Taphonomy of the Chengjiang Biota: Using a combination of sedimentology, geochemistry and experimental taphonomy to determine preservation

Lindsay Ann MacKenzie

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TAPHONOMY OF THE CHENGJIANG BIOTA: USING A COMBINATION OF
SEDIMENTOLOGY, GEOCHEMISTRY AND EXPERIMENTAL TAPHONOMY
TO DETERMINE PRESERVATION

By

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Dissertation

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Taphonomy of the Chengjiang Biota: Using the combination of sedimentology, geochemistry and experimental taphonomy to determine preservation

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The Chengjiang Biota, found in the Early Cambrian Maotianshan Shale (Yunnan, China), represents the Cambrian “explosion of life,” providing a snapshot of the early evolution and development of complex life. This snapshot is possible because high fidelity processes transfer information from living organisms to preserved fossils. Taphonomy is everything that occurred during this information transfer from death to fossilization and discovery, including chemical and/or sedimentological processes. The Chengjiang fossils’ biology is well documented and taphonomical processes have been proposed. Tests of these models are limited because the depositional environment remains poorly understood. To fill this gap and better constrain the taphonomy, I examined the sedimentology and geochemistry of the Maotianshan Shale.

Newly identified sedimentological facies and depositional units indicated the Maotianshan Shale was deposited between the lower shoreface and the offshore transition zone during changing sea levels. The exceptionally preserved fossils are always found in fine-grained event mudstones, but not all such mudstones yield exceptional fossils. Those that do are typically found near the top of the sedimentary succession and occur in a time-transgressive pattern across the study area.

The mineralogical suite, REE distributions, and elemental ratios constrained the sediment source of the Maotianshan Shale to granitic and/or felsic volcanic protoliths. Terrigenous sediments are comparable to those of other deposits documenting the explosion of life. Organic carbon concentrations were low and sediments were anoxic during fossilization. These conditions opened a distinct taphonomic window allowing early preservation of the tissues, which largely non-mineralized, comprising thin organic carbon films. Surface analyses identified areas of concentrated trace element accumulations associated with weathering bands and fossil tissues. It is unclear when these accumulations occurred taphonomically and if they were important to the preservation of the organisms. Excellent preservation is predicated on the early formation of films or on effective mineralization of fossil tissues.

Biofilms, thin microbial communities, are known to promote fossilization. Indeed, they form quickly on organisms in aqueous taphonomy experiments, but the effect of sediments on biofilms formation was unclear. Fine-grained, felsic-sediment experiments limited biofilm growth on hard and soft tissues. If the organic films observed on Chengjiang fossils represent biofilms then other as yet unidentified processes play a critical role in the fossilization.
ACKNOWLEDGEMENTS

There are so many people who have helped and supported me through this long journey. I have had the opportunity to meet and work with an abundance of people, without whom would not have made this possible.

Mom and Dad, you have stood by me continuously and let me know this was possible.

I would like to thank my committee, all of whom believed in me even when I wasn’t sure it was possible: Nancy Hinman, George Stanley, Michael Hofmann, Sandy Ross, David Patterson.

Of course this work could not have been done without the tremendous support from our collaborator in China, Professor Chen Junyuan and his assistant/driver/procurer of things Mr. Wu.

There are several faculty and staff at UM who have aided me in this process: Jim Driver, Loreen Skeel, Christine Foster, Kallie Moore, Aaron Deskins, Jim Staub, Heiko Langner, Matt Young, Wendy Woolett, Lisa Eby, Katie George, Marc Hendrix, Kathleen Harper.

My fellow graduate students in the Geosciences Department have been an important part of my experience at UM. So many students have been there for discussion, beers, tears and celebration. A few who have stood out over the years are: Megan Rosenblatt, Liane Stevens, Adam Johnson, Warren Roe, Amy Singer, Vicky Balfour, Paul Skudder, Neal Auchetr, Elyse Rector.

Other notable people and businesses who have been imperative in me completing this project are: Mark Wilson, Alex Dutchak, Christopher Hayes, Ted Fremd and Skylar Rickbau, Matt Kohn, Paul Olin, Jill Scott, Andrew Fisher, Jessica Hawthorne, James Fivecoats, David Linzmeyer, The Rhino, Reef Missoula, Pattee Creek Market.
Taphonomy Song

This song was initially developed on a UM geology club field trip to Carters of the Moon National Monument with the help of Dr. Nancy Hinman and the students on the trip. I took it upon myself to finish it to be included in this dissertation. It should be read to the tune of the 1952 song “That’s Amore”.

When a poor fishy dies
And he lands on his side, that’s taphonomy!
When he’s coved with mud,
Becomes buried in sludge, that’s taphonomy!

Fossilization is neat, sure is sweet, what a treat!
An important part of geology!
Many parts to fit, quite a bit, like a mitt!
All parts of paleontology!

Oxygen levels will sink
Anoxia sets in real quick, that’s taphonomy
Next the microbes will come
And they’ll eat tissues, yum! That’s taphonomy!

Biofilms cover all
Rapidly, they don’t stall! That’s taphonomy!
Tissues mineralize
Muscles, skin, bones and eyes. That’s taphonomy!

Fossilization is neat, sure is sweet, what a treat!
An important part of geology!
Many parts to fit, quite a bit, like a mitt!
All parts of paleontology!

Sediments lithify
Cement’s in high supply, that’s taphonomy!
Rocks are thrust up by faults
Or displaced by basalts, that’s taphonomy!

Weathering of the rock
Makes the colors all pop, that’s taphonomy!
Excavations begin
Search for fossils within, that’s taphonomy!

Fossilization is neat, sure is sweet, what a treat!
An important part of geology!
Many parts to fit, quite a bit, like a mitt!
All parts of paleontology!
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PREFACE

The following dissertation is based on my doctoral research conducted in the Geosciences Department of the University of Montana between 2007 and 2015. The goal of this dissertation was to further our understanding of conditions responsible for the preservation of the Chengjiang Biota through a combination of sedimentology, geochemistry and experimental taphonomy. The majority of the data discussed in this dissertation were collected from multiple fossil-bearing localities of the Maotianshan Shale, in Yunnan Province, China. This dissertation is composed of six chapters. The four main chapters (Chapters 2-5) have either been published or are being prepared for publication and are each formatted for different journals.

Chapter one is a general introduction to the dissertation research and outlines the organization of the dissertation. It includes general background about Konservat-Lagerstätten, taphonomical processes and experiments, early Cambrian Burgess Shale-type deposits, as well as some background about the Maotianshan Shale and the Chengjiang Biota.

Chapter two focuses on sedimentological data collected from five different Chengjiang Biota fossil-bearing localities of the Maotianshan Shale to determine the environment in which the Maotianshan Shale was deposited and was published in *Palaeogeography, Palaeoclimatology, Palaeoecology*. This study resulted in the identification of depositional trends across the study area and taphonomical processes related to the fossilization of the Chengjiang Biota.
Chapter three examines the mineralogy and trace element geochemistry of the Maotianshan Shale to determine the provenance of the sediments and the paleoredox conditions in the sediments. This study focuses on fresh samples that are considered to preserve original chemical signatures. This paper is being prepared for publication in *Chemical Geology*.

In chapter four, inductively coupled plasma mass spectrometry (ICP-MS) was used to determine the elemental composition of the surfaces of fossil worms compared to the surrounding matrix. From this we examined how the accumulation of trace elements and biofilms likely aided in fossilization of the tissues. This paper was submitted to *Geochimica et Cosmochimica Acta* and was rejected. Additional analyses have been performed as suggested by reviewers. The manuscript is currently being revised and will be resubmitted to *Paleobiology*.

Chapter five describes a taphonomy experiment examining the effects of fine-grained sediments on the formation of biofilms on hard and soft tissues. In this study freshwater snails were introduced into a marine environment and buried in fine-grained granitic sediments and examined after different time intervals for the presence of microbial biofilms. This paper is being prepared for submission to *Palaios*.

Chapter six is a summary of the research presented in this dissertation and describes an overall taphonomical model for the preservation of the Chengjiang Biota. It summarizes all of the data collected from the dissertation and draws conclusions about the environment in which the Maotianshan Shale was deposited and the processes that led to the fossilization of the Chengjiang Biota.
There are three appendices at the end of the dissertation that consist of ongoing research that will be the focus of future publications. Appendix I is a geochemical model of early Cambrian seawater created using the geochemical modeling program PHREEQC, version 3. Major elements and redox-sensitive trace elements were included in the input file to determine when the each trace element was reduced and which mineral phases precipitated and/or dissolved at changing pH values. Appendix II is a list of the geochemical data of the fresh and weathered samples of the Maotianshan Shale. Appendix III is a review of Konservat-Lagerstätten throughout geologic history. Several different categories were collected to characterize the Lagerstätten to look for commonalities and differences between them. Ten different categories were used to compare the different Lagerstätten: 1) mineralogies of preserved fossils, 2) sediment type, 3) siliciclastic versus chemical sediments; 4) were the sediments laminated?; 5) were the fossils found in concretions?; 6) obrution or stagnation deposit?; 7) deep or shallow depositional environment?; 8) anoxia detected at time of burial?; 9) are soft tissues preserved?; 10) the geologic age (Period) of the deposit. IBM SPSS Statistics version 22 was used to calculate frequencies and hierarchical classifications.
CHAPTER 1 – INTRODUCTION

1.1 – Overview

The study of the processes that occurred to an organism from death until discovery is referred to as taphonomy (Efremov, 1940) and integrates paleontology, sedimentology and geochemistry (Behrensmeyer and Kidwell, 1985; Brett et al., 1997; Martin, 1999; Behrensmeyer et al., 2000). The sedimentary and fossil records preserve information about past environments, providing insight into past life and the taphonomical processes required for fossilization. These records are incomplete; sedimentary rocks were not continuously preserved throughout geologic time and only a fraction is exposed on the Earth’s surface. Additionally, only a small fraction of extinct organisms are preserved in the fossil record (Foote and Raup, 1996).

Fossils, found within sedimentary rocks, typically consist of skeletal elements, commonly disarticulated and fragmented, limiting the ability to interpret information about the organisms (Hendy, 2011). Exceptionally preserved fossils, those that are fully articulated and conserve soft tissues (muscles, skin, etc.), occur rarely and are important because they contain information about organisms that would not normally be preserved (Allison and Briggs, 1993). Occasionally deposits with an abundance of these exceptionally preserved fossils are found; these deposits contribute to our understanding of animal interactions and past ecosystems (Brett et al., 1997; Bottjer et al., 2002).

Commonly asked questions about these deposits are: 1) What were the sedimentological conditions at the time? 2) What were the chemical conditions in the water and sediments? 3) How did the sedimentology and geochemistry aid in the fossilization? 4) What other
taphonomical processes are responsible for the exceptional preservation of the fossils?

This dissertation focuses on the Early Cambrian Maotianshan Shale and Chengjiang Biota to address these questions.

1.1.1 – *Sedimentary Rocks*

Sedimentary rocks preserve information about past depositional processes and allow for the reconstruction of past environments, climates, and major events (e.g., extinctions) through time. Sedimentary rocks can be characterized using a facies model, focusing on the size, shape, sorting and composition of the sedimentary grains, as well as any sedimentary structures and fossils (trace or body) in the sediments to distinguish different depositional processes that occurred (Anderton, 1985; Suter, 2006). The stacking patterns of the different facies help determine the depositional environment. If correlations can be identified between outcrops, basin-wide depositional trends can be observed (Dalrymple, 2010). Chapter 2 of this dissertation uses a facies model to determine the depositional environment of the Maotianshan Shale and infer the sedimentary processes responsible for the fossilization of the Chengjiang Biota.

The geochemistry and mineralogy of sedimentary rocks reveal the source of the sediments as well as any post-depositional diagenesis (Berner, 1970; Blatt, 1985; Lanson et al., 2002). Trace and rare earth elements tend to be immobile after deposition and unaffected by weathering, recording original chemical signatures (Jones and Manning, 1994; Morford and Emerson, 1999; Rimmer, 2004; Bracciali et al., 2007). Chapter 3 focuses on the geochemistry of the Maotianshan Shale to infer the provenance of the sediments and the conditions within the sediments at time of burial. Appendix I consists
of a geochemical model of Cambrian seawater to infer redox reactions and mineral phases present at changing pe values.

1.1.2 – Fossils

The discoveries of complete, articulated skeletons and soft-bodied fossils have broadened our understanding of the evolution of life throughout geologic time. Soft tissue are rarely fossilized but when found give clues about organisms and ecosystems that cannot be determined from skeletal elements alone. There are many invertebrate taxa that do not possess hard parts or exoskeletal elements; these forms would be unknown without the preservation of soft tissues. The discovery of a complete conodont fossil in the Granton Shrimp Bed (Aldridge et al., 1993) is an example of this; prior to its discovery conodonts were an enigmatic animal known only by their oral elements. The newly discovered body fossils revealed a new type of body plan and added to the understanding of the origins of chordates and vertebrates (Donoghue et al., 2000).

Many factors contribute to soft-tissue preservation including the chemical composition of the tissues, the composition of the depositional setting and the enclosing sediments and the chemical processes that occurred prior to, during, and post burial (Allison, 1986; Briggs, 2003). Chapters 2 and 3 infer the sedimentological and geochemical processes responsible for the preservation of the Chengjiang Biota. Chapter 4 presents the results of the chemistry of soft tissues of fossilized worms (*Palaeoscolex sinensis*), determined using laser ablation inductively coupled plasma mass spectrometry (LA-ICP-MS), to uncover the factors for their preservation.
1.2 – The Study of Taphonomy

Taphonomical studies are becoming increasingly of interest with the discoveries of fossil localities with an abundance of articulated and/or soft-bodied and lightly sclerotized organisms (Behrensmeyer et al., 2000). These fossil tissues are preserved via multiple pathways, some examples include the phosphatized metazoan embryos in the Precambrian Doushantuo Formation (Xiao et al., 1998; Xiao and Knoll, 1999), the pyritized invertebrates of the Devonian Hunsrück Slate (Bartels et al., 1998; Etter, 2002a), and the original color in insects and vertebrates from the Eocene Messel Shale (Wuttke, 1992; McNamara, 2013). The focus of this dissertation is to determine the taphonomical processes responsible for one such locality, the Chengjiang Biota.

1.2.1 Experimental Taphonomy

Taphonomy experiments model specific factors responsible for the mineralization of different tissues present in the fossil record (Briggs and Kear, 1993; Briggs, 1995). In natural systems there are many unknown factors responsible for fossilization. In a laboratory setting each condition (temperature, pH, salinity, nutrients, etc.) can be controlled and tested to observe its role in preserving soft tissues (Raff et al., 2014). These experiments highlight the rates at which fossils form and the important role microbes play in early mineralization of the tissues (Briggs, 2003; Darroch et al., 2012); if the tissues are not rapidly isolated under low oxygen conditions and mineralized they will not be preserved (Briggs and Kear, 1994; Briggs, 2003). Chapter 5 describes a taphonomy experiment examining the effects sediments have on biofilm formation and the preservation of hard and soft tissues.
1.3 – Fossil-Lagerstätten

A Lagerstätte (plural Lagerstätten) is a German term for a geological deposit of economic interest. Thus a fossil Lagerstätte is a geologic deposit containing an abundance of fossils and/or exceptionally preserved fossils that are valuable to the scientific community (Seilacher, 1970; Seilacher 1990). Fossil-Lagerstätten are found throughout much of the geologic record and can consist of a single geologic interval, like the ~4 cm thick Beecher’s Trilobite Bed (Etter, 2002b) or represent significant geologic time like the Pioche Shale found in sediments ranging from Early to Middle Cambrian age (Lieberman, 2003). These deposits are important as they yield a large amount of information about past geologic environments.

Fossil-Lagerstätten can be divided into two groups: Konzentrat- and Konservat-Lagerstätten. Konzentrat-Lagerstätten are concentration deposits where large amounts of fossils are preserved. The fossils are generally concentrated due to a trap (e.g., a cave or below storm wave base under anoxic conditions) where the fossils will not be destroyed, accumulate in large numbers, and can represent a large amount of time (Seilacher, 1990). In these deposits the fossils are not always well-preserved, but the abundance preserved in Konzentrat-Lagerstätten provides a great deal of information about past forms and populations (Nudds and Selden, 2008). One example of this is the Middle Holocene Konzentrat-Lagerstätte on Mauritius Island; this fossil deposit consists of an abundance of bone fragments, invertebrates, plants and microfossils. Although none of the fossils show exceptional preservation, the assemblage is a representation of the ecosystem prior to the arrival of humans (Rijsdijk et al., 2009).
1.3.1 Konservat-Lagerstätten

Fossils found within Konservat-Lagerstätten show exceptional preservation, often preserving soft tissues (skin, muscles, etc.; Allison and Briggs, 1993) and, occasionally, original pigments (McNamara, 2013). There are over 90 Konservat-Lagerstätten in the fossil record containing a rare abundance of fully articulated and/or soft-bodied and lightly sclerotized organisms. These deposits provide information about past organisms, their diversification and evolution, and the ecosystem in which they lived (Nudds and Selden, 2008; Bottjer et al., 2002); much of this information would not normally be preserved. The presence of these exceptional fossils expands our knowledge of the past. For example, the diversification of metazoan life would not be well understood had Cambrian Lagerstätten, such as the Burgess Shale and Sirius Passett, which preserve soft-bodied organisms, not been found (Conway Morris, 1985; Conway Morris et al., 1987; Hagadorn, 2002).

There are two main divisions of Konservat-Lagerstätten: stagnation and obrution (Seilacher et al., 1985). In stagnation deposits the organisms are preserved due to low oxygen conditions in stagnant and/or hypersaline environment; these fossils were not rapidly buried. An excellent example of a stagnation Lagerstätte is the Posidonia Shale/Holzmaden in Germany. The Posidonia Shale contains exceptionally preserved fish, marine reptiles and cephalopods that were preserved in a stratified, anoxic basin and slowly buried by organic black shales (Röhl, 2001; Etter and Tang, 2002; Frimmel et al., 2004). Obrution deposits are those in which the organisms are rapidly buried, typically by fine-grained sediments, the best known of which is the Burgess Shale in Canada. In
this Middle Cambrian fossil locality, the organisms were swept from off of the Cathedral escarpment by fine-grained, storm-derived flows below wave base where they were preserved under anoxic conditions (Conway Morris, 1999; Gould, 2000).

Konservat-Lagerstätten can also be formed by a combination of obrution or stagnation. In the Jurassic Voulte-sur-Rhône Lagerstätte cephalopods and other invertebrates were preserved in great detail aided by anoxic bottom waters, episodic sedimentation, and possible hydrothermal fluids (Etter, 2002c; Charbonnier et al., 2010). Appendix III is a summary of frequency data and hierarchical classifications of many of the Konservat-Lagerstätten found globally from the Precambrian through the Cenozoic.

1.4 Burgess Shale-Type Deposits

Early Paleozoic Burgess Shale type (BST) Lagerstätten are of particular interest. BST deposits are obrution Konservat-Lagerstätten that share similarities with the famous Middle Cambrian Burgess Shale fossil Lagerstätten in Canada (Conway Morris, 1989). The Burgess Shale, found in British Columbia, Canada, was the first Lagerstätten to depict the “Cambrian explosion” (Gould, 1989; Conway Morris, 1999). There are over 20 BST deposits around the globe, the majority of which are found in the Cambrian, with a few found in the Ordovician and Silurian (Table 1.1; Collins et al., 1983; Butterfield, 1995; Hagadorn, 2002; Han et al., 2008; Zhang et al., 2008). BST deposits contain diverse fossil assemblages illustrating the origination, diversification and evolution of metazoans. The fossils are largely non-mineralized in fine-grained siliciclastic sediments. The majority of the fossils are preserved as carbonaceous compressions (Gaines et al., 2008), but other mineralizing pathways have also been identified (e.g.,
Table 1.1 – Some of the Burgess Shale-type (BST) localities found around the globe

<table>
<thead>
<tr>
<th>Name</th>
<th>Location</th>
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<td>China</td>
<td>Early Cambrian</td>
</tr>
<tr>
<td>Chengjiang Biota</td>
<td>China</td>
<td>Early Cambrian</td>
</tr>
<tr>
<td>Zawiszany Formation</td>
<td>Poland</td>
<td>Early Cambrian</td>
</tr>
<tr>
<td>Sirius Passet</td>
<td>Greenland</td>
<td>Early Cambrian</td>
</tr>
<tr>
<td>Indian Springs/Poleta Formation</td>
<td>USA</td>
<td>Early Cambrian</td>
</tr>
<tr>
<td>Guanshan Fauna</td>
<td>China</td>
<td>Early Cambrian</td>
</tr>
<tr>
<td>Sinsk Biota</td>
<td>Russia</td>
<td>Early Cambrian</td>
</tr>
<tr>
<td>Emu Bay Shale</td>
<td>Australia</td>
<td>Early Cambrian</td>
</tr>
<tr>
<td>Kinzers Formation</td>
<td>USA</td>
<td>Early Cambrian</td>
</tr>
<tr>
<td>Parker Slate</td>
<td>USA</td>
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</tr>
<tr>
<td>Lantham Shale</td>
<td>USA</td>
<td>Early Cambrian</td>
</tr>
<tr>
<td>Mount Cap Formation</td>
<td>Canada</td>
<td>Early to Middle Cambrian</td>
</tr>
<tr>
<td>Pioche Shale</td>
<td>USA</td>
<td>Early to Middle Cambrian</td>
</tr>
<tr>
<td>Kaili Formation</td>
<td>China</td>
<td>Middle Cambrian</td>
</tr>
<tr>
<td>Wheeler Shale</td>
<td>USA</td>
<td>Middle Cambrian</td>
</tr>
<tr>
<td>Spence Shale</td>
<td>USA</td>
<td>Middle Cambrian</td>
</tr>
<tr>
<td>Marjum Formation</td>
<td>USA</td>
<td>Middle Cambrian</td>
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<tr>
<td>Burgess Shale</td>
<td>Canada</td>
<td>Middle Cambrian</td>
</tr>
<tr>
<td>Conasauga Formation</td>
<td>USA</td>
<td>Middle Cambrian</td>
</tr>
<tr>
<td>Stephen Formation</td>
<td>Canada</td>
<td>Middle Cambrian</td>
</tr>
<tr>
<td>Huaqiao Formation/Paibi Section</td>
<td>China</td>
<td>Middle Cambrian</td>
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<tr>
<td>Eldon Escarpment</td>
<td>Canada</td>
<td>Middle Cambrian</td>
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<tr>
<td>Fezouata Formation</td>
<td>Morocco</td>
<td>Early Ordovician</td>
</tr>
<tr>
<td>Walcott-Rust Quarry</td>
<td>USA</td>
<td>Late Ordovician</td>
</tr>
<tr>
<td>Soom Shale</td>
<td>South Africa</td>
<td>Late Ordovician</td>
</tr>
</tbody>
</table>

pyrite, phosphate; Gaines et al., 2012). Not only do these deposits share similar biological and depositional conditions, but they may also share similar taphonomical conditions specific to the Early Paleozoic environment (Gaines et al., 2008; Gaines and Droser 2010; Lin and Bricker, 2010; Gaines et al., 2012).

1.5 The Maotianshan Shale and the Chengjiang Biota

This dissertation focuses on the Chengjiang biota in Yunnan, China, one of the earliest BST Lagerstätten. The Chengjiang biota is a predominantly non-mineralized
Figure 1.1 – Stratigraphic column of Early Cambrian sediments in Yunnan, China (modified from MacKenzie et al., 2015).
fossil fauna found within the early Cambrian (Atdabanian stage, ~520 Ma) Maotianshan Shale (Chen and Lindström 1991; Hu, 2005; Hou et al., 2007). The Maotianshan Shale as the middle member of the Yu’anshan Formation (Figure 1.1; Hu, 2005; Zhao et al., 2012). The bottom member is a carbonaceous black shale. The upper member consists of a siltstone. The Shiyantou Formation, consisting of fissile black shales, underlies the Yu’anshan formation. The Shuiyantou Formation is underlain by the Zhuijiajing Formation phosphorites. The Yu’anshan Formation is overlain by the interbedded sandstones and siltstones of the Hongjingshao Formation (Zhu et al., 2001; 2005a).

The Maotianshan Shale is found exposed in outcrops around Kunming in Yunnan province (Figure 1.2). It consists of interbedded sandstones, siltstones, and mudstones, with the exceptionally preserved fossils found in clean mudstones (Babcock and Zhang, 2001). When freshly exposed, the Maotianshan Shale sediments are grey to black in color, and the fossils are difficult to see. When weathered, the rocks are a tan to yellow color, and the fossils are much more visible (Bergström, 2001; Zhu et al, 2005b; Forchielli, 2013). The majority of the exposed Maotianshan Shale deposits are weathered, and few geochemical analyses have been performed on these rocks (e.g., Gabbott et al., 2004; Zhu et al., 2005b; Forchielli et al., 2014).

The depositional environment of the Maotianshan Shale is still under debate; it has been interpreted as a shallow, normal marine environment (Pu et al., 1992), a partially closed embayment (Chen and Erdtmann, 1991), a nearshore and foreshore shelf setting (Pu et al., 1992), fine-grained suspension settle-out in an offshore environment (Zhu et al., 2001; Hu, 2005) and a proximal offshore to distal shoreface (Zhao et al.,
Figure 1.2 – Locations of Early Cambrian rocks near Kunming, Yunnan, China (modified from Hu, 2005). Localities visited in this dissertation are noted.
2009). Additionally, the lack of continuous exposures and of marker beds had made correlations of stratigraphic layers and fossil occurrences between localities difficult.

The Chengjiang biota is one of the earliest occurrences of BST fossils, pushing back the timing of the “Cambrian Explosion” by at least 15 Ma (Hagadorn, 2002; Steiner et al., 2005). The fossils are preserved with exceptional fidelity and represent all modern phyla as well as some Ediacara holdover taxa (Chen and Erdtmann, 1991; Hou et al., 2007). The fossils preserve mineralized and non-mineralized elements including soft tissues, gills, muscles, and other organs (Chen and Erdtmann, 1991; Hou et al., 1991; Hou et al., 2005). The diversity of the Chengjiang biota represents infaunal, epifaunal and nektobenthic organisms, providing information on community interactions as well the diversification and evolution of several taxonomic forms (Vannier and Chen, 2005; Chen et al., 2007; Hou et al., 2007; Chen, 2009; Zhao et al., 2009; 2010; 2012; 2014). The fossils are predominantly preserved as thin carbonaceous films, but pyrite, apatite and clay minerals have also been identified (Gabbott et al., 2004; Zhu et al., 2005b; Gaines et al., 2008). The fossils are generally considered to have been rapidly buried by event beds (turbidites, storm-generated flows) and promptly mineralized under anoxic conditions, similarly to other BST deposits (Gaines et al., 2008; 2012). The exact taphonomical processes responsible for the fossils remain unclear.

In this dissertation I use a multidisciplinary approach to better understand the depositional processes of the Maotianshan Shale and the taphonomy of the Chengjiang Biota. The major goals of this dissertation are to answer the following questions:

- What are the sources of the sediments of Maotianshan Shale?
• What was the depositional environment of the Maotianshan Shale? How did this aid in preserving the Chengjiang Biota?

• What chemical conditions (paleoredox, temperature, etc.) were present in the sediments and how did they preserve the Chengjiang biota?

• What can the trace element geochemistry of the soft tissues tell us about the taphonomy of the fossils?

• How do sediments affect biofilm formation and fossil preservation?

A combination of sedimentology and stratigraphy, mineralogical and geochemical analyses, and experimental taphonomy were used to answer these questions. These results are discussed in the following chapters.

1.6 – References


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CHAPTER 2 – STRATIGRAPHIC CONTROLS OF SOFT-BODIED FOSSIL OCCURRENCES IN THE CAMBRIAN CHENGJIANG BIOTA LAGERSTÄTTE, MAOTIANSHAN SHALE, YUNNAN PROVINCE, CHINA

Lindsay A. MacKenzie, Michael H. Hofmann, Chen Junyuan, Nancy W. Hinman

2.1 – Abstract

The Early Cambrian Maotianshan Shale is exposed near Kunming, China, is one of the oldest and best-preserved Konservat Lagerstätten and contains the Chengjiang Biota. Although the fossils have been well-studied the depositional processes and stratigraphic relationships among the fossil-bearing localities are poorly understood, largely due to limited continuous exposures and structural complexity in this area.

In this study we use a detailed facies and stacking pattern analysis to delineate the depositional history and stratigraphic relationship of the Maotianshan. Sedimentologic observations from five prolific fossil localities of the Chengjiang Biota have yielded six distinct facies within the Maotianshan Shale. The three coarse-grained facies, composed of siltstones and very fine sandstones (F1-F3), contain sedimentary structures including hummocky cross stratification and combined flow ripples that are consistent with deposition within storm wave base. The fine grained facies, composed of interbedded silty mudstones and clay-rich mudstones (F4-F6), were deposited below wave base and represent episodes of lower storm frequency and pelagic sedimentation.

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These six facies have specific stacking patterns, indicating the presence of five distinct depositional units. Each of those depositional units is bound on its upper and lower contact by a flooding surface. Fining-upward successions near the base of each stratigraphic unit are interpreted as short-lived marine transgressions; these are overlain by coarsening-upward stacked facies interpreted as a period of shoreline progradation.

The fossils of the Chengjiang fauna are exclusively found in the coarsening-upward part of the depositional units. The oldest fossil-bearing intervals are associated with the most western sections in the study area. In contrast, the youngest fossil-bearing intervals are found in the most eastern sections. This time-transgressive relationship is interpreted to reflect a genetic relation between distinct sedimentary processes and environments and the burial and preservation of the Chengjiang Biota.

2.2 – Introduction

*Konservat-Lagerstätten* deposits are important snapshots into past environments that provide abundant geological and biological information (Seilacher et al., 1985; Bottjer et al., 2002). These deposits often contain soft-bodied fossils, which require specific taphonomic conditions for preservation (e.g., Seilacher et al., 1990; Allison, 1988a,b; Allison and Briggs, 1991; Brett et al., 1997; Briggs, 2003).

In the early Paleozoic there are multiple fossil *Lagerstätten* that, based on their general similarities in taxonomy and depositional history to the Burgess Shale, are termed Burgess Shale-type (BST) deposits (Conway Morris, 1989; Butterfield, 1995; Gaines et al., 2008). The Chengjiang Biota (Maotianshan Shale) in Yunnan province, China, is these oldest of the BST *Lagerstätten* deposits, depicting the emergence and
Figure 2.1 – a) Map showing the exposures of Early Cambrian rocks. The black rectangle highlights the study area, shown in detail in Figure 2.8. Location of exposures and ancient landmasses based on Geological Mineral Bureau of Yunnan Province (1995), Zhu et al. (2001a) and Hu (2005); b) Simplified stratigraphic column of the Early Cambrian strata in central Yunnan.

diversification of metazoan life during the Early Cambrian. It contains exceptionally well-preserved, autochthonous, soft-bodied fossils that are found in situ (e.g., Hou et al., 2005; Dornbos and Chen, 2008) or show signs of local transport (Zhang et al., 2006; Zhang and Hou, 2007), unlike the Burgess Shale (Hagadorn, 2002), and are considered to represent the original composition and diversity of the marine community (Chen and Zhou, 1997; Chen et al., 2007; Hou et al., 2007; Dornbos and Chen, 2008; Zhao et al., 2009; Zhao et al., 2010; Zhao et al., 2012; Zhao et al., 2014).
Although the Maotianshan Shale has been studied in great detail and many depositional hypotheses have been proposed (e.g., Hou, 1987; Chen and Lindström, 1991; Pu et al., 1992; Babcock et al., 2001; Hu, 2005; Dornbos and Chen, 2008; Zhao et al., 2009; Zhao et al., 2012), little attention has been given to the detailed sedimentological and stratigraphic relationships amongst the various fossiliferous localities.

In this study we present a new facies framework from five prolific fossil localities of the Chengjiang biota located to the west and south of Kunming (Figure 2.1a). The fossil-bearing facies are found near the top of each stratigraphic interval in a coarsening-upward succession. Based on the facies characteristics, such as sedimentary structures and trace fossil assemblages, and facies associations observed in the sediments the Maotianshan Shale is interpreted as having been deposited primarily within the offshore transition zone, between the lower shoreface and the shallow shelf. Based on the distinct stacking of facies throughout the study area, we identified five distinct depositional units that reveal a time-transgressive relationship of facies development and soft-bodied fossil occurrence between the different Lagerstätten localities.

2.3 – Geological Setting

The Early Cambrian stratigraphy in Yunnan province is complex and has resulted in a variety of stratigraphic hierarchies for the Maotianshan Shale (Zhu et al., 2001a). In this study we follow the organization of Hu (2005) and Zhao et al. (2012), both defining the Maotianshan Shale as the middle member of the Yu’anshan Formation; it is underlain by a lower black shale member and overlain by an upper siltstone member (Hu, 2005;
The exceptionally well-preserved fossils of the Chengjiang biota are found primarily in fine-grained mudstones in the middle to upper parts of the Maotianshan Shale, although some fossils are found in the overlying siltstone member (Babcock and Zhang, 2001). Fossils preserved in the Maotianshan Shale are considered to represent the original composition and diversity of the marine community at each of the various localities (Zhao et al., 2009; Zhao et al., 2012; Zhao et al., 2014). More controversial is the interpretation of the depositional environment in which the Chengjiang fauna lived and where it was eventually deposited. Previously published interpretations for the depositional environment of the Maotianshan Shale include: (1) a partially restricted marine basin (Chen and Erdtman, 1991), (2) turbiditic storm deposition in a basin ranging in depths of tens to hundreds of meters (e.g. Hou 1987; Zhao et al., 2012), (3) foreshore and nearshore environments (Pu et al., 1992), and (4) fine-grained suspension settle-out deposition in an offshore environment (Zhu et al., 2001a; Hu, 2005).

The Yu’anshan Formation is underlain by the black shales of the Shiyantou Formation which itself is underlain by the phosphorite-bearing rocks of the Zhuijiajing Formation, both serving as good marker beds in the study area (Zhu et al., 2001a, Zhu et al 2005). Interbedded sandstones and siltstones of the Hongjingshao Formation overlie the Yu’anshan Formation (Figure 2.1b). In the localities near Haikou the Maotianshan Shale is disconformably overlain by the Devonian Haikou Formation (Zhu et al., 2001a; Zhao et al., 2009).

The depositional ages of the Early Cambrian sedimentary rocks are poorly constrained and primarily stem from relative dating using small shelly fossil (SSF) and
trilobite biozones. The Zhuhijiang Formation is characterized by the *Anabarites trisulcatus-Protohertzina anabarica* SSF assemblage (basal Meishucunian) and the Shiyantou Formation by the *Sinosachites flabelliformis-Tannuolina zhangwentangi* SSF assemblage (upper Meishucunian) (Qian and Bengston, 1989; Steiner et al., 2007; Rozanov et al., 2008). The lower black shale member of the Yu’anshan Formation is characterized by the *Parabadiella* biozone, late Atdabanian (530-524 Ma), and the Maotianshan Shale and the upper siltstone are characterized by the *Eoredlichia* and *Wutangaspis* biozones (~520 Ma) (Zhu et al., 2003; Rozanov et al., 2008). The overlying Hongjingshao Formation is characterized by the *Drepanuroides-Yilingella* trilobite zone (Botomian) (Rozanov et al., 2008).

### 2.2.1 – Fossil Assemblages

The study area (Figure 2.1a) comprises five localities that have good sedimentary exposure and are known to yield abundant soft-bodied fossils (Zhang et al., 2001; Hu, 2005; Zhao et al., 2012). Three distinct, geographically separated fossil assemblages have been previously identified in this study area based on the relative abundance and diversity of the fossils. The first of these is the Anning assemblage, represented in this study by the Shankou locality, which is dominated by the bradoriidid arthropod *Kunmingella*, the trilobite *Wutingaspis* and the brachiopod *Obolus* (Zhang et al., 2001; Zhu et al., 2001b). The second is the Haikou assemblage, represented by the Jianshan and Ercai localities, and is dominated by the bradoriidid arthropod *Kunmingella* and the priapulid *Cricocosmia* (Zhang et al., 2001; Steiner et al., 2005). This third and final fossil assemblage is the Chengjiang assemblage, represented by the Xiaolantian and
Maotianshan localities, and is dominated by the bivalved arthropods *Kunmingella* and *Isoxys* and the arthropod *Naraoia* and also contains an abundance of the enigmatic, medua-like *Eldonia* (Hou et al., 1991; Zhang et al., 2001).

2.4 – Methods

The results presented here are largely based on field observations made between 2009 and 2013. Detailed stratigraphic sections were measured at five localities in the greater Kunming area, Yunnan, China, where scores of soft-bodied fossils of the Chengjiang biota have been previously found and described (e.g., Chen and Zhou, 1997; Chen, 2004; Hou et al., 2007; Figure 2.1a). At the Jianshan, Ercai, Xiaolantian and Maotianshan localities sedimentological data was recorded on a millimeter to centimeter scale. The section at Shankou was exposed along a narrow trench, preventing such detailed measurements, and was measured at decimeter scale. The different scales used to record sedimentological changes had no influence on the overall results presented in this paper, as stratigraphic correlations and interpretations of depositional environment were made at a decimeter scale. The Ercai composite section is a mosaic of three individual stratigraphic sections separated by densely covered areas that were structurally restored to their respective stratigraphic positions in the composite section.

2.5 – Ichnofauna

Bioturbation in the study area is mainly confined to four different types of traces that can be found throughout the Maotianshan Shale spatially and stratigraphically.
Of those traces *Planolites* is the most common (Figure 2.2a). Morphologic characteristics of this trace include a round (elliptical) shape, they are relatively straight and unbranched, and are bedding plane parallel to slightly oblique. They occur most commonly in silty mudstones and muddy siltstones and are always infilled with coarser sediments from the overlying facies (often sandy siltstone or very fine sandstone). *Planolites* is not linked to one particular depositional environment, but they have been identified in sediments deposited between the proximal lower shoreface and the distal offshore (Pemberton, 1992). In addition, *Planolites* traces have been previously reported in the Maotianshan Shale (Luo et al., 1994).

*Arthrophycus* traces are also common in the Maotianshan Shale (Figure 2.2b). They are large, branching traces that often cut slightly oblique to bedding. They most commonly occur in silty mudstones and muddy siltstones. They are often lined and, like *Planolites*, are infilled with coarser sediments. *Arthrophycus* traces were first identified in the Maotianshan Shale by Luo et al (1994) and are often found in sediments ranging from the lower shoreface to the offshore environment (Uchman, 1998).

*Chondrites* traces (Figure 2.2c) appear to be less common in the Maotianshan Shale than *Planolites* and *Arthrophycus*. They are small (sub-mm in diameter), rounded branching burrows that occur in small clusters within silty mudstones. They are infilled with mudstone and silty mudstone. *Chondrites* traces can occur in a wide range of depositional environments, but are often found in proximal lower shoreface to proximal offshore deposits (Pemberton, 1992).

The fourth type of bioturbation observed in the Maotianshan Shale was interpreted as *Helminthopsis* (Figure 2.2d). These trace fossils are small, round, burrows
Figure 2.2 – Polished slabs showing examples of the four ichnotaxa in the Maotianshan Shale: a) *Planolites* (shown in circle; 28/06/09-02); b) *Arthrophycus* (17/07/09-01); c) *Chondrites* (11/07/09-03); d) *Helminthopsis* (01/09/11-04). All samples are from the Xiaolantian section.
that occur in laminated silty mudstone facies and are infilled with muddy sediment. They are recognized by a mottled appearance of the bioturbated sediment. Like *Chondrites*, *Helminthopsis* burrows are often associated with fine-grained deposits in the lower shoreface to proximal offshore environments (Pemberton, 1992).

### 2.6 – Description of Sedimentary Facies of the Maotianshan Shale

The deposits of the Maotianshan Shale are best described by six sedimentary facies. Distinct lithologies, ichnological assemblages, fossil occurrences, and sedimentary structures are characteristic of each facies, which are described in detail below and summarized in Table 2.1. The bioturbation index used is based on the classification scheme of Droser and Bottjer (1986). The facies are interpreted as having been deposited in different parts of a marine shoreface and shelf setting. For our nomenclature we adapted the shoreface interpretation of Reading and Collinson (1996); the shoreface defined as the area below the shoreline ranging between the mean low water line and mean fair-weather wave base, the offshore-transition zone defined as the area between mean fair-weather and mean storm weather wave base, and the shelf (offshore) as the depositional environment below mean storm wave base.

#### 2.6.1 – Facies 1 (F1) – Massive siltstone/sandstone (*Figure 2.3a, 2.3a’*)

*Description:* F1 includes hummocky cross-stratified (HCS) sandy siltstones and very fine sandstones. Bed thickness is variable ranging from 1-12 cm. Most of the upper boundaries are hummocky and swaley, and the internal laminations dip at very low
Table 2.1 – Characteristics of the six facies identified in the Maotianshan Shale.

<table>
<thead>
<tr>
<th></th>
<th>Facies 1</th>
<th>Facies 2</th>
<th>Facies 3</th>
<th>Facies 4</th>
<th>Facies 5</th>
<th>Facies 6</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Lithology</strong></td>
<td>siltstone and sandstone</td>
<td>siltstone and sandstone</td>
<td>siltstone and sandstone</td>
<td>silty mudstone and muddy</td>
<td>mudstone</td>
<td>mudstone</td>
</tr>
<tr>
<td><strong>Grain size</strong></td>
<td>very fine sand and coarse silt; minor fine sand</td>
<td>very fine sand and coarse silt; minor fine sand</td>
<td>very fine sand and coarse silt; minor fine sand</td>
<td>silt and clay</td>
<td>silt and clay</td>
<td>silt and clay</td>
</tr>
<tr>
<td><strong>Sorting</strong></td>
<td>moderately sorted</td>
<td>moderately sorted</td>
<td>moderately sorted</td>
<td>moderately sorted</td>
<td>n.a</td>
<td>n.a</td>
</tr>
<tr>
<td><strong>Rounding</strong></td>
<td>Sub-rounded</td>
<td>Sub-rounded</td>
<td>Sub-rounded</td>
<td>n.a</td>
<td>n.a</td>
<td>n.a</td>
</tr>
<tr>
<td><strong>Bed thickness</strong></td>
<td>1-12 cm</td>
<td>5-20 cm</td>
<td>3-70 cm</td>
<td>0.5 cm to &gt; 1 m</td>
<td>0.5 cm to &gt; 1 m</td>
<td>0.5-5 cm thick</td>
</tr>
<tr>
<td><strong>Laminae thickness</strong></td>
<td>n.a</td>
<td>1-3 mm</td>
<td>n/a</td>
<td>1-3 mm</td>
<td>1-2 mm</td>
<td>1-2 mm</td>
</tr>
<tr>
<td><strong>Sed structures</strong></td>
<td>hummocky cross stratification and slightly normal grading when present</td>
<td>normally graded; laminations, cross-bedding, ripples</td>
<td>normally graded; laminations, cross-bedding, ripples</td>
<td>laminations; occasional low-angle cross beds</td>
<td>laminations</td>
<td>faint laminations to structureless</td>
</tr>
<tr>
<td><strong>Upper contact</strong></td>
<td>wavy</td>
<td>gradational, sometimes erosional</td>
<td>sharp, occasionally wavy</td>
<td>sharp</td>
<td>sharp</td>
<td>sharp</td>
</tr>
<tr>
<td><strong>Lower contact</strong></td>
<td>sharp to wavy</td>
<td>sharp; occasional tool marks and groove casts</td>
<td>sharp; occasional tool marks and groove casts</td>
<td>sharp, sometimes gradational</td>
<td>sharp</td>
<td>sharp</td>
</tr>
<tr>
<td><strong>Bioturbation index</strong></td>
<td>BI = 1</td>
<td>BI = 2</td>
<td>BI = 1</td>
<td>BI = 2</td>
<td>BI = 1-4</td>
<td>BI = 1</td>
</tr>
<tr>
<td><strong>Fossil present?</strong></td>
<td>no</td>
<td>no</td>
<td>few exoskeletal fragments</td>
<td>exoskeletal fragments</td>
<td>exoskeletal and chitinous fragments, algae</td>
<td>Chengjiang biota</td>
</tr>
<tr>
<td><strong>Depositional processes and environment</strong></td>
<td>storm-derived deposits</td>
<td>turbidites</td>
<td>combined flows</td>
<td>distal turbidites</td>
<td>pelagic/background sedimentation</td>
<td>muddy event beds</td>
</tr>
</tbody>
</table>
angles that follow the hummocky or swaley topography. The lower contacts of both the sandstone and coarse siltstone beds are sharp or wavy. Bioturbation is absent (BI = 1).

**Interpretation:** F1 is interpreted as storm-generated event deposits; HCS beds are most commonly found deposited on the lower shoreface and the offshore transition zone (e.g. Dumas and Arnott, 2006). Wavy upper and lower surfaces as well as small-scale (<1 cm ripple crests), symmetric ripples related to some of these beds support this rendition and are interpreted to be the result of oscillatory waves with shorter wave periods and likely lower oscillatory velocities during small storm events or waning storm conditions.

### 2.6.2 – Facies 2 (F2) – Amalgamated Ripple-Laminated Sandstones/Siltstones (Figure 2.3b, 2.3b’)

**Description:** F2 primarily consists of very fine sandstone/siltstone couplets that are either massive or symmetrical ripple laminated and have wave-reworn erosional surfaces. These couplets are separated by an erosional contact that, on occasion, is accentuated by a thin mud drape. Beds are 3-70 cm thick; the thicker beds are typically coarser, composed of very fine sandstone, while the thinner beds are composed of siltstone. The basal contact of the bottom flow is always sharp, occasionally containing grooves or flutes. The middle and upper contacts of the amalgamated beds are either sharp or wavy. Bioturbation is absent in this facies (BI=1).

**Interpretation:** F2 is interpreted as combined flow deposits and wave-modified turbidites, consisting of steady flow and oscillatory flow components, which are found deposited between the lower shoreface and the offshore transition zone (Myrow et al.,
Figure 2.3 – The six major lithofacies within the Maotianshan Shale: a) F1, hummocky cross stratification (HCS) underlain by parallel bedded sandy siltstones (F4) and draped with silty mudstones (F5), Jianshan section; a’) annotated sketch of F1 seen in (a); b) F3, two wave-modified turbidites forming a combined flow, polished slab of the flow inset, Xiaolantian section; b’) annotated polished slab of the lower flow pictured in (b), note the parallel laminae truncated by wave-reworked foresets; c) F2, turbidite (showing only Ta-Tb portion of the Bouma succession), Xiaolantian section; d) F4, distal turbidite, Ercai section; e) F5 pelagic mudstones, Jianshan section; f) F6, event mudstones, Jianshan section (annotated image in Figure 2.5).
The wavy contacts between many of the beds are interpreted as minor scouring from syn-depositional wave-reworking of genetically-related, unidirectional and oscillatory flows as they might occur in storm-generated density currents in combination with gravity currents. In contrary, the thin mud drapes between beds suggest a short, temporal separation between deposition of some of the couplets, allowing for the mud-sized fraction to settle out of suspension. This suggests that the primary density currents might not have been directly triggered by storms in the basin, but by other mechanisms including slope failure in a more proximal position of the depositional system or large, high-density flood events in the source area. Subsequent wave modification, as indicated by the wavy upper and lower contacts, occurred shortly after primary deposition.

2.6.3 – Facies 3 (F3) – Normally graded sandstones/siltstones (Figure 2.3c)

Description: This facies consists of normally graded fine and very fine sandstones and siltstones. Individual beds are 5 to 20 cm thick. The basal contact is always sharp and sometimes exhibits tool marks and groove casts. The upper contact can be sharp or wavy. In general, thicker sandy beds are composed of massive sandstones at the base, grading into laminated sandstones and cross-bedded sandstones; thinner beds grade from laminated and ripple laminated very fine sandstones to siltstones. Bioturbation is largely absent (BI = 1); only one Planolites burrow was recognized in all of the sections.

Interpretation: F3 shows all sedimentary characteristics of Bouma-type turbidites (Bouma, 1962). The massive beds are interpreted as Bouma Ta beds, laminated deposits as Tb, ripple laminated sandstones and siltstones as Tc, laminated siltstones as Td and
mudstones as Te. The thicker sandy beds contain incomplete successions of Ta-Tb (Tc) beds that are interpreted as having been deposited in a relative proximity to the shoreline. Thinner beds contain Tb-Td beds and are interpreted as more distal deposits. Te beds are interpreted as hemipelagic, background sedimentation, described in detail in F5, and are preserved during times of relative quiescence. The turbulent, sediment-laden gravity flows of F3 can be triggered by a variety of events including, but not limited to, slope failure, storms, sea level fluctuation and tectonic activity (Bouma, 2004; Meiburg and Kneller, 2010). Turbidites can be deposited along the shoreface and into the offshore depending on the size of the flow and the triggering mechanisms (Pattison, 2005; Pattison and Hoffman, 2008; Basilici et al., 2012). The lone Planolites burrow suggests rare times during which habitable conditions were established. Wavy upper contacts indicate wave-reeking of some of the upper surfaces of the turbidites.

2.6.4 – Facies 4 (F4) – Laminated silty mudstones (Figure 2.3d)

*Description:* F4 is primarily composed of finely-laminated silty mudstones and muddy siltstones, occasionally containing small-scale ripples. Beds of facies 4 range from 0.5-50 cm in thickness. On occasion ripples and small, low angle trough cross beds are observed in this facies and are associated with an increase in coarse silt content. The upper and lower contacts of the beds are usually sharp; in rare cases F4 grades into hemipelagic facies (F5). Bioturbation is subordinate (BI = 2) in this facies; occasional *Planolites* and *Arthrophycus* burrows were identified. When present, exoskeletal fragments (most commonly hyolithids and bivalved arthropods) are found along bedding
planes; they are typically disarticulated, angular and < 2cm in diameter. No soft-bodied fossils are associated with this facies.

**Interpretation:** This facies encompasses sedimentary characteristics that are typical for partially preserved fine-grained turbidites, which are typically deposited along the offshore transition zone and into the offshore (Bouma, 1962; Bouma 2000). The laminated silty mudstones and muddy siltstones are interpreted as Td deposits and the rippled muddy siltstones as Tc deposits (sensu Bouma 1962). The presence of low-angle cross beds and ripples requires the presence of bedload transport processes and supports this facies interpretation as turbidites rather than passive, suspended load-driven hemipelagic sedimentation. The exoskeletal fragments present in this facies were either disarticulated from decaying on the seafloor prior to transport or abraded during bedload transport. The lack of soft-bodied fossils in this facies suggests the taphonomic conditions were not conducive to tissue preservation. The few *Planolites* and *Arthrophycus* burrows suggests that conditions within the water column may have occasionally been suited to support limited organisms, but these times were uncommon.

2.6.5 – **Facies 5 (F5) – Laminated and algal mudstones (Figure 2.3e)**

**Description:** This facies is characterized by laminated silty mudstones with algal fragments and fecal strings present on some of the bedding planes (Figure 2.4a). The beds of F5 range in thickness from 1-15 cm when interlaminated with F4 and from 2 cm to > 1 m thick in all other facies associations. Lower contacts are sharp and drape over underlying facies. The upper contacts are typically sharp but are occasionally scoured into or deformed when overlain by a coarser facies (e.g., F1-4). Bioturbation is variable
Figure 2.4 – Examples of pelagic mudstones (F5) a) showing laminations with algal fragments and b) with a partial trilobite exoskeleton. Both samples are from the Xiaolantian section. Samples stored at Field Station of Early Life Research Center (NIGPAS).

(BI = 1-4); common ichnotaxa include *Planolites, Arthrophycus, Chondrites,* and *Helminthopsis* (Figure 2.2). The algae found on some of the bedding planes consist of *Sinocylindra yunnanensis, Megaspirellus houi,* and *Fuxianospira gyrata,* with the latter being the most common form within the Maotianshan Shale (Chen and Erdtmann, 1991; Chen and Zhou, 1997). The algae range from small fragments 1-4 mm in length to complete algal strings, up to ~2 cm in length. Exoskeletal fragments are occasionally found in association with both the algal-rich and algal-poor bedding planes. Unlike in F4 the exoskeletons in F5 are typically complete, most often consisting of arthropods (Figure 2.4b). No soft-bodied fossils were identified in F5 but they have been identified in sediments described as background mudstones in other studies (e.g., Hu et al., 2005).  

**Interpretation:** F5 is interpreted to represent hemipelagic, suspension settle-out deposits, deposited below fair-weather wave base (Kuehl et al., 1982). The thin laminae with abundant algal material suggest episodic low sedimentation rates with breaks in
sediment fall-out, enabling the establishment of extensive algal communities within the photic zone. The fragile algae filaments are either complete or in large fragments, suggesting *in situ* preservation or minimal transport prior to burial. The presence of these well-preserved algal fossils in this facies may be due in part to tissues more resistant to degradation. The presence of a variety of mostly articulated exoskeletons and bioturbation also suggest aerobic conditions and a position within the photic zone allowing algae to thrive. Unlike the exoskeletal fragments in F4, the exoskeletons in F5 are mostly complete indicating minimal transport prior to burial. These sediments were deposited anywhere between the lower shoreface to the offshore. The algal-rich intervals indicate relatively shallow conditions while the algal-poor intervals may reflect either deeper water, below the photic zone, or times of higher sedimentation rates or other conditions unfavorable to the establishment of algal communities.

2.6.6 – *Facies 6 (F6) – Structureless Mudstones (Figure 2.3f)*

*Description:* This facies is the finest-grained facies of the Maotianshan Shale recognized in the study area, mainly composed of clay-sized particles. The beds are typically 0.5-5 cm thick and have sharp upper and lower contacts (Figure 2.5) and do not exhibit lateral or vertical grading at outcrop scale. The beds are structureless but other researchers have observed a slight fining-upward within these beds (e.g., Hu, 2005). Bioturbation is absent in this facies (BI = 1). Most of the well-preserved soft-bodied fossils of the Chengjiang biota are associated with this facies (for faunal lists see Chen and Zhou, 1997; Hou et al., 2007). The fossils are mostly complete and are often found *in situ* with appendages in life position (Hou et al., 2005; Dombos and Chen, 2008),
Figure 2.5 - Event mudstones (F6) interbedded with pelagic mudstones (F5), Jianshan section. The event mudstones upper and lower boundaries are noted with dashed lines and highlighted with arrows.
while others show sign of minor, localized transport (Zhang et al., 2006; Zhang and Hou 2007). Some of the fossils are preserved with a 3-dimensional component (e.g., Hou et al., 2005). The well-preserved algae found in F5 are rarely found in F6.

*Interpretation:* F6 is interpreted as event mudstones associated with highly concentrated, fine-grained sediment plumes. Deposition was rapid (instantaneous) resulting in mudstones that lack any visible sedimentary structures. The detailed mechanism of sediment delivery to the basin is speculative at this point and outside the scope of this study but the absence of algae and trace fossils in F6 suggests a different depositional mechanism or depositional environment (suboxic to anoxic conditions (Gaines et al., 2012a, 2012b)), than for the other fine-grained facies (F4-F5).

Previous interpretations of facies similar to F6 have been focused on the structureless to slightly gradational nature of the beds. These studies focused on modern analogs, laboratory experiments, and microfabric observations and have proposed that the structureless mudstones in the Maotianshan Shale are the result of rapid suspension settle out associated with flocculation of clay particles (O’Brien, 1994; Zhu et al., 2001b; Hu, 2005; Steiner et al., 2005; Zhao et al., 2009; Gaines et al., 2012). Although the fossils of the Burgess Shale are encased in structurally similar clay-rich event beds (Hagadorn, 2002), which are also believed to have been associated with rapid event deposition (Gabbott et al., 2008), we do not consider the Burgess Shale as a good analog due to its different depositional environment in a setting much deeper than the Maotianshan basin and the Burgess Shale fossils show evidence of transport (Hagadorn, 2002).
Figure 2.6 – Simplified stratigraphic correlations of the 5 depositional units identified within the Maotianshan Shale in the study area. Arrows show coarsening and fining upward successions.
2.7. Basin Evolution during deposition of the Maotianshan Shale

The six lithofacies recognized in the Maotianshan Shale are the basic building blocks used to interpret the various depositional conditions that occurred during the early Cambrian in the Kunming area. They become even more meaningful when studied in the context of facies successions (facies stacking, facies associations), trace fossil occurrences, and accompanying fossil assemblages. The facies analysis and stacking patterns of the Maotianshan Shale suggests the presence of five genetically related depositional units that are bound by flooding surfaces observed across the study area (Figure 2.6).

2.7.1 – Depositional Unit 1

Unit 1 is the basal-most interval of the Maotianshan Shale overlying the black shale member of the Yu’anshan Formation and ranges from 3.5-9 meters thick across the study area (Figure 2.7). The contact with the underlying black shale member is covered or structurally deformed in all sections. The exposed upper part of this depositional unit consists of a coarsening-upward succession interpreted, based on paleoflow measurements, to reflect an overall eastward shoreline progradation (Table 2.2). In the Shankou section the coarse-grained facies primarily consist of stacked turbidites that contain the coarsest sediments (fine sand) within the study area (F3), while in the Jianshan and Xiaolantian sections coarse-grained beds containing HCS and combined flows (F1 and F2) are encased in the finer-grained facies (F4-F5). The progradational stacked turbidites in the Shankou section are interpreted as lower delta front deposits, implying that this section was the most proximal of the localities to an active sediment
Figure 2.7 – Stratigraphic column showing the correlation of depositional unit 1 in the Shankou, Jianshan and Xiaolantian sections.
Table 2.2 - Sedimentary and stratigraphic characteristics of depositional unit 1.

<table>
<thead>
<tr>
<th>Locality</th>
<th>Contact to underlying unit</th>
<th>Contact to overlying unit</th>
<th>Facies present</th>
<th>Stacking patterns</th>
<th>Bioturbation</th>
<th>Soft-bodied fossils</th>
<th>Paleoflow</th>
<th>Depositional environment</th>
<th>Sedimentary Processes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Xiaolantian</td>
<td>contact with Shiyantou Fm is covered</td>
<td></td>
<td>Facies 1-3 interbedded with facies 4 and 5</td>
<td>coarsening upward, gradual increase of sandy interbeds (facies 1-3)</td>
<td>no</td>
<td>no</td>
<td>NE</td>
<td>lower shoreface to offshore transition zone</td>
<td>abundant gravity flows and wave-reworking</td>
</tr>
<tr>
<td>Jianshan</td>
<td>contact with Shiyantou Fm is faulted out</td>
<td>sharp, regional flooding surface</td>
<td>Facies 4 and 5 interbedded with facies 2 and 3</td>
<td>coarsening upward, gradual increase of facies 2 and 3 towards top</td>
<td>no</td>
<td>no</td>
<td>n.a</td>
<td>offshore transition zone</td>
<td>offshore transition zone</td>
</tr>
<tr>
<td>Shankou</td>
<td>contact with the black shale member is covered</td>
<td></td>
<td>Facies 5 interbedded with facies 3</td>
<td>coarsening upward, gradual increase of facies 2 interbeds towards top</td>
<td>no</td>
<td>no</td>
<td>E-SE</td>
<td>prodelta to offshore transition zone</td>
<td>gravity flows and secondary wave-reworking</td>
</tr>
</tbody>
</table>
source at the time, whereas the Jianshan and Xiaolantian sections occupied a more distal location along a storm-influenced shoreface system. Paleoflow indicators suggest the sediment transport in the Shankou area was in a northeasterly direction (Figure 2.8a).

**Figure 2.8** – Paleogeographic reconstructions of the Kunming area during deposition of the Maotianshan Shale: a) progradation of the shoreline in depositional unit 1; b) retrogradation during the maximum flooding surface in depositional unit 2; c) progradation of the shoreline resuming in the upper part of depositional unit 3; d) final progradation of the shoreline in depositional unit 5. Facies belts are modified from those used by Hu (2005): I – exposed land, II – nearshore sandstone facies, III – shoreface silty mudstone facies, IV – offshore mudstone and siltstone facies. Stars denote the locations with soft-body fossil-bearing intervals. Black arrows show the direction of shoreline movement (progradation or retrogradation).

2.7.2 – Depositional Unit 2

The contact between units 1 and 2 is interpreted as a flooding surface that can be recognized across the study area and is associated with a landward shift in facies (Figure 2.9). Depositional unit 2 ranges from 8 to 15.5 meters thick across the study area.
Figure 2.9 – Stratigraphic columns showing the correlation of depositional unit 2 in the Shankou, Jianshan, Ercai and Xiaolantian sections. Legend in Figure 2.7.
Table 2.3 – Sedimentary and stratigraphic characteristics of depositional unit 2.

<table>
<thead>
<tr>
<th>Locality</th>
<th>Contact to underlying unit</th>
<th>Contact to overlying unit</th>
<th>Facies present</th>
<th>Stacking patterns</th>
<th>Bioturbation</th>
<th>Soft-bodied fossils</th>
<th>Paleoflow</th>
<th>Depositional environment</th>
<th>Sedimentary Processes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Xiaolantian</td>
<td>sharp, regional flooding surface</td>
<td>within a fault zone</td>
<td>Facies 1-3 interbedded with facies 4 and 5</td>
<td>fining upward (basal 3 m) followed by gradual coarsening upward</td>
<td>no</td>
<td>no</td>
<td>NE</td>
<td>offshore transition zone to lower shoreface</td>
<td>storm-derived deposits and abundant gravity flows</td>
</tr>
<tr>
<td>Ercai</td>
<td>n.a</td>
<td>sharp, regional flooding surface</td>
<td>Facies 4 and 5 with interbeds of facies 3</td>
<td>coarsening upward</td>
<td>Planolites</td>
<td>no</td>
<td>n.a</td>
<td>lower shoreface</td>
<td>gravity flows and times of quiescence</td>
</tr>
<tr>
<td>Jianshan</td>
<td>sharp, regional flooding surface</td>
<td>sharp, regional flooding surface</td>
<td>Facies 1-3 and 6 interbedded with facies 4 and 5</td>
<td>fining upward (basal 1m) followed by gradual coarsening upward (increase in interbeds of facies 1 and 3)</td>
<td>no</td>
<td>Haikouella</td>
<td>NE</td>
<td>offshore transition zone to lower shoreface</td>
<td>abundant gravity flows and storm-derived deposits</td>
</tr>
<tr>
<td>Shankou</td>
<td>sharp, regional flooding surface</td>
<td>sharp, regional flooding surface</td>
<td>Facies 4 and 5 interbedded with facies 3</td>
<td>coarsening upward, gradual increase of interbeds of facies 2</td>
<td>no</td>
<td>no</td>
<td>E</td>
<td>offshore transition zone to prodelta</td>
<td>gravity flows and times of quiescence</td>
</tr>
</tbody>
</table>
Following this initial flooding event, the facies stacking patterns in all sections show a continued fining-upward trend in the basal parts of unit 2, interpreted as resulting from a sustained, basin-wide transgression (Table 2.3). This basal fining upward succession is overlain by a thick, coarsening-upward facies succession, forming the majority of unit 2. Similar to unit 1, the coarse facies in the Shankou section are dominated by turbidites (F3) and by storm influenced beds (F1 and F2) in the Jianshan, Xiaolantian and Ercai sections. The abundance of storm-derived and reworked facies (F1 and F2) in the latter sections suggests continuous storm activity during the deposition of this unit. These
storms continued to only affect part of the basin, as there is little evidence for storm-derived facies in the Shankou area; there is a possibility that some of the turbidites resulted from re-suspension of nearshore deposition during storm events. The F3 beds in the Shankou are thicker and more closely spaced than in the other localities, and are interpreted as having been deposited in a more proximal position relative to a sediment source.

In the lower part of the coarsening-upward succession in the Jianshan section there is an abundance of 2-5 cm thick beds of clean mudstones (F6) that are interbedded with F5 and F4. The early vertebrate *Haikouella* (Chen et al., 1999) is found preserved in these clean mudstones, but only the heads and branchial arches are present (Figure 2.10), not the entire body with soft tissues.

2.7.3 – *Depositional Unit 3*

Depositional Unit 3 is 7.5 to 15 meters thick and consists of a thin succession of fining-upward facies overlain by a thicker, coarsening-upward facies succession (Table 2.4, Figure 2.11). The highest abundance of hemipelagic mudstones (F5) is found at the transition between the retrogradationally stacked basal interval and the progradationally stacked facies of the upper interval of unit 3. This transition zone is interpreted as the most distal depositional environment (lower offshore transition zone to shelf) of the Maotianshan Shale, and associated with the most landward shoreline position (Figure 2.8b). Differences in sediment transport directions between the Xiaolantian (paleoflow to the NE-SE) and the Jianshan (paleoflow to the NE-E) might be the result of multiple sediment point sources to the west of the study area, supplying sediments to the basin.
Figure 2.11 – Stratigraphic columns showing the correlation of depositional unit 3 within the Shankou, Jianshan, Ercai and Xiaolantian sections. Legend is in Figure 2.7.
Table 2.4 – Sedimentary and stratigraphic characteristics of depositional unit 3.

<table>
<thead>
<tr>
<th>Locality</th>
<th>Contact to underlying unit</th>
<th>Contact to overlying unit</th>
<th>Facies present</th>
<th>Stacking patterns</th>
<th>Bioturbation</th>
<th>Soft-bodied fossils</th>
<th>Paleo-flow</th>
<th>Depositional environment</th>
<th>Sedimentary Processes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Xiaolantian</td>
<td>sharp, regional flooding surface</td>
<td>sharp, regional flooding surface</td>
<td>Facies 5 interbedded with facies 3, 4 and 6</td>
<td>minor fining upward (bottom 2.5 m) followed by gradual coarsening upward</td>
<td>no</td>
<td>no</td>
<td>NE-SE</td>
<td>offshore transition zone to lower shoreface</td>
<td>suspension settle-out and abundant gravity flows and wave-modified flows</td>
</tr>
<tr>
<td>Ercai</td>
<td>sharp, regional flooding surface</td>
<td>n.a</td>
<td>Facies 5 interbedded with facies 3 and 6</td>
<td>minor fining upward in the bottom 2.5 m; small interval of coarsening upward, facies 6 interbedded with facies 5</td>
<td>no</td>
<td>no</td>
<td>S (doubtful)</td>
<td>offshore transition zone</td>
<td>suspension settle out and distal turbidites</td>
</tr>
<tr>
<td>Jianshan</td>
<td>slightly gradational flooding surface</td>
<td>slightly gradational flooding surface</td>
<td>Facies 5 interbedded with facies 2-4</td>
<td>fining upward (bottom 2.25 m); gradual coarsening-upward from facies 4 near the base to facies 2 near the top of the interval</td>
<td>no</td>
<td>no</td>
<td>NE</td>
<td>offshore transition zone to lower shoreface</td>
<td>suspension settle-out, gravity flows and wave-modified flows</td>
</tr>
<tr>
<td>Shankou</td>
<td>sharp, regional flooding surface</td>
<td>n.a</td>
<td>Facies 5 interbedded with facies 3, 4 and 6</td>
<td>coarsening upward from facies 5 to facies 2 interbedded with facies 4, 5 and 6, yielding soft-body fossils</td>
<td>Planolites, Arthropycus</td>
<td>Naraonia, nematode and priapulid worms</td>
<td>n.a</td>
<td>offshore transitions zone to delta front</td>
<td>suspension settle-out wave-reworked gravity flows and fine-grained, high concentration sediment plumes</td>
</tr>
</tbody>
</table>
Sandstones and siltstones (F1-F3) in the Shankou section are the coarsest observed in this unit, followed by the sandstones in the Xiaolantian section, the Jianshan section and lastly the Ercai. The grain size relationship, together with the overall abundance of sand beds is interpreted to reflect the inferred position of these sections to the shoreline (Figure 2.8c); the Shankou section is interpreted to be located closest to the sediment source and the Xiaolantian, Jianshan and Ercai sections in a more distal position. A relative proximal position of the Shankou section is further supported by an increase in frequency of wave-reworked coarse beds (F3 and F4), event mudstones (F6) as well as the presence of Arthrophycus burrows in the hemipelagic mudstones (F5). Many of the F6 beds contain soft-bodied fossils such as priapulid worms and the arthropod Naroia, suggesting that high-density sediment plumes repeatedly affected biological communities in this area.

2.7.4 – Depositional Unit 4

The contact between units 3 and 4 is only exposed in the Jianshan and Xiaolantian sections and is interpreted as a basin-wide flooding surface (Figure 2.12). This flooding surface is overlain by a gradual fining-upward facies succession followed by a coarsening-upward pattern that is truncated in all localities (Table 2.5); in the Jianshan and Ercai sections the upper part of unit 4 is covered and truncated by the Devonian Haikou Formation, and in the Xiaolantian the contact between units 4 and 5 is within a fault zone. The coarsening-upward succession in the Jianshan and Xiaolantian sections is characterized by abundant storm and wave-reworked beds (F1-F4) as well as event mudstones (F6). In situ soft-bodied fossils occur in this part of the sedimentary
Figure 2.12 – Stratigraphic columns showing the correlation of depositional unit 4 in the Jianshan, Ercai and Xiaolantian sections. Legend is in Figure 2.7.
Table 2.5 – Sedimentary and stratigraphic characteristics of depositional unit 4.

<table>
<thead>
<tr>
<th>Locality</th>
<th>Contact to underlying unit</th>
<th>Contact to overlying unit</th>
<th>Facies present</th>
<th>Stacking patterns</th>
</tr>
</thead>
<tbody>
<tr>
<td>Xiaolantian</td>
<td>n.a</td>
<td>Facies 5 interbedded with facies 2-4</td>
<td>Minor fining upward (basal 1 m) followed by coarsening upward</td>
<td>n.a</td>
</tr>
<tr>
<td>Ercai</td>
<td>n.a</td>
<td>Facies 5 interbedded with facies 3, 4 and 6</td>
<td>Coarsening upward of facies 2 and 4 interbedded with facies 6</td>
<td>Isoxys, Heliomedusa, nematode and priapulid worms</td>
</tr>
<tr>
<td>Jianshan</td>
<td>n.a</td>
<td>Facies 5 interbedded with facies 1-4 and 6</td>
<td>Fining upward (basal 1.5 m) followed by coarsening upward of facies 1-3, interbedded with facies 6</td>
<td>Planolites, Arthropycus, Chondrites, Helminthopsis Leptomitella, Cardiodictyon, Microdictyon, Fuxianhuia, Leanchoilia</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Bioturbation</th>
<th>Soft-bodied fossils</th>
<th>Paleoflow</th>
<th>Depositional environment</th>
<th>Sedimentary Processes</th>
</tr>
</thead>
<tbody>
<tr>
<td>n.a</td>
<td>no</td>
<td>n.a</td>
<td>offshore transition zone to lower shoreface</td>
<td>suspension settle-out grading and abundant gravity flows</td>
</tr>
<tr>
<td>n.a</td>
<td>Isoxys, Heliomedusa, nematode and priapulid worms</td>
<td>n.a</td>
<td>offshore transition zone to lower shoreface</td>
<td>suspension settle-out and fine-grained sediment plumes with minor gravity flows</td>
</tr>
<tr>
<td>n.a</td>
<td>Planolites, Arthropycus, Chondrites, Helminthopsis</td>
<td>Leptomitella, Cardiodictyon, Microdictyon, Fuxianhuia, Leanchoilia</td>
<td>offshore transition zone to lower shoreface</td>
<td>suspension settle out and fine-grained sediment plumes with gravity flows and wave-modified flows</td>
</tr>
</tbody>
</table>
succession. In the Xiaolantian area sandy interbeds are less common; when present they are mostly turbidites (F3) and show little wave-reeking. All sedimentary characteristics taken together infer the Jianshan/Ercai area was located more proximally to the inferred shoreline than the Xiaolantian. Due to the missing or truncated upper contacts of this unit, its true thickness is unknown; the exposed portions of depositional unit 4 range from 5.5 to 11.5 meters across the study area.

2.7.5 – Depositional Unit 5

Unit 5 is the uppermost unit of the Maotianshan Shale observed in this study and is only present in the Xiaolantian and Maotianshan localities. Unit 5 is also the thickest of all the units observed in the study area, with an overall thickness of over 21 meters exposed at the Xiaolantian locality (Figure 2.13). The lower contact of the unit is not exposed in the study area, but based on its similarity to the other units, the contact between units 4 and 5 is likely another flooding surface. Unit 5 consists of a thin fining-upward succession overlain by a thick package of coarsening-upward stacked facies (Table 2.6). In the coarsening upward succession the sandy facies (mostly F3 with some F1 and F2) become more frequent and more closely stacked up-section. All four types of ichnotaxa identified in the Maotianshan Shale are present in this interval and are all associated with F5. Planolites and Arthrophycus burrows are infilled with coarser sediments and are often found in association with a thin (1-2 cm thick) bed of F3 or F4. F5 also contains many exoskeletal fragments, but no well-preserved soft tissues. The lower to middle portions of the retrogradational interval consist of interbedded packages of F3, F4, F5, and F6, the latter of which yields abundant soft-bodied fossils. In the
Figure 2.13 – Stratigraphic columns showing the correlation of depositional unit 5 in the Xiaolantian and Maotianshan sections. Legend is in Figure 2.7.
### Table 2.6 – Sedimentary and stratigraphic characteristics of depositional unit 5.

<table>
<thead>
<tr>
<th>Locality</th>
<th>Contact to underlying unit</th>
<th>Contact to overlying unit</th>
<th>Facies present</th>
<th>Stacking patterns</th>
<th>Bioturbation</th>
<th>Soft-bodied fossils</th>
<th>Paleoflow</th>
<th>Depositional environment</th>
<th>Sedimentary Processes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maotianshan</td>
<td>n.a</td>
<td>n.a</td>
<td>Facies 5 interbedded with facies 2-4 and 6</td>
<td>coarsening upward facies 2-3 interbedded with facies 6</td>
<td>no</td>
<td>Naroia, Eldonia, Isoxys</td>
<td>NE</td>
<td>offshore transition zone to lower shoreface</td>
<td>suspension settle-out and fine-grained sediment plumes with abundant wave-modified gravity flows and combined flows</td>
</tr>
<tr>
<td>Xiaolantian</td>
<td>n.a</td>
<td>n.a</td>
<td>Facies 5 interbedded with facies 1-4 and 6</td>
<td>fining upward (basal 3.5 m) followed by coarsening upward with an increase in frequency of facies 2 up-section; middle to upper interval interbedded with facies 6</td>
<td>Planolites, Arthropycus, Chondrites, Helminthopsis</td>
<td>Naroia, Onychodictyon, Cindarella, nematode and priapulid worms</td>
<td>E-NE</td>
<td>offshore transition zone to lower shoreface</td>
<td>suspension settle out and abundant sediment plumes interbedded with abundant wave-modified gravity flows</td>
</tr>
</tbody>
</table>
Maotianshan locality (type section) there is limited exposure and only 5 m of section were available for study but was included due to its importance in the original identification of the Chengjiang biota. The Maotianshan section consists of an abundance of coarse-grained facies, dominated by F3, which often have wave-reeformed upper surfaces. Event mudstones (F6) containing soft-bodied fossils are also frequently found in this interval interbedded with hemipelagic mudstones (F5). The abundance of wave-reeformed coarse-grained beds and the abundance of autochthonous, soft-body fossils and bioturbation place these localities above storm wave base. Paleoflow measurements indicate the sediment transport direction was generally SW-NE (Figure 2.8d).

2.8 – Overall sedimentological trends and implications for fossil occurrences and taphonomy

The soft-bodied fossils of the Chengjiang biota are generally interpreted as being preserved in situ or having undergone localized transport prior to burial, indicating they were buried in or close to the environment in which they were living (Hou et al., 2005; Zhang and Hou, 2007; Hou et al., 2009). This model for fossil deposition is supported by the occurrence of well-defined fossil assemblages in different areas within the study area (Zhao et al., 2012). In the study area, these well-preserved fossils are always found in event-mudstones (F6) interbedded with hemipelagic sediments (F5) and storm-derived deposits and turbidites (F1-F3). The interbedded nature of those deposits indicates that hemipelagic sedimentation in the study area was interrupted by episodic storm events and turbidity currents in the lower offshore transition zone and on the shelf along a wave-dominated delta or shoreline (Figure 2.14).
Figure 2.14 – Schematic shoreface reconstruction based on Reading and Collinson (1996); horizontal and vertical axes are not drawn to scale. The depositional ranges of the six identified facies are noted. MLW – mean low water; FWWB – fair weather wave base; SWB – storm wave base.
Although data presented in this study do not allow for the interpretation of a distinct provenance, paleoflow data indicates multiple directions of sediment transport, possibly indicating that the sediments were delivered to the basin by multiple sources located to the west of the study area (Figures 2.8a-d), likely from the Cambrian Central Yunnan Hills, landmass (e.g., Geological Mineral Bureau of Yunnan Province, 1995; Zhu et al., 2001a; Hu 2005).

Ichnofaunal communities identified in the Maotianshan Shale also indicate a depositional environment on the upper shelf and offshore transition zone. In modern nearshore marine environments, biotic communities associated with soft sediments are typically found on the offshore transition zone where they are less affected by waves and sediment re-working but are still within the photic zone (Pemberton et al., 1992; Clifton, 2006; Buatois and Mangano, 2012; Byers and Grabowski, 2014), suggesting the offshore transition zone as a plausible location for where the Chengjiang biota was thriving. The presence of abundant algal material in the background mudstones (F5) also constrains the depositional environment to within the euphotic zone. This depositional environment is slightly shallower than inferred in previous interpretations that place the event beds (F6) containing the Chengjiang biota offshore (e.g., Zhu et al., 2001b; Hu, 2005).

In all measured sections the soft-bodied fossils and ichnofossils are associated with the coarsening-upward interval of the individual depositional units (Figure 2.15). This suggests that during episodic shoreline progradation, the conditions for diverse communities to develop became successively more favorable in the Maotianshan Basin and at the same time increased the preservation potential of the organisms. With the exception of the Haikouella-rich beds in Unit 2 of the Jianshan section, the exceptionally
Figure 2.15 - Simplified stratigraphic column of the Maotianshan Shale showing a transgressive-regressive cycle within a depositional unit. The soft-bodied fossils of the Chengjiang biota most frequently occur in the lower to middle part of the regressive phase. The most diverse occurrence of ichnofauna also occur sin this interval.
preserved fossils are found in the upper parts of the Maotianshan Shale, equivalent to Units 3-5 in this study.

The fossil-rich beds found in Unit 2 of the Jianshan section are dominated by the early vertebrate *Haikouella*, often found in death assemblages in which only parts of the heads and branchial arches are preserved (Figure 2.10). The incompleteness of the fossils, the lack of biodiversity in these event mudstones, and the frequent death assemblages in this stratigraphic interval suggests that these pelagic organisms might have been displaced from their original habitat more proximal to the shoreline and carried more distally within the depositional basin. As an alternative it needs to be considered that the incompleteness of the fossils may also be attributed to the taphonomic factors, or lack thereof in that particular depositional unit. Soft-tissue preservation in the fossil record is rare, and there are multiple taphonomic factors required for this to occur (Brett et al., 1997; Briggs, 2003); it is possible the taphonomic conditions were not optimal for exceptional preservation during deposition of this stratigraphic interval.

The incomplete preservation of the *Haikouella* fossils in the Jianshan section starkly contrasts with that of the fossils found higher up-section in the Jianshan and in the other localities included in this study, where entire organisms are preserved (Chen et al., 1999; Zhu et al., 2001c). Although the well-preserved fossils always occur in the upper parts of the Maotianshan Shale, the data presented in this study suggests that the fossil beds are not time correlative amongst the different localities. In the Shankou section, the exceptionally well-preserved soft-bodied fossils first occur in depositional unit 3. In the Jianshan and Ercai sections, these fossils first occur in unit 4, and in the Xiaolantian and Maotianshan sections, they do not occur below depositional unit 5. This time-
transgressive nature for the developments of the fossil communities, and/or taphonomic conditions favorable for their preservation, seems to correlate with the eastward progradation of the Early Cambrian shoreline. The earliest fossils appear in the Shankou section, interpreted as the most proximal location to the shoreline at the time of deposition of unit 3. Through time, fossils appear in sections successively located farther to the east (Figure 2.16). This allows for the interpretation that the depositional environments associated with the time-transgressive progradation of the shoreline from southwest to northeast also created conditions suitable for the development of diverse biotic communities, coinciding with taphonomic conditions appropriate for the preservation of the Chengjiang biota.

This time-transgressive shoreline progradation, from the Shankou locality to the Jianshan and Ercai localities and finally to the Xiaolantian and Maotianshan localities, also corresponds to the geographical distribution of distinct fossil assemblages. Fossil assemblages in the Anning (Shankou), Haikou (Jianshan and Ercai), and Chengjiang (Xiaolantian and Maotianshan) areas are characterized by well-defined communities although some species overlap is present between these localities (Zhao et al., 2012). Based on the data presented here, the time-transgressive nature of the eastward-prograding shoreline may have contributed to the development of these different biotic assemblages in this overall “taphonomic goldilocks zone”. Taphonomic conditions required for the fossilization of the Chengjiang biota were not present outside of this facies belt, paralleling the northwest-southeast trending shoreline. Areas further to the east would have been too far offshore to be reached by the highly-concentrated, fine-grained sediment plumes (F6); areas to the west would have been too shallow on a
Figure 2.16 – A schematic showing the time-transgressive depositional model for the occurrence of well-preserved fossils of the Chengjiang biota. At time 1 the Chengjiang biota is preserved in the Shankou area during depositional unit 3. At time 2 the high fidelity fossils are preserved in the Haikou area (Jianshan and Ercai sections) during deposition of unit 4. Finally, at time 3 the soft-bodied fossils are preserved in the Maotianshan section (Xiaolantian and Maotianshan localities) during deposition of unit 5. Histograms show the diversity of each area based on phyla from Zhao et al (2012), n equals the total number of phyla identified (Ar – Arthropoda, Pr – Priapulida, Po – Porifera, Br – Brachiopoda, Lo – Lobopodia, Hy – Hyolitha, Vt – Vetulicolia, Cn – Cnidaria, Ct – Ctenophora, Cd – Chordata, Ch – Chancelloriida, Si – Sipuncula, Ec – Echinodermata, An – Annelida, Ph – Phoronida, Mo – Molluska, Cg – Chaetognatha, Un – Unknown).
shoreface where high-energy conditions would have prevented the preservation of fragile organisms.

Although there is no definitive evidence for any particular mechanism, the following environmental (sedimentological) conditions associated with shoreline progradation might have contributed to the fossilization of the soft tissues: (1) gradual increased proximity to sediment sources resulting in an increased flux of nutrients to the basin, creating a habitable environment; (2) increased sediment flux of fine-grained plumes increasing the potential for preservation; (3) a flux of oxygenated waters from a freshwater source creating aerobic conditions in the basin favorable to the development of a diverse marine community; (4) suboxic to anoxic conditions in the event mudstones (F6) shortly after deposition preventing bioturbation and aiding in fossilization; (5) shoreline progradation over a habitable substrate, creating a livable environment within the photic zone where productive, diverse communities are able to thrive; (6) times of diminished storm activity allowing diverse communities to develop.

2.9 – Conclusions

Based on the sedimentological and stratigraphic data presented herein, the following conclusions can be drawn:

- Three coarse-grained facies are present in the Maotianshan Shale (F1-F3): hummocky cross-stratified (HCS) beds, turbidites, and combined flow deposits.
- Three fine-grained facies were identified in the Maotianshan Shale (F4-F6): distal turbidites, pelagic sediments, and structureless mudstones containing the Chengjiang Biota.
• Five distinct depositional units were identified within the study area; they are bound by flooding surfaces observed throughout the study area and contain a minor fining-upward facies succession overlain by a coarsening upward facies succession interpreted as a minor marine transgression followed by shoreline progradation, respectively.

• At each of the measured sections, the exceptionally preserved fossils of the Chengjiang biota are always found in the coarsening-upward, prograding packages.

• The soft-body fossil-bearing event beds are interpreted as having been deposited on the offshore transition zone, which is shallower than previous interpretations.

• Taphonomic conditions allowing for the preservation of the Chengjiang Biota shifted eastward though time. Fossil beds in Shankou section are stratigraphically older than those in the more eastward Xiaolantian and Maotianshan sections.

The time-transgressive model presented herein will guide addition sedimentological and stratigraphic studies of other localities known to yield Chengjiang fossils as well as other BST deposits.

2.10 – Acknowledgements

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2.11 – References


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CHAPTER 3 – THE MINERALOGY AND TRACE ELEMENT GEOCHEMISTRY OF THE MAOTIANSHAN SHALE, YUNNAN, CHINA

3.1 – Introduction

Provenance, redox conditions during deposition and diagenetic history of sedimentary rocks can be obtained from mineralogical and geochemical analyses (e.g., Fairbridge, 1970; Bhatia and Crook, 1986; Condie, 1991; Rimmer, 2004). All of these factors contribute to the sequence of events and processes that promote or prevent fossilization (Efremov, 1940; Seilacher et al., 1985; Briggs, 2003). Exceptionally well-preserved fossils are rare, and there are a multitude of factors responsible for their preservation (Brett et al., 1997; Bottjer et al., 2002, and references therein). In this study we analyzed the mineralogy and geochemistry of the early Cambrian Maotianshan Shale to determine (i) the source of the sediments and any diagenetic changes and (ii) the impacts these may have had in the fossilization of the Chengjiang Biota.

3.1.1 – Maotianshan Shale and the Chengjiang Biota

The Early Cambrian (~520 Ma; Adtabanian) Maotianshan Shale is the middle member of the Yu’anshan Formation, exposed in outcrops surrounding Kunming, Yunnan, (Figure 3.1) on the western edge of the South China Block of the Yangtze craton (Lin and Zhang, 1985; Wang et al., 2010). The sediments consist of a succession of interbedded mudstones, siltstones and sandstones interpreted as having been deposited along a wave-dominated deltaic/shoreface environment (Babcock and Zhang, 2001; Hu,
Figure 3.1 – Locality map and stratigraphic column of the study area. All but one of the samples were collected from the Jianshan locality. The core sample was taken from a mine near the type section at Maotianshan. The samples from the Jianshan section and the core are grey in color and considered fresh. When the sediments are weathered they become tan-yellow in color. Both outcrop images are in color. Figure modified from MacKenzie et al. (2015).

2005; MacKenzie et al., 2015). The sedimentology discussed in this paper is taken from MacKenzie et al (2015) and is summarized in Table 3.1.

The Maotianshan Shale is most famously known for containing the exceptionally well-preserved fossils of the Chengjiang Biota, the earliest known deposits of the “Cambrian Explosion”. The Chengjiang Biota is categorized as a Burgess Shale-type
Table 3.1 – Description of the sedimentology and depositional environments of the facies sampled for this study (after MacKenzie et al., 2015).

<table>
<thead>
<tr>
<th>Facies</th>
<th>Description of sediments</th>
<th>Interpreted depositional environment</th>
</tr>
</thead>
<tbody>
<tr>
<td>combined flow</td>
<td>sandstones/siltstones; normally graded with evidence of wave-reworking</td>
<td>combined unidirectional and oscillatory flows; possibly co-occurring gravity flows and storm-generated flows</td>
</tr>
<tr>
<td>turbidite</td>
<td>sandstones/siltstones showing a Bouma-type grading (massive base, grading into laminations and cross beds to ripples)</td>
<td>turbulent, sediment-laden gravity flows triggered by storms, changes in sea level, slope failure, etc.</td>
</tr>
<tr>
<td>distal turbidite</td>
<td>finely laminated muddy siltstones and silty mudstones</td>
<td>fine-grained turbidites; often the tail end of the turbidite</td>
</tr>
<tr>
<td>pelagic mudstone</td>
<td>laminated mudstones with minor silt; exoskeletal fragments and algal material occasionally present</td>
<td>hemipelagic/background sedimentation</td>
</tr>
<tr>
<td>event mudstone</td>
<td>laminated to structureless, clean mudstones; contain the well-preserved fossils of the Chengjiang biota</td>
<td>event mudstones deposited by highly concentrated, fine-grained sediment plumes</td>
</tr>
</tbody>
</table>

(BST) deposit based on its similarity to the Burgess Shale and other early Paleozoic fossiliferous localities (Hagadorn, 2002a,b,c). The Chengjiang Biota fossils preserve a diverse community that depicts the evolution and diversification of metazoans (Chen and Lindström, 1991; Chen, 2009). The exoskeletons organisms are calcified or phosphatized, and the soft tissues are preserved as thin carbon films (Hou et al., 2007; Gaines et al., 2008). Exoskeletal fragments of the Chengjiang fossils are found throughout the Maotianshan shale, but the exceptionally preserved fossils are found in clean mudstones interpreted as rapidly deposited event beds (MacKenzie et al., 2015).
The Maotianshan Shale has not undergone significant alteration at high temperatures like some other BST Lagerstätten (i.e., greenschist metamorphism of the Burgess Shale; Powell, 2003) but it has undergone weathering and minor diagenesis, as observed by Forchielli et al. (2014), across the depositional basin. The degree of weathering and/or diagenesis at the various fossil localities is unknown and further data are needed. Newly excavated samples and those taken from cores are grey to black in color (Bergström, 2001; Zhu et al., 2001) and contain minerals easily removed during weathering (oxidation of pyrite and dissolution of dolomite) while samples from exposed outcrops are yellow, indicating extensive surface weathering (“…(the Chengjiang Biota is) influenced by late alteration and weathering that led to some major transformations in clay and iron minerals, as well as dissolution of dolomite and skeletal biominerals”, Forchielli et al., 2014; see also: Chen et al., 1991; Bergström, 2001; Zhu et al, 2005). Until recently continuous sections of freshly exposed rocks of the Maotianshan Shale have been unavailable so little trace element geochemistry has been performed on these sediments.

3.1.2 – Mineralogy

Primary mineral assemblages, insofar as they are primary, limit possible source environments, or provenance (e.g., Blatt, 1985), and also provide information about diagenesis and authigenic mineralization that occurred within the sediments (e.g., Berner, 1970; Lanson et al., 2002). Many minerals can be formed via weathering of parent rocks as well as authigenically or via diagenetic processes. Quartz is one such mineral; it can be weathered from other rocks (Blatt, 1985), but can also be formed authigenically
Hendry and Trewin, 1995; Lynch et al., 1997) or diagenetically (Schieber et al., 2000; Vorhies and Gaines, 2009). Because of this its presence alone does not reveal information about the history of the sediments. Detrital minerals can be identified through hand sample and petrographic analysis, and their distribution can be used to determine the provenance of the sediments (Blatt, 1985). Secondary minerals such as pyrite, depending on the form present, will form during early diagenesis and provide information about the conditions within the sediments prior to lithification (Berner, 1970). Clay minerals illite and chlorite can form via primary or secondary processes; if formed secondarily, they can provide information about the burial and thermal history the sediments underwent (Lanson et al., 2002).

3.1.3 – Sediment Source and Provenance Plots

Rare earth elements (REEs) are incorporated into igneous rocks. REE concentrations and distributions are dependent on the type of rocks being formed; light rare earth elements (LREE; La-Sm) are incorporated more frequently in felsic rocks while heavy rare earth elements (HREE; Gd-Lu) are incorporated into rocks of mafic composition. REEs are not easily fractionated during physical or chemical weathering and considered immobile, retaining the original composition of the source rocks (Taylor and McLennan, 1988; McLennan, 1989). REE plots normalized to a known standard (e.g. the world shale average or the C1 chondrites) are used to determine the original source of the sediments (McLennan, 1989; Condie, 1991). The C1 chondrites are calcium-rich meteorites considered to have a mafic composition similar to the earliest rocks in the solar system and the composition of Earth’s primitive mantle (Haskin et al.,
1964; Morgan et al., 1978; McDonough and Sun, 1995; Barrat et al., 2012). The WSA is a standard used to represent average shale values for the Phanerozoic and is considered to represent sediments derived from continental weathering and deposited under normal marine conditions (Turekian and Wedepohl, 1961; Wedepohl, 1971).

Many proxies have been developed to determine the provenance and/or tectonic regime from which sediments were derived (e.g., Bhatia and Crook, 1986; Floyd and Leveridge, 1987; McLennan et al., 1990; Plank and Langmuir, 1998). The provenance plots used in this study were chosen because they have been adapted for both sandstones and fine-grained sedimentary rocks and contain elements unaffected by weathering.

3.1.4 – TC and TOC

Total carbon (TC) includes both organic carbon (TOC) and inorganic carbon (TIC). TIC (carbonate) originates from the remains of shelly organisms, by direct precipitation, or during diagenesis and may or may not be associated with high organic carbon concentrations (Burdige, 2006).

Organic carbon comes from terrestrial and marine sources. Terrestrial sources of organic matter predominantly consist of plant litter. The marine fraction is composed of bacterial biomass (bacterially derived organic matter) and phytoplankton debris (Burdige, 2007). Depositional environments with high production of organic matter include areas of upwelling, near coastal and equatorial zones (Stein, 1991).

In the Cambrian, life was limited to the oceans thus all TOC was derived from the marine fraction. The lack of vascular plants is cited as a reason that early Paleozoic sediments generally have lower TOC values than younger sediment (Raiswell and
Berner, 1986); phytoplankton-derived organic carbon is easily consumed, effectively removing it from the system prior to deposition in the sediments. Organic carbon is removed from the water column and sediments via remineralization, infaunal uptake, and only sediments that are in areas of high productivity and are not overprinted by high sedimentation rates deposited under suboxic to anoxic conditions will result in organic-rich sediments with high TOC values (Arthur and Sageman, 1994; Canfield, 1994; McKee, 2003a,b).

3.1.5 – Paleoredox indicators

Many minor and trace elements behave differently depending on the redox conditions within the sediments and their presence or absence can be used to infer paleoredox conditions. U/Th, Ni/Co, V/Cr ratios, and Mo concentrations are some of the more reliable paleoredox indicators considered in this study. The U/Th ratio is considered a good redox indicator because U is redox sensitive while Th is not redox-sensitive and is considered immobile once deposited (Adams and Weaver, 1958; Klinkhammer and Palmer, 1991; Arthur and Sageman, 1994; Morford and Emerson, 1999), allowing the changes in uranium concentrations to be normalized to Th. Under reducing conditions U is reduced from U(VI) to U(IV), which is insoluble and precipitates, increasing the overall U/Th ratio; ratios less than 0.75 represent oxic conditions, between 0.75 and 1.25 suboxic conditions, and greater than 1.25 anoxic conditions.

The Ni/Co ratio is another redox indicator. Co is not redox sensitive while Ni is reduced from Ni(III) to Ni(II) under suboxic to anoxic conditions, respectively. In
addition, Ni(II) is preferentially incorporated into sulfide minerals, leading to higher Ni/Co ratios under sulfidic conditions. Ni/Co values below 5 are considered to represent oxic conditions, values between 5-7 represent suboxic conditions and those greater than 7 representing anoxic conditions (Dypvik, 1984; Jones and Manning, 1994).

V/Cr is another widely used paleoredox indicator (Ernst, 1970; Jones and Manning, 1994; Rimmer, 2004; Yang et al., 2004). Cr(VI) is soluble under oxic condition and will be adsorbed onto Mn- or Fe-oxides and clay minerals. Under sulfidic conditions reduced Cr is found in sulfide minerals. So under anoxic/sulfidic conditions dissolved Cr concentrations would be low because Cr, if present, would be immobilized. Under oxic conditions V is present as V(V). Under suboxic conditions it is reduced to V(IV), which adsorbs strongly to Fe-oxides. Under anoxic conditions V will be reduced to V(II), which is incorporated into sulfide minerals. V/Cr values less than 2 indicate oxic conditions, between 2-4 suboxic conditions, and values greater than 4 anoxic conditions (Morford and Emerson, 1999; Piper and Isaacs, 2001).

Molybdenum is present as soluble Mo(VI) under oxic conditions; this form can be slightly adsorbed onto Fe-oxides. Under anoxic and sulfidic conditions the reduced form Mo(IV), dominates. Mo(IV) is insoluble and will precipitate as MoS₂ or co-precipitate with iron sulfides (Bertine, 1972; Morford and Emerson, 1999). In this study we measured for the presence of molybdenum in the sediments, but do not know in which form it is present. Mo concentrations above 2 ppm are considered to be indicators of anoxic conditions (Dean et al., 1997; Rimmer, 2004).
3.2 – Methods

3.2.1 – Sample collection

Samples were collected in 2013. Standard field techniques were used for sample collection at the Jianshan locality, in the Kunyang Phosphate Mine, Jinning County, Yunnan (Figure 3.1). The basal portions of the exposed outcrops were newly exposed and were grey in color, which the samples collected form the upper portions of the section were tan to yellow in color.

3.2.2 – Mineralogy and Major Elements

Bulk sample X-Ray diffraction (XRD) was used to determine the mineralogical composition of the sediments. Diffraction patterns were collected on an X’Pert PRO XRD with X’Pert Data Collector software. Triplicate diffractograms were collected between 2 and 70 degrees two-Θ at a scan speed of 0.10445 °/sec and a step size of 0.0167°. The resulting diffractograms were summed to increase signal to noise ratio. Diffractograms were analyzed using X’Pert HighScore Plus.

Scanning electron microscopy with energy dispersive X-ray spectroscopy (SEM-EDS) was used to determine the major element chemistry of the sediments to correlate spatial and chemical data. Images and chemical maps were collected using a Tescan Vega 3 SEM and Oxford Instruments X-Max EDS.
3.2.3 – Geochemical Analyses

Elemental Analyses

Elemental concentrations were determined using combined inductively coupled plasma atomic emission spectroscopy (ICP-AES) and inductively coupled plasma mass spectroscopy (ICP-MS). Samples were extracted using sodium peroxide fusion and analyzed at SGS Laboratories, Canada.

Carbon

Total carbon (TC) and total organic carbon (TOC) concentrations were obtained from a CE Instruments EA 1110 Elemental Analyzer. 15-20 mg of ground and dried sample was used for each analysis; samples were loaded into tin boats and analyzed for TC and for TOC, samples were loaded into silver boats, acidified with 100 µl of 50% HCl, dried at 80°C and analyzed at the Environmental Biogeochemistry Laboratory at the University of Montana.

3.3 – Results

3.3.1 – REE concentrations and Provenance Plots

The Maotianshan Shale samples are enriched in light rare earth elements (LREEs) and generally depleted in middle rare earth (MREEs) and heavy rare earth elements (HREEs) (Table 3.2). There are no significant differences in REE concentrations between the different sedimentary facies identified in the Maotianshan Shale. C1 chondrite normalized (Morgan et al., 1978) plots of the samples again show a similar distribution


<table>
<thead>
<tr>
<th>ANALYTE</th>
<th>Type</th>
<th>Locality</th>
<th>Th</th>
<th>Sc</th>
<th>Hf</th>
<th>La</th>
<th>Ce</th>
<th>Pr</th>
<th>Nd</th>
<th>Sm</th>
<th>Eu</th>
<th>Gd</th>
<th>Tb</th>
<th>Dy</th>
<th>Ho</th>
<th>Er</th>
<th>Tm</th>
<th>Yb</th>
<th>Lu</th>
</tr>
</thead>
<tbody>
<tr>
<td>LM-28/07/09-01</td>
<td>pelagic mud</td>
<td>core</td>
<td>16.3</td>
<td>16.0</td>
<td>5.0</td>
<td>40.9</td>
<td>83.2</td>
<td>9.8</td>
<td>36.3</td>
<td>6.5</td>
<td>1.2</td>
<td>5.3</td>
<td>0.9</td>
<td>5.3</td>
<td>1.0</td>
<td>3.0</td>
<td>0.5</td>
<td>3.2</td>
<td>0.5</td>
</tr>
<tr>
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<tr>
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</table>
Figure 3.2 – Rare earth element plot of the Maotianshan Shale samples normalized to the C1 Chondrites (range of sample values shaded in grey). Standards used for comparison include the world shale average (WSA), North American shale composite (NASC), upper continental crust (UCC), Yangtze craton (YC) and the post-Archaean average Australian shale (PAAS).

3.3.2 – Mineralogy and Major elements

Comparison of the grey versus yellow samples identified minerals (i.e., pyrite, dolomite and chlorite) in the grey samples that were not present in the yellow samples.
Figure 3.3 – Samples from the Maotianshan Shale normalized to the various standards used to compare the samples. WSA - world shale average. NASC - North American shale composite. PAAS - post-Archean average Australian shale. UCC - upper continental crust. YC - Yangtze craton.
Figure 3.4 – X-ray diffractograms showing the mineralogical differences between fresh (red; JS-4/4/2013-2-14) and weathered (black; JS-9/4/2013-4C-2) sediments. Both samples are laminated, hemipelagic mudstones from the Jianshan section. Mineral key: C - chlorite, D - dolomite, I - illite, M - muscovite, O - orthoclase, P - pyrite, Q - quartz. The arrows indicate minerals lost during weathering.

(Figure 3.4). These minerals were likely lost during weathering. When the rocks are exposed to air and freshwater the pyrite (FeS₂) is oxidized creating sulfuric acid (H₂SO₄), which dissolves dolomite (CaMg(CO₃)₂) (Ritsema and Groenenberg, 1993; Eberts and George, 2000; Torres et al., 2014).

\[
\text{FeS}_2 + 15/4 \text{O}_2 + 7/2 \text{H}_2\text{O} \rightarrow 2\text{H}_2\text{SO}_4 + \text{Fe(OH)}_3
\]

\[
\text{CaMg(CO}_3\text{)}_2 + \text{H}_2\text{SO}_4 \rightarrow \text{Ca}^{2+} + \text{Mg}^{2+} + \text{SO}_4^{2-} + 2\text{HCO}_3^{-}
\]
The oxidation of pyrite will also cause chlorite to alter to smectite, which will in turn alter to illite given enough time (Gilkes and Little, 1972; Chigira and Oyama, 1999). Additional analyses (petrographic, etc.) are needed to confirm this mechanism of removal. Due to this loss only the grey samples will be discussed in this paper. Differences between the fresh and weathered samples are addressed elsewhere (MacKenzie et al., in prep).

Table 3.3 – Mineralogy of the Maotianshan Shale sediments determined by X-ray diffraction (n.d - not detected). Stars (*) denote algal-rich pelagic mudstones.

<table>
<thead>
<tr>
<th>Sample Number</th>
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<th>Ill</th>
<th>Chl</th>
<th>Mus</th>
<th>Rut</th>
<th>Orth</th>
<th>Dol</th>
<th>Pyr</th>
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<td>X</td>
<td>X</td>
<td>X</td>
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<td>JS</td>
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Figure 3.5 – Secondary electron SEM-EDS image and elemental maps of the core sample (28/07/09-01), a pelagic mudstone from the Maotian Mine. The majority of the scan is composed of clay minerals, with a larger quartz grain in the middle right of the image. The EDS scan of this quartz grain is only partially mapped because the relief of the left side of the grain likely shadowed the rest of it. Mineral key: Al - aluminum, Si - silica, K - potassium, Mg - magnesium, Fe - iron, S - sulfur, Ti, titanium, Ca - calcium. Scale bar equals 10 microns.
Mineralogical analyses of the Maotianshan Shale showed all of the samples contained quartz (SiO$_2$), muscovite (KAL$_2$(ALSi$_3$O$_{10}$)(F,OH)$_2$), rutile (TiO$_2$), dolomite (CaMg(CO$_3$)$_2$), illite ((K,H$_3$O)(Al,MgFe)$_2$(Si,Al)$_4$O$_{10}$[(OH)$_2$(H$_2$O)]), and chlorite ((Mg$_5$Al)(AlSi$_3$)O$_{10}$(OH)$_8$). Orthoclase (KAlSi$_3$O$_8$) was present in all but one of the samples. At least half of the samples contained pyrite (FeS$_2$) (Table 3.3). SEM-EDX of event mudstones detected aluminum, silica, potassium, magnesium, iron, sulfur, titanium and calcium (Figure 3.5), which are consistent with the identified mineral assemblage.

3.3.3 – TC and TOC

Total carbon (TC) concentrations in the Maotianshan Shale ranged from 0.79% to 4.48%, the majority of which is total inorganic carbon (TIC), 79.9% to 97%. Total organic carbon (TOC) values varied between 0.13% and 0.21%, within the reported instrumental error (± 10%) (Table 3.4).

**Table 3.4** – Total carbon (TC), total organic carbon (TOC) and total inorganic carbon (TIC) concentrations (as wt %) of select samples. Algal-rich pelagic mudstones noted with a star (*). Calculated (not measured) values are noted by (**).

<table>
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<th>Sediment type</th>
<th>TC (%)</th>
<th>TOC (%)</th>
<th>TIC (%)**</th>
<th>%TIC (of TC)**</th>
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3.3.4 – Paleoredox

Paleoredox indicators indicate redox conditions within the Maotianshan Shale sediments ranging from oxic to anoxic (Table 3.5). U/Th ratios range from 0.23-0.39, indicating oxic conditions. Ni/Co ratios range between 1.97-4.09, also suggesting oxic conditions in the sediments. V/Cr ratios vary between 1.51-2.58, indicating paleoredox conditions between oxic to suboxic conditions. Molybdenum concentrations in the Maotianshan Shale sediments were greater than 2 ppm in all but one of the samples.

Table 3.5 – Paleoredox proxies used to determine the redox conditions within the Maotianshan Shale sediments at time of deposition. Sediment type key: PM – pelagic mudstone, CF – combined flow, EM – event mudstone; Algal-rich pelagic mudstones are noted with a star (*). bdl - below detection limit.

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3.4 – Discussion

3.4.1 – Source of the sediments

Previous studies have identified the Maotianshan Shale having been derived from terrigenous sources (e.g., Babcock and Zhang, 2001). Volcanic sediments have been suggested, and ash beds have been reported within the Maotianshan Shale (Hou et al., 1991; Zhang et al., 2004). Our data show the sediments from the Maotianshan Shale most likely originated from a continental, felsic source and deposited in a continental island-arc setting.

The C1 chondrite-normalized REE plots of Maotianshan Shale plots have a similar shape to several standards used to identify terrigenous sediments (Figure 3.2). When plotted against each standard the ratios of the Maotianshan samples are close to one (Figure 3.3). Of the shale standards, the world shale average (WSA) has a trend similar to the upper range of the concentrations of the Maotianshan Shale. The North American Shale composite (NASC) trends closer to the lower range of values from the Maotianshan Shale. The post-Archean average Australian shale (PAAS) plots similarly to the NASC, with slight depletions in MREEs and HREEs. The two crustal standards, the Yangtze Craton (YC) and the upper continental crust (UCC) follow similar trends of the other standards with the LREEs but are depleted in MREEs and HREEs compared to the Maotianshan Shale samples. The depletions in some of the standards compared to the Maotianshan Shale samples are likely due to differences in the provenance of the sediments used in the different standards and varying degrees of weathering of the standards. Zircons, minor minerals, and clay minerals can take up a significant amount
Figure 3.6 – Diagrams identifying the provenance and tectonic setting of the Maotianshan Shale. (a) La/Th versus Hf indicates the sediments came from a felsic, acidic island arc setting. (b) La/Sm versus Yb/Sm indicates the Maotianshan Shale is generally enriched in LREEs with some signs of scavenging by hydrous oxides and/or apatite. (c) La-Th-Sc ternary diagram indicates the sediments were derived from a continental island arc setting. Key to standards: WSA - world shale average, NASC - North American shale composite, PAAS - post-Archean average Australian Shale, YC - Yangtze craton, UCC - upper continental crust.
of REEs (Gromet et al., 1984); extensive sorting of the shale standards compared to the
crustal standards explains the enrichment in MREEs and HREEs in the shale standards
and the Maotianshan Shale samples.

Plotting La/Th to Hf identified an acidic arc source for the sediments (Figure
3.6a), which is typically composed of felsic volcanic and plutonic rocks (Floyd and
Leveridge, 1987; Ochoae et al., 2007; Wang et al., 2014). A general enrichment in
LREEs (La/Sm) and depletion in HREEs (Yb/Sm) indicates a felsic source area (Gu et
al., 2002; Ochoa et al., 2007; Figure 3.6b). The La-Th-Sc ternary plot indicates a
continental island-arc tectonic setting; these sediments and are primarily composed of
felsic volcanics (Bhatia, 1983; Bhatia and Crook, 1986; Figure 3.6c).

Quartz, orthoclase, muscovite and titanium oxide are the major detrital minerals
(determined from petrographic analysis by Hofmann et al., (in press)) in the Maotianshan
Shale; this mineral assemblage suggest the Maotianshan Shale was weathered from
granites and/or felsic volcanics. The illite present likely derived from the diagenetic
alteration of feldspars and/or other phyllosilicates (including kaolinite and muscovite)
(Fairbridge, 1967; Krauskopf and Bird, 1995; Lanson et al., 2002). Hofmann et al (in
press) identified alteration rinds and secondary clay minerals associated with orthoclase
in sandstones of the Maotianshan Shale, and it is likely that a similar process occurred in
the finer grained sediments analyzed in this study. Chlorite and pyrite are commonly
formed during alteration or diagenesis of the sediments (Berner, 1970; DeSegonzac,
1970) and will be discussed more in the next section. Hofmann et al (in press)
determined the majority of dolomite was formed authigenically by late-stage quartz
replacement. Detrital dolomite may also have been introduced into the Maotianshan
Shale sediments via weathering of the underlying Dahai Member of the Zhuhijajing Formation (Figure 3.1).

The exact of source(s) of the terrigenous minerals are still under investigation. Paleocurrent data presented by MacKenzie et al (2015) indicated sediments were delivered from multiple sources west of the study area, possibly the Central Yunnan Hills landmass. Hofmann et al (in press) cite the Kham-Dian Oldland (another name for the Central Yunnan Hills) and the southern Song Ma zone as possible sources. Both of these potential sources are felsic (Hofmann et al (in press) and references therein), consistent with the composition of the Maotianshan Shale sediments.

The provenance and composition of the sediments of the Maotianshan Shale are important as they likely played an important role in the preservation of the Chengjiang Biota and other BST deposits. BST deposits share commonalities in their overall depositional environments and taphonomic processes that fossilized their respective biotas. Several studies report that BST sediments were derived, at least in part, from continental sources (Gabbott et al., 2008; Brett et al., 2009; Gaines et al., 2011; Gehling et al., 2011; Ineson and Peel, 2001; Harvey et al., 2012; Le Boudec et al., 2014). This would suggest that the composition of the sediments could be a key factor to the taphonomy of BST deposits. One of the links between all of the BST Lagerstätten is that the exceptionally preserved fossils are found in rapidly deposited fine-grained mudstones (Conway Morris, 1985; Hagadorn, 2002c; Gaines et al., 2012), with the soft tissues being preserved as carbonaceous films (Gabbott et al., 2004; Zhu et al., 2005; Gaines et al., 2008). It is possible that the mineralogical and chemistry of terrigenous sediments promoted the fossilization of the tissues, preserving the organisms in such detail. Further
investigations into the composition and provenance of the mudstones entombing the fossils in various BST deposits are needed to verify this.

### 3.4.2 – Carbon and Temperature of the Sediments

Most of the carbon (79.9-97%; Table 3.4) in the Maotianshan Shale is inorganic carbon, the source of which is dolomite or other undetected carbonate minerals. All of the fresh samples contained dolomite, which is either detrital or diagenetic in origin. The remaining 2-21% of the total carbon was organic carbon. The concentrations of TOC detected in the samples range between 0.13 wt% to 0.21 wt%, which are considered low.

The low TOC values in the Maotianshan Shale may represent originally low concentrations of organic matter due to low productivity. Zhu et al (2001) reported TOC concentrations from fresh samples of lithofacies corresponding to the Maotianshan Shale Mbr between 0.3 and 0.4 wt.%, which are higher than our values between 0.13 and 0.21 wt.%. Zhu et al. (2001) also reported TOC concentrations up to 3.57 wt.% in lithofacies corresponding to the Black Shale Mbr, directly beneath the Maotianshan Shale Mbr. The sedimentation between these two members was continuous and any alteration (heating, etc.) would have affected both the Black Shale and Maotianshan Members equally. The differences in TOC values between these two members suggest that the Maotianshan Shale had initial low concentrations of organic matter compared to more typical marine black shales. The following discussion addresses possible explanations for these low organic carbon concentrations.

In modern upwelling zones, such as the west coast of South America (Brink et al., 1983; Figueroa and Moffat, 2000; Capone and Hutchins, 2013), high concentrations of
organic matter are produced when dense, nutrient-rich waters are transported from deep areas along the slope/shelf environment to surface waters, promoting primary productivity (Stein, 1991; Capone and Hutchins, 2013). Paleogeographic reconstructions of the South China block in the early Cambrian show a gently sloping basin that accumulated the Maotianshan Shale sediments (Lin et al., 1985; Wang et al., 2010; Wang et al., 2013), with no bathymetric evidence for deep water near the coast. This lack of nearshore deep water would have resulted in little to no upwelling (Brink et al., 1983; Stein, 1991), resulting in low concentrations of organic matter in the water column during deposition of the Maotianshan Shale.

Oxic conditions in the bottom waters (Goldberg et al., 2007) could have prevented the preservation of organic matter, derived from organic carbon settling from the water column or resuspended from the sediments, resulting in low TOC concentrations. Remineralization of TOC occurs under oxic conditions when microbes consume available organic matter (CH$_2$O) during aerobic respiration, resulting in carbon dioxide (CO$_2$) (Bjørlykke, 1983; Burdige, 2007), via the following process:

\[ \text{CH}_2\text{O} + \text{O}_2 \rightarrow \text{CO}_2 + \text{H}_2\text{O} \]

Remineralization of organic carbon also occurs during photodegradation/photolysis where the organic matter is degraded by photons from the sun and only occurs within the photic zone (Tett, 1990; McKee, 2003a). The Maotianshan Shale sediments were deposited in a relatively shallow environment, between the lower shoreface and the offshore transition zone (MacKenzie et al., 2015), well within the photic zone based on the occurrence of the Chengjiang Biota; photodegradation of organic carbon may have contributed to low TOC concentrations preserved within the sediments.
Higher TOC concentrations may have been expected in fossiliferous layers especially in the algal-rich, hemipelagic facies, but significant differences in TOC values between these facies and the other facies of the Maotianshan Shale were not observed. Although there are abundant algal-rich layers, the algal fossils are very thin, likely not containing detectable TOC concentrations. Likewise, the carbonaceous films preserving the fossils (Gabbott et al., 2004; Gaines et al., 2008) are also very thin, a probably do not contain detectable concentrations of organic matter.

If the TOC concentrations were originally higher than those observed in this study, the organic matter may have been lost after deposition due to one of two mechanisms: (1) resuspension and remineralization or (2) thermal maturation and migration. MacKenzie et al (2015) interpreted the depositional environment of the Maotianshan Shale as between the lower shoreface and the offshore transition zone between fair-weather and storm wave base, within the photic zone. The sediments were eroded and remobilized by waves, making buried organic matter available for remineralization (McKee, 2003b).

Loss of organic carbon during burial can occur via bacterial methanogenesis, thermal maturation/catagenesis, or by metamorphosis, the latter of which require elevated temperatures. Bacterial methanogenesis is a form of anaerobic respiration in which microbes consume the available carbon in the organic matter (generic CH$_2$O) and produce methane (CH$_4$) and carbon dioxide (CO$_2$) (Claypool and Kaplan, 1974; Burdige, 2007) as depicted by the following equation:

$$2(CH_2O) \rightarrow CH_4 + CO_2$$
When sediments are compacted and heated, the organic matter degrades, forming kerogen. Kerogen includes all solid organic matter found in sedimentary rocks (Hutton et al., 1994). The process of organic matter degradation is known as thermal maturation. Catagenesis converts kerogen to hydrocarbons (oil and gas) at elevated temperatures and/or pressures. Once hydrocarbons are formed they will migrate from the source rock (organic-rich shale) into a reservoir rock. Typically catagenesis requires temperatures between 50°C-150°C and pressures between 300-1500 bars; conditions usually found at burial depths greater than 1000 meters (Hunt, 1979; Tissot and Welte, 1984; Raiswell and Berner, 1986). The Maotianshan Shale sediments could have been exposed to slightly elevated temperatures and pressures during burial, leading to catagenesis and possibly migration of oil and gas out of the system. To better understand whether this process occurred, information about the thermal maturity of the Maotianshan Shale and surrounding sediments is required.

There is little information available describing the thermal maturity of the Maotianshan Shale and underlying organic-rich sediments. The black shales of the Yangtze Platform on the South China Block are considered to be important source rocks for hydrocarbons, especially in the Sichuan basin where these shales are 100-200 meters thick (Tan et al., 2013; Wang et al., 2013). These deposits are ~400 km to the NW of our study area; they are found on the same tectonic block as the Maotianshan Shale outcrops and can provide some information of the burial history of early Paleozoic sediments. The black shales of the Sichuan basin contain between 5% and 10% TOC and are considered over mature for oil production, having a vitrinite reflectance (Ro) greater than 2% (Tan et al, 2015). These sediments were buried to depths of up to 6000 m and experienced
temperatures up to 220°C by the end Jurassic (Tan et al., 2013), providing a maximum temperature for the black shales on this part of the tectonic block. Areas of the Yangtze platform surrounding the Sichuan basin, including our study area, were not as deeply buried (Tan et al., 2013); some areas were actually undergoing erosion in the middle Mesozoic (Tan et al., 2015).

There are two major NW/WNW trending faults, the Ziyun-Luodian fault and the Youjiang fault, separating our field area from the Sichuan basin (Wang et al., 2013). These faults likely displaced the Maotianshan Shale sediments relative to those in the Sichuan basin. Regardless of this, the thermal history of the Sichuan basin provides a maximum temperature for the region. If the Maotianshan Shale had been subjected to similar temperatures and burial depths, it is possible the TOC concentrations in the sediments were reduced via thermal maturation resulting in the low organic matter measured in this study.

The presence of clay minerals, illite and chlorite, in the Maotianshan Shale may support elevated temperatures. Illitization of kaolinite and smectites typically occurs between temperatures of 50°C and 100°C (Worden and Morad, 2003), well within the maximum temperature identified within the Sichuan basin. During this process kaolinite first alters to smectite during burial. With additional heating the smectite transforms into randomly ordered illite-smectite (I-S) mixed layer clays. Over time the I-S layers become ordered and, eventually, all the smectite alters to illite (Meunier, 2005). Illitization can also occur at lower temperatures over a long period of time (Smart and Clayton, 1985), so its presence in the Maotianshan Shale is not a definitive indicator of elevated temperatures. Diagenetic chlorite is formed through thermal alteration of mafic
igneous rocks around 100°C (Hillier, 1993); evidence of a minor mafic component in the Maotianshan Shale was identified by Hofmann et al (in press). During this process, smectites initially form, followed by mixed layer corrensite and chlorite phases with increasing temperatures, similar to the smectite-illite transition. Diagenetic chlorite can also be formed through the alteration of biotites and smectites (Dimberline, 1986; Hillier, 1993). Chlorite can form authigenically in sandstones at temperatures as low as 20°C-40°C (Grigsby, 2001), preventing it from being an indicator for high temperatures.

Illite and chlorite can also be formed during low-grade metamorphism at temperatures over 200°C (DeSegonzak, 1970; Kisch, 1987), which occurred to the Burgess Shale sediments. In the Burgess Shale, the sediments underwent greenschist metamorphism (200-280°C; ~10 km), resulting in a slaty texture of the sediments and the carbonaceous fossils being replaced by clay minerals (Orr et al., 1998; Powell, 2003; Butterfield et al., 2007). The sedimentary rocks of the Maotianshan Shale retain their original sedimentary textures and show no evidence of metamorphism. Additionally, many of the Chengjiang Biota fossils are preserved as their original carbon films (Gabbott et al., 2004; Gaines et al., 2008), suggesting they did not undergo heating to high temperatures; Burgess Shale fossils have lost their carbon films and only clay mineral films are preserved (Hagadorn, 2002b). Clay minerals have been identified in association with the Chengjiang Biota fossils, but they were determined to have formed authigenically as coatings during late diagenesis (Gabbott et al., 2004; Zhu et al., 2005).

A summary of the different paleotemperature proxies discussed is depicted in Figure 3.7. Based on our results and discussion above we do not have significant evidence that the Maotianshan Shale was subjected to extreme temperatures or that it had
originally high TOC values. Older marine shales have been documented to preserve lower organic carbon concentrations than younger ones (Raiswell and Berner, 1986), so our results are not surprising. These lower TOC values in the early Paleozoic are attributed to diagenetic loss of organic matter (through thermal maturation etc.) as well as lower original concentrations compared to younger sediments. In the early Paleozoic the
majority of the organic matter in the oceans was composed of easily consumed algal material (Burdige, 2007; Raiswell and Berner, 1986), which would have been rapidly removed from the water column before deposition. This is in contrast with organic matter derived from vascular plants, which evolved in the middle Paleozoic, and are not as easily consumed and are more likely to be preserved in the sediments (Raiswell and Berner, 1986; Canfield, 1994). Additionally, the contrast in TOC values for the Black Shale Member compared to the Maotianshan Shale Member (reported by Zhu et al., 2001) also supports our theory that the Maotianshan Shale originally had relatively low TOC concentrations. These low concentrations are similar to those reported from the Emu Bay Shale (TOC between 0.25-0.55%; McKirdy et al., 2011), another BST deposit, and may represent similar depositional conditions in these Lagerstätten.

3.4.3 – Paleoredox Conditions

The paleoredox indicators used in this study indicate conditions ranging from oxic to anoxic/sulfidic for the same samples (Table 3.5). These confounding data caused for reevaluation of the various proxies used to determine if they were appropriate for the Maotianshan Shale samples. Nickel, cobalt and vanadium are all elements that are typically found in association with organic matter and their presence is used to determine the paleoredox conditions of organic-rich black shales (Jones and Manning, 1994; Rimmer, 2004). As discussed above, the Maotianshan Shale is not rich in organic matter, and would likely be depleted in Ni, Co and V compared to typical black shales. Because of this the Ni/Co and V/Cr paleoredox indicator were not used in this study. U/Th indicator was also ultimately excluded as uranium, although immobile and stable, can be
overprinted by thorium in areas of high rates of sedimentation (Arthur and Sageman, 1994). The Maotianshan Shale sediments were deposited in a shoreface environment between the lower shoreface and offshore transition zone, and were subjected to wave reworking and relatively high sedimentation rates (MacKenzie et al., 2015), making the U/Th indicator potentially misleading.

Molybdenum concentrations greater than 2ppm suggest anoxic conditions were present in the porewaters of the Maotianshan Shale sediments prior to lithification (Dean et al., 1997; Rimmer, 2004). Molybdenum is an immobile trace element that is considered stable for long periods of time (Morford and Emerson, 1999; Rimmer, 2004), making it a good indicator of paleoredox conditions. For molybdenum to be present in the sediments it must be reduced from Mo(VI) to Mo(IV) under anoxic conditions where it will precipitate (Bertine, 1972; Morford and Emerson, 1999; Wang, 2012). The reliability of molybdenum as a redox indicator has been questioned in euxinic environments (e.g., Algeo and Lyons, 2006). Paleocurrent data in the Maotianshan Shale (see MacKenzie et al., 2015) preclude euxinic conditions during deposition of the sediments; because of this we feel comfortable using molybdenum as a paleoredox indicator.

Pyrite was identified in many of the sediments and in association with the fossils in the Maotianshan Shale. The pyrite is not extensive and does not make up a large component of the sediments or the fossils. Authigenic pyrite forms in anoxic sediments where there are sources of sulfide, organic matter and iron minerals (Berner, 1984). Elevated iron concentrations in the Maotianshan Shale compared to the rest of the BST deposits have been reported (Hammarlund, 2007), indicating iron was not limiting. Lack
of abundant, widespread pyrite throughout the sediments and fossils may have been limited by low organic carbon concentrations or limited available sulfate. Gaines et al (2012) analyzed the isotopic composition of sulfur in sedimentary pyrite associated with the Chengjiang fossils as well as in other BST deposits and identified low sulfate concentration in the Cambrian seawater, suggesting sulfate was a limiting factor to widespread pyrite formation; sulfidic conditions require anoxia but anoxic conditions do not require sulfide (Berner, 1984; Raiswell and Berner, 1986). Limited sulfate may also explain why pyrite was not detected in all the Maotianshan samples (Table 3.3).

The fossils of the Chengjiang Biota are preserved as original carbon films, like many other BST deposits, but framboidal pyrite has also been identified in association with the fossils (Gabbott et al., 2004; Zhu et al., 2005). Framboidal pyrite is commonly found in marine sediments and is considered to take months to years to form (Luther et al., 1982). In order for soft tissues to be preserved, they must be rapidly isolated (hours to days) to prevent decay, suggesting pyrite was not the primary mode of preservation of the fossils. Gaines et al (2012) cited localized anoxia and limited microbial activity as factors responsible for the preservation of the carbonaceous compressions in the Chengjiang fossils and other BST deposits. Unfortunately the majority of the exceptionally preserved, fossil-bearing event beds in the Maotianshan Shale are found in weathered parts of the stratigraphic sections and were thus excluded from this study. Analyses of newly exposed, grey samples of the event beds are needed to verify anoxic conditions associated with fossilization.

The presence of bioturbated firmgrounds (Gingras and Pemberton, 2000) and pelagic mudstones in the Maotianshan Shale (Mackenzie et al., 2015) suggest that oxic or
suboxic conditions were, at least occasionally, present in the bottom-waters as well as in the top of the sediments. Firmgrounds are formed by dewatering and/or compaction of the sediments and can take months to years to develop in modern environments. The dewatered sediments would have remained oxic to suboxic long enough to be bioturbated. During the Cambrian it is assumed that the high amounts of fine-grained sediments and the lack of abundant bioturbating organisms caused little sediment mixing and rapid dewatering of the sediments (Droser et al., 2002; Droser et al., 2004; Dzik, 2005), allowing for faster firmground development. The majority of the bioturbated intervals are located stratigraphically above the sediments discussed in this study, in weathered outcrops precluded from geochemical analyses. It is possible the Maotianshan Shale shifts from anoxic to suboxic or anoxic conditions up-section. Analyses of freshly exposed samples from the upper part of the section are needed to look for the presence of pyrite and to compare molybdenum concentrations.

3.5 – Conclusions

This study has highlighted several important features of the Maotianshan Shale directly bearing on the preservation of the Chengjiang Biota:

- The sediments were derived from a felsic source. This is similar to many other BST deposits and is likely an important factor in the preservation of BST fossils.
- The sediments are relatively poor in TOC, which is likely an original signature of organic carbon.
• The Maotianshan Shale was subjected to temperatures no higher than 220°C; this did not greatly alter the rocks or fossils, allowing for geochemical analyses to be conducted.
• The paleoredox conditions for the Maotianshan Shale were mostly anoxic, based on Mo values and the presence of pyrite, which would have aided in the rapid mineralization of the soft tissues.

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CHAPTER 4 – BIOFILMS AND TRACE ELEMENT FILMS AID SOFT TISSUE FOSSILIZATION

NOTE**

This manuscript was written based on original data. Subsequent scans on weathered event mudstone samples with Liesegang bands and without fossils demonstrated similar accumulations of trace elements on the Liesegang bands (Kohn pers. comm.).

4.1 – Abstract

Biofilms are thin microbial communities encased in extracellular polymeric substances, which are known to aid in fossilization, particularly in cases of extraordinary soft-tissue preservation in Konservat-Lagerstätten deposits. Modern marine biofilms take up trace elements (e.g., Cu, P, Fe, Zn, Cd). Here we report increased concentrations of trace elements (transition metals, heavy metals, P, actinides) on the surfaces of worm fossils from the Chengjiang biota of Yunnan, China. Abiotic processes cannot explain increased concentrations of all of these elements. Instead, we propose that biofilm surfaces take up trace elements to forms a thin, trace-element-rich film that prohibits rapid (hours to days) post-mortem decay. This allows early diagenetic cements to form a rigid rock matrix and results in excellent preservation of the fossils. This process may help explain soft-tissue preservation in other Lagerstätten, providing information into past environments and communities.
4.2 – Introduction

Biofilms are communities of microbes that are bound together by extracellular polymeric substance (EPS) and can survive in a wide range of environments. Modern biofilms have the ability to concentrate metals via biosorption causing an electronegative charge on the outer surface of the biofilm community and precipitation of mineral phases (Beveridge, 1989; Schultze-Lam et al., 1996). A geochemical microenvironment is created on the surface of the biofilm, promoting the absorption of trace elements and nutrients that would, under normal conditions, not be taken up (Schultze-Lam et al., 1996; van Hullebusch et al., 2003). Biofilm communities tolerate high concentrations of metals, such as copper and antimony, by sequestering the metal ions within the EPS layer as well as through interactions with metal-binding proteins on the microbial surfaces (Beveridge, 1989; Silver and Phung, 2005; Workentine et al., 2008). Reactive sites on the bacterial surfaces serve as nucleation sites for the precipitation of mineral phases, sometimes fossilizing the microbial cellular morphologies (Schultze-Lam et al., 1995). These microbial fossils are found throughout the geological record, from the Proterozoic Gunflint Chert (Barghoorn and Tyler, 1965) to Recent deposits in Yellowstone National Park (Cady and Farmer, 1996).

Biofilms are key for the preservation of soft tissues in Konservat-Lagerstätten deposits (Briggs, 2003); Konservat-Lagerstätten are exceptional fossil deposits in which soft tissues, such as muscles and organs, are preserved in addition to exo- and endoskeletal material (Bottjer et al., 2002). These deposits provide an unparalleled opportunity to study past life, diversity, and paleoecology (Briggs, 2003). Rapid burial in fine-grained sediments, suboxic to anoxic pore waters, low SO₄ concentrations (Allison,
1988; Gaines et al., 2012) and early mineralization (Seilacher et al., 1985), among other conditions, cause the rare fossilization of soft tissues.

In Lagerstätten deposits, biofilms create a microcosm around the organisms, isolating them from the environment and protecting them from degradation and disarticulation (Briggs, 2003; O’Brien, 2008). Examples of this fossilization process include the Ediacara biota, where biofilms and microbial mats are found coating the organisms and preserving them as casts and molds in coarse sediments (Gehling, 1999; Laflamme et al., 2011) and the Eocene Florissant Formation in which diatomaceous biofilms are found preserved as thin coatings fossilizing the insect and plant fossils (Harding and Chant, 2000; O’Brien et al., 2008). There is not always direct evidence of biofilms on exceptionally preserved fossils, and other techniques such as experimental taphonomy and geochemical analyses, as discussed in this manuscript, are used to infer microbial processes responsible for the preservation of soft-bodied fossils.

Experimental taphonomy highlights the importance of biofilms and microbes for fossilization (Briggs, 2003; Briggs et al., 2005). In these experiments, biofilms form within a few hours post-mortem, creating a chemically reduced microcosm around the organism (Briggs et al., 2005; Raff et al., 2008). Thin mineral films quickly form on the surfaces of the biofilms (Fein et al., 2001; Harrison et al., 2005), encasing and preserving the organism prior to cementation and compaction of the sediments.

The Chengjiang biota, the focus of this study, is the oldest (~520 Ma) (Babcock and Zhang, 2001) of the Burgess Shale-type (BST) deposits, a series of Konservat-Lagerstätten that depict the explosion of metazoan life and globally range in age from Early Cambrian to Ordovician (Hagadorn, 2002). BST deposits contain faunas that are
Figure 4.1 – (a) Map of China showing Yunnan Province and the field area (detail in (b)); (b) Exposures of the Chengjiang Biota localities (modified from Hu (2005)): 1) Shankou, 2) Ercai, 3) Xiaolantian, 4) Ma’anshan; (c) Image of *Palaeoscolex sinensis* showing the LA-ICP-MS scans across the fossil; (d) Simplified stratigraphic column of Early Cambrian strata, highlighting the Maotianshan Shale.

taxonomically similar to those of the Burgess Shale in Canada (Bottjer et al., 2002). The Chengjiang biota comprises a diverse fauna of soft-bodied organisms preserved in the Maotianshan Shale, a unit within the Yu’anshan Formation, exposed near Kunming, Yunnan, China (Chen and Erdtmann, 1991; Babcock et al., 2001; Hu, 2005) (Fig. 4.1a).
The fauna of the Chengjiang biota exhibits preserved soft tissues that provide insight into the evolution and diversification of several taxonomic groups (Chen and Erdtmann, 1991; Babcock et al., 2001; Chen, 2009). The soft-bodied fossils are found in fine-grained, clay-rich mudstone layers interpreted as event beds that rapidly buried the organisms (Babcock and Zhang, 2001). The exact mechanisms of preservation of these fossils, in particular the soft tissues, are still debated (Zhu et al., 2005; Gaines and Droser, 2010), but there is general consensus that microbial communities were somehow involved in this process (Chen and Erdtmann, 1991; Gaines et al., 2012), possibly under SO$_4$-poor conditions (Gaines et al., 2012). Here we report elevated concentrations of trace elements on the surface of fossil tissues compared to the surrounding sediments. We attribute this to biofilms encasing the organisms, although there are alternative explanations, such as local diagenetic processes and post depositional weathering, are also discussed.

4.3 – Materials and Methods

4.3.1 – Sample collection

Fossil samples of the fossil worm *Palaeoscolex sinensis* (Hou and Sun, 1988) were collected from four localities of near the cities of Chengjiang and Kunming, Yunnan, China (Fig. 4.1b). All of the samples were found in weathered event mudstones; the weathered rocks are more easily available than fresh ones, and the fossils are more recognizable in the weathered rocks.
4.3.2 – Sample Analysis

Trace-element profiles were collected across fossils using laser-ablation inductively coupled plasma mass spectrometry (LA-ICP-MS) at Boise State University based on methods described in Hinz and Kohn (2010). This method preferentially samples the surface of the fossil and emphasizes elemental concentrations across preserved tissues of the fossil compared to the matrix. The fossils were analyzed using a Thermo XSeries2 Quadrupole ICP-MS in conjunction with a NewWave UP-213 laser-ablation system. We used a laser spot diameter of 40 µm, a scan speed of 10 µm/sec, and a repetition rate of 10 Hz, with a fluence of 4-5 J/cm². As the fossils are preserved as thin films, we used a larger spot size to acquire more of the fossilized tissues. In addition, a low repetition rate and fluence were used to avoid penetrating though the fossil into the underlying sediment matrix, diluting the signal. We collected counts on \(^{23}\text{Na}\) (5 ms), \(^{24}\text{Mg}\) (10 ms, etc.), \(^{27}\text{Al}\), \(^{29}\text{Si}\), \(^{31}\text{P}\), \(^{39}\text{K}\), \(^{43}\text{Ca}\), \(^{44}\text{Ca}\), \(^{45}\text{Sc}\), \(^{49}\text{Ti}\), \(^{51}\text{V}\), \(^{52}\text{Cr}\), \(^{53}\text{Cr}\), \(^{54}\text{Fe}\), \(^{55}\text{Mn}\), \(^{57}\text{Fe}\), \(^{59}\text{Co}\), \(^{60}\text{Ni}\), \(^{63}\text{Cu}\), \(^{65}\text{Cu}\), \(^{88}\text{Sr}\), \(^{95}\text{Mo}\), \(^{121}\text{Sb}\), \(^{123}\text{Sb}\), \(^{137}\text{Ba}\), \(^{138}\text{Ba}\), \(^{140}\text{Ce}\), \(^{208}\text{Pb}\), \(^{232}\text{Th}\), and \(^{238}\text{U}\).

Background-corrected raw counts from samples were calibrated against NIST 610 and NIST 612 glasses, using Si as an internal standard and the average Si content from mudstones at our localities. Bulk mudstone XRF data were obtained commercially from SGS Canada. Traverses varied in length depending on the size of the fossils and 5 parallel scans that started in the matrix, crossed into the fossil and ended again in the matrix were acquired for each fossil. The 5 parallel scans were averaged and further smoothed with 3- and 5-point running averages. Statistical comparisons were made using two-tailed t-tests, (see Table A1), however in our experience, systematic errors up to 10% are possible in relative concentrations as measured by LA-ICP-MS. Consequently, elements exhibiting
concentrations in the fossils between 90 and 110% of matrix are not considered significantly different.

**4.4 – Results**

We collected ~2-4-mm long transects from the matrix, across the surface of the ~1-2-mm wide worm fossils, and back into the matrix to create matrix-worm-matrix geochemical profiles. Analyses were performed on six worm fossils (Fig. 4.2). All fossils show statistically significant, higher concentrations of P, Sc, V, Cr, Fe, Ni, Cu, Mo, Sb, Th, and U than the matrix (based on two-tailed t-tests with p≤0.05; see Table A1; Table 4.1, Fig. 4.3). Statistically significant increases in Na, Mg, Al, K, Ca, Mn, Co, Sr, Ba, Ce, and Pb were found in some, but not all samples. None of the samples showed significant increases in Ti.

**4.5 – Discussion**

In principle, accumulation of trace elements on the surface of the fossil might occur via two processes: 1) through abiotic uptake driven by local redox or geochemical conditions after the initial fossilization of the organism or 2) via biotic processes where biofilms rapidly form on the dead organism and accumulate trace elements on their surfaces prior to and during fossilization. We cannot rule out the first process although trace elements that show significant increases on the fossil worms have a wide range of geochemical behaviors. The elements analyzed include immobile vs. mobile elements, redox sensitive vs. insensitive elements, and elements that sorb onto particles over a range of redox conditions, including oxic, suboxic, and anoxic conditions (Table 4.1),
Figure 4.2 – Plots of composition vs. distance documenting elevated concentrations of (a) Fe and (b) Cu in the tissues compared to the matrix in the six fossils of *Palaeoscolex sinensis* used in this study. The scan lengths are listed on the right of each scan in (a). Fe concentrations range between 1E+4 and 2E+5 ppm. Cu concentrations range between 1E+1 and 1E+3 ppm.
Figure 4.3 – Plots of composition vs. distance documenting certain trace element elevated concentrations across the fossils.
making abiotic uptake of all of the elements unlikely. More specifically, organic-rich
tissues would be expected to impart local suboxic to anoxic conditions that would prevent
accumulation of P, V, Cr, Ni, and Sb, yet these elements have elevated concentrations in
the fossils compared to the matrix. Consequently, we favor our hypothesis that biofilms
concentrated trace elements and that the resulting trace metal films facilitated fossil
preservation. However, scans collected by Kohn (pers. comm.) on weathered, event,
Liesegang-banded mudstone samples lacking fossils demonstrated similar accumulations
of trace elements on the Liesegang bands. If these trace element accumulation events are
related then the enrichments occurred later, during the weathering process, enhancing the
fossils rather than aiding preservation.

Biofilms are known to accumulate trace elements and minerals on their surfaces
(Beveridge,1989; Schultze-Lam et al., 1996). They do this by forming
microenvironments in which conditions are favorable for concentration and accumulation
of these elements where they would otherwise not be expected due to the overall
chemical environment (Lion et al., 1988; Schultze-Lam et al., 1996; Huang et al., 2000;
Briggs, 2003; van Hullebusch et al., 2003). Elements accumulated by a biofilm must be
available as dissolved ions either in the water column or in pore waters (Fein et al.,
2001). Many trace elements enter the marine system adsorbed onto particles (e.g.,
sinking organic matter, clay minerals or iron oxides) and are subsequently released
(Morford and Emerson, 1999; Rimmer, 2004; Bruland and Lohan, 2006) during (i)
consumption of organic matter during aerobic respiration, (ii) reduction of iron oxides
from insoluble Fe(III) to soluble Fe(II) under suboxic conditions, or (iii) desorption of
ions from clay minerals due to changes in pH during respiration (Piper and Isaacs, 2001;
Table 4.1 - Trace element behavior under different redox conditions.

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<th>Sorbed under anoxic/sulfidic conditions</th>
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<th>Toxic**</th>
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† - elements showing significant increases in at least 50% of the samples
‡‡ - elements showing significant increases in 100% of the samples
* - immobile means insoluble under most, common marine conditions
** - toxicity based on Workentine et al. (2008).

Bruland and Lohan, 2005). Each of these processes involves reduced oxygen levels, suggesting suboxic to anoxic porewater developed at or shortly after burial of the organism.

Elements enriched in the fossil worms are redox sensitive except for P, Ni, Sb, and Pb (Table 4.1). The increased abundance of redox-sensitive elements on the fossils is consistent with their release from surfaces under suboxic to anoxic conditions and subsequent accumulation by biofilms (Morford and Emerson, 1999; Piper and Isaacs, 2001; Rimmer, 2004). Increased P on the fossil worm most likely results from its initial association with organic matter (Ruttenberg, 2005). Its continued presence suggests the
sediments became suboxic or anoxic shortly after deposition, preventing loss of P from the sediments through organic degradation. Titanium shows no change across the fossils because it is neither redox sensitive nor mobile once deposited in the sediments (Piper and Isaacs, 2001; Bruland and Lohan, 2006). In addition, there is no known biological role of titanium in organisms. Several reviews make no mention of Ti as having a biological role (the most recent reviews confirm this) (Workentine et al., 2008).

Although we favor post-mortem concentration of trace elements from nearby sediment, trace elements might alternatively have been derived from original worm tissues. Some modern worms (e.g. polychaete *Nereis diversicolor*) accumulate trace elements (Mn, Cu, Zn, Cd) as a means of detoxifying their environment (Bryan and Hummerstone, 1971; Zhou et al., 2003). If the trace elements came directly from the worms instead of from interstitial waters, the biofilms would have taken up the elements from the tissues shortly after the death of the worm. Although this possibility changes the source of trace elements, the basic processes remain unchanged – trace metals films retarded complete degradation of the fossils.

Low trace element concentrations in the tissues suggests it is unlikely that specific trace-element-rich mineral phases would be identified in the fossils of the Chengjiang or other BST Lagerstätten. Trace-element-rich films impart information about preservation and are detectable at lower concentrations by LA-ICP-MS than are mineral phases. The detection of thin, trace element-rich films on preserved soft tissues in other Lagerstätten may indicate a common mechanism of preservation for soft tissues in the fossil record, acting either alone or in concert with other factors such as rapid sedimentation and/or low O₂ (Gaines et al., 2012) or during post-depositional fluid transport.
4.6 – Acknowledgements

Professor Chen Junyuan was instrumental; his insight into the Chengjiang biota helped guide sample collection. Adam Johnson helped with sample collection. Drs. Jill Scott, Carrine Blank, and David Patterson discussed and evaluated aspects of this work. LAM was supported by NSF-EAPSI, Sigma Xi – Grants-in-Aid of Research, and the International Association of GeoChemistry PhD Student Research Grant, with additional support from The University of Montana. This work was supported by NSF grant EAR0819837 to MJK and by Boise State University.

4.7 – References


4.8 – Appendix
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CHAPTER 5 – USING EXPERIMENTAL TAPHONOMY TO DETERMINE RATES OF BIOFILM FORMATION ON HARD AND SOFT TISSUES IN BURIED SNAILS

5.1 - Introduction

_Konservat-Lagerstätten_ are deposits containing fossils with exceptional fidelity, including soft tissues (Allison and Briggs, 1993). These deposits are snapshots into past environments, revealing information about ecosystems and the diversification of taxonomic forms (Brett et al., 1997; Bottjer et al., 2002). Fossilized microorganisms (microbes) have been identified in such deposits including the Ediacara biota (Laflamme et al., 2011), Nusplingen (Briggs et al., 2005), and the Florrisant Formation (O’Brien et al., 2008; Henning et al., 2012). Taphonomic experiments have greatly improved our understanding of the roles microbes play in soft tissue preservation by controlling the driving factors such as anoxia, nutrient availability, pH, etc. (Briggs, 1995; Raff et al., 2014). These experiments have clarified the conditions required for the mineralization of tissues and the rates at which preservation occurs and have found that microbial biofilms play a significant role in the mineralization of tissues (Raff et al., 2014).

Biofilms are densely packed, surface bound communities of microbes encased in an extracellular polymeric substance (EPS) (Costerton et al., 1995; Harrison et al., 2005) that aids in the architecture and protection of the biofilm (Branda et al., 2005). Biofilms are often made up of a diverse community of microbes, including archaea, bacteria, protozoans, algae and fungi, which act in concert to survive in a variety of different environments (McLean et al., 1999; Branda et al., 2005; Harrison, 2005). The microbes
found within a biofilm can have multiple morphological forms (Figure 5.1), which are controlled by available energy sources and the chemical environment in which they are found (Gorby et al., 2006; Jiang et al., 2011). Biofilm EPS helps attract carbon and oxidants to keep the microbial community healthy and can help promote mineralization (Harrison, 2005). The chemistry of the EPS and the metabolic activity of the microbes create a microenvironment resulting in an anionic surface that will attract available dissolved cations, such as \( \text{Ca}^{2+} \), \( \text{Fe}^{3+} \), \( \text{Mn}^{2+} \), onto the surface of the biofilm. Experiments have shown that
mineral films will form when these ions are accumulated in large enough concentrations (Schultze-Lam et al., 1996; McLean et al., 1999; Channing and Edwards, 2003; Hippler et al., 2012).

Few taphonomical experiments have included sediments, which affect the mobility of dissolved cations and, as such, would affect the accumulation of such elements onto biofilms. With sediments in the experiments we can better model processes that occurred in many Lagerstätten deposits. In this study we buried freshwater snails (*Biomphalaria galabrata*) snails in fine-grained sediments to observe the effects sediment has on the rates of biofilm formation.

5.2 – Methods

In this experiment we buried snails in fine-grained sediments and inundated the sediments with artificial seawater. We sealed the experimental vessels to promote anoxia and subsequently sampled at three different times. The snails were harvested and examined for biofilms. The methods used are detailed below.

5.2.1 – Experimental Taphonomy

Granite, ground and passed through a 250-micron sieve, was used in the experiments to simulate the fine-grained, terrigenous sediments associated with many *Konservat-Lagerstätten* (Seilacher, 1990; Brett et al., 1997). Sediment was added to the 10mL mark in 68mL digestion tubes (Environmental Express) and artificial seawater (Coralife Scientific Grade Marine Salt Mix) was added to the 25mL mark. Once the sediment settled, two drops of methylene blue indicator were added to test for anoxia. One to three freshwater snails
(Biomphalaria glabrata) were added to each tube where the change in salinity caused them to retract their bodies into their shells. Additional sediment was added to the 40mL mark to cover the snails. Some snails floated up to the sediment-water interface, probably because air was trapped in the shells. All of the buried snails, regardless of whether they were partially or fully buried, are referred to as “buried in sediment” (BS) snails. Tubes were sealed and left at ambient room temperature for the duration of the experiment.

Triplicate tubes were opened at 1, 2, and 8 weeks, and the excess saltwater was extracted and replaced with an equal volume of 50% glutaraldehyde solution to stop the experiment. The tubes were covered and refrigerated for a week to allow the glutaraldehyde solution to penetrate the pore spaces, fixing the snail tissues and any biofilms present in situ. After one week, the snails were carefully removed from the tubes and dehydrated for analysis following methods of Nation (1983).

Control samples were analyzed to observe the differences between buried snails and those without sediments. At the beginning of the experiment, snails were placed directly in the glutaraldehyde solution, they did not undergo any experimental conditions; these are referred to as “freshwater start” (FS) snails. These samples provide information about the condition of the tissues and the distribution and morphology of any biofilms present before the experiment. “Saltwater control” (SC) snails were placed in the artificial seawater solution for the same time intervals as the experimental snails to observe any tissue deterioration and biofilm formation in the absence of sediment. The FS and SC snails were prepared and analyzed in the same manner as the experimental snails.
5.2.2 – Sample Imaging

Macro-scale observation of the exoskeletons and soft tissues were done using a Leica M26 stereomicroscope and images were taken with a Lumenera INFINITY 1-3C camera mounted to the microscope. Micro-scale observations of exoskeletons and soft tissues were made by scanning electron microscopy and energy-dispersive x-ray spectroscopy (SEM-EDS). Snails were mounted on SEM stubs with double-sided carbon tape and liquid silver, to prevent charging, and sputter-coated with carbon. Snails that charged in the SEM were additionally coated using a gold-palladium alloy. Snail exoskeletons were imaged with a Hitachi S-4700 Field Emission SEM. Soft tissues were analyzed with a Tescan Vega 3 SEM and Oxford Instruments X-Max EDS.

The morphology of the biofilms were described by noting relative percentages of microbes of bacillus, coccoid and filamentous forms (Figure 5.1). Point counts of the microbes were done by overlying a 1x1 micron grid on SEM images and recording the shape of the microbes that fell under the nodes of the grid. Relative percentages of the microbial forms for each of the samples were calculated. These counts were also used to determine the percentages of shell covered by the biofilms.

5.3 – Results

5.3.1 – Freshwater Snails

The FS snails had a diverse biofilm community on their shells (Figure 5.2A) composed of microbes of bacillus (77.3%), of coccoid (16.7%), and filamentous (6.1%)
Figure 5.2 - Freshwater snail (FS) prior to the beginning of the experiment. (A) Sketch of the snail shell, stippling represents the distribution of the microbial biofilm on the snail shell. (A’) SEM image of the bacillus-dominated biofilm on the FS snail (scale bar equals 10 microns). (B) Sketch of the FS snail soft body (body in grey). (B’) Photograph showing the preserved morphology of the FS snail body (scale increments equal 1 mm). No biofilms were detected on the soft tissues of FS snail.
forms (Tables 5.1 and 5.2). The biofilms, on average, covered 31% of the snail shell (Table 5.3).

The soft bodies of the FS snails were retracted into their shells, but the general morphology of the snail mantle was preserved (Figure 5.2B). Biofilms were not found on any of the soft tissues of the FS snails.

5.3.2 – Experimental Snails

The SC and BS tubes became anoxic within 24 hours. Anoxic conditions persisted throughout the experiment. A dark grey halo was observed in the sediment surrounding experimental snails located near the side of the tube; these halos were 2-3 mm thick surrounding the shell.

A thin layer of sediment coated the shells of all of the BS snails, regardless of whether they were partially or fully buried. The biofilms observed on the snail shells were located beneath the thin layer of sediments and are described in detail below and summarized in Figure 5.3 and Table 5.1. Tables 5.2 and 5.3 summarize the morphological diversity of the microbes and their distributions on the snail shell, respectively.

All the snails used in the experiment, including the FS and SC snails, retracted their bodies into their shells. The FS snails did so when added to the glutaraldehyde solution. The SC and BS snails retracted into their shells when added to the artificial seawater mixture. A solid sediment plug formed in many of the BS snails and remained intact in the opening of the shell after removal of the snail from the sediments. The sediment plugs extended 2-4 mm into the shell, never reaching the soft tissues, leaving open space within the shell.
Table 5.1 – Descriptions of the biofilms detected on the snail exoskeletons. Freshwater starts (FS) and saltwater controls (SC) snails are noted in the sample number column. Positions of the snail in the sediment are fully buried (B) or partially exposed at the surface-water interface (SW).

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<td>patchy to extensive distribution; single layer to overlapping microorganisms EPS visible in areas with densely packed organisms</td>
</tr>
<tr>
<td>8 weeks</td>
<td>5</td>
<td>SW</td>
<td>patchy to extensive distribution; single layer of microorganisms; EPS visible in densely packed patches of microorganisms</td>
</tr>
<tr>
<td>8 weeks</td>
<td>6</td>
<td>B</td>
<td>patchy to extensive distribution; single layer to overlapping microorganisms EPS visible in areas with densely packed organisms</td>
</tr>
<tr>
<td>8 weeks</td>
<td>6.2</td>
<td>B</td>
<td>patchy to extensive distribution; single layer of microorganisms; EPS visible in densely packed patches of microorganisms</td>
</tr>
<tr>
<td>8 weeks</td>
<td>SC</td>
<td>no sediment</td>
<td>extensive; often overlapping layers of cells; EPS extensive</td>
</tr>
</tbody>
</table>
Table 5.2 – Morphology of the biofilms detected on the exoskeletons (shells) of the snails used in this study. Microbial counts were done by overlaying a 1x1 micron grid over SEM images of the biofilms and counting the microbes that were located on the nodes of the grid.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Total# of microbes</th>
<th>Bacillus</th>
<th>Coccoidal</th>
<th>Filamentous</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>FRESHWATER STARTS</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Freshwater 1</td>
<td>129</td>
<td>97 (75.2%)</td>
<td>29 (22.9%)</td>
<td>3 (2.3%)</td>
</tr>
<tr>
<td>Freshwater 2</td>
<td>151</td>
<td>116 (76.8%)</td>
<td>20 (13.2%)</td>
<td>15 (10%)</td>
</tr>
<tr>
<td>Freshwater 3</td>
<td>190</td>
<td>153 (80.5%)</td>
<td>28 (14.7%)</td>
<td>9 (4.7%)</td>
</tr>
<tr>
<td>Freshwater 4</td>
<td>308</td>
<td>221 (71.7%)</td>
<td>64 (20.8%)</td>
<td>23 (7.5%)</td>
</tr>
<tr>
<td>Freshwater 5</td>
<td>523</td>
<td>431 (82.4%)</td>
<td>61 (11.7%)</td>
<td>31 (5.9%)</td>
</tr>
<tr>
<td>**Freshwater %</td>
<td>77.32%</td>
<td>16.66%</td>
<td>6.08%</td>
<td></td>
</tr>
<tr>
<td><strong>SALTWATER CONTROLS</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Week 1-1</td>
<td>275</td>
<td>226 (82.2%)</td>
<td>17 (6.2%)</td>
<td>32 (11.6%)</td>
</tr>
<tr>
<td>Week 1-2</td>
<td>136</td>
<td>112 (82.3%)</td>
<td>11 (8.1%)</td>
<td>13 (9.5%)</td>
</tr>
<tr>
<td>Week 1-3</td>
<td>302</td>
<td>267 (88.4%)</td>
<td>8 (2.6%)</td>
<td>27 (8.9%)</td>
</tr>
<tr>
<td>Week 1-4</td>
<td>677</td>
<td>522 (91.8%)</td>
<td>38 (5.6%)</td>
<td>17 (2.5%)</td>
</tr>
<tr>
<td>Week 1-5</td>
<td>162</td>
<td>155 (95.7%)</td>
<td>0 (0%)</td>
<td>7 (4.3%)</td>
</tr>
<tr>
<td><strong>WEEK 1 TOTALS</strong></td>
<td>88.08%</td>
<td>4.50%</td>
<td>7.36%</td>
<td></td>
</tr>
<tr>
<td>Week 2-1</td>
<td>200</td>
<td>161 (80.5%)</td>
<td>27 (13.5%)</td>
<td>12 (6%)</td>
</tr>
<tr>
<td>Week 2-2</td>
<td>371</td>
<td>305 (82.2%)</td>
<td>23 (6.2%)</td>
<td>43 (11.6%)</td>
</tr>
<tr>
<td>Week 2-3</td>
<td>137</td>
<td>121 (88.3%)</td>
<td>3 (2.2%)</td>
<td>13 (9.5%)</td>
</tr>
<tr>
<td>Week 2-4</td>
<td>213</td>
<td>162 (76%)</td>
<td>18 (8.4%)</td>
<td>33 (15.5%)</td>
</tr>
<tr>
<td>Week 2-5</td>
<td>463</td>
<td>450 (97.2%)</td>
<td>7 (1.5%)</td>
<td>6 (1.3%)</td>
</tr>
<tr>
<td><strong>WEEK 2 TOTALS</strong></td>
<td>84.84%</td>
<td>6.36%</td>
<td>8.78%</td>
<td></td>
</tr>
<tr>
<td>Week 8-1</td>
<td>201</td>
<td>173 (86%)</td>
<td>12 (6%)</td>
<td>16 (8%)</td>
</tr>
<tr>
<td>Week 8-2</td>
<td>250</td>
<td>183 (73.2%)</td>
<td>31 (12.4%)</td>
<td>36 (14.4%)</td>
</tr>
<tr>
<td>Week 8-3</td>
<td>111</td>
<td>98 (88.3%)</td>
<td>4 (3.6%)</td>
<td>9 (8.1%)</td>
</tr>
<tr>
<td>Week 8-4</td>
<td>176</td>
<td>160 (90.1%)</td>
<td>10 (5.7%)</td>
<td>6 (3.4%)</td>
</tr>
<tr>
<td>Week 8-5</td>
<td>338</td>
<td>312 (80.4%)</td>
<td>14 (3.6%)</td>
<td>12 (3.1%)</td>
</tr>
<tr>
<td><strong>WEEK 8 TOTALS</strong></td>
<td>83.60%</td>
<td>6.26%</td>
<td>7.40%</td>
<td></td>
</tr>
<tr>
<td><strong>BURIED IN SEDIMENT</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>W1 exp1</td>
<td>16</td>
<td>11 (69%)</td>
<td>5 (31%)</td>
<td>0</td>
</tr>
<tr>
<td>W1 exp2</td>
<td>40</td>
<td>33 (82%)</td>
<td>5 (12%)</td>
<td>2 (5%)</td>
</tr>
<tr>
<td>W1 exp3</td>
<td>41</td>
<td>25 (61%)</td>
<td>11 (27%)</td>
<td>5 (12%)</td>
</tr>
<tr>
<td>W1 exp4</td>
<td>9</td>
<td>8 (89%)</td>
<td>1 (11%)</td>
<td>0</td>
</tr>
<tr>
<td>W1 exp5</td>
<td>17</td>
<td>11 (65%)</td>
<td>5 (29%)</td>
<td>1 (6%)</td>
</tr>
<tr>
<td><strong>WEEK 1 EXP TOTALS</strong></td>
<td>73.20%</td>
<td>21.80%</td>
<td>4.60%</td>
<td></td>
</tr>
<tr>
<td>W2 exp1</td>
<td>45</td>
<td>31 (69%)</td>
<td>13 (29%)</td>
<td>1 (2%)</td>
</tr>
<tr>
<td>W2 exp2</td>
<td>49</td>
<td>39 (79%)</td>
<td>9 (18%)</td>
<td>1 (2%)</td>
</tr>
<tr>
<td>W2 exp3</td>
<td>55</td>
<td>44 (80%)</td>
<td>9 (16%)</td>
<td>2 (4%)</td>
</tr>
<tr>
<td>W2 exp4</td>
<td>158</td>
<td>140 (89%)</td>
<td>16 (10%)</td>
<td>2 (1%)</td>
</tr>
<tr>
<td>W2 exp5</td>
<td>57</td>
<td>51 (89%)</td>
<td>6 (10%)</td>
<td>1 (2%)</td>
</tr>
<tr>
<td><strong>WEEK 2 EXP TOTALS</strong></td>
<td>81.20%</td>
<td>16.60%</td>
<td>2.20%</td>
<td></td>
</tr>
<tr>
<td>W8 exp1</td>
<td>44</td>
<td>33 (75%)</td>
<td>6 (14%)</td>
<td>5 (11%)</td>
</tr>
<tr>
<td>W8 exp2</td>
<td>25</td>
<td>15 (60%)</td>
<td>4 (16%)</td>
<td>6 (24%)</td>
</tr>
<tr>
<td>W8 exp3</td>
<td>132</td>
<td>100 (76%)</td>
<td>30 (23%)</td>
<td>2 (1%)</td>
</tr>
<tr>
<td>W8 exp4</td>
<td>150</td>
<td>121 (81%)</td>
<td>26 (17%)</td>
<td>3 (2%)</td>
</tr>
<tr>
<td>W8 exp5</td>
<td>45</td>
<td>29 (64%)</td>
<td>5 (11%)</td>
<td>11 (24%)</td>
</tr>
<tr>
<td><strong>WEEK 8 EXP TOTALS</strong></td>
<td>71.20%</td>
<td>16.20%</td>
<td>12.40%</td>
<td></td>
</tr>
</tbody>
</table>
### Table 5.3 – Calculated percentages of the morphological distribution of the biofilms on the snail exoskeletons.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Total # of nodes</th>
<th>Nodes with microbes</th>
<th>Percentage of shell covered</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>FRESHWATER STARTS</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Freshwater 1</td>
<td>468</td>
<td>129</td>
<td>27.6%</td>
</tr>
<tr>
<td>Freshwater 2</td>
<td>330</td>
<td>151</td>
<td>45.8%</td>
</tr>
<tr>
<td>Freshwater 3</td>
<td>900</td>
<td>190</td>
<td>21.1%</td>
</tr>
<tr>
<td>Freshwater 4</td>
<td>925</td>
<td>308</td>
<td>33.3%</td>
</tr>
<tr>
<td>Freshwater 5</td>
<td>2052</td>
<td>523</td>
<td>25.5%</td>
</tr>
<tr>
<td>Freshwater %</td>
<td></td>
<td></td>
<td>30.6%</td>
</tr>
<tr>
<td><strong>SALTWATER CONTROLS</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Week 1-1</td>
<td>682</td>
<td>275</td>
<td>40.3%</td>
</tr>
<tr>
<td>Week 1-2</td>
<td>330</td>
<td>136</td>
<td>41.2%</td>
</tr>
<tr>
<td>Week 1-3</td>
<td>682</td>
<td>302</td>
<td>44.3%</td>
</tr>
<tr>
<td>Week 1-4</td>
<td>2052</td>
<td>677</td>
<td>33.0%</td>
</tr>
<tr>
<td>Week 1-5</td>
<td>408</td>
<td>162</td>
<td>39.7%</td>
</tr>
<tr>
<td><strong>WEEK 1 TOTALS</strong></td>
<td></td>
<td></td>
<td>39.7%</td>
</tr>
<tr>
<td>Week 2-1</td>
<td>682</td>
<td>200</td>
<td>29.3%</td>
</tr>
<tr>
<td>Week 2-2</td>
<td>682</td>
<td>421</td>
<td>61.7%</td>
</tr>
<tr>
<td>Week 2-3</td>
<td>408</td>
<td>137</td>
<td>33.6%</td>
</tr>
<tr>
<td>Week 2-4</td>
<td>600</td>
<td>213</td>
<td>35.5%</td>
</tr>
<tr>
<td>Week 2-5</td>
<td>682</td>
<td>463</td>
<td>67.9%</td>
</tr>
<tr>
<td><strong>WEEK 2 TOTALS</strong></td>
<td></td>
<td></td>
<td>45.6%</td>
</tr>
<tr>
<td>Week 8-1</td>
<td>408</td>
<td>201</td>
<td>49.3%</td>
</tr>
<tr>
<td>Week 8-2</td>
<td>408</td>
<td>250</td>
<td>61.3%</td>
</tr>
<tr>
<td>Week 8-3</td>
<td>330</td>
<td>111</td>
<td>33.6%</td>
</tr>
<tr>
<td>Week 8-4</td>
<td>408</td>
<td>176</td>
<td>43.1%</td>
</tr>
<tr>
<td>Week 8-5</td>
<td>962</td>
<td>338</td>
<td>35.1%</td>
</tr>
<tr>
<td><strong>WEEK 8 TOTALS</strong></td>
<td></td>
<td></td>
<td>44.5%</td>
</tr>
<tr>
<td><strong>BURIED IN SEDIMENT</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>W1 exp 1</td>
<td>330</td>
<td>16</td>
<td>4.8%</td>
</tr>
<tr>
<td>W1 exp 2</td>
<td>962</td>
<td>40</td>
<td>4.2%</td>
</tr>
<tr>
<td>W1 exp 3</td>
<td>408</td>
<td>41</td>
<td>10.0%</td>
</tr>
<tr>
<td>W1 exp 4</td>
<td>425</td>
<td>9</td>
<td>2.1%</td>
</tr>
<tr>
<td>W1 exp 5</td>
<td>425</td>
<td>17</td>
<td>4.0%</td>
</tr>
<tr>
<td><strong>WEEK 1 EXP TOTALS</strong></td>
<td></td>
<td></td>
<td>5.0%</td>
</tr>
<tr>
<td>W2 exp 1</td>
<td>513</td>
<td>45</td>
<td>8.8%</td>
</tr>
<tr>
<td>W2 exp 2</td>
<td>408</td>
<td>49</td>
<td>12.0%</td>
</tr>
<tr>
<td>W2 exp 3</td>
<td>330</td>
<td>55</td>
<td>16.7%</td>
</tr>
<tr>
<td>W2 exp 4</td>
<td>1290</td>
<td>158</td>
<td>12.2%</td>
</tr>
<tr>
<td>W2 exp 5</td>
<td>425</td>
<td>57</td>
<td>13.4%</td>
</tr>
<tr>
<td><strong>WEEK 2 EXP TOTALS</strong></td>
<td></td>
<td></td>
<td>12.6%</td>
</tr>
<tr>
<td>W8 exp 1</td>
<td>425</td>
<td>44</td>
<td>10.4%</td>
</tr>
<tr>
<td>W8 exp 2</td>
<td>513</td>
<td>25</td>
<td>4.9%</td>
</tr>
<tr>
<td>W8 exp 3</td>
<td>682</td>
<td>132</td>
<td>19.4%</td>
</tr>
<tr>
<td>W8 exp 4</td>
<td>962</td>
<td>150</td>
<td>15.6%</td>
</tr>
<tr>
<td>W8 exp 5</td>
<td>330</td>
<td>54</td>
<td>16.4%</td>
</tr>
<tr>
<td><strong>WEEK 8 EXP TOTALS</strong></td>
<td></td>
<td></td>
<td>13.3%</td>
</tr>
</tbody>
</table>
The presence/absence of these plugs as well as observations of the soft tissues in SC and BS snails are summarized in Figure 5.4 and Table 5.4.

SEM-EDS identified no mineral films associated with biofilms identified in this experiment. Snails in replicate tubes all had similar results.

The microbes forming the biofilm in our experiments may have come from several sources: 1) the DI water used, 2) the salts in the artificial seawater mixture, 3) the glassware and/or the tubes used, 4) the sediments and/or the snails themselves. The aim of this study was to observe the formation and development of biofilms in the presence of sediments, the source(s) of the biofilms are not important and no attempt was made to keep the system sterile. Regardless of the origin of the microbes, new biofilms formed under the controlled conditions and their development was slowed due to the presence of the sediments.

5.3.3 – Results After 1 Week

After 1 week, the shells of the BS snails had poorly developed biofilms composed of a single layer of cells with a patchy distribution across the shell (Figure 5.3a). EPS was only observed where microbes were very densely packed. The majority of the microbes had bacillus morphology (73.2%). Some coccoid (21.8%) and filamentous (4.6%) forms were also observed. Surface analyses using point counts indicate the biofilms after week 1 covered on average 5% of the shell, much less than the biofilms on the FC shells.

The soft tissues of the week 1 BS snails were shrunken and cracked, but the overall morphology of the foot and mantle of the snail was still visible (Figure 5.4a). There were no morphological differences in the snail tissues between those with sediment plugs and
Figure 5.3 – Comparisons of biofilm distributions and morphologies between experimental (BS) and control (SC) snail exoskeletons during the three experimental time steps (stippling on sketches represents the biofilm coverage on the shell). Scale bars equal 10 microns.
<table>
<thead>
<tr>
<th>WEEK</th>
<th>EXPERIMENTAL</th>
<th>CONTROL</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>![Image 1]</td>
<td>![Image 2]</td>
</tr>
<tr>
<td>2</td>
<td>![Image 3]</td>
<td>![Image 4]</td>
</tr>
<tr>
<td>8</td>
<td>![Image 5]</td>
<td>![Image 6]</td>
</tr>
</tbody>
</table>

**Figure 5.4** - Sketches and images of soft tissues of the experimental (BS) and control (SC) snails for each time step. Experimental snails are figured with and without sediment plugs. Scale increment equals 1 mm.
those without. Very patchy biofilms with microbes of bacillus and filamentous morphologies were observed on anterior portions of the bodies of the snails without sediment plugs. No biofilms were identified in snails with sediment plugs.

The SC snail shells had an extensive biofilm, covering approximately 40% of the shell, composed of overlapping cells connected with EPS (Figure 5.3b). These biofilms were dominated with microbes of bacillus morphology (88%) with subordinate coccoidal (4.5%) and filamentous (7.4%) morphologies. The tissues of the one-week SC snails were dehydrated but, like the buried snails, the overall tissue morphology was preserved (Figure 5.4b). Patchy biofilms, dominated by bacillus-form microbes, were identified on the soft tissues of some of the SC snails.

5.3.4 – Results After 2 Weeks

After 2 weeks the biofilms on the shells of the BS snails were composed of a single layer of microbes and had a patchy distribution (averaging 12.6% of the shell; Figure 5.3c). Microbes of bacillus form were the dominant morphology in the biofilms (81.2%) with coccoid (16.6%) and filamentous (2.2%) forms also present. The biofilm EPS was observed more frequently at 2 weeks; more densely packed groups of microbes were observed in these snails than those from week 1.

The soft tissues of the week 2 BS snails were increasingly shrunken and cracked compared to the snails after week 1 (Figure 5.4c). The soft tissues of the snails without sediment plugs were completely dehydrated, shrunken, and flattened to the inner whorl of the shell, limiting identification of morphological detail. In some cases, the main body had started to disintegrate and isolated pieces were found in the hollow of the shell. Snails with
sediment plugs were less shrunken and more complete, still preserving some of the original shape of the snail body. Biofilms were not detected on any of the soft tissues of the BS snails.

**Table 5.4** – Description of the soft tissues of the snails used in the study. The morphology of the microbes are identified as bacillus (B), coccoid (C), or filamentous (F). Freshwater starts (FS) and saltwater controls (SC) are noted. Positions of the snail in the sediment are fully buried (B) or exposed at the surface-water interface (SW).

<table>
<thead>
<tr>
<th>Harvest Date</th>
<th>Sample Number</th>
<th>Position in Sediment</th>
<th>Biofilm Present?</th>
<th>Morphology of Microbes</th>
<th>Sediment Plug?</th>
<th>Description of Soft Tissues</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 weeks</td>
<td>FS</td>
<td>no sediment</td>
<td>no</td>
<td>no</td>
<td>no</td>
<td>intact soft tissues preserving morphological detail</td>
</tr>
<tr>
<td>1 week</td>
<td>1</td>
<td>B</td>
<td>yes</td>
<td>x</td>
<td>x</td>
<td>no slight dehydration of the tissues; much of the morphological detail preserved; very patchy biofilms present</td>
</tr>
<tr>
<td>1 week</td>
<td>4.2</td>
<td>SW</td>
<td>yes</td>
<td>x</td>
<td>no</td>
<td>no slight dehydration of the tissues; much of the morphological detail preserved; very patchy biofilms present</td>
</tr>
<tr>
<td>1 week</td>
<td>6.3</td>
<td>SW</td>
<td>no</td>
<td>yes</td>
<td></td>
<td>no slight dehydration of the tissues; much of the morphological detail preserved</td>
</tr>
<tr>
<td>1 week</td>
<td>SC</td>
<td>no sediment</td>
<td>yes</td>
<td>x</td>
<td>x</td>
<td>n/a intact soft tissues preserving much of the anatomical detail; patchy biofilms present</td>
</tr>
<tr>
<td>2 weeks</td>
<td>2</td>
<td>SW</td>
<td>no</td>
<td>no</td>
<td></td>
<td>tissues dehydrated but intact, compressed to inner whorl of shell; morphology partially preserved</td>
</tr>
<tr>
<td>2 weeks</td>
<td>3</td>
<td>B</td>
<td>yes</td>
<td>yes</td>
<td></td>
<td>tissues dehydrated but intact, slightly compressed; morphology partially preserved</td>
</tr>
<tr>
<td>2 weeks</td>
<td>5</td>
<td>B</td>
<td>no</td>
<td>no</td>
<td></td>
<td>tissues dehydrated and flattened; some tissue sloughed off; little morphology preserved</td>
</tr>
<tr>
<td>2 weeks</td>
<td>5.3</td>
<td>SW</td>
<td>no</td>
<td>yes</td>
<td></td>
<td>tissues dehydrated, mostly intact; some pieces sloughed off but overall morphology preserved</td>
</tr>
<tr>
<td>2 weeks</td>
<td>SC</td>
<td>no sediment</td>
<td>yes</td>
<td>x</td>
<td>x</td>
<td>n/a intact soft tissues preserving much of the anatomical detail; patchy to extensive biofilms present</td>
</tr>
<tr>
<td>8 weeks</td>
<td>2</td>
<td>B</td>
<td>yes</td>
<td>yes</td>
<td></td>
<td>tissues dehydrated, mostly intact; only slight morphology preserved</td>
</tr>
<tr>
<td>8 weeks</td>
<td>4.2</td>
<td>SW</td>
<td>no</td>
<td>no</td>
<td></td>
<td>tissues dehydrated and mostly disarticulated; morphological detail not preserved</td>
</tr>
<tr>
<td>8 weeks</td>
<td>5</td>
<td>SW</td>
<td>no</td>
<td>no</td>
<td></td>
<td>tissues dehydrated and mostly disarticulated; morphological detail not preserved</td>
</tr>
<tr>
<td>8 weeks</td>
<td>6.2</td>
<td>B</td>
<td>no</td>
<td>yes</td>
<td></td>
<td>tissues dehydrated and partially disarticulated; some morphology still preserved</td>
</tr>
<tr>
<td>8 weeks</td>
<td>SC</td>
<td>no sediment</td>
<td>yes</td>
<td>x</td>
<td>x</td>
<td>n/a intact soft tissues preserving much of the anatomical detail; patchy to extensive biofilms present</td>
</tr>
</tbody>
</table>

The week 2 SC snails, like the week 1 SC snails, had an extensive biofilm that covered almost half (45.6%) the snail shell (Figure 5.3d). The biofilms had extensive EPS and overlapping microbes. These biofilms were dominated by microbes of bacillus form (84.8%) with microbes of coccoid (6.4%) and filamentous (8.8%) form also present. The
soft tissues of the SC were dehydrated by retained the overall shape of the foot and mantle of the snail (Figure 5.4d). The soft tissues had patchy to extensive biofilms dominated by microbes of bacillus morphologies.

5.3.5 – Results After 8 Weeks

At the end of the experiment 8 weeks later, the biofilms on the BS snail shells had a patchy distribution covering, on average, 13.3% of the snail shell (Figure 5.3e). The biofilms were composed of a thin layer of microbes dominated by bacillus morphology (71.2%) with coccoid (16.2%) and filamentous (12.4%) forms also present. The EPS layer was frequently observed in the week 8 BS snails, especially where the microbes were densely packed.

After 8 weeks, the soft tissues of the BS snails without sediment plugs were shrunken and most disintegrated with only small bits of tissue left in the shell; little morphological detail was preserved (Figure 5.4e). The remnants of the tissues in these snails were largely identified using SEM-EDX analyses (Figure 5.5). The snails with sediment plugs were also shrunken, but the basic shape of the mantle was still visible. No biofilms were observed on the soft tissues of the BS snails harvested at 8 weeks.

The SC snails from week 8, like the other saltwater controls, had an extensive biofilm (averaging 44.5% of shell) of overlapping cells and extensive EPS on their shell (Figure 5.3f). The biofilm was dominated by microbes of bacillus morphology (83.6%) with some coccoidal (6.3%) and filamentous (7.4%) morphologies. The soft tissues of the SC snails were intact, preserving the morphology of the snail body (Figure 5.4f). Patchy to extensive biofilms were observed on the soft tissues and consisted of microbes of bacillus morphology.
**Figure 5.5** – Left: SEM image of a week 8 snail without a sediment plug. Most of the soft tissues are dehydrated and disarticulated throughout the shell. The rectangle on the image highlights the area that was analyzed using EDS. Right: SEM image of the inner whorl of the shell with some remnants of the soft tissues preserved. An elemental map of carbon (C) helped with our identification of the tissues. The other elements identified are noted: Al-aluminum, Ca-calcium, K-potassium, Si-silicon.

**5.4 – Discussion**

Haloes in the sediment surrounding the BS snails were likely sulfidic zones formed around the decaying organism. Darroch et al (2012) and Martin et al (2004) both reported similar dark haloes in the sediments around decaying animals. Both studies identified the darker sediments as containing iron sulfide minerals formed by sulfate reducing bacteria. The haloes observed in our study were likely formed via similar processes, although we did not test for the presence of sulfide minerals in the sediments.

These dark haloes surrounding the buried snails indicate the microbes in the biofilms lived heterotrophically as reactants diffused into the sediments around the snails; the conditions for chemo- and photoautotrophy were not present. Heterotrophic biofilms need
two main factors to thrive: an oxidant (O\(_2\), Fe(III), S(VI)) and a carbon source (Burdige, 2006). Aerobic respiration yields the most energy of all the oxidants available and will be the first to be consumed. Anaerobic respiration (denitrification, manganese and iron reduction, and sulfate reduction) yields less energy (Froelich et al., 1979; Strumm and Morgan, 1996). The snails were the dominant source of carbon in all of the tubes. The haloes surrounding the buried snails appeared after the tubes went anoxic indicating anaerobic respiration occurred. The sediments limited the availability of oxidants to the biofilms and may have limited access to the carbon source, resulting in the small radius of the haloes around the snails.

5.4.2 – Exoskeletons

The morphologies and distribution of the biofilms on the FS snail shells compared to those on the SC snails suggests that the original community changed during the transition from a freshwater to saltwater environment; the SC snails have up to 10% more bacillus microbes than the FS snails and have more filamentous microbes than coccoidal forms. Changes in salinity are known to have a great impact on freshwater microbial biofilms; certain microbes within a biofilm are not tolerant to changes in salinity, while others will be less affected, changing the diversity of the biofilm community (Hart et al., 1995; Nielson et al., 2003; Zhang et al, 2014). This would explain the changes in diversity between the FS and experimental snails.

The presence of the sediment in the BS snails greatly slowed biofilm colonization on the shells. After one week biofilms colonized, on average, close to 40 percent of the SC snail shell whereas the BS snails only had an average of 5 percent biofilm coverage. It also appears that after widespread biofilm formation on the SC snails after one week the biofilm
did not greatly change in growth or diversity over time. The differences between the biofilm community in the SC and BS snails, particularly the concentrations of coccoidal versus filamentous forms suggest that the sediment affected the microbial diversity of the biofilms.

Diffusion rates of ions in sediments are much lower than diffusion rates in solutions (Manheim, 1970). The sieved sediments consisted of poorly-sorted particles ranging from very fine sand to clay size, resulting in a relatively low permeability compared to a well-sorted sand or silt. Low permeability would limit chemical diffusion of oxidants, slowing biofilm growth. In all of the SC and BS tubes the microbial biofilms consumed all of the available oxygen, rendering the tubes anoxic. In the BS tubes the pore waters likely became anoxic prior to the water above the sediments; the closed system and low diffusion rates would have limited the available oxygen in the pore waters, eliminating aerobic microbes. Anaerobic respiration would have been the dominant source of energy for the biofilms for the duration of the experiment, which does not yield nearly the same amount of energy as aerobic respiration. This would have slowed biofilm development on the snail shells.

The sediments may have provided additional surfaces for biofilm colonization; it is possible the biofilms formed close to the sediment-water interface. Biofilms that formed at this interface would have consumed all of the oxygen in the water and formed an additional layer further limiting diffusion into the sediments (Stewart, 2003). We did not analyze the sediments for biofilms but it should be considered in future experiments.

In the SC tubes the snails were not isolated, and aerobic microbes likely formed the majority of the biofilms on the snail shell prior to the tubes becoming anoxic. The biofilm morphology on the SC snails during aerobic conditions was not documented. These biofilms would have formed and reconfigured during the transition from aerobic to anaerobic
conditions. This could explain the relatively even distribution of microbes in biofilms observed in the SC snails throughout the experiment. Additionally, initially aerobic microbes forming the biofilms in the SC tubes and anaerobic microbes dominating the biofilms in the BS tubes could account for the delayed biofilm formation on the BS snail shells and different morphologies of the biofilms between the SC and BS snails (Tables 5.2 and 5.3).

Phylogenetic analyses of the biofilms are needed to identify different microbial populations and, thereby, infer whether aerobic or anaerobic respiration dominated in each tube. Regardless, the sediments clearly slowed biofilm growth and affected the microbial diversity.

5.4.2 – Soft Tissues

All the SC and BS snail bodies showed signs of tissue dehydration compared to the freshwater starts. This occurred within the first week, driven by the change in salinity. When freshwater organisms are introduced into saline environments their tissues become dehydrated by the uptake of salt and the loss of water (Hart et al., 1991); when this osmotic imbalance occurs the tissue cells will cease proper function, killing the organism. The snails used in this experiment were chosen because they were readily available at the University of Montana and were used to standardize protocols. Marine snails would not have dehydrated and would not have died of salinity shock as the freshwater snails did. Nevertheless, the SC snails allowed us to document the effects of sediments on biofilm formation in comparison to the absence of sediments.

Biofilms observed on the soft tissues of SC and BC snails were formed de novo as no biofilms were detected on the soft tissues of the FS snails from time zero. Biofilms,
dominated by microbes of bacillus form, were identified on the soft tissues of all of the SC snails and some of the week 1 BS snails. The biofilms on the SC snails had a patchy distribution all over the snail’s body whereas the BS snails had very patchy biofilms located only on the anterior portion of the body, close to the opening of the shell. The biofilms were detected only on week 1 BS snails lacking sediment plugs. Further, neither the week 2 nor week 8 BS snails showed any evidence of biofilms on their soft tissues.

The soft tissues of the snails would have provided a carbon source for the microbes and were not limiting in either the SC and BS snails. The biofilms on the week 1 BS snail soft tissues were found only on the anterior portion of the body, near the shell’s opening, where they would have had access to the low amounts of oxidants available by diffusion. Like the biofilms on the BS snail shells, anoxic environments likely slowed biofilm growth on the soft tissues. The lack of any detectable biofilms on the week 2 and week 8 BS snails suggests the sediments created an environment unable to support long-term biofilm development.

The development of sediment plugs in the opening of some of the BS snail shells appear to have delayed degradation of the soft tissues, retaining more of the original shape of the snail mantle compared to the BS snails without sediment plugs. The plugs isolated the tissues from the sediments, creating a microcosm inside of the shell. Although the plugs seem to extend the amount of time the shape of the snail body is distinguishable, our experiments demonstrate the presence of sediments has an overall have a negative effect on the long-term preservation of the snail tissues due to the lack of biofilm formation, which has proven to aid in fossilization (e.g., Briggs, 2003). The appearance of the soft tissues of in the SC snails throughout the experiment do not change, suggesting the biofilms played a role in
the preservation of their form. This is consistent with what has been observed in other
taphonomical studies (e.g., Raff et al., 2014).

In all BS snails, the shells, both with and without sediments plugs, isolated the soft
tissues, inhibited biofilm formation and actually prevented preservation of the tissues. This
study leaves unanswered questions about the preservation of soft tissues of organisms
without exoskeletons (e.g., worms, anemones, etc.) under the experimental conditions.

5.5 – Conclusions

Our experiment demonstrated that sediments affect biofilm formation on the shells
and soft tissues of buried snails. Biofilms formed and developed more slowly on snails that
were buried in fine-grained sediments compared to those not buried. Previous taphonomy
experiments, lacking sediments, demonstrated biofilms formed and enveloped deceased
organisms within 24-48 hours and that, under the right circumstances, formation of mineral
films on the tissues rapidly followed (e.g., Raff et al., 2008). Our SC samples showed rapid
biofilm formation on both the exoskeletons and soft tissues, similar to results in other
taphonomy experiments. After 8 weeks the soft tissues of the SC snails retained their
original form, indicating a well-nourished biofilm is important in preservation. Sediment
plugs helped isolate the soft tissues, and extended the period in which the original shape of
the body was preserved, but ultimately the tissues disintegrated. The delay in biofilm
formation in the BS snails is interpreted as slowed diffusion of oxidants and caused the soft
tissues to disintegrate, preventing preservation.

Based on the results presented here, if soft tissues disintegrate in the absence of
biofilms and biofilms form slowly in fine-grained sediments, then biofilms cannot be the sole
mechanism responsible for exceptional fossilization. This must be taken into account when considering the taphonomy of obrution Lagerstätten. The grain size and sorting of the sediments encasing the fossils in various obrution Lagerstätten may play an important role in fossilization; sediments with a higher permeability (better sorting, larger grain size) would provide more oxidants and cations to feed biofilms and produce mineral films and cements, which could better the chances of early mineralization of tissues. If the permeability of sediments in these Lagerstätten is the same or less than that of sediments used in our experiment then biofilm growth could have been inhibited and other, yet unidentified mechanisms are responsible for fossilization.

5.6 – References


HARRISON, J.J., TURNER, R.J., MARQUES, L.L.R., and CERI, H., 2005, Biofilms: A new understanding of these microbial communities is driving a revolution that may transform the science of microbiology: American Scientist, v. 93, p. 508-515.


CHAPTER 6 – SUMMARY

Overall the individual chapters from this dissertation document processes that resulted in the deposition and lithification of the Maotianshan Shale. From these a taphonomical model revealing the sedimentological and geochemical conditions that preserved the Chengjiang Biota was created (Figure 6.1). The combination of this information allowed me to place the different processes into a clear taphonomical sequence of events that preserved the fossils. The approach I used, combining information from multiple studies in different fields, has not been used by other researchers. For example, Gabbott et al. (2004) and Zhu et al. (2005) focused on the geochemistry of the sediments and fossils but did not take into account the overall sedimentology and different fossil assemblages in the basin. Zhao et al. (2009; 2012) examined the taphofacies and paleocommunities in several fossil-rich localities but did not examine the chemical environment, nor did they look for correlations across the basin. In my dissertation I combined information from sedimentological and geochemical analyses with results from taphonomical experiments to get an overall understanding of the different processes responsible for the deposition of the Maotianshan Shale and the preservation of the Chengjiang Biota.

The depositional facies of the Maotianshan Shale and the stratigraphic controls for the preservation of the Chengjiang Biota (chapter 2) constrained the depositional environment of the Maotianshan Shale to between the lower shoreface and the offshore transition zone. Five distinct depositional units were identified in the Maotianshan Shale; each was composed of a progradational and retrogradational packages of sediments separated by flooding surfaces. These newly described facies and depositional units provided a more complete understanding
of the sedimentological changes that occurred in the across the study area and allowed for correlations between different fossiliferous localities. The sediments were deposited in a time-transgressive pattern across the study area from west to east, corresponding to the three main fossil assemblages identified in the basin. The exceptionally preserved fossil-yielding event mudstones are typically found near the top of the sedimentary succession at each locality within a prograding package of sediments. This stratigraphic position of the fossiliferous event beds is similar to what has been observed in some other BST deposits (e.g., The Wheeler Shale (Halgedahl et al., 2009)) and is likely a taphonomic control for the preservation of these exceptional fossils. Sedimentological and stratigraphic studies from many other BST sites are needed to determine if this is a shared taphonomic trait between these localities.

Previous interpretations of the depositional environment of the Maotianshan Shale include an epieric sea (Chen and Lindström, 1991), a shallow shelf to coastal marine environment (Babcock and Zhang, 2001; Babcock et al., 2001) and turbiditic sedimentation ranging 10’s to 100’s of meters deep (Hou et al., 2007); most of these interpretations focused on the mudstones preserving the fossils and not the overall, detailed sedimentology including all the facies within the Maotianshan Shale. The sedimentological analyses in this dissertation distinguished the depositional environment of the Maotianshan Shale to be much shallower than many other BSTs, many of which are interpreted as deep water deposits (e.g., Burgess Shale (Hagadorn, 2002), Kinzers Formation (Skinner, 2005)). The differences in depositional environment between the Maotianshan Shale and other BST sites suggest that a specific depositional environment is not the driving factor behind BST fossil preservation. That being said, it is likely that the specific depositional processes of the exceptional fossil-
yielding mudstones are similar in all BST deposits. A detailed analysis of the fine-grained, event mudstones in the Maotianshan Shale is needed. This will clarify the specific depositional processes responsible for this facies and will explain how the Chengjiang fossils were encased within the fine-grained sediments with little damage to their delicate tissues. Once this is understood comparative analyses between the event mudstones of the Maotianshan Shale and those from other BST deposits can be conducted to determine if they do indeed share a common mode of deposition.

The mineralogy and geochemistry of the Maotianshan Shale (chapter 3) provides insight into the provenance of the sediments as well as the conditions within the sediments prior to lithification and changes that occurred during diagenesis. The sediments were derived from felsic granitic and volcanic sources. These are compositionally similar to other BST deposits (e.g., Sirius Passett (Le Boudec et al., 2014)), suggesting the composition of the sediments played a role in the taphonomy of these exceptionally preserved fossils. Differences in sediment provenance between BST deposits may explain, at least in part, differences in the preservation style and composition of the biotas identified at each of the localities. Although the majority of the soft tissues in BST deposits are preserved as carbonaceous films there are other minerals found preserving soft tissue present in several BST deposits (e.g., calcium carbonate in the Emu bay Shale (Briggs and Nedin; 1997); iron oxide in the Pioche Shale (Moore and Lieberman, 2009)), which may have resulted from different sediment sources providing the ions needed for these minerals. Further examination of the composition and provenance of other BST deposits is necessary to better understand how the composition of the sediments aids in preserving the fossils.
Low TOC concentrations were measured in the Maotianshan Shale; these likely reflect an originally low concentration in organic matter as the underlying black shale member records much higher TOC concentrations. The presence of illite and chlorite in the rocks indicates that the sediments might have undergone some heating but not enough to completely remove all the organic carbon from the system.

The presence of molybdenum indicates anoxic conditions were present in the sediments at time of deposition. Pyrite, either primary or diagenetic, supports the interpretation that the conditions in the sediments were anoxic; this does not require the surface of the sediments, nor the water column to be anoxic. These conditions likely aided in the fossilization of the Chengjiang biota. Geochemical analyses to better understand the chemical conditions required for soft tissue preservation have only been conducted in detail on a few BST deposits (e.g., Sirius Passett (Le Boudec et al., 2014), Emu Bay Shale (McKirdy et al., 2011)). Further studies of the detailed geochemistry of different BST Lagerstätten will help highlight the similarities and differences in the chemical environments, and will elucidate the common factors for these fossils throughout the Cambrian and early Paleozoic.

Surface analyses of the tissues of fossilized worms demonstrated elevated trace element concentrations on the soft tissues compared to the matrix (chapter 4). Statistically significant elevated concentrations of U, P, Sc, V, Cr Fe, Ni, Cu, Mo, Sb and Th were identified in the tissues of all of the fossils. These elevated concentrations may have occurred via 1) active or passive uptake on biofilms prior to fossilization and/or 2) accumulation on the tissues after fossilization. Additional analyses identified similar elemental increases in Liesegang bands that cut across the sediments and fossils. This
suggests abiotic accumulation of these elements occurred during diagenesis, but further analyses need to be done to confirm this. Local redox or geochemical conditions in the sediments after initial fossilization were considered as the cause for the trace element uptake but ultimately ruled out because the trace elements with elevated concentrations have a wide range of geochemical behaviors such as immobile versus mobile elements, redox sensitive versus insensitive, etc. It is unclear whether biofilms formed on the tissues and if they aided, at least partly, in the uptake of the trace elements. This leaves the questions as to how and when the organic films preserving the tissues formed. Since the elevated elemental concentrations are found in both the fossilized tissues and the Liesegang bands, at least some trace element accumulations likely occurred during weathering. Fossils found in weathered (tan to yellow color) samples were used in the fossil-surface study because the organisms are more clearly visible in these samples and they were available at the time. To better understand the timing of trace element accumulation, similar analyses should be conducted on soft tissues of fossils from fresh, unweathered (grey to black color) samples.

Experiments designed to test the effects of biofilm formation on organisms buried in fine-grained sediments demonstrated that sediments delayed and, in some cases, prevented biofilm formation (chapter 5). The biofilms on the control snails, those without sediments, formed rapidly covering both the exoskeletons and the soft tissues and had different morphologies than those that were buried, suggesting different processes occurred between the control and buried snails. The buried snails had biofilms with patchy distributions on their shells and no long-term biofilm development on the soft tissues. The delay in biofilm formation in the buried snails caused disarticulation of the soft tissues, indicating the experimental conditions would not have promoted soft-tissue preservation. The sediments
likely slowed the diffusion of oxidants, preventing widespread biofilm formation in the buried snails. These results were unexpected and contradict our interpretation based on the results in chapter 4, that biofilms associated with organisms buried in fine-grained sediments promote accumulation of trace elements on their surfaces. If the sediment type and size used in these experiments are comparable to those in BST deposits it is unclear if biofilms ever formed on the fossils and, if they did, the role they played the preservation of the organisms found in these Lagerstätten. Gaines et al (2012) cite rapid entombment of the organisms in fine-grained sediments capped by carbonate cement as cause for the preservation of organic films in BST deposits. In their model, the fine-grained sediments and carbonate cement would restrict oxidants and slow microbial decay of the tissues. My taphonomy experiment shows that even without a carbonate cement sealing the top of the sediments oxidant discussion is greatly slowed in fine-grained sediments, but it also demonstrated that this alone was not enough to prevent tissue decay and preserve the films. It is unlikely the mechanism proposed by Gaines et al (2012) would have been sufficient to preserve the detailed carbonaceous films preserving the fossils. If that is the case there must have been other factors, such as fluid transport during weathering, responsible for preserving the organic carbon films and the fossils in such detail.

The use of snails in this experiment may not have been the best analog to use for the organisms found in BST deposits. Snails have a hard exoskeleton composed of calcium carbonate into which they can retract their bodies for protection. The majority of fossils in the Chengjiang Biota are composed of soft tissues, those with hard parts had exoskeletons were composed primarily of chitin. Additionally, the fossils of the Chengjiang biota were killed instantly; many are sill found in life position, showing no evidence of struggle (Chen
2004; Chen, 2009). During the taphonomy experiments, the snails retracted into their shells, which added another protective layer for the tissues, limiting access by microbial biofilms, preventing tissue preservation. If the taphonomy experiment was conducted on organisms with chitinous exoskeletons (e.g., shrimp, velvet worms) and on organisms composed solely of soft tissues (e.g., anemones) biofilms may have formed on the exposed hard and soft tissues, preserving them. Further experiments are needed to confirm this.

A taphonomical model for the preservation of the organisms of the Chengjiang biota was created from the results of this dissertation. Sedimentological analyses indicate the Chengjiang Biota were living on the lower shoreface or prodelta, depending on the locality (Figure 6.1a). The organisms were rapidly killed and buried by event muds, highly concentrated, fine-grained sediment plumes, during times of shoreline progradation. The fine-grained sediments would have suffocated the Chengjiang Biota. The majority of the fossils were buried in situ, but some were transported downslope to the offshore transition zone (Figure 6.1.b).

The sediments became suboxic to anoxic through microbial degradation of organic matter, with anoxic/sulfidic zones forming rapidly around the dead organisms. Biofilms may have formed on the tissues within 12-48 hours of burial, although my data show that sediments delay this process. Carbon films, which preserved the fossils, likely formed within a month after burial to prevent decay and disarticulation of the tissues. Trace metals may have begun accumulating on the carbon films at this time (Figure 6.1c). From the early Cambrian to the middle Paleozoic sediments continued to accumulate in the basin under oxic conditions (Tan et al., 2015). During this time the sediments would have compacted and begun to lithify (up to 90% porosity loss). Pore fluids may have deposited trace metals,
Figure 6.1 - A chronological model of the taphonomy of the Chengjiang Biota. (a) The Chengjiang Biota was living on the lower shoreface to offshore transition zone under normal marine conditions. (b) Death and burial of the organisms. (c) 1 day to 1 month after burial (*based on taphonomy experiments such as Raff et al. (2008)). (d) Early to Middle Paleozoic (** based on Tan et al., 2015). (e) Middle Paleozoic to Middle Mesozoic (+ based on Tan et al. (2013) and Wang et al (2013)). (f) Middle Mesozoic to Present (++ based on Tan et al. (2015)). The Chapters in the dissertations that cover information for each taphonomical step are noted on the left.
which accumulated on the carbon films associated with the fossils (Figure 6.1 d). Between the middle Paleozoic and the middle Mesozoic additional sediments accumulated (Tan et al., 2013; Wang et al., 2013), further burying the Maotianshan Shale. During this time minor heating of the sediments occurred, altering some of the primary minerals (e.g., kaolinite, k-feldspars) to illite and chlorite. This heating may have caused catagenesis, reducing the amount of organic carbon in the sediments.

Trace metal accumulations on the carbon films could have occurred during this time interval (Figure 6.1e). From the middle Mesozoic to the Present uplift began on the Yangtze platform, eventually exposing the Maotianshan Shale (Tan et al., 2015). During this time the sediments underwent weathering, changing from grey-black in color to yellow tan. Minerals pyrite and dolomite were lost during weathering; trace elements may also have accumulated on the fossils during this time (Figure 6.1f).

Generalized taphonomical models for BST preservation have been previously proposed (e.g., Gaines et al., 2008; Gaines et al., 2012). These models include evidence from a multitude of BST deposits and look at commonalities between the deposits but overlook the individual deposits comparing their similarities and differences (e.g., water depth, fossil assemblage). The results from this dissertation do not differ from the overall model for BST preservation, but they highlight the site-specific details within the Maotianshan Shale/Chengjiang Biota. Although the overall BST taphonomical model is useful, these Lagerstätten show variations in depositional environment, sediment composition, geologic age, faunal assemblages, and modes of fossilization, which should be considered when discussing the taphonomy of each locality. This will provide a more complete history of various depositional environments and processes that formed the many BST deposits. This
will clarify the specific taphonomical pathways responsible for the preservation of these exceptional fossils.

6.1 – References


APPENDIX I – REDOX REACTIONS AND PRECIPITATION/DISSOLUTION OF MINERAL PHASES IN EARLY CAMBRIAN SEAWATER CALCULATED USING PHREEQC

A geochemical model of early Cambrian seawater was created using the computer program PHREEQC (Glasby and Schulz, 1999) to determine the redox reactions as well as the precipitation and dissolution (as saturation indices (SI)) of various mineral phases with changing pe values. The major element concentrations for Cambrian seawater were taken from Brennan et al. (2004) who analyzed fluid inclusions in marine halites. Concentrations for Na\(^+\), Cl\(^-\) and alkalinity (as HCO\(_3^-\)) were calculated using the charge balance, assuming the [Na\(^+\)],[Cl\(^-\)] ratio is the same as in modern oceans, using the following equations:

\[
[\text{Na}^+] + [\text{K}^+] + 2[\text{Ca}^{2+}] + 2[\text{Mg}^{2+}] = [\text{Cl}^-] + 2[\text{SO}_4^{2-}] + [\text{HCO}_3^-]
\]
\[
538.45 + 9 + 2(37) + 2(44) = 605 + 2(8) + \text{[HCO}_3^-]
\]
\[
700.45 = 621 + \text{[HCO}_3^-]
\]
\[
[\text{HCO}_3^-] = 79.45
\]

The PHREEQC input file follows below. Trace elements present in the Maotianshan Shale sediments were included. The pe values were lowered from 13.0 to -6.0 by one unit per step and the redox reactions and mineral phases present at each pe value were noted.
PHREEQC input file:

TITLE Example 1.-- Minerals in Early Cambrian seawater.  
SOLUTION 1 -- Determine the minerals that will precipitate out of solution with changing pe values  
units mmol/kgw  
pH 8  
pe 13.0  
temp 25.0  
Ca 37  
Mg 44  
Na 538.35  
K 9  
Cl 605  
Alkalinity 79.45 as HCO3  
S 8 pe  
Fe 5.37e-7 pe  
Mn 3.64e-7 pe  
Cu 2.36e-6 pe  
Ni 8.18e-6 pe  
V 3.93e-5 pe  
Co 2.36e-8 pe  
Cr 4.04e-6 pe  
U 1.34e-5 pe  
Mo 1.04e-4 pe  
Save solution 1  
END

The results from this model are listed below in a table showing the redox reactions and saturation indices obtained from the PHREEQC output files at differing Eh values (calculated from pe).
<table>
<thead>
<tr>
<th>Eh (mV)</th>
<th>Value taken from Langmuir (1997)</th>
<th>Redox reactions from PHREEQC</th>
<th>Data from PHREEQC</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>O$_\text{2-}$ + 4H$^+$ + 4e$^-$ = 2H$_2$O (pH 7.5)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>500</td>
<td>NO$_3^-$ + 6H$^+$ + 5e$^-$ = 0.5N$_2$ + 3H$_2$O (pH 6.42)</td>
<td>MnO$_2$ + 4H$^+$ + 2e$^-$ = Mn(II) + 2H$_2$O (pH 5.90)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>MnO$_{2-3}$ + 4H$^+$ + 2e$^-$ = Mn(II) + 2H$_2$O (pH 4.26)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>NO$_2^-$ + 2H$^+$ + 2e$^-$ = NO$_3^-$ + H$_2$O (pH 3.71)</td>
<td>CrO$_2^{2-}$ + 6H$^+$ + 3e$^-$ = Cr(OH)$_3^{3-}$ + 2H$_2$O (pH 3.54)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>NO$_3^-$ + 8H$^+$ + 6e$^-$ = NH$_4^+$ + H$_2$O (pH 2.61)</td>
<td>Cu(II) + e$^-$ = Cu(I) (pH 2.36)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Fe(OH)$_3^{+3}$ + 3H$^+$ + e$^-$ = Fe(II) + 3H$_2$O (pH -1.64)</td>
<td>Fe$^{2+}$ + 6H$^+$ + 2e$^-$ = 2Fe$^{3+}$ + 3H$_2$O (pH 0)</td>
<td></td>
</tr>
<tr>
<td>250</td>
<td>2SO$_4^{2-}$ + Fe$^{3+}$ + 16H$^+$ + 14e$^-$ = FeS$_2$ + 8H$_2$O (pH -2.24)</td>
<td>U(VI) + 2H$_2$O = UO$_2^-$ + 4H$^+$ (pH -2.36)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>SO$_4^{2-}$ + 10H$^+$ + 8e$^-$ = H$_2$O + 4H$_2$O (pH -2.91)</td>
<td>MoO$_4^{2-}$ + 6H$^+$ + 2e$^-$ = MoS$_2$ + 4H$_2$O (pH -2.36)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>HVO$_4^{2-}$ + H$^+$ + 2e$^-$ = V(III) (pH -2.95)</td>
<td>Cu$^{2+}$ + Fe$^{3+}$ + 2HS = CuFeS$_2$ + 2H$^+$ (pH -2.95)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>2Fe$^{2+}$ + HS = Fe$S_2$ + H$^+$ (pH -2.95)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Fe$^{2+}$ + HS = FeS$_2$ + 2H$^+$ + 2e$^-$ (pH -2.95)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>V$^{5+}$ + 1.5H$_2$O = V$_2$O$_5$ + 3H$^+$ (pH -3.54)</td>
<td></td>
</tr>
</tbody>
</table>
APPENDIX II – GEOCHEMICAL DATA
(in attached CD)
APPENDIX III – REVIEW OF KONSERVAT-LAGERSTÄTTEN

THROUGHOUT GEOLOGIC HISTORY

An extensive and thorough literature review was conducted to characterize Konservat-Lagerstätten deposits through geologic time. This was done in an effort to better understand the controls responsible for the preservation of the biotas present within these deposits. A total of 146 Lagerstätten were identified, of which 63 had sufficient published data for this investigation. They were categorized by their geologic age, geographic location, whether or not soft tissues were preserved, the mineralizing agents of the tissues, the sediment type in which the fossils were found, whether the sediments were laminated, if the fossils were found in concretions, whether the Lagerstätten was an obrution or stagnation deposit (or both), whether it was shallow or deep, if microbes (biofilms or microbial mats) were identified as aiding in tissue preservation, and whether the sediments were anoxic; these data are listed in the data matrix. The other 82 identified Lagerstätten are still under investigation but their name, age and geographic location are listed in Table 2.

The data from the data matrix was put into a data matrix and run in SPSS for frequency information as well as hierarchical cluster analysis. These results are listed below.
### Frequency Data:
Distribution of Lagerstätten based on geologic age

<table>
<thead>
<tr>
<th>Geologic Age</th>
<th>Freq.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Precambrian</td>
<td>3</td>
</tr>
<tr>
<td>Cambrian</td>
<td>14</td>
</tr>
<tr>
<td>Ordovician</td>
<td>11</td>
</tr>
<tr>
<td>Silurian</td>
<td>2</td>
</tr>
<tr>
<td>Devonian</td>
<td>4</td>
</tr>
<tr>
<td>Carboniferous</td>
<td>3</td>
</tr>
<tr>
<td>Permian</td>
<td>1</td>
</tr>
<tr>
<td>Triassic</td>
<td>2</td>
</tr>
<tr>
<td>Jurassic</td>
<td>6</td>
</tr>
<tr>
<td>Cretaceous</td>
<td>6</td>
</tr>
<tr>
<td>Paleocene</td>
<td>0</td>
</tr>
<tr>
<td>Eocene</td>
<td>6</td>
</tr>
<tr>
<td>Oligocene</td>
<td>0</td>
</tr>
<tr>
<td>Miocene</td>
<td>4</td>
</tr>
<tr>
<td>Pliocene</td>
<td>0</td>
</tr>
<tr>
<td>Pleistocene</td>
<td>1</td>
</tr>
</tbody>
</table>
### Distribution of Lagerstätten based on geographic location

<table>
<thead>
<tr>
<th>Location</th>
<th>Freq.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Africa</td>
<td>3</td>
</tr>
<tr>
<td>Antarctica</td>
<td>1</td>
</tr>
<tr>
<td>Asia</td>
<td>5</td>
</tr>
<tr>
<td>Australia</td>
<td>4</td>
</tr>
<tr>
<td>Europe</td>
<td>17</td>
</tr>
<tr>
<td>North America</td>
<td>30</td>
</tr>
<tr>
<td>South America</td>
<td>3</td>
</tr>
</tbody>
</table>

### Distribution of Lagerstätten based on depositional environment

<table>
<thead>
<tr>
<th>Environment</th>
<th>Count</th>
</tr>
</thead>
<tbody>
<tr>
<td>Deep</td>
<td>37</td>
</tr>
<tr>
<td>Shallow</td>
<td>26</td>
</tr>
</tbody>
</table>

---

[Distribution of Lagerstätten based on geographic location diagram]

[Distribution of Lagerstätten based on depositional environment diagram]
Lagerstätten with soft tissues preserved

- Soft tissues preserved: 59
- Only hard parts: 4

Lagerstätten with laminated sediments

- Laminated sediments: 59
- Massive sediments: 4

- 94% Soft tissues preserved
- 6% Only hard parts
- 59 Laminated
- 4 not laminated
Fossils with evidence of biofilms and/or microbial mats preserving the tissues

<table>
<thead>
<tr>
<th>Present</th>
<th>Absent</th>
</tr>
</thead>
<tbody>
<tr>
<td>27</td>
<td>36</td>
</tr>
</tbody>
</table>

Lagerstätten preserved under anoxic conditions

<table>
<thead>
<tr>
<th>Anoxic</th>
<th>Oxic</th>
</tr>
</thead>
<tbody>
<tr>
<td>61</td>
<td>2</td>
</tr>
</tbody>
</table>
### Type of Lagerstätten

<table>
<thead>
<tr>
<th>Type</th>
<th>Count</th>
</tr>
</thead>
<tbody>
<tr>
<td>Obrution</td>
<td>39</td>
</tr>
<tr>
<td>Stagnation</td>
<td>20</td>
</tr>
<tr>
<td>Both</td>
<td>4</td>
</tr>
</tbody>
</table>

![Pie Chart](chart.png)
### Distribution of mineralizing agents of the preserved tissues

<table>
<thead>
<tr>
<th>Mineralizing Agent</th>
<th>Freq</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cast/mold</td>
<td>9</td>
</tr>
<tr>
<td>pyrite</td>
<td>31</td>
</tr>
<tr>
<td>calcite</td>
<td>20</td>
</tr>
<tr>
<td>Dolomite</td>
<td>2</td>
</tr>
<tr>
<td>phosphate</td>
<td>40</td>
</tr>
<tr>
<td>Carbon film</td>
<td>39</td>
</tr>
<tr>
<td>Clay minerals</td>
<td>10</td>
</tr>
<tr>
<td>Silica</td>
<td>13</td>
</tr>
<tr>
<td>Iron oxide</td>
<td>6</td>
</tr>
<tr>
<td>siderite</td>
<td>5</td>
</tr>
<tr>
<td>amber</td>
<td>2</td>
</tr>
<tr>
<td>carbonate</td>
<td>1</td>
</tr>
<tr>
<td>Evaporite</td>
<td>1</td>
</tr>
<tr>
<td>Trace metals</td>
<td>3</td>
</tr>
</tbody>
</table>
References


revealed via synchrotron imaging. Proceedings of the National Academy of Science 107, 9060-9065.


Middle Cambrian faunal complex. Science 222, 163-167.


Gabbott, S.E., 1998. Taphonomy of the Ordovician Soom Shale Lagerstatte: An example
of soft tissue preservation in clay minerals. Palaeontology 41, 631-667.


Acta Palaeontologica Sinica 45, 460-472.


