Microbially induced pseudotraces from a Pantanal soda lake, Brazil: Alternative interpretations for Ediacaran simple trails and their limits

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ABSTRACT

Despite the absence of metazoans, structures resembling animal traces are common in a soda lake from the western Brazil Pantanal wetland. Pantanal soda lakes are ecologically extreme environments that preclude complex life, allowing extremophiles to flourish. Direct observation indicates that these structures are pseudotraces, representing groove marks that result from the interaction of wave-transported floating rafts of epibenthic microbial mat fragments with the substrate. Variations in wind/wave direction and intensity result in marks of different size and morphology. The most common pseudotraces are simple and slightly curved, narrow grooves (type 1), whereas others are straight and present raised lateral ridges (type 2). Both are V-shaped in cross section. Type 3 comprises long, sinuous, shallow grooves, displaying internal crescentic laminated infill and U-shaped cross section. The similarity of these pseudotraces to Ediacaran structures usually interpreted as animal trace fossils suggests that care should be exercised in their analysis. A set of criteria is proposed to differentiate microbially induced pseudotraces from trace fossils. Analysis of Ediacaran structures needs to be performed on a case-by-case basis, taking into account morphology, orientation, and preservation style of the structure, sedimentary environment, and presence or absence of microbial mats.

INTRODUCTION

The earliest records of complex life are epitomized by Ediacaran macroscopic fossils (635–541 Ma), comprising the Avalon, White Sea, and Nama assemblages (Waggoner, 2003; Narbonne, 2005). The presence of trace fossils in the latter two has been regarded as evidence of late Ediacaran bilaterians (Seilacher, 1989; Jensen, 2003; Buatois and Mángano, 2016). Ediacaran ichnodiversity has been historically overinflated, with many supposed ichnogenera being body fossils or inorganic structures (Seilacher et al., 2005; Jensen et al., 2006; Mángano and Buatois,

2014; Buatois and Mángano, 2016). Alternative interpretations that question the bilaterian origin of some of these structures have been put forward (Jensen et al., 2007; Matz et al., 2008). Experimental studies show that the interaction between microbial aggregates and substrate may generate structures reminiscent of simple trails (Mariotti et al., 2016). Our study documents for the first time the formation of pseudotraces under natural conditions in the field as a result of wind-induced waves dragging microbial flocs on lake margins. The Pantanal wetland (western Brazil) is an unusual landscape composed of small-scale lakes (Assine and Soares, 2004). Some of these are salty and highly alkaline, comprising a physically and geochemically

extreme environment. Extremophiles flourish, covering the lake bottoms and forming microbial mats (see the Supplemental Material[1] for descriptions and methods). Despite the absence of animals (McGlue et al., 2017), especially benthic metazoans, horizontal structures bearing a resemblance to Ediacaran bilaterian trails are common on soda lake margins. The aims of this study were to document the morphology and generation of these pseudotraces and discuss their implications regarding interpretations of similar structures in late Ediacaran strata.

ALKALINE SODA LAKES OF THE BRAZILIAN PANTANAL

The Pantanal Basin (Fig. 1A) comprises a 150,000 km^2 seasonally flooded area, characterized by distributive fluvial systems, alluvial fans, and a trunk river (Assine et al., 2015). The Taquari River forms the Taquari megafan (Fig. 1B), comprising an alluvial area >50,000 km^2. The Nhecolândia region (Fig. 1C) contains >10,000 shallow, mostly elliptical, floodplain lakes, separated by sand ridges, a configuration that results in a complex hydrochemical system, with lakes varying from freshwater to hypersaline, and pH values from 5 to 10 (Bergier et al., 2014). The soda lakes comprise ~10% of the lakes in the study area and are surrounded by sand beaches. Recent studies pointed to the synergy between planktonic and benthic extremophiles and the increase in alkalinity (McGlue et al., 2017). The unusual lake ecology (salinity >4000 ppm and pH > 9) precludes
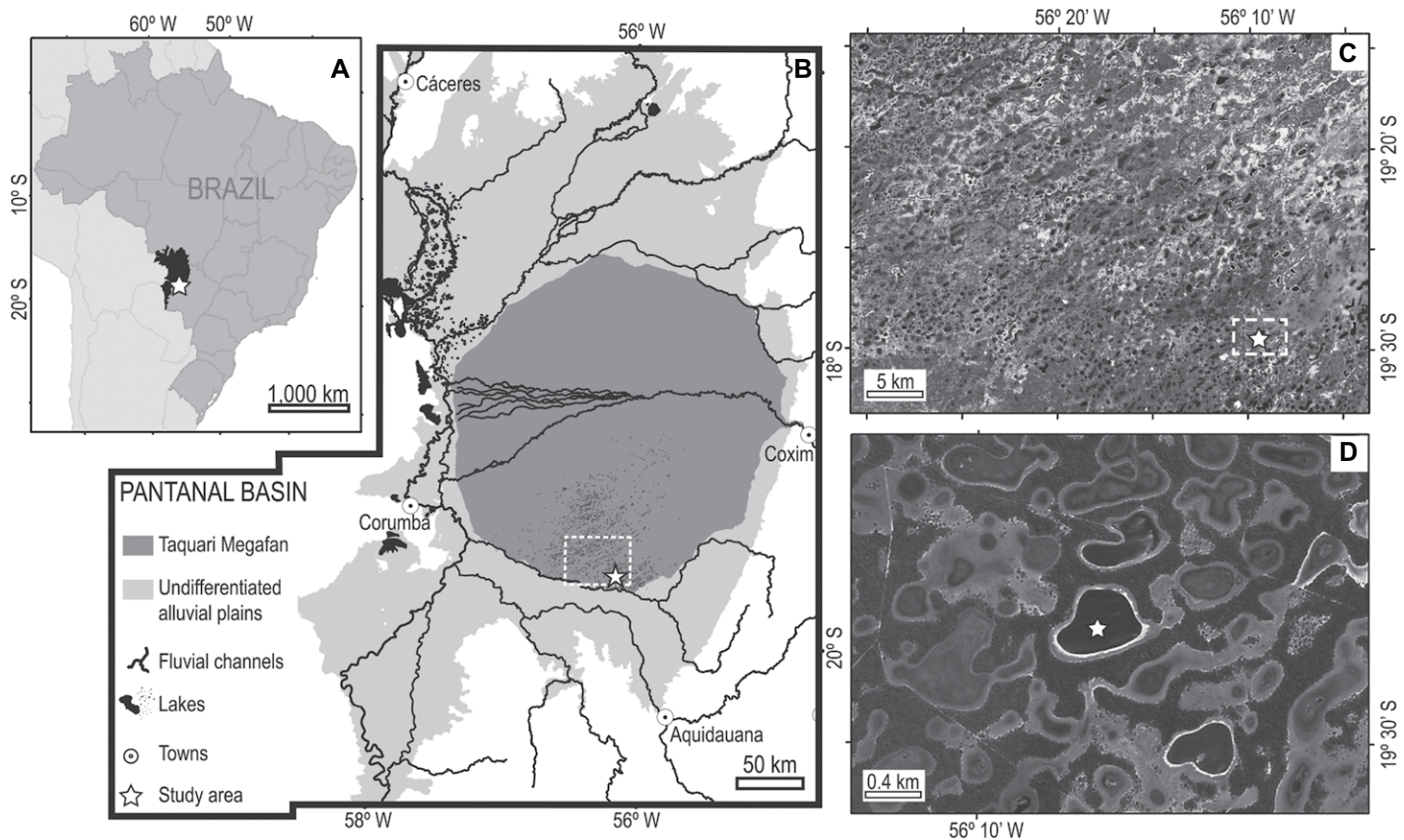
Figure 1. (A) Location of the Pantanal Basin in South America. (B) Pantanal Basin, highlighting the Taquari megafan and Nhecolândia area (white square). (C) Detail of Nhecolândia area (white square in B) showing tens of natural lakes. (D) Detail of square in C showing Burro Branco Lake (white star).

the presence of metazoans, and the sediment is occupied by epibenthic microbial mats and biofilms produced by archaea, viruses, cyanobacteria, and heterotrophic bacteria (Bergier et al., 2014). Occasionally, small alligators may enter these waters, stay shortly, and return to nearby freshwater bodies, without influencing the lake ecosystem.

PSEUDOTRACES FROM PANTANAL SODA LAKES

The pseudotraces described here are from a single lake, Burro Branco, located in southeastern Nhecolândia (see the Supplemental Material). We observed 23 pseudotraces formed on wave-rippled, very fine-grained sandy substrate (Fig. 2A) locally mantled by thin epibenthic microbial mats or bacterial biofilms (Fig. 2B). The density of the pseudotraces was low (<1 per m^2) and depended on the availability of microbial rafts at the water-sediment interface, but may be slightly higher locally (1–3 per m^2; Fig. 2E). Almost all pseudotraces were oriented perpendicular to the lake margin and parallel to the main wind direction; slight variations were related to small irregularities in the substrate and variations in wind direction. The floating rafts were small (0.5–6.4-cm-wide) fragments of epibenthic microbial mats with increased buoyancy due to

gas production. The width range of the pseudotraces was coincident with that of the rafts, and the observed length was 0.5–36 cm. All structures were produced in very shallow water (<1.5 cm). These fragments were detached from the substrate by erosion due to wind-generated waves. The density of these medium-buoyancy microbial rafts was similar to that of the saline lagoon water (~1 g/cm^3). Ribonucleic acid (RNA) sequencing revealed that the mats formed in the Nhecolândia lagoons were mainly constituted by extracellular polymeric substances (EPS); coccoidal, filamentous, and spiraled cyanobacteria; and Archaea (Malone et al., 2012).

Three types of pseudotraces were recognized. Type 1 was the most abundant and comprised simple and dominantly straight, narrow grooves, flanked by small lateral raised ridges (Figs. 2C and 3E). Ornamentation and crescent marks were absent. Type 2 consisted of wide grooves with flat bases flanked by lateral raised ridges. These structures were slightly curved, having linear and parallel erosion marks on the groove (Figs. 2D, 2E, and 3C). Types 1 and 2 are V-shaped in cross section. Type 3 consisted of straight to locally curved grooves with internal crescent marks, the convexity of which was opposite to particle motion, and U-shaped cross section (Figs. 2F and 3A).

The three types occurred on substrates not covered by microbial mats and did not present morphologic variation along their length. Rare marks produced over surfaces protected by biomats were significantly smaller in length (<1.5 cm), penetration depth (<0.5 cm), and width (<0.2 cm), formed at water depth <0.5 cm, and were less complex morphologically.

ORIGIN OF THE PSEUDOTRACES

The three types of pseudotraces are similar to groove or tilting marks (Wetzel, 1999, 2013). They result from the transport of floating rafts of biomat fragments by oscillatory flows (Figs. 2B–2F; see Movies S1–S4 in the Supplemental Material). These objects are subject to transport by wind-generated waves, producing gutters and grooves resulting from raft interaction on a soft sandy substrate. These structures are absent at water depths >10 cm, due to the size of the biomat floating rafts and the relatively low transport capacity of the waves. The morphology is controlled by variations in direction and strength of waves and by the size and shape of the flocs. Differences in morphology are related to (1) variation in wind velocity and direction, (2) how much of the object is touching the substrate, (3) local bottom irregularities, and (4) presence or absence of epibenthic microbial
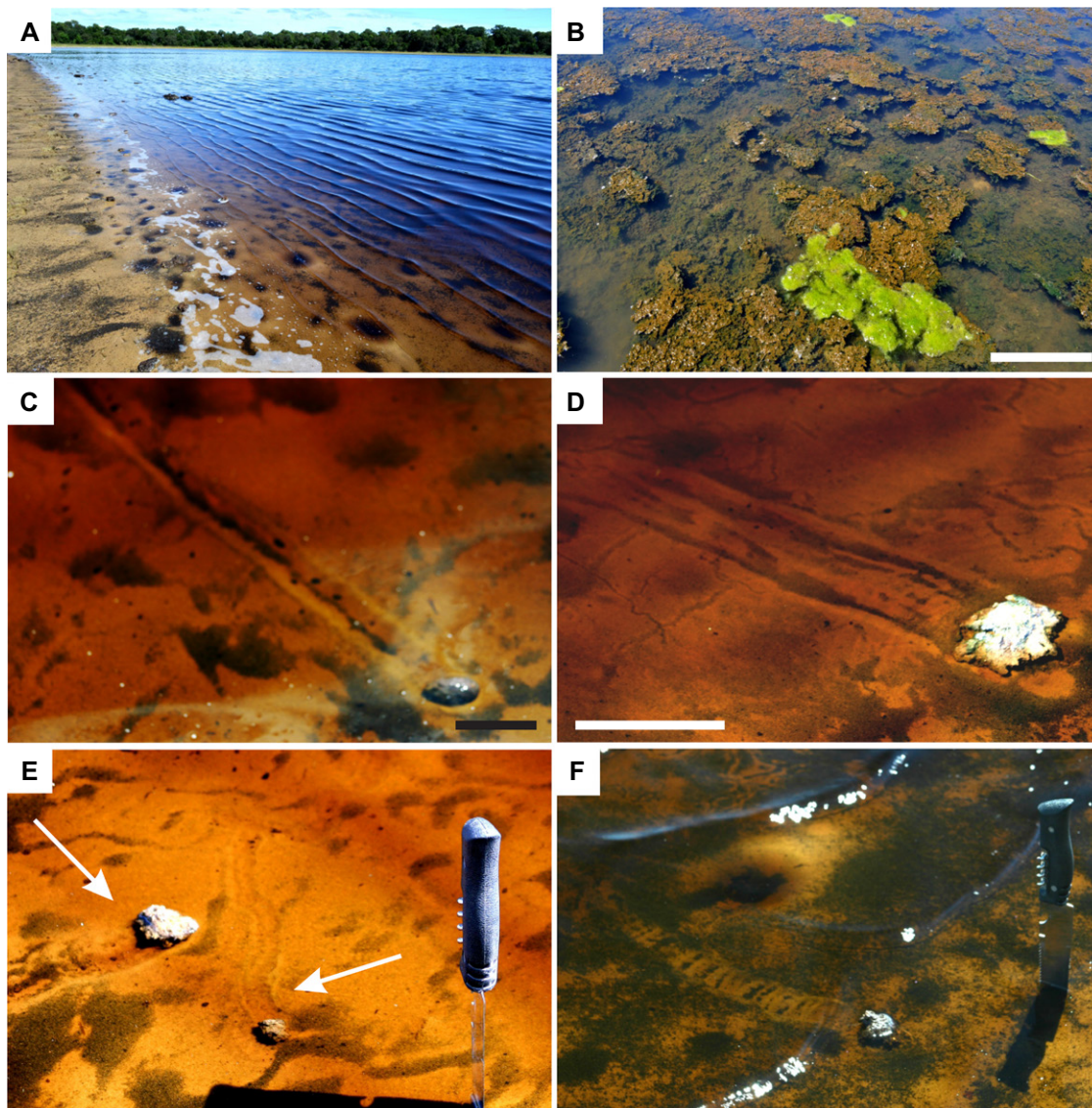
Figure 2. Pantanal (western Brazil) soda lakes and pseudotraces. (A) Lake shoreline showing incidence of wind-produced waves. (B) Microbial mat locally covering substrate. Bright green material in the lower part is detached microbial raft. (C) Simple, rectilinear groove with lateral raised lobes (type 1). (D) Type 2 groove with well-preserved linear and parallel erosion marks. (E) Types 1 and 2 curved grooves produced by variations in wind direction. (F) Slightly curved type 3 groove presenting crescentic pattern in opposite direction to particle movement. White and black scale bars in B, C, and D are 5 cm long. Knife in E and F is 20 cm long.

mats. The degree of sinuosity is related to slight changes in wind direction that cause deviations in the trajectory of the rafts. Types 1 and 2 are formed by the dragging of fragments of mats on the sandy substrate under increasing wind velocity conditions (>4 m/s), while the formation of type 3 requires high-frequency oscillatory motion of partially floating flocs under continuous action of waves produced by low-velocity winds (<4 m/s). The crescents resembling backfill menisci are produced by the oscillatory impact of the medium-buoyancy rafts against the substrate, promoting the accumulation of sandy crests separated by small troughs. The continuous up-and-down oscillatory movement of spherical and discoidal fragments eroding and deforming the substrate results in the formation of crescentic features in the inner part of the pseudotraces, similar to tilting marks (Wetzel, 2013). During periods of stronger winds (>6 m/s), some rafts are dragged over the substrate, producing rectilinear to slightly curved grooves (type 1; Fig. 2F). The frequency of the wave orbitals directly affects the spacing between the crescents (i.e., decrease in wave frequency increases spacing between crescents). The structures in Figure 2F were generated by an average frequency of 1 Hz (one oscillatory cycle per second). Despite their nonbiogenic origin, types 1 and 2 (Figs. 2C and 2D) resemble simple grazing trails with lateral levees, such as *Archaeonassa* (Jensen, 2003). Type 3 resembles meniscate trace fossils, such as *Beaconites* and *Taenidium*, or horizontal trails with transverse bars and furrows, such as *Steinsfjordichnus* (Buatois et al., 2017).

IMPLICATIONS AND CAVEATS FOR INTERPRETING THE EDIACARAN TRACE-FOSSIL RECORD

Despite important advances in unravelling the phylogenetic affinities of members of the Ediacara biota (for an updated perspective, see Dunn and Liu [2019]), the occurrence of simple trace fossils (*Helminthoidichnites*, *Helminthopsis*, *Archaeonassa*, *Gordia*) has been historically taken as solid evidence of the presence of bilaterians (Seilacher, 1989). The same ichnotaxa persisted all through the Phanerozoic (Buatois and Mángano, 2018). However, these have been subject to scrutiny, in cases resulting in alternative interpretations. First, compressed carbonaceous filaments may resemble simple trails (Jensen et al., 2006). However, this interpretation does not apply to structures with levees, which provides evidence of sediment displacement, supporting a trace-fossil origin (Droser et al., 2005). Second, giant protists may form straight and short trails (Matz et al., 2008) with discontinuous levees and axial ridges, features that are rare in metazoan traces (Buatois and Mángano, 2016). In contrast, long trails point to a metazoan origin (Gehling and Droser, 2009). A third scenario involves the production of similar structures from interactions between oscillatory flows and microbial aggregates (Mariotti et al.,
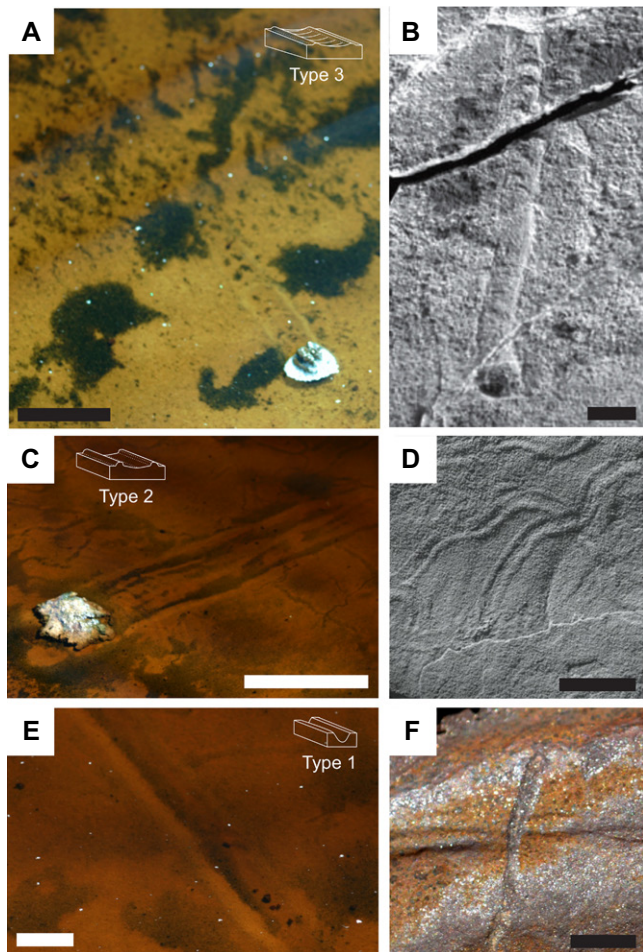
Figure 3. Comparison between Pantanal (western Brazil) pseudotraces and Ediacaran structures regarded as trace fossils. (A) Type 3 pseudotrace with similar morphology to the structure shown in B. (B) Structure with crescentic internal divisions interpreted as a trace fossil, Mistaken Point Formation, Canada (see Liu et al., 2010). The paleoenvironmental context of Ediacaran structures is not consistent with this origin due to oscillatory flows. (C) Type 2 pseudotrace with lateral raised levees similar to the structure illustrated in D. (D) *Archaeonassa* isp., Ust Pinega Formation, northwest Russia (see Jensen, 2003). Winding nature is inconsistent with a microbially induced origin, reinforcing its interpretation as a trace fossil. (E) Type 1 pseudotrace similar to the structure shown in F. (F) Simple linear structure interpreted as a trace fossil, Cerro Negro Formation, Argentina (see Arrouy et al., 2016). An inorganic origin is hard to disregard. Scale bars are 1 cm long in A, C, E, and F, and 5 cm in B and D.

2014, 2016). These structures have been generated in the laboratory, but the observation of similar pseudotraces in the Pantanal shows their production in a natural environment. We propose a set of criteria that allows us to distinguish between microbially generated pseudotraces and trace fossils.

First, evaluation of associated facies is paramount to assess the conditions under which the suggested mechanism may be invoked. Lake margins and low-energy intertidal areas are ideal for the mechanism documented in our study. However, Ediacaran trails are known from slightly deeper-water environments, namely, between fair-weather and storm wave bases. Regardless, Wetzel (2013) noted that similar tilting marks may be formed in the open ocean. This is consistent with our observations showing that under very intense winds, microbial objects can be dragged on the bottom, and with experiments showing the formation of similar pseudotraces by wave-induced near-bed flow, which may operate in subtidal regions (Mariotti et al., 2016). Geostrophic flows during storms can transport particles of different sizes and densities hundreds of meters toward the offshore. Thus, microbially induced pseudotraces may form along a wider range of bathymetries than

that represented in the Pantanal. However, considering the central role of oscillatory flows, we disagree with the idea that these structures may form at any water depth (Mariotti et al., 2016). Trails occur in Ediacaran deposits formed well below storm wave base, where oscillation at the sea bottom is negligible (MacNaughton et al., 2000; Carbone and Narbonne, 2014), limiting the application of the wave-generated model. Type 3 (Fig. 3A) resembles horizontal trails with crescent marks from the Mistaken Point Formation (Newfoundland and Labrador; Fig. 3B) interpreted as deposited in deep water (Liu et al., 2010). Their morphology is consistent with that of tilting marks (Retallack, 2010), but the deep-marine setting is inconsistent with this interpretation.

Second, the structures from the Pantanal are produced by removal and accumulation of disaggregated sand, which is more difficult on a microbially bound substrate. Similar structures associated with microbial mats may be assigned to the activity of animals with more confidence.

Third, the mode of occurrence and morphology help to differentiate between pseudotraces and trace fossils. The Pantanal pseudotraces tend to be straight and parallel to each other, but they occasionally are curved and oblique

due to changes in flow orientation. By contrast, animal traces may wind, meander, or loop, and they are not commonly oriented parallel to each other. Type 2 impressions display levees (Fig. 3C), which are also present in *Archaeonassa* isp. from the Ust Pinega Formation (northwest Russia; Fig. 3D; Jensen, 2003), but the winding nature of the latter is inconsistent with a microbial floc genesis. In contrast, type 1 (Fig. 3E) appears to be indistinguishable from a straight structure regarded as a trace fossil from the Cerro Negro Formation (Argentina) (Fig. 3F; Arrouy et al., 2016).

Fourth, pseudotraces are formed on the sediment surface. Preservation of structures as both positive and negative reliefs on the same surface supports intrastratal formation (Gehling, 1999) and is a key argument in favor of a trace-fossil origin.

Finally, size provides information for distinguishing between trace fossils and pseudotraces. The Pantanal structures tend to be larger than Ediacaran trails and show a wider size range, reflecting the variable size of the flocs, although size overlap does exist (Fig. S1). In addition, pseudotraces generated in the laboratory by the motion of microbial aggregates tend to be smaller than those observed in nature, reflecting the small size of the particles used (Mariotti et al., 2016).

CONCLUSIONS

The occurrence of pseudotraces in the Pantanal soda lake offers a unique opportunity to test alternative interpretations for the origin of Ediacaran structures interpreted as trace fossils. Our observations indicate that (1) pseudotraces may form by the interaction of microbial flocs, wind action, and sandy substrates in a shallow-water body under natural conditions; (2) the studied pseudotraces resemble some Ediacaran structures regarded as trace fossils; and (3) a set of criteria can be established to help differentiate between biogenic and abiogenic structures in the fossil record. Careful analysis of the morphology, orientation, and preservation style of the structures, sedimentary environments, and presence or absence of microbial mats should be done on a case-by-case basis, avoiding interpretations based on limited material.

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