Biologic
ENVIRONMENTAL SURVEY

McPhee Creek Short-range Endemic Survey

Molecular Systematics Analysis

Report to Atlas Iron Pty Ltd

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Glossary

12S Mitochondrially encoded 12S ribosomal RNA, a component of the small subunit of the mitochondrial ribosome, which is useful in phylogenetic studies.

Bootstrap Value between 0 and 100 that indicates the robustness of the node in a phylogenetic tree.

COI Cytochrome Oxidase subunit 1, a mitochondrial gene commonly used in phylogenetic studies and used as a DNA barcode to identify species.

GenBank Annotated open access sequence database of all publicly available nucleotide sequences and their protein translations.

Monophyletic A grouping of specimens that all share a common ancestor, inferred by sequence data. The sequences within the monophyletic group will all be more closely related to each other, relative to sequences outside of the monophyletic group. This grouping is foten referred to as a lineage or clade, and are graphically represented in phylogenies/trees by sharing a single node with a high bootstrap value.

OTU Operational taxonomic unit – species-equivalent taxonomic unit based on COI or 12S cluster similarity.

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1 Introduction

Atlas Iron Pty Ltd (Atlas Iron) commissioned Biologic Environmental Survey (Biologic) to undertake a molecular systematics analysis (DNA barcoding) of 30 specimens collected from McPhee Creek (the Study Area).

1.1 Aims and objectives

The aims and objectives of the molecular systematics analysis were to:

- Undertake DNA sequencing of 30 short-range endemic invertebrate specimens to obtain barcoding sequences of the mitochondrial gene Cytochrome Oxidase subunit 1 (COI; Hebert *et al.*, 2003b);
- Investigate the interspecific and intraspecific relationships between sequences of each higher taxonomic group (*i.e.* use the results of the DNA analysis to indicate how many different species/ OTUs are likely to occur within each genus or relevant higher taxon, based on published species-thresholds); and
- Investigate the relationships between sequences from the Study Area and relevant previous sequences from the wider Pilbara region, using available DNA databases (*i.e.* compare the results of the current analysis with accessible DNA databases to assess whether any of the species/ OTUs from the Study Area have been collected previously or more widely beyond the Study Area).

This document reports the methods and results of the molecular systematics analysis. All sequence data will be uploaded to GenBank (https://www.ncbi.nlm.nih.gov/genbank/) as per Biologic Molecular Systematics standard procedure.

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2 Methods

2.1 Sub-sample Preparation

A total of 30 specimens collected from the Study Area by Biologic were selected for molecular systematics analysis (Table 2.1). The specimens were chosen based on their geographic spread across the Study Area to assist with understanding species distributions. Adequate redundancy in specimen selection was incorporated to account for any potential sequence generation failure. Specimens in good condition were chosen to increase their DNA extraction potential.

Where whole specimens were available, tissue preparation was undertaken by removing a leg or another body part less important for taxonomic identification, briefly drying off the ethanol, and placing the tissue in ATL buffer. In some instances, for very small and/or juvenile specimens, the entire animal was utilised. Again, these were briefly dried and placed in ATL buffer. Greatest care was taken to decontaminate all tools and equipment between samples, using bleach and repeated rinsing in deionised water, Table 2.1 provides details of the taxonomic orders chosen for molecular analysis. Further taxonomic clarification for each specimen included in the analysis can be found in Appendix A.

Table 2.1: Taxonomic groups from the Study Area included in the analysis, with a summary of PCR and sequencing success

Order	Fail	Pass	Total
Araneae	0	1	1
Pseudoscorpiones	3	26	29
Total	3	27	30

2.2 DNA Extraction, Amplification and Sequencing

DNA extraction and sequencing methods followed standard methods (e.g. Edgecombe *et al.*, 2019; Framenau *et al.*, 2018; Huey *et al.*, 2019; Perina *et al.*, 2018), as follows:

Subsampled tissue/specimen was placed directly into ATL buffer for extraction using the QIAGEN DNeasy Blood and Tissue extraction kit, and DNA extraction followed the manufacturer's protocols. DNA extractions were amplified by Polymerase Chain Reaction (PCR) using Folmer PCR primers (LCO1490, HCO2198; Folmer *et al.*, 1994) to assess the variability of COI. For some specimens that did not amplify using the Folmer primers, alternative primers amplifying the same part of COI were used, such as C1-J2329 and C1-J1718 (Perina *et al.*, 2018; Simon *et al.*, 1994).

The resulting PCR product was cleaned up and sequenced by the Australian Genomic Research Facility (AGRF) Perth node. Molecular laboratory workflows were managed using

GENEIOUS Prime (Kearse et al., 2012) with the Biocode plugin (http://www.mooreabiocode.org). Raw sequence data were edited and assembled in GENEIOUS, and final consensus sequences were then available for downstream analysis.

2.3 Specimen Selection for Comparative Analysis

DNA comparisons were typically conducted at the order level (Table 2.1). Comparative sequences were from GenBank (a publicly available DNA sequence database) and Biologic's unpublished DNA sequence libraries, using two separate methods.

- BLAST (Basic Local Alignment Search Tool): a method for rapidly searching a DNA sequence library to identify similar sequences. Sequences were searched using the "blastn" function, which returns similar matches.
- Taxonomic Curation: BLAST occasionally fails to identify sequences that could be considered useful for comparison, such as species that might be genetically distant, but are required to be included in the analysis for comparison. Taxonomically relevant specimens were identified using the available taxonomic classifications and identifications in those databases.

The final phylogenies and distance matrices in this report were pruned back to those sequences that can be provided to the Client, with any matches to sequences that cannot be provided to the Client discussed in the relevant sections.

2.4 Analysis and Interpretation of Alignments and Phylogenies

For each taxonomic group, the selected sequences were aligned using the MAFFT (Multiple Alignment using Fast Fourier Transform) algorithm (Katoh *et al.*, 2002). Trees were constructed on resulting alignments using the RaxML (Stamatakis, 2014) plugin in GENEIOUS Prime, using 1,000 bootstrap replicates and the GTR+G substitution model.

To delimit taxonomic units using molecular data, we integrated multiple lines of evidence, including:

- Genetic distance threshold method (~8% pairwise distances at COI, see below);
- Morphological identifications, where available;
- Geographic information; and
- Interpretation of phylogenetic topology.

Fauna-specific genetic distance thresholds for delimiting species and OTUs were used wherever possible, based on published literature and available previous reports. Where these thresholds were not available, the assessment used average divergence thresholds for related groups or higher taxa developed by broad-level studies (e.g. Hebert *et al.*, 2003a). In general, ≤8% COI divergence is seen as appropriate to determine OTUs (Hebert et al., 2003a),

however, higher or lower divergences are sometimes justified depending on the organism studied. Unless otherwise stated, we considered sequences that exhibited COI divergences ≤8% to belong to the same OTU.

The branching pattern and statistical robustness of the nodes (measured using bootstrap support) is also used to inform OTU delimitation. OTUs form monophyletic groups (or lineages), and so if an unknown sequence falls within a lineage comprised of other sequences that have already been identified as a single OTU or species, then that unknown sequence likely shares the same OTU/species as those sequences it is nested within. Additionally, distinct OTUs typically have large internode distances separating OTUs, with short internode distances within the OTU/species.

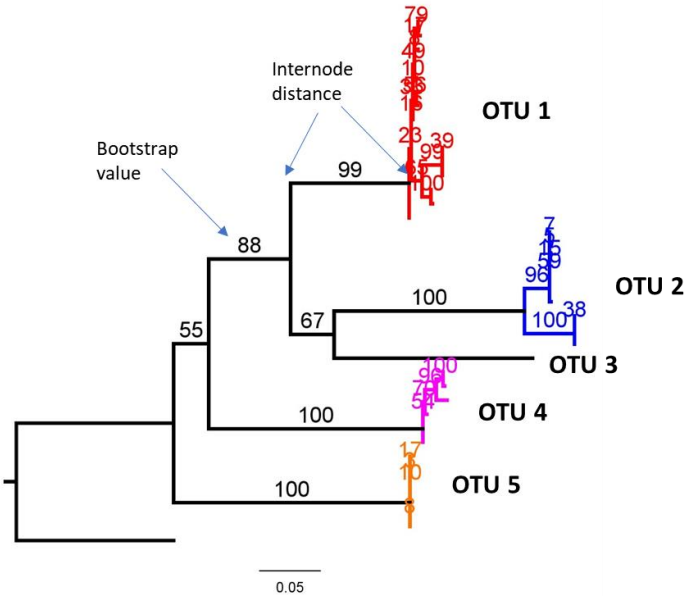


Figure 2.1. Example phylogeny showing delimited OTUs with internode distances and bootstrap values indicated

2.5 Constraints and Limitations

The analysis was constrained by the breadth of data available to undertake comparisons, the accessibility of pre-existing regional sequences, and the success rate of genetic sequencing, which can be affected by specimen collection, preservation, storage methods and contamination. Best practises were followed during specimen collection, preservation, and storage, prior to specimens arriving at Biologic's laboratories. All care was taken to ensure that the risks of laboratory contamination, data handling issues, and specimen management issues were minimised within Biologic's laboratories throughout the subsampling, processing and genetic analysis.

The databases used for regional comparisons included GenBank and Biologic's sequence libraries. While these sequence databases, in combination, comprise a large portion of the subterranean fauna genetic work undertaken in the Pilbara region, it is acknowledged that there may be many other relevant sequences from third party project areas nearby or elsewhere in the region that were not available for comparison at the time of the study. GenBank is dynamic database, and the addition of new sequences and altered taxonomic classifications could not be included into this report if they occurred after 20/1/23.

DNA barcoding using the mitochondrial gene COI, while useful for explaining genetic differences between closely related or moderately related species, is limited in its ability to resolve deeper phylogenetic relationships among taxa at higher taxonomic levels (e.g. genus, family, order). In the current study, phylogenetic relationships among species/OTUs >25% COI divergence are treated with caution. If further resolution of deeper phylogeny is important for project goals, this could be investigated using a multiple gene approach.

3 Results and Discussion

A total of 30 specimens were processed for sequencing by Biologic (Table 1). Sequences were successfully derived for 28 of these specimens (93% of specimens), with two failing to produce a PCR product. Of these 28 sequences, two did not produce a high-quality sequence (less than 80% of untrimmed bases in the sequence were of high quality) or were high quality sequences of an organism that was not the target organism (likely contamination). This left 26 high quality sequences for analysis (93% of sequences). The orders of the sequences are tabulated in Table 2.

In total, five OTUs have been designated to specimens from the Study Area, all of these being specific to this study (Table 2). The results of each taxonomic group's analysis are described in the subsequent sections.

Table 3.1: Summary of species and OTUs recovered from samples successfully sequenced in this study, organised by taxon

Original Taxon ID	OTU (genetic taxon)	Sequenced specimens	Matches to external sequences	Linear Range
Araneae				
Idiopidae?	*Conothele* `sp. Biologic-ARAN053`	1	no	singleton
Pseudoscorpiones				
Austrohorus AES03	*Austrohorus* `sp. Biologic-PSEU118`	11	no	8.7 km
Beierolpium '8/3'	*Beierolpium* `sp. Biologic-PSEU115`	3	no	4.3 km
Indolpium AES01	*Indolpium* `sp. Biologic-PSEU116`	8	no	10.0 km
Indolpium AES02 & AES03 (merge)	*Indolpium* `sp. Biologic-PSEU117`	5	no	1.2 km
Euryolpium	*Euryolpium* `sp. Biologic-PSEU064`	1	yes	>200 km
Failed to sequence				
Olpiidae Gen. nov. ??	n/a PCR Failed	1	N/A	N/A

3.1 Araneae

A single mygalomorph spider was included in the analysis. This juvenile specimen was tentatively identified as an idiopid using morphology. Analysis of the COI region placed it firmly within the genus *Conothele*, from the family Halonoproctidae (Figure 3.1). This specimen was more than 10% divergent from all other available sequences (Table 3.2). *Conothele* species are generally short ranging in distribution and commonly follow drainage

lines. However, this specimen was collected in Breakaway/Cliff habitat which is of moderate to high suitability for SRE species. It remains as a Potential SRE.

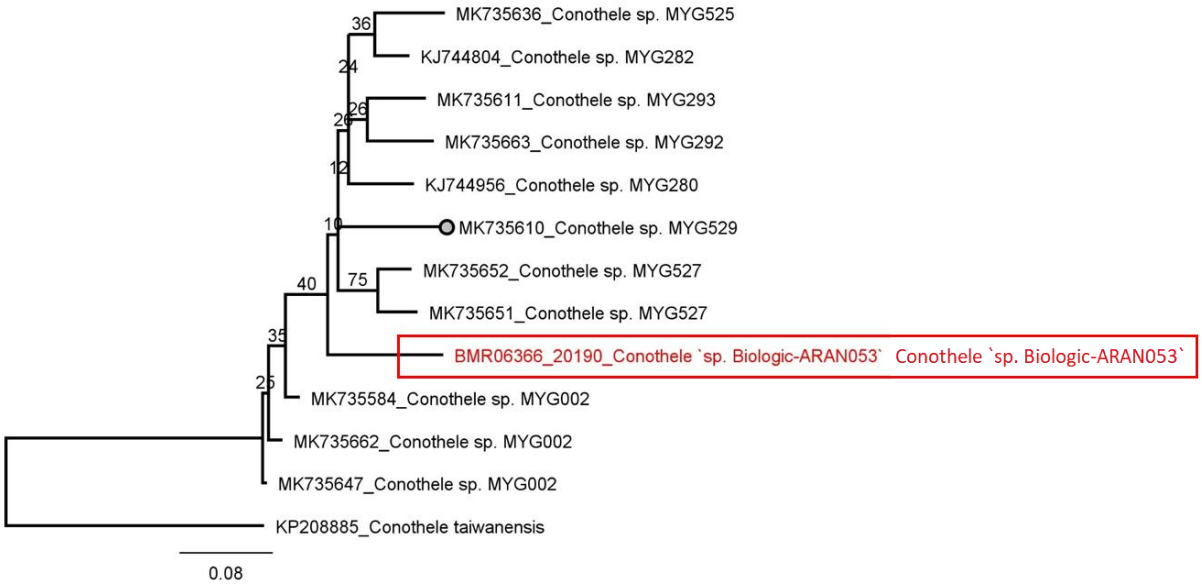


Figure 3.1: Maximum Likelihood phylogeny for Araneae, with bootstrap values.

Table 3.2: Pairwise distances (%) for the Araneae dataset.

COI Pairwise Distances (%)	BMR06366	KJ744804	MK735636	MK735663	MK735611	MK735584	MK735647	MK735662	MK735651	MK735652	KJ744956	MK735610	KP208885
BMR06366_20190_Conothele `sp. Biologic-ARAN053`		12.6	12.8	11.7	12.8	10.4	11.7	11.0	11.9	12.3	11.0	12.8	20.0
KJ744804_Conothele sp. MYG282	12.6		7.7	10.4	9.3	9.3	9.3	9.0	9.0	9.3	9.0	10.4	16.1
MK735636_Conothele sp. MYG525	12.8	7.7		8.4	9.9	11.2	11.7	11.2	9.5	10.1	9.7	11.7	17.8
MK735663_Conothele sp. MYG292	11.7	10.4	8.4		8.4	11.2	11.7	10.8	10.6	9.9	9.7	13.2	19.4
MK735611_Conothele sp. MYG293	12.8	9.3	9.9	8.4		11.0	12.1	11.5	11.0	10.1	9.5	12.1	17.6
MK735584_Conothele sp. MYG002	10.4	9.3	11.2	11.2	11.0		3.5	4.0	8.8	9.7	9.7	11.7	18.5
MK735647_Conothele sp. MYG002	11.7	9.3	11.7	11.7	12.1	3.5		2.2	8.8	9.7	11.0	12.6	18.1
MK735662_Conothele sp. MYG002	11.0	9.0	11.2	10.8	11.5	4.0	2.2		8.4	9.7	10.1	11.9	18.3
MK735651_Conothele sp. MYG527	11.9	9.0	9.5	10.6	11.0	8.8	8.8	8.4		5.9	10.1	11.2	16.7
MK735652_Conothele sp. MYG527	12.3	9.3	10.1	9.9	10.1	9.7	9.7	9.7	5.9		10.6	11.7	17.0
KJ744956_Conothele sp. MYG280	11.0	9.0	9.7	9.7	9.5	9.7	11.0	10.1	10.1	10.6		10.4	17.6
MK735610_Conothele sp. MYG529	12.8	10.4	11.7	13.2	12.1	11.7	12.6	11.9	11.2	11.7	10.4		17.2
KP208885_Conothele taiwanensis	20.0	16.1	17.8	19.4	17.6	18.5	18.1	18.3	16.7	17.0	17.6	17.2	

3.2 Pseudoscorpiones

Twenty-five successfully sequenced pseudoscorpion specimens formed four OTUs (Figure 3.2), all within the family Olpiidae. These four OTUs came from three genera, and these generic identifications broadly conformed with available genetic data. The OTUs all had intraspecific genetic variation less than 8.8% and were more than 13% divergent from all other sequences in the analysis (Table 3.3).

Austrohorus `sp. Biologic-PSEU118` had intraspecific divergences of over 8%, driven by two lineages within the OTU. However, most pairwise comparisons between these lineages were below 8%. The two lineages are found at either end of the Study Area, and the divergence likely represents geographic structuring. It is likely sequencing of specimens between these locations would collapse the two lineages, leaving a clearly supported OTU with high intraspecific genetic variation. Eighteen specimens were collected and identified as *Austrohorus* AES03, 11 were sequenced (with one failing to yield a viable sequence) to indicate one morphospecies with a wide distribution through the Study Area (Figure 3.3). However, as it did not match any regional sequences it is still considered a Potential SRE.

Three specimens of *Beierolpium* '8/3` were collected and all three were sequenced. These yielded three viable sequences and were given the OTU *Beierolpium* `sp. Biologic-PSEU115`. While the morphological identification of this species at the time was considered a widespread group, sequencing work has shown there to be a lot of cryptic diversity in this family. It is considered a Potential SRE.

Thirty-five specimens of *Indolpium* were collected from the Study Area morphological identified as representing three morphospecies. Thirteen specimens were sequenced (with one failing to yield a viable sequence) and yielded two OTUs. One of the morphospecies was a small juvenile (*Indolpium* AES02) and it is not surprising that it happens to represent one of two more abundant OTUs (*Indolpium* AES03) in the Study Area. Both OTUs were well represented in the Study Area but as they did not match any regional sequences are still considered Potential SRE.

Euryolpium sp. indet. was sequenced as *Euryolpium* `sp. Biologic-PSEU064`which has been previously collected from the Newman area. This makes it a widespread species with a range of greater than 200 km. Unfortunately, the single female specimen of Olpiidae gen nov, possibly representing a new genus for the Pilbara did not yield a viable sequence. It was collected from Breakaway/Cliff habitat and remains a Potential SRE.

There is currently no taxonomic framework beyond genus level for Olpiidae and advice is that the likelihood for short-range endemism is low, partly due to an expectation of very high diversity (M. Harvey pers. comm. 2021). Substantial sequencing of this group has been undertaken throughout the Pilbara and so continual, routine morphological and genetic

treatment of this family will continue to improve the current state of taxonomy, however the species presented herein should be treated as tentative.

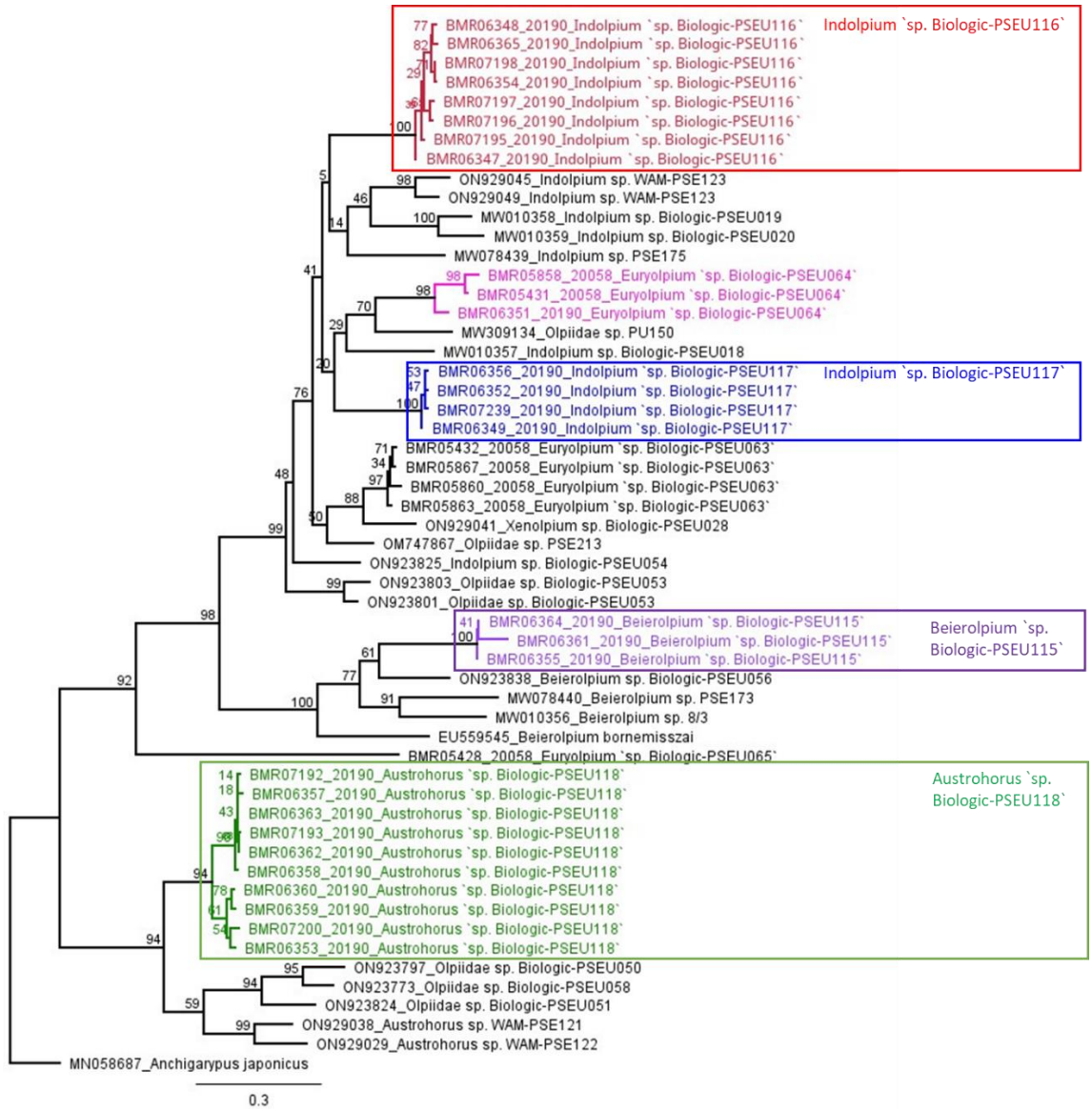


Figure 3.2: Maximum Likelihood phylogeny for Pseudoscorpiones, with bootstrap values.

Table 3.3: Pairwise distances (%) for the Pseudoscorpiones dataset.

COI Pairwise Distances (%)	EU559545	MW010356	ON923838	BMR06355	BMR06364	BMR06361	MW078440	MW010357	MW010358	MW010359	ON929045	ON929049	MW309134	BMR05431	BMR05858	BMR06351	OM747867	ON929041	BMR05432	BMR05867	BMR05863	BMR05860	BMR06347	BMR07195	BMR07196	BMR07197	BMR06348
EU559545_Beierolpium bornemisszai		18.1	17.3	18.8	18.9	18.4	18.1	22.1	20.2	22.7	22.2	22.5	21.2	24.2	25.6	25.3	19.6	21	18.8	19	19	19.1	20.3	21	21.4	21.1	23.1
MW010356_Beierolpium sp. 8/3	18.1		15.8	18.2	20	19.3	16.2	25.6	24.2	21.6	23	22.7	23	25.1	26.7	24.5	22.2	22.7	21.6	21.8	21.8	21.6	22.4	22.8	23	22.7	23.6
ON923838_Beierolpium sp. Biologic-PSEU056	17.3	15.8		16.2	15.9	16.4	17.6	22.3	23.8	23.6	23.3	22.1	22.4	26.2	27.8	25.8	21.1	24.1	22.8	22.8	23.1	23.3	22.7	22.2	22.2	22.1	22.5
BMR06355_20190_Beierolpium `sp. Biologic-PSEU115	18.8	18.2	16.2		0.44	5.04	19.1	23.6	24.9	24.1	25.3	24.1	21.9	23.4	24.2	24	22.6	25.6	23.7	23.8	24.1	23.8	22.3	22.4	22.4	22.4	22.3
BMR06364_20190_Beierolpium `sp. Biologic-PSEU115	18.9	20	15.9	0.44		5.07	20	23.6	24.9	24.1	25.3	24.3	22.1	23.1	24	23.8	22.3	25.8	23.1	23.1	23.7	23.6	22	22	22.7	23.1	22.5
BMR06361_20190_Beierolpium `sp. Biologic-PSEU115	18.4	19.3	16.4	5.04	5.07		18.2	25.6	26.2	25.2	25.7	25.2	24.1	25.1	25.6	26.2	23.4	26.3	24	24.4	24.7	24.7	23.2	23.1	23.4	23.4	24.7
MW078440_Beierolpium sp. PSE173	18.1	16.2	17.6	19.1	20	18.2		25.2	24	23	25	22.7	24.1	26.4	25.8	25.1	22.5	23.4	22.5	23.2	22.5	23.2	22.5	22.8	23	22.8	25.1
MW010357_Indolpium sp. Biologic-PSEU018	22.1	25.6	22.3	23.6	23.6	25.6	25.2		20	19.2	19	17	16.4	18.1	19	17.7	18.3	18.5	19.2	18.8	18.4	18.5	16.6	16.3	17.4	17.9	17.2
MW010358_Indolpium sp. Biologic-PSEU019	20.2	24.2	23.8	24.9	24.9	26.2	24	20		11.3	16.9	15.8	16.9	17.8	19.8	18.7	20	20	18.7	19.1	19	19.8	19.1	19.8	20	20.7	20.2
MW010359_Indolpium sp. Biologic-PSEU020	22.7	21.6	23.6	24.1	24.1	25.2	23	19.2	11.3		18.1	16.8	18.6	21	22.5	20.5	20.8	20.5	19.7	19.7	19.5	19.9	20.1	19.9	20.1	20.8	20.3
ON929045_Indolpium sp. WAM-PSE123	22.2	23	23.3	25.3	25.3	25.7	25	19	16.9	18.1		8.39	16.6	18.3	18.1	19.2	16.2	16.6	15.3	15.1	15.7	15.9	16.5	17.1	17	17	18.7
ON929049_Indolpium sp. WAM-PSE123	22.5	22.7	22.1	24.1	24.3	25.2	22.7	17	15.8	16.8	8.39		15.7	19	19.2	18.3	17.2	17.7	17.4	17.9	18	17.7	15.9	16.6	17.4	17.4	17.9
MW309134_Olpiidae sp. PU150	21.2	23	22.4	21.9	22.1	24.1	24.1	16.4	16.9	18.6	16.6	15.7		14.4	15.7	14.6	15.9	17	16.8	16.6	16.5	17	16.8	17	17.3	17.5	17.3
BMR05431_20058_Euryolpium `sp. Biologic-PSEU064`	24.2	25.1	26.2	23.4	23.1	25.1	26.4	18.1	17.8	21	18.3	19	14.4		3.08	7.71	17	17.2	18.3	18.3	18.2	18.3	17.8	18.1	17.6	18.5	18.3
BMR05858_20058_Euryolpium `sp. Biologic-PSEU064`	25.6	26.7	27.8	24.2	24	25.6	25.8	19	19.8	22.5	18.1	19.2	15.7	3.08		8.81	17.2	18.5	18.7	18.7	18.6	18.7	18.7	19.4	19.8	20.3	20
BMR06351_20190_Euryolpium `sp. Biologic-PSEU064`	25.3	24.5	25.8	24	23.8	26.2	25.1	17.7	18.7	20.5	19.2	18.3	14.6	7.71	8.81		17.2	15.2	17.4	17.8	17.7	17.8	16.5	17.6	17.6	17.2	17.4
OM747867_Olpiidae sp. PSE213	19.6	22.2	21.1	22.6	22.3	23.4	22.5	18.3	20	20.8	16.2	17.2	15.9	17	17.2	17.2		14.8	12.1	12.2	11.9	11.3	13.7	14.7	14.5	14.4	15.9
ON929041_Xenolpium sp. Biologic-PSEU028	21	22.7	24.1	25.6	25.8	26.3	23.4	18.5	20	20.5	16.6	17.7	17	17.2	18.5	15.2	14.8		11.5	11.3	11.4	11.7	16.3	16.3	16.3	15.7	15.9
BMR05432_20058_Euryolpium `sp. Biologic-PSEU063`	18.8	21.6	22.8	23.7	23.1	24	22.5	19.2	18.7	19.7	15.3	17.4	16.8	18.3	18.7	17.4	12.1	11.5		1.07	2.14	3.21	15.6	15.7	15.3	15.1	18.7
BMR05867_20058_Euryolpium `sp. Biologic-PSEU063`	19	21.8	22.8	23.8	23.1	24.4	23.2	18.8	19.1	19.7	15.1	17.9	16.6	18.3	18.7	17.8	12.2	11.3	1.07		2.14	3.05	14.8	15.6	15.4	15	17.8
BMR05863_20058_Euryolpium `sp. Biologic-PSEU063`	19	21.8	23.1	24.1	23.7	24.7	22.5	18.4	19	19.5	15.7	18	16.5	18.2	18.6	17.7	11.9	11.4	2.14	2.14		2.9	15.3	15.7	15.4	15.3	17.7
BMR05860_20058_Euryolpium `sp. Biologic-PSEU063`	19.1	21.6	23.3	23.8	23.6	24.7	23.2	18.5	19.8	19.9	15.9	17.7	17	18.3	18.7	17.8	11.3	11.7	3.21	3.05	2.9		15.6	16	16.2	16	17.6
BMR06347_20190_Indolpium `sp. Biologic-PSEU116`	20.3	22.4	22.7	22.3	22	23.2	22.5	16.6	19.1	20.1	16.5	15.9	16.8	17.8	18.7	16.5	13.7	16.3	15.6	14.8	15.3	15.6		1.68	3.05	2.9	3.96
BMR07195_20190_Indolpium `sp. Biologic-PSEU116`	21	22.8	22.2	22.4	22	23.1	22.8	16.3	19.8	19.9	17.1	16.6	17	18.1	19.4	17.6	14.7	16.3	15.7	15.6	15.7	16	1.68		1.98	2.44	3.08
BMR07196_20190_Indolpium `sp. Biologic-PSEU116`	21.4	23	22.2	22.4	22.7	23.4	23	17.4	20	20.1	17	17.4	17.3	17.6	19.8	17.6	14.5	16.3	15.3	15.4	15.4	16.2	3.05	1.98		1.37	3.3
BMR07197_20190_Indolpium `sp. Biologic-PSEU116`	21.1	22.7	22.1	22.4	23.1	23.4	22.8	17.9	20.7	20.8	17	17.4	17.5	18.5	20.3	17.2	14.4	15.7	15.1	15	15.3	16	2.9	2.44	1.37		2.86
BMR06348_20190_Indolpium `sp. Biologic-PSEU116`	23.1	23.6	22.5	22.3	22.5	24.7	25.1	17.2	20.2	20.3	18.7	17.9	17.3	18.3	20	17.4	15.9	15.9	18.7	17.8	17.7	17.6	3.96	3.08	3.3	2.86	
BMR06365_20190_Indolpium `sp. Biologic-PSEU116`	23.4	23.6	22.7	21.8	22	24.2	24.7	17	20.4	19.9	18.5	17.2	17.3	18.3	20	17.8	16.3	16.8	17.6	17.2	17.5	17.4	4.19	2.86	3.52	3.52	1.54
BMR06354_20190_Indolpium `sp. Biologic-PSEU116`	21.4	22.7	21.9	22.3	21.8	23.7	23.5	17.4	19.8	19.9	17.6	17.9	16.8	17.8	19.6	17.4	14.4	15.7	15.9	15.4	16	16.2	3.66	2.9	3.66	3.36	1.76
BMR07198_20190_Indolpium `sp. Biologic-PSEU116`	21.3	22.5	21.8	22.4	22	23.5	23.3	17.7	19.6	19.2	17.4	17.2	16.6	18.1	19.8	17.6	14.5	15.9	15.7	15.6	16	16.3	3.21	2.44	3.21	2.9	1.54
ON923825_Indolpium sp. Biologic-PSEU054	20	22.1	21.6	22.6	22	23.7	22.1	18.5	18	18.8	18.4	18.8	16.6	19.4	20.5	18.1	14.4	15	15	14.4	15.3	15.6	14.4	15.6	15.8	15.3	16.7
ON923801_Olpiidae sp. Biologic-PSEU053	20.9	22.4	21.8	21.6	21.8	24	24.4	15.8	17.3	18.9	18.7	16.7	16	20	20.7	19.6	16	17.1	15.6	15.8	15.2	15.1	18	18	19.1	18.2	18.4
ON923803_Olpiidae sp. Biologic-PSEU053	20.3	22.4	23.3	23.6	23.6	24.5	23.3	16.6	18	18.3	18.5	19	17.7	21.2	22.3	20	15.1	17.9	15.6	15.9	15.6	14.7	16.5	16.5	16.8	16.7	17.4
BMR06349_20190_Indolpium `sp. Biologic-PSEU117`	22.9	24.7	23.6	24.5	24.7	25.3	24.2	15.9	15.3	16.8	18.3	17.4	16.8	16.5	18.1	16.3	16.1	17.9	17.6	17.6	17.7	18.3	17.6	17.2	17.2	17.6	16.7
BMR07239_20190_Indolpium `sp. Biologic-PSEU117`	22.5	24.7	23.4	24.7	24.9	25.1	24	15.5	15.3	16.8	18.1	16.8	17.5	17	18.1	16.3	16.1	18.3	17.6	18.1	18	18.7	17.4	17.4	17.4	17.8	17.8
BMR06352_20190_Indolpium `sp. Biologic-PSEU117`	22.3	24.7	23.4	24.5	24.7	24.9	24.7	15.7	15.6	16.3	18.3	17.2	16.8	17.2	18.3	17	16.5	19	17.8	17.8	18.2	18.9	17.2	17.4	17.8	18.3	17.8
BMR06356_20190_Indolpium `sp. Biologic-PSEU117`	21.6	23.3	23.1	24.4	24.7	24.6	23.3	15.7	15.6	17	18.2	16.6	17.5	17.4	18.1	17.2	14.7	18.8	15.4	15.6	16.2	16.2	16.2	16.3	16.6	16.5	17.6
MW078439_Indolpium sp. PSE175	21.6	25.2	23	24.7	24.7	25.4	23	18.8	18.2	20.3	17.4	17.4	19.5	18.5	19	18.3	15.7	16.6	18.1	17.9	17.3	17.2	17	17.7	18.5	17.9	17.7
MN058687_Anchigarypus japonicus	20.8	25.9	24.8	23	23	23.5	28.3	23.5	23.6	23.5	24.8	23.5	22.6	23.9	24.6	24.8	21.5	24.6	23.9	23.7	24	23.5	22.4	22.8	24.3	24.6	23.2
ON923773_Olpiidae sp. Biologic-PSEU058	23.6	23.7	22.7	25.2	26.4	25.3	25.5	28.3	24.4	25.2	24.1	22.5	25.4	26.9	27.8	25.3	25.5	23.6	25.3	25.5	25.8	25.8	24.3	24.3	23.8	23.5	24.9
ON923797_Olpiidae sp. Biologic-PSEU050	24.5	22.4	24	25.7	26.7	24.9	24.5	26.1	24.9	23.4	24.4	23	24.8	25.3	26.7	24.2	23.8	25.4	25	25.5	25.5	25.2	26.1	25.8	26.3	25.7	26.4
ON923824_Olpiidae sp. Biologic-PSEU051	23.8	26.5	24.7	26.7	26.7	26.9	27.4	24.9	22.9	23.8	24.1	23	24.3	25.6	26.9	26.1	24.7	25.4	23.6	23.6	23.7	24.5	25.2	25.6	25.6	26.3	26.9
ON929029_Austrohorus sp. WAM-PSE122	24.2	25.9	25.2	26.3	25.6	26.6	25.7	26.8	27.4	28.3	25.8	26.1	26.8	28.3	28.5	30.2	23.6	28.3	24.3	24.6	24.5	24.2	25.5	25.4	25.2	25.5	26.5
ON929038_Austrohorus sp. WAM-PSE121	25.5	24	27	26.7	26.2	26.7	25	27.8	25.1	24.7	26.9	25.8	26.8	27.8	28	27.3	25	27.8	24.7	24.7	25.3	25.5	25	25.2	25.3	25.3	27.3
BMR06353_20190_Austrohorus `sp. Biologic-PSEU118	23.1	27.3	26	25.1	25.6	26.7	28.9	26.7	23.1	25.8	25.6	24.7	23	23.1	24	23.4	24.7	24.1	25.3	25.3	25	26	25.3	25.1	25.6	24.5	24.9
BMR07200_20190_Austrohorus `sp. Biologic-PSEU118	22	26	26	25.6	26	26.2	28	25.8	23.3	25.4	25.3	24.5	23.2	23.8	24.2	24	24	24.1	25.6	25.1	24.3	25.3	24.7	24.9	25.3	24.7	24.2
BMR06359_20190_Austrohorus `sp. Biologic-PSEU118	22.9	27.8	26.9	25.3	25.8	26.4	29.5	26.3	22.9	25.8	24.5	23.2	23.7	23.4	23.8	23.1	24.2	24.1	25.3	25.3	24.6	25.6	24.5	24.7	24.7	23.6	24.5
BMR06360_20190_Austrohorus `sp. Biologic-PSEU118	23.1	26.2	26.7	26.4	25.8	27.2	27	26.5	22.9	25.8	25.2	23.6	23.2	23.6	24	22.9	23.8	23.6	24.6	24.7	23.8	24.9	24.9	25	24.9	24	24.7
BMR06357_20190_Austrohorus `sp. Biologic-PSEU118	24.8	26.8	26	27.8	26.9	29	26.5	26.7	23.8	24.7	27.3	25.2	25	25.1	25.6	25.3	26.3	24.1	24.6	24.7	24.4	25.2	26.4	26.4	26.9	26.1	26
BMR06362_20190_Austrohorus `sp. Biologic-PSEU118	24.3	26.5	26.3	27.9	27.1	28.9	26.5	26.3	23.3	24.3	26.6	24.3	24.8	25.1	25.6	25.3	25.8	23.6	25	24.9	24.4	25	25.8	26.1	26.6	25.8	25.8
BMR07193_20190_Austrohorus `sp. Biologic-PSEU118	24.3	26.5	26.3	28.1	27.3	29	26.5	26.3	23.3	24.3	26.7	24.5	24.8	25.1	25.6	25.3	26	23.8	24.9	24.7	24.3	24.9	26	26.3	26.7	26	26
BMR06363_20190_Austrohorus `sp. Biologic-PSEU118	24.9	26.9	26.4	28.3	27.6	29.2	26.8	26.8	23.7	24.6	26.8	24.4	25.3	25.7	26.1	25.9	26.2	24	25.4	25.3	24.8	25.4	26.3	26.6	27.1	26.3	26.3
BMR07192_20190_Austrohorus `sp. Biologic-PSEU118	24.9	26.8	26.3	28.4	27.8	29.3	26.9	26.9	23.3	24.3	26.9	24.9	25.4	25.8	25.8	26	25.7	23.8	25.5	25.3	24.9	25.2	26.4	26.7	27	26.3	25.8
BMR06358_20190_Austrohorus `sp. Biologic-PSEU118	24.9	26.3	25.9	27.8	27.1	28.7	27.3	26.5	23.8	24.7	26.4	24.3	24.8	25.3	25.8	25.1	26	23.4	24.6	24.7	24.3	24.9	26	26.3	26.7	25.7	26
BMR05428_20058_Euryolpium `sp. Biologic-PSEU065`	23.4	25.7	24.4	26	26	26.4	26.5	29.4	27.6	27.6	24.7	24.7	26.3	29.3	30.2	29.1	23.2	26.7	23.4	23.8	24.3	24.1	24.4	24.9	25.2	25	27.5

COI Pairwise Distances (%)	BMR06365	BMR06354	BMR07198	ON923825	ON923801	ON923803	BMR06349	BMR07239	BMR06352	BMR06356	MW078439	MN058687	ON923773	ON923797	ON923824	ON929029	ON929038	BMR06353	BMR07200	BMR06359	BMR06360	BMR06357	BMR06362	BMR07193	BMR06363	BMR07192	BMR06358	BMR05428
EU559545_Beierolpium bornemisszai	23.4	21.4	21.3	20	20.9	20.3	22.9	22.5	22.3	21.6	21.6	20.8	23.6	24.5	23.8	24.2	25.5	23.1	22	22.9	23.1	24.77	24.31	24.31	24.85	24.92	24.92	23.39
MW010356_Beierolpium sp. 8/3	23.6	22.7	22.5	22.1	22.4	22.4	24.7	24.7	24.7	23.3	25.2	25.9	23.7	22.4	26.5	25.9	24	27.3	26	27.8	26.2	26.8	26.49	26.49	26.88	26.8	26.34	25.73
ON923838_Beierolpium sp. Biologic-PSEU056	22.7	21.9	21.8	21.6	21.8	23.3	23.6	23.4	23.4	23.1	23	24.8	22.7	24	24.7	25.2	27	26	26	26.9	26.7	26.03	26.34	26.34	26.42	26.34	25.88	24.35
BMR06355_20190_Beierolpium `sp. Biologic-PSEU115	21.8	22.3	22.4	22.6	21.6	23.6	24.5	24.7	24.5	24.4	24.7	23	25.2	25.7	26.7	26.3	26.7	25.1	25.6	25.3	26.4	27.79	27.94	28.09	28.32	28.4	27.79	25.95
BMR06364_20190_Beierolpium `sp. Biologic-PSEU115	22	21.8	22	22	21.8	23.6	24.7	24.9	24.7	24.7	24.7	23	26.4	26.7	26.7	25.6	26.2	25.6	26	25.8	25.8	26.87	27.09	27.31	27.64	27.75	27.09	25.99
BMR06361_20190_Beierolpium `sp. Biologic-PSEU115	24.2	23.7	23.5	23.7	24	24.5	25.3	25.1	24.9	24.6	25.4	23.5	25.3	24.9	26.9	26.6	26.7	26.7	26.2	26.4	27.2	29.01	28.85	29.01	29.24	29.31	28.7	26.41
MW078440_Beierolpium sp. PSE173	24.7	23.5	23.3	22.1	24.4	23.3	24.2	24	24.7	23.3	23	28.3	25.5	24.5	27.4	25.7	25	28.9	28	29.5	27	26.54	26.54	26.54	26.77	26.85	27.31	26.54
MW010357_Indolpium sp. Biologic-PSEU018	17	17.4	17.7	18.5	15.8	16.6	15.9	15.5	15.7	15.7	18.8	23.5	28.3	26.1	24.9	26.8	27.8	26.7	25.8	26.3	26.5	26.71	26.27	26.27	26.82	26.93	26.49	29.36
MW010358_Indolpium sp. Biologic-PSEU019	20.4	19.8	19.6	18	17.3	18	15.3	15.3	15.6	15.6	18.2	23.6	24.4	24.9	22.9	27.4	25.1	23.1	23.3	22.9	22.9	23.78	23.33	23.33	23.67	23.33	23.78	27.56
MW010359_Indolpium sp. Biologic-PSEU020	19.9	19.9	19.2	18.8	18.9	18.3	16.8	16.8	16.3	17	20.3	23.5	25.2	23.4	23.8	28.3	24.7	25.8	25.4	25.8	25.8	24.72	24.28	24.28	24.61	24.28	24.72	27.59
ON929045_Indolpium sp. WAM-PSE123	18.5	17.6	17.4	18.4	18.7	18.5	18.3	18.1	18.3	18.2	17.4	24.8	24.1	24.4	24.1	25.8	26.9	25.6	25.3	24.5	25.2	27.33	26.56	26.72	26.79	26.87	26.41	24.73
ON929049_Indolpium sp. WAM-PSE123	17.2	17.9	17.2	18.8	16.7	19	17.4	16.8	17.2	16.6	17.4	23.5	22.5	23	23	26.1	25.8	24.7	24.5	23.2	23.6	25.17	24.28	24.5	24.39	24.94	24.28	24.72
MW309134_Olpiidae sp. PU150	17.3	16.8	16.6	16.6	16	17.7	16.8	17.5	16.8	17.5	19.5	22.6	25.4	24.8	24.3	26.8	26.8	23	23.2	23.7	23.2	25	24.78	24.78	25.33	25.44	24.78	26.33
BMR05431_20058_Euryolpium `sp. Biologic-PSEU064`	18.3	17.8	18.1	19.4	20	21.2	16.5	17	17.2	17.4	18.5	23.9	26.9	25.3	25.6	28.3	27.8	23.1	23.8	23.4	23.6	25.11	25.11	25.11	25.66	25.77	25.33	29.3
BMR05858_20058_Euryolpium `sp. Biologic-PSEU064`	20	19.6	19.8	20.5	20.7	22.3	18.1	18.1	18.3	18.1	19	24.6	27.8	26.7	26.9	28.5	28	24	24.2	23.8	24	25.55	25.55	25.55	26.1	25.77	25.77	30.18
BMR06351_20190_Euryolpium `sp. Biologic-PSEU064`	17.8	17.4	17.6	18.1	19.6	20	16.3	16.3	17	17.2	18.3	24.8	25.3	24.2	26.1	30.2	27.3	23.4	24	23.1	22.9	25.33	25.33	25.33	25.88	25.99	25.11	29.07
OM747867_Olpiidae sp. PSE213	16.3	14.4	14.5	14.4	16	15.1	16.1	16.1	16.5	14.7	15.7	21.5	25.5	23.8	24.7	23.6	25	24.7	24	24.2	23.8	26.26	25.8	25.95	26.18	25.65	25.95	23.21
ON929041_Xenolpium sp. Biologic-PSEU028	16.8	15.7	15.9	15	17.1	17.9	17.9	18.3	19	18.8	16.6	24.6	23.6	25.4	25.4	28.3	27.8	24.1	24.1	24.1	23.6	24.06	23.62	23.84	23.95	23.84	23.4	26.71
BMR05432_20058_Euryolpium `sp. Biologic-PSEU063`	17.6	15.9	15.7	15	15.6	15.6	17.6	17.6	17.8	15.4	18.1	23.9	25.3	25	23.6	24.3	24.7	25.3	25.6	25.3	24.6	24.58	25.04	24.89	25.42	25.5	24.58	23.36
BMR05867_20058_Euryolpium `sp. Biologic-PSEU063`	17.2	15.4	15.6	14.4	15.8	15.9	17.6	18.1	17.8	15.6	17.9	23.7	25.5	25.5	23.6	24.6	24.7	25.3	25.1	25.3	24.7	24.73	24.89	24.73	25.27	25.34	24.73	23.82
BMR05863_20058_Euryolpium `sp. Biologic-PSEU063`	17.5	16	16	15.3	15.2	15.6	17.7	18	18.2	16.2	17.3	24	25.8	25.5	23.7	24.5	25.3	25	24.3	24.6	23.8	24.43	24.43	24.27	24.81	24.89	24.27	24.27
BMR05860_20058_Euryolpium `sp. Biologic-PSEU063`	17.4	16.2	16.3	15.6	15.1	14.7	18.3	18.7	18.9	16.2	17.2	23.5	25.8	25.2	24.5	24.2	25.5	26	25.3	25.6	24.9	25.19	25.04	24.89	25.42	25.19	24.89	24.12
BMR06347_20190_Indolpium `sp. Biologic-PSEU116`	4.19	3.66	3.21	14.4	18	16.5	17.6	17.4	17.2	16.2	17	22.4	24.3	26.1	25.2	25.5	25	25.3	24.7	24.5	24.9	26.41	25.8	25.95	26.34	26.41	25.95	24.43
BMR07195_20190_Indolpium `sp. Biologic-PSEU116`	2.86	2.9	2.44	15.6	18	16.5	17.2	17.4	17.4	16.3	17.7	22.8	24.3	25.8	25.6	25.4	25.2	25.1	24.9	24.7	25	26.41	26.11	26.26	26.64	26.72	26.26	24.89
BMR07196_20190_Indolpium `sp. Biologic-PSEU116`	3.52	3.66	3.21	15.8	19.1	16.8	17.2	17.4	17.8	16.6	18.5	24.3	23.8	26.3	25.6	25.2	25.3	25.6	25.3	24.7	24.9	26.87	26.56	26.72	27.1	27.02	26.72	25.19
BMR07197_20190_Indolpium `sp. Biologic-PSEU116`	3.52	3.36	2.9	15.3	18.2	16.7	17.6	17.8	18.3	16.5	17.9	24.6	23.5	25.7	26.3	25.5	25.3	24.5	24.7	23.6	24	26.11	25.8	25.95	26.34	26.26	25.65	25.04
BMR06348_20190_Indolpium `sp. Biologic-PSEU116`	1.54	1.76	1.54	16.7	18.4	17.4	16.7	17.8	17.8	17.6	17.7	23.2	24.9	26.4	26.9	26.5	27.3	24.9	24.2	24.5	24.7	25.99	25.77	25.99	26.32	25.77	25.99	27.53
BMR06365_20190_Indolpium `sp. Biologic-PSEU116`		1.98	1.76	17	18.7	17.8	16.7	17.4	17.4	17.2	17.4	23	25.1	27.3	26.5	26.9	27.3	25.8	25.1	25.3	25.6	27.31	27.09	27.31	27.64	27.09	27.31	27.53
BMR06354_20190_Indolpium `sp. Biologic-PSEU116`	1.98		0.46	15.4	18.2	17	15.9	17	17	16.3	17	23	24.1	26	25.6	25.8	25.7	24.2	24.5	24.7	25.5	26.26	26.26	26.41	26.49	26.11	26.11	25.19
BMR07198_20190_Indolpium `sp. Biologic-PSEU116`	1.76	0.46		15.6	17.6	16.5	16.1	16.7	16.7	16.2	17.2	22.8	24	25.8	25.4	26	25.5	24.5	24.2	24.5	25.3	26.11	26.11	26.26	26.34	25.95	25.95	25.34
ON923825_Indolpium sp. Biologic-PSEU054	17	15.4	15.6		14.4	14.4	18.5	18.9	18.5	16.5	19.2	21.7	23.9	24.3	24.7	23.9	23.9	22.5	22.3	22.7	23.6	26.15	25.84	25.99	26.22	26.3	25.38	23.55
ON923801_Olpiidae sp. Biologic-PSEU053	18.7	18.2	17.6	14.4		6	17.1	17.1	17.3	17.1	18.2	22.9	24.9	24	24	26.3	25.6	22.9	23.1	22.7	22.9	24	23.78	23.78	24.11	24.22	23.33	26.22
ON923803_Olpiidae sp. Biologic-PSEU053	17.8	17	16.5	14.4	6		16.7	16.7	17	16.4	18.5	23	25.5	25.1	23.6	24.8	25.5	24	23.4	23.8	22.8	24.16	23.85	23.85	24.24	24.01	23.7	25.54
BMR06349_20190_Indolpium `sp. Biologic-PSEU117`	16.7	15.9	16.1	18.5	17.1	16.7		1.15	1.1	1.32	19	23	25.3	25.3	24.9	27.6	24.5	23.8	22.9	24.2	24.5	24.67	24.67	24.89	25	24.67	24.67	27.75
BMR07239_20190_Indolpium `sp. Biologic-PSEU117`	17.4	17	16.7	18.9	17.1	16.7	1.15		1.1	1.32	18.8	22.6	25.1	24.9	24.1	28	24.5	23.8	22.5	23.8	24	24.67	24.23	24.45	24.56	24.23	24.23	27.53
BMR06352_20190_Indolpium `sp. Biologic-PSEU117`	17.4	17	16.7	18.5	17.3	17	1.1	1.1		1.1	19.2	22.1	25.3	25.3	24.1	28	24.2	23.6	22.7	24	24.2	24.89	24.89	25.11	25.22	24.89	24.89	27.31
BMR06356_20190_Indolpium `sp. Biologic-PSEU117`	17.2	16.3	16.2	16.5	17.1	16.4	1.32	1.32	1.1		19	22.1	24.3	24	24.5	25.2	23.5	23.8	22.9	24.2	24.6	25.04	24.89	25.04	25.27	25.04	24.89	24.27
MW078439_Indolpium sp. PSE175	17.4	17	17.2	19.2	18.2	18.5	19	18.8	19.2	19		25.9	26.1	26.5	25.6	26.8	31.1	25.6	25.4	25.6	25.6	24.28	23.84	24.06	24.17	23.84	24.06	28.92
MN058687_Anchigarypus japonicus	23	23	22.8	21.7	22.9	23	23	22.6	22.1	22.1	25.9		22.1	23	20.8	21.5	21.5	17.9	16.8	17.7	18.1	20.8	20.58	20.8	20.69	20.8	20.8	22.12
ON923773_Olpiidae sp. Biologic-PSEU058	25.1	24.1	24	23.9	24.9	25.5	25.3	25.1	25.3	24.3	26.1	22.1		9.47	13.3	17	16.6	18.7	19.4	18.3	17.7	19.08	18.63	18.63	18.55	18.32	18.17	22.6
ON923797_Olpiidae sp. Biologic-PSEU050	27.3	26	25.8	24.3	24	25.1	25.3	24.9	25.3	24	26.5	23	9.47		14.6	16.7	16.5	18.7	19.6	18.5	18	18.63	18.17	18.17	18.4	18.32	18.02	22.6
ON923824_Olpiidae sp. Biologic-PSEU051	26.5	25.6	25.4	24.7	24	23.6	24.9	24.1	24.1	24.5	25.6	20.8	13.3	14.6		20.6	18.3	17.7	18.3	18.8	18.3	17.44	17.88	17.88	17.55	17.44	17.88	24.94
ON929029_Austrohorus sp. WAM-PSE122	26.9	25.8	26	23.9	26.3	24.8	27.6	28	28	25.2	26.8	21.5	17	16.7	20.6		11.8	17.2	16.6	17.9	16.1	17.58	17.43	17.58	17.97	17.89	17.28	23.55
ON929038_Austrohorus sp. WAM-PSE121	27.3	25.7	25.5	23.9	25.6	25.5	24.5	24.5	24.2	23.5	31.1	21.5	16.6	16.5	18.3	11.8		17	16.5	17	16.3	16.34	16.64	16.79	16.72	16.64	16.03	23.82
BMR06353_20190_Austrohorus `sp. Biologic-PSEU118	25.8	24.2	24.5	22.5	22.9	24	23.8	23.8	23.6	23.8	25.6	17.9	18.7	18.7	17.7	17.2	17		2.42	3.3	3.08	7.93	7.71	7.93	8.26	8.37	7.71	25.11
BMR07200_20190_Austrohorus `sp. Biologic-PSEU118	25.1	24.5	24.2	22.3	23.1	23.4	22.9	22.5	22.7	22.9	25.4	16.8	19.4	19.6	18.3	16.6	16.5	2.42		3.96	3.74	8.59	7.93	8.15	8.48	8.59	7.93	25.99
BMR06359_20190_Austrohorus `sp. Biologic-PSEU118	25.3	24.7	24.5	22.7	22.7	23.8	24.2	23.8	24	24.2	25.6	17.7	18.3	18.5	18.8	17.9	17	3.3	3.96		1.1	9.69	8.15	8.37	8.26	8.81	8.15	25.77
BMR06360_20190_Austrohorus `sp. Biologic-PSEU118	25.6	25.5	25.3	23.6	22.9	22.8	24.5	24	24.2	24.6	25.6	18.1	17.7	18	18.3	16.1	16.3	3.08	3.74	1.1		7.94	6.72	6.87	7.25	7.33	7.02	24.73
BMR06357_20190_Austrohorus `sp. Biologic-PSEU118	27.3	26.3	26.1	26.2	24	24.2	24.7	24.7	24.9	25	24.3	20.8	19.1	18.6	17.4	17.6	16.3	7.93	8.59	9.69	7.94		1.22	1.37	1.15	1.53	1.68	25.65
BMR06362_20190_Austrohorus `sp. Biologic-PSEU118	27.1	26.3	26.1	25.8	23.8	23.9	24.7	24.2	24.9	24.9	23.8	20.6	18.6	18.2	17.9	17.4	16.6	7.71	7.93	8.15	6.72	1.22		0.15	0.53	0.92	1.07	25.04
BMR07193_20190_Austrohorus `sp. Biologic-PSEU118	27.3	26.4	26.3	26	23.8	23.9	24.9	24.5	25.1	25	24.1	20.8	18.6	18.2	17.9	17.6	16.8	7.93	8.15	8.37	6.87	1.37	0.15		0.69	1.07	1.22	25.19
BMR06363_20190_Austrohorus `sp. Biologic-PSEU118	27.6	26.5	26.3	26.2	24.1	24.2	25	24.6	25.2	25.3	24.2	20.7	18.6	18.4	17.6	18	16.7	8.26	8.48	8.26	7.25	1.15	0.53	0.69		0.84	0.99	25.57
BMR07192_20190_Austrohorus `sp. Biologic-PSEU118	27.1	26.1	26	26.3	24.2	24	24.7	24.2	24.9	25	23.8	20.8	18.3	18.3	17.4	17.9	16.6	8.37	8.59	8.81	7.33	1.53	0.92	1.07	0.84		1.37	25.34
BMR06358_20190_Austrohorus `sp. Biologic-PSEU118	27.3	26.1	26	25.4	23.3	23.7	24.7	24.2	24.9	24.9	24.1	20.8	18.2	18	17.9	17.3	16	7.71	7.93	8.15	7.02	1.68	1.07	1.22	0.99	1.37		24.89
BMR05428_20058_Euryolpium `sp. Biologic-PSEU065`	27.5	25.2	25.3	23.6	26.2	25.5	27.8	27.5	27.3	24.3	28.9	22.1	22.6	22.6	24.9	23.6	23.8	25.1	26	25.8	24.7	25.65	25.04	25.19	25.57	25.34	24.89	

812753 818753 824753

7610106

7604106

SRE Invertebrate Fauna

- Araneae: *Conothele* `sp. Biologic-ARAN053`
- Pseudoscorpiones: *Austrohorus* `sp. Biologic-PSEU118`
- Pseudoscorpiones: *Beierolpium* `sp. Biologic-PSEU115`
- Pseudoscorpiones: *Indolpium* `sp. Biologic-PSEU116`
- Pseudoscorpiones: *Indolpium* `sp. Biologic-PSEU117`
- Pseudoscorpiones: Olpiidae gen. nov.
- Pseudoscorpiones: Olpiidae sp. indet.
- Scorpiones: Scorpiones sp. indet.
- Polydesmida: Paradoxosomatidae sp. indet.
- Isopoda: Armadillidae sp. indet.

Marble Bar Rd

LEGEND

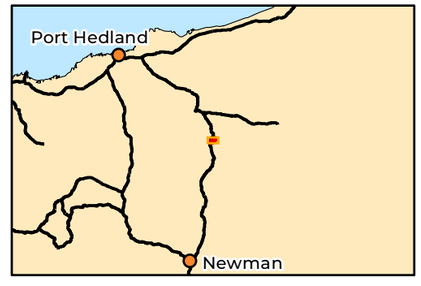
- Study Area
- State Road

Biologic

Scale 1:45,000

0 0.5 1 1.5 Km

Coordinate System: GDA2020 MGA Zone 50
Projection: Transverse Mercator
Datum: GDA2020 Created 03/02/2023



ATLAS IRON

McPhee Creek: SRE Molecular Systematics Analysis

Figure 3.3: SRE invertebrates recorded in the Study Area

4 Summary

Using well-established DNA extraction and sequencing methods, this molecular systematics analysis designated five distinct species/ OTUs to 27 high quality sequences from the Study Area. All OTUs, the areas in which they were found, and the specimen numbers per OTU are shown in Appendix A. The following are the key findings at the species/ OTU level:

- Araneae (COI): one OTUs, unique lineage to Study Area, and
- Pseudoscorpiones (COI): four unique lineages to Study Area and one widespread OTU.

5 References

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Appendix A: Specimen Data

BMR	Unique ID code	Site	Dec_Lat	Dec_Long	Coll_Date	Coll_Method	Lowest_ID_Legacy	OTU_Name	Reaction_State
Araneae: Halonoproctidae									
BMR06366	6500	SMCP-03	-21.564063	120.13717	2019-10-03	Foraging	Idiopidae?	Conothele `sp. Biologic-ARAN053`	PASS
Pseudoscorpiones: Olpiidae									
BMR06347	6197	SMCP-31	-21.616964	120.07716	2019-10-05	Soil Sieving	Indolpium AES01	Indolpium `sp. Biologic-PSEU116`	PASS
BMR06348	6050	SMCP-01	-21.551048	120.14286	2019-10-02	Opportunistic	Indolpium AES01	Indolpium `sp. Biologic-PSEU116`	PASS
BMR06349	6650	SMCP-20	-21.613038	120.10178	2019-10-04	Soil Sieving	Indolpium AES02	Indolpium `sp. Biologic-PSEU117`	PASS
BMR06350	5974b	SMCP-29	-21.584852	120.14144	2019-10-05	Soil Sieving	Olpiidae Gen. nov. ??		FAIL; PCR
BMR06351	5974	SMCP-29	-21.584852	120.14144	2019-10-05	Soil Sieving	poss. Euryolpium sp.	*Euryolpium* `sp. Biologic-PSEU064	PASS
BMR06352	6758	SMCP-17	-21.60562	120.09361	2019-10-04	Soil Sieving	Indolpium AES03	Indolpium `sp. Biologic-PSEU117`	PASS
BMR06353	7185	SMCP-06	-21.570625	120.1313	2019-10-03	Soil Sieving	Austrohorus AES03	Austrohorus `sp. Biologic-PSEU118`	PASS
BMR06354	6472	SMCP-22	-21.557411	120.14056	2019-10-04	Soil Sieving	Indolpium AES01	Indolpium `sp. Biologic-PSEU116`	PASS
BMR06355	7193	SMCP-20	-21.613038	120.10178	2019-10-04	Soil Sieving	Beierolpium '8/3'	Beierolpium `sp. Biologic-PSEU115`	PASS
BMR06356	6499	SMCP-21	-21.611579	120.10195	2019-10-04	Soil Sieving	Indolpium AES03	Indolpium `sp. Biologic-PSEU117`	PASS
BMR06357	6966	SMCP-19	-21.613819	120.10066	2019-10-04	Soil Sieving	Austrohorus AES03	Austrohorus `sp. Biologic-PSEU118`	PASS
BMR06358	6268	SMCP-18	-21.606013	120.0932	2019-10-04	Soil Sieving	Austrohorus AES03	Austrohorus `sp. Biologic-PSEU118`	PASS
BMR06359	7382	SMCP-03	-21.564063	120.13717	2019-10-03	Foraging	Austrohorus AES03	Austrohorus `sp. Biologic-PSEU118`	PASS
BMR06360	6246	SMCP-23	-21.559351	120.13707	2019-10-06	Leaf Sieving	Austrohorus AES03	Austrohorus `sp. Biologic-PSEU118`	PASS
BMR06361	6761	SMCP-24	-21.580647	120.11541	2019-10-04	Soil Sieving	Beierolpium '8/3'	Beierolpium `sp. Biologic-PSEU115`	PASS
BMR06362	6249	SMCP-12	-21.608056	120.09685	2019-10-03	Soil Sieving	Austrohorus AES03	Austrohorus `sp. Biologic-PSEU118`	PASS
BMR06363	6506	SMCP-35	-21.602861	120.07442	2019-10-07	Soil Sieving	Austrohorus AES03	Austrohorus `sp. Biologic-PSEU118`	PASS
BMR06364	6028	SMCP-33	-21.614447	120.09436	2019-10-06	Soil Sieving	Beierolpium '8/3'	Beierolpium `sp. Biologic-PSEU115`	PASS
BMR06365	6367	SMCP-30	-21.585877	120.14736	2019-10-05		Indolpium AES01	Indolpium `sp. Biologic-PSEU116`	PASS
BMR07192	17238	SMCP-15	-21.617734	120.08055	2019-10-04	Soil Sieving	Austrohorus AES03	Austrohorus `sp. Biologic-PSEU118`	PASS
BMR07193	17296	SMCP-21	-21.611579	120.10195	2019-10-04	Soil Sieving	Austrohorus AES03	Austrohorus `sp. Biologic-PSEU118`	PASS
BMR07194	17406	SMCP-24	-21.580647	120.11541	2019-10-04	Soil Sieving	Austrohorus AES03		FAIL; bad seq
BMR07195	17379	SMCP-18	-21.606013	120.0932	2019-10-04	Soil Sieving	Indolpium AES01	Indolpium `sp. Biologic-PSEU116`	PASS

BMR	Unique ID code	Site	Dec_Lat	Dec_Long	Coll_Date	Coll_Method	Lowest_ID_Legacy	OTU_Name	Reaction_State
BMR07196	17224	SMCP-25	-21.597634	120.10047	2019-10-04	Soil Sieving	Indolpium AES01	Indolpium `sp. Biologic-PSEU116`	PASS
BMR07197	17322	SMCP-35	-21.602861	120.07442	2019-10-07	Soil Sieving	Indolpium AES01	Indolpium `sp. Biologic-PSEU116`	PASS
BMR07198	17475	SMCP-23	-21.559351	120.13707	2019-10-06	Leaf Sieving	Indolpium AES01	Indolpium `sp. Biologic-PSEU116`	PASS
BMR07199	17532	SMCP-20	-21.613038	120.10178	2019-10-04	Soil Sieving	Indolpium AES03		FAIL; bad seq
BMR07200	17248	SMCP-27	-21.588828	120.15	2019-10-05	Soil Sieving	Austrohorus AES03	Austrohorus `sp. Biologic-PSEU118`	PASS
BMR07239	17325	SMCP-19	-21.613819	120.10066	2019-10-04	Soil Sieving	Indolpium AES03	Indolpium `sp. Biologic-PSEU117`	PASS