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EARLY FRESHWATER DIATOMS FROM THE UPPER CRETACEOUS BATTLE FORMATION IN WESTERN CANADA

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ABSTRACT: Despite the rise of marine diatoms in the world's oceans throughout the Cretaceous, only a handful of fossil localities worldwide detail invasion of freshwater habitats by diatoms commencing in the Late Cretaceous. We report on the occurrence of numerous freshwater diatom specimens and species from the Battle Formation, an extensive freshwater locality in western Canada that formed in the Late Cretaceous approximately 66.5 million years ago (Ma). The formation represents one of the oldest known localities worldwide harboring definitive remains of freshwater diatoms, contains the oldest known freshwater specimens of the centric diatom genus *Aulacoseira*, and confirms that these early *Aulacoseira* colonizers formed filaments linked together with interdigitating spines. We further document a high diversity of araphid pennate diatoms belonging to the order Fragilariales. Seven pennate morphotypes were uncovered, six of which definitively lacked a raphe and whose closest modern relatives are in the genera *Fragilariforma*, *Fragilaria*, and *Stauroforma*. Given the extensive coverage of the Battle Formation, it is possible that it represents a network of numerous smaller shallow waterbodies that collectively offered a diversity of environments for colonization, making it a unique deposit for examination of early freshwater diatoms.

INTRODUCTION

Diatoms are photosynthetic, unicellular organisms with siliceous cell walls that can dominate planktonic, periphytic, and benthic habitats in aquatic systems worldwide (Round et al. 1990; Medlin et al. 2000). High cell abundances, often coupled with impressive species diversity, make diatoms integral elements of aquatic food chains and significant components of biogeochemical cycles in both freshwater and marine systems (Field et al. 1998; Armbrust et al. 2004; Katz et al. 2005). Diatoms form a cornerstone of paleobiological research and have contributed immensely to our understanding of environmental issues such as lake-water acidification, eutrophication, and climate change (Smol and Stoermer 2010).

Current estimates yield a Mesozoic origin for the diatoms and since that time period the group has become one of the most successful protistan lineages in recent geologic history (Kooistra and Medlin 1996; Sims et al. 2006; Medlin 2016). Their radiation in the world's oceans commenced in the early Cretaceous (ca. 145 Ma) and despite the rapid and prolific rise of marine diatoms throughout the Cretaceous (Harwood and Nikolaev 1995), invasion of freshwater habitats, presumably from marine environments, appears to have occurred later near the end of the Cretaceous (Chacón-Baca et al. 2002; Ambwani et al. 2003; Singh et al. 2006; Buatois et al. 2016). Further, based on examination of numerous fossil sites, diatoms do not begin to establish a prominence in freshwater environments until at least the middle (Strelnikova and Lastivka 1999; Siver and Wolfe 2009) to late (Lohman and Andrews 1968; Benson and Kociolek 2012) Eocene. Significant diversification of the group does not occur in North America until the Miocene (Bradbury and Krebs 1995), possibly correlated with expansion of grasslands (Kidder and Gierlowski-Kordesch 2005). Despite recent discoveries, the fossil record surrounding the early invasion of freshwater habitats by diatoms, including the initial colonizing taxa, is poor.

The Battle Formation represents an extensive freshwater environment that was deposited across much of southern Alberta and into southwestern Saskatchewan, Canada, during the Late Cretaceous, approximately 66.5 Ma (Binda 1970; Picard and High 1972; Srivastava and Binda 1984). The lake represented by the Battle Fm. is one of the largest lakes in the geologic record formed, in part, due to reduced drainage caused by a gentle tectonic uplifting to the east (Binda 1970; Picard and High 1972). Whether the environment was one contiguous shallow water body, or a series of interconnected smaller lakes, marshes and bogs is unknown. Remains of freshwater lycopods, ferns, chrysophyte cysts and sponge gemmoscleres were previously documented by Srivastava and Binda (1984). The purpose of this paper is to report on the occurrence of freshwater diatoms from the Battle Fm. Species representing centric diatoms, araphid pennate diatoms, and a possible raphe-bearing form, have been uncovered and are described using both light and scanning electron microscopy. Early colonization of freshwater environments by siliceous algae is discussed.

SITE DESCRIPTION

The Battle Fm. outcrops in the southern part of the provinces of Alberta and Saskatchewan (Canada), and is stratigraphically located immediately above the Maastrichtian Whitemud Fm. and below the Maastrichtian–Paleocene Scollard and Ravenscrag formations of Alberta (AB) and Saskatchewan (SK), respectively (Binda 1970; Lerbekmo et al. 1987). The Battle Fm. stretches from just south of Edmonton, AB, in a northwest to southeast direction to the Cypress Hills region of southwestern SK, outcropping extensively in Red Deer, Drumheller and Cypress Hills (Fig.1A, 1B). The formation is a 3–12 m thick, massive, dark brown claystone deposit (Irish 1970; Gibson 1977) situated approximately 50 m below the K–Pg boundary (Binda 1970). A thin silicified tuffaceous layer, known as the Kneehills Tuff, was deposited in the upper portion of the Battle Fm. and used to estimate the age of the formation based on K/Ar
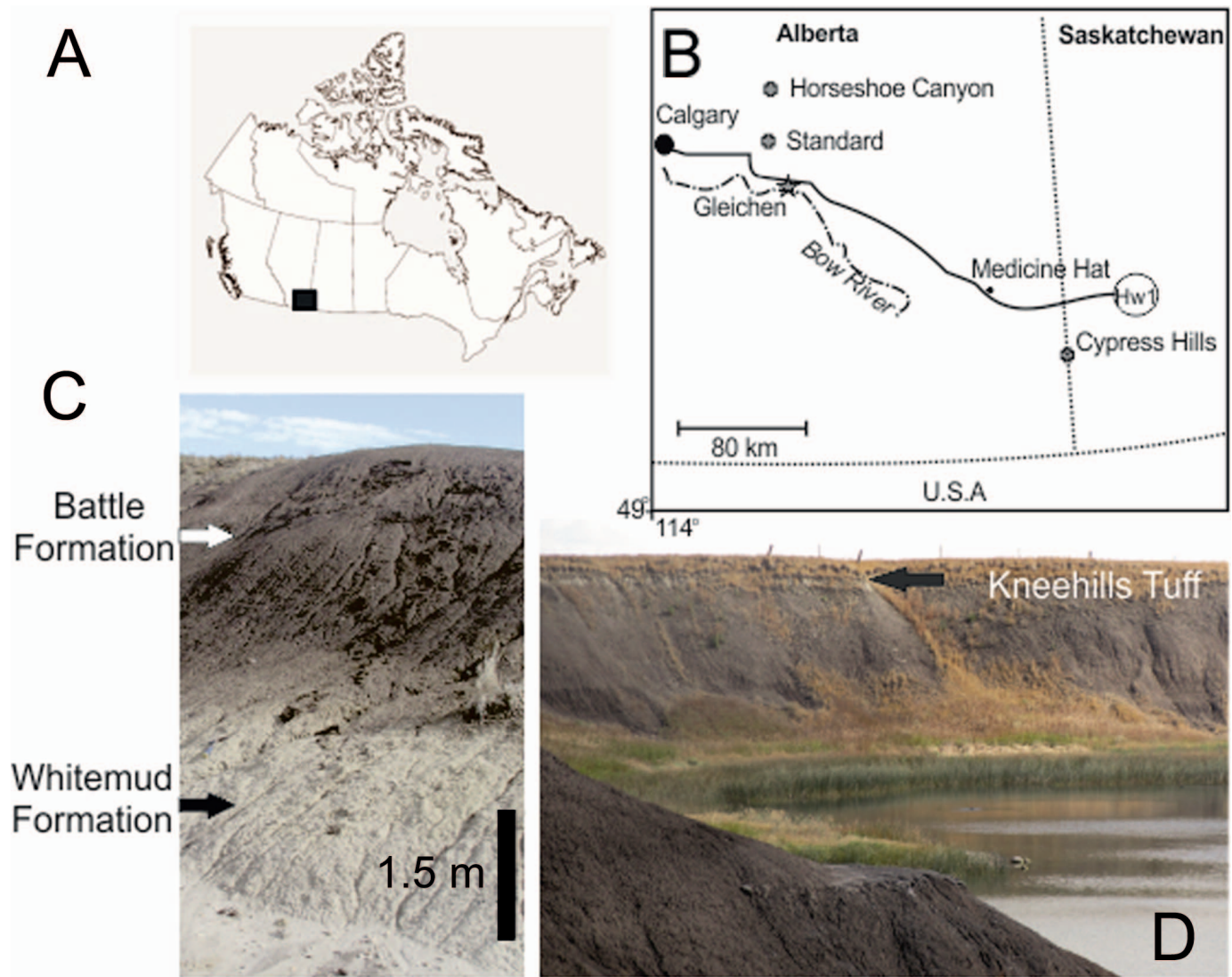
FIG. 1.—Details of the study location. **A, B**) Geographic location of the Gleichen locality relative to other known areas (Horseshoe Canyon, Standard and Cypress Hills) where outcrops of the Battle Formation exist. **C**) The Gleichen site showing the underlying Whitemud Fm. and the overlying Battle Fm. Note the chromatic contrast between the two units. **D**) The Kneehills Tuff layer capping the Battle Fm. (arrow).

dating of sanidine and biotite minerals (Allan and Sanderson 1945; Lerbekmo et al. 1987). The chromatic contrast between the Battle and Whitemud formations is striking (Fig. 1C).

North of the town of Gleichen (AB), on Range Road 231, adjacent to the Hamlet of Gleichen water reservoir (50°52′49.7″N/113°04′21″W) there is an outcrop displaying 3 m of Battle Fm. (Fig. 1B). This site, referred to as GP, was sampled in September of 2017. The contact with the underlying Whitemud Fm. is visible west of the reservoir (Fig. 1C); to the east, the outcrop is capped by a continuous indurated layer of the Kneehills Tuff (Fig. 1D). The 3 m deep outcrop was systematically sampled every 20 cm starting at the top of the Whitemud Fm. and labeled as GP1, GP2, etc. through to the top of the Battle Fm. A minimum of 50 gm of sediments was taken from cleaned surfaces at each interval. Sediments are composed of shale displaying the typical fracture of the formation reported in Binda (1970) with varying degrees of organic content, from highly organic, carbonaceous roots at the bottom, to coaly shale. Shale is intermixed with light colored silt in the top two samples.

MATERIALS AND METHODS

Mudstone samples from the Gleichen Pond locality were prepared for microscopy and examined for microfossil remains using light (LM) and scanning electron microscopy (SEM). Although all samples contained microfossil remains and were examined as part of this project, samples GP4, GP5, GP6, and GP7, representing approximately 80 cm of the strata, yielded the highest number of diatom specimens.

Samples for LM observation were prepared by oxidizing 100–500 mg of mudstone fragments with 30% H_2O_2 under low heat for a minimum of an hour. Two different cleaning procedures were used, both yielding similar results. In one case, 300 ml of distilled water was added to the oxidized slurry and left at room temperature for 24 hours to settle. Two-hundred ml were then discarded leaving a 100 ml sample. In the second procedure, the oxidized slurry was washed five times with distilled water using repeated centrifugation and the final pellet diluted to 15 ml and stored in a glass vial. Aliquots of each preparation were air dried onto glass coverslips, mounted onto glass slides with Naphrax, and observed with either an

Olympus CX10 coupled with an Infinity 1-2C camera, or a Leica DMR coupled with a Zeiss Axiocam 506 color camera.

For SEM observation, an aliquot of each slurry was air dried onto a piece of heavy duty aluminum foil. The aluminum foil samples were trimmed and attached to aluminum stubs with Apiezon® wax. Samples were coated with a mixture of gold and palladium for two minutes with a Polaron Model E sputter coater, and examined with an FEI Nova NanoSEM 450 FESEM. Measurements of specimens were taken directly from electron micrographs.

RESULTS

Numerous diatom specimens were uncovered from the Battle Fm. at the GP site, with especially high numbers from GP6 and GP7. The specimens can be divided into three groups: centric forms representing the genus *Aulacoseira* (Figs. 2, 3), small pennate specimens assigned to the order Fragilariales, and a few pennate specimens of unknown taxonomic affinity (Figs. 4, 5).

Specimens Assigned to the Genus Aulacoseira

Specimens of a species belonging to the genus *Aulacoseira* were common. Single valves, whole frustules, valves from adjacent frustules linked together, and short filaments of two to three cells were uncovered (n > 300 specimens) (Figs. 2A–2H, 3A, 3B). Valves were cylindrical, ranged in diameter from 4–15 μm with a mean of 7.7 μm, and in mantle height from 6–12 μm (n = 25). The length to width ratio ranged from 0.75–2 with a mean of 1.3. Mantle striae were straight and composed of circular to slightly elongate areolae (Figs. 3A, 3B, 3D). The mantle areolae were usually, but not always, aligned between adjacent striae. The collum was 1.0–1.5 μm wide, and consisted of a series of short, closely spaced, parallel ribs (Fig. 3B, white arrows). The valve face appeared to be solid and lacked areolae (Fig. 3E). A series of short, evenly spaced spines lined the valve margin (Figs. 2D (arrow), 3A, 3C–3E). The spines were pointed, with a mean length of 0.5 μm, and on most specimens positioned on each costae. On a few specimens the spines were spaced more widely apart, slightly wider, and possibly on every other costae; these specimens often had wider areolae, a larger length:width ratio, and may represent a second species. No other spine type was observed. A shallow ringleiste was present (Figs. 2 (longer double black arrows), 3F). Given the condition of the specimens, and the fact that most valves were filled with sediment, we were unable to determine the presence and position of rimoportulae.

Remains of Pennate Diatoms

Compared to the majority of early freshwater fossil localities, the Battle Fm. samples from Gleichen Pond had a relatively high diversity of pennate diatoms, with the majority being small forms representing the order Fragilariales (Figs. 4, 5). Seven morphotypes were included in this group, including five small forms that lacked a raphe and were appropriately placed in order Fragilariales. A sixth small morphotype also had a shape that resembled taxa belonging to order Fragilariales, but on some specimens a thin line within the sternum was observed that may represent a raphe. The seventh morphotype was larger, lacked a raphe, and may also be related to taxa within order Fragilariales.

Morphotype #1 (Figs. 4I–4N, 5F) was the most common pennate taxon with numerous partial valves (n >100) observed with both LM and SEM, especially in samples GP6 and GP7. Valves were linear to slightly elliptical with size reduction, narrow, with more or less parallel margins in the center of the valve, becoming attenuated with acutely rounded-rostrate apices. Striae were parallel and often unevenly spaced. The axial area was narrow and often difficult to discern. Smaller and more elliptical specimens (Figs. 4I, 4J) may represent a separate taxon. Valves ranged in size from 16–34 ×

3.5–5 μm (n = 20). Valve shape resembled species in the genera *Fragilaria*, *Fragilariforma*, or *Stauroforma*.

Morphotype #2 (Figs. 4A–4C, 5C), also a common form in samples GP5–GP7 with over 50 specimens observed, had linear valves with wavy margins and attenuated to slightly rostrate apices. One valve type was slightly constricted in the center with biundulate margins (Fig. 5C), while others were slightly swollen in the center forming triundulate margins (Fig. 4A–4C). Striae were mostly parallel and the axial area was narrow or unresolved. Valves ranged in size from 15–23 × 4.5–6 μm (n = 15). Valve shape resembled varieties within the *Fragilariforma constricta* complex.

Morphotype #3 (Figs. 4E, 4F, 5A) consisted of small valves, ranging in size from 14–20 × 2.5–4.2 μm (n = 20), with parallel margins and protracted and capitate apices. Details of the striae could not be resolved. Valve shape resembled those of *Fragilaria virescens* var. *capitata*, and related taxa.

Morphotype #4 (Fig. 4G, 4H) had small elliptical-shaped valves ranging in size from 15–19 × 4.6–5.2 μm (n = 14), and with distinctly capitate apices. Except for the elliptical valve shape, morphotype #4 is similar to morphotype #3. Details of the striae could not be resolved. Valve shape resembled those of *Fragilariforma marylandica*.

Morphotype #5 (Fig. 5B) was a rare taxon with long, narrow, linear valves and rounded apices. Valve margins were largely parallel over most of the valve. Only four whole specimens were observed, ranging in size from 38–44 × 5.5–6.3 μm. This shape could fit many species, especially ones within *Fragilaria* and smaller taxa within *Synedra/Ulnaria*.

Morphotype #6 (Figs. 4D, 5D) had linear valves with wavy margins and rostrate to slightly capitate apices. Four specimens of this morphotype were found. Valve shape was similar to that of morphotype #2, but specimens had a linear structure, possibly a raphe, along the sternum. Based on SEM observations, the striae were closely spaced, unevenly positioned, with relatively large areolae and possibly several small spines on the apices. We could not definitively resolve the presence or absence of a raphe and these specimens could belong to morphotype #2.

Morphotype #7 (Fig. 5E) was a rare taxon with only fractured valves recovered. Valves were centrally swollen, tapered to rounded apices, and estimated to be approximately 45–50 μm long (n = 3). Striae were short, restricted to the margins, consisted of only 2–3 areolae, and continued onto the mantle. The axial region was wide, especially in the center region, and specimens lacked a raphe.

DISCUSSION

In a review of fossil diatom records, Bradbury and Krebs (1995) reported the earliest non marine diatoms were from the Eocene, including the Dewey Beds of Idaho (45–50 Ma) (Sims et al. 2006). Subsequent investigations have pushed back this important marker by about 20 Ma to the latest Cretaceous (Chacon-Baca et al. 2002; Ambwani et al. 2003; Beraldi-Campesi et al. 2004; Singh et al. 2006), and possibly much further back to the Permian at 294–299 Ma if findings from Farooqui et al. (2015) prove true. In addition, other fossil localities bearing numerous freshwater diatoms from the Paleocene (Siver et al. 2016) and Eocene (e.g., Bullwinkle and Reigel 2001; Wolfe et al. 2006; Wolfe and Siver 2009; Siver et al. 2010; Benson and Kociolek 2012) have added significantly to our knowledge of early freshwater diatom communities. Ambwani et al. (2003) reported specimens of *Aulacoseira* from the Late Cretaceous Deccan Intertrappean beds (India) that they state as "slightly older" than 65 Ma. Although the specimens are in poor condition and the SEM images are questionable, the light micrographs do present characters of *Aulacoseira*. A later study by Singh et al. (2006), also based on samples from the Deccan Intertrappean beds, reported raphe-bearing freshwater diatoms found within tests of *Thecamoeba*. In this case, the taxa appear to represent species from the genera *Nitzschia* and *Planothidium*. Slightly older remains of freshwater diatoms in carbonaceous cherts from the Upper
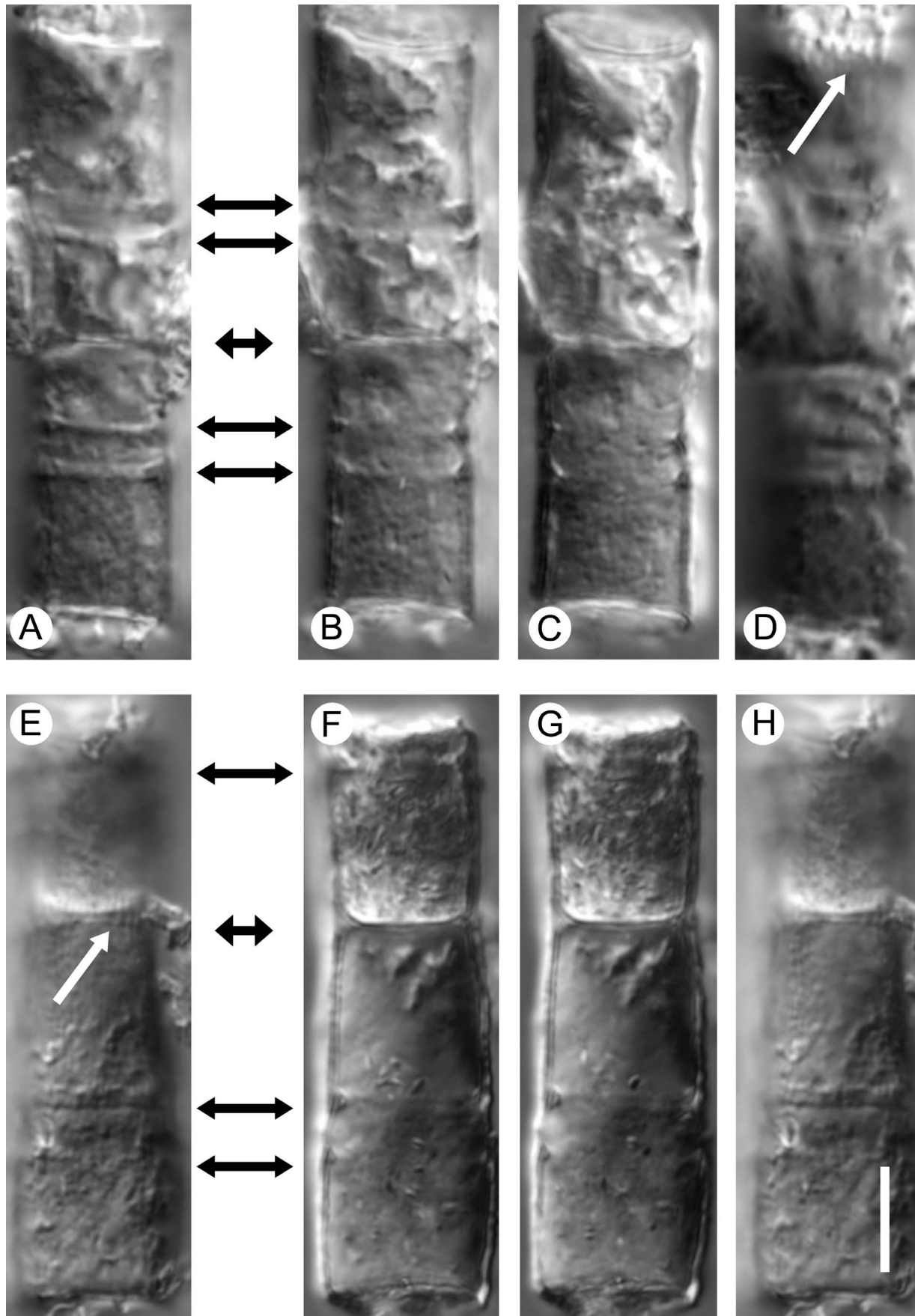
Cretaceous Tarahumara Formation in Mexico were reported by Chacón-Baca et al. (2002) and Beraldi-Campesi et al. (2004). The specimens illustrated from the Tarahumara sediments, which dated to ca. 70 Ma, indeed appear to represent diatoms based on form, shape and size, but the lack of resolution precludes positive taxonomic assignment (Sims et al. 2006). Sims et al. (2006) further questioned the age of the Tarahumara Fm. and whether the specimens were indeed indicative of a freshwater environment. However, the strata bearing the Tarahumara specimens were situated between rhyolitic tuffs dated using U-Pb zircon (Chacón-Baca et al. 2002), a fairly reliable dating method. Our findings from the Battle Fm., dating to 66.5 Ma, give definitive evidence that diatoms had invaded freshwater environments by the Late Cretaceous, supporting findings based on specimens of similar age from the Deccan Intertrappean beds and the Tarahumara Fm.

Farooqui et al. (2015) reported well-preserved pennate fossil diatoms as xenosomes within thecamoebian tests, presumably in ancient Permian sediments from the Chamba basin, India (251–299 Ma). Remarkably, the well-preserved specimens appear quite modern and some are said to have affinities with the raphe-bearing genus *Nitzschia*. The Farooqui et al. (2015) findings, however, do not align with numerous studies regarding diatom evolution. First, the oldest known and undisputable diatom fossils are marine specimens from the early Cretaceous (ca. 140 Ma; Harwood et al. 2007). An older record was reported by Rothpletz (1896) from the Early Jurassic (ca. 190 Ma), but this remains unverified (Brown and Sorhannus 2010). Thus, the findings reported by Farooqui et al. (2015) are close to, or over, 100 Ma older than all other known records. Second, whereas the early Cretaceous specimens presented by Harwood et al. (2007) represent largely extinct lineages and forms distinct from modern diatoms, the much older ones illustrated by Farooqui et al. (2015) resemble modern taxa. This is of further concern since Farooqui et al. (2015) believe their specimens represent the modern genus *Nitzschia*, a highly derived raphe bearing form. Third, the Permian specimens do not align with molecular-based phylogenies that place the origin of diatoms at ca. 240–250 Ma in the Middle Triassic (Kooistra and Medlin 1996; Medlin et al. 2000; Sims et al. 2006; Medlin 2016). If the Permian specimens prove true, especially if they represent derived raphid forms, then the origin of the diatoms far surpasses the Middle Triassic as reviewed by Medlin (2016).

Many of the earliest freshwater diatom specimens from Upper Cretaceous localities are poorly preserved and specific details often hard to resolve. Rows of areolae on the mantle, short spines along the valve face margin, and a ringleist can be discerned on specimens from the Deccan Intertrappean beds with light microscopy (Ambwani et al. 2003), supporting placement in *Aulacoseira*. However, in our opinion, the SEM images presented are likely not of *Aulacoseira*, and specific details of the valve face, collum and presence of rimoportulae were not resolved. The specimens from the Upper Cretaceous Tarahumara Fm. illustrated by Chacón-Baca et al. (2002) and Beraldi-Campesi et al. (2004) are clearly diatoms, but valve ornamentation is not resolved making generic placement tenuous (Sims et al. 2006). For example, girdle bands with septa, a hallmark of the genus *Tabellaria*, are not resolved on specimens described as *Tabellaria sonorensis* (Beraldi-Campesi et al. 2004), and it is equally possible that these specimens represent another member of the Fragilariaceae. Likewise, specimens potentially assigned to the raphe-bearing genus *Amphora* (Chacón-Baca et al. 2002, figs. E–G) are questionable, especially since the presence of a raphe was difficult to resolve and the shape of the specimens also resembled species from other genera such as *Eunotia*. Regardless of the inability to assign the

Tarahumara Fm. specimens to specific taxonomic units, they do represent diatoms.

As is true of other freshwater diatoms reported from the Upper Cretaceous, our specimens from the Battle Fm. are also poorly preserved. Despite preservation issues, the Battle Fm. fossils clearly represent the genus *Aulacoseira*, and members of the araphid order Fragilariales based on size, form, shape and other characters observed with SEM. Since many of the valves and frustules uncovered from the Battle Fm., especially the cylindrical specimens of *Aulacoseira*, are infiltrated with sediment and mineral deposits, ornamentation could not often be resolved with LM. However, unlike previous studies, surface details were resolved with SEM. In the case of the centric specimens found in Battle deposits, a suite of characters can be used to readily place this organism into *Aulacoseira*. The frustules are clearly cylindrical and link together, valve-to-valve, forming chains. In addition, the presence of areolae forming striae on the mantle, spines aligning the margin of the valve face, a distinct collum, and presence of a ringleiste all support placement in *Aulacoseira*.

According to Sims et al. (2006), the earliest pennate forms had a central sternum, uniseriate striae, often with large areolae, and one or two apically positioned rimoportulae per valve. Battle Fm. specimens had uniseriate striae, often consisting of relatively large areolae that continued onto the mantle, and many possessed a central sternum. Other Battle Fm. specimens appeared to have striae continuous across the valve face, lacking a central sternum, and on a few specimens we could not rule out the presence of a raphe. Further, our specimens included ones with pointed, rounded, rostrate and capitate apices, as well as ones with smooth and wavy margins, all common forms found within the order Fragilariales. Based on size and valve shape, the Battle Fm. specimens resemble species in the genera *Fragilariforma*, such as *F. constricta* (Ehrenberg) Williams and Round or *F. marylandica* Edlund, Laub, Siver, Hamilton and Morales, and *Fragilaria*, such as *F. virescens* var. *capitata* Østrup. In many respects, the few broken specimens of morphotype #7 resemble another member of the order Fragilariales, *Ambistria hyalina*, described from late Eocene deposits (see Lohman and Andrews 1968, pl. 1, figs. 21, 29, 30). The overall shape, size, and short marginal striae are similar, suggesting that *Ambistria* may be older than previously thought. Likewise, the araphid species represented by morphotype #6 may also belong to this order. Lastly, although the size and shape of morphotype #5 valves are similar to members of the order Fragilariales, this taxon most likely represents an early freshwater raphe-bearing species. This is especially important because other than the study by Singh et al. (2006) based on samples from the Deccan Intertrappean beds, the oldest known raphid diatom is from Paleocene remains (Sims et al. 2006). Thus, if the structure observed on morphotype #5 is a raphe, this taxon would be the oldest known raphid diatom.

Aulacoseira and members of the Fragilariales (e.g., *Fragilaria*) are often among the most common diatom taxa uncovered from the oldest known freshwater environments represented in the geologic record (Bradbury and Krebs 1995; Ambwani et al. 2003; Sims et al. 2006). Findings from the Battle Fm. not only confirm previous reports, but may represent the oldest such definitive records for freshwater deposits. Species of *Aulacoseira*, the closely related *Eoseira* (Wolfe and Edlund 2006), and *Fideliacyclus* (Siver et al. 2016) are the oldest representatives of centric diatoms found to date in early freshwater localities, with the first definitive record of *Aulacoseira* reported from uppermost Cretaceous deposits (65 Ma) in India (Ambwani et al. 2003). The Battle specimens of *Aulacoseira* are slightly older, but complement those

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Fig. 2.—Light micrograph images of *Aulacoseira* specimens from Upper Cretaceous mudstones of the Battle Fm. Intact remains of two frustules (A–D) and three valves (E–H) imaged at four focal planes. Positions of the ringleiste (long double black arrows), connection between adjoining frustules (short double black arrows), and position of marginal spines (white arrows) are noted. Scale bar = 5μm.
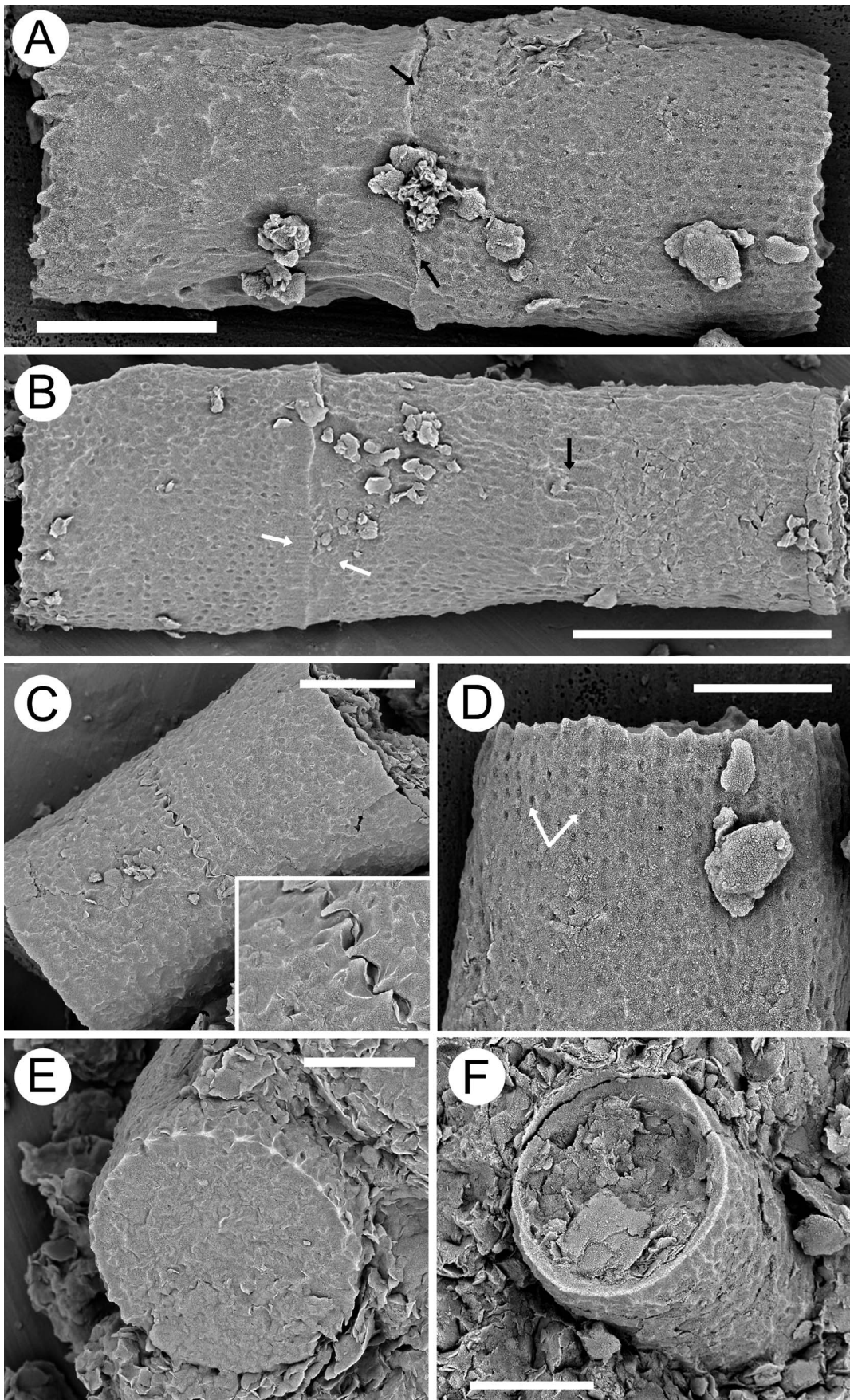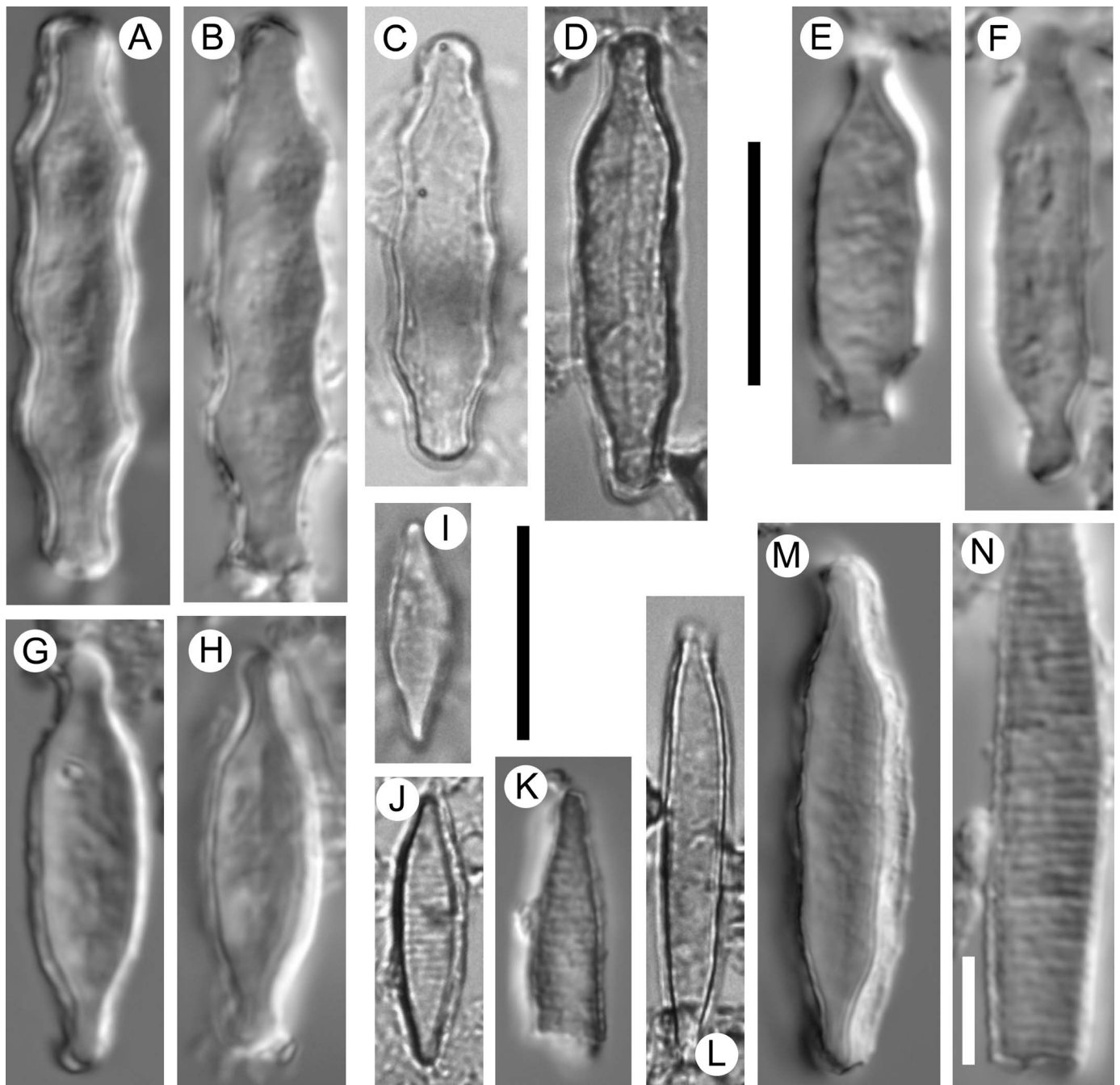
FIG. 4.—Light micrograph images of araphid pennate diatoms representing order Fragilariales from Upper Cretaceous mudstones of the Battle Fm. **A–C**) Morphotype #2. **D**) Morphotype #6. **E, F**) Morphotype #3. **G, H**) Morphotype #4. **I–N**) Morphotype #1. Note the wavy margins (morphotype #2), linear valves with capitates apices (morphotype #3), elliptical valves with capitate apices (morphotype #4), striae on morphotype #1 (J, K, N), and possible raphe on morphotype #6 (D). Top black scale bar = 10 μm (A, B, E, F, G, H, K, M); lower black scale bar = 15 μm (C, D, I J, L); white scale bar = 5 μm (N).

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FIG. 3.—SEM micrographs of *Aulacoseira* specimens from Upper Cretaceous mudstones of the Battle Fm. **A**) Whole frustule showing rings of small marginal spines on both valves, mantle striae and areolae, and connection between the two valves (black arrows). **B**) An intact frustule (left) and adjoining valve (right). Note mantle striae, marginal spines, series of parallel ribs on the collum (white arrows), and region connecting neighboring cells (black arrow). **C**) Marginal spines linking adjoining valves, including close-up view (insert). **D**) Valve displaying mantle striae (white arrows) and short marginal spines. **E**) Valve face view with marginal ring of spines. **F**) Internal view showing the shallow ringleiste. Scale bars = 3 μm (D–F); 5 μm (A, C) and 10 μm (B).
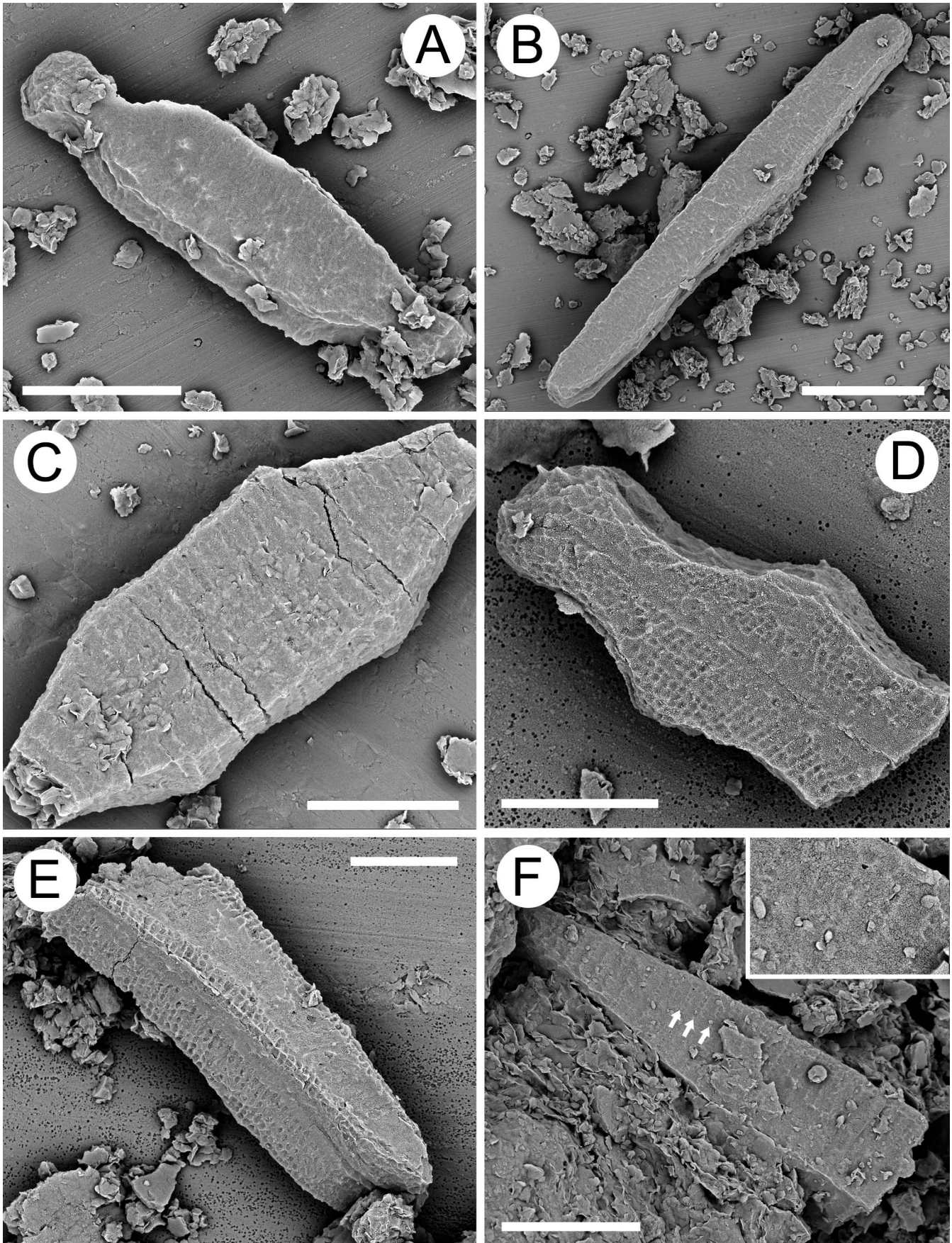
uncovered in the Cretaceous locality from India. Specimens possibly representing *Aulacoseira* were reported in amber from an upper Albian marine locality by Girard et al. (2009), but details of the specimens could not be resolved and the authors list this finding as questionable. Moving forward from these earliest records, *Aulacoseira* specimens were recently reported from the Wombat locality, a Paleocene site estimated at 60 Ma (Siver et al. 2016), and this taxon is well established in many Eocene freshwater deposits, for example Horsefly (44–52 Ma; Wolfe and Edlund 2006), Giraffe Pipe (48 Ma; Wolfe et al. 2006), Eckfeld (44 Ma; Bullwinkel and Riegel 2001; Lutz et al. 2000) and the Beaver Divide fossil assemblage in Wyoming (40–45 Ma, Lohman and Andrews 1968). As is true for the late Upper Cretaceous specimens of *Aulacoseira*, those from the Cenozoic localities also possess characteristics commonly found on modern species.

Eoseira, an extinct close relative of *Aulacoseira*, is another member of the family Aulacoseiraceae found in early Cenozoic deposits (Wolfe and Edlund 2006). Large populations were first described from the Eocene Horsefly locality (Wolfe and Edlund 2006), and this taxon has since also been reported from the slightly older Wombat locality (Siver et al. 2016). This diatom genus bears valves with large spines (megaspines) positioned in recessed grooves that lack a geometric relationship with respect to mantle striae. These features differ significantly from fossil and modern *Aulacoseira*, but resemble features found on the extinct marine diatom *Archepyrgus melosiroides* (Gersonde and Harwood 1990). *Archepyrgus* is an Early Cretaceous diatom that formed short filaments held together with large tetrahedral spines that, as is the case for *Eoseira*, show no apparent geometric relationship with respect to mantle striae. The fact that *Archepyrgus* shares characteristics with *Eoseira* and *Aulacoseira*, including interdigitizing spines used to link cells together, mantle striae, and presence of a ringleiste, has led some researchers (Gersonde and Harwood 1990; Sims et al. 2006; Wolfe and Edlund 2006; Farooqui et al. 2015) to postulate that *Archepyrgus* represents a distant relative, and possible basal member of the family Aulacoseiraceae.

The fact that specimens of the order Fragilariales were common members of the freshwater community represented in the Battle Fm. is not surprising given that araphid pennate diatoms have been uncovered in early freshwater environments. Our findings clearly support those reported by Beraldi-Campesi et al. (2004), also based on Upper Cretaceous sediments, making the Battle Formation among the oldest known deposits with freshwater araphid pennate diatoms. Sims et al. (2006) stated that the appearance of marine pennate species at ca. 75 Ma in the Upper Cretaceous signaled a significant change in diatom evolution and community structure of marine habitats. These early marine araphid forms most likely provided the seed source for early invasion of freshwater environments in the Late Cretaceous. Still, given the closeness in age of the marine and freshwater fossils, a freshwater origin for pennate diatoms can't be ruled out. Regardless, members of the order Fragilariales become increasingly more diverse after the K–Pg boundary, often forming abundant populations in many freshwater localities by the Eocene and into the Oligocene in North America (Lohman and Andrews 1968; Bradbury and Krebs 1995; Benson and Kociolek 2012) and Europe (Lupkina and Dolmatova 1975).

It is well known that the K–Pg boundary is associated with mass extinction caused by a large asteroid strike on the Yucatán Peninsula in Mexico. This event resulted in extinction of some marine diatom genera and an overall decline in diversity, but the level of extinction was not as great as observed with other groups of marine organisms (Katz et al. 2005). On the other hand, virtually nothing is known about the impact of the asteroid on freshwater diatom communities. Interestingly, most of the

deposits harboring early freshwater diatom records date to time periods close to the asteroid strike at ca. 66 Ma (Renne et al. 2013). Both the Tarahumara Fm. (ca. 70 Ma) and the Battle Fm. (66.5 Ma) provide data that predates the K–Pg boundary. In fact, given that the Battle Fm. is situated just below the K–Pg boundary (Binda 1970), it provides an especially valuable benchmark for examination of the pre-impact conditions. However, to fully evaluate effects the asteroid impact had on freshwater diatoms, additional fossil records are needed on both sides of the K–Pg boundary.

In conclusion, the Battle Fm. represents one of the oldest freshwater localities worldwide bearing diatoms, is the oldest locality harboring *Aulacoseira*, and is certainly one of the oldest, if not the oldest, site containing members of the Fragilariales. In addition, it may harbor the oldest known raphe-bearing diatom. The Battle Fm. is extensive, spanning much of southern Alberta and part of southern Saskatchewan, Canada. Further, it was likely a large remnant or a series of numerous smaller waterbodies left behind as the Western Interior Seaway regressed from central North America at the end of the Cretaceous (Binda 1970; Picard and High 1972). Since freshwater diatom floras are believed to have been derived from marine taxa able to make the transition across the salt-to-freshwater continuum (Sims et al. 2006; Wolfe and Edlund 2006), the extensive Battle Fm. represents a logical and potentially important deposit for further examination of the early evolution of diatoms in freshwater habitats.

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FIG. 5.—SEM micrographs of pennate diatoms representing the order Fragilariales from Upper Cretaceous mudstones of the Battle Fm. **A)** Morphotype #3. **B)** Morphotype #5. **C)** Morphotype #2. **D)** Morphotype #6. Note the striae and possible raphe slit. **E)** Morphotype #7. **F)** Morphotype #1. Insert = close-up of striae (white arrows). Scale bars = 4 μm (C, D), 5 μm (A, E, F) and 10 μm (B).

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