## Laminae as Structural Biosignatures in NASA's Life Detection Knowledge Base

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## Abstract

Laminae are millimeter-scale features in rocks created by physiochemical processes that can be influenced by the presence and activities of communities of organisms that occur as biofilms and microbial mats. The structure and composition of laminae reflect the processes involved in their formation and can be preserved in the rock record over geologic time, however diagenetic and metamorphic alteration can lead to the loss of primary information and confusion over the interpretation of their origins. As potential records of ancient life, laminae can preserve evidence of microbial activity over billions of years of Earth's history. On planetary bodies beyond Earth, laminae in sedimentary rocks are the most common sedimentary structure and represent significant features of interest that can record habitable conditions (e.g., the presence of liquid water) at the time of their formation. Here we review the significance of laminae as targets for astrobiological exploration. We discuss common mechanisms by which laminae form in natural environments on Earth, present arguments and evidence for laminae as biosignatures, and describe how such information is incorporated into the NASA Life Detection Knowledge Base.

## 1. Introduction

In the search for fossilized evidence of microorganisms in the earliest rock record on Earth—and potentially among other planetary bodies—laminae produced from biofilm growth in and on sediments constitute a prime morphological feature of interest. Laminae can form through a variety of non-biological depositional processes that involve detrital sediment accumulation and chemical precipitation; their formation, however, can be influenced by the presence, behavior and metabolic activities of organisms, especially when those communities live at sediment surfaces in biofilms (even those only a few cell-layers thick) and in thicker (mm-cm) trophically segregated microbial mats.

In the context of planetary robotic exploration, laminae are detectable using a wide variety of imaging and analytical instruments. They are distinguishable by marked changes in color, granulometry, and chemical composition compared to their surrounding host deposits. In rocky outcrops, laminae appear in cross section as thin dark or alternating light and dark layers and exhibit a variety of geometries. Lamina range in size from micrometers (i.e., requiring microscopic observation) to millimeters (i.e., visible to the naked eye). They can occur as individual layers or be superimposed upon antecedent laminae to form thicker stacked units. In this paper we provide an overview of the processes involved in the formation of abiotic and biotically-influenced laminae, their significance as astrobiological targets, and how they are incorporated as features of interest into NASA's Life Detection Knowledge Base (LDKB).

The LDKB is an online community tool that presents collated information relevant for understanding potential biosignatures that may be detected on rocky planets beyond Earth (Hoehler et al., this issue). A variety of potential biosignatures are presented in the LDKB and are organized into broad categories of "Chemistry", "Structure", and "Activity" (Davila et al., Parenteau et al., Pohorille et al., Shkolyar et al., Perl et al. Des Marais et al., this issue). Morphological features of interest, such as laminae, are grouped together in the Structure category of the LDKB. Arguments and evidence regarding the biogenicity of structural features are organized by observable morphological characteristics, namely "size", "spatial distribution", "shape/form", "orientation", and "texture/fabric" (Shkolyar et al., this issue). We focus here on the description and interpretation of these morphological characteristics with respect to laminae, which represent a key indicator of microbial life in the sedimentary rock record on Earth (e.g., Walter et al., 1980; Buick 1992; Walsh 1992; Riding, 2000; Noffke, 2000, 2001, 2010, 2021; Cady et al, 2003; Westall et al., 2001, 2006, 2015a, 2015b; Awramik and Grey, 2005; Oehler and Cady, 2014; Homann et al., 2015, 2016 2018; Homann 2019; Trower and Lowe 2016; Hickman-Lewis et al., 2016, 2018, 2023; Bosak et al., 2021; Noffke et al., 2024). In addition to the structural characteristics discussed here, biogenic laminae may also retain other key signatures of life that include carbonaceous remnants of the original microbial community (e.g., biomolecules and their degraded residues), characteristic stable isotope fractionation patterns, and trace element concentrations consistent with biological activity, among others. These additional lines of evidence for biogenicity are represented within the "Chemistry" and "Activity" categories of the LDKB, and are reviewed further by Des Marais et al., and Parenteau et al., this issue.

Here we focus on structural observations relevant to the identification and interpretation of laminae as potentially biogenic features with an emphasis on examples described from Precambrian rock samples obtained through geological field studies. We discuss information about possible geological, physical, chemical, and biological processes that are signified by the presence of laminae, as well as how they occur as defining characteristics of laminated microbialites that include stromatolites and microbially induced sedimentary structures (MISS; *cf.* Noffke et al., 1996). We also discuss the specific attributes of laminae that are highlighted in the LDKB based on preliminary efforts to populate the database with evidence from the literature, a requirement of LDKB entries. Lastly, we summarize considerations with regard to the interpretation of laminae as well as knowledge gaps that were identified through this exercise.

### 2. Laminae and their Characteristics

In the field of sedimentology, individual lamina are typically regarded as having thicknesses less than 1 cm when observed in vertical cross section, whereas thicker units are considered "beds" (*e.g.*, Collinson & Mountney 2019; Tucker & Jones 2023). Consistent with this, we define laminae as micrometer-to-millimeter scale layers in sedimentary rock that consist of detrital and/or chemically precipitated material deposited commonly, though not exclusively, in an aquatic setting. In depositional environments, laminae can form as a result of changes in fluid energy and/or geochemistry (*e.g.*, temperature, pH, Eh, ion saturation, and density), changes in sediment supply that reflect shifts in the environment, and by changes/interruptions at the sediment water interface during mineral accretion or detrital grain deposition. These mechanisms may reflect abiotic processes, biological activity, or a combination thereof. Evidence of such processes can be preserved in lithified laminated sedimentary rocks for geological timescales and thus serve as attractive targets for understanding the paleoenvironmental conditions and habitability of the original depositional environment. Although not intended to be an exhaustive list of all possible processes and environments responsible for the formation of laminae, some of the major abiotic and biotic mechanisms involved are described below.

#### 2.1. Occurrences and Composition

Laminae form in nearly every sedimentary environment and exhibit wide range of structures (both at the macro- and micro-scale) and compositions. On Earth, laminae most commonly form in subaqueous environments at the sediment–water interface, such as shallow- and deep-water lacustrine, riverine, and marine settings. They can also form in environments where evaporative and/or cooling chemical precipitates accumulate around hot and cold springs, desert playas, and on cave walls (e.g., Spear et al., 2007; Handley *et al.*, 2008; Gonzalez-Pimentel et al., 2018; Lingappa et al., 2021). Laminae also form in the subsurface, for example within the void space of vesicular igneous rocks and fractured openings that can infiltrate all types of hard rock over geological time (e.g., Homann et al., 2016; Ivarsson et al., 2020; Neubeck et al., 2021; Cavalazzi et al., 2007, 2021; Lima-Zaloumis et al., 2022). Additionally, laminae can form either sub-aerially or sub-aqueously via a high flow regime in settings such as turbidites, rivers, and in sand sheets at the edge of eolian ergs (Boggs, 2006).

Laminae and laminated sedimentary structures formed in association with microbial activity are common features in Archean and Proterozoic sedimentary rocks (*e.g.*, Grotzinger and Knoll, 1999; Riding, 2000; Bosak et al., 2013; Grey and Awramik, 2020). Such structures can be found today forming in a variety of surface and subsurface environments (e.g., Hoffman, 1976; Reid et al.,

2000; Heim et al., 2017; Vahrenkampf et al., 2024). Thus, laminae as structural features provide a long-term record of both direct and indirect aspects of biological processes that have been occurring *in situ* since the early appearance of life on our planet. We hypothesize that laminated structures preserved within detrital and chemical sedimentary rocks beyond Earth could contain similar paleobiological information where microbial life has evolved. The lack of evidence on other planetary bodies in our Solar System for plate tectonics, a mega-scale process that can significantly alter the fidelity of preservation of fossilized biosignatures in surficial sedimentary deposits, suggests that primary laminae may preserve a significant amount of information about their depositional processes and possible biogenic input on their formation.

The environments where laminae form are broadly categorized into detrital and chemical sedimentary systems, each of which is dominated by specific depositional processes that can result in characteristic textures that manifest in lithified rocks. It is important to note that physical sedimentary and chemical sedimentary processes are not mutually exclusive, as both detrital accumulation and chemical precipitation often occur simultaneously.

#### 2.1.1. Detrital Laminae

Detrital laminae commonly develop when clastic particles accumulate atop bottom sediments of a water body (e.g., rivers, lakes, and marine environments that include deltas, lagoons, and estuaries). Detrital components can range from mud-sized ( $<4 \mu m$ ) to silt-sized ( $<62.5 to >4 \mu m$ ) to sand-sized (<2 mm to >62.5  $\mu$ m) particles, or a combination thereof. The particles may be composed of individual minerals, rock fragments, biologically derived organic material and, in Phanerozoic beds, shelly material. The composition and grain size of depositing clastic materials can change gradually over time owing to shifts in sediment sources and supply (e.g., through weathering and erosion), water energy and depth, or they may oscillate regularly due to seasonal processes, such as glacial meltwater influx (resulting in the deposition of laminated couplets known as varves; e.g., Anderson and Dean 1988). The characteristics of sediments may also change suddenly via bioturbation or during catastrophic events such as storms and earthquakes (e.g., Kreisa 1981; Sabatier et al., 2022). In any given depositional environment, the final grain sizes and shapes of detrital sediments are influenced by the weathering of continental source rocks, transport of particles by flowing fluid (wind, water, or ice), and by reworking and abrasion during bed load transport. In combination, these effects contribute to variations in detrital sediment that eventually manifest as distinctive layers visible in lithified sedimentary rock.

#### 2.1.2. Laminated Chemical Sediments

Laminated chemical sediments develop within ion-supersaturated aqueous environments such as evaporative subaerial settings and, in combination with cooling, in hot spring settings via the precipitation of amorphous mineraloids and crystalline minerals from aqueous solutions. Environments where chemical sediments commonly form include shallow marine environments, hot and cold springs, lakes and sabkhas, and subsurface environments that include fractures and vesicles. Chemical precipitates commonly found in such settings include opaline silica, carbonates (*e.g.*, calcite, dolomite, aragonite, and magnesite), anhydrous and hydrous sulfates (e.g., gypsum, barite, and mirabilite), chlorides (*e.g.*, halite and sylvite), phosphates (*e.g.*, apatite), and iron oxides (*e.g.*, ferrihydrite and hematite). The size of crystals in such systems is influenced by several factors that include the rate of nucleation and the duration of time that fluid supersaturation is

maintained within the local environment; crystalline precipitates can continue to increase in size depending on the duration of undisturbed growth during relatively quiescent super-saturated conditions (e.g., García-Ruiz and Otálora 2015). In aqueous environments, chemical precipitates may form at or beneath the sediment–water interface, or they may precipitate within the water column or at the air–water interface and settle onto the benthic floor. Such processes lead to distinctive textures that can be recognized as primary forms (e.g., Warren 2006). Laminae may also form by secondary diagenetic processes, as is the case with concretions, geodes, and botryoids where chemically oscillating reactions result in the precipitation of fine scale laminar patterns that consist of quartz, apatite, and/or carbonate (Papineau et al., 2021). Chemical sediments may become altered to form secondary diagenetic textures and/or may be replaced by more stable mineral forms due to their generally high solubilities as compared to resistant siliciclastic material (e.g., Warren 2006; Lima-Zaloumis et al., 2024).

#### 2.2. Common Lamination Geometries

#### 2.2.1. Parallel, Flat Lamination

In the rock record, laminae can exhibit wide range of shapes and orientations. Parallel horizontal laminae that display a flat, smooth morphology are formed in aqueous environments by the settling of fine-grained sand, silt, and mud through the water column, often from slow-moving sediment clouds above the depositional surface, or by low-density turbidity currents (Tucker 2001). They can also form in an upper flow regime where the energy of flow is high enough to plane off the dunes such as in turbidites and sand sheets in eolian environments. In chemically precipitating environments, flat laminae can form by evaporation and ion supersaturation of fluids; Seasonal precipitation and settling of chemical sedimentary particles through the water column, as well as direct precipitation on the benthic floor, lead to the development of parallel, laterally continuous laminae that are sometimes traceable for tens of kilometers (Fig. 1A; Warren, 2006).

Although flat laminae can contain biological constituents, such as organics transported from a surrounding basin or formed in place (e.g., by non-lithifying biofilm growth or settling of phytoplankton through the overlying water column), their geometry is not strongly indicative of past biological activity. It should be noted, however, that some flat laminated structures have been interpreted to form in the presence of or directly influenced by microorganisms (e.g., in the case of "stratiform" stromatolites (Walter, 1976), "biolaminites" (Gerdes and Krumbein, 1987) and "cryptalgal laminites" (Monty, 1976)). Although flat laminar geometries in sediments may not be uniquely indicative of biological activity, some post-depositional activities that act on flat laminae may more strongly constitute structural biosignatures, for example in the case of disruptions caused by grazing or burrowing by Metazoa (see review of ichnofossils by Taylor et al., 2003). Similar activities by prokaryotes have been inferred at the microscale among glass and carbonate phases (e.g., Garcia-Pichel, 2006; Ivarsson and Holm, 2008; Fisk et al. 2019).

#### 2.2.2. Ripples and Cross Lamination

These structures commonly form in aqueous environments characterized by unidirectional or bidirectional currents, i.e., wave or current-dominated settings (Fig. 1B), but they can also form due to aeolian (wind-driven) processes in subaerial settings (Fryberger et al., 1988). These

lamination geometries typically exhibit distinctive crests and trough patterns that may be sinuous, linguoid, or asymmetric, among other forms (e.g., Rubin and Carter 1987; Tucker 2001). Migration of ripple marks over time results in the deposition of cross-laminated sequences, which in vertical cross section reflect two dimensions of the original three-dimensional structure of ripples or larger-scale subaerial dunes. Paleoenvironmental information such as paleocurrent direction and strength may be inferred by analyzing cross laminae and their orientations in space.

The presence of ripples and cross laminae may indicate the past presence of water based on the ripple index (a calculation of wave length to wave height; Tanner 1967), therefore such structures serve as important astrobiological targets to understand an environment's past habitability potential. As with flat laminae described above, however, cross or ripple laminae as structural features do not directly represent potential evidence of past life. Such structures may incidentally be associated with biosignatures such as burrows and other ichnofossils, kerogen, microfossils and EPS (e.g., Noffke et al., 2006, 2008, 2013a). In such environments, biofilm-forming microorganisms often do not influence the morphology of laminae, but instead template onto existing sediment relief (a process termed "imprinting", Noffke et al., 2001).

#### 2.2.3. Concentric and Botryoidal Lamination

Concentric laminae can form as cements around moving grains in shallow wave dominated environments. Concentrically laminated spherical to subspherical (egg-shaped) particles smaller than 2 mm are termed ooids, while larger grains termed pisoids can take on a variety of non-spherical shapes. Biofilms may develop on the surfaces of moving grains and result in the precipitation of irregular laminae, which can manifest as couplets of dark organic-rich laminae alternating with organic-poor lighter colored laminae (Flugel 2010). Such structures are termed oncoids (originally described by Heim, 1916), where lamination thickness is dependent upon the activity of cyanobacteria and changes in fluid geochemistry over time (e.g. seasonal conditions affecting carbonate saturation).

Concentric laminae may also form within diagenetic concretions such as botryoids, geodes, granules, and rosettes (Fig. 1C–D). Generally, spheroidal concretions precipitate via diffusive transport of reactants and elongate forms precipitate via advective flow of reactants. Concretions precipitate via two endmember growth mechanisms: radial and pervasive. Radial growth occurs when cements preferentially precipitate in the subsurface around a nucleus and the concretion grows radially outward forming concentric laminae (e.g., Mozley, 1996; Raiswell and Fisher, 2000); pervasive growth does not produce laminae. Such structures can be associated with organics, which may be derived from biological or abiotic sources and can be preserved over billion-year timescales (Sugitani et al., 2007, 2013, Brasier et al., 2011; Wacey et al., 2012; Papineau et al., 2021; Papineau, 2024), although some ancient examples may have been subject to younger post-depositional oil migration (Coutant et al., 2021).

Diagenesis can also produce Liesegang bands where reactants diffuse towards each other in sediments, creating laminae with spacing controlled by the rate of diffusion (Liesegang, 1896;

Ostwald, 1897). These forms can be identified because they typically exhibit crosscut stratification. They can occur as poorly cemented, only creating a coloration pattern in the rocks, or they can be so well-cemented that they weather differentially and produced positive topographic features in weathered rocks.

#### 2.2.4. Irregular Laminae and the Role of Biology

In sedimentary rocks, sub-millimeter-scale irregular laminae may be associated with remnants of biological activity (Fig. 2). Microbially associated laminae are commonly described as wavy, which Walter (1972) defined as having 'flexures of wavelength greater than 2 mm', and wrinkled, defined as having flexures of wavelength less than or equal to 2 mm (see also Grev and Awramik. 2020, p. 129). Other descriptions of irregular laminae in the literature include terms such as undulating, non-isopachous (i.e., uneven thickness), crenulated, wispy, and crinkly, among others. Irregular laminae may exhibit discontinuities in lateral extent, bifurcation, anastomoses, and tapering along ends. Additionally, they can include structural elements such as tufts (Fig. 3) and encrusted microfossil structures (e.g., Flugel 2010; Homann et al. 2015; Hickman-Lewis et al. 2018; Noffke et al., 2013; Hickman-Lewis & Westall (2021)). In sedimentary rocks on Earth, irregular laminae are commonly interpreted as remnants of microbial biofilm communities, particularly when combined with additional lines of evidence that support their biogenicity such as their occurrence in a habitable environment (as interpreted from local geologic context) and the presence of microfossils, organic carbon, and isotope fractionation patterns consistent with metabolic activity (e.g., Buick et al., 1981; Awramik and Grey, 2005; Schopf et al., 2007; Wacey, 2009; Wagstaff and Corsetti, 2010; Homann et al., 2018; Homann 2019; Lepot, 2020). Unlike abiotic processes that produce relatively uniform deposits of flat lying, isopachous (eventhickness), cross-layered, or regularly oscillating layers of sediment, micrometer-to-centimeter scale benthic biofilm and mat communities can directly produce or influence the formation of irregularly shaped laminae at comparable scales; it is this irregularity that suggests biogenicity.

The distribution and morphology of microbially associated laminae reflects non-uniform colonization and growth characteristics of biofilms, as well as their responses to a dynamic environment with changing fluid energy regimes, sediment burial rates, and nutrient availability among other factors. Due to the minimum energy requirements for cellular upkeep and reproduction, active biofilm communities must occupy zones with sufficient access to nutrients and energy supplies. Chemical and radiative energy availability can vary over micrometer scales in space and time owing to dynamic environmental processes (e.g., Jørgensen and Des Marais 1990). For example, zones inhabited by photosynthetic communities at the sediment-water interface may become subject to sediment accumulation and burial, as is the case in most active shallow marine, lacustrine, and hot spring environments on Earth. As zones of energy availability migrate in space (e.g., upward as deposition proceeds), biofilm communities can react via active motility or passive dispersion, preferentially colonizing zones with greater access to nutrients and energy supplies (e.g., Sauer et al., 2022). In addition to minimum energy requirements, biofilm colonization depends upon surface adhesion and roughness characteristics, favorable fluid flow velocities, and temperature (Donlan, 2002). The thickness of a biofilm reflects community maturity and can be influenced by species composition (Murga et al., 1995). Biofilms can mature into cm-thick layered microbial mats with complex, stratified community structure (e.g., Des Marais, 1995, Dupraz et al., 2009), although thicker mats with high biomass production may hinder

sedimentation (and thus preservation in the rock record) by limiting ion diffusion (e.g., Reid et al., 2024).

Biofilms can directly affect the depositional characteristics of the sediment-water interface that they inhabit. Biofilm communities are typically composed of a consortium of species that utilize a variety of autotrophic and heterotrophic metabolisms. Some metabolisms, such as photosynthesis and microbial sulfate reduction, contribute toward shifting the pH of the surrounding microenvironment such that mineral (particularly carbonate) supersaturation and precipitation is favored (the so-called "alkalinity engine"; Dupraz et al., 2009; Gallagher et al., 2012). Biofilms also contribute to sedimentation by reducing energy barriers to nucleation, thus serving as favorable sites for mineral precipitation (Braissant et al., 2007; Dupraz et al., 2009; Gallagher et al., 2010) even in cases when the ambient aqueous medium is undersaturated with respect to the precipitating phase (Moore et al., 2020). Cell lysis and EPS degradation, caused in part by microbial heterotrophy, may also release cations that contribute to mineral saturation and precipitation (Dupraz et al., 2009). In addition to extracellular mineral precipitation, some organisms precipitate minerals intracellularly for a variety of purposes that include use as navigational aids within the Earth's magnetic field (e.g., Bazylinski et al. 2007) and as possible carbon stores (Benzerara et al., 2014). Sulfur-metabolizing microbial consortia may form pyrite deposits with specific morphologies (e.g., amorphous nanopyrites and framboids) that can serve as evidence for biogenicity (Marin-Carbonne et al., 2018, 2020). These and other biominerals can become preserved and contribute to the overall lithic texture and/or elemental component of biologically associated laminae (e.g., Chang et al., 1989).

Due to the production of extracellular polysaccharides, biofilms can exhibit sediment baffling and trapping (microbial accumulation of sediment), biostabilization (microbial protection against the erosion of sediment), and binding (incorporating of sediment into the biofilm fabrics, e.g., Reid et al, 2000, Gerdes et al., 2000; Noffke et al., 2001, 2024; Frantz et al. 2015; Noffke 2021). Such activities influence sedimentation and result in specific structural features (Noffke et al., 2001); for example, the occurrence of laminae and adhered detrital grains at steep interfaces beyond the angle of repose (i.e., the maximum angle at which particles will remain in place without sliding) is often interpreted as evidence of organic mat cohesion and associated trapping and binding by past biofilm communities (e.g., Mitchell, 1985; Schieber 1998; Hofmann et al., 1999; Riding 2000; Kenter et al., 2005; Allwood et al., 2006, 2009; Murphy and Sumner 2008; Le Heron et al., 2013, Homann et al., 2015; Tice et al., 2017, Wilmeth et al., 2019). Such interpretations are supported by laboratory experiments that have investigated grain trapping among living mat communities (Frantz et al., 2015). In detrital (clastic) deposits, laminae formed in association with biofilms may also exhibit biostabilization characteristics when compared with surrounding host sediments (Noffke et al., 1997; 2024); eroded fragments of biofilms in the form of mat chips and roll-up structures preserved in rocks (Fig. 4) document the initial plasticity and cohesiveness of an organic-rich layer and a strong enough water current able to transport bioclasts (Simonson and Carney, 1999; Tice and Lowe, 2004; Noffke et al., 2001; 2023; Tice and Lowe, 2006; McLoughlin et al., 2023; Howard et al., 2024). The hydrodynamic conditions of the local environment can also affect biofilm/mat lamination morphology, where low energy regimes favor the formation of smoother mat textures, and higher energy fluid regimes (e.g. inter- to supratidal environments) favor the formation of increasingly wrinkled and tufted textures exhibiting higher vertical relief (Homann et al., 2015). The combined effects of biology on both detrital and chemical sediment accumulation can be preserved as irregular laminae, which can retain carbonaceous remnants of the original microbial community and/or mineral textures and fabrics associated with microbial activity.

#### 2.2.5. Irregular Laminae Formed by Nonbiological Processes

Several abiotic processes can result in the formation of laminae with irregular geometries (e.g., non-isopachous, discontinuous, wavy, etc.). Stylolites represent one such example of irregular abiotic structure (Fig. 1E), which exhibit zig-zagged laminae formed by pressure solution that – especially in carbonate rocks - results in the removal of material leaving behind an insoluble residue, the morphologies of which can potentially be confused with biofilms. Although in some cases stylolites may be difficult to distinguish from crinkled remnant microbial mat material by observation of their structural features alone (e.g., thickness, orientation), they can be distinguished by their chemical composition and texture (Noffke et al., 2008), particularly by their highly irregular undulation at the microscale, which is inconsistent with microbialite morphogenesis (Hickman-Lewis et al., 2024). Soft-sediment deformation caused by slumping, slope failure, shear stress, and liquefaction can also result in the formation of irregular "convolute laminae", which are particularly common among turbidites (Fig. 1F; Mills, 1983; Tinterri et al., 2016). Additionally, "hummocky" and "swaley" cross laminae can form irregular curved geometries that occur in fine-to-medium grained sandstones in shelf environments. These structures can exhibit nonuniform thicknesses with thinning toward convex up hummocks (topographically high areas) and thickening around swales (low areas of sedimentation) atop complex wave-scoured surfaces (Dott and Bourgeois, 1982; Collinson and Mountney, 2019).

Metamorphism can also produce oriented fabrics that can resemble biogenic laminae (Fig 1G). In the Isua supracrustal belt (SW Greenland), laminated structures originally interpreted as stromatolites (Nutman et al., 2016) were reinterpreted by others as products of metamorphism (Allwood et al., 2018; Zawaski et al., 2020). In each case above, multiple lines of evidence are key to deciphering the possible biological origins of irregular laminae, such as their three-dimensional orientation in space, the presence of microfossils, diagnostic biomolecules, or characteristic isotopic fractionation patterns indicative of microbial activity. Additionally, the local geologic context must be well characterized, ideally by detailed mapping efforts, in order to reconstruct the likely paleoenvironment and potential for habitability, and to rule out effects such as regional metamorphic activity and deformation associated with deep burial and exhumation.

#### 2.3. Laminae as Defining Features of Stromatolites and MISS

Biologically influenced laminae can occur as isolated layers sporadically distributed in a rock, or they can occur as well-defined stacks that form structures of flat to positive (convex-upward) relief, the latter being known as stromatolites. Sedimentary structures composed of stacked laminae are often large enough to be visible to the naked eye in rocky outcrops. Grey and Awramik (2020) define stromatolites as "a laminated organosedimentary structure produced by precipitation or by sediment trapping and binding as a result of the growth, behaviour, and metabolic activity of microorganisms, principally cyanobacteria". Internal laminae are the defining feature of stromatolites, where the word "stromatolite" [Stromatolith] was derived originally from the laminated appearance of microbial bioherms (Kalkowski, 1908). Stromatolites can exhibit a variety of lamination architectures that Grey and Awramik (2020) describe as "banded", "filmy",

"streaky", "tussocky", "pillared", "veriform", and "alveolar" (Fig. 5). Although similarly biological in origin, other types of microbialites occur that do not exhibit layering, namely thrombolites (clotted internal fabric; Burne and Moore, 1987), dendrolites (dendritic internal fabric, Riding, 2001), and leolites (structureless internal fabric; Braga, et al., 1995). As products of microbial activity, stromatolites and laminated Microbially-Induced Sedimentary Structures (MISS) represent some of the oldest evidence of life on Earth; Notable examples are found in 3.4–3.5 billion year old (Ga) rocks from the Dresser and Strelley Pool formations in the Pilbara, Western Australia, (e.g., Buick et al., 1981; Hofmann et al., 1999; Allwood et al., 2006; Noffke et al., 2013; 2024), and among 3.4–3.3 Ga Kromberg and Mendon formations in the Barberton greenstone belt in southern Africa (e.g., de Wit et al., 1982; Byerly et al., 1986). Additional examples of ancient, laminated structures included in the LDKB are discussed in Section 3.

As with the development of individual biological laminae, the same microbial interactions influence the formation of stromatolites such as passive trapping and binding of detrital sediment by sticky extracellular polysaccharide (EPS), metabolic processes that contribute to mineral saturation and precipitation, and organic-mineral templating. Informed by modern microbialite growth in Shark Bay, Western Australia, and the Bahamian islands, Reid et al. (2024) describe a "microbial balancing act" in stromatolite formation that involves the production of biomass, mineral grain agglutination, and the precipitation/dissolution of chemical sediments. Key to the formation of mineral layering is the ability of a benthic microbial community as either an unstructured biofilm or stratified mat to lithify, generating a stable substrate on which additional biofilm or mat communities may colonize and grow. Mat lithification depends upon the ratio of mineral precipitation to biomass production, which is influenced by *extrinsic* factors (e.g. environmental pH, temperature, and cation concentrations) and *intrinsic* factors (e.g., microbial biomass and metabolic activity) relative to the microbial mat community. Changes in extrinsic and intrinsic processes that shift the balance of biomass growth and mineral precipitation will influence the preservation potential of any morphological biosignatures in the deposit, including laminae. Regular episodic variation in this balance, for example, due to seasonal environmental changes over time along with the potential for hiatuses in sediment accumulation, can result in the generation of stromatolites with distinctive laminae.

Stromatolite and MISS development can result in a range of macroscale morphologies (Walter, 1976, Noffke et al., 2001). In plan view, individual stromatolite "heads" may appear circular, ovate, or linear. In cross-sectional view, stromatolites can take a variety of shapes from domes forming columns, to conical and branching morphologies. The spacing of stromatolite heads can occur as isolated unlinked masses, closely spaced or continuous heads, or they may merge to form linked masses. MISS also include a wide variety of morphologies and sizes that include multidirectional ripple marks, polygonal oscillation cracks, erosional remnants and pockets, mat chips and roll-ups, and wrinkle structures (Noffke, 2021; Noffke et al., 2024).

As structures preserved in the sedimentary rock record, microbialites represent the cumulative sedimentary record of biofilm and microbial mat activities over time (Hofmann, 1969; Gray and Awramik, 2020). In stromatolites, each lamina represents a snapshot of growth and development, marking past positions of the sediment–water interface, i.e., the "synoptic profile," whereas the height of an ancient stromatolite documents the progress of the stromatolite building process (Hofmann, 1969). As described by Hofmann (1969), "The fundamental feature of a stromatolite is its mineralogically or texturally differentiated lamination. The synoptic morphology of the laminae

in an assemblage represents the dynamic equilibrium conditions as well as the micro-bathymetries of the biologically, chemically, and mechanically active interfaces in the environment". This is also true for MISS; however, chemical and mineral precipitation leading to upward-accretion does not play a role in MISS formation.

# **3. Laminae as Features of the Life Detection Knowledge Base (LDKB)**

#### 3.1. LDKB Structure and Language

The Life Detection Knowledge Base is a community tool designed to cover a wide range of possible biosignatures. By featuring supporting evidence from peer-reviewed literature, the LDKB is intended to convey the requirements needed to identify possible signatures of life. To coherently present information about disparate biosignature categories (e.g., isotopes, lipids, laminae, technosignatures, and beyond; Hoehler et al., Pohorille et al., Davila et al., Shkolyar et al., Parenteau et al., Des Marais et al., Perl et al., this issue) in a standardized format, the LDKB employs a formal organizational structure and language (Fig. 6). Each presented piece of evidence and their supporting arguments are organized into two main categories: *prevalence* and *signal strength*.

#### 3.1.1. Prevalence

Prevalence expresses the likelihood that a feature is produced by a particular source, such as by life or a nonbiological process. For features whose prevalence are not universal and may instead be environment-specific, the term *congruence* is used to classify that particular piece of evidence. "Prevalence" in epidemiology indicates the proportion of a population that has a specific characteristic (e.g., a disease; Krug and McNutt, 2008). Nominally, "prevalence" in astrobiology applies across all biological systems and all environments, therefore it includes all occurrences of a feature on Earth and beyond. The implication is that an argument about general prevalence is essentially independent of environmental context and rather about the intrinsic aspects of a feature. However, some features, such as biogenic laminae in sedimentary rocks, are expected to be prevalent in certain environments and unlikely in others. Therefore, it should be specified whether the prevalence of a feature is considered in the context of all environments or specific ones. The evaluation of prevalence within a specific environmental context is an evaluation of its "congruence".

#### 3.1.2. Signal Strength

Signal strength is the prominence of a feature (e.g., its abundance, rate, structure, patterns, intensity, etc.) that provides evidence for the past or extant presence of life or a nonbiological process. The prominence of a feature is determined by the balance between its rates of production and its rates of modification and/or destruction. For a biosignature, this balance is determined by its biological production, its survivability, and the patchiness of its occurrence in a target

environment. The "target environment" includes the environment prevailing during either feature production or its modification, degradation, and/or destruction. These environments may be different (separated by time and/or space).

#### 3.2. Arguments and Evidence in the LDKB

In the following sections, we discuss example LDKB entries of laminae found in Precambrian rocks from the Barberton greenstone belt, South Africa, and from the Pilbara, Western Australia. The entries discussed relate to three types of observations that might be made by a current or future life detection mission, namely 1) "Size Distribution", 2) "Spatial Distribution", and 3) "Texture & Fabric". These measurable parameters are applicable for all potential "Structural" biosignatures in the LDKB (see Shkolyar et al., this issue). These broad categories were determined with reference to well-established geological terminologies and are intended to be sufficiently "agnostic" to describe a given visual feature of interest.

#### **3.2.1. Biotic Prevalence Arguments for Laminae**

The following examples describe LDKB entries that address the *prevalence* of laminae that are generally considered attributable to the presence or influence of microorganisms or microbial populations or communities, as supported by evidence presented in the scientific literature.

*Argument* - Ancient laminae that display ripped up and plastically deformed rolled up fragments may be interpreted as remnants of cohesive microbial mats.

*Evidence* - In the 3.42 Ga Buck Reef Chert (Barberton greenstone belt), fine laminae deposited in open-marine volcanic platform facies contain ripped up, plastically deformed, rolled up fragments and occur within black bands in banded chert; These fabrics–consistent with cohesion displayed by modern microbial mats– are interpreted as remnants of microbial mats (Simonson and Carney, 1999; Tice and Lowe, 2004). These fine laminae incorporate and loosely drape detrital grains and form silica-filled lenses. They also form fine meshworks of filament-like strands <5 um in diameter that drape over detrital grains. "Gamma-type" laminae display even-spaced, flat laminae that tightly drape underlying sediments and exhibit lightly-fractionated  $\delta^{13}C_{org}$  values consistent with a biological origin (see LDKB entry under "Chemistry" – isotope ratio patterns: carbon redox states (e.g. Des Marais et al., this issue)).

*Evidence* – Rip-up and roll-up structures have been identified in the 2.5 Ga Hamersley Group (Simonson and Carney, 1999), ~2.5 Ga Campbellrand Subgroup (Gandin et al., 2005), 3.22 Ga Moodies Group (Noffke et al., 2006; Homann et al., 2015), 3.48 Ga Dresser Formation, Pilbara, Western Australia (Noffke et al., 2013), 3.33 Ga Josefsdal Chert (Westall et al., 2015b), 3.46 Ga stratiform 'Apex chert' (Hickman-Lewis et al., 2016), and 3.47 Ga Middle Marker horizon

(Hickman-Lewis et al. 2018) where geometries and geochemical signals coincide and support biogenicity.

*Argument* - Precambrian laminae that display micro-tufting may be interpreted as microbial mat remains.

*Evidence* - Precambrian laminae from the Barberton greenstone belt occur as horizontally continuous packets and display macro- and micro-tufted spatial distribution. Homann et al. (2015), Hickman-Lewis et al. (2018), and Hickman-Lewis & Westall (2021) argue that the micro-tufted spatial organization observed in the 3.22 Ga Moodies Group and 3.47 Ga Middle Marker horizon is consistent with growth phenomena associated with phototrophic microbial mats, namely the accumulation of gasses produced as a result of microbial metabolic activity as shown in modern microbial mats (e.g., Bosak et al., 2010).

*Evidence* – Micro-tufted laminae occur in the 3.48 Ga Dresser Formation, Pilbara, Western Australia. The tufts have a similar height/base ratio of 10/50 to 25/75 um, arranged in regular distances of 100-125 um from each other. Raman analyses showed a composition of pyrite and relic original carbonaceous matter. Raman spectra show two carbon peaks at 1350 cm<sup>-1</sup> (D1 disordered peak) and 1600 cm<sup>-1</sup> (the G graphite peak). The positions and widths of these peaks and the D1/G peak heights are consistent with maturation related to the metamorphic grade of the Dresser Formation. Based on their morphological similarity to modern tufted microbial mats and carbonaceous composition, these features are interpreted as biogenic microbial mat remains (Noffke et al., 2013).

*Argument* - Tufted laminae that exhibit hollow cavities are interpreted as evidence of gas accumulation formed by metabolic processes.

*Evidence* – In the Moodies Group, Barberton greenstone belt, chert-filled cavities in the interior of tufts were interpreted to represent silicified gas bubbles that were trapped within a microbial mat fabric (Homann et al., 2015; Homann et al., 2016) as they are common features in cyanobacterial mats that produce oxygen-rich bubbles (Bosak et al., 2010).

*Evidence* - Tufts with gas bubbles occur in the 3.48 Ga Dresser Formation, Pilbara, West Australia (Noffke et al., 2013) and within stromatolitic laminae in the 2.7 Ga Ventersdorp Formation, South Africa (Wilmeth et al., 2019, 2022). Gas bubbles concentrated along laminae in tufts are interpreted to form due to the upward migration of biogenic gases such as O<sub>2</sub> and H<sub>2</sub>S (Noffke et al., 2003).

*Argument* - Laminae that drape underlying sediments and entrain detrital material are consistent with microbial baffling and trapping processes.

*Evidence* - Within the Moodies Group, Barberton greenstone belt, Homann et al. (2015, 2018) and Homann (2019) describe laminae as draped and stabilized horizontally laminated and rippled

sandstones that have an enrichment in fine-grained quartz and feldspar, as well as heavy mineral grains (zircon and rutile or anatase), interpreted to be caused by microbial baffling and trapping; such microbe–sediment interactions are commonly observed in epibenthic microbial mats (Gerdes et al., 2000).

*Evidence* - In the Moodies Group, Barberton greenstone belt, laminae include oriented grains and heavy minerals such as rutile (Noffke et al., 2006; 2024). Grains interpreted to be derived from the substrate beneath a microbial mat are interpreted to become oriented as they are dragged upward by growth of microbial communities. The fine-grained particles enriched in organic material are interpreted as microbial mat remains that were engaged in baffling and trapping activities (Noffke et al., 2001). In addition, the carbonaceous materials that formed these microbial mats have carbon and nitrogen isotope distributions that are consistent with a biological origin (Noffke et al., 2006; Homann et al., 2018).

*Evidence* – Laminae composed of carbonaceous materials in the Middle Marker horizon, Barberton greenstone belt, drape underlying sedimentary layers and exhibit petrographic evidence for grain trapping (Hickman-Lewis et al., 2018). This grain trapping behavior has also been detected in three-dimensional renderings of microbial mat horizons, where organic laminae enwrap denser mineral particles (Hickman-Lewis et al., 2019).

#### 3.2.2. Signal Strength Arguments for Laminae

The following examples describe LDKB entries that address the *signal strength* of laminae or the prominence of a feature as determined by the balance between its rates of production and its rates of modification and/or destruction. The arguments below present a useful framework for users of the LDKB to recognize that laminae as structural features may persist on planetary surfaces for billion-year time scales and may be laterally extensive at the km scale.

**HIGH** *Argument* - Laminae several micrometers-to-millimeters thick that are interpreted as mineralized remains of biofilms and microbial mats can be preserved for billion-year timescales on Earth.

*Evidence* - Laminae within the Middle Marker horizon 3.472 Ga) of the Barberton Greenstone Belt that have been interpreted as fossil microbial mats occur as packets with thickness of 200  $\mu$ m to 2.5 mm (Hickman-Lewis et al., 2018, 2019; Homann et al., 2015, 2018).

*Evidence* - Within the Buck Reef Chert (3.416 Ga) of the Barberton Greenstone Belt, laminae interpreted as microbial mat remains occur as fine, anastomosing (i.e., branching and merging) carbonaceous laminae (cf., Tice and Lowe, 2004; Tice, 2009; Greco et al., 2018; Hickman-Lewis et al., 2022; Pellerin et al., 2023; Reinhardt et al., 2024).

*Evidence* - Within the Josefsdal Chert (3.33 Ga) of the Barberton greenstone belt, wavy carbonaceous laminae that entrain fine-grained detrital volcanic clasts and quartz grains occur

within banded black and white cherts as layered packets. These are interpreted as microbial matremains (Westall et al., 2006; 2011; 2015b; Homann, 2019).

*Evidence* – Black and white cherts of the 3.26 Ga Mendon Formation (Barberton greenstone belt) preserve layered packets of laminae formed of thin, wavy carbonaceous laminae up to several tens of micrometers in thickness (Trower and Lowe, 2016).

*Evidence* – The presence of 0.5 - 1 mm thick crinkly laminae within in the Saddleback Syncline in the Moodies Group (~3.22 Ga) of the Barberton Greenstone Belt are interpreted as examples of siliciclastic tidal mats (Noffke et al., 2006; Heubeck, 2009; Gamper et al., 2012; Homann et al., 2015). Terrestrial (fluvial) environments within the Moodies Group also preserve laminae interpreted as the fossilized remains of microbial mats with thickness of up to 4 mm (Homann 2019). Similar laminae occur in sandy tidal flat horizons of the 3.48 Ga Dresser Formation, Pilbara, Western Australia (Noffke et al., 2013; 2024).

*Evidence* – Numerous horizons of the Campbellrand–Malmani carbonate platform sequence are associated with layered carbonaceous materials and form bifurcating anastomosing laminae that locally form more complex fenestral (cavity-forming) stromatolitic mesostructuress (Sumner, 1997; Gandin et al., 2005).

*Argument* - Ancient laminae interpreted as having a biological origin are traceable in geologic units laterally over kilometer scales.

*Evidence* - Within the Precambrian Moodies Group on the Barberton greenstone belt, fossil mats are laterally traceable for ~15 km in a ~1000 m-thick succession in the lower part of the Saddleback Syncline and show distinct morphological adaptations to different hydrodynamic settings: (1) planar-type in coastal floodplain, (2) wavy-type in intertidal, and (3) tufted-type in upper inter- to supratidal facies (Homann et al., 2015).

*Argument* – Laminated features interpreted as MISS are preserved over billion-year timescales on Earth.

*Evidence* - Archean MISS that include laminae consistent with microbial origin are described from numerous sites, including the 3.48 Ga Dresser Formation, Pilbara, Western Australia (Noffke et al., 2013; 2024); the 3.2 Ga Moodies Group, Barberton Greenstone Belt, South Africa (Noffke et al. 2006); and the 2.8 Ga Pongola Supergroup, South Africa (Noffke et al., 2003; 2008).

*Argument* - Laminae associated with filamentous structures may be interpreted as microbial remains that have survived in the rock record for billions of years.

*Evidence* - The fossilized remains of 1-4  $\mu$ m thick biofilms are exposed on fresh fractured bedding surfaces of the Josefsdal Chert in the Barberton greenstone belt Westall et al. (2001; 2006; 2011). SEM observations reveal the presence of multiple layers of parallel filament-like structures with

constant diameter of 0.25  $\mu$ m embedded in a granular-to-smooth film interpreted as extracellular polymeric substance (Westall et al., 2006, 2011).

*Evidence* - Filamentous microstructures of carbonaceous composition were identified in a single 2 cm thick black-and-white banded chert layer in the upper Hooggenoeg Formation of the Barberton greenstone belt (Walsh and Lowe, 1985; Walsh, 1992). These thread-like or cylindrical, unbranched filaments are 0.2–2.5  $\mu$ m in diameter, up to 200  $\mu$ m long, and occur in association with carbonaceous laminae that are interpreted as microbial mats (Homann, 2019).

*Evidence*: Filaments associated with fossil mats in several localities of the Buck Reef Chert of the Barberton greenstone belt consist of hollow cylindrical filaments,  $1.2-1.4 \mu m$  in diameter and  $10-150 \mu m$  in length, that are composed of carbonaceous matter and fine pyrite, mostly occur oriented subparallel to bedding or sometimes as interwoven, tangled clumps (Walsh and Lowe, 1985; Walsh, 1992; Homann 2019).

*Evidence* - Ancient (~3.42 Ga) filamentous microfossils discovered in the Barberton greenstone belt occur in specific associations (single or in clusters) within different parts of vein microhabitats and in association with biofilms (Cavalazzi et al., 2021).

**LOW** *Argument* – The textures of ancient laminae (e.g., within some geological units of the Barberton greenstone belt) rarely include associated filamentous structures. (Note: LOW arguments indicate weaker signal strength, increasing the probability of false negative detections).

*Evidence* - Putative microfossils in the carbonaceous cherts of the Onverwacht Group are extremely rare. The systematic analysis of such materials by Walsh (1992) and Walsh and Lowe (1999) revealed that only 9 of > 400 analyzed samples contained possible microfossils of filamentous, lenticular, and spheroidal shapes.

#### **3.2.3.** Abiotic Prevalence and Signal Strength Arguments

In addition to presenting evidence and arguments for a variety of biogenic features, the LDKB seeks to present information on abiotic features that can closely mimic those produced by biology. Where possible, this context is provided to make clear the possible ambiguities of interpretation, particularly as they relate to individual types of measurements/observations. Below are some examples of abiotic prevalence and signal strength arguments that may contribute toward false positive identifications of signs of life.

Abiotic Prevalence *Argument* - Abiotic laminated structures can form in sedimentary deposits that undergo metamorphic processes.

*Evidence* - Laminated stromatolite-like or stromatoloid structures that have been interpreted as deformational features produced by metamorphic activity occur within the Isua Supracrustal Belt from southwest Greenland (Allwood et al., 2018).

*Evidence* - Pseudocolumnar and cylindroidal deformation structures that contain laminae that strongly resemble stromatolitic laminae occur in the Upper Onverwacht and Fig Tree groups (de Wit et al., 1982); however, detailed observation shows that these structures are medusoid-like surface impressions that are more consistent with abiotic formation in mudpools associated with hydrothermal activity.

Evidence - Stromatoloidal structures produced during laboratory spray deposition experiments suggest that similar structure in environments characterized by spray deposition, e.g. splash pools silica sinter-forming settings and desert varnish crusts, may also be formed by abiotic processes (McLoughlin et al., 2008), though it is worth noting that these experiments produced similar 2D rather than 3D structures.

Abiotic Prevalence Argument - Ancient laminae with hollow tufts may be formed by abiotic processes.

*Evidence* - Cavities within tufts may form by tidally driven hydraulic pumping of the ambient air trapped in pore space (Homann et al., 2016). Such structures may be misconstrued as biological in the absence of further information, e.g., *in situ* geochemistry within the tufts.

Abiotic Signal Strength *Argument* - Pseudo-tufted laminae may be generated abiotically and preserved for billions of years.

*Evidence* - Pseudo-tufted structures that occur up to 0.8 mm in height within the Middle Marker horizon, Barberton greenstone belt, may form abiotically as a result of plastic deformation caused by settling of dense particles (Hickman-Lewis et al., 2018). Such structures may be incorrectly interpreted as fossilized (micro-)tufted microbial mats.

**Abiotic Prevalence** *Argument* – Filamentous structures associated with laminae may be formed by abiotic processes.

*Evidence* – McMahon (2019) demonstrate the growth of abiotic filamentous structures in laboratory experiments mimicking plausible natural geochemical environments (i.e., "chemical gardens").

*Evidence* – McMahon et al., (2021) describe filaments associated with laminae in Jurassic mineralized veins formed in association with serpentinization. Here, filamentous structures are recognized as "dubiofossils" of uncertain origin, which may have formed by abiotic processes such as those described in chemical laboratory experiments. For example, experiments by Garcia-Ruiz (2000) under comparable alkaline conditions produced "biomimetic" microstructures of abiotic origin.

*Evidence* - In the upper Hooggenoeg Formation (Barberton greenstone belt), Walsh (2010) interpret solid filaments as having formed abiotically and thereby represent simple mineral filaments.

## 4. Discussion

## **4.1.** Considerations for interpreting laminae as biosignatures in the context of planetary exploration

1) Microbially influenced laminae can contain micro-scale information that includes detrital and/or chemically precipitated components. Some of the examples above retain evidence of trapping and binding of detrital sediments and precipitation/cementation by chemical sediments. As discussed by Reid *et al.* (2024), lamination formation is a balance between extrinsic and intrinsic forces that result in a variety of textural characteristics that may be preserved over geologic timescales. It is crucial that laminae suspected to be of biogenic origin are examined in detail at the micro scale, where observations of grain size, mineralogy, and orientation may be characterized. Spaceflight missions focused on characterizing evidence of past life should be capable of resolving these features using imaging and chemical mapping techniques. The combination of the SHERLOC (including WATSON camera) and PIXL instruments on the Mars 2020 Perseverance rover are examples of such an instrument combination capable of resolving sub-millimeter rock textures and their compositional variation (Allwood et al., 2020; Bhartia et al., 2021; Hickman-Lewis et al., 2022).

**2)** Lamination geometry alone is not diagnostic, though should be consistent with microbial biofilm morphologies if considered biogenic. Flat parallel and cross bedded/rippled laminae may form as a result of either biological or non-biological activity. Biologically influenced laminae are typically influenced by the irregular distribution and geometry of the architect biofilm community. Therefore, if encountered during current and future life exploration missions, irregular laminae should be examined in close detail as potential products of past biofilm communities.

**3) Physiochemically formed laminae may also preserve biotic or abiotic organics.** Uniform laminated sediments, particularly those containing significant clay fractions, may also retain biosignature-derived material. Though the general structural character of laminae in these cases may not be directly attributable to the activity of microorganisms, they may still act as repositories of biosignature-derived material (e.g., diagnostic organic material) concentrated from the wider sedimentary basin. Given the likelihood of elevated influx of meteoritic materials to rocky bodies during the early history of the Solar System, it is also likely that a higher contribution from carbonaceous chondrites would have delivered organic materials to sedimentary basins. Compelling evidence for impact events is recorded in the strata of the Barberton greenstone belt and Pilbara craton (Lowe et al., 1989; Glikson et al., 2004, 2016; Koeberl et al., 2015); one such horizon preserves geochemical evidence for extraterrestrial carbonaceous materials (Gourier et al., 2019).

4) Metamorphic activities and soft sediment deformation processes can create irregular lamination geometries. A number of depositional and post-depositional processes may result in

the generation of irregular laminae that resemble those produced by biofilm communities. In these cases, it is crucial to consider the wider geological context of the studied area. For example, evidence of metamorphism can be gleaned by the investigation of surrounding geologic units in search for diagnostic metamorphic textures, microtectonic trends, and high temperature and pressure mineral assemblages. A robust interpretation of the original depositional environment can assist in deciphering potential signs of life that include biogenic laminae (as is the case for most other structural potential biosignatures; Shkolyar et al., this issue). In the case of soft sediment deformation features resembling biological laminae, careful study of the broader context of such structures may lead to important insights, for example continuous parallel laminae may grade laterally into irregular displacive textures exhibiting iso- or monoclinal deformation folds or shear zones indicative of physical deformation. Such features should also be investigated with regard to their three-dimensional orientations in space, which may reveal a postdepositional deformation origin, particularly in the case where planar folds may resemble domal or conical stromatolites (e.g., Allwood et al., 2018). Additionally, traditional facies analysis applied to exposed sedimentary rocks would likely benefit towards recognizing environments susceptible of soft sediment deformation, such as steep-sloped subaqueous platforms and shelves (Tinterri et al., 2016).

5) Identifying structural characteristics of laminae alone is not sufficient to determine biogenicity. Structural observations such as lamination thickness, geometry, and lateral extent will likely fail to provide sufficient evidence for the unambiguous role of biology in their formation. Additional lines of evidence, such as the presence of carbonaceous material, diagnostic isotope fractionation patterns, and elevated trace element concentrations will be required to strengthen interpretations of possible biogenicity (see also Des Marais et al.; Parenteau et al.; Shkolyar et al., this issue). Similar constraints apply to microfossils and other structural biosignatures that may be encountered during life detection missions. It is therefore crucial that future missions include instruments capable of characterizing the composition of materials at the scale of individual laminae (cm to  $\mu$ m) or result in sample return efforts where materials can be analyzed in well-equipped laboratories on Earth.

#### 4.2. Existing Knowledge Gaps

1) Details of how microbial mats create successive mineral layers are not well explored. As indicated by Reid et al. (2024), "detailed study of environmental characteristics on multiple scales (diurnal to seasonal) must be integrated with investigation of accretion rate and initial architecture, achieved through petrographic, biogeochemical, and metabolomic approaches." Additionally, although important insights have been gained from the study of modern microbialites, further laboratory experimentation and theoretical studies are warranted to explore the various parameters that influence microbial mat lithification and upward growth, such as community composition, mineral saturation states, and the role of physiochemical processes such as evaporation and degassing.

**2) Biologically influenced lamination formation is not well studied outside of carbonate systems.** To date, the dynamics between microbial mat growth and lithification have been best explored primarily in modern carbonate precipitating systems such as the Bahamian islands and Shark Bay, Australia. These dynamics should be further explored in systems that exhibit dominant precipitation of minerals such as silica, sulfate, and iron and in a range of

environments, such as epicontinental basins, lakes, hot springs, evaporative systems, and subaerial settings.

**3) Diagenetic processes that influence the long-term preservation of biogenic laminae are not well understood.** The detection of biogenic laminae ultimately depends on whether they can be preserved in the rock record (through the so-called "taphonomic window"). It remains unclear how biologically induced laminae, along with their associated geochemical signs of life, may become altered or lost throughout early-to-late stage diagenesis. For example, the relative importance of early *post mortem* mineralization, short- and long-term diagenesis, and metamorphism should be experimentally evaluated with reference to the rock record. Further work is warranted to explore such processes in active systems where microbiology has played a role.

## **5.** Conclusions

Throughout the astrobiological and paleontological literature, researchers have utilized a wide variety of methods to characterize morphological signs of life in terrestrial samples. The development and application of ever more advanced laboratory instrumentation, miniaturized flight instruments, and analytical methods and protocols (including data treatment) are likely to be driven by the search for life at the surface and near-surface of Mars and other bodies of astrobiological significance, as well as within returned samples, for example, those collected as part of future Mars Sample Return (MSR) mission(s). The LDKB represents a growing library of key observations required to interpret possible signs of life in planetary analog materials through the use of existing instruments. By using and contributing to the LDKB, researchers will likely identify gaps in life detection capabilities and technical strategies, helping to guide the development of new tools that explicitly fill such gaps for use in different types of geological settings and, in the case of ancient signs of life, on different types of rocks and sedimentary materials.

Here we have described laminae as featured in the LDKB, which serve as key structures of interest in astrobiological exploration activities beyond Earth. Laminae form by a wide range of processes, some of which are influenced by the activity of microorganisms. On Earth, evidence of their activity can be preserved over geological time. Laminae therefore represent potential targets of interest for current and future space missions focused on life detection. Although we describe some structural attributes of laminae that may be consistent with a biological origin, such observations must be supported by other data, particularly geochemistry, to generate a robust interpretation. Overall, the LDKB is designed to collate information and help highlight knowledge gaps that remain for understanding the explicit processes that contribute to lamination formation, as well as the technological challenges that exist for the design of robotic instrument deployment on other worlds.

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