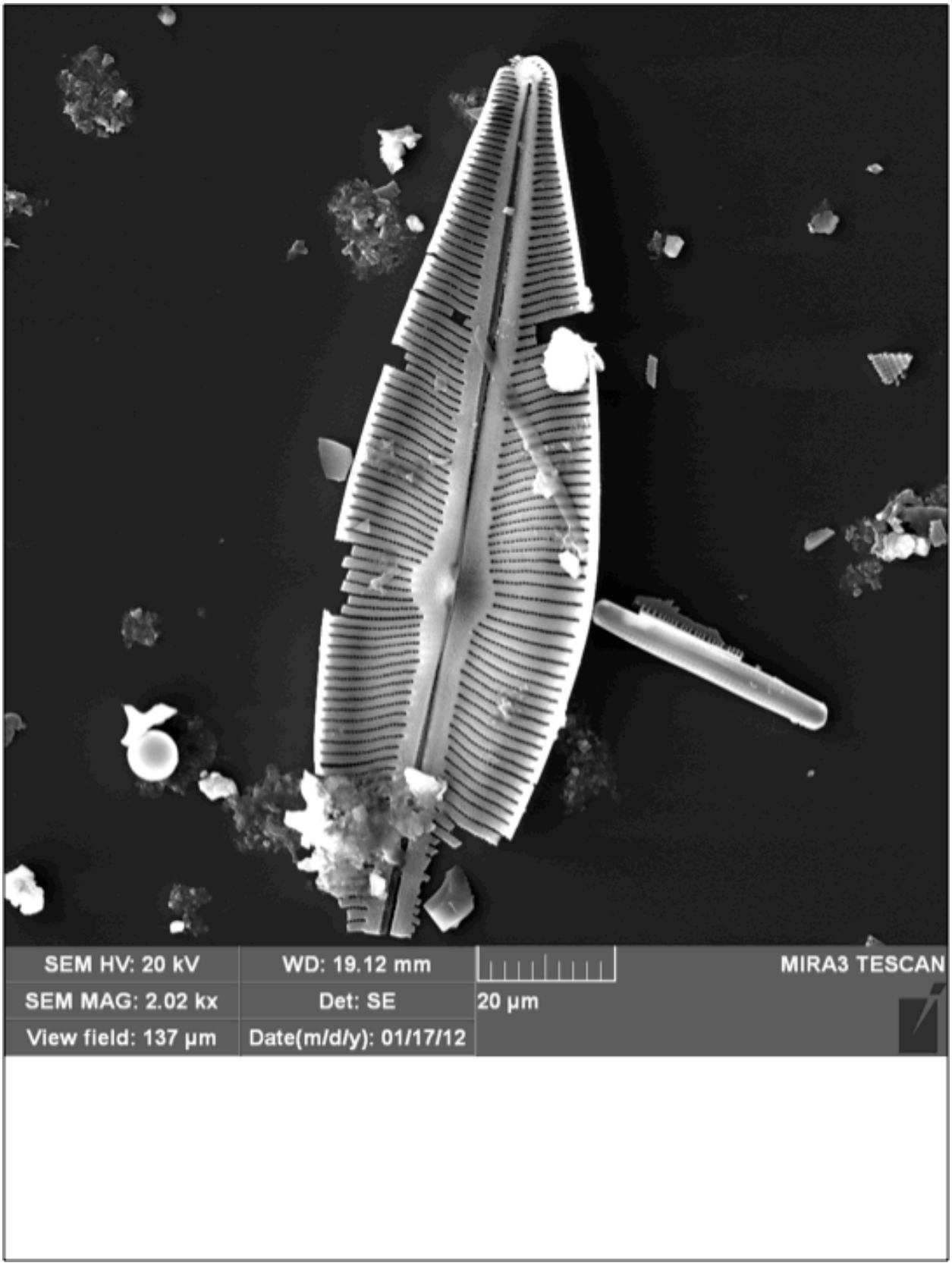


Figure A1.100: SEM identification plate for *Cymbella ehrenbergii*

Figure A1.101: SEM identification plate for *Cymbella neocistula*

Figure A1.102: SEM identification plate for *Cymbella muelleri*

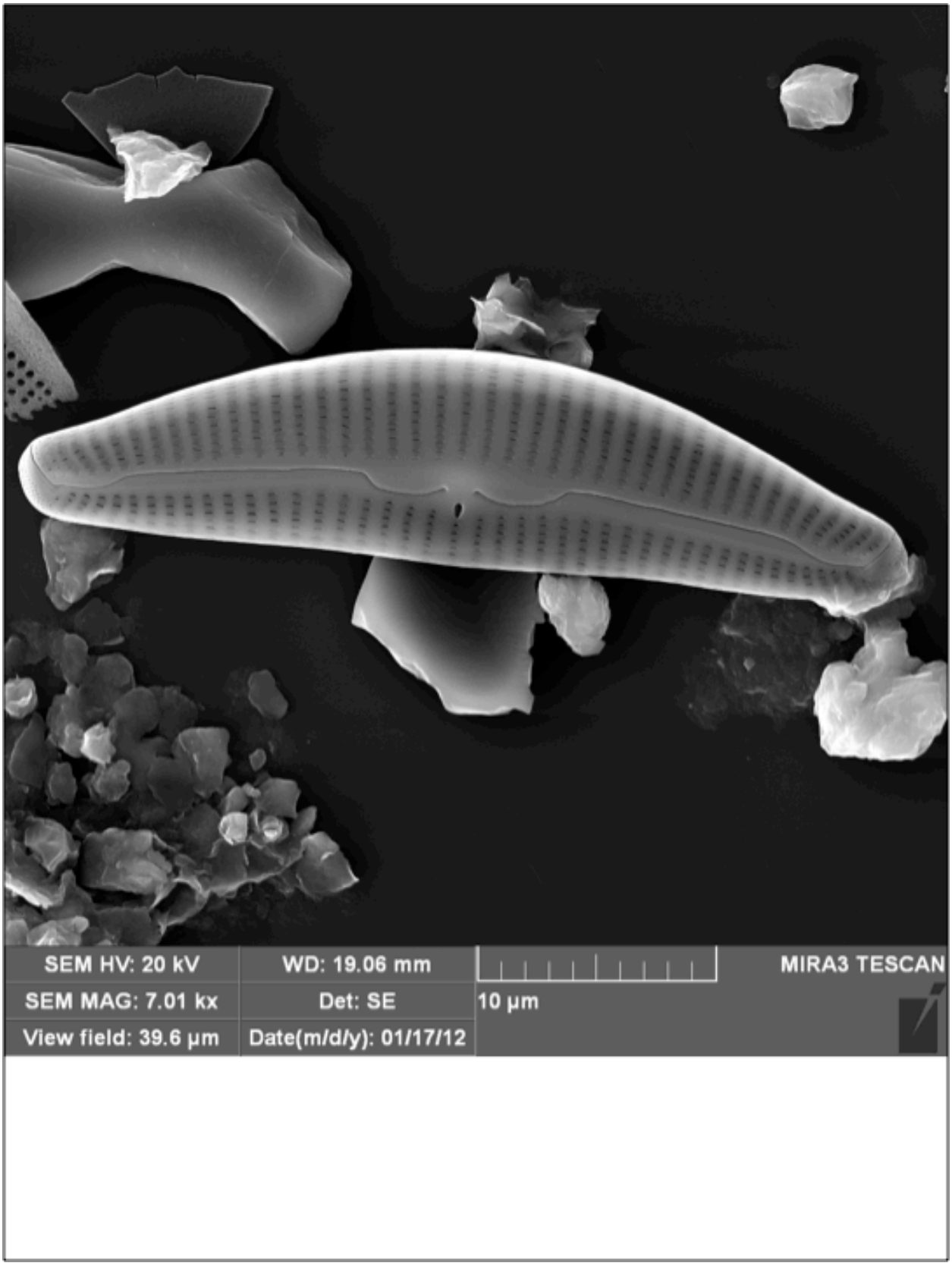


Figure A1.103: SEM identification plate for *Cymbella neocistula*

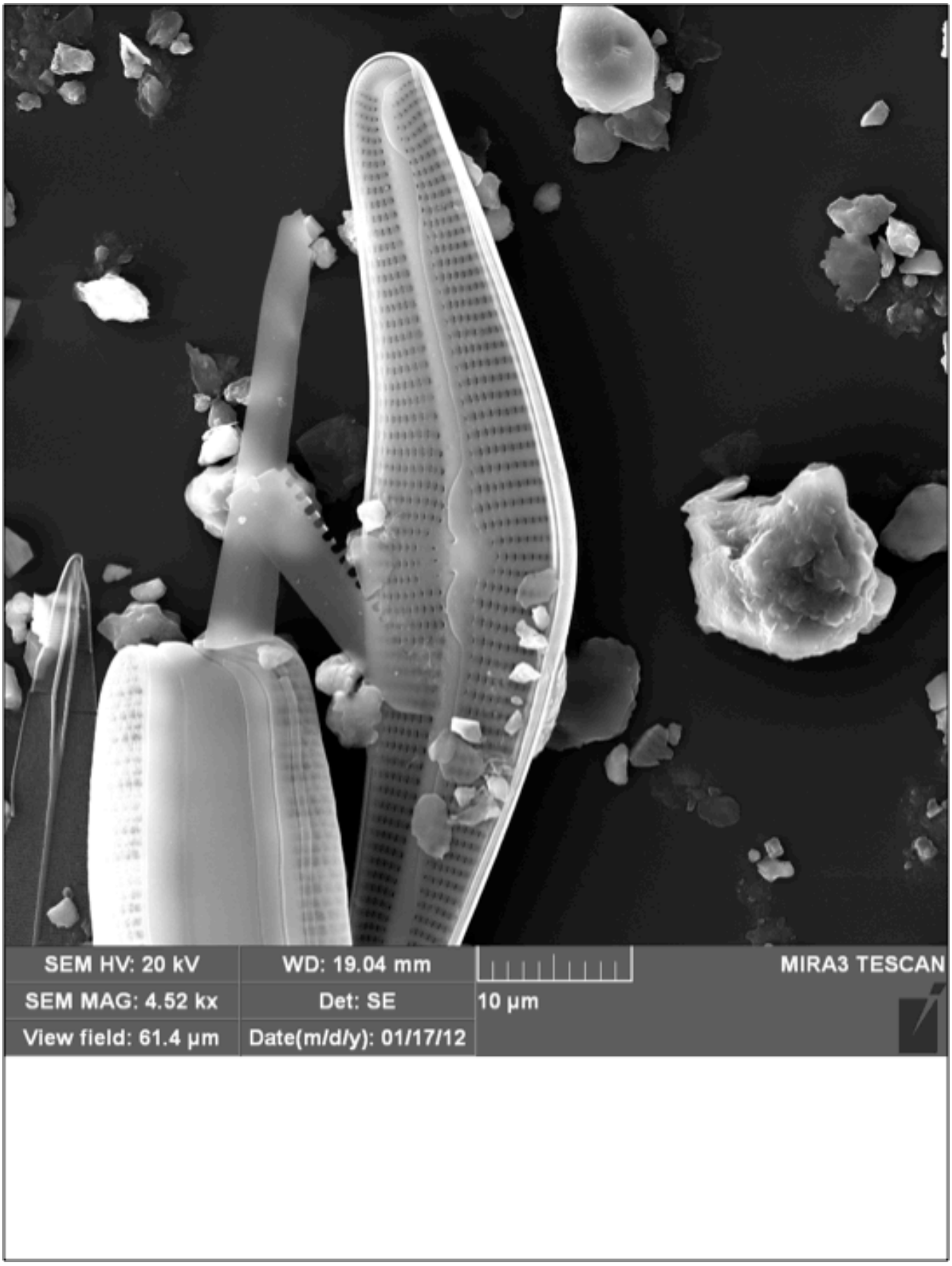


Figure A1.104: SEM identification plate for *Cymbella subcistula*

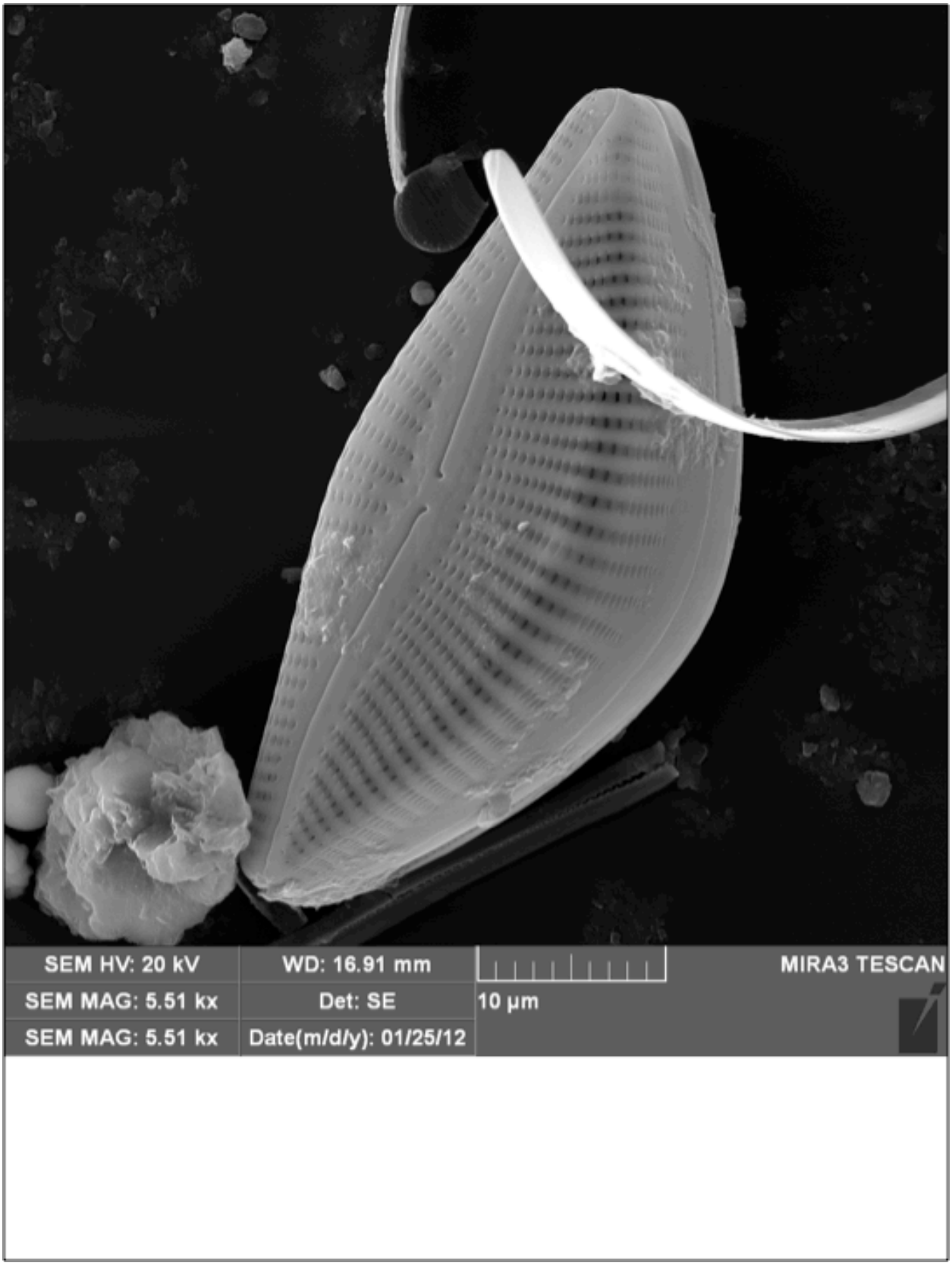


Figure A1.105: SEM identification plate for *Cymbella muelleri* var. *ventricosa*

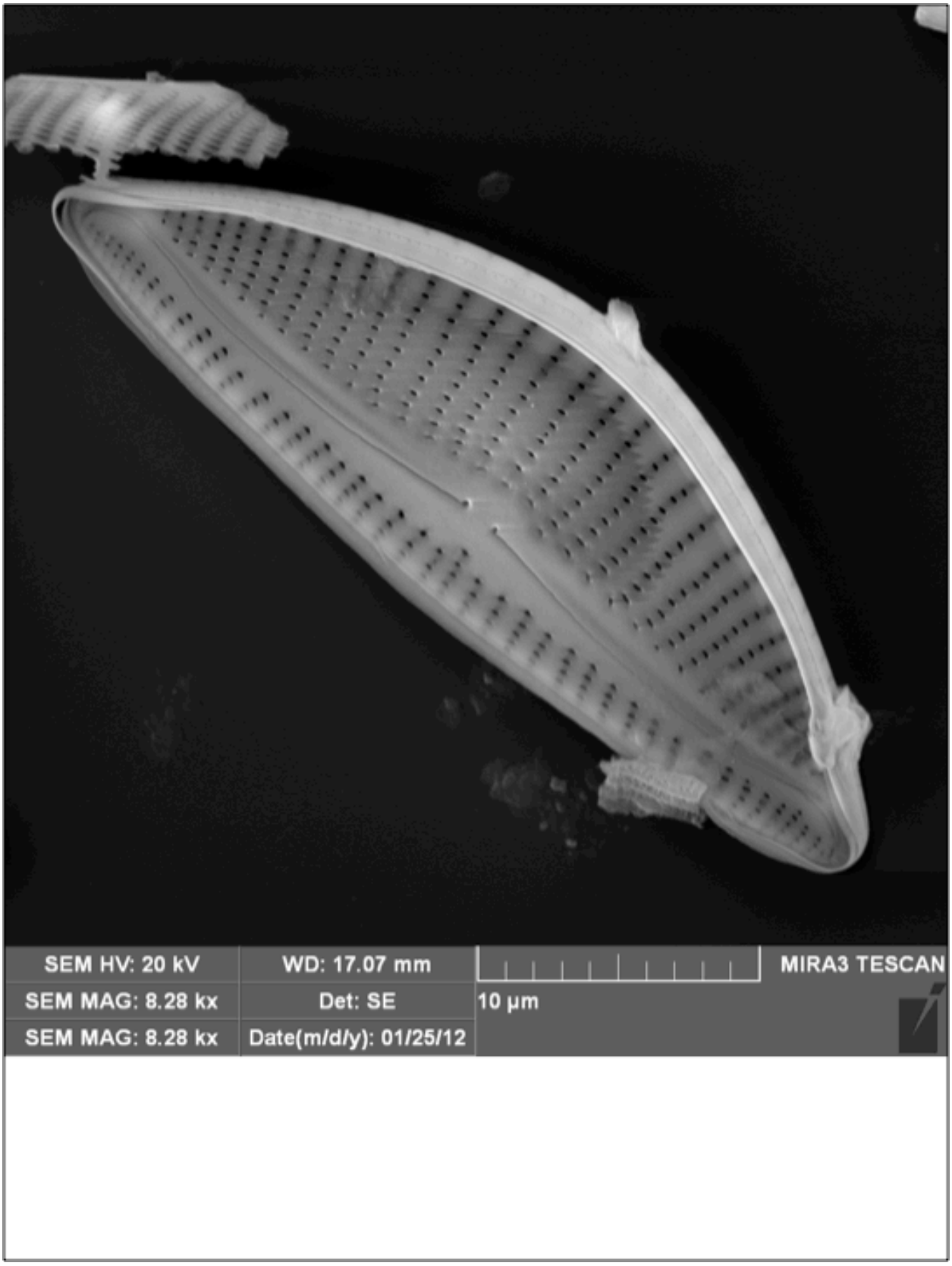


Figure A1.106: SEM identification plate for *Cymbella muelleri* var. *ventricosa*

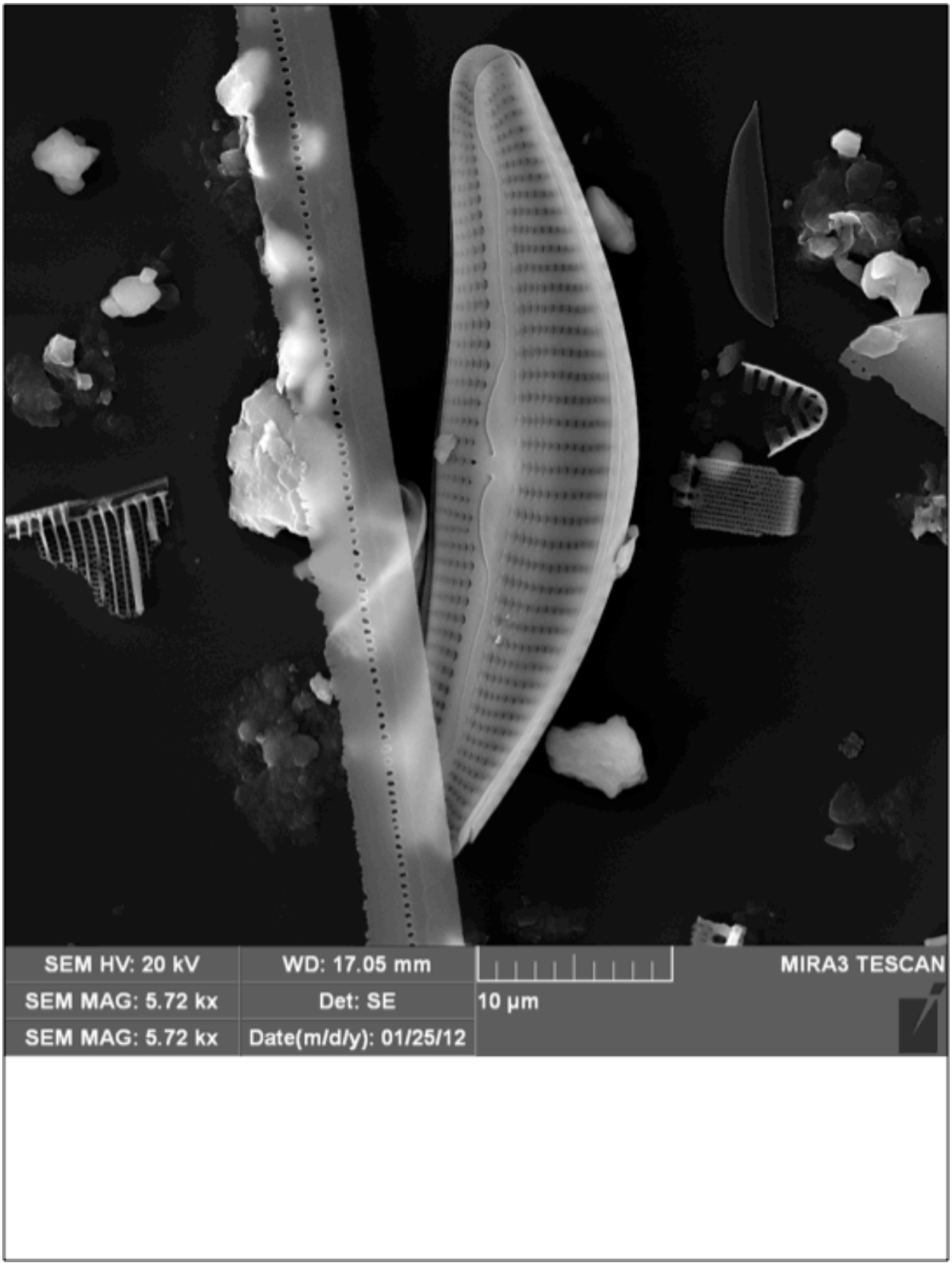


Figure A1.107: SEM identification plate for *Cymbella neocistula*

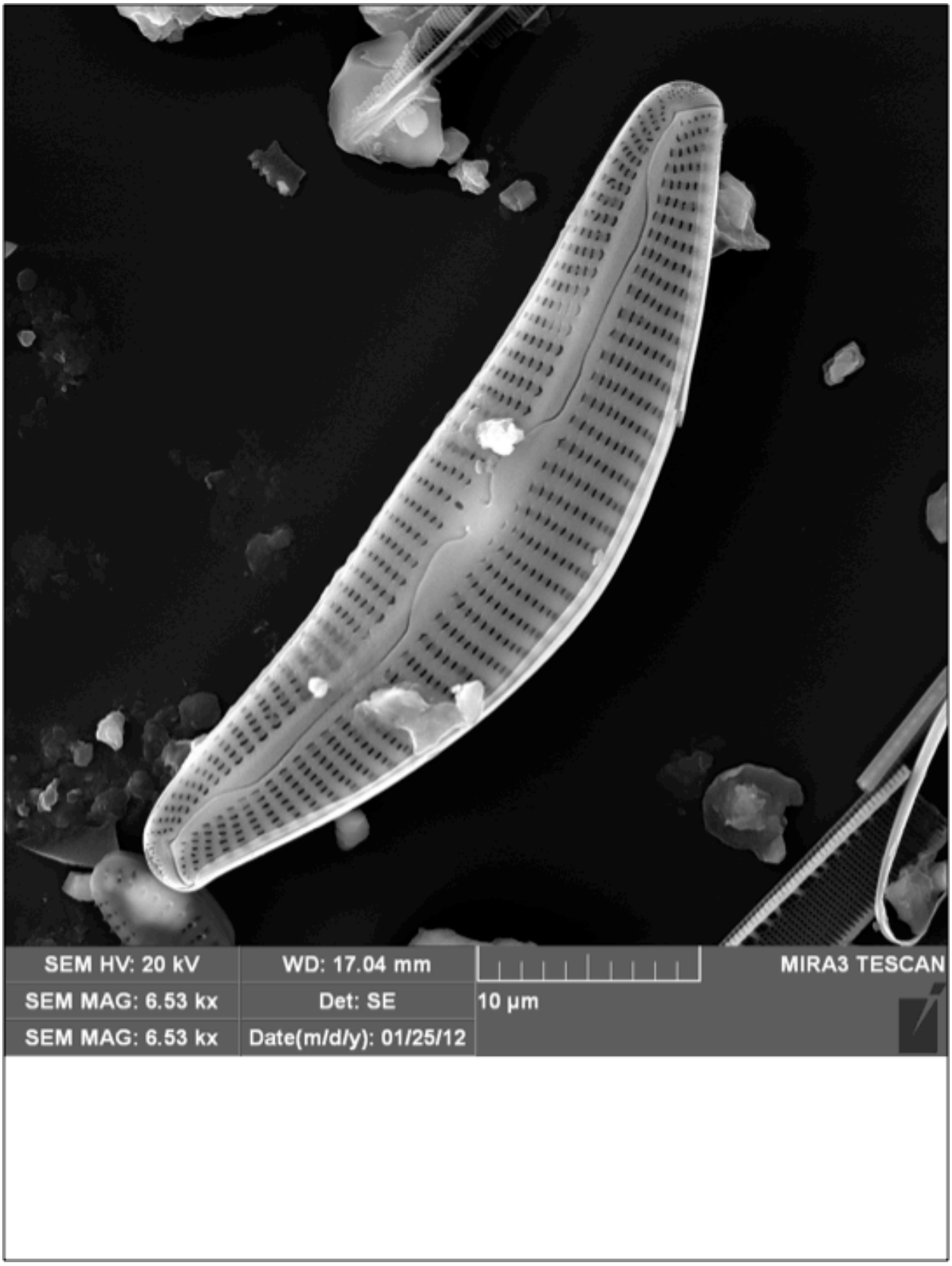


Figure A1.108: SEM identification plate for *Cymbella neocistula*

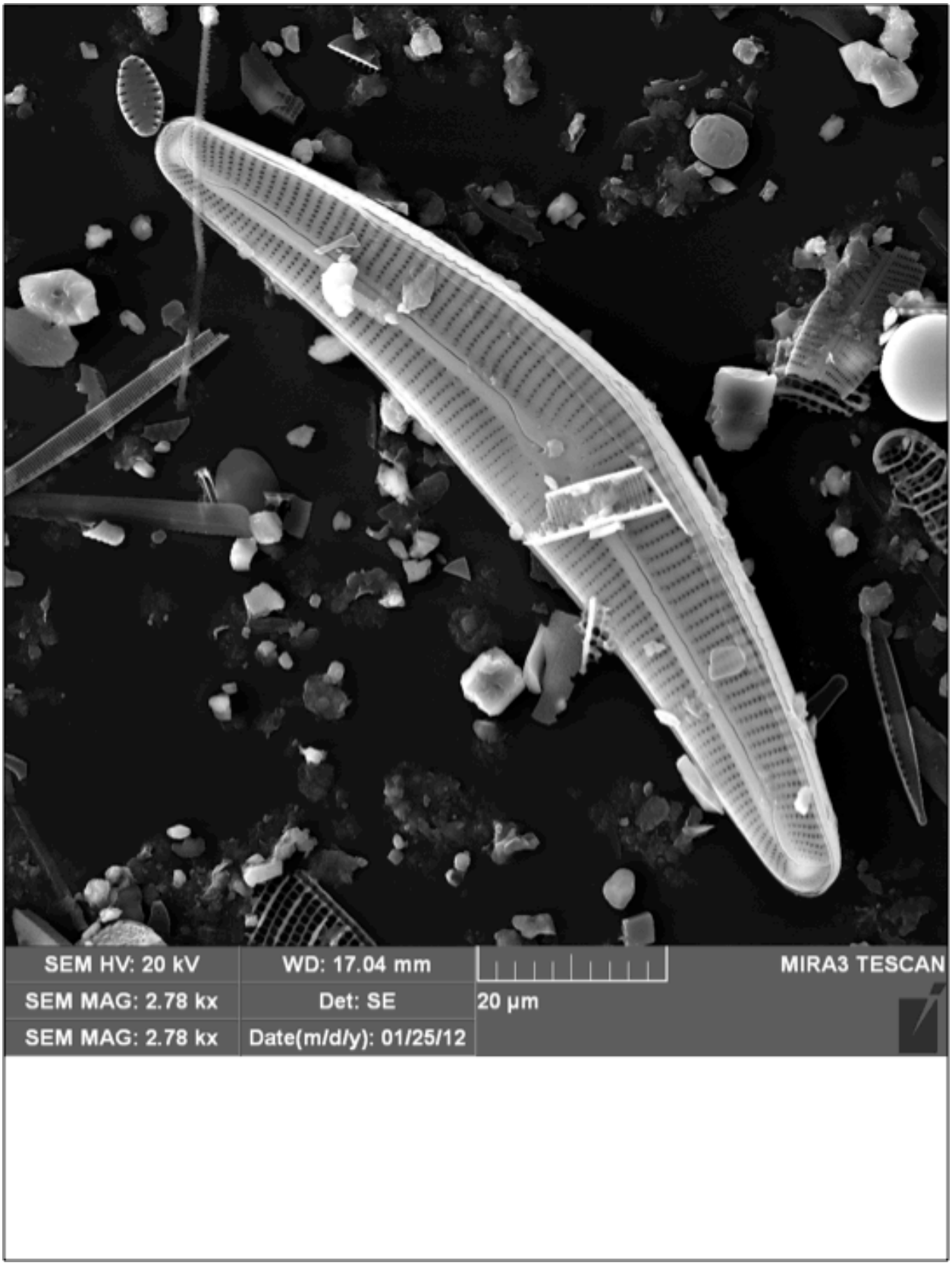


Figure A1.109: SEM identification plate for *Cymbella subcistula*

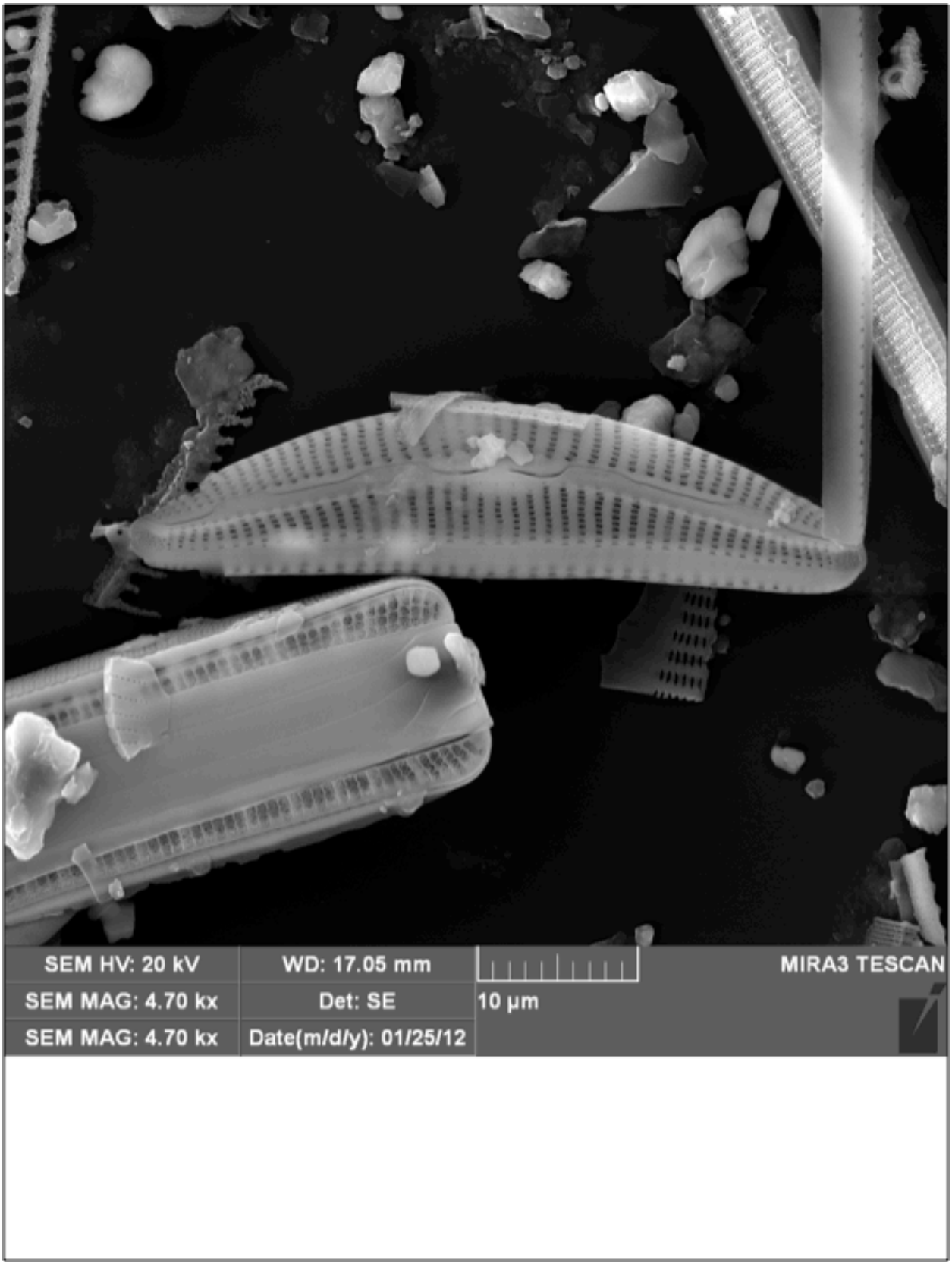


Figure A1.110: SEM identification plate for *Cymbella muelleri*

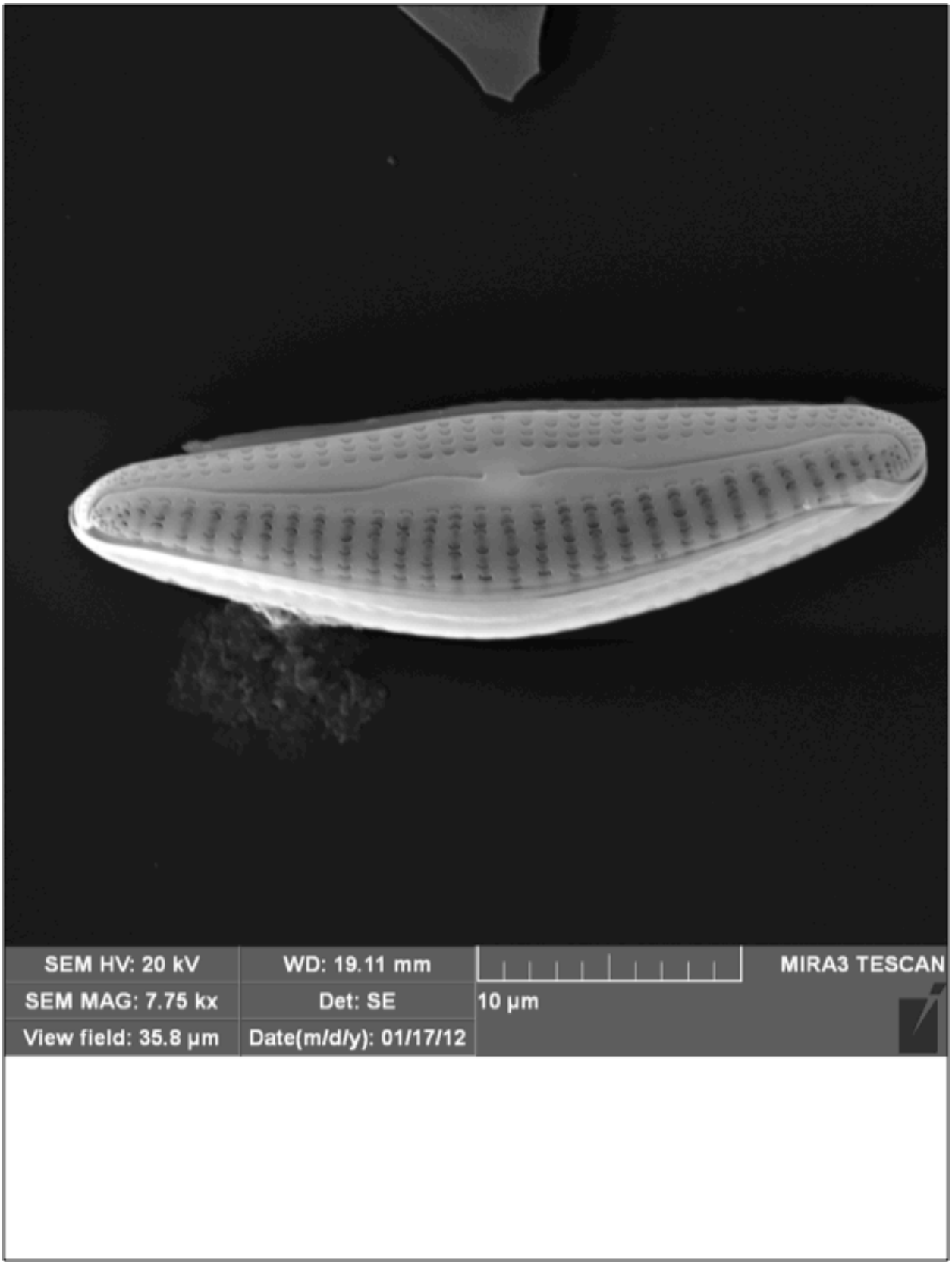


Figure A1.111: SEM identification plate for *Cymbella minuta*

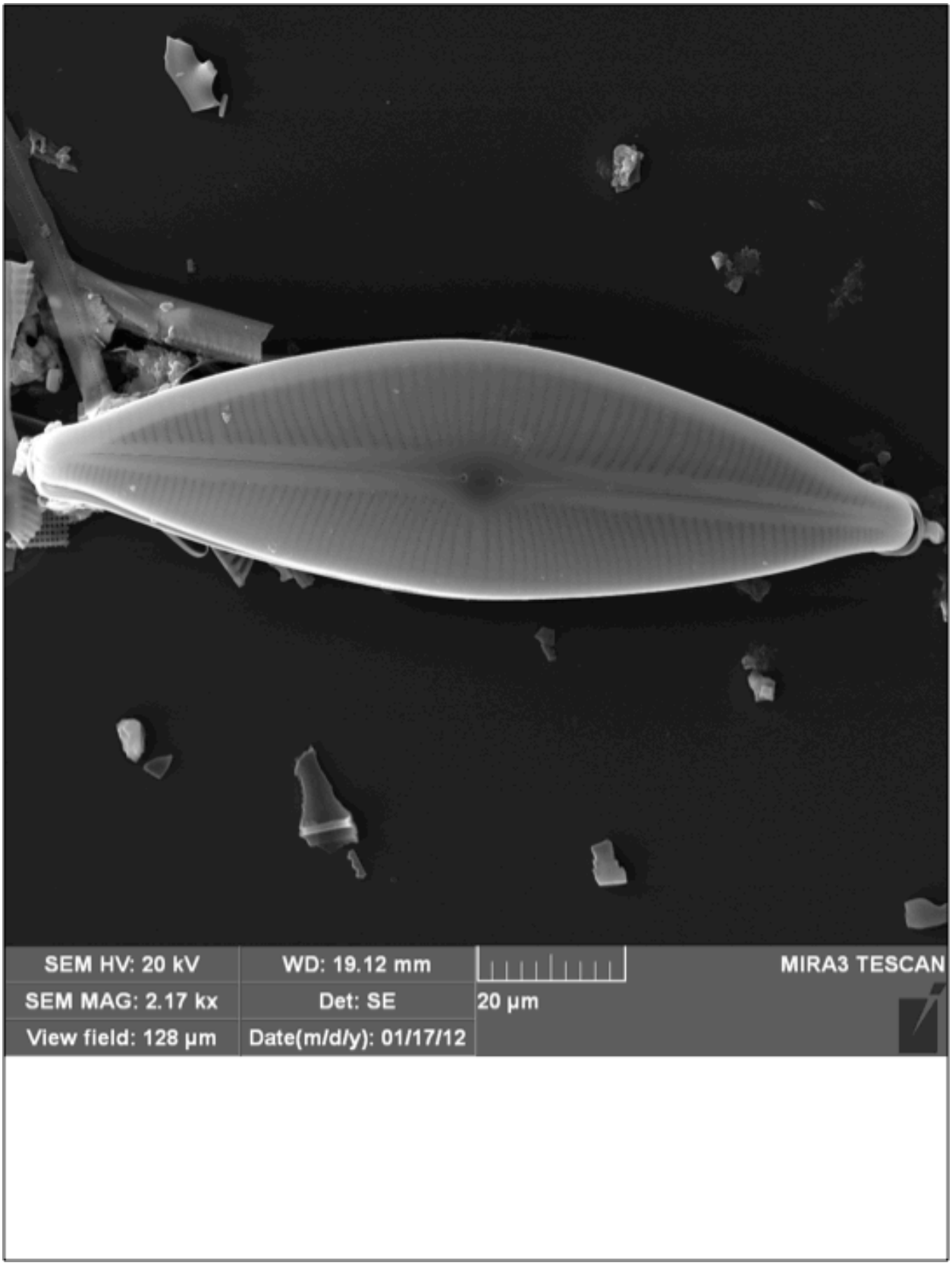


Figure A1.112: SEM identification plate for *Cymbella ehrenbergii*

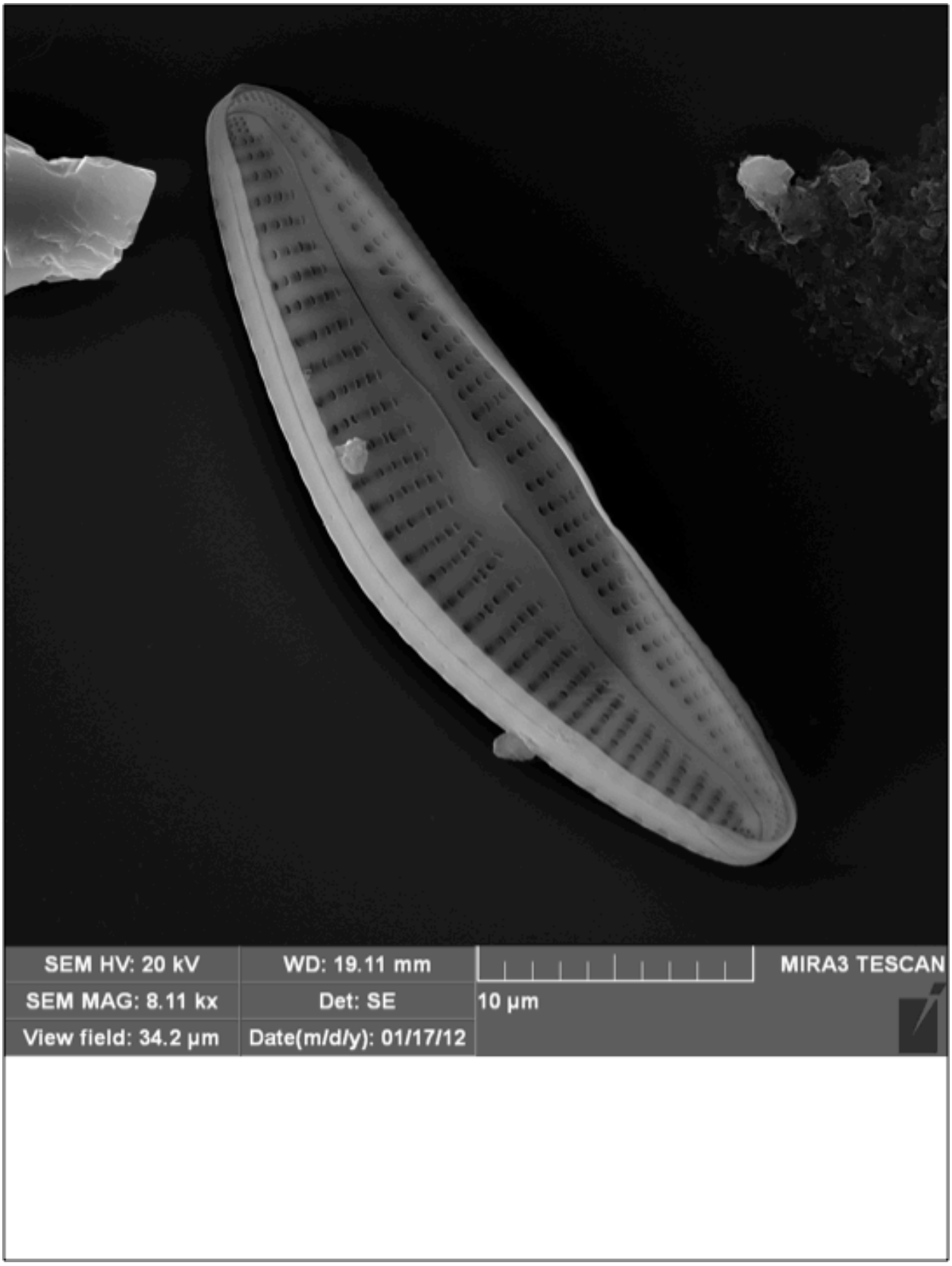


Figure A1.113: SEM identification plate for *Cymbopleura hustedtii*

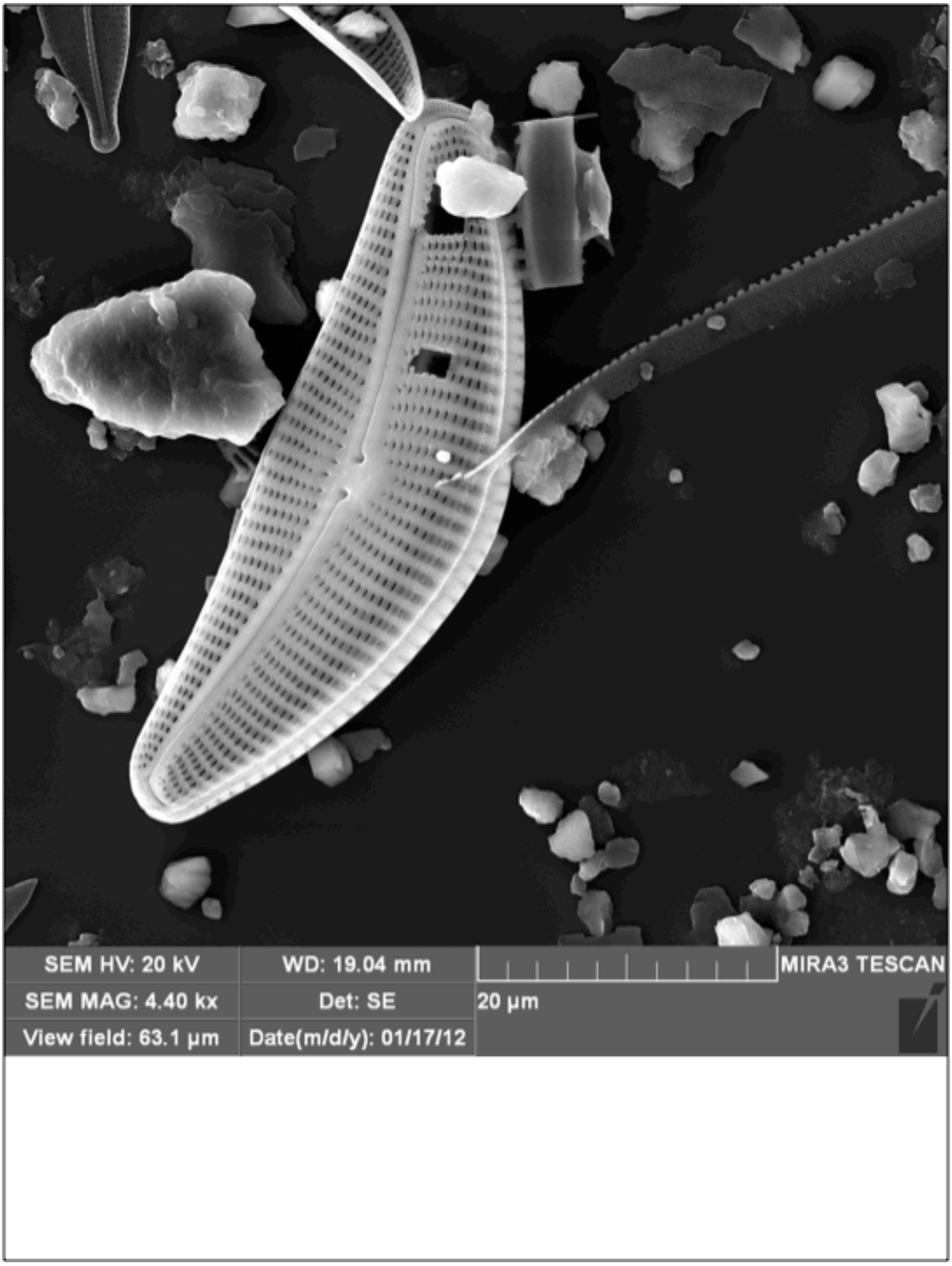


Figure A1.114: SEM identification plate for *Cymbella neocistula*

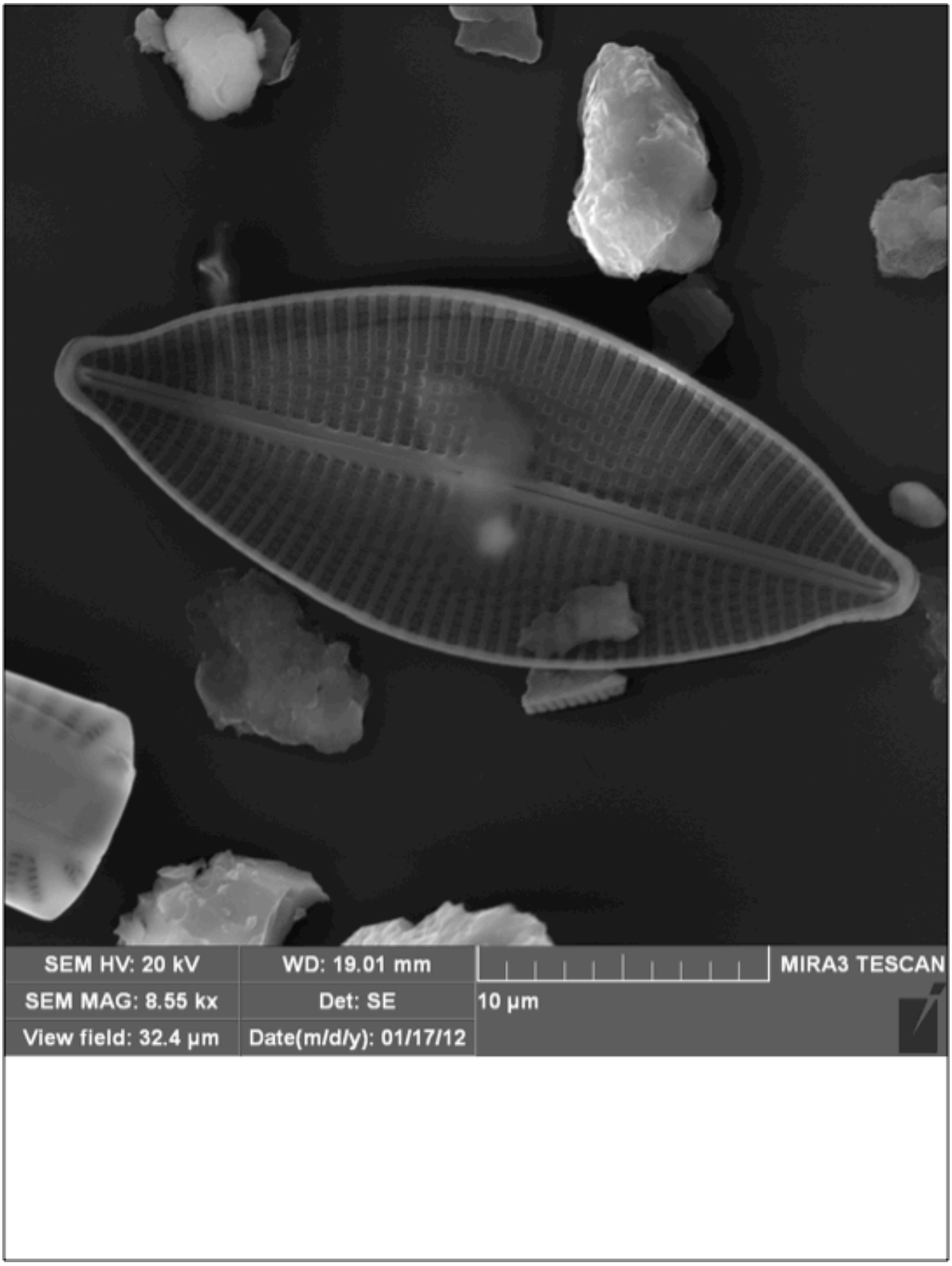


Figure A1.115: SEM identification plate for *Navicula tuscula*

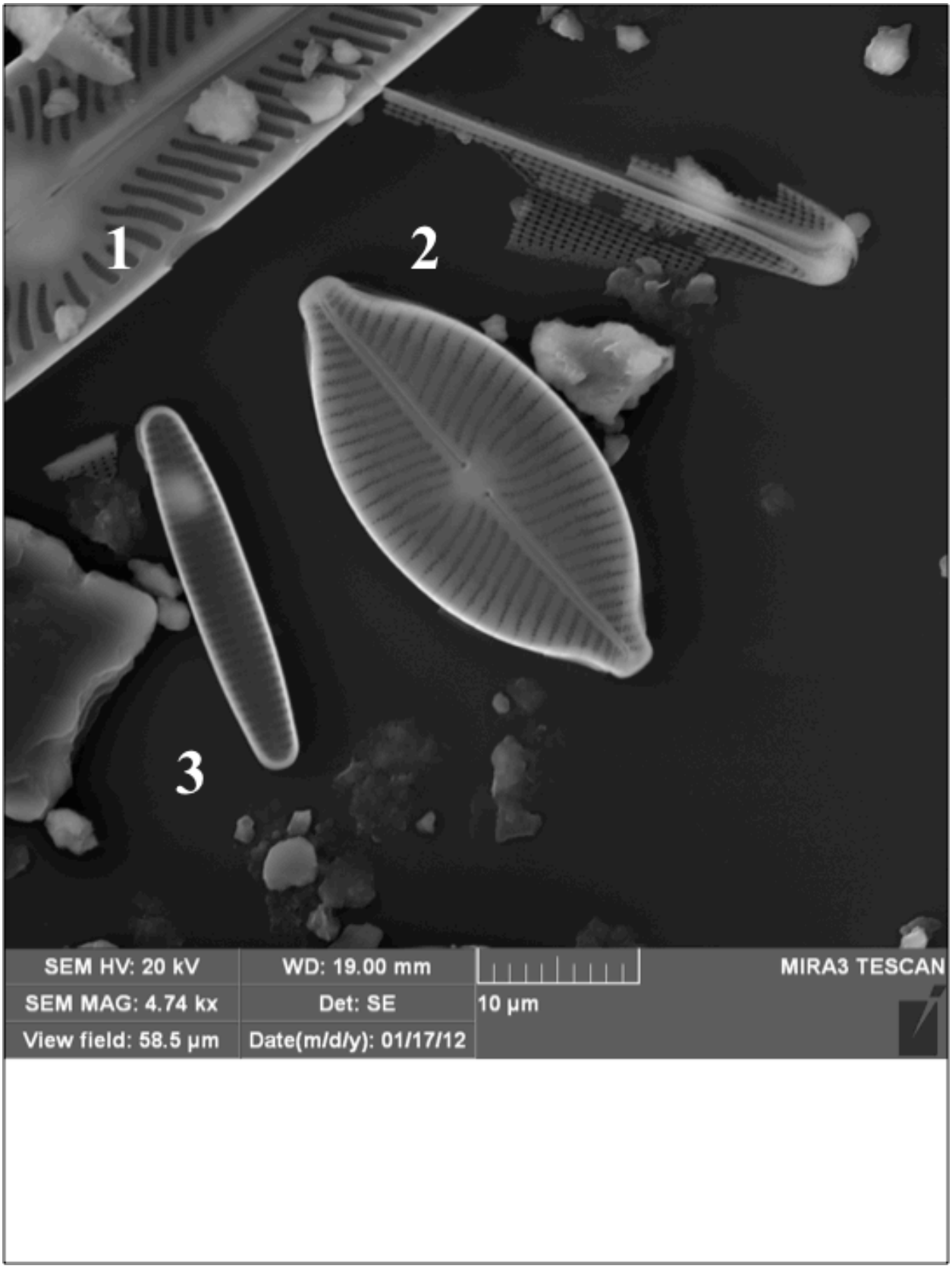


Figure A1.116: SEM identification plate for: *Navicula oblonga, Navicula gastrum,* and *Fragilaria brevistriata*
1 – *Navicula oblonga,* 2 – *Navicula gastrum,* 3 – *Fragilaria brevistriata*

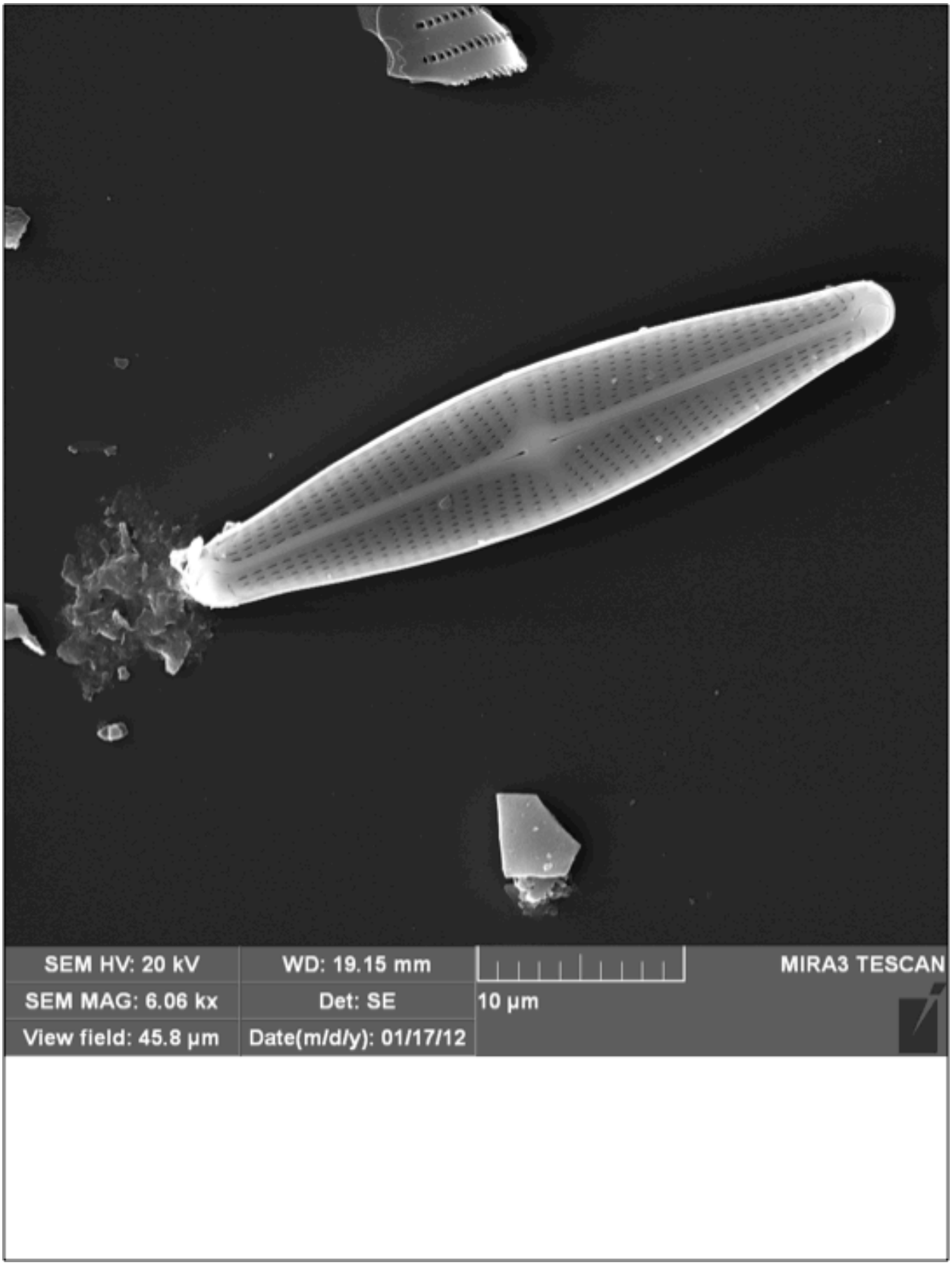


Figure A1.117: SEM identification plate for: *Navicula veneta*

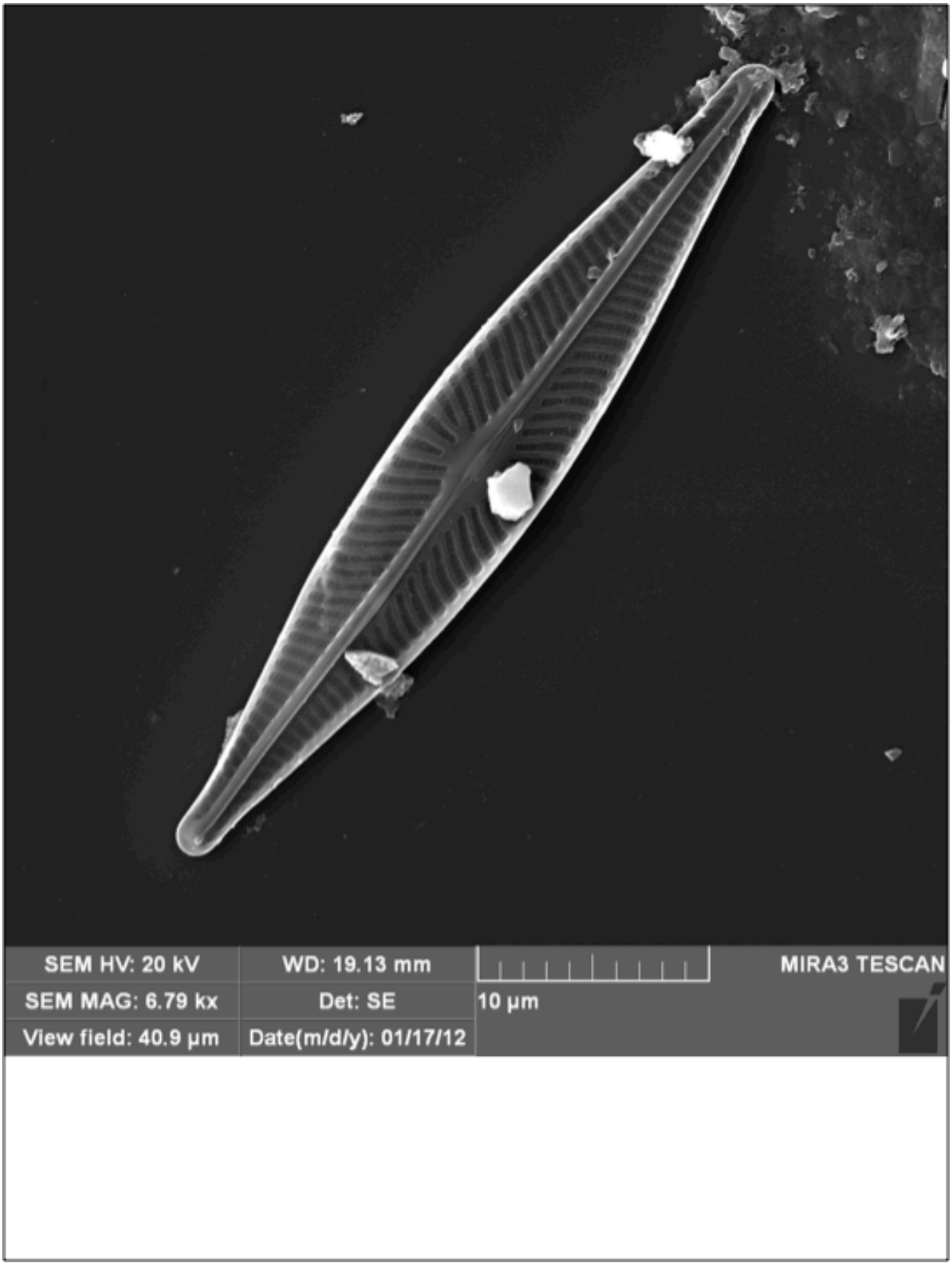


Figure A1.118: SEM identification plate for: *Navicula radiosa*

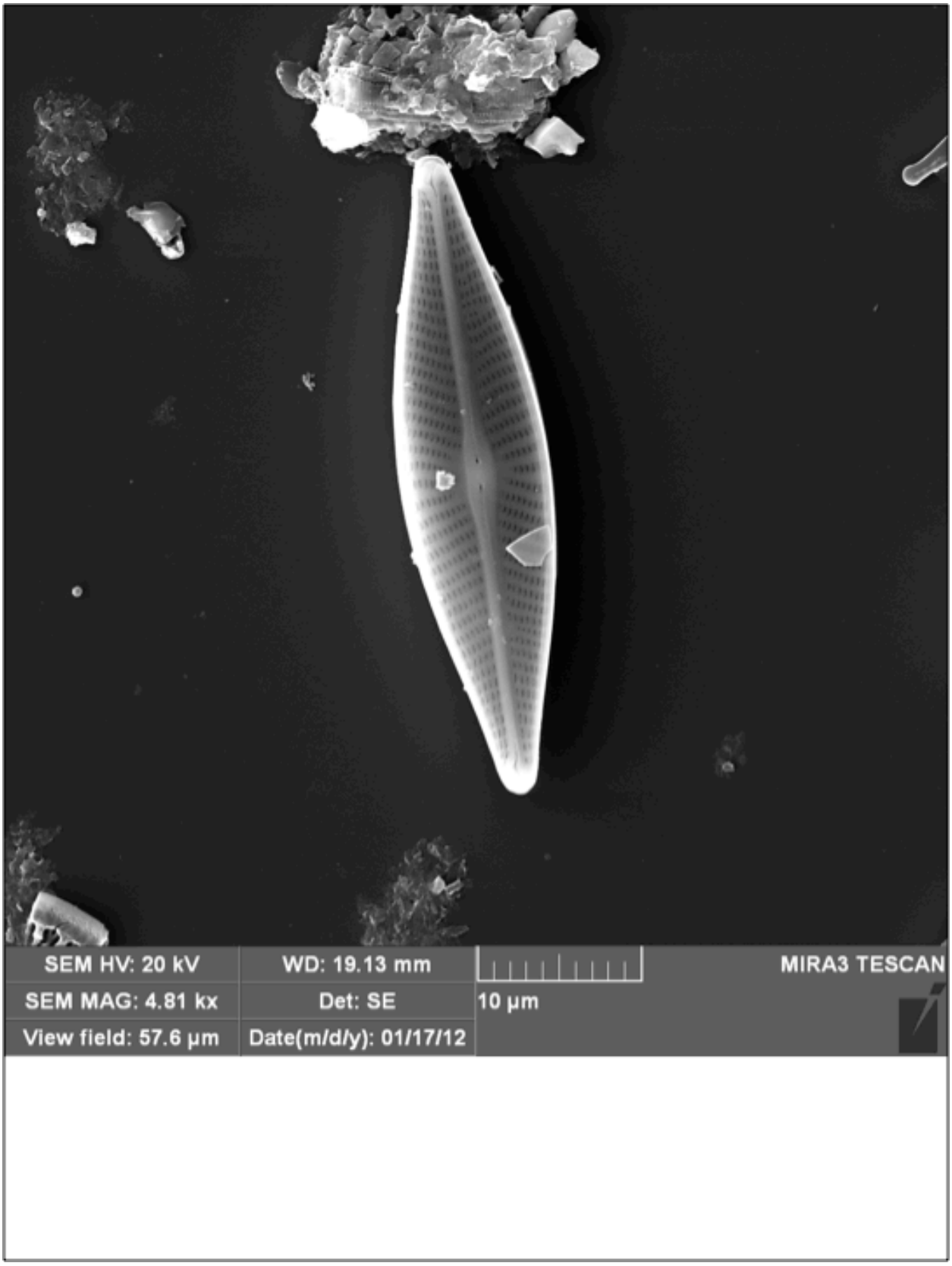


Figure A1.119: SEM identification plate for: *Navicula rhynchocephala*

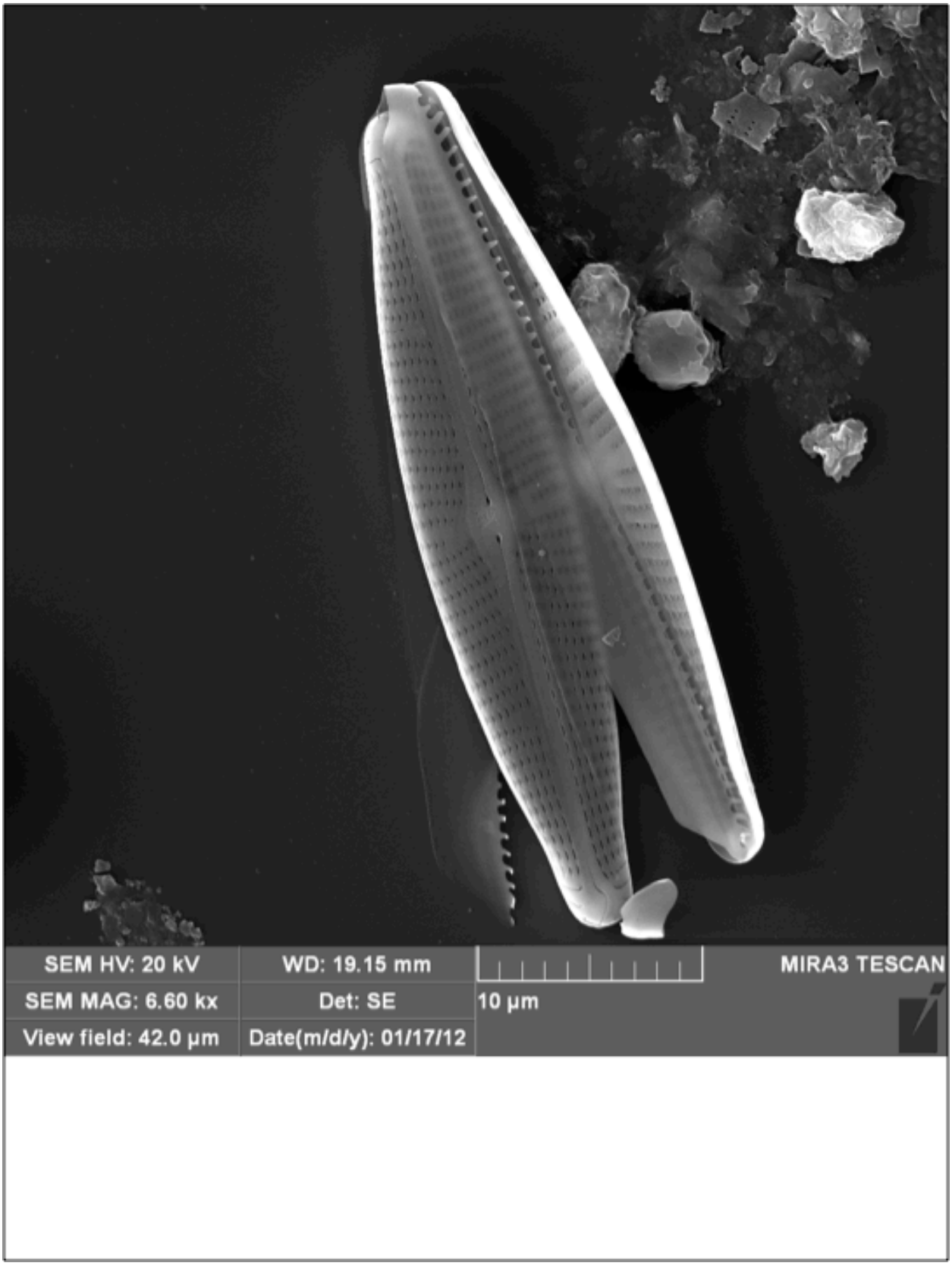


Figure A1.120: SEM identification plate for: *Navicula veneta*

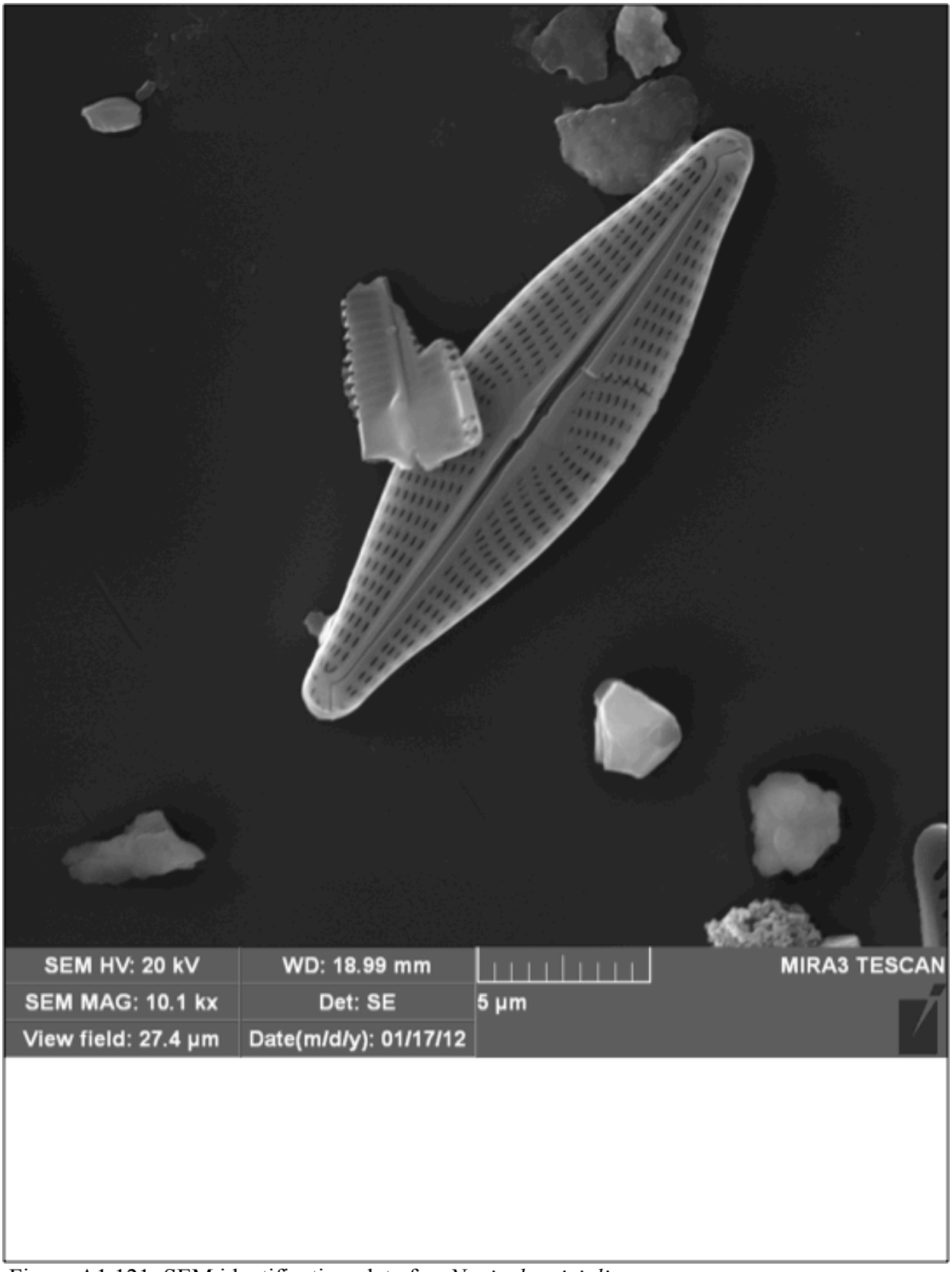


Figure A1.121: SEM identification plate for: *Navicula trivialis*

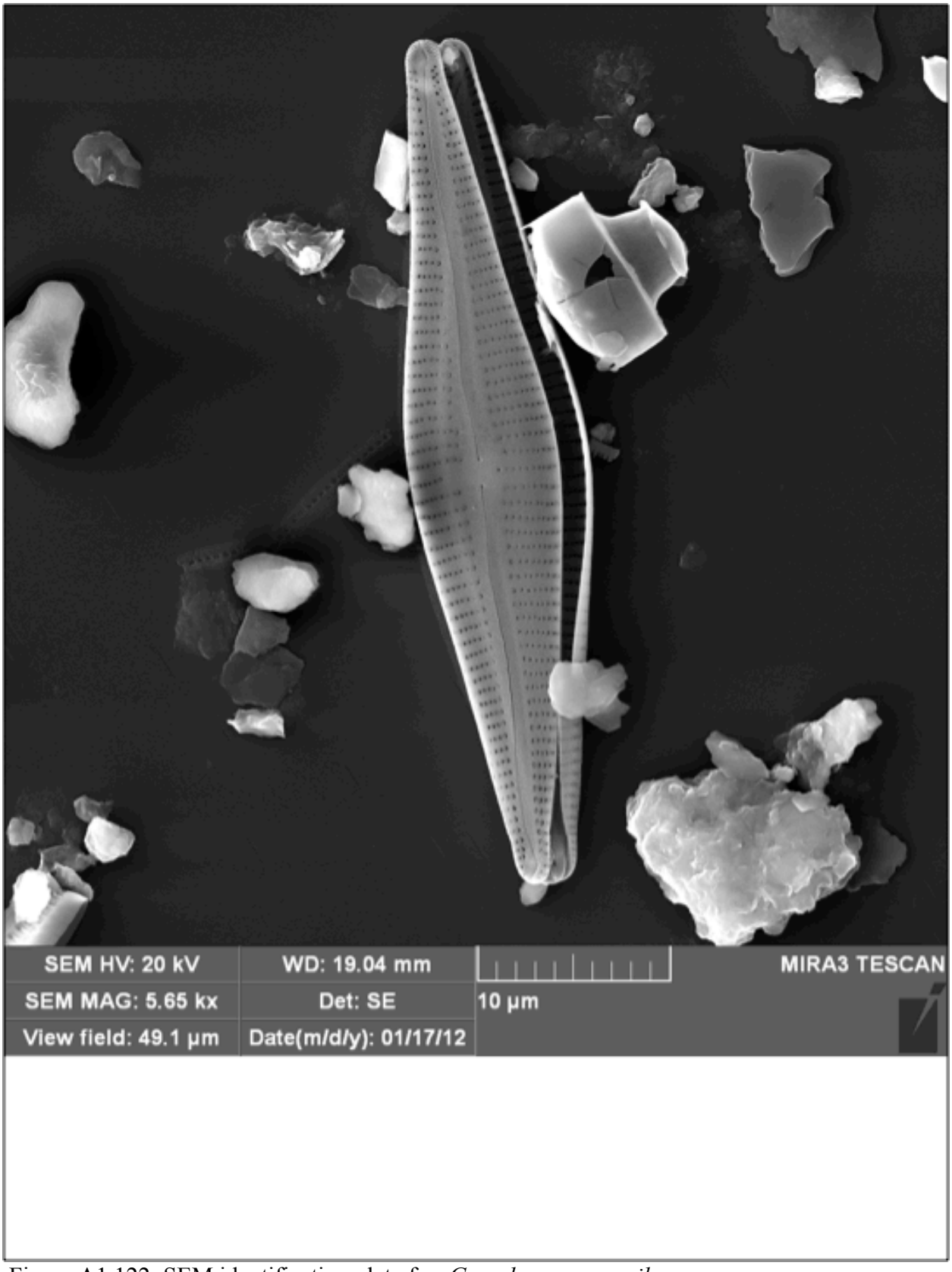


Figure A1.122: SEM identification plate for: *Gomphonema gracile*

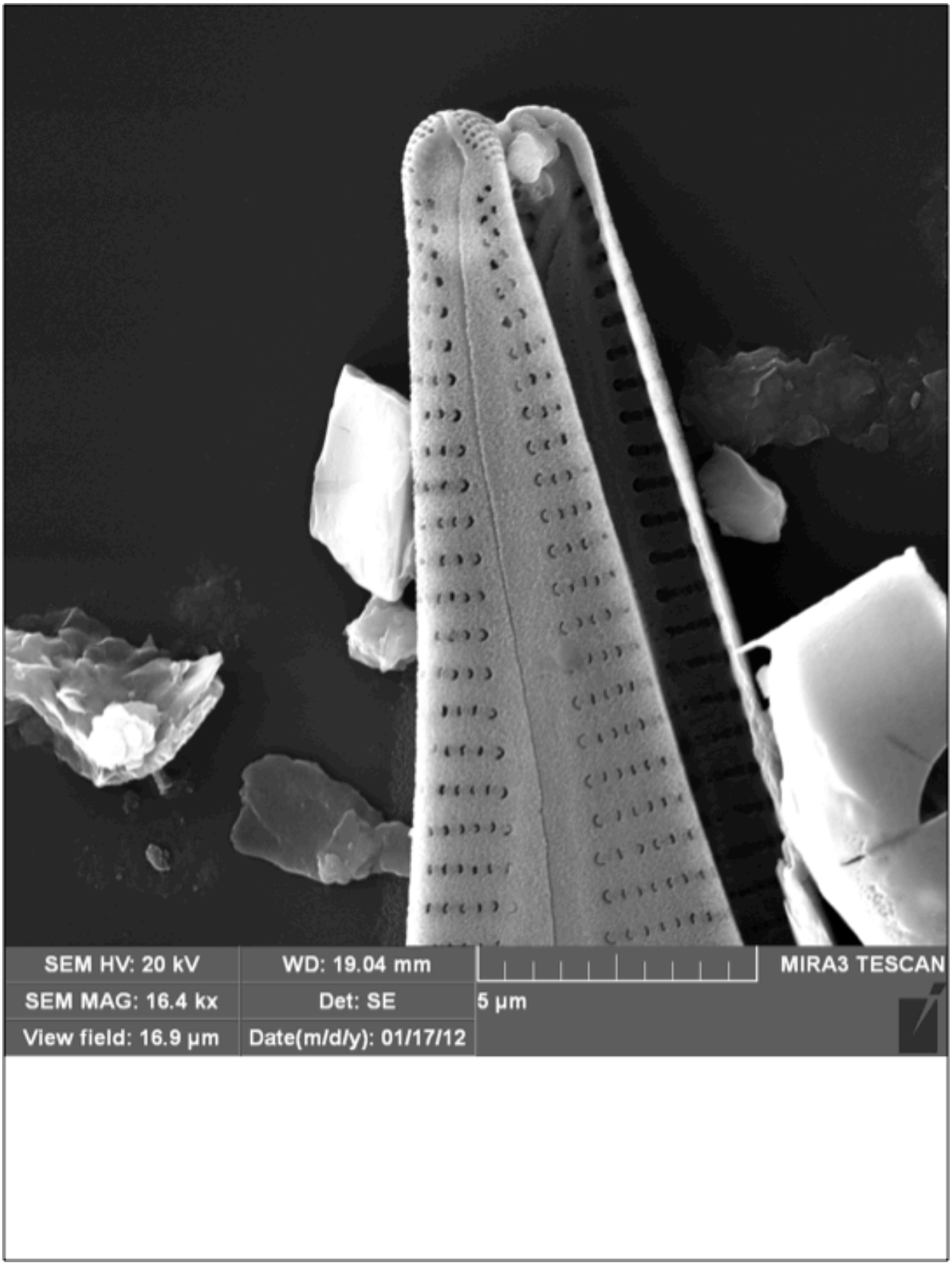


Figure A1.123: SEM identification plate for: *Gomphonema gracile*

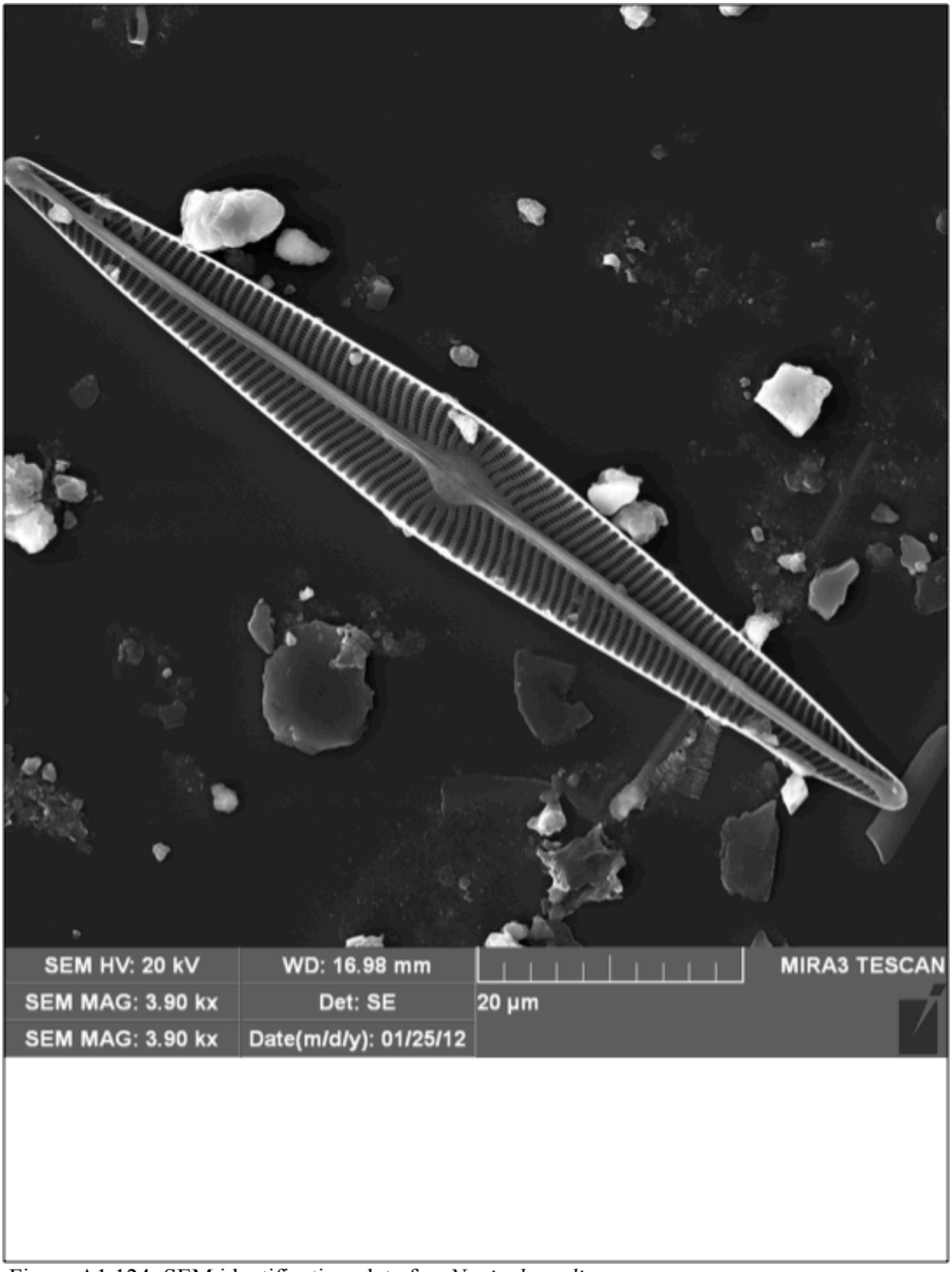


Figure A1.124: SEM identification plate for: *Navicula radiosa*

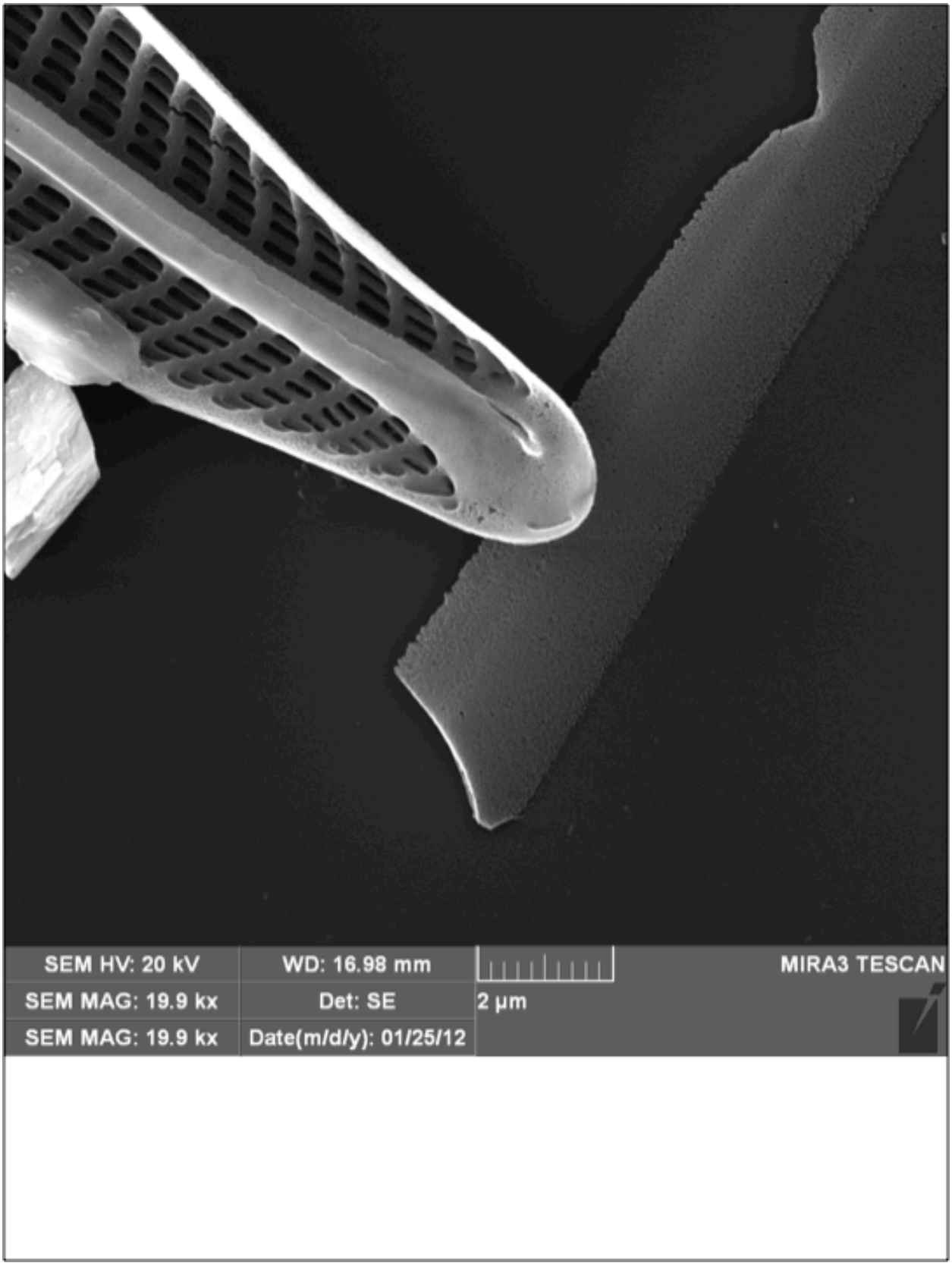


Figure A1.125: SEM identification plate for: *Navicula radiosa* (distal raphe end)

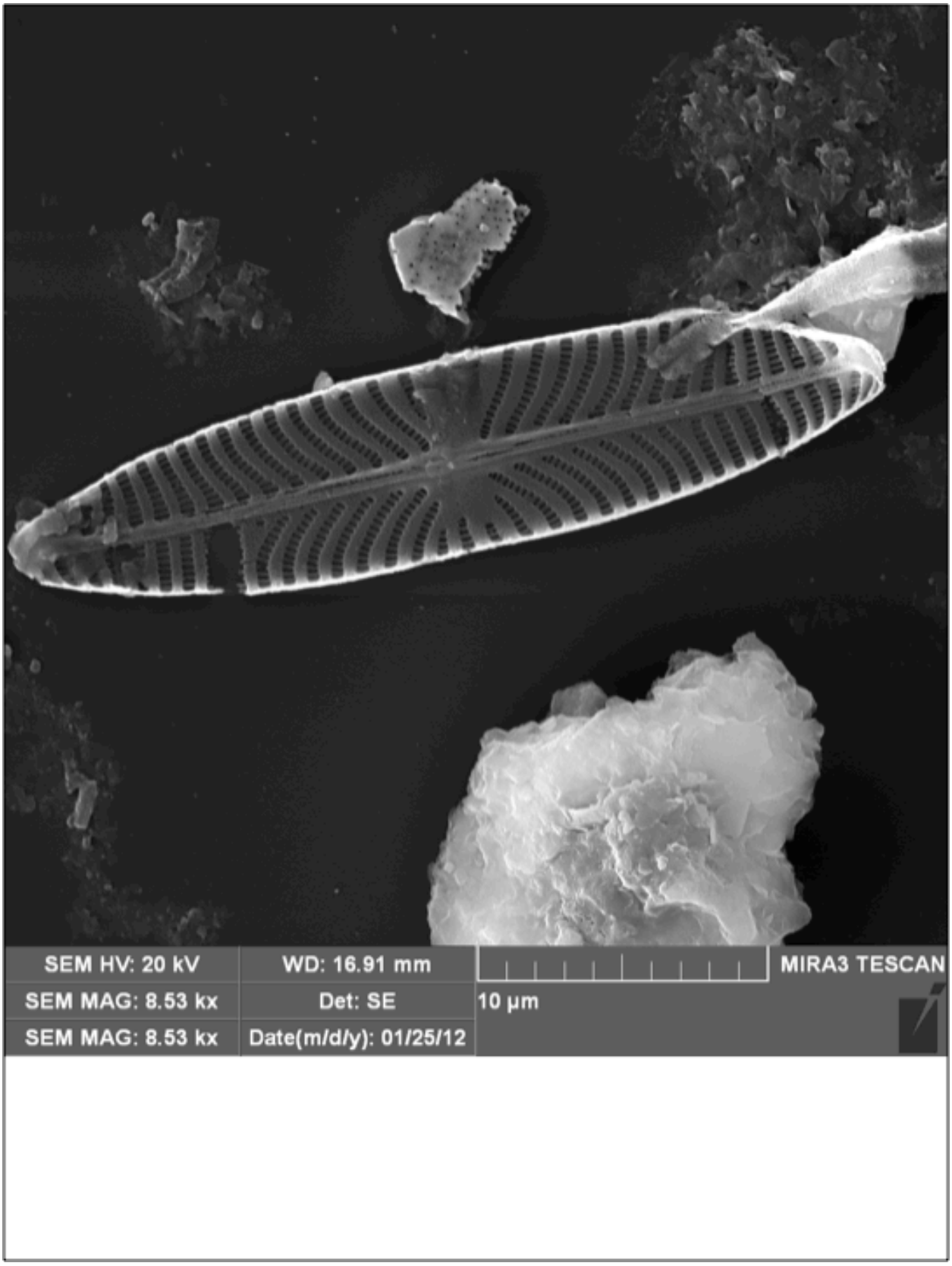


Figure A1.126: SEM identification plate for: *Navicula cari*

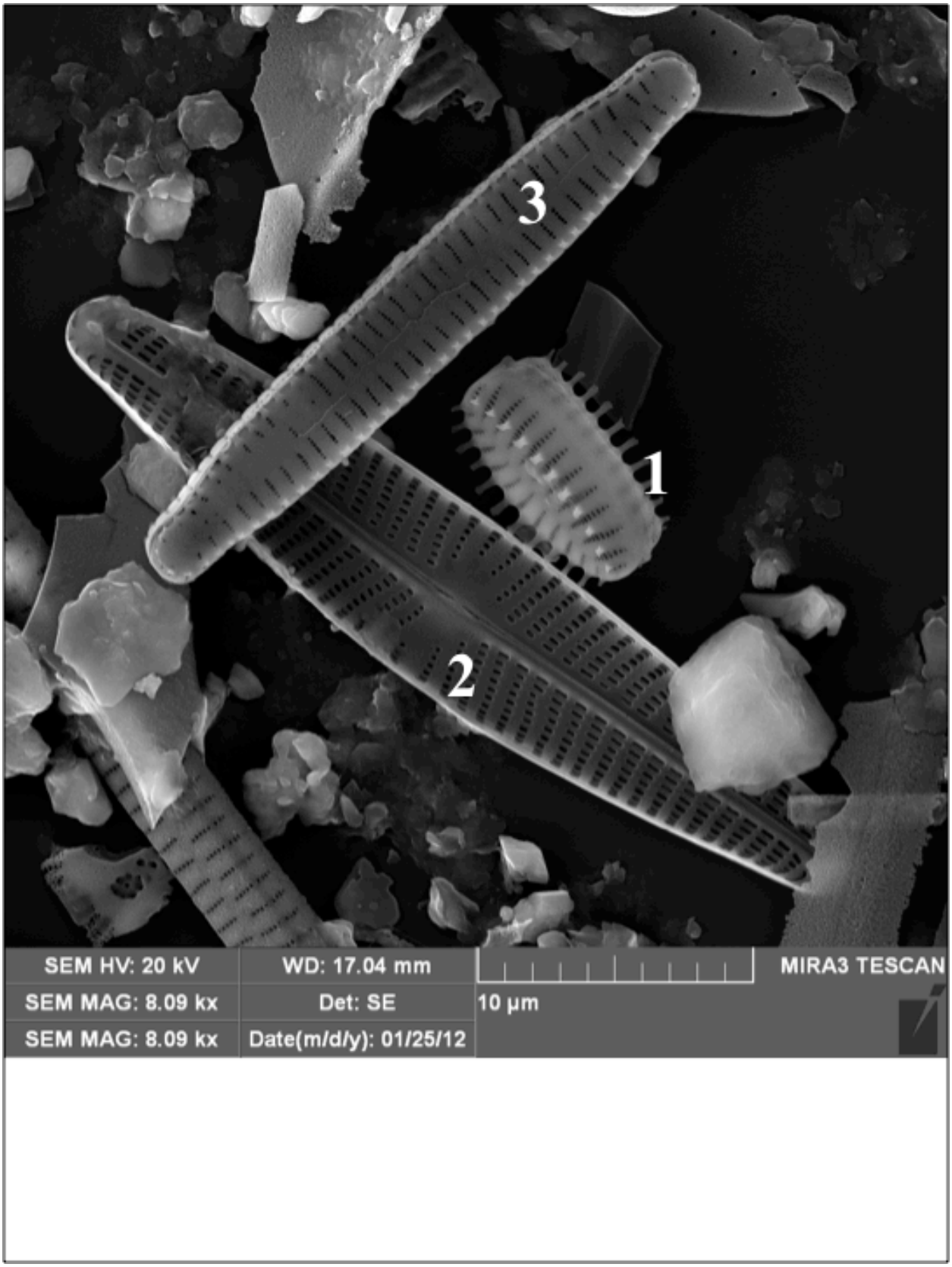


Figure A1.127: SEM identification plate for: *Navicula veneta, Fragilaria construens,* and *Fragilaria brevistriata*
1 – *Fragilaria construens*, 2 – *Navicula veneta*, 3 – *Fragilaria brevistriata*

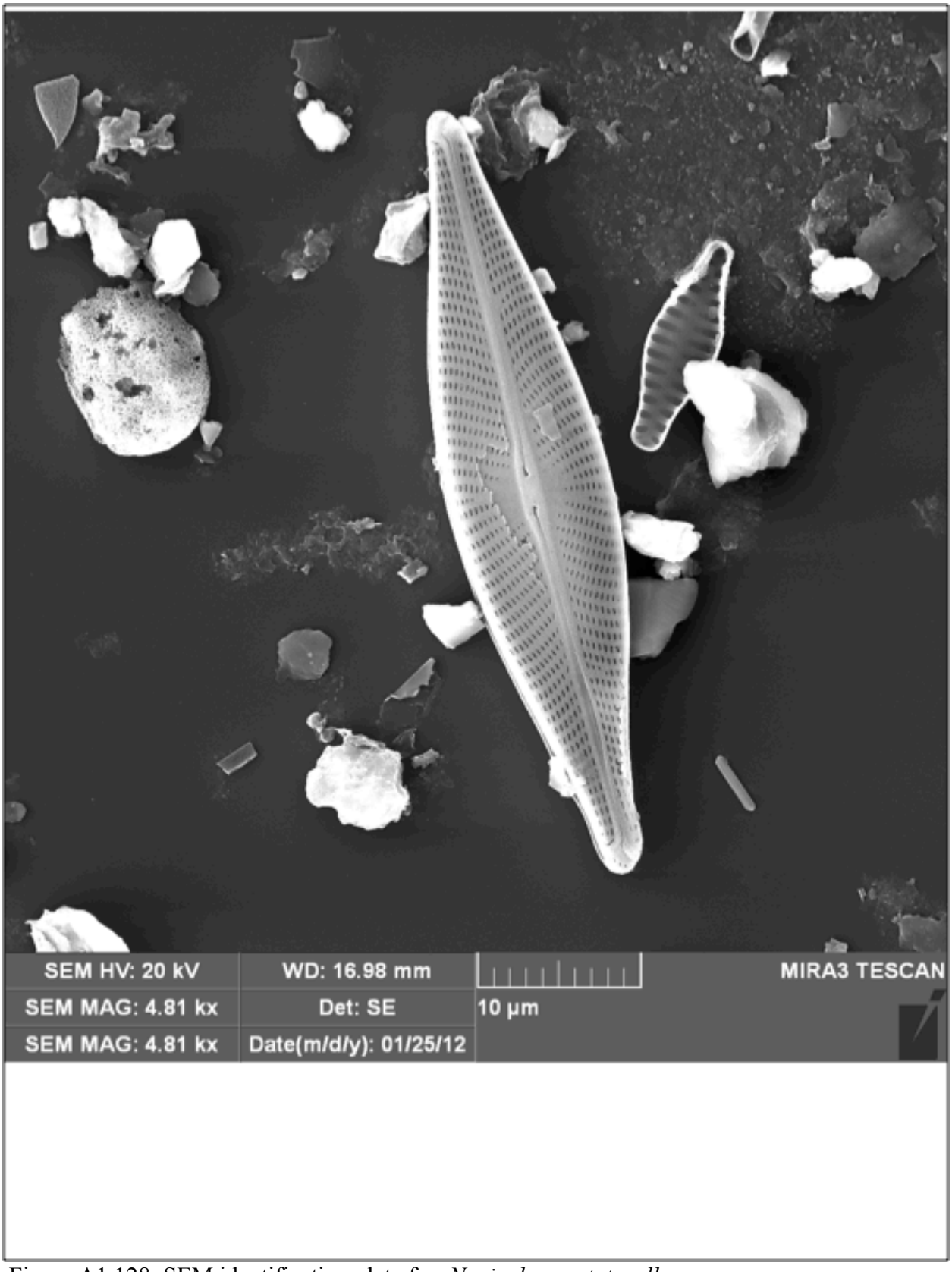


Figure A1.128: SEM identification plate for: *Navicula cryptotenella*

Figure A1.129: SEM identification plate for: *Navicula trivialis* (distal raphe end)

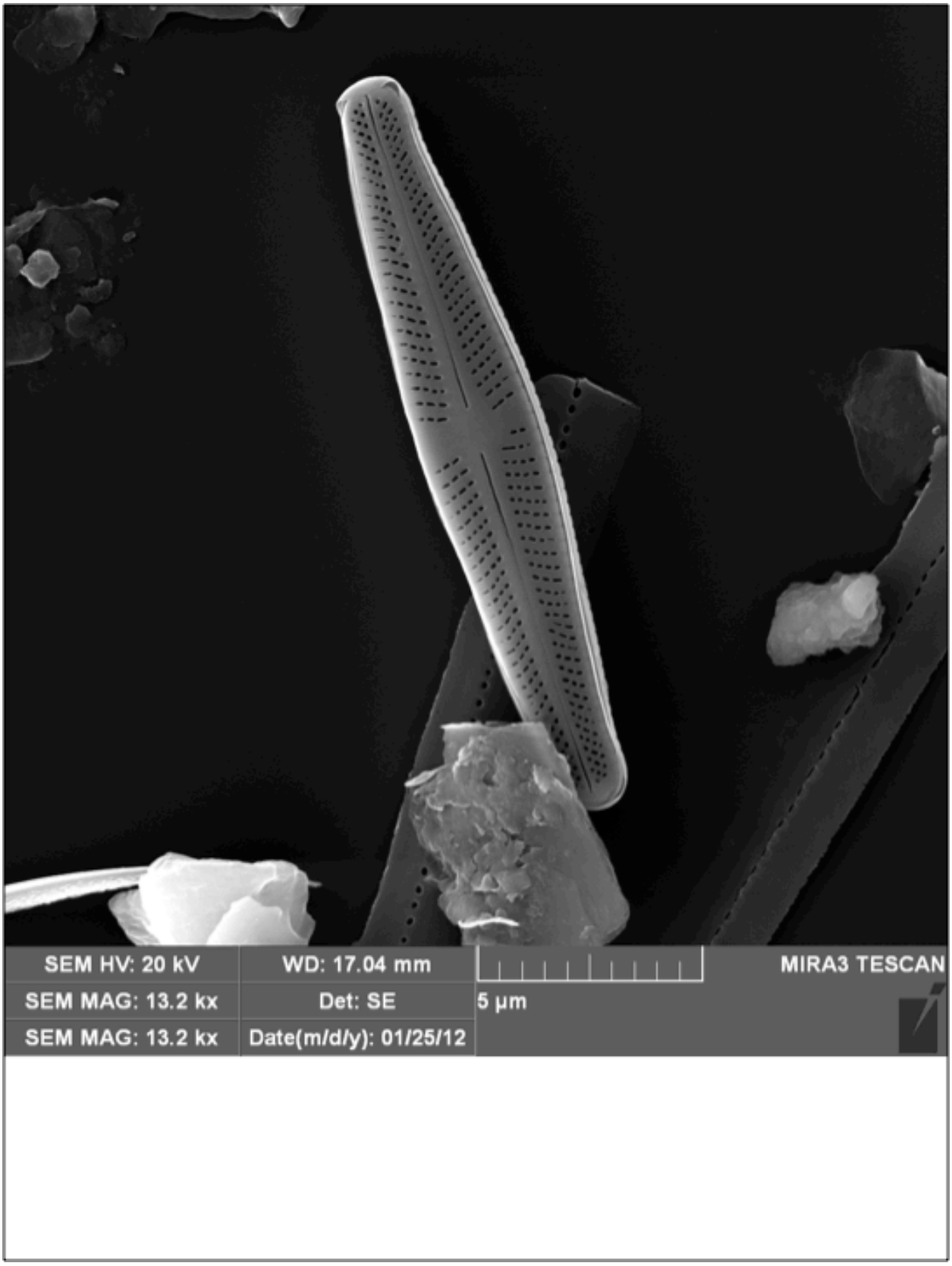


Figure A1.130: SEM identification plate for: *Achnanthes minutissima*

Figure A1.131: SEM identification plate for: *Navicula recens*

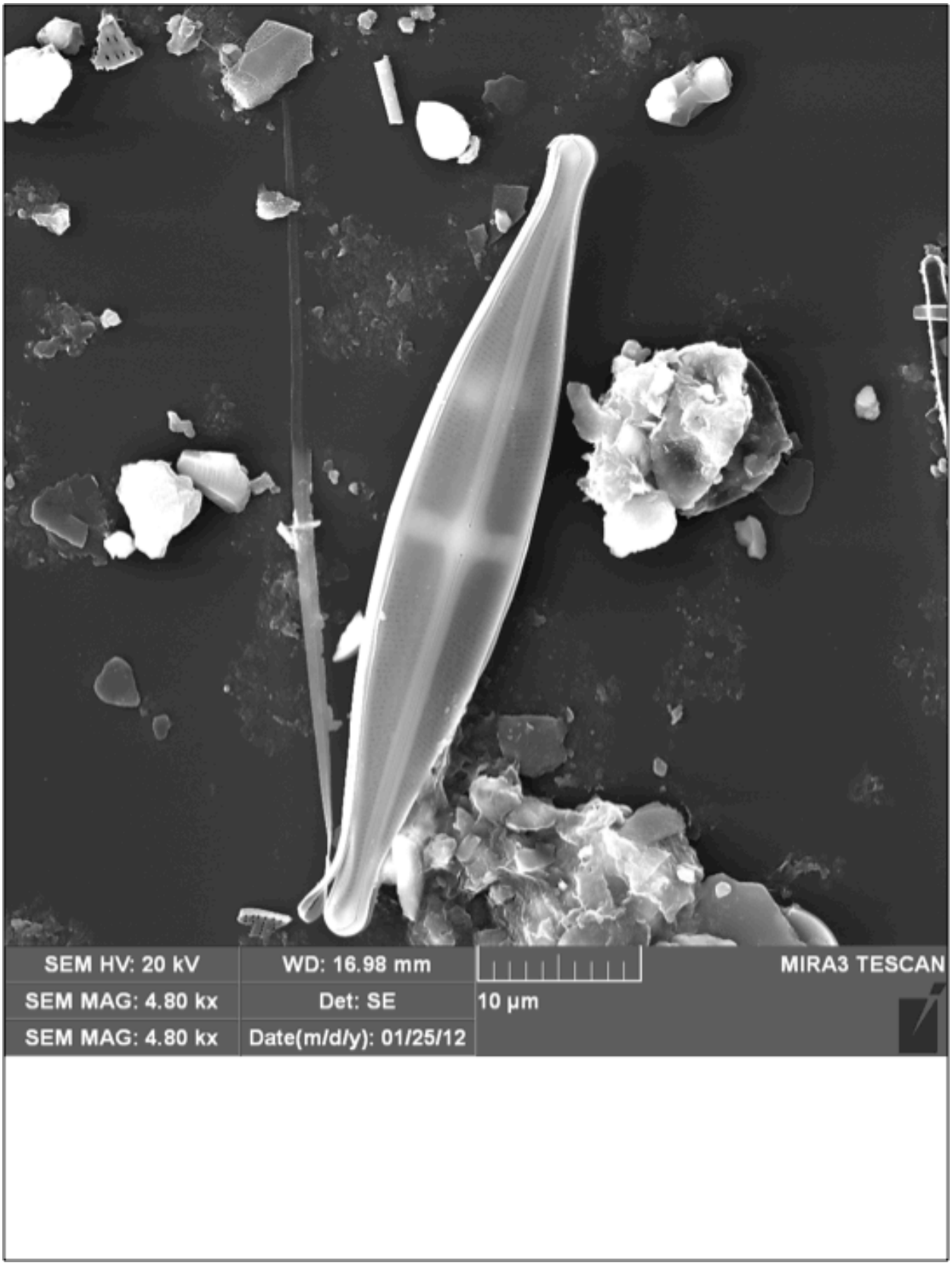


Figure A1.132: SEM identification plate for: *Stauroneis italica*

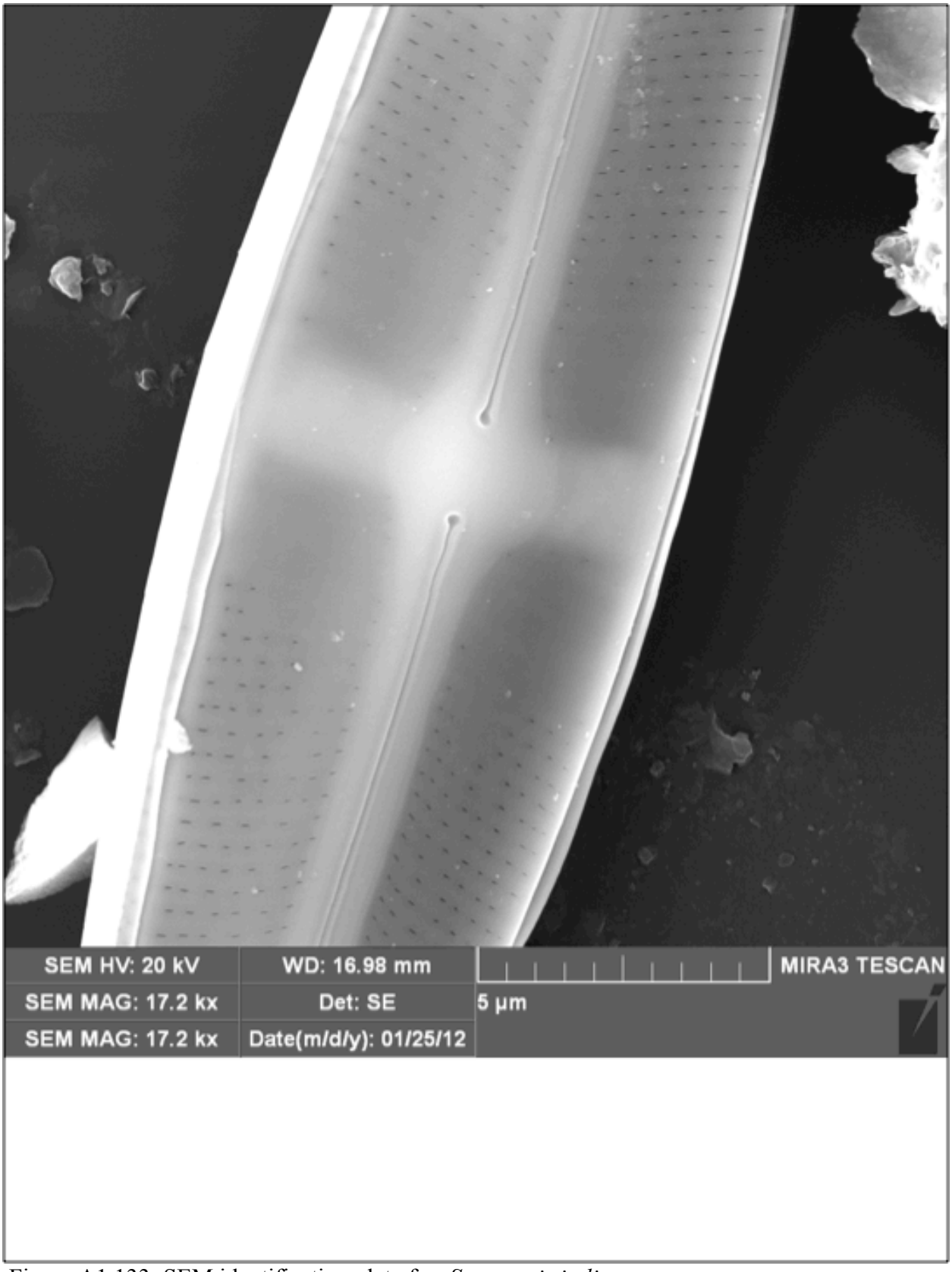


Figure A1.133: SEM identification plate for: *Stauroneis italica*

Figure A1.134: SEM identification plate for: *Navicula rhynchocephala*

Figure A1.135: SEM identification plate for: *Navicula rhynchocephala*

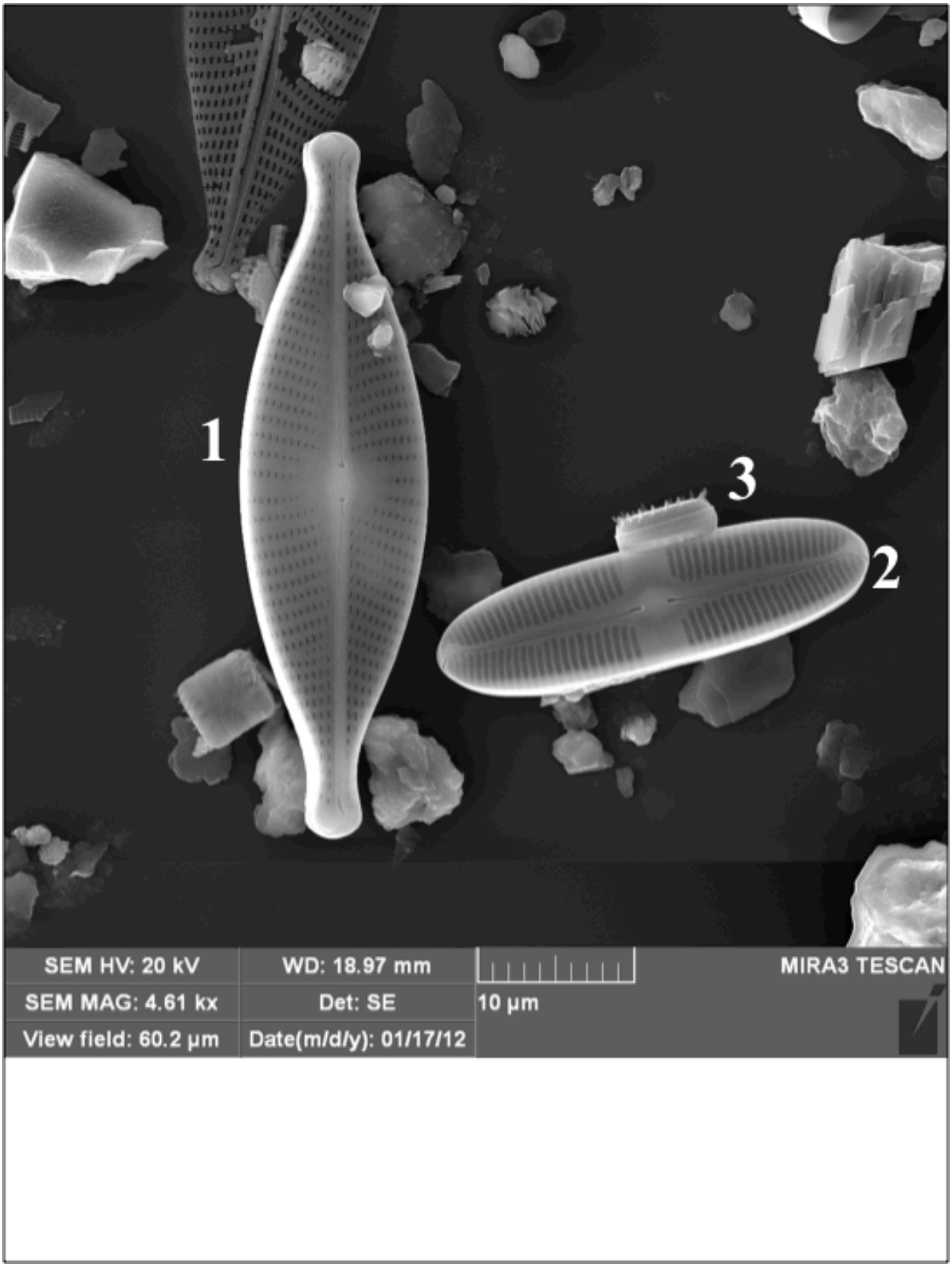


Figure A1.136: SEM identification plate for: *Navicula rhynchocephala, Caloneis silicula,* and *Stephanodiscus minutulus*

1 – *Navicula rhynchocephala*, 2 – *Caloneis silicula*, 3 – *Stephanodiscus minutulus*

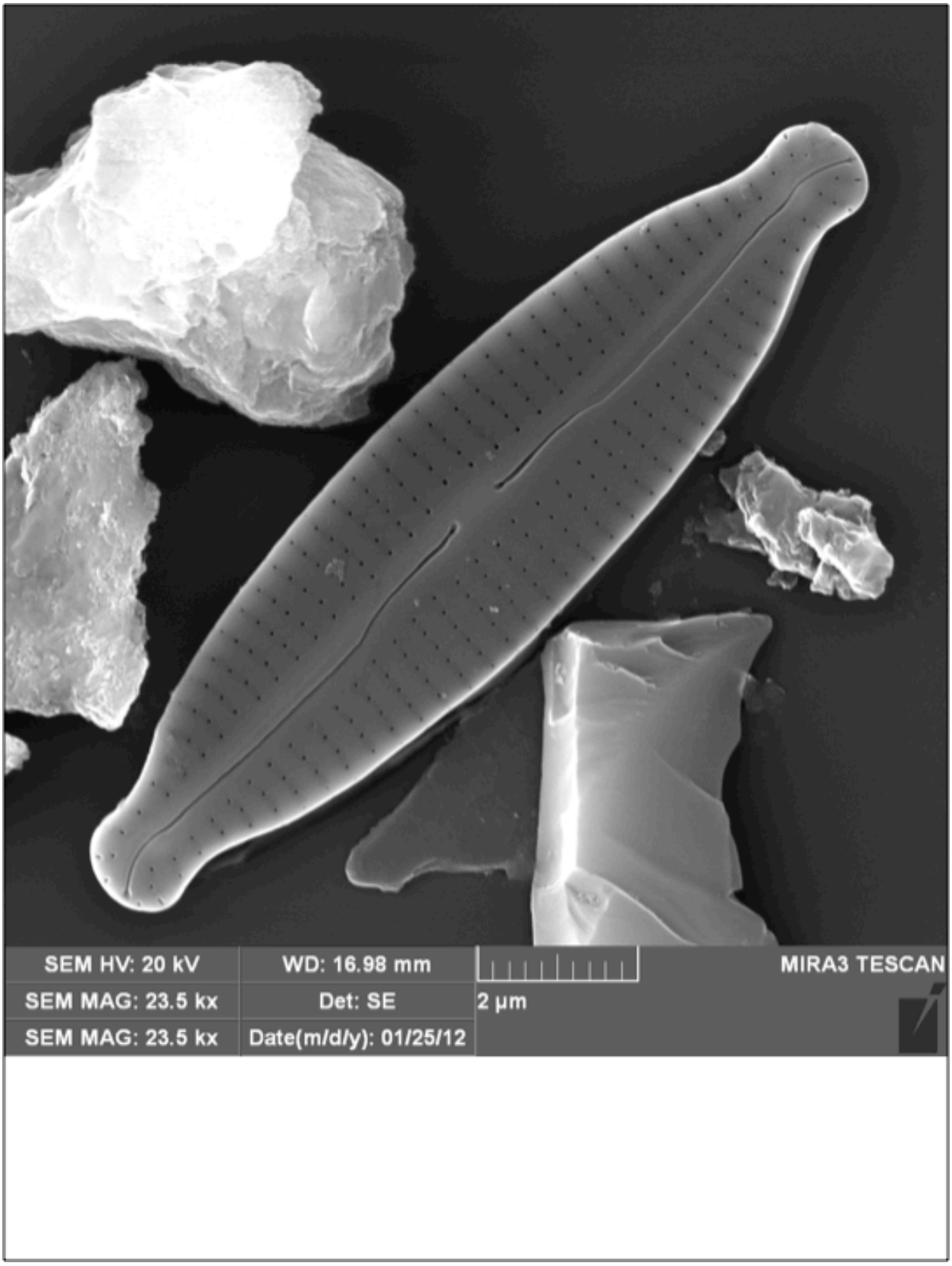


Figure A1.137: SEM identification plate for: *Cymbella amphicephala* (*Cymbopleura amphicephala*)

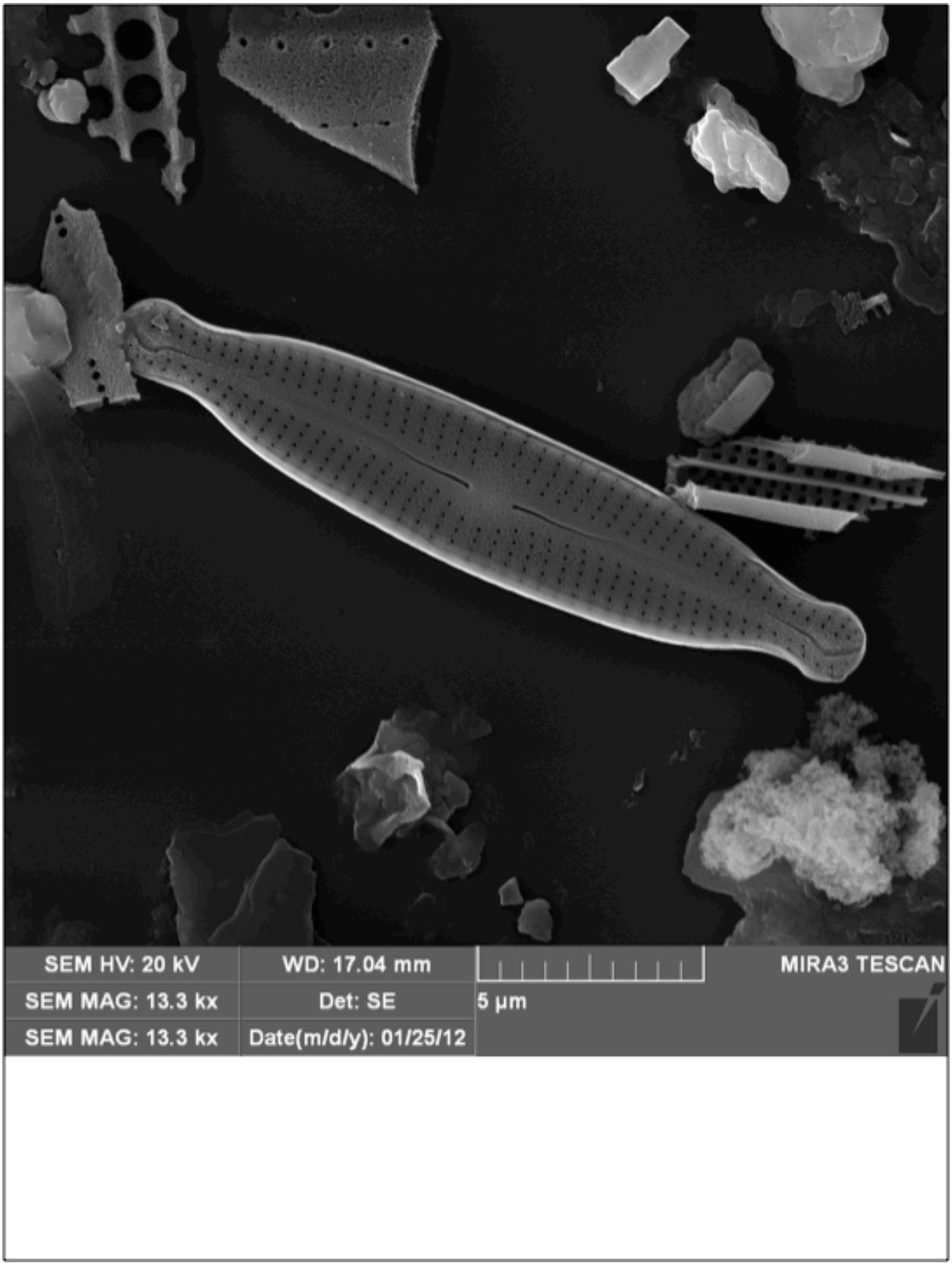


Figure A1.138: SEM identification plate for: *Cymbella amphicephala* (*Cymbopleura amphicephala*)

Figure A1.139: SEM identification plate for: *Navicula pupula*

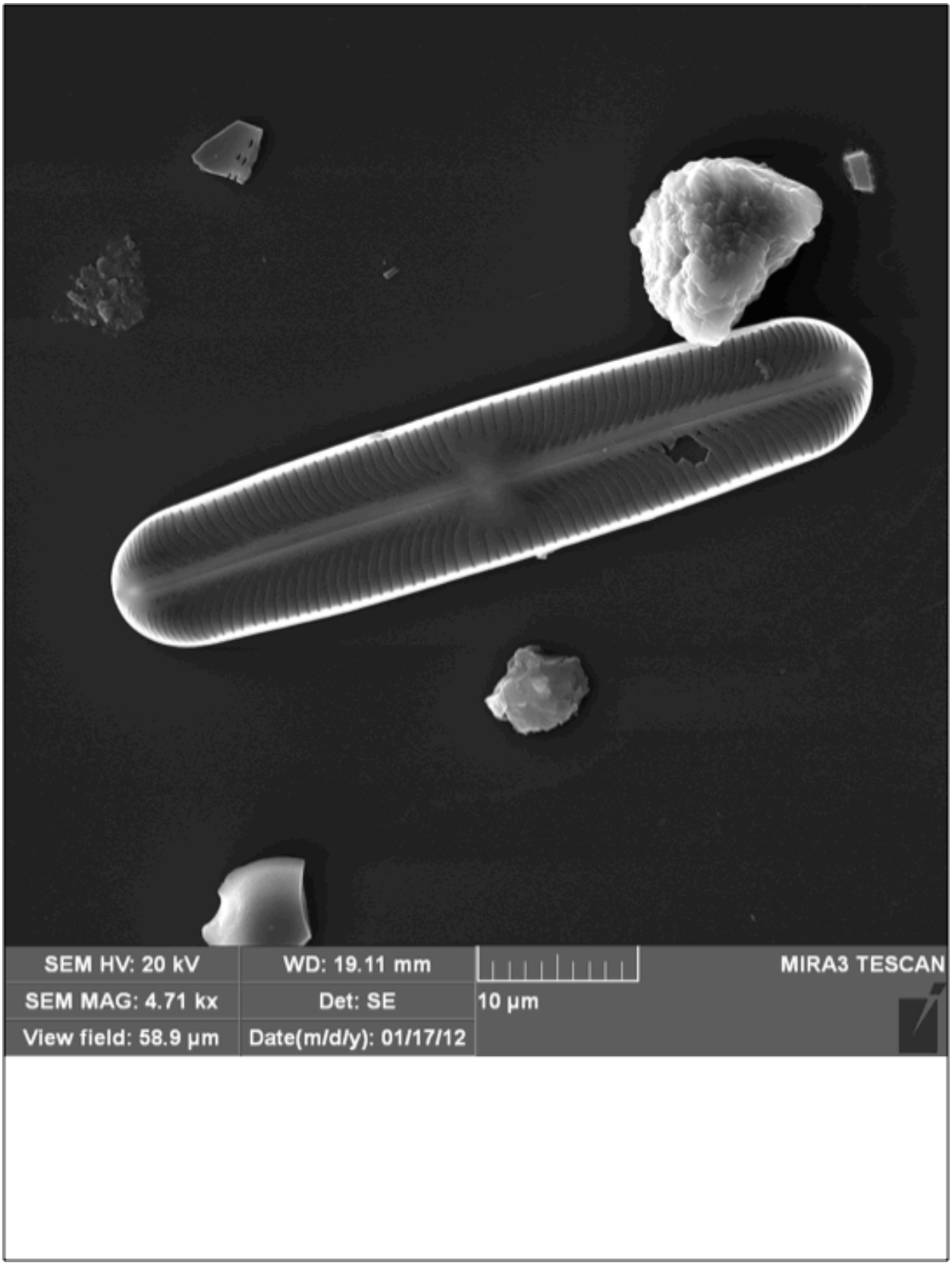


Figure A1.140: SEM identification plate for: *Navicula pupula*

Figure A1.141: SEM identification plate for: *Pinnularia abaujensis*

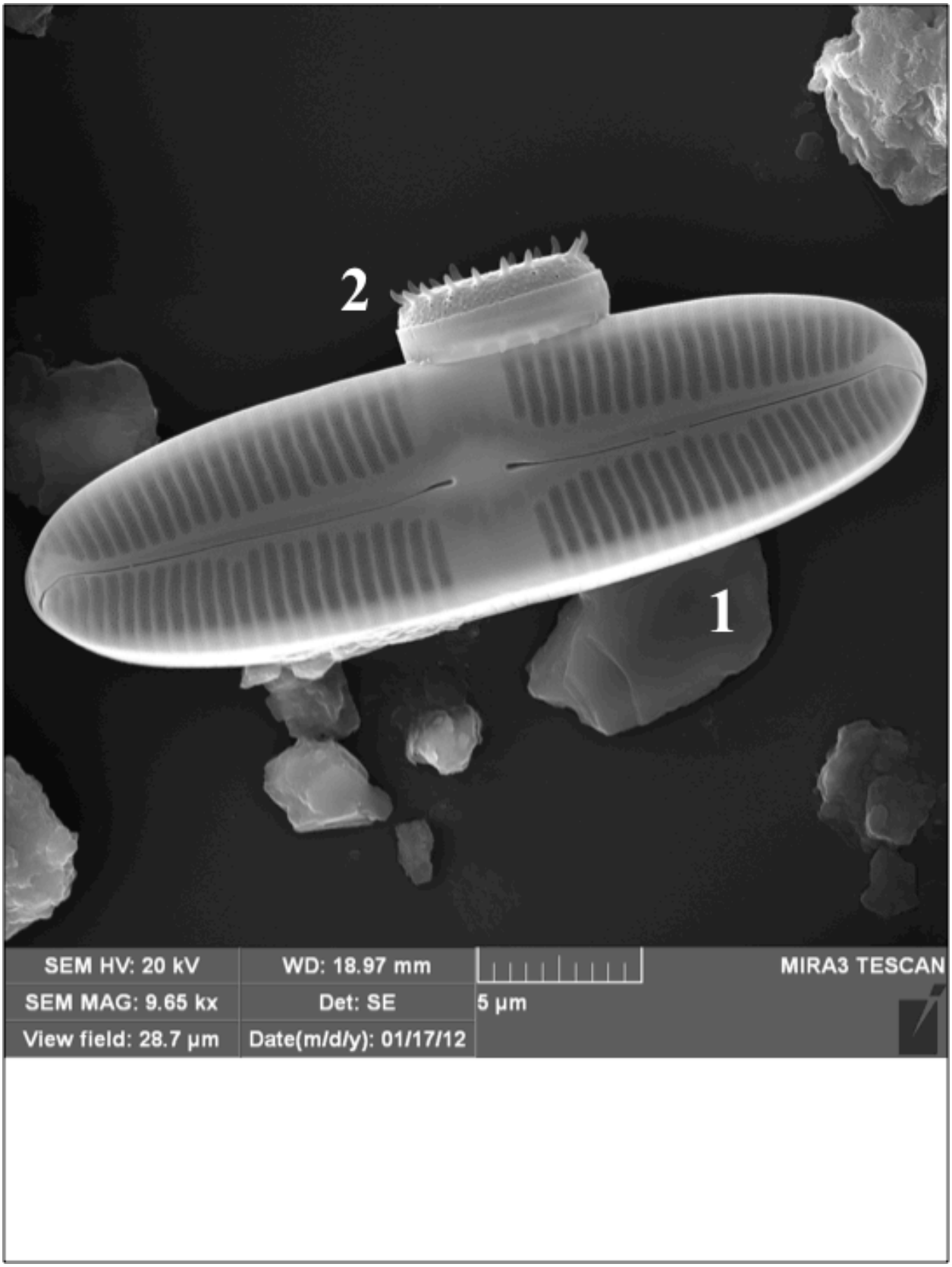


Figure A1.142: SEM identification plate for: *Caloneis silicula* and *Stephanodiscus minutulus*

1 – *Caloneis silicula,* 2 – *Stephanodiscus minutulus*

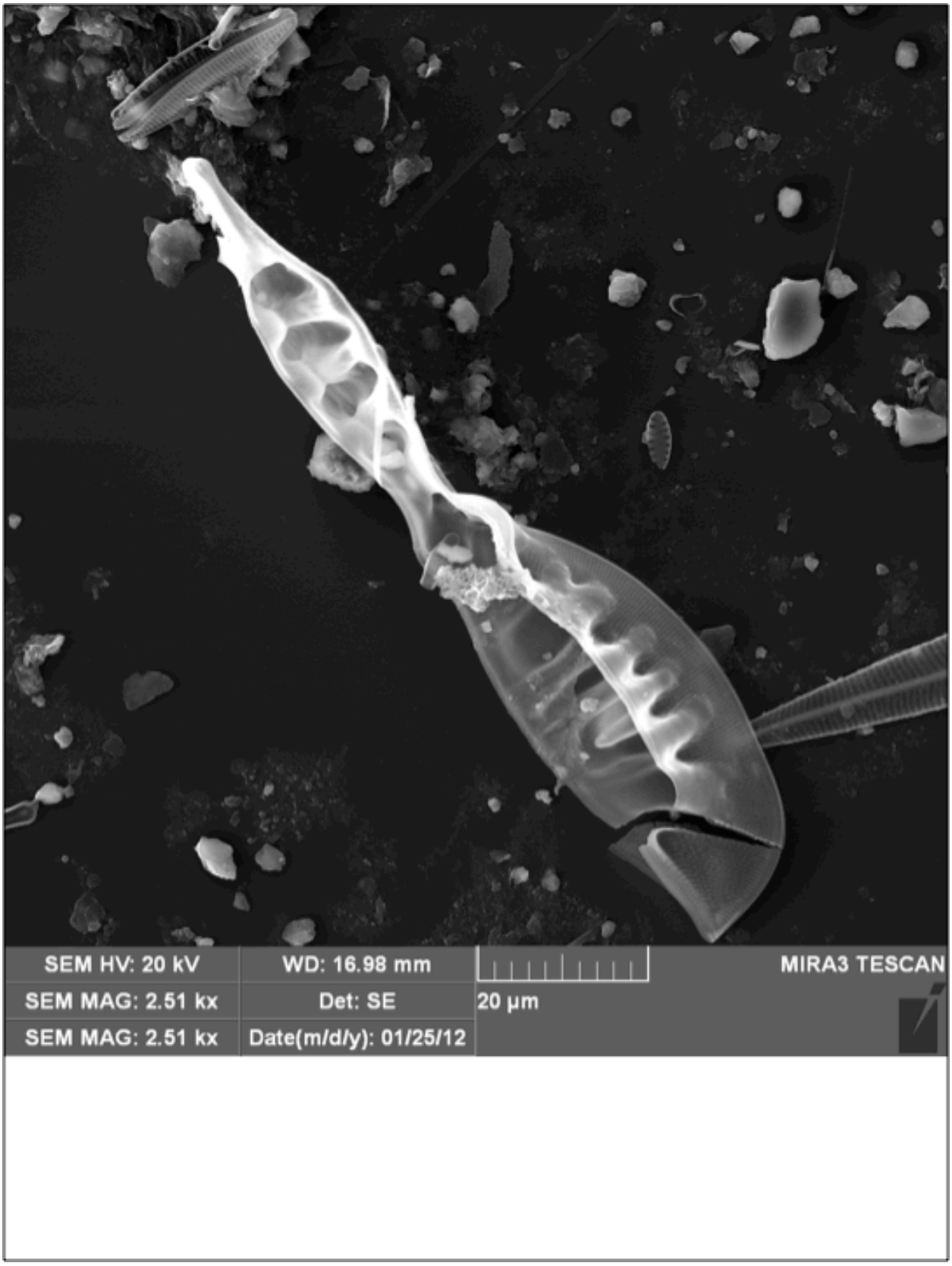


Figure A1.143: SEM identification plate for: *Entomoneis ornata*

Appendix 2. Additional analyses and data

A2.1: Outlier lake analysis

DCA and PCA analysis was used to determine the presence or absence of outlier lakes following Hall & Smol (1992). The associated axis scores for each lake with respect to environmental data (PCA) and diatom taxa (DCA) are in Table A2.1. Those lakes in which all four axis scores fell outside the 95% confidence interval were considered to be outliers.

Table A2.1: PCA and DCA axis scores of 47 sample lakes and associated confidence intervals used to determine presence of outlier lakes within dataset. Those values outside the confidence intervals highlighted in gray, those lakes where all four scores are outside confidence limits are considered outliers within the dataset and are indicated by a (*).

Lake	PCA Axis 1	PCA Axis 2	DCA Axis 1	DCA Axis 2
1	0.0242	-0.113	1.8327	2.7929
2	2.9199	0.3141	1.7015	1.4151
3	-0.3519	0.6904	2.6523	1.6919
4	-1.8086	0.0126	0.0153	1.4404
5	-0.1763	2.3805	2.2446	1.7927
6	-0.1195	0.7841	0	1.4538
7	-1.0669	-0.5933	1.7909	0.6337
8	-1.6633	-0.3229	1.8759	0
9	0.1902	-0.5958	1.7963	1.0424
10	-2.2831	-0.8301	1.7395	0.6013
11	-1.2488	1.213	1.7838	0.4783
13*	0.6447	-1.1578	2.3001	0.8862
14	0.2543	-0.505	1.1458	0.579
15	0.4959	0.2237	0.5582	1.8313
16	-0.5943	-0.1546	0.7614	1.736
17*	-0.4416	-0.5499	0.9967	2.3432
18	0.9707	1.1835	1.5125	2.0458
19	-0.7507	-0.579	1.9053	1.5834
30	-0.9597	-0.1896	0.7163	1.025
21	0.254	-0.4904	2.3979	1.7571
22	2.8952	-0.4277	0.6559	1.6154
23*	-1.8633	0.3295	0.6108	1.1325
24*	-0.7865	-0.8507	0.179	0.3913
25	0.0269	0.5938	1.779	1.6449
26	-0.1161	3.2566	2.5449	2.2456
27*	0.4	2.4204	0.9311	0.6003
28*	1.1925	0.4535	2.7595	2.3772
29	0.2455	-0.7126	2.9189	1.8792
30*	0.3642	0.6573	2.4594	2.0322
31	0.9933	-0.1783	1.5035	2.3075
33	-0.3618	-0.0816	0.4594	3.2756
34	-0.0532	1.4807	2.0957	3.1953
35	0.1416	1.0695	2.0977	1.9414
36	-0.3123	0.1134	1.6634	2.8943
37*	0.6263	-0.4186	3.2607	2.2276

Lake	PCA Axis 1	PCA Axis 2	DCA Axis 1	DCA Axis 2
38	-0.1465	-0.516	3.6957	1.5655
39*	0.7771	-1.3012	2.8982	2.1378
40*	0.4623	-1.0837	2.9576	1.3564
43	0.6263	-0.5008	1.7756	2.4394
44	0.2776	0.1983	3.6046	1.9286
45	-0.4541	-0.2318	2.0061	2.368
46	0.4747	-1.572	2.8738	1.54
48	-0.2147	-0.1669	2.2954	1.5772
49	-0.6704	-1.3784	2.5975	1.8568
50	-0.0079	-0.9764	3.343	1.7691
51*	1.1945	-0.8969	0.8765	3.5385
Confidence+	0.2922	0.2922	2.1145	1.9419
Confidence-	-0.2922	-0.2922	1.5624	1.4915

A2.2: Diatom analysis and DI-TP for South Lake

South Lake is directly adjacent to Clear Lake and is separated from it only by a narrow sandy berm. It covers 2.06 km^2 and in contrast with Clear Lake it is much more shallow, has murky waters, with abundant macrophyte and phytoplankton populations. South Lake has two distinct basins: the North Basin, which is directly adjacent to Clear Lake, and the South Basin. The North Basin is dominated by emergent bulrush, whereas the South Basin is lacking in emergent vegetation (Scott & Kling 2006). For the purposes of this study, the North Basin was not included in analysis; only samples from the South Basin were analyzed. The South Basin productivity is limited by phosphorus input and exists in a hyper-eutrophied and phytoplankton-dominated state (Scott & Sellers 2003). South Lake is much more eutrophic in comparison with Clear Lake and was analysed extensively with respect to diatoms by Scott and Kling (2006).

South Lake sediment core characteristics are summarized in Figure A2.1, with respect to ^{210}Pb activity, sediment accumulation rate, and age (as estimated by ^{210}Pb dating). A chronology for the core sediments was provided using ^{210}Pb dating, using a constant rate of supply model. South Lake has a much slower rate of sediment accumulation, as indicated by the 130 years contained within the top 10cm of the core sample. ^{210}Pb activity peaked at the surface of the core at 0.319 Bq/g and was below the 0.020 Bq/g level from the base of the core to a depth of 10cm. The sediment accumulation rate peaked at 390 g/m2/year in 1986. The error surrounding the calculated ^{210}Pb chronology increases with depth however, from the 8cm onward, the error is $\leq \pm 10$ years. Dates obtained using the ^{210}Pb CRS model developed for South Lake are used in subsequent diatom species abundance diagrams (Figure A2.2).

Species relative abundances are shown in Figure A2.2 with corresponding species cluster analysis (CONISS) on the far right hand side of figure red dotted line indicating the approximate time of RMNP establishment and the blue dotted line approximately the establishment of the areas a Timber Reserve. South Lake samples were analysed at a 4cm resolution.

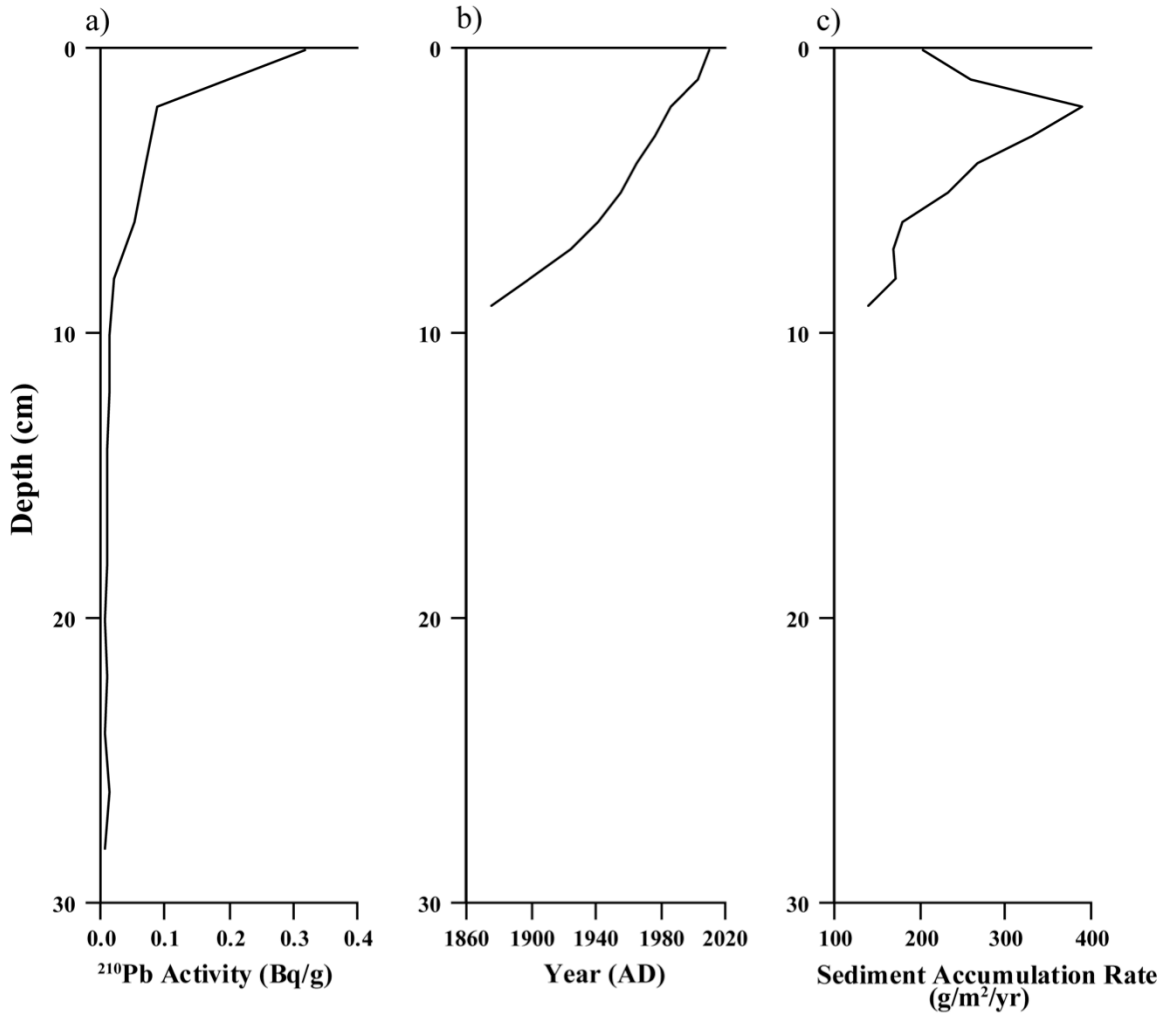


Figure A2.1: South Lake core data, (a) ^{210}Pb activity (Bq/g), (b) Year (AD) and (c) Sediment accumulation rate (g/m^2/year).

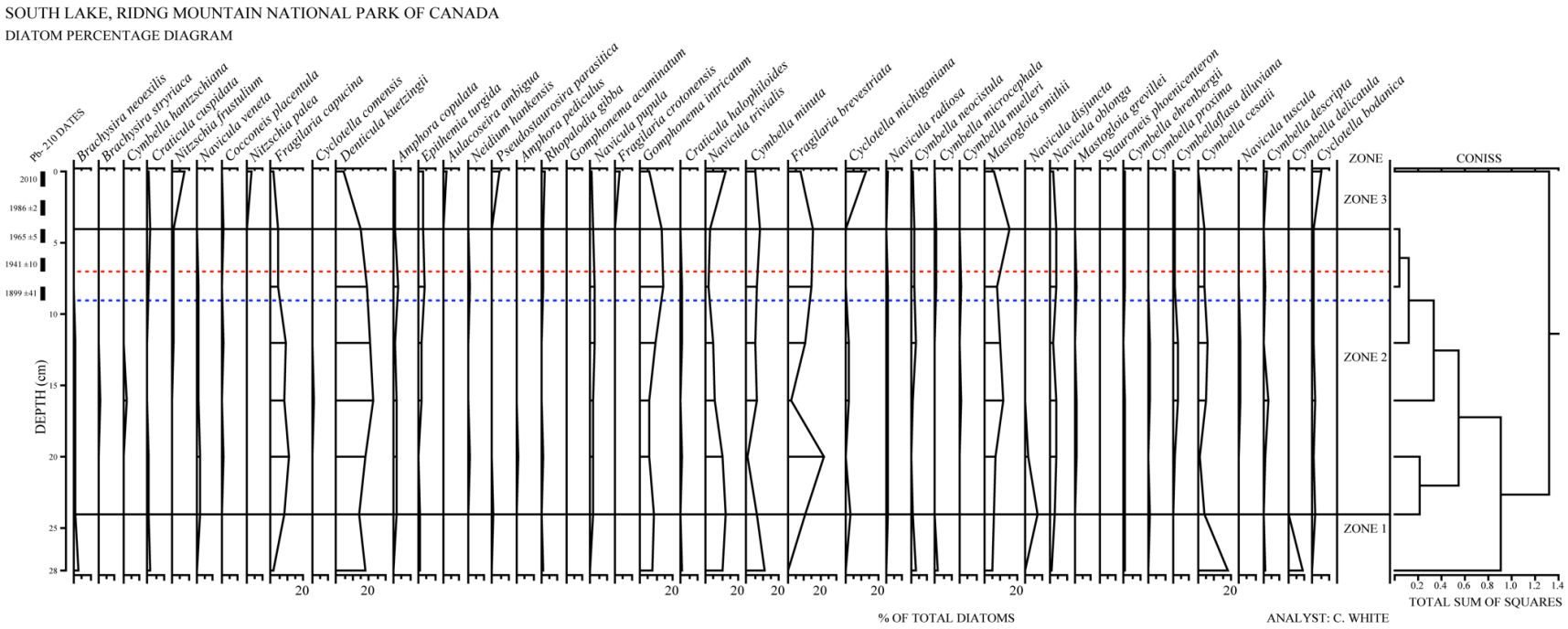


Figure A2.2: Diatom stratigraphy of the most important and abundant taxa from South Lake arranged according to calculated weighted averaging bootstrapped TP optima and grouped into corresponding trophic groups. The solid black horizontal lines indicate zone boundaries, as determined by CONISS sum of squares analysis (far right). The blue dashed line represent the approximate time of establishment of the region as a Timber Reserve (1895), and the red dashed line indicates the approximate time of park establishment (1933), as determined by ^{210}Pb dating techniques.

South Lake was excluded from further discussion and analysis due to the previous extensive study completed by Scott & Kling (2006) and the relatively little variation present in the diatom communities throughout the core. Preliminary reconstruction of TP from the DI-TP model developed in Chapter 3 of this thesis was conducted and does not appear to reveal much variation in the TP levels throughout the length of the core (Figure A2.3). Scott & Kling determined that the most significant changes in the diatom fossil assemblages occurred within the last 40 years, most likely due to increased nutrient inputs, and an increase in trophic state in the last 20 years (2006). This increase in trophic state manifests as a switch from a clear, macrophyte-dominated, and abundant benthic algae state to a phytoplankton-dominated system that is turbid and unvegetated (Scott & Kling, 2006).

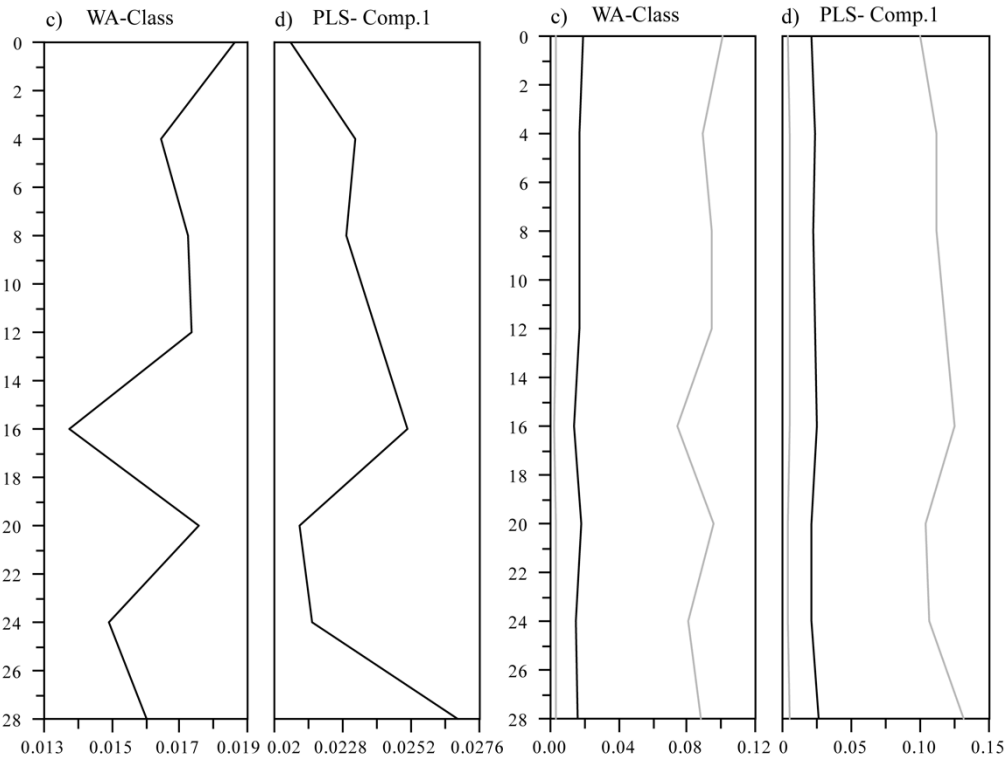


Figure A2.3: Diatom Inferred Total Phosphorus (mg L^{-1}) South Lake comparing Weighted Averaging (classical deshrinking) modelling techniques (a,c) and Partial Least Squares (first component) modelling techniques (b,d,) with (c,d) and without (a,d) error bars.

A2.3: Diatom analysis and DI-TP for Moon Lake

Moon Lake is the most northern lake included in this study and is not accessible to the public for any motorized boating. Moon Lake experiences frequent algal blooms, in the form of cyanobacteria, and as such, visitors to Moon Lake are often not permitted to enter the lake. There

is a great amount of macrophytes present within Moon Lake, in particular on the north shore where dense mats form.

Moon Lake sediment core characteristics are summarized in Figure 3.8, with respect to ^{210}Pb activity, sediment accumulation rate, and age (as estimated by ^{210}Pb dating). A chronology for the core sediments was provided using ^{210}Pb dating, using a constant rate of supply model. There are no drastic shifts in the activity level of ^{210}Pb as it increases at a relatively constant rate over the length of the core. It peaks at the top of the core at 0.671 Bq/g. Calculated ^{210}Pb dates have an error of $\leq \pm 10$ years from 1927 (28cm) upward. Sediment accumulation rate is decreasing in the upper 7 cm of the core. Sediment accumulation rate is lowest at the base of the core, and also has a significant decrease to 168 g/m^2/year at a depth of 22 cm. Dates obtained using the ^{210}Pb CRS model developed for Moon Lake are used in subsequent diatom species abundance diagrams (Figure A2.4).

Species relative abundances are shown in Figure A2.5 with corresponding species cluster analysis (CONISS) on the far right hand side of figure, the red dotted line indicating the approximate time of RMNP establishment and the blue dotted line approximately the establishment of the areas a Timber Reserve. Moon Lake samples were analysed at a 4cm resolution, to a depth of 10 cm as below this depth diatom were not present in high enough concentration to be enumerated.

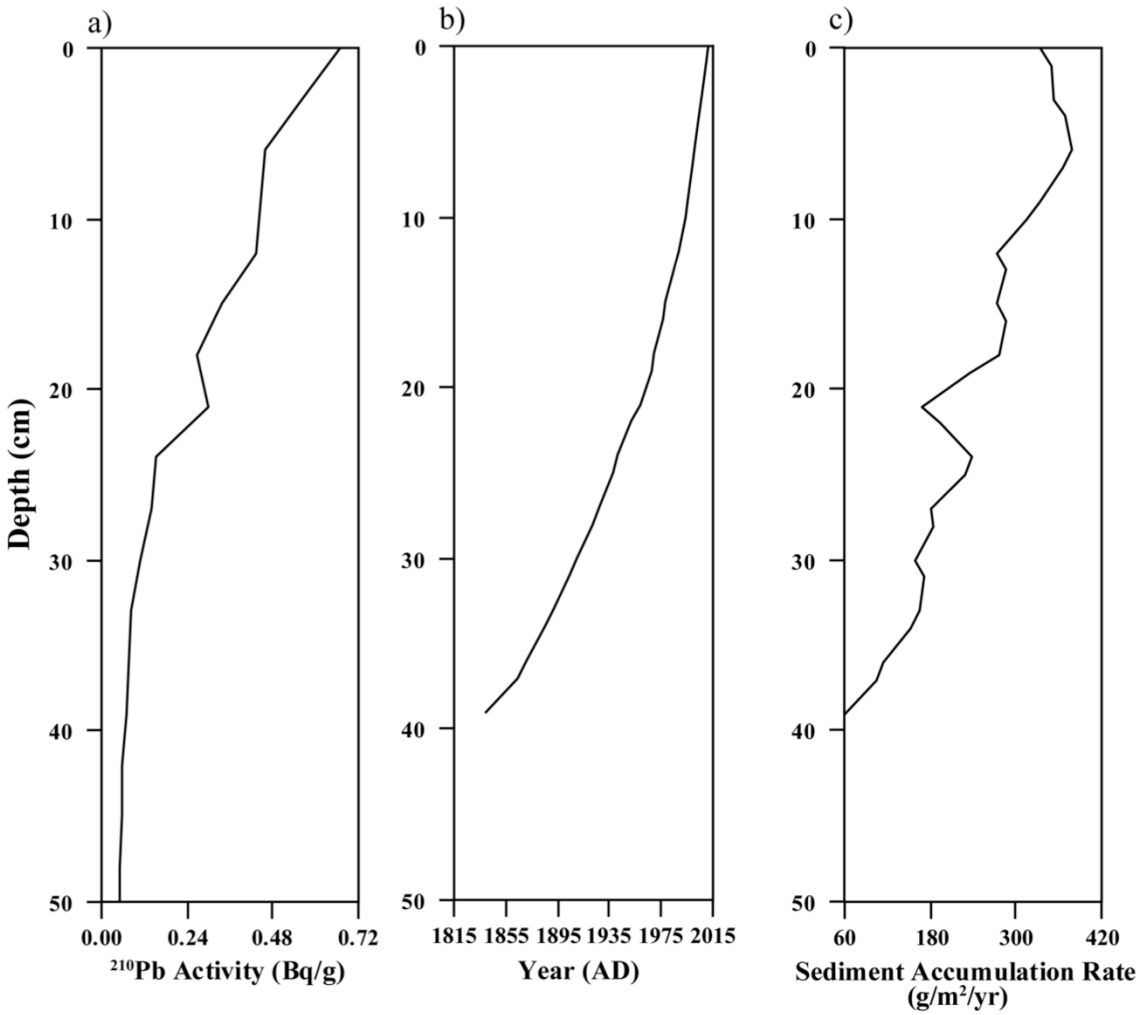


Figure A2.4: Moon Lake core data, (a) ^{210}Pb activity (Bq/g), (b) Year (AD) and (c) Sediment accumulation rate (g/m^2/year).

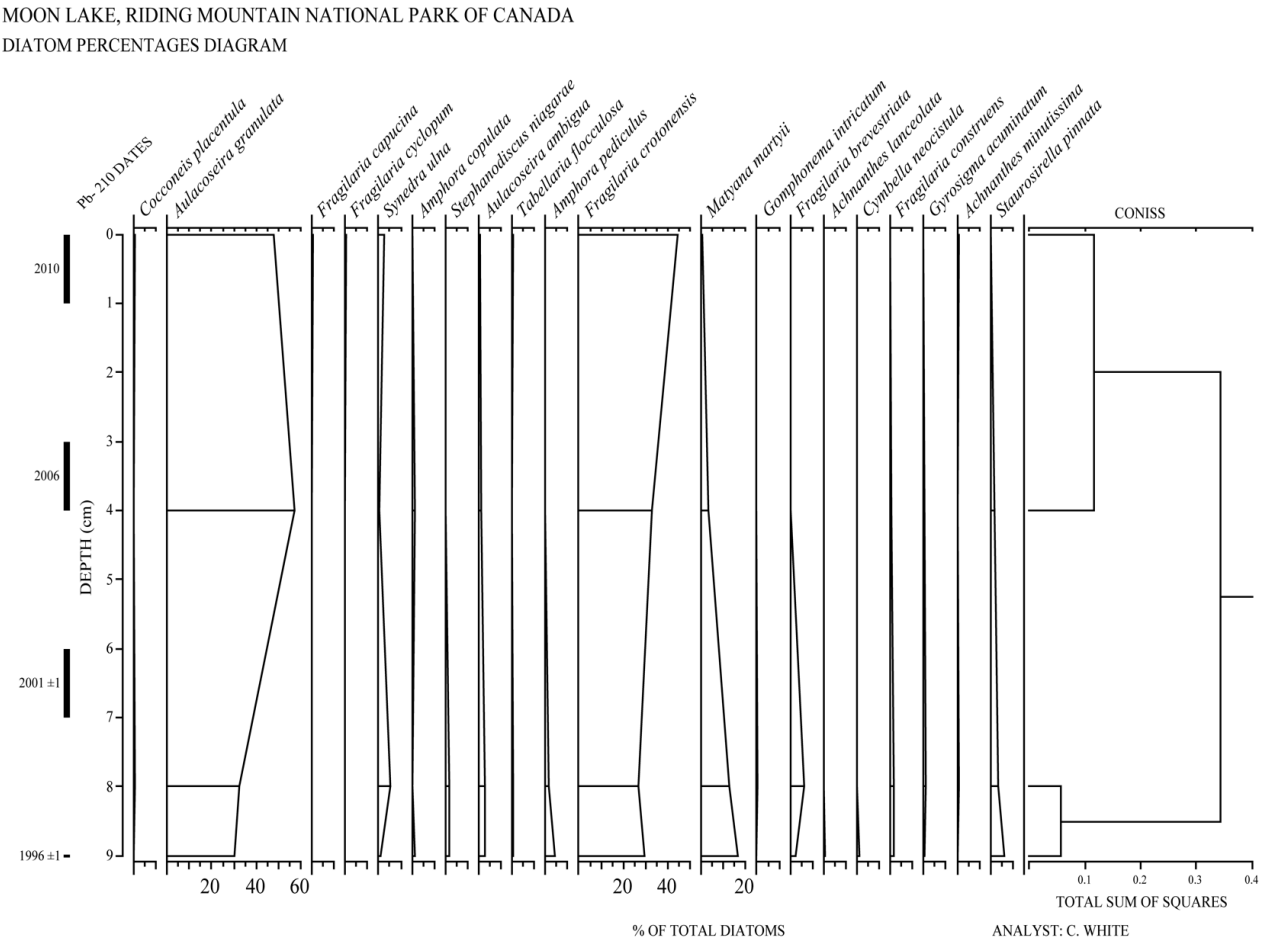


Figure A2.5: Diatom stratigraphy of the most important and abundant taxa from Moon Lake arranged according to calculated weighted averaging bootstrapped TP optima and grouped into corresponding trophic groups. CONISS sum of squares analysis on the far right of the diagram.

Moon Lake was excluded from further discussion and analysis due to the lack of diatoms in the sediments. Preliminary reconstruction of TP from the DI-TP model developed in Chapter 3 of this thesis was conducted and does not appear to reveal any useful information due to the poor sample resolution (Figure A2.6).

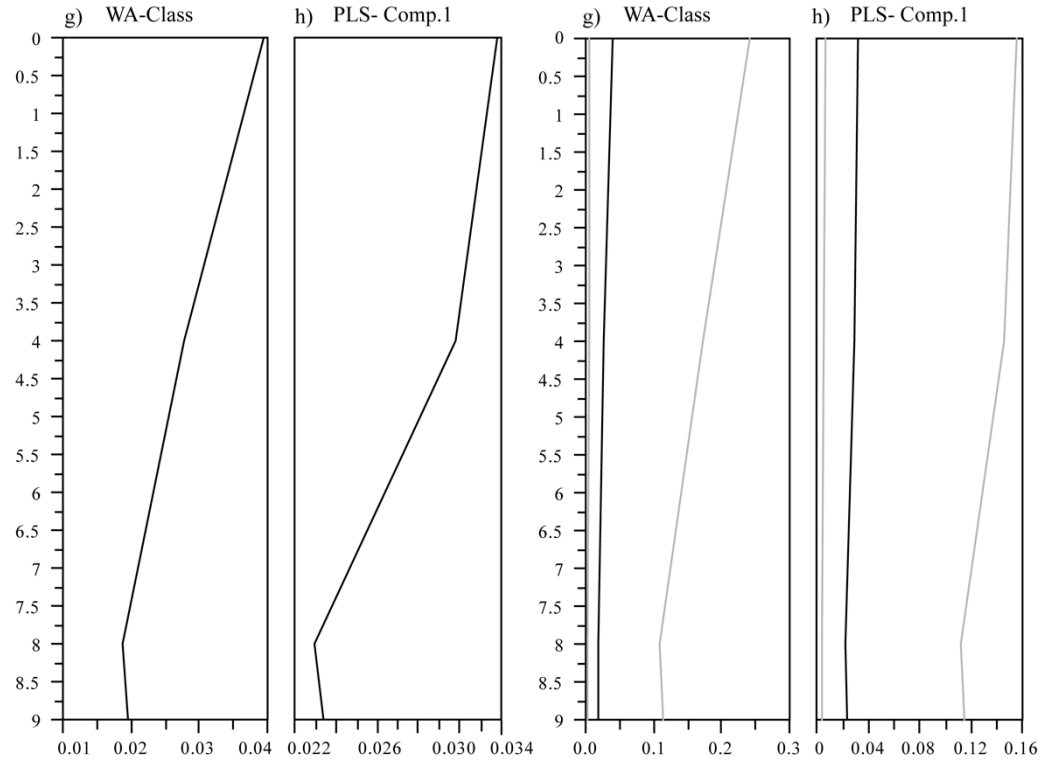


Figure A2.6: Diatom Inferred Total Phosphorus (mg L^{-1}) Moon Lake comparing Weighted Averaging (classical deshrinking) modelling techniques (a,c) and Partial Least Squares (first component) modelling techniques (b,d,) with (c,d) and without (a,d) error bars.

A2.4: Surface sediment assemblages in relation macrophyte populations

Relating the diatom communities to the presence or absence of macrophyte populations is an area that required further analysis in the RMNP dataset. The change in the abundance of macrophytes and benthic/planktonic diatom species is useful in terms of lake management. The relationship between trophic state and the alternating stable state of macrophyte populations with planktonic diatom populations has been well explored (Timms & Moss 1984; Scheffer 1989).

Diatoms can be employed indirectly to reconstruct change in macrophyte vs. plankton dominance. TP levels can have an impact on the macrophyte populations, with heightened levels of TP generally associated with the disappearance or reduction in macrophyte populations, in previously macrophyte dominant aquatic systems (Jeppesen et al. 1990). This is related to a shift from benthic or epiphytic diatom populations to one where planktonic species are prevalent. Such a shift was noted by Scott & Kling (2006) in the South Lake system.

Diatoms assemblages may also be used in a direct fashion to quantitatively reconstruct changes in macrophyte cover (Vermaire & Gregory-Eaves, 2008). This is useful as fossils of macrophyte specimens may be rare and unevenly distributed (Vermaire & Gregory-Eaves 2008).

In the RMNP dataset two CCA ordinations were performed to assess the relationship between macrophyte species and diatom species. One in which all macrophyte species were included as environmental variables (Figure A2.7), and a second where only those significant were included (Figure A2.8)

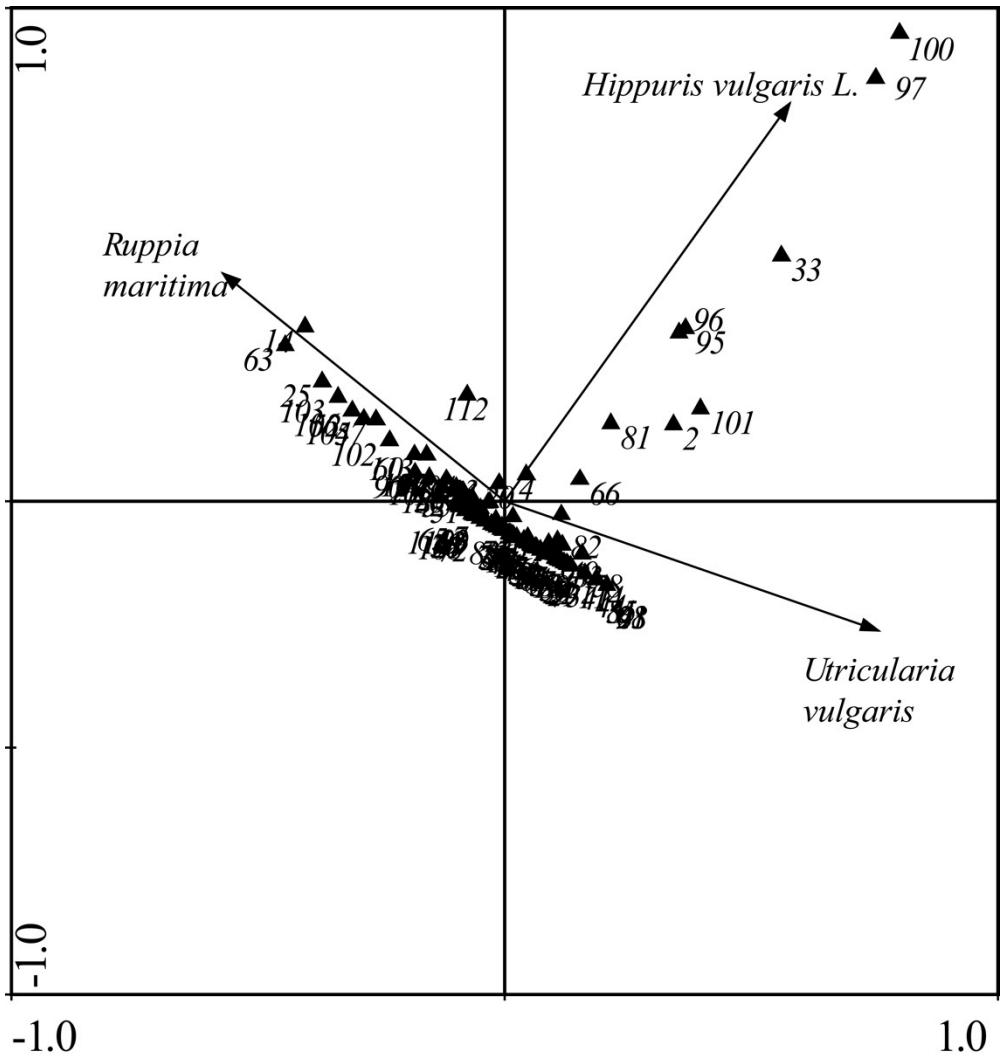


Figure A2.7: CCA ordination diagram of diatoms and macrophytes, including signficant macrophyte species and all diatom species, with macrophytes included in analysis as environmental variables.

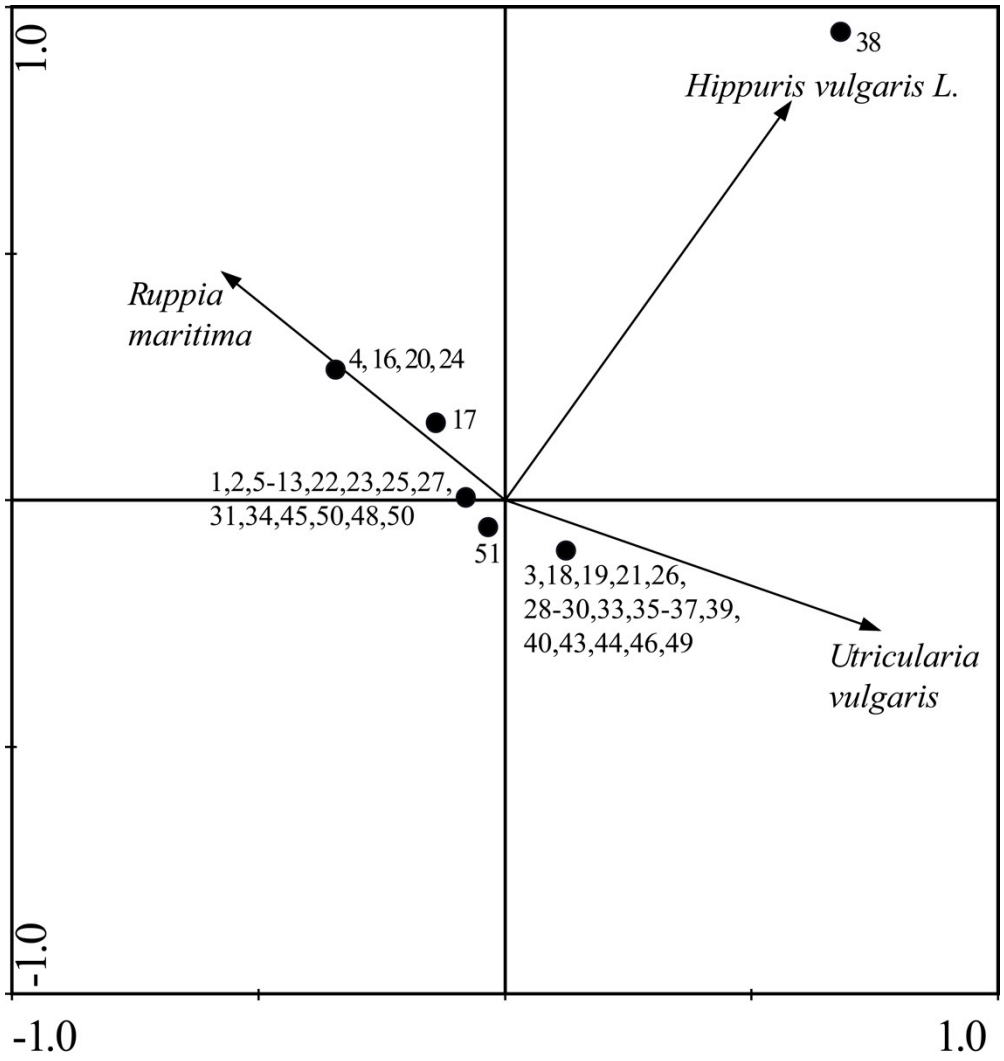


Figure A2.8: CCA ordination diagram of sample sites and macrophytes, including signficant macrophyte species and all sites sampled, with macrophytes included in analysis as environmental variables.