Head of College Scholars List Scheme

Summer Studentship

Report Form

This report should be completed by the student with his/her project supervisor. It should summarise the work undertaken during the project and, once completed, should be sent by email to: jill.morrison@glasgow.ac.uk within four weeks of the end of the studentship.

1. Student

   Surname: Rankin Forename: Aime

   E-mail address: 1002993r@student.gla.ac.uk

2. Supervisor:

   Surname: Page Forename: Roderic

   E-mail address: Roderic.Page@glasgow.ac.uk

3. Research Project Report

   3.1 Project Title (maximum 20 words):

   Are the parasites of marsupials ancient heirlooms or recent souvenirs?

   3.2 Project Lay Summary (copied from application):

   The parasites of an organism comprise a mixture of "heirlooms" and "souvenirs". Heirlooms are parasites retained from that species ancestor, and can be recognised by shared patterns of evolutionary history. Souvenirs are parasites that have been more recently acquired, for example as a result of a species moving into a new environment or evolving a new behaviour. This project seeks to determine what fraction of parasites on marsupial mammals are heirlooms reflecting their ancient past, and which parasites are souvenirs, perhaps of recent encounters with placental mammals.

3.3 Start Date: June 23rd Finish Date: July 19th

3.4 Original project aims and objectives (100 words max):

The null hypothesis is that the parasites of any marsupial mammal are more closely related to parasites found on other marsupials than to those on placental mammals. That is, marsupial parasites are heirlooms.

If we find parasites that are consistent with this hypothesis we can test a second hypothesis, namely that the marsupials and their parasites have cospeciated, by comparing evolutionary trees for both host and parasite. If cospeciation has occurred the trees should be more similar than we would expect due to chance.

If there are parasites that are shared between marsupial and placentals, we will test the hypothesis that sharing of parasites is related to degree of historical geographic isolation.

3.5 Methodology: Summarise and include reference to training received in research methods etc. (250 words max):

Lists of marsupial host-parasite associations were obtained by searching through the Bionames database (http:bionames.org) or by searching the literature. Using phylogenies on Bionames and from the literature, close relatives of these parasites were found as well as their hosts and country of residence. Appropriate graphs and diagrams were then created using programs such as Mintab to summarise this data. Then the sequence database GenBank was searched for DNA sequences from those parasites. For each parasite sequence in GenBank, similar sequences were retrieved using BLAST and used to build a phylogeny (this process is automated using http://iphylo.org/~rpage/phyloinformatics/blast). For each resultant phylogeny the hosts for the parasites were identified, from GenBank or the literature. Through interpreting the tree, it was determined whether marsupial parasites tended to cluster together on the tree, suggesting evolutionary association with their hosts, or scattered across the tree, suggesting episodes of host switching. This process required the employment of various data mining and analysis skills relevant to evolutionary biology.

3.6 Results: Summarise key findings (300 words max). Please include any relevant tables or images as an appendix to this report:

It was found that the close relatives of marsupial parasites, parasitized on a broad range of chordate groups. The majority of these were found on placental animals or mammalian animals in general (Figure 1). Only 5.5% of the parasites' relatives lived exclusively on marsupials and these came only from Australian Macropods (Figure 1). A small proportion of the parasites was found on Cephalopods, Insects or were free-living (Figure 1).

The range of countries in which these close relatives resided was equally as broad. The majority of the parasites' relatives (34%) had a worldwide presence, followed by 16.1% residing in Australia (Figure 2). Very few resided in South America and fewer still in New Guinea (Figure 2). However, for New Guinean parasites, the majority of their close relatives also resided there (Figure 2). Half of Australian parasites had relatives in Australia (Figure 2). Very few South American parasites had relatives exclusive to South America itself (Figure 2).

When comparing the taxonomic distance between host and parasite pairs, there was a slight positive correlation between the taxonomic distance in host and parasite pairs (Figure 3). However, the taxonomic distance between the host pairs was greater than that of the parasite pairs in general (Figure 3). Several outliers were found and identified. These included *Ophidascaris robertsi* and *Hepatozoon vivernus*.

Similarly, a positive correlation between taxonomic distance of parasite pairs and geographic distance between host pairs was observed (Figure 4). Again, interesting outliers were identified. They included *Toxoplasma* and *Heterodoxus octoseriatus.*

The majority of parasite trees did not show obvious clustering of marsupial hosts. A surprising find was the occurrence of distantly related taxa placed within parasite groups. An example of this came from the phylogeny for *Parastrongyloides* (Figure 5). Within this tree, frogs, bears and even maize were included in the parasite's phylogeny (Figure 5). However, some trees did show clusters of marsupial parasites. These included *Sarcocystis, H. octoseriatus, Brachylaima dasyuri*, *Macropodinium ennuensis* and *M. yalabense* (Figures 6 to 9 respectively).

3.7 Discussion (500 words max):

It was observed that few relatives of marsupial parasites were exclusive to marsupial hosts and that a majority were found to parasitize a great number of vertebrate groups (Figure 1). An explanation could be the presence of sampling error. Parasite phylogenies may not represent the diversity of parasites actually present. Not every parasite studied will be sequenced, so not every potential relative can be included in the tree. This could result in two parasites appearing more closely related than they are. For example, *Aggregata* appears as the closest relative of *H. vivernus* yet this would suggest a close relationship between marsupials and its host Cephalopods, which is not likely since there are no marine marsupials or for that matter, terrestrial Cephalopods.

Furthermore, the geographic distribution of the parasites was found to be diverse (Figure 2). Only parasites of New Guinea had the majority of relatives in the same area. This may be because the island has been relatively isolated since the end of the last ice age (Gascoigne, 1998). Regards the number of parasites found worldwide, it may be that the parasites are not host specific such as *Strongyloides* and can infect a range of vertebrates, hence their global presence. Again, sampling error would affect these results, so geographically distant parasites may not be as closely related to the marsupial parasites as suggested. This may explain why so few parasites of South American marsupials appear to have relatives in the same region.

As might be expected, the more taxonomically distant a parasite was from its closest relative, the more taxonomically distant the hosts of these parasites would be (Figure 3). Similarly, increasing geographic distance between host pairs correlated with the taxonomic distance between parasite pairs (Figure 4). Similar results of this nature have been found in other studies looking at helminth communities (Poulin, 2003).

The abundance of phylogenies not showing obvious clustering of marsupial hosts could be due to the sampling error discussed earlier. The occurrence of distantly related taxa in the tree itself may be down to using sequences from conserved genes or it could be that very short sequences were used. Additionally, it could be the case that the sequence used may have at some point integrated itself into the host's genome making it appear that the parasite and host were closely related. Therefore, an improvement to BLAST would be the addition of confidence values to give an impression of the reliability of the tree.

For the trees that did exhibit marsupial hosts clustered around groups of parasites, this could be evidence of cospeciation and further research should be carried out to test this. Studies on the protozoan *Sarcocystis* (Figure 6) have produced results suggesting that Sarcocystids from Australia and South America are monophyletic and have co-evolved with their marsupial hosts (Merino et al., 2010). Another theory has been that Boopiidae lice, including Australian *H. octoseriatus* (Figure 7), share a common ancestor with South American lice of marsupials (Barker, 1994). However, characters have been discovered to suggest a sister-group of bird lice and Boopiids and that the transfer of bird lice to marsupials creating the Boopiidae family is relatively recent (Barker, 1994). Many associations have been suggested for the genus *Macropodinium* (Figure 9). Coevolution and host switching have been observed while some speciation events are thought to have occurred due to vicariation in the Pleistocene, when marsupial populations split (Cameron and O'Donoghue, 2004). Another theory is that the associations of these ciliates with their hosts are not by co-descent but by resource tracking the diets of their hosts (Cameron and O'Donoghue, 2004). For each of these examples however, definite associations cannot be assumed until robust parasite and host phylogenies are formed.

To conclude, only a small number of the marsupial parasites investigated showed evidence of cospeciation but of these parasites, most were present in literature which reinforced their status as heirlooms. For the remainder of the parasites investigated, it was hard to establish their provenance as there was much variety in the taxonomic and geographic distances between themselves and the assumed close relatives. Perhaps most likely due to sampling error, further study would be benefitted by closing the gaps in our sequence databases.

Cameron, S. L., 2004. Morphometric and Cladistic Analyses of the Phylogeny of Macropodinium (Ciliophora: Litostomatea: Macropodiniidae). *Acta Protozoologica,* 43 (1), pp.43-53.

Gascoigne, I., 1998. *Papua New Guinea (Cultures of the World).* 2nd ed. New York: Benchmark Books.

Merino, S., Martinez, J., Vasquez, R. A. and Slapeta, J., 2010. Monophyly of Marsupial Intraerythrocytic Apicomplexan Parasites From South America and Australia. *Parasitology,* 137, pp.37-43.

Poulin, R., 2003. The decay of similarity with geographical distance in parasite communities of vertebrate hosts. *Journal of Biogeography,* 30 (10), pp.1609-1615.

4. Reflection by the student on the experience and value of the studentship (300 words max):

As a Zoology student hoping to forge a career in academia, I found the studentship provided by the HoCSLS an invaluable introduction to the world of research in the field of taxonomy.

Although the process of designing and carrying out successful experiments is prominent in the current curriculum, it is limited to the subject areas being taught at the time as well as the resources and time available during term time. By applying for the studentship, I was able to pursue an area of zoology that interested me and conduct a study over a reasonable timescale. I believe that this is very good experience for carrying out the fourth year honours project and beyond.

Another advantage that the studentship has brought has been the opportunity to learn new skills from an established researcher in the field, such as building and interpreting phylogenies. Such skills are not focused on in the practical elements of my course so by learning them through the studentship it has greatly added to my learning experience at the university.

To conclude, the HoCSLS studentship has been a very enjoyable experience that has allowed me to expand upon my current studies and has given me confidence to pursue a career in research once I graduate.

5. Dissemination: (note any presentations/publications submitted/planned from the work):

A full report will be published on Figshare in the near future

(http://figshare.com/authors/Aime%20Rankin/434982) .

6. Signatures:	Supervisor	Date	Student	Date
			Aime Rankin	22/07/2013

Appendix

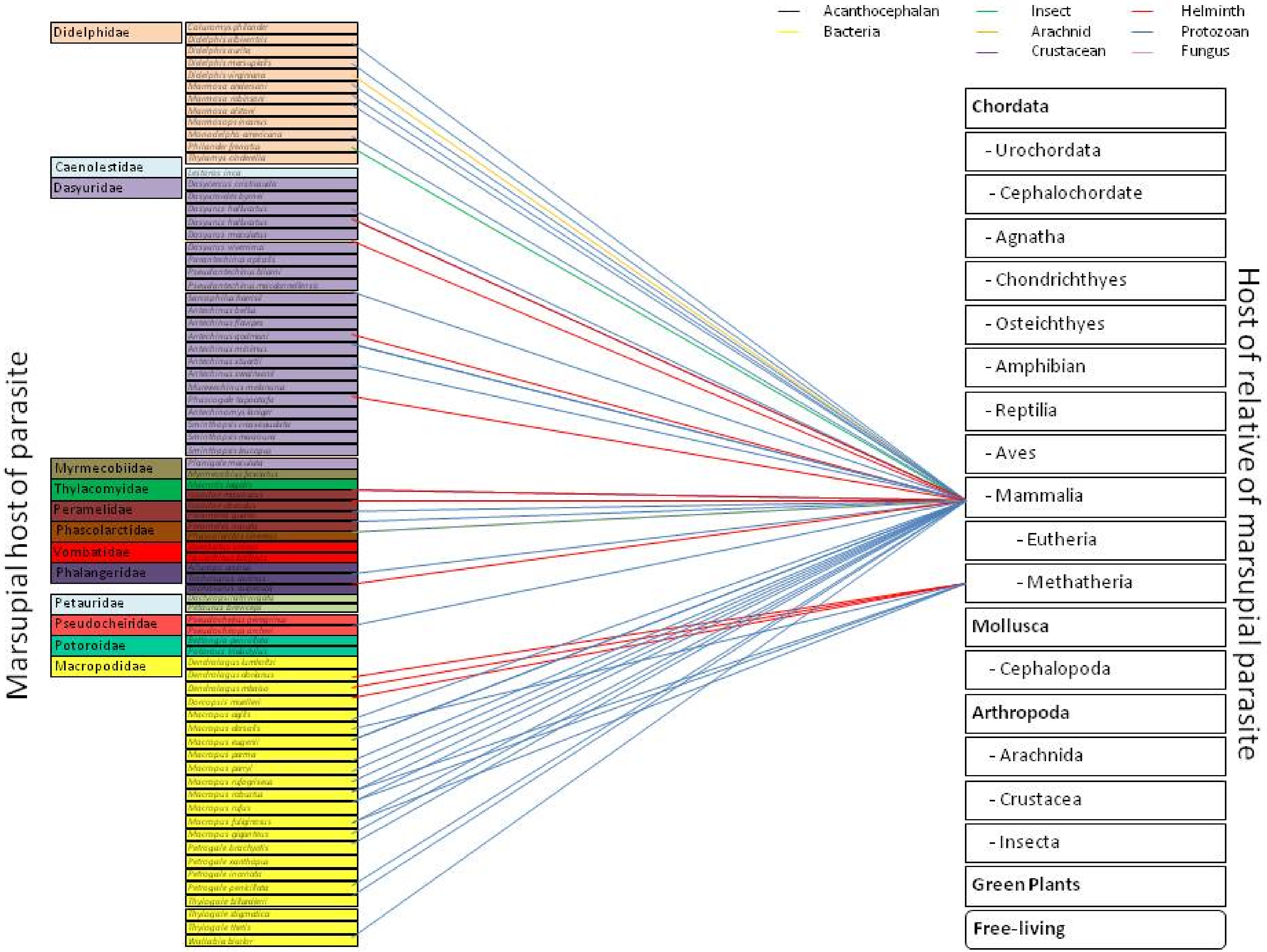

Figure 1: Diagram showing marsupial species and the hosts of the close relatives of their parasites. The colour of the line indicates the group to which the parasite belongs.

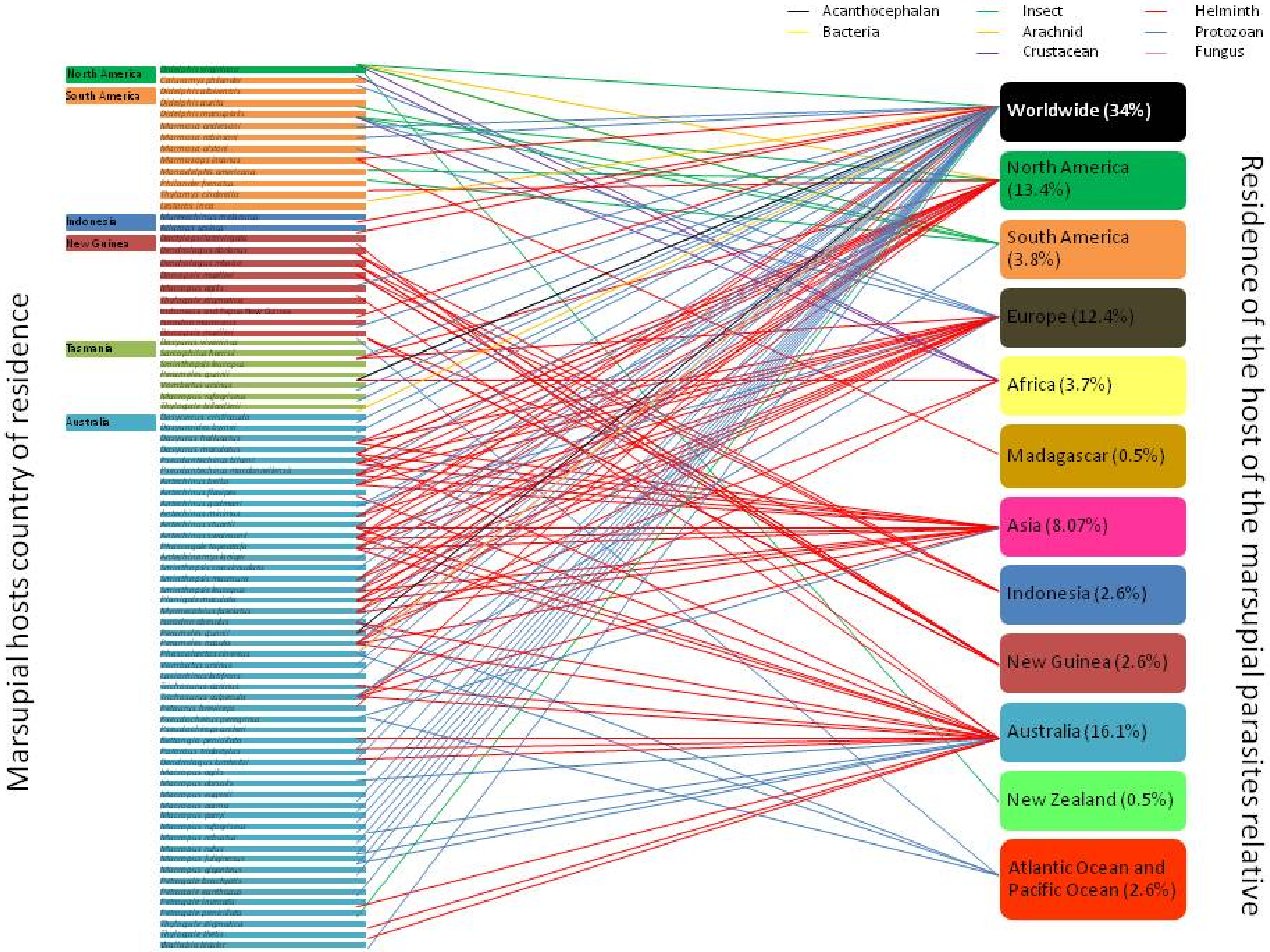

Figure 2: Diagram depicting the nationality of a marsupial and the country to which the host of their parasites closest relative lives. The colour of the line indicates the group to which the parasite belongs.

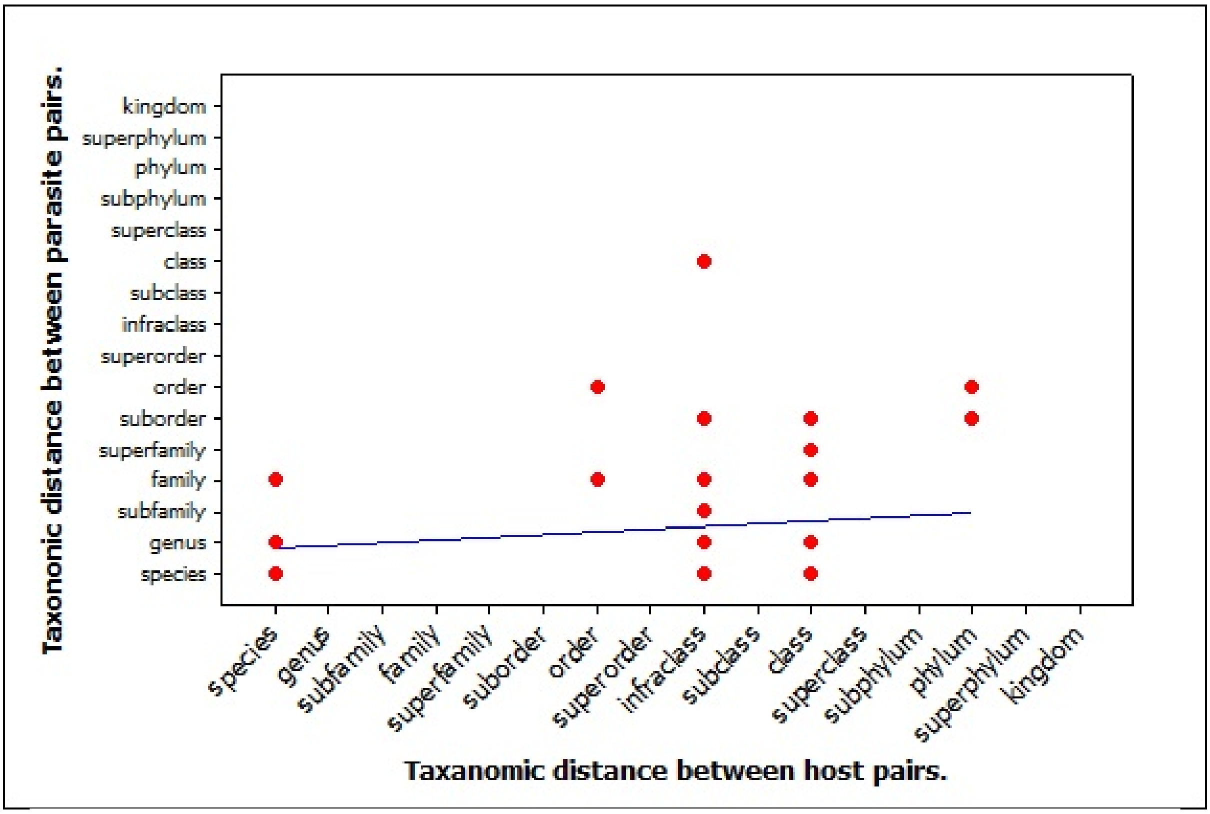

Figure 3: Graph depicting the taxonomic distance between marsupial parasites and their closest relatives, against the taxonomic distance between the marsupial host and the host of the parasites closest relative.

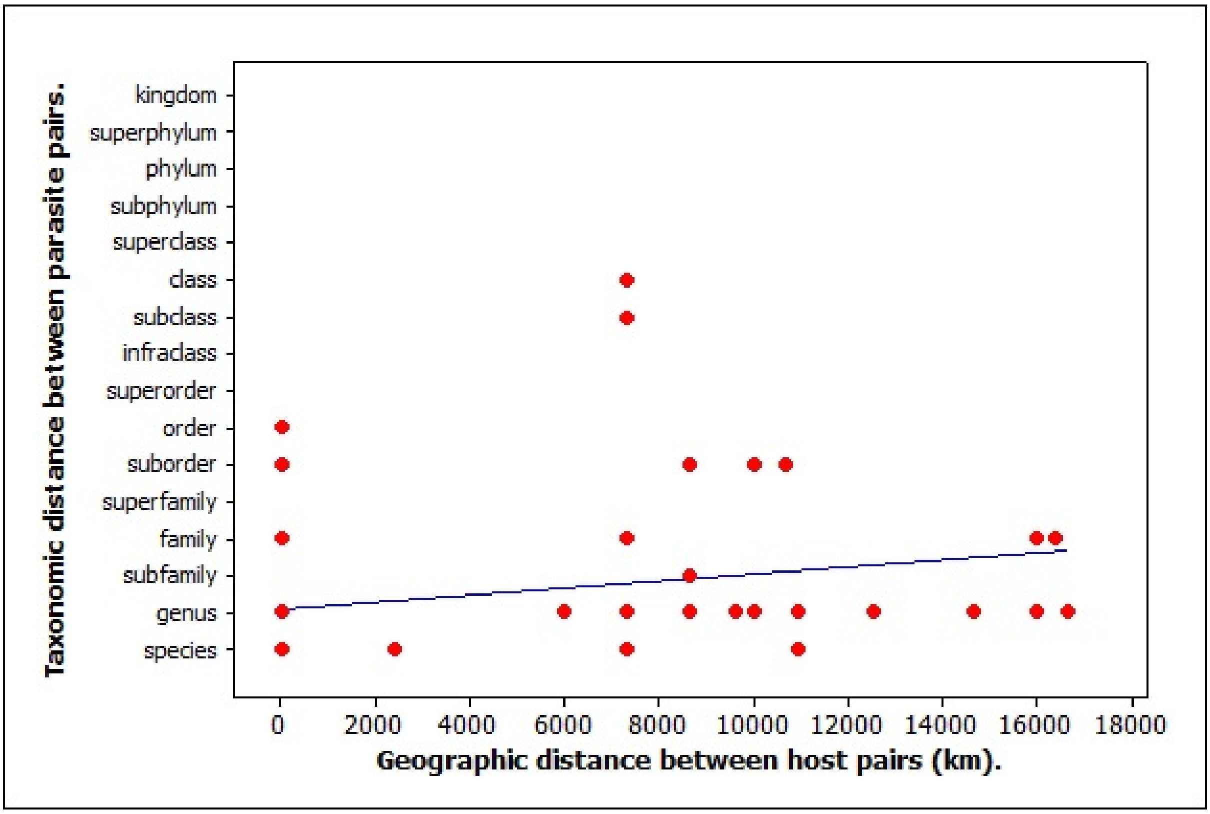

Figure 4: Graph depicting the taxonomic distance between marsupial parasites and their closest relatives, against the geographic distance between the marsupial host and the host of the parasites closest relative.

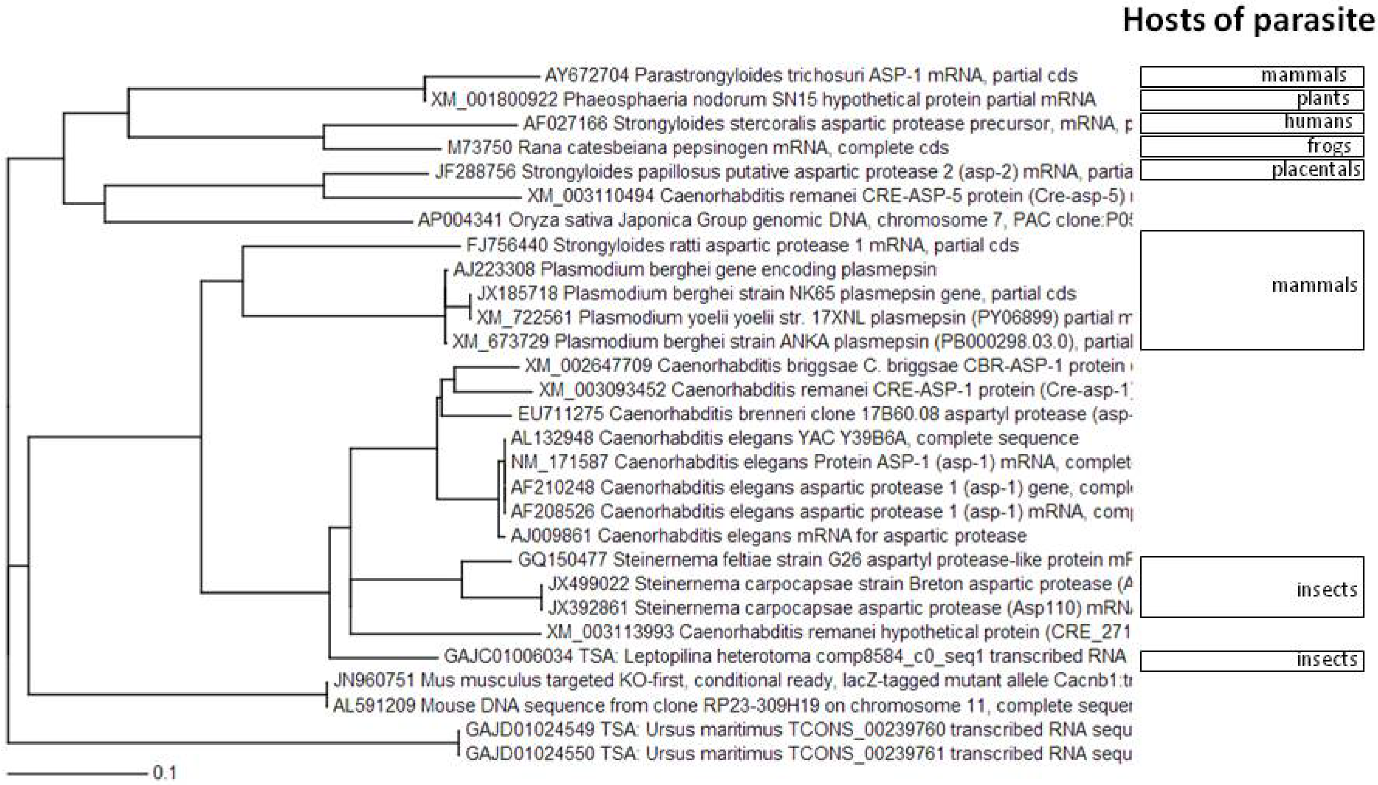

Figure 5: Phylogeny of *Parastrongyloides* generated by BLAST

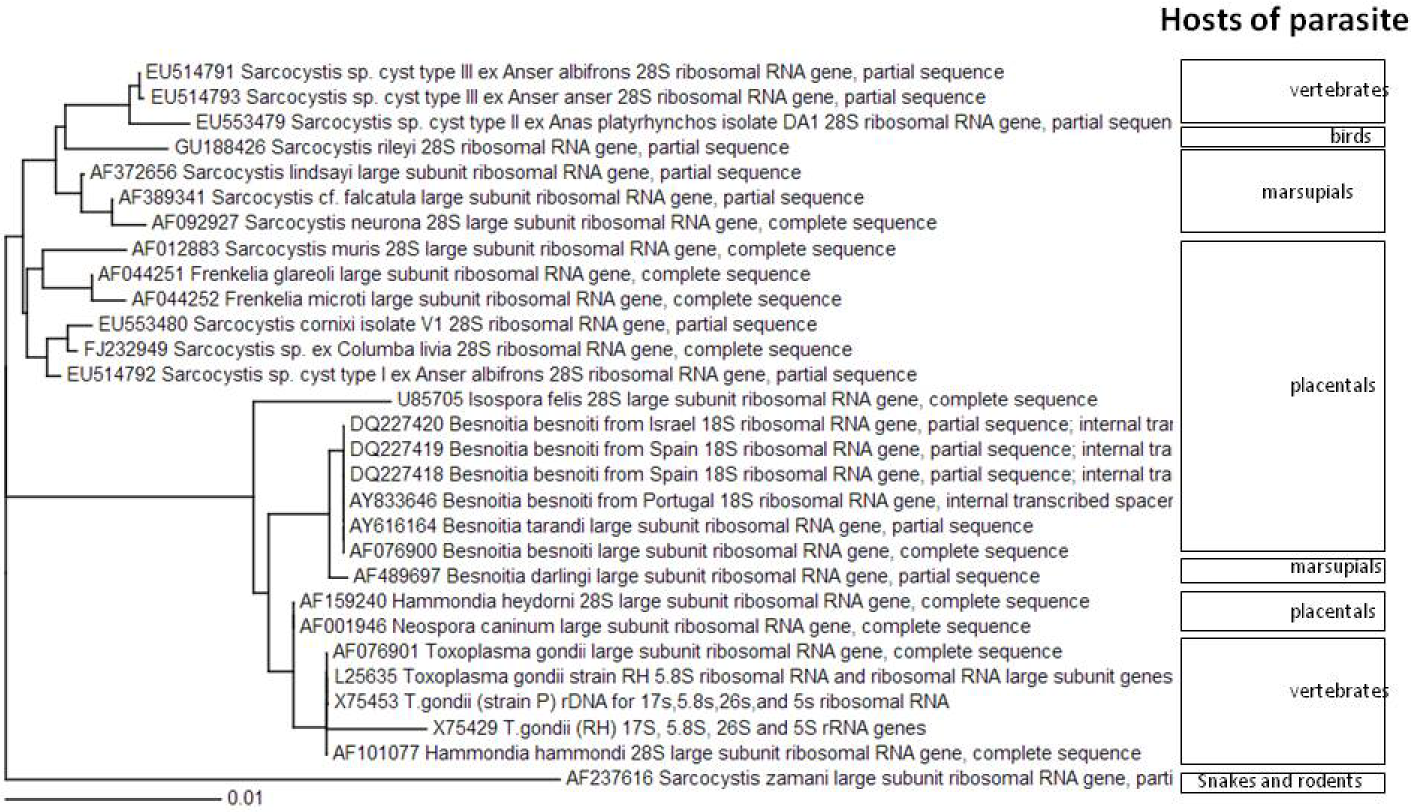

Figure 6: Phylogeny of *Sarcocystis* generated by BLAST.

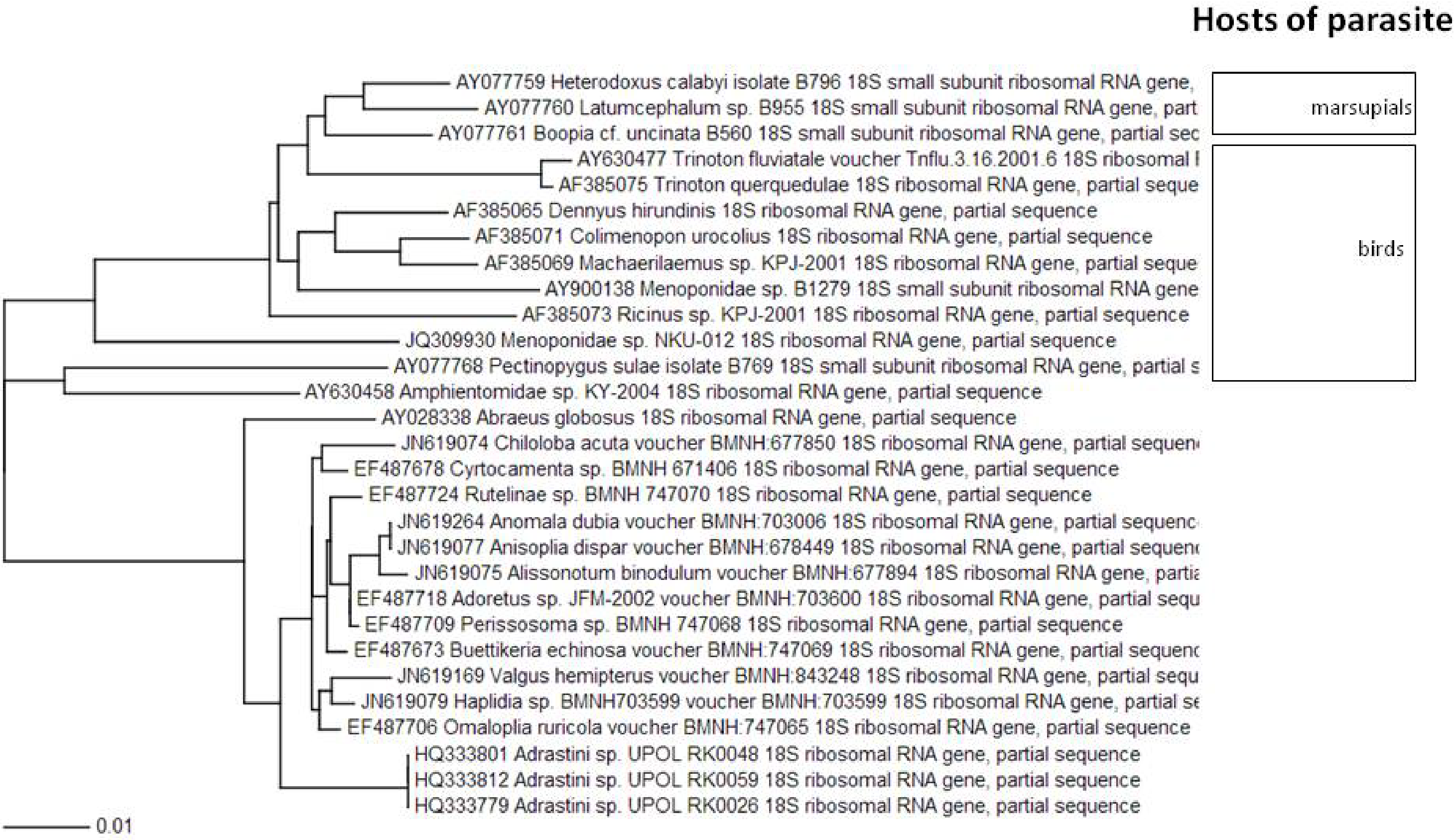

Figure 7: Phylogeny of *H. octoseriatus* generated by BLAST.

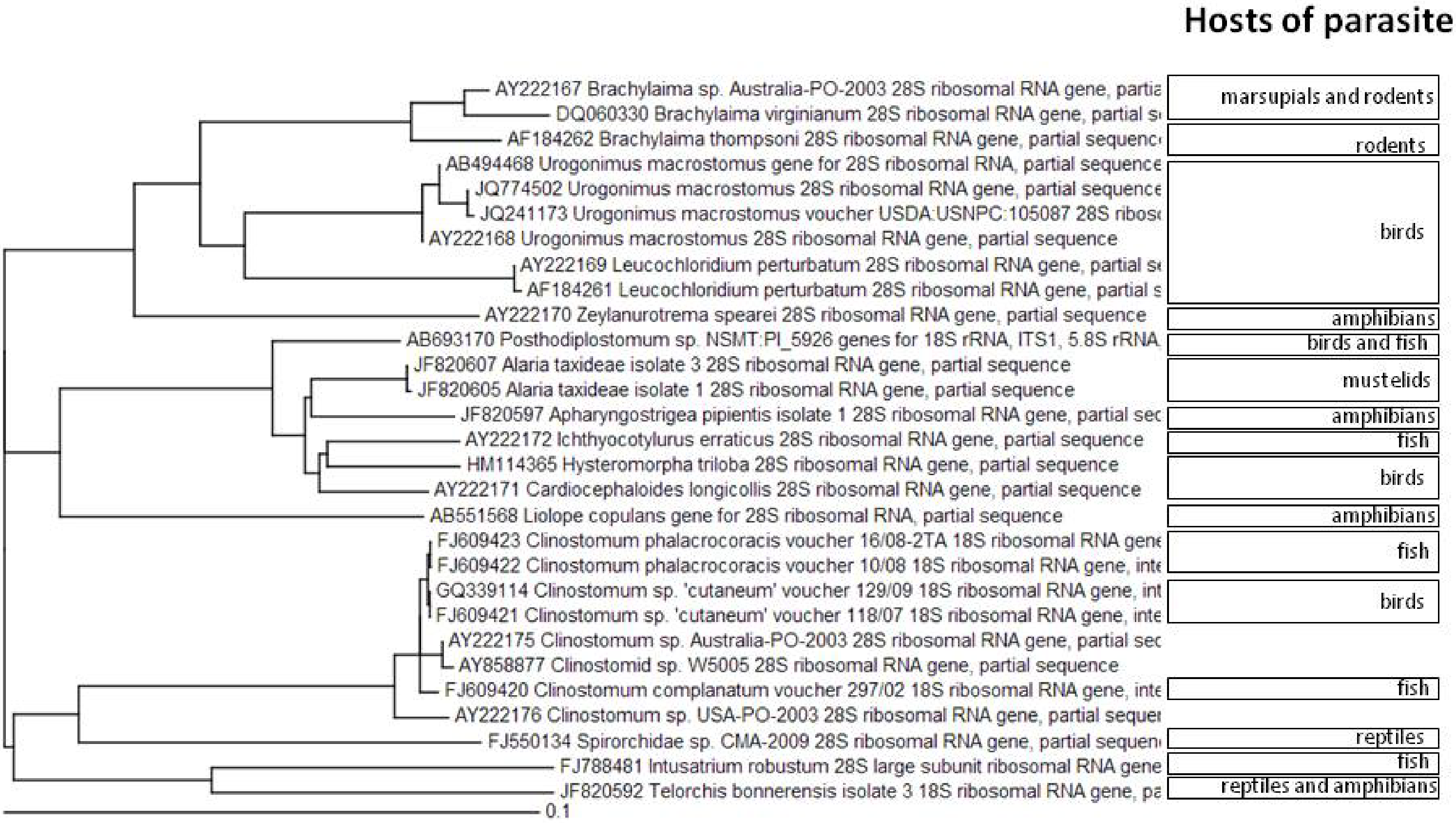

Figure 8: Phylogeny of *Brachylaima dasyuri* generated by BLAST.

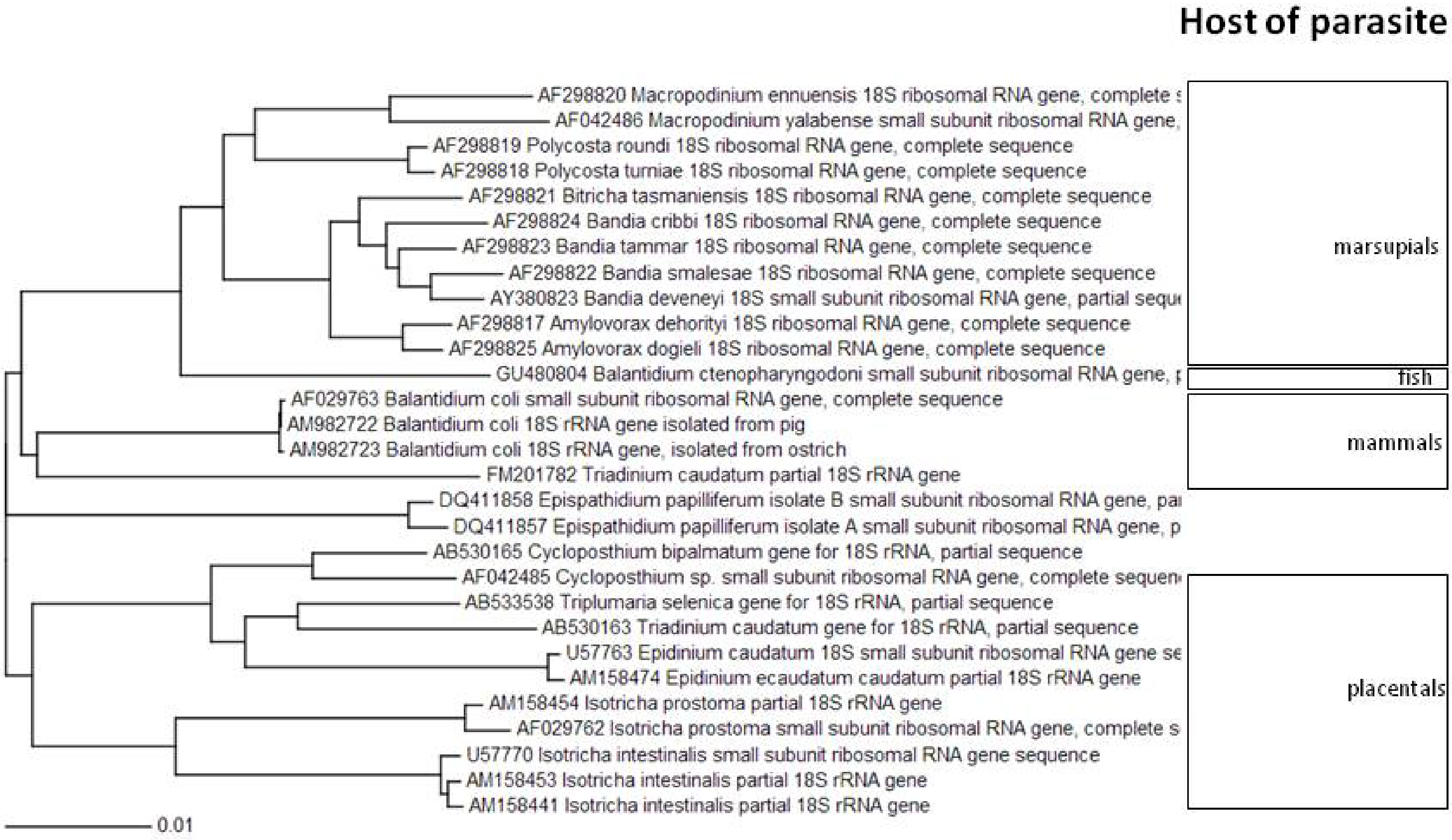

Figure 9: Phylogeny of *Macropodinium ennuensis* and *M. yalabense generated* by BLAST.