

Siphonophore Phylogeny

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Abstract

Introduction

Siphonophores are...

Methods

This manuscript is an executable document computed directly from the data, providing an explicit and reproducible description of all findings. All scripts for the analyses are available in a git repository at https://github.com/caseywdunn/siphonophore_phylogeny_2017. The most recent commit at the time of the analysis presented here was 4b88bf878524d97752471c3e564584a5c8b2041d.

Collecting

Collection data on all examined specimens, a description of the tissue that was sampled from the colony, collection mode, sample processing details, mRNA extraction methods, sequencing library preparation methods and sequencing details are summarized in supplementary table 1. Monterey Bay and Gulf of California specimens were collected by remotely operated underwater vehicle (ROV) or during blue-water scuba dives. *Chelophyes appendiculata* and *Hippopodius hippopus* specimen were collected in the bay of Villefranche-sur-Mer, France, during a plankton trawl on 04/13/11. Available physical vouchers have been deposited at the Museum of Comparative Zoology (Harvard University), Cambridge, MA, and at the United States National Museum (Smithsonian Institution), Washington, DC. Accession numbers are given in supplementary table X. In cases where physical vouchers were unavailable we provide photographs to document species identity (table x).

mRNA isolation and RNAseq library preparation

When possible specimens were starved overnight in filtered seawater at temperatures close to ambient water temperatures at the time point of specimen collection (supplementary table 1). mRNA was extracted directly from tissue using Magnetic mRNA Isolation Kit (NEB, #S1550S), Invitrogen Dynabeads mRNA Direct Kit (Ambion, #61011) or from total RNA after Trizol (Ambion, #15596026) extraction and through purification using Dynabeads mRNA Purification Kit (Ambion, #61006). Extractions were performed according to

the manufacturer’s instruction. In case of very small total RNA quantities, only a single round of bead purification was performed (supplementary table 1). Resulting higher rRNA read counts were dealt with further downstream in the bioinformatics workflow. Libraries were prepped for sequencing using the Illumina TruSeq RNA Sample Prep Kit (Illumina, #FC-122-1001, #FC-122-1002), the Illumina TruSeq Stranded Library Prep Kit (Illumina, #RS-122-2101) or the NEBNext RNA Sample Prep. Master Mix Set (NEB, #E6110S). We collected long read paired end Illumina data for *de novo* transcriptome assembly. Summary statistics for expression libraries are given in Table 1.

Analysis

Sequence assembly, annotation, phylogenetic analysis, and derivation of expression counts were conducted with the tool Agalma (Dunn et al. 2013), v0.XXX. Subsequent analyses were conducted in R and integrated into this manuscript with the `knitr` package. See Supplementary Information for R package version numbers.

Results and Discussion

Sample collecting and sequencing

Table 1 (at the end of file): Summary statistics for libraries.

Species phylogeny

Found more than one class "phylo" in cache; using the first, from namespace 'treeio'

Also defined by 'RNeXML'

Found more than one class "phylo" in cache; using the first, from namespace 'treeio'

Also defined by 'RNeXML'

The phylogenetic relationships between siphonophores species presented here is based on XXX genes from XXX siphonophore species, including new data presented here for XXX species. These findings are entirely consistent with a previous analysis based on two genes (16S and 18S ribosomal RNA) (Dunn et al. 2005). Cystonectae is the sister group to the remaining siphonophores, and Calyophorae is nested within the paraphyletic “Physonectae”. In addition, multiple nodes that were not resolved in the previous two-gene analysis do receive strong support in this XXX-gene transcriptome analysis. These findings include XXX.

Character Evolution

Siphonophores have evolved a fascinating diversity of morphological features, zooid types, life history traits, and habitats. Here we explore the evolutionary history of some of these features.

In our previous siphonophore phylogenetic analysis (Dunn et al. 2005) there were several characters left with equivocal evolutionary histories, due to unresolved relationships between physonects. With our current cladistic resolution, we were able to determine the following results:

Evolution of Monoecy

[Citation needed] noticed for the first time that some siphonophores were monoecious and others were dioecious. Our analyses in 2005 reconstructed this character and found a great amount of phylogenetic conservatism, with an unambiguous resolution of the MRCA as dioecious, and the appearance of monoecy in several taxa and clades (including Calyophorae) within the polytomy. Figure X shows the evolution of sex distribution in siphonophores under the current better-resolved tree model, and it strongly indicates that

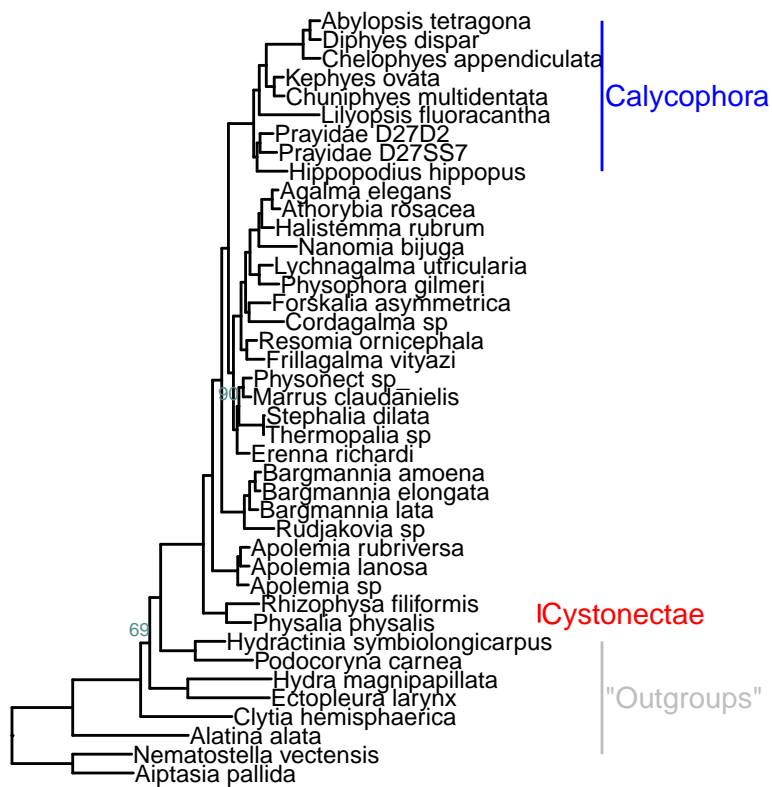


Figure 1: Phylogram of siphonophore relationships. Node labels indicate bootstrap support percent, unnumbered nodes have 100% support. The image was rendered with ggtree (Yu et al. 2016)

monoecy in siphonophores from a dioecious ancestor occurred twice, in the branch leading to Calyptophorae and in the branch leading to Agalmatids (sensu lato). There is a small probability for an alternative scenario featuring a single gain of monoecy before the split of Calyptophorae, with a subsequent derived shift back to dioecy in the *Marrus-Erenna* clade.

The Evolution of Zooid Types

One of the most striking aspects of siphonophore biology is their diversity of unique zooid types. Other colonial cnidarians (such as Hydractinia) and some bryozoans (example) have been found to have up to X different zooid types [Citation1, Citation 2]. The siphonophore genus *Forskalia* has 6 basic zooid types (pneumatophore, nectophore, gastrozooid, palpon, bract, and gonophore), and a total of 10 counting subtypes (4 types of bract, male & female gonophores). Diphyomorphs have more than 1 type of nectophore, while Cystonects have none. Here we reconstruct the evolutionary origins of the different zooid types and subtypes on the present transcriptome tree.

Nectophores are retained modified medusae Codonophora use for swimming. The nectosome is the region of the colony that develops from the nectosomal growth zone. Unlike the siphosomal growth zone, the nectosome does not bud gastrozooids, but nectophores (and in the case of *Apolemia*, also palpons). If fact, with the exception of *Physalia physalis* (which grows small nectophores near the gonodendra), siphonophore nectophores are exclusively found on the nectosome. It is likely that the MRCA of siphonophores had a nectosome, which has lost on the branch leading to Cystonects. We cannot exclude with certainty the alternative hypothesis of a nectosome-less ancestor followed by a gain of the nectosome in the branch leading to the Codonophora. The nectosome probably arose as a duplication of the siphosome, followed by functional specialization in propelling the colony. Figure X shows the origin of the dorsal nectosome in the Agalmatidae (sensu stricto) from an ancestral ventral nectosome.

All Codonophora have a nectosome, but the number and subtypes of nectophores present varies greatly between species. As shown in Figure X, most Codonophora presents the ancestral nectosome with multiple nectophores of the same subtype. However, Calyptophorans evolved a different system with just 2 nectophores of one type. This shift may be associated with the loss of the pneumatophore. Not all Calyptophorans remained with this arrangement. The Hippopodidae returned to bearing multiple identical nectophores, many of which are inactive and serve functions of defense (like a shell to retract in) and buoyancy. As in the rest of Calyptophorans, the Hippopodids only use 2 nectophores to propel the colony. Another interesting shift occurs in the branch leading to Diphyomorpha, where the 2 nectophores specialize into 2 subtypes, associated with a shift into a vertically aligned position and pointed bell shapes. The 2 types function together in a coupled hydrodynamic system that allows very fast escape responses (Mackie 1964).

Bracts also present a complex evolutionary history of subtypes (Figure X). Bracts are highly reduced zooids unique to siphonophores, but they are only present in the Codonophora. As with the nectosome, we have ambiguity determining whether the MRCA of siphonophores had bracts or not. The MRCA of Codonophora had only one bract subtype, which was lost in Hippopodidae and independently evolved into 2 subtypes in *Erenna* and *Nanomia*. During the branch leading to Forskalidae, bract subtypes evolved from 1 to 4 morphologically distinct forms. Bracts are important for protection of the delicate zooids and to help maintain neutral buoyancy. Some calyptophorans are able to actively regulate the Na/NH₄ balance to adjust their buoyancy (ref).

The ancestral siphonophore certainly had a pneumatophore (Figure X). This zooid type helps the colony float, maintain its pose, and regulate geotaxis (Church, 2013). Despite its diverse biological functions, it is lost in Calyptophorae and never gained again in that clade. Calyptophorans rely on the ionic balance of their gelatinous nectophores and bracts to retain posture and neutral buoyancy.

Palpons are modified mouthless gastrozooids used for digestion and circulation of the gastrovascular fluid. They were present in the MRCA of siphonophores (Figure X), retained in most species, but lost twice independently in the branches leading to *Bargmannia* and Calyptophorae. These taxa must have found other avenues to effectively circulate nutrients across the colony.

The Gain and Loss of Tentilla

The most complex nematocyst batteries of Cnidaria can arguably be found among the siphonophores, hanging in regularly spaced tentacle side branches called tentilla. Most hydrozoans, including the clade that contains siphonophores, bear simple tentacles (tentacles with no side branches). It is still an open question whether the most recent common ancestor (MRCA) of Siphonophora had simple or branched tentacles. The only two siphonophore genera regarded as lacking tentilla are *Physalia* and *Apolemia*, at each side of the earliest split in the siphonophore tree, with an immediate sister group with all members bearing tentilla (*Rhizophysidae* and *{Bargmannia, Diphyes}*) (FigXX). Figure X shows the evolution of this character on the current phylogeny, indicating a most-likely tentilla bearing MRCA, with two independent losses. However, this reconstruction bears some uncertainty on the state of the MRCA, suggesting a ~30% probability of 2 independent gains of tentilla in the branches leading to *Rhizophysidae* and *{Bargmannia, Diphyes}*.

A key issue here is how we code for absence of tentilla, especially for the case of *Physalia physalis*. The tentacles of this species, when uncoiled, show very prominent, evenly spaced, 3D buttons which contain all active and functionally arranged nematocysts used by the organism for prey capture. Siphonophore tentilla are complete diverticular branchings of the tentacle ectoderm, mesoglea, and gastrovascular canal (lined by endoderm). Hessinger & Ford 1988 (in the Biology of Nematocysts) described these bands as enclosing individual fluid-filled chambers connected by narrow channels to the tentacular canal, lined by endoderm. This suggests they are not just ectodermal swellings, but probably are reduced tentilla. When we code *Physalia physalis* as tentilla bearing, the results for the character reconstruction change to a more parsimonious scenario (Figure X), indicating there was a single loss of tentilla in the branch leading to *Apolemiidae*. This result suggests an unambiguous tentilla-bearing state of the MRCA.

The Evolution of Vertical Habitat Use

Siphonophores are abundant predators in the pelagic realm, ranging from the surface (*Physalia physalis*) to bathypelagic depths (ref , *Bargmannia sp* 3888m VARS unpublished). While there are some pleustonic (*Physalia*) and benthic (*Rhodaliidae*) siphonophores, the phylogeny suggests the siphonophore MRCA was planktonic, as most extant taxa are. Some interesting questions arise from these facts, including 1) what was the bathymetric niche of the siphonophore MRCA, and 2) how did siphonophore's vertical habitat use of the water columns evolve along the phylogeny. Our results indicate a mesopelagic MRCA, with several convergent transition events to epipelagic and bathypelagic waters. There was only a single transition to benthic lifestyle on the stem of *Rhodaliidae*.

Discussion

The strong phylogenetic signal in the characters traditionally used for taxonomic diagnostics is a positive indicator of the applicability and unambiguity of these characters.

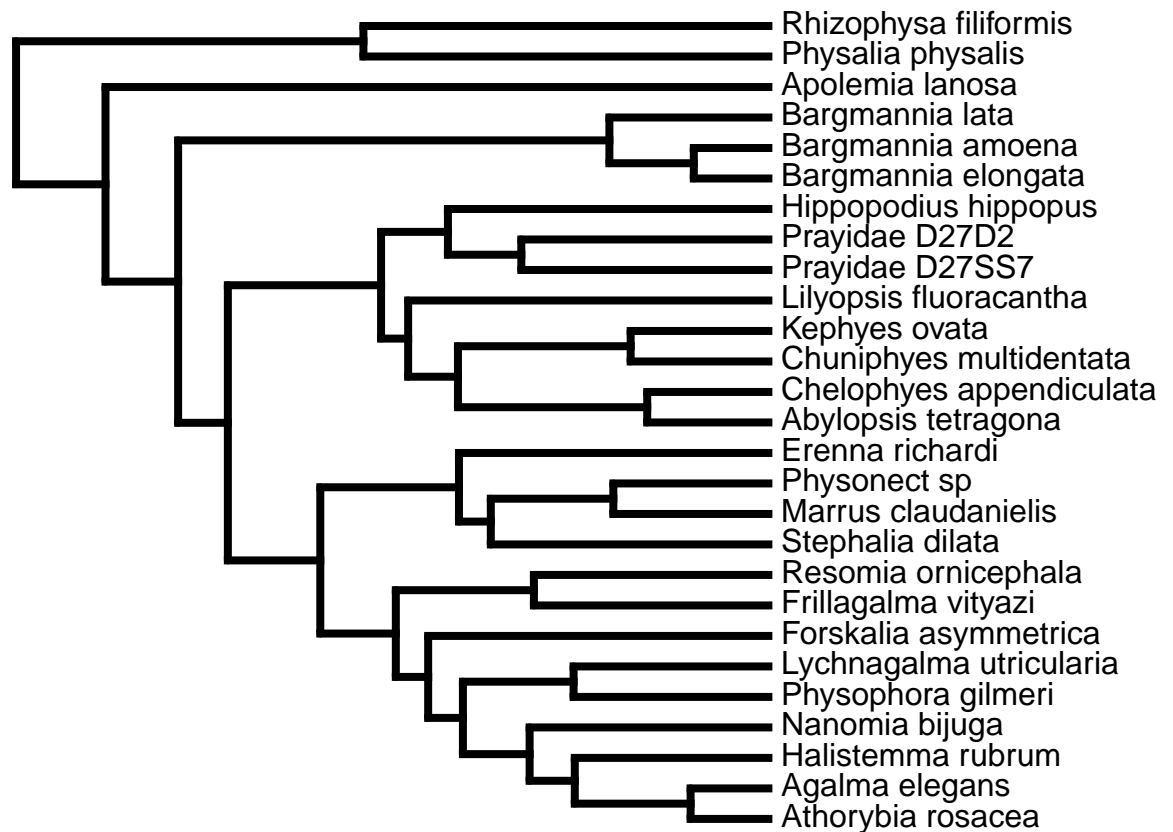
Conclusions

Acknowledgements

This work was supported by the National Science Foundation (DEB-1256695 and the Waterman Award). Sequencing at the Brown Genomics Core facility was supported in part by NIH P30RR031153 and NSF EPSCoR EPS-1004057. Data transfer was supported by NSF RII-C2 EPS-1005789. Analyses were conducted with computational resources and services at the Center for Computation and Visualization at Brown University, supported in part by the NSF EPSCoR EPS-1004057 and the State of Rhode Island. We also thank the MBARI crews and ROV pilots for collection of the specimens.

Supplementary Information

Supplementary Analyses



The Tree:

Stochastic trait mapping:

```
## [1] "SIMMAP Sex Distribution"

## make.simmap is sampling character histories conditioned on the transition matrix
##
## Q =
##      Dioecious Monoecious
## Dioecious -0.2043091  0.2043091
## Monoecious  0.2043091 -0.2043091
## (estimated using likelihood);
## and (mean) root node prior probabilities
## pi =
##      Dioecious Monoecious
##      0.5        0.5

## [1] "SIMMAP Palpons"

## make.simmap is sampling character histories conditioned on the transition matrix
##
## Q =
##      0        1
## 0 -0.1915304  0.1915304
## 1  0.1915304 -0.1915304
```

```

## (estimated using likelihood);
## and (mean) root node prior probabilities
## pi =
## 0 1
## 0.5 0.5

## [1] "SIMMAP Tentilla"

## make.simmap is sampling character histories conditioned on the transition matrix
##
## Q =
## 0 1
## 0 -0.1876166 0.1876166
## 1 0.1876166 -0.1876166
## (estimated using likelihood);
## and (mean) root node prior probabilities
## pi =
## 0 1
## 0.5 0.5

## [1] "SIMMAP Tentilla2"

## make.simmap is sampling character histories conditioned on the transition matrix
##
## Q =
## 0 1
## 0 -0.08687465 0.08687465
## 1 0.08687465 -0.08687465
## (estimated using likelihood);
## and (mean) root node prior probabilities
## pi =
## 0 1
## 0.5 0.5

## [1] "SIMMAP Nectosome"

## make.simmap is sampling character histories conditioned on the transition matrix
##
## Q =
## 0 1
## 0 -0.09152961 0.09152961
## 1 0.09152961 -0.09152961
## (estimated using likelihood);
## and (mean) root node prior probabilities
## pi =
## 0 1
## 0.5 0.5

## [1] "SIMMAP Nectosome Position"

## make.simmap is sampling character histories conditioned on the transition matrix
##
## Q =
## Dorsal None Ventral
## Dorsal -0.1011359 0.0000000 0.1011359
## None 0.0000000 -0.1011024 0.1011024
## Ventral 0.1011359 0.1011024 -0.2022383
## (estimated using likelihood);

```

```

## and (mean) root node prior probabilities
## pi =
##      Dorsal      None      Ventral
## 0.3333333 0.3333333 0.3333333

## [1] "SIMMAP Nectophore Types"

## make.simmap is sampling character histories conditioned on the transition matrix
##
## Q =
##           Multiple of one type      None Two of one type
## Multiple of one type      -0.5519187  0.5519187      0.000000
## None                      0.5519187 -3.5583410      2.457982
## Two of one type           0.0000000  2.4579816     -2.457982
## Two of two types          0.0000000  0.5484407      0.000000
##           Two of two types
## Multiple of one type      0.0000000
## None                      0.5484407
## Two of one type           0.0000000
## Two of two types          -0.5484407
## (estimated using likelihood);
## and (mean) root node prior probabilities
## pi =
## Multiple of one type      None      Two of one type
##           0.25              0.25              0.25
##      Two of two types
##           0.25

## [1] "SIMMAP Pneumatophore"

## make.simmap is sampling character histories conditioned on the transition matrix
##
## Q =
##           0      1
## 0 -0.0871947  0.0871947
## 1  0.0871947 -0.0871947
## (estimated using likelihood);
## and (mean) root node prior probabilities
## pi =
##      0      1
## 0.5 0.5

## [1] "SIMMAP Bract Types"

## make.simmap is sampling character histories conditioned on the transition matrix
##
## Q =
##           0      1      2      4
## 0 -0.3301839  0.3301839  0.0000000  0.000000
## 1  0.3301839 -0.7921012  0.4619173  0.000000
## 2  0.0000000  0.4619173 -2.0898685  1.627951
## 4  0.0000000  0.0000000  1.6279512 -1.627951
## (estimated using likelihood);
## and (mean) root node prior probabilities
## pi =
##      0      1      2      4
## 0.25 0.25 0.25 0.25

```

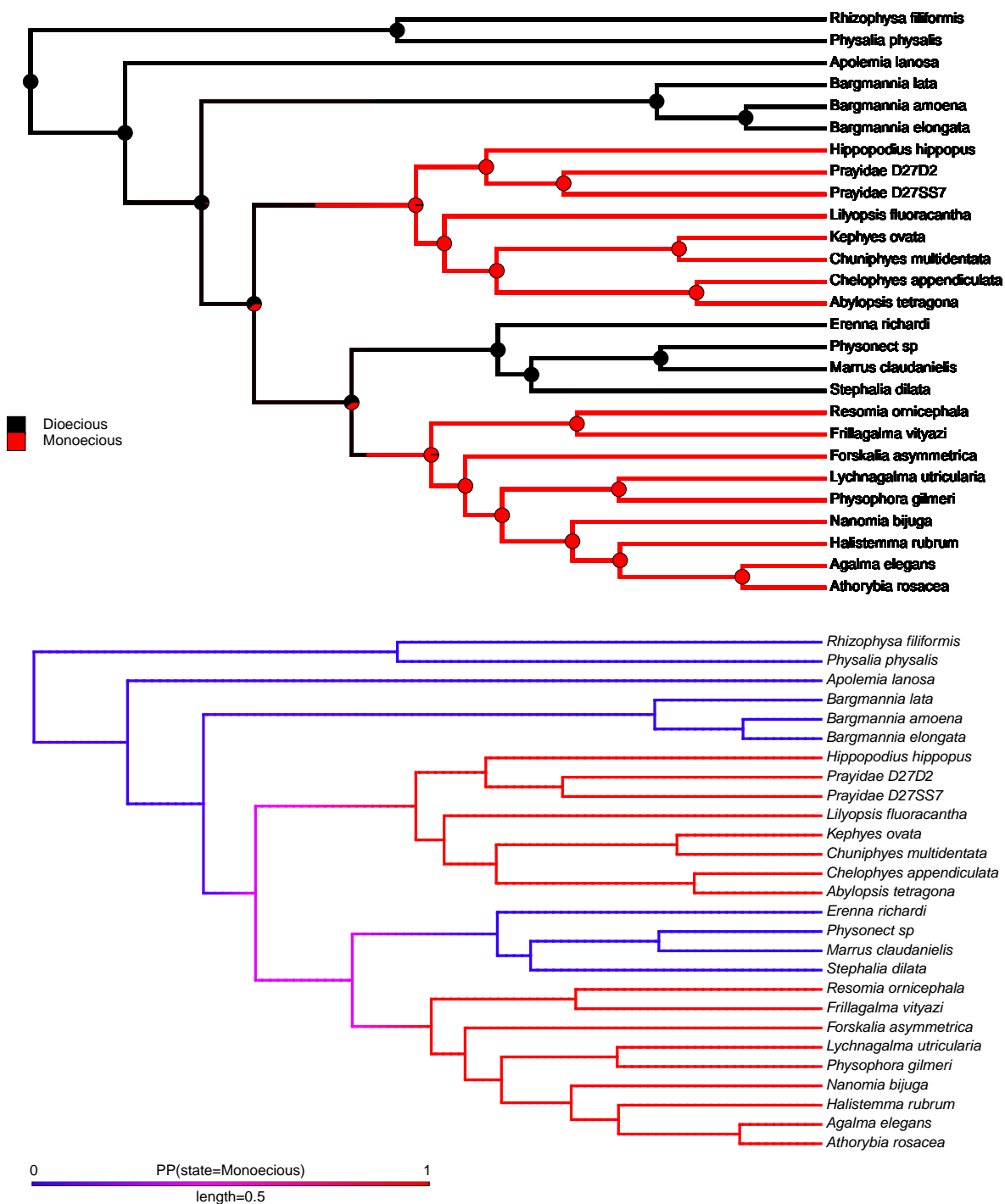


```
## [1] "SIMMAP Habitat"

## make.simmap is sampling character histories conditioned on the transition matrix
##
## Q =
##           Bathypelagic Deep benthic Epipelagic Mesopelagic   Surface
## Bathypelagic -1.0094091   0.5464764   0.0000000   0.4629328   0.000000
## Deep benthic  0.5464764  -0.5464764   0.0000000   0.0000000   0.000000
## Epipelagic    0.0000000   0.0000000  -2.115283   1.7792722   0.336011
## Mesopelagic   0.4629328   0.0000000   1.779272  -2.2422050   0.000000
## Surface       0.0000000   0.0000000   0.336011   0.0000000  -0.336011
## (estimated using likelihood);
## and (mean) root node prior probabilities
## pi =
## Bathypelagic Deep benthic   Epipelagic   Mesopelagic       Surface
##           0.2           0.2           0.2           0.2           0.2
```

SIMMAP Sex distribution:

[illegible]



Posterior probabilities of states (0 Dioecious, 1 Monoecious).

SIMMAP Presence of palpons:

no colors provided. using the following legend:

0 1

"black" "red"

no colors provided. using the following legend:

[illegible]

[illegible]

[illegible]

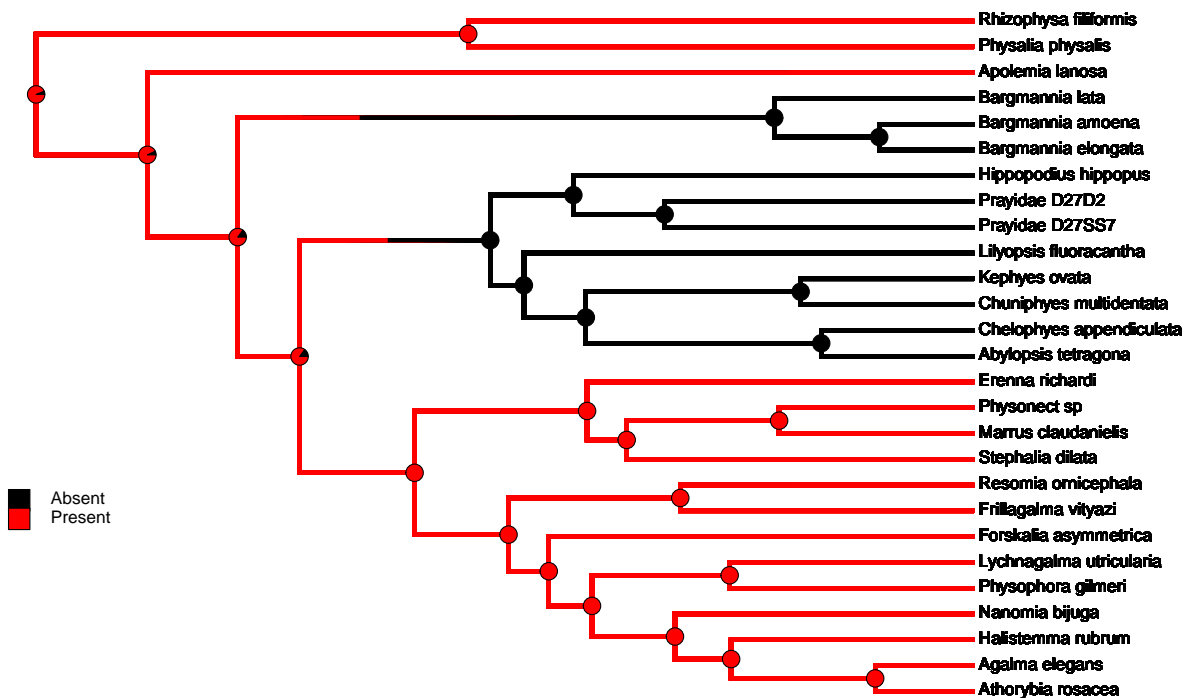
[illegible]

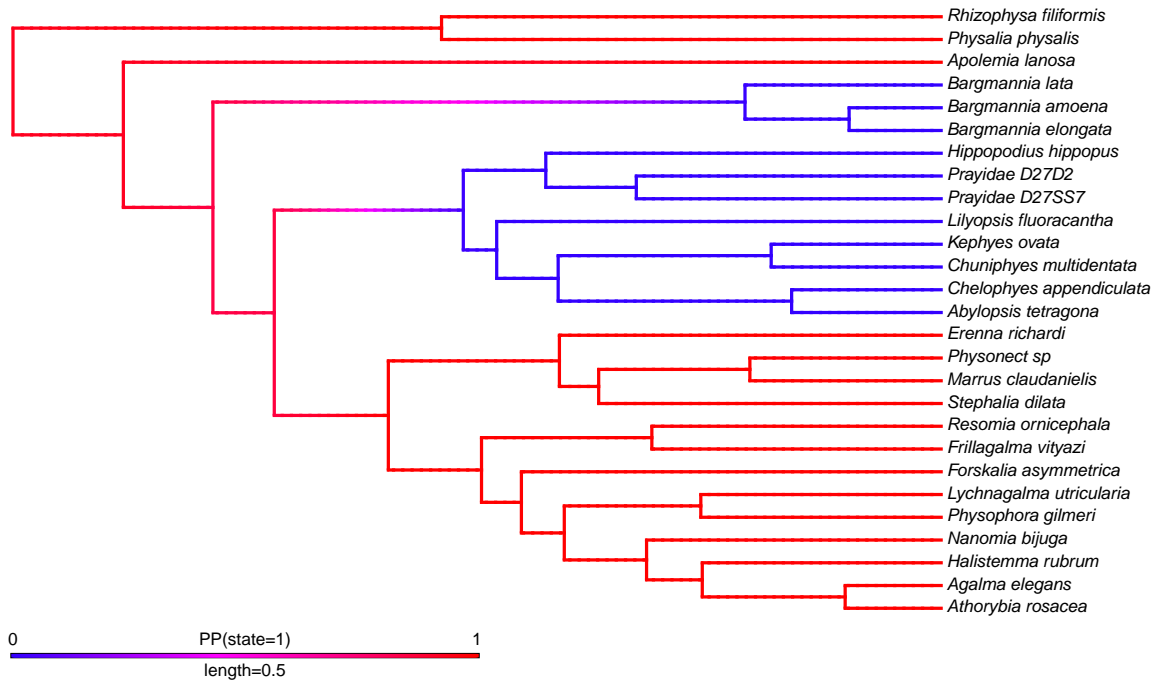
[illegible]

```

##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"

```





Posterior probabilities of states (1 Present, 0 Absent).

SIMMAP Presence of tentilla:

```
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
```

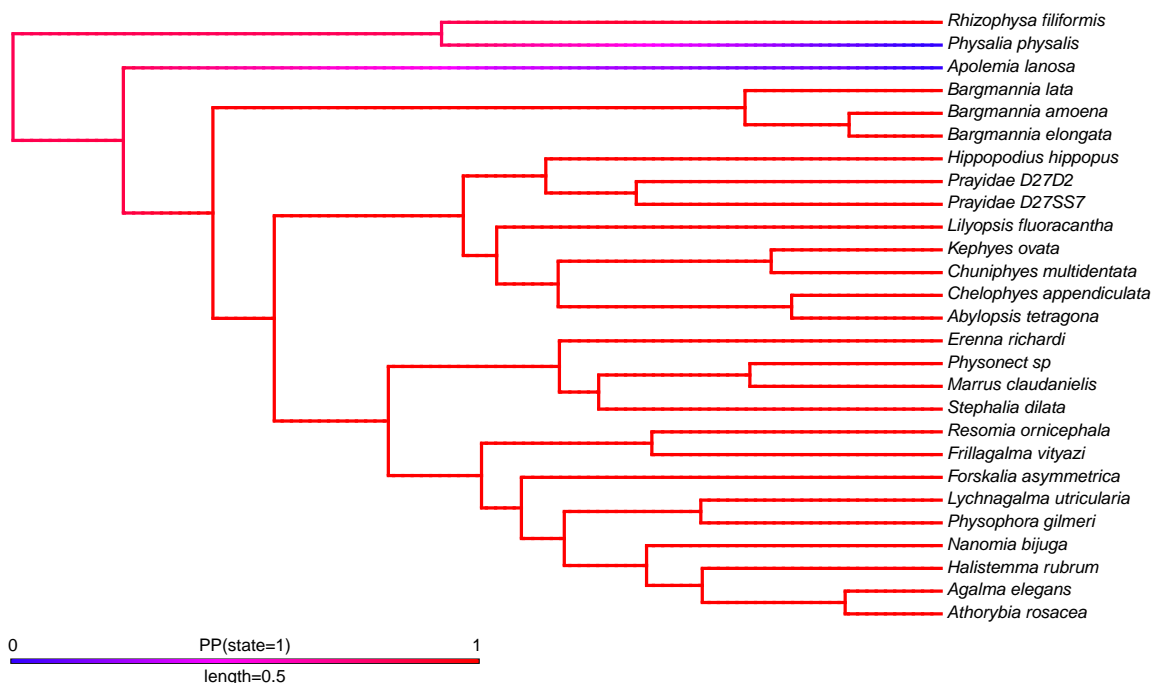
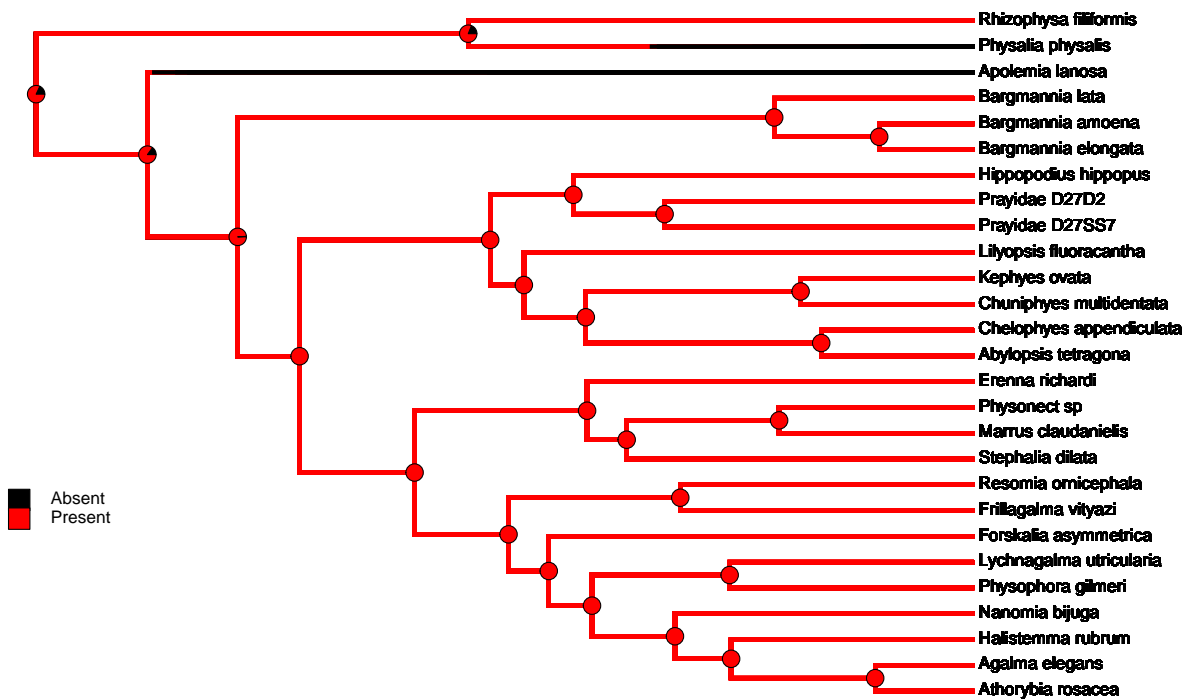
[illegible]

[illegible]

[illegible]

[illegible]

```
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
```



Posterior probabilities of states (1 Present, 0 Absent).

Physalia physalis may have reduced tentilla, which would indicate there was only one loss of tentilla at the branch leading to *Apolemia*.

SIMMAP Presence of tentilla - *Physalia* corrected:

[illegible]

[illegible]

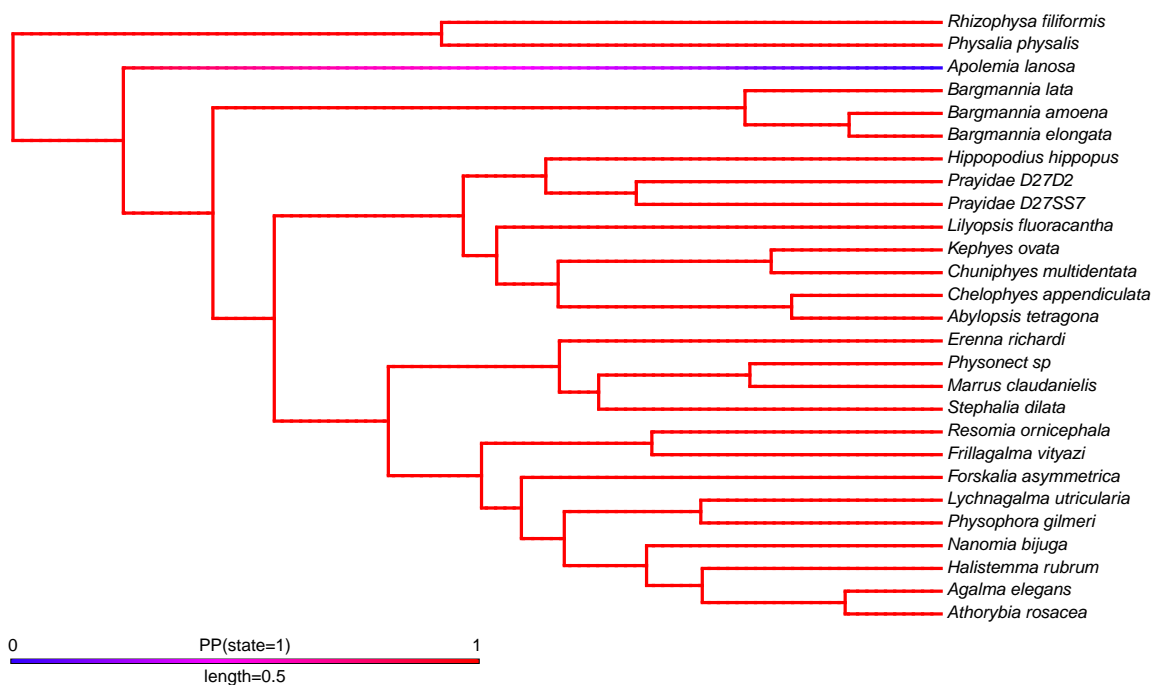
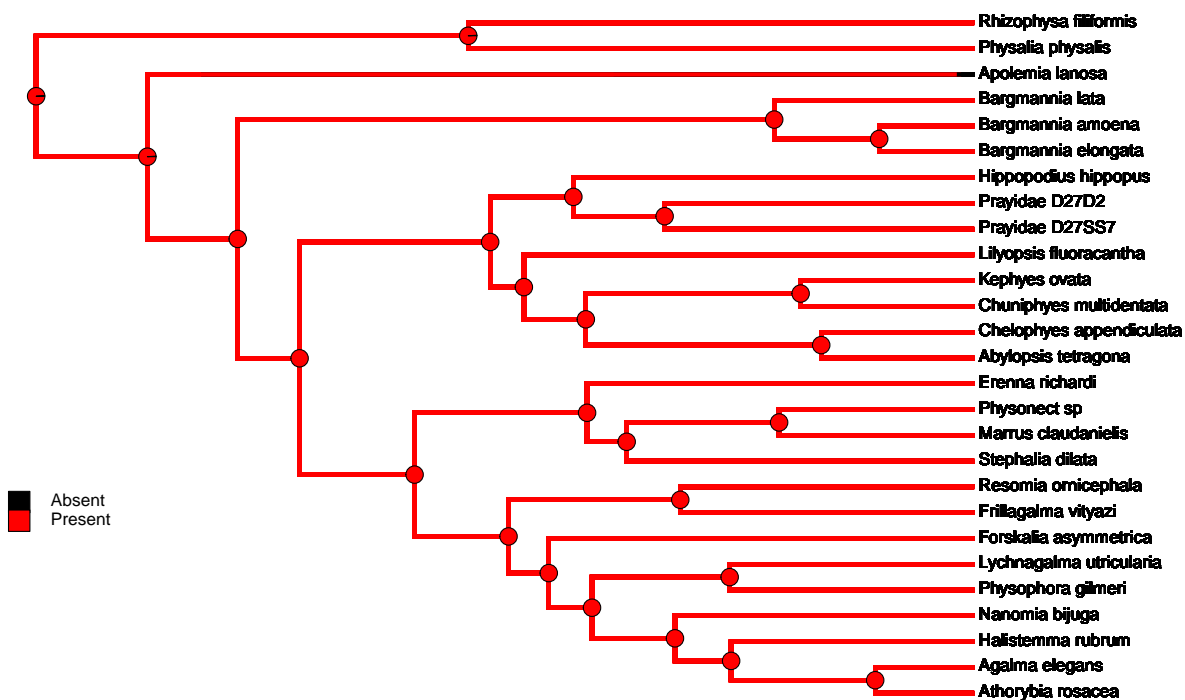
[illegible]

[illegible]

```

##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"

```



Posterior probabilities of states (1 Present, 0 Absent).

SIMMAP Presence of Nectophores:

```
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
```

[illegible]

[illegible]

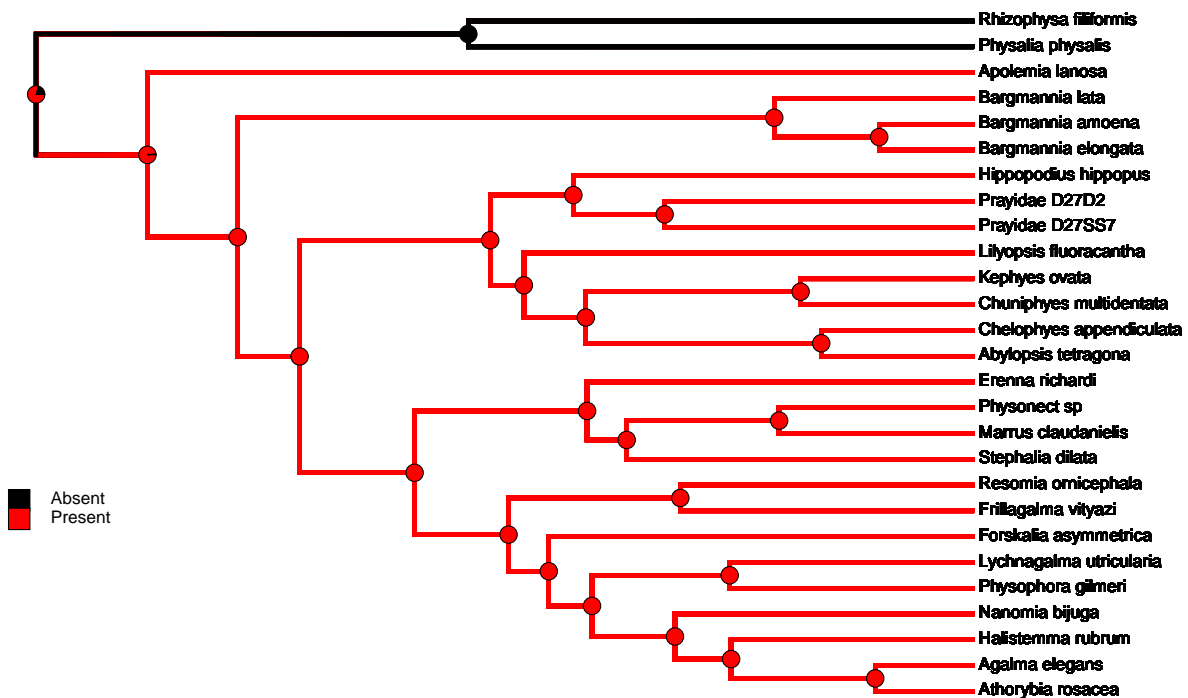
[illegible]

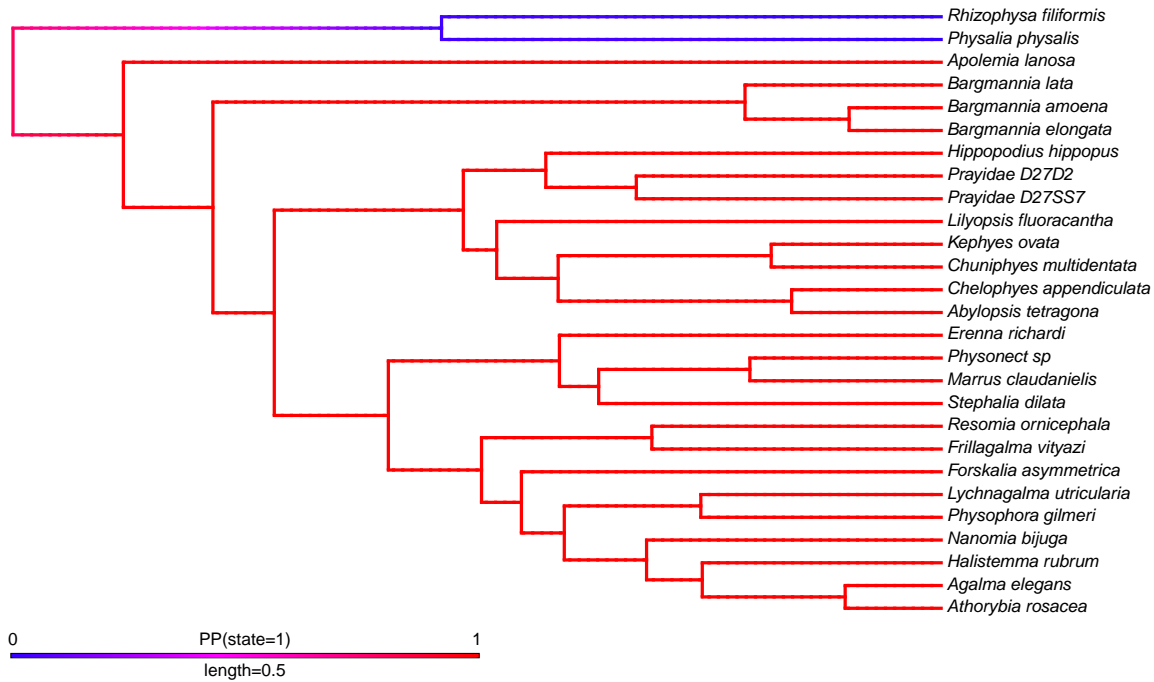
[illegible]


```

##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"

```





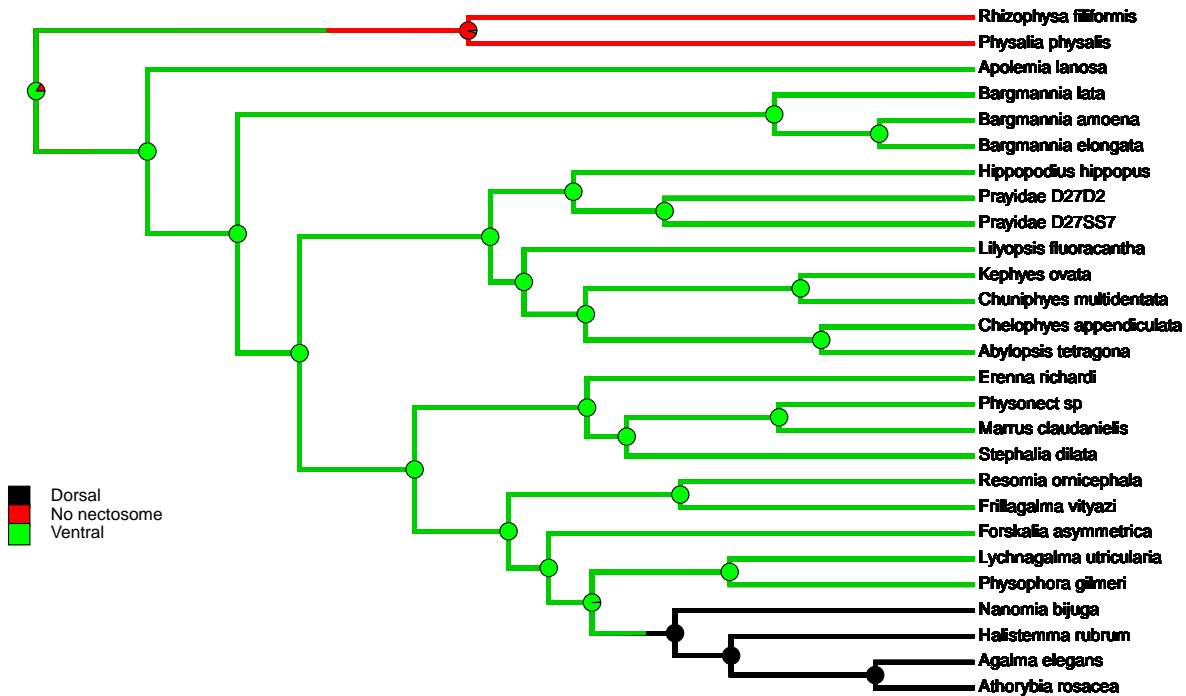
Posterior probabilities of states (1 Present, 0 Absent).

SIMMAP Position of Nectosome:

```
## no colors provided. using the following legend:
## Dorsal None Ventral
## "black" "red" "green3"
## no colors provided. using the following legend:
## Dorsal None Ventral
## "black" "red" "green3"
## no colors provided. using the following legend:
## Dorsal None Ventral
## "black" "red" "green3"
## no colors provided. using the following legend:
## Dorsal None Ventral
## "black" "red" "green3"
## no colors provided. using the following legend:
## Dorsal None Ventral
## "black" "red" "green3"
## no colors provided. using the following legend:
## Dorsal None Ventral
## "black" "red" "green3"
## no colors provided. using the following legend:
## Dorsal None Ventral
## "black" "red" "green3"
## no colors provided. using the following legend:
## Dorsal None Ventral
## "black" "red" "green3"
```


[illegible]


```
## no colors provided. using the following legend:
##   Dorsal      None  Ventral
##   "black"     "red" "green3"
```



SIMMAP Nectophore number and types:

```
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##   "black"                 "red"      "green3"
##   Two of two types
##   "blue"

## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##   "black"                 "red"      "green3"
##   Two of two types
##   "blue"

## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##   "black"                 "red"      "green3"
##   Two of two types
##   "blue"

## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##   "black"                 "red"      "green3"
##   Two of two types
##   "blue"
```



```

## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"

```

[illegible]

```

##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##      "black"              "red"      "green3"
##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##      "black"              "red"      "green3"
##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##      "black"              "red"      "green3"
##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##      "black"              "red"      "green3"
##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##      "black"              "red"      "green3"
##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##      "black"              "red"      "green3"
##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##      "black"              "red"      "green3"
##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##      "black"              "red"      "green3"
##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type

```

```

##          "black"          "red"          "green3"
##      Two of two types
##          "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##          "black"          "red"          "green3"
##      Two of two types
##          "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##          "black"          "red"          "green3"
##      Two of two types
##          "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##          "black"          "red"          "green3"
##      Two of two types
##          "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##          "black"          "red"          "green3"
##      Two of two types
##          "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##          "black"          "red"          "green3"
##      Two of two types
##          "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##          "black"          "red"          "green3"
##      Two of two types
##          "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##          "black"          "red"          "green3"
##      Two of two types
##          "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##          "black"          "red"          "green3"
##      Two of two types
##          "blue"
## no colors provided. using the following legend:

```

```

## Multiple of one type          None      Two of one type
##         "black"                "red"        "green3"
##     Two of two types
##         "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##         "black"                "red"        "green3"
##     Two of two types
##         "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##         "black"                "red"        "green3"
##     Two of two types
##         "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##         "black"                "red"        "green3"
##     Two of two types
##         "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##         "black"                "red"        "green3"
##     Two of two types
##         "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##         "black"                "red"        "green3"
##     Two of two types
##         "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##         "black"                "red"        "green3"
##     Two of two types
##         "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##         "black"                "red"        "green3"
##     Two of two types
##         "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##         "black"                "red"        "green3"
##     Two of two types
##         "blue"

```

```

## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##           "black"              "red"      "green3"
##           Two of two types
##           "blue"

```

[illegible]

```

##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##      "black"              "red"      "green3"
##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##      "black"              "red"      "green3"
##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##      "black"              "red"      "green3"
##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##      "black"              "red"      "green3"
##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##      "black"              "red"      "green3"
##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##      "black"              "red"      "green3"
##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##      "black"              "red"      "green3"
##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type
##      "black"              "red"      "green3"
##      Two of two types
##      "blue"
## no colors provided. using the following legend:
## Multiple of one type      None      Two of one type

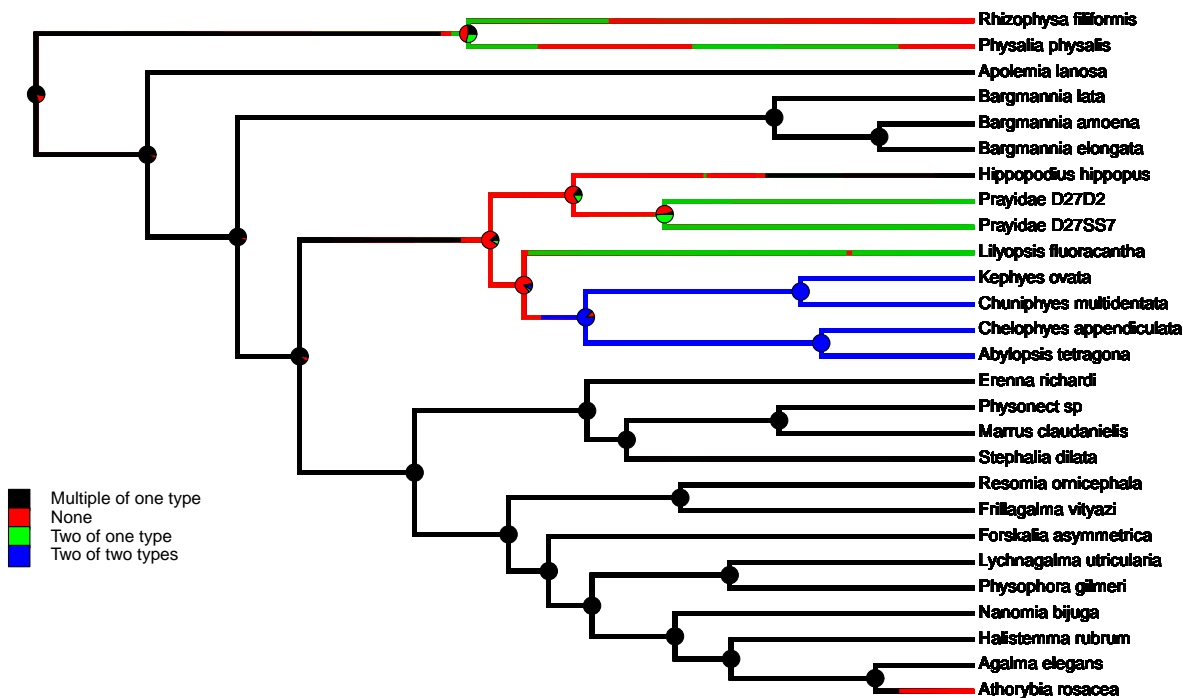
```



```

##          "black"          "red"          "green3"
##      Two of two types
##          "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##          "black"          "red"          "green3"
##      Two of two types
##          "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##          "black"          "red"          "green3"
##      Two of two types
##          "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##          "black"          "red"          "green3"
##      Two of two types
##          "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##          "black"          "red"          "green3"
##      Two of two types
##          "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##          "black"          "red"          "green3"
##      Two of two types
##          "blue"
## no colors provided. using the following legend:
## Multiple of one type          None      Two of one type
##          "black"          "red"          "green3"
##      Two of two types
##          "blue"

```



SIMMAP Presence of Pneumatophore:

```
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
## no colors provided. using the following legend:
##      0      1
## "black"  "red"
```

[illegible]

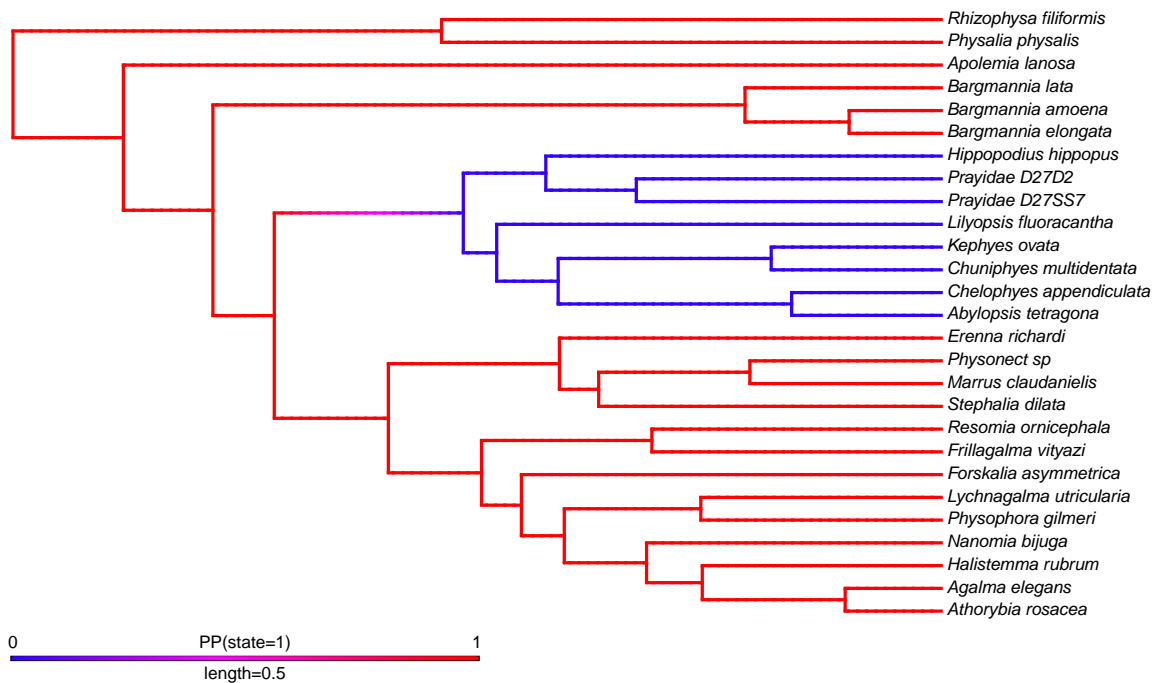
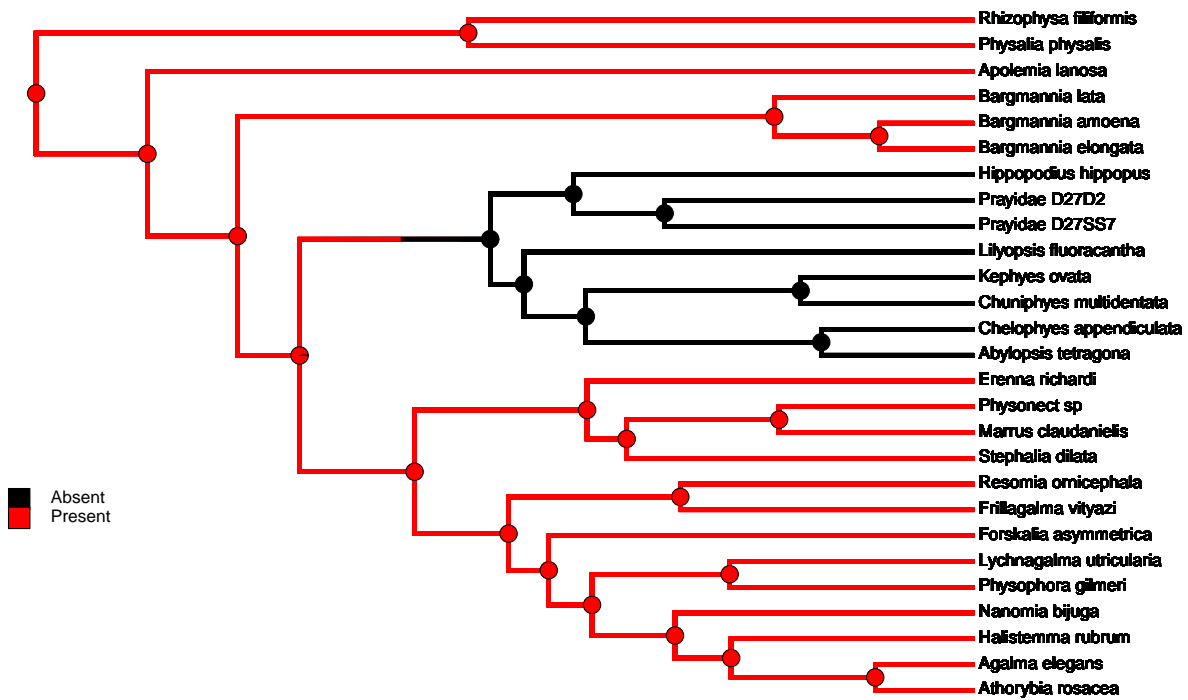
[illegible]

[illegible]

[illegible]

[illegible]

"black" "red"



Posterior probabilities of states (1 Present, 0 Absent).

It is not clear whether or not the stem group of siphonophores had a pneumatophore.

SIMMAP Number of Bract types:

no colors provided. using the following legend:

0 1 2 4


```

## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4

```

```

## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4

```

```

## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4

```

[illegible]

```

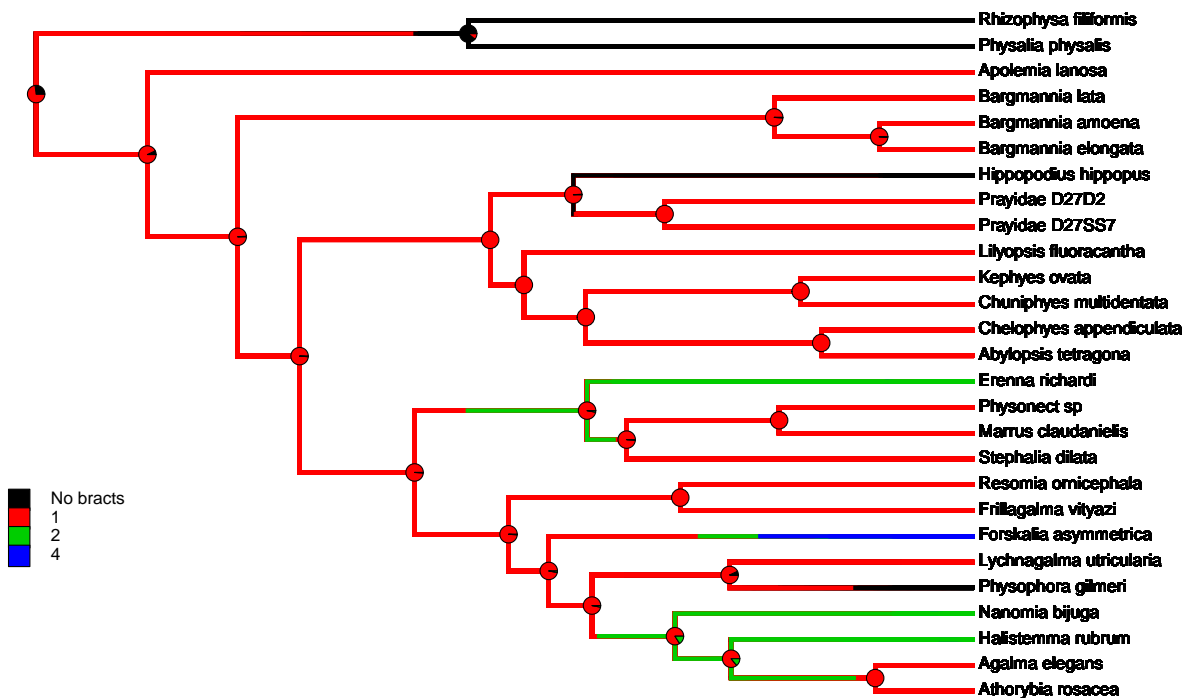
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4

```

```

## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"
## no colors provided. using the following legend:
##      0      1      2      4
## "black"      "red" "green3"  "blue"

```

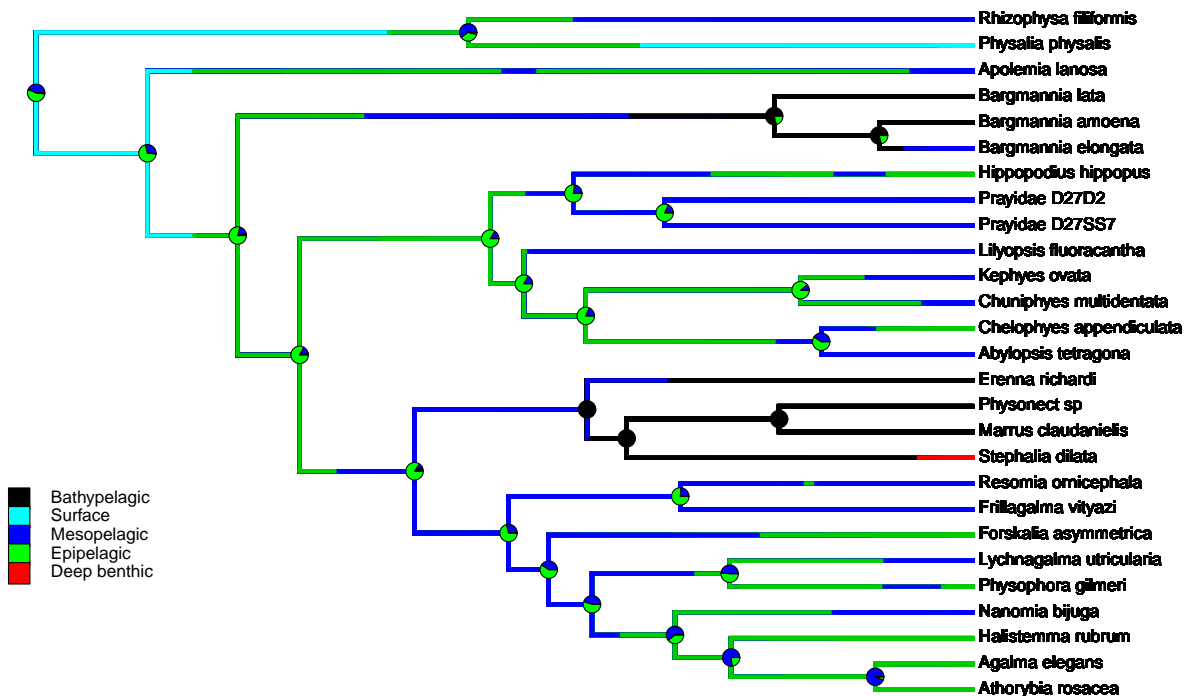


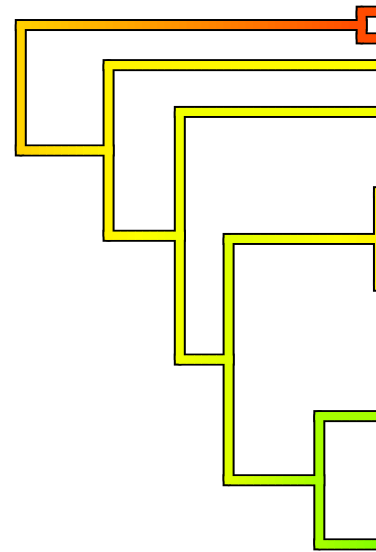
SIMMAP Habitat:


```

## no colors provided. using the following legend:
## Bathypelagic Deep benthic  Epipelagic  Mesopelagic  Surface
##    "black"      "red"      "green3"      "blue"      "cyan"
## no colors provided. using the following legend:
## Bathypelagic Deep benthic  Epipelagic  Mesopelagic  Surface
##    "black"      "red"      "green3"      "blue"      "cyan"
## no colors provided. using the following legend:
## Bathypelagic Deep benthic  Epipelagic  Mesopelagic  Surface
##    "black"      "red"      "green3"      "blue"      "cyan"
## no colors provided. using the following legend:
## Bathypelagic Deep benthic  Epipelagic  Mesopelagic  Surface
##    "black"      "red"      "green3"      "blue"      "cyan"
## no colors provided. using the following legend:
## Bathypelagic Deep benthic  Epipelagic  Mesopelagic  Surface
##    "black"      "red"      "green3"      "blue"      "cyan"
## no colors provided. using the following legend:
## Bathypelagic Deep benthic  Epipelagic  Mesopelagic  Surface
##    "black"      "red"      "green3"      "blue"      "cyan"
## no colors provided. using the following legend:
## Bathypelagic Deep benthic  Epipelagic  Mesopelagic  Surface
##    "black"      "red"      "green3"      "blue"      "cyan"
## no colors provided. using the following legend:
## Bathypelagic Deep benthic  Epipelagic  Mesopelagic  Surface
##    "black"      "red"      "green3"      "blue"      "cyan"
## no colors provided. using the following legend:
## Bathypelagic Deep benthic  Epipelagic  Mesopelagic  Surface
##    "black"      "red"      "green3"      "blue"      "cyan"
## no colors provided. using the following legend:
## Bathypelagic Deep benthic  Epipelagic  Mesopelagic  Surface
##    "black"      "red"      "green3"      "blue"      "cyan"
## no colors provided. using the following legend:
## Bathypelagic Deep benthic  Epipelagic  Mesopelagic  Surface
##    "black"      "red"      "green3"      "blue"      "cyan"

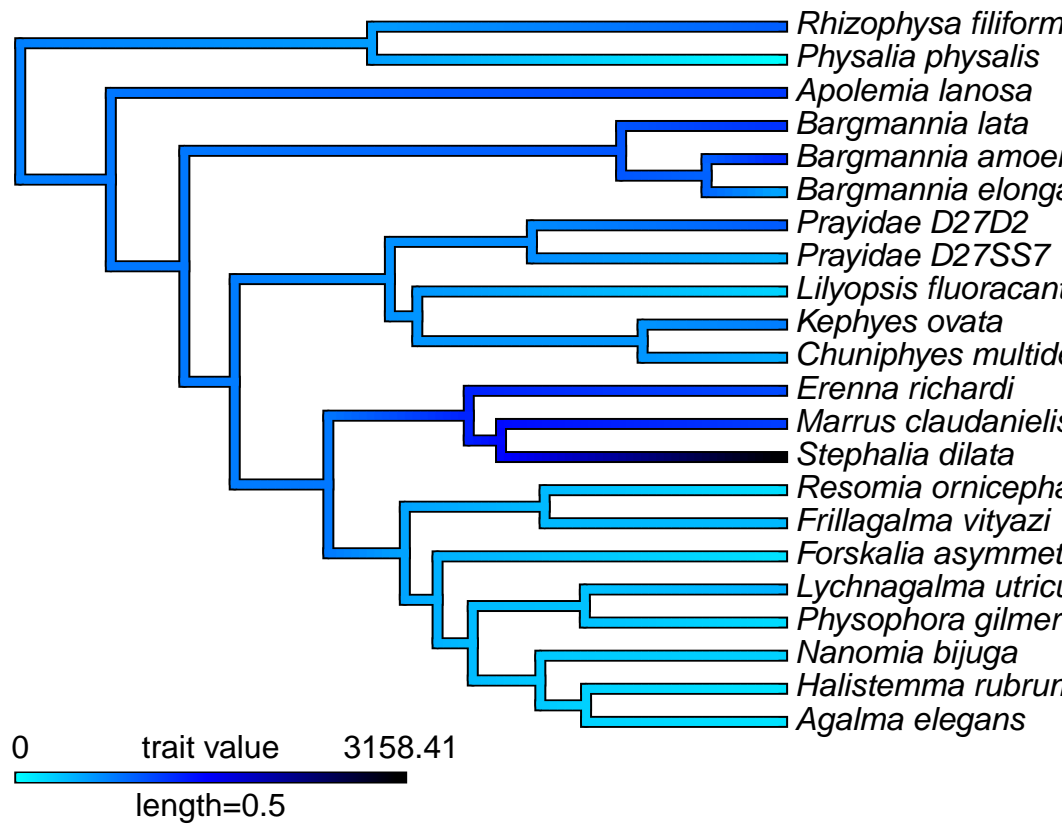
```



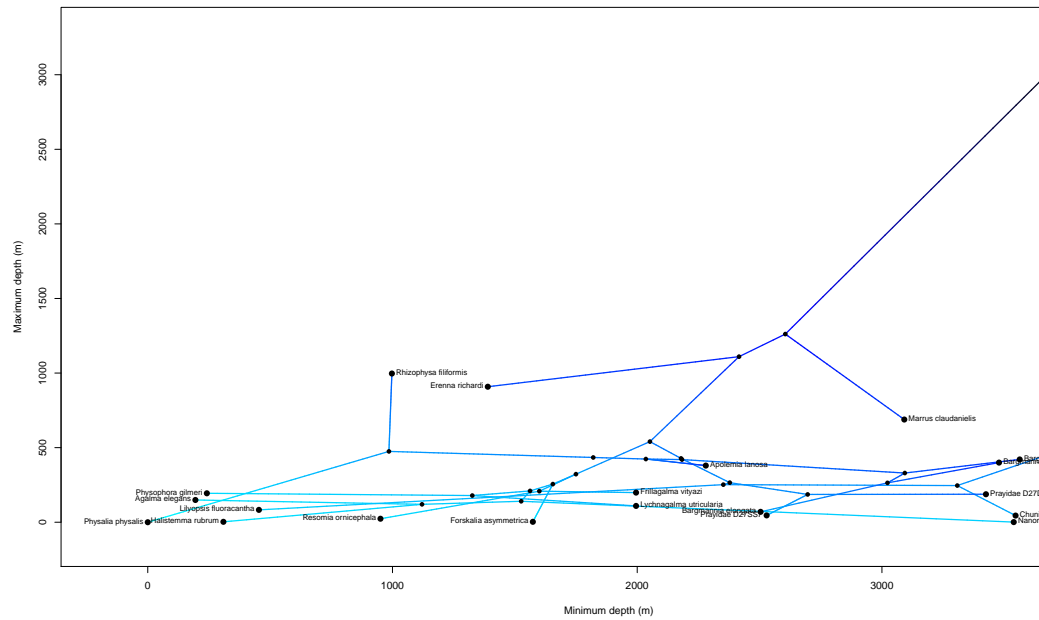


0 trait value
length=0.5

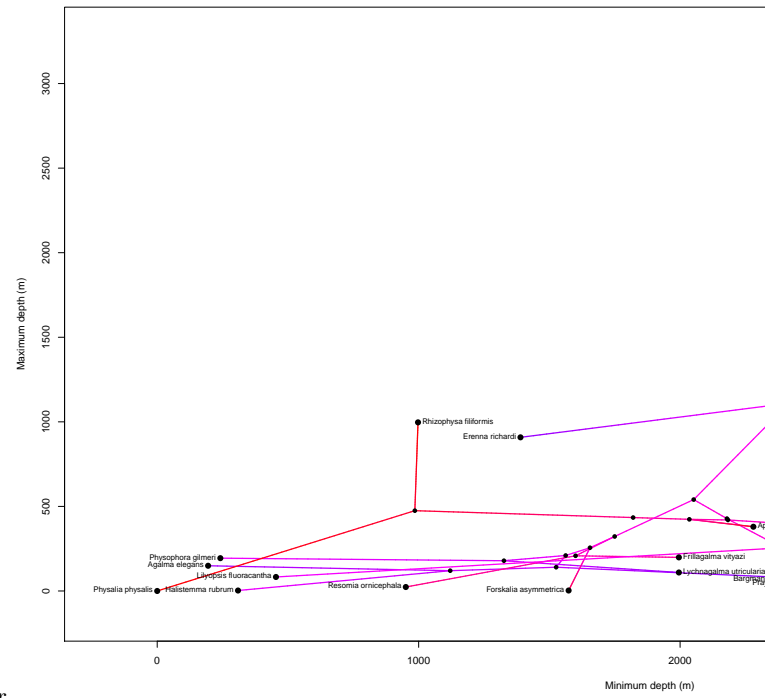
Continuous traits: Brownian motion reconstructions Number of bract types reconstruction:
Trait = Number of Bract Types



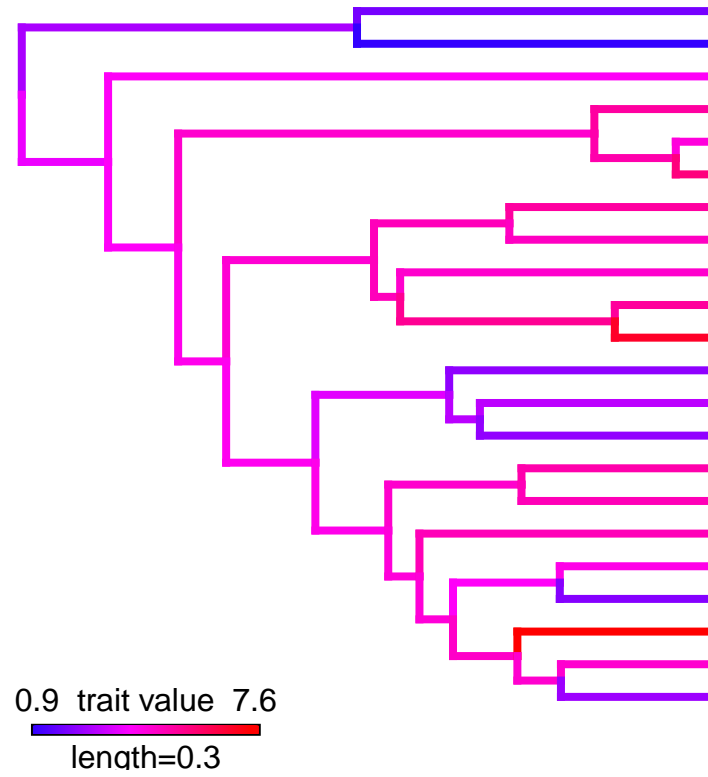
Median depth reconstruction:



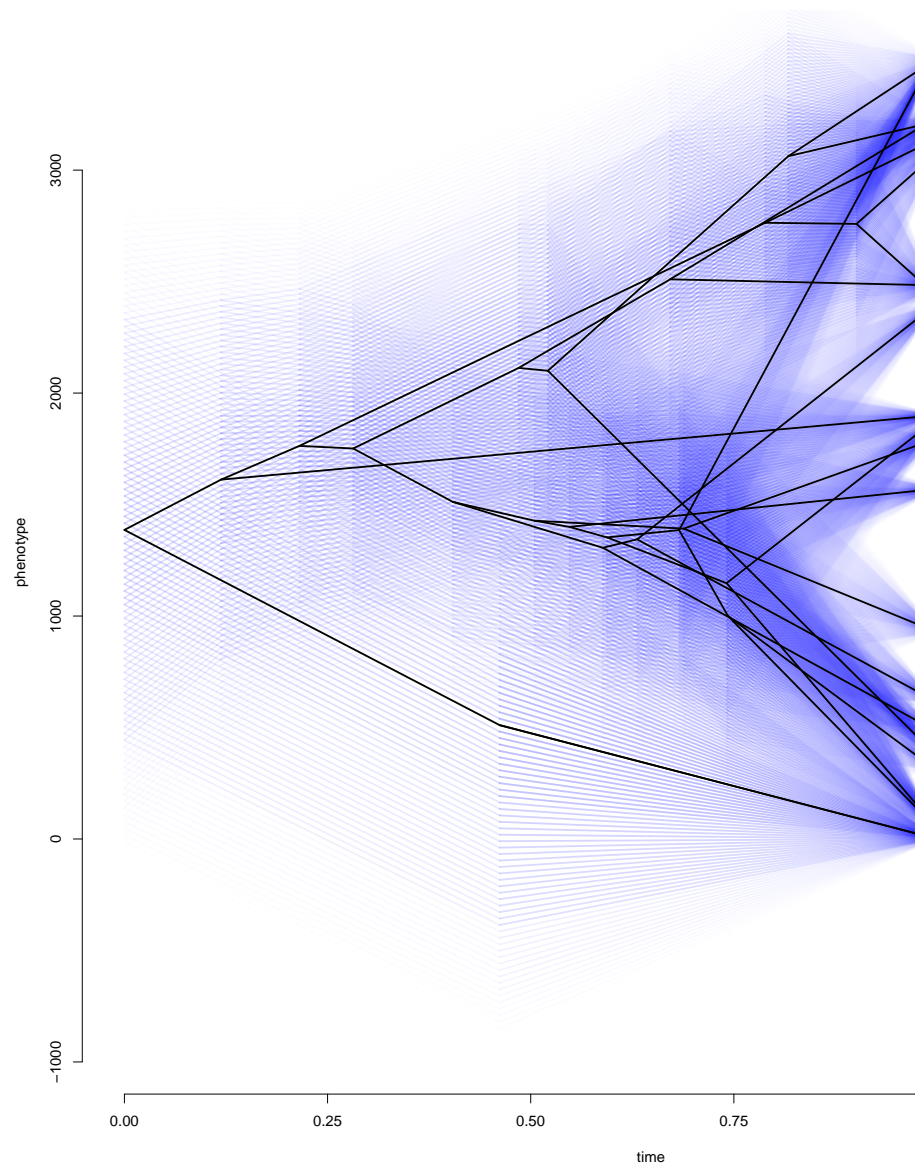
Bathymetrics phylomorphospace



Bathymetrics phylomorphospace with time from root as color



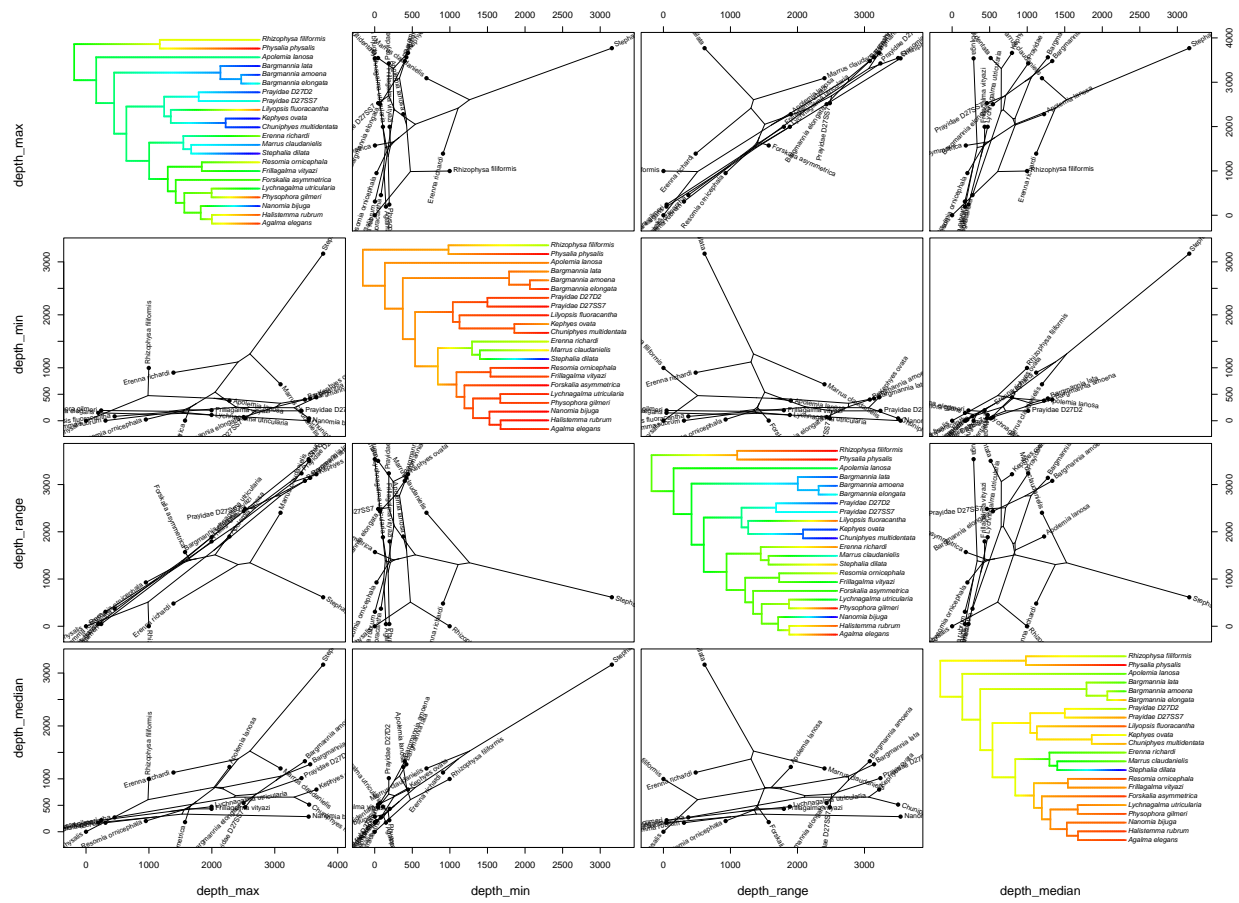
Abundance of each taxon in the Monterey Bay sampling site

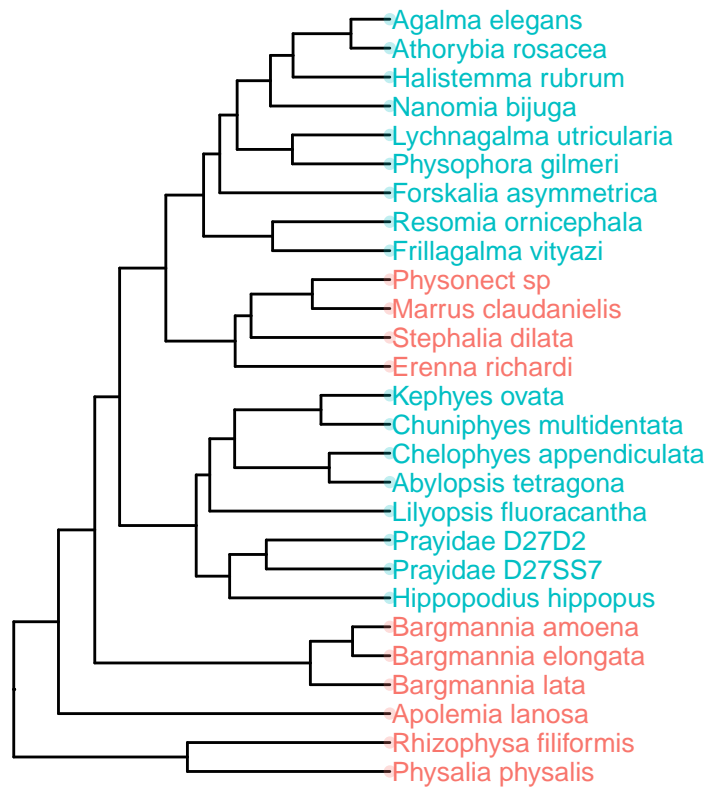


Uncertainty inclusive traitgram for depth range

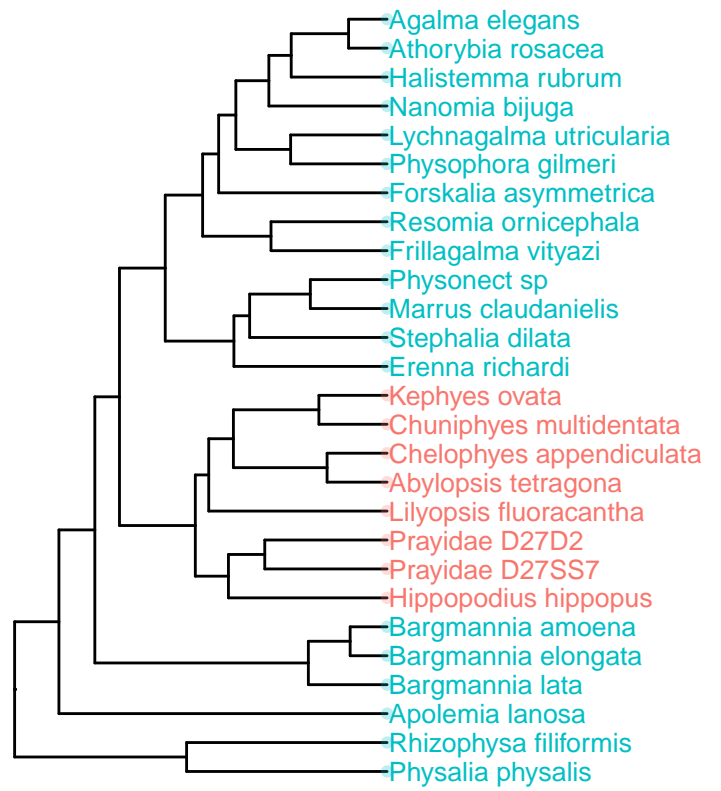
Bathymetrics scattergram array

```
## Computing multidimensional phylogenetic scatterplot matrix...
```

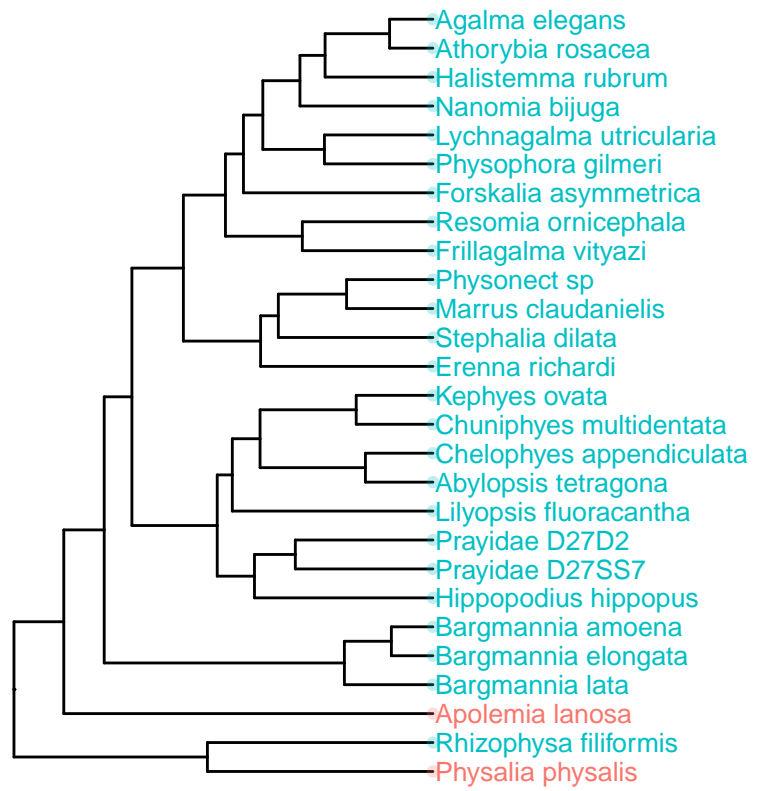





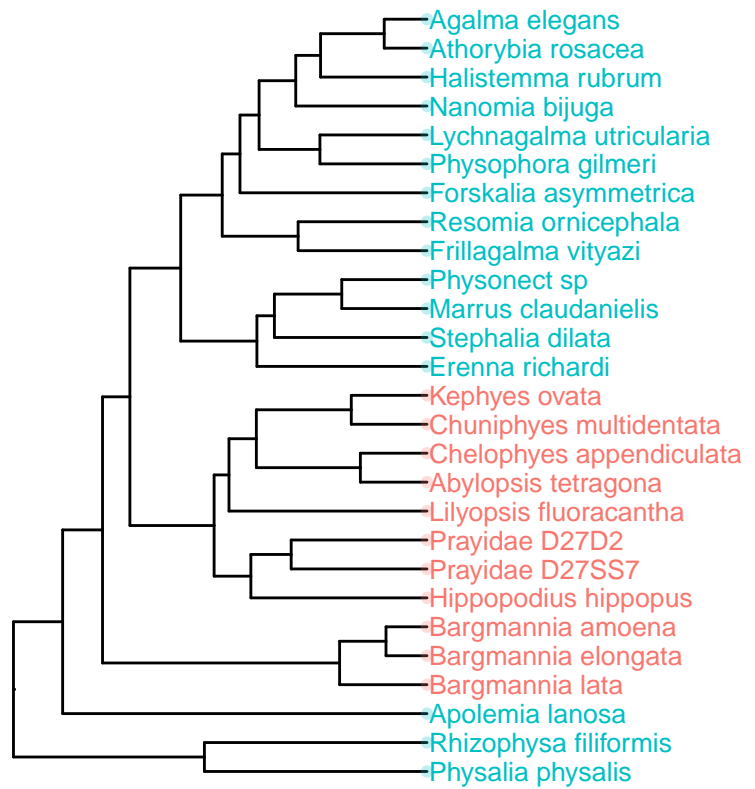
Distribution of sex across siphonophore taxa



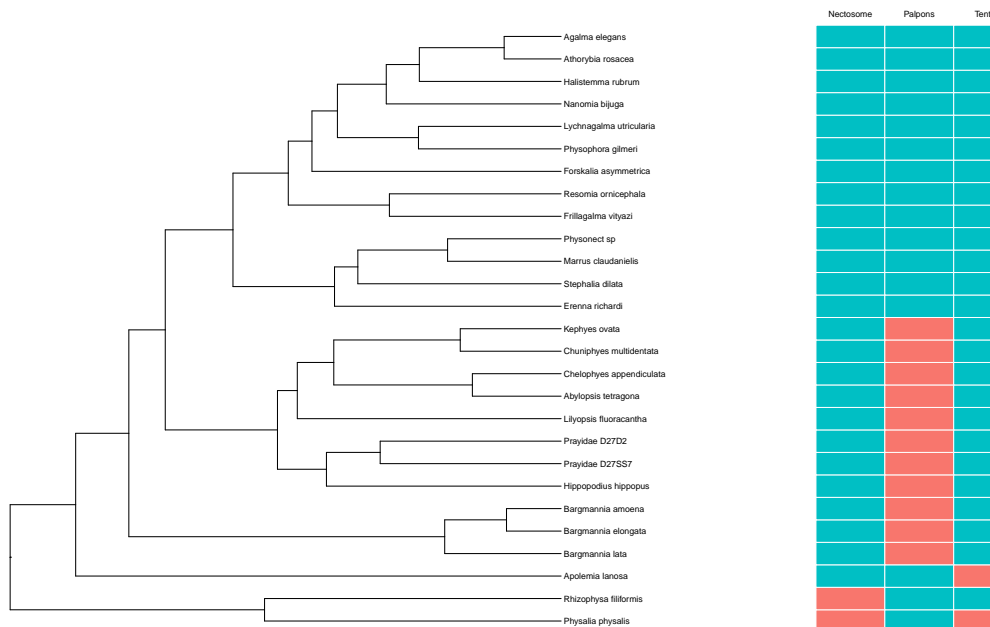
Pneumatophore presence across siphonophore taxa



Tentilla presence across siphonophore taxa



Palpon presence across siphonophore taxa



Overview grid of main binary traits

Phylogenetic signal in the character data Agnostic branch length tree generation:

```
flat_tree = tree
flat_tree$edge.length = rep(mean(flat_tree$edge.length), length(flat_tree$edge.length))
```

Phylogenetic signal in bract number

```
##           K PIC.variance.obs PIC.variance.rnd.mean PIC.variance.P
## 1 0.8039207      1.114354      1.905134      0.027
## PIC.variance.Z
## 1      -1.328822
##
## Call:
## physignal(A = bracts, phy = ultratree)
##
##
## Observed Phylogenetic Signal (K): 0.8039
##
## P-value: 0.051
##
## Based on 1000 random permutations
```

Phylogenetic signal in Binary Traits

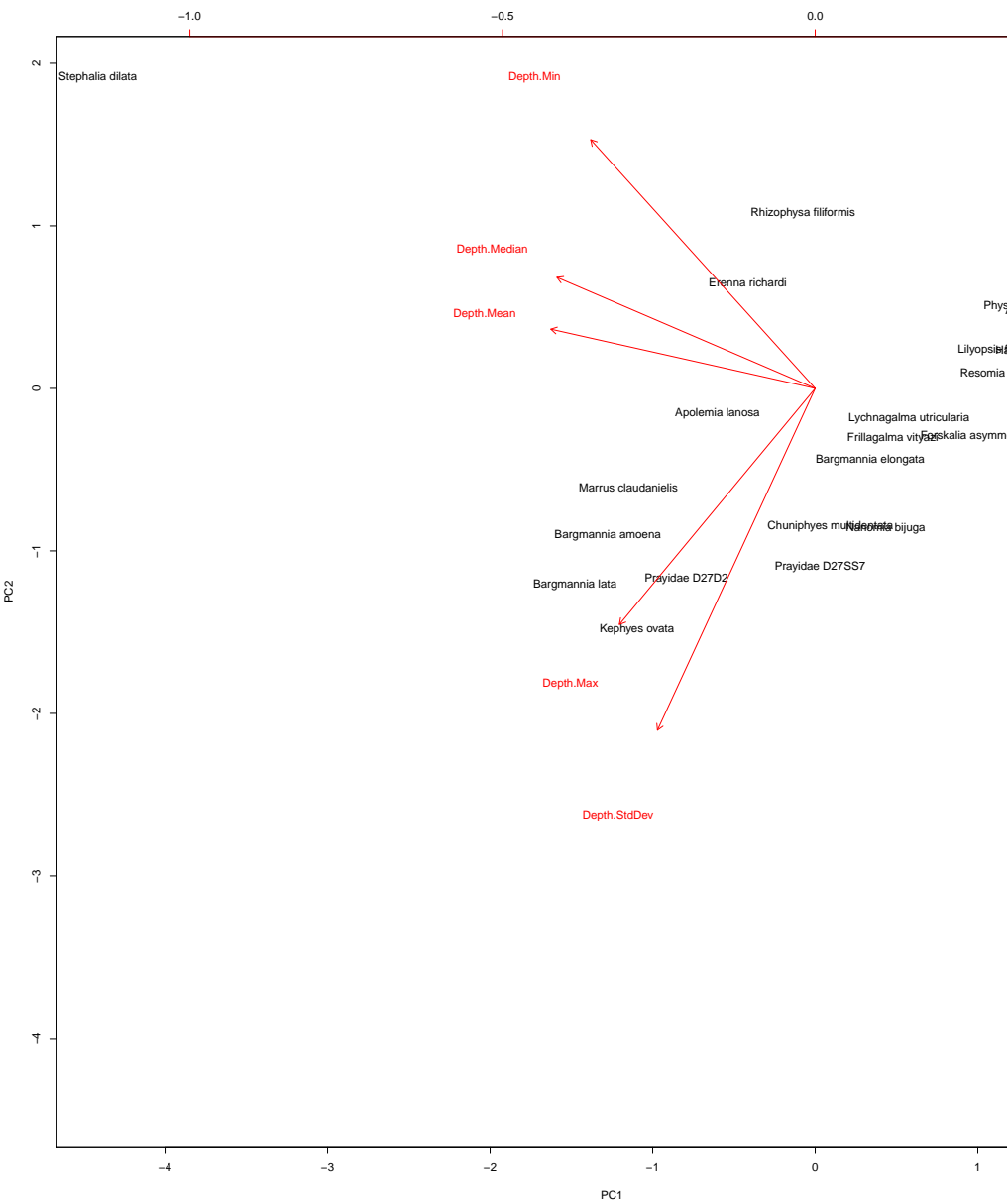
```
##           K PIC.variance.obs PIC.variance.rnd.mean
## Nectosome      4.198327      0.03822813      0.2235080
## Palpons        3.190846      0.10473403      0.7775871
## Tentilla       2.171223      0.07535593      0.2206210
## Pneumatophore  3.388250      0.08703055      0.6733229
## PIC.variance.P PIC.variance.Z
## Nectosome      0.0050      -2.354699
## Palpons        0.0010      -4.467321
## Tentilla       0.0065      -1.921329
## Pneumatophore  0.0010      -4.312803
```

Phylogenetic signal in VARS bathymetrics

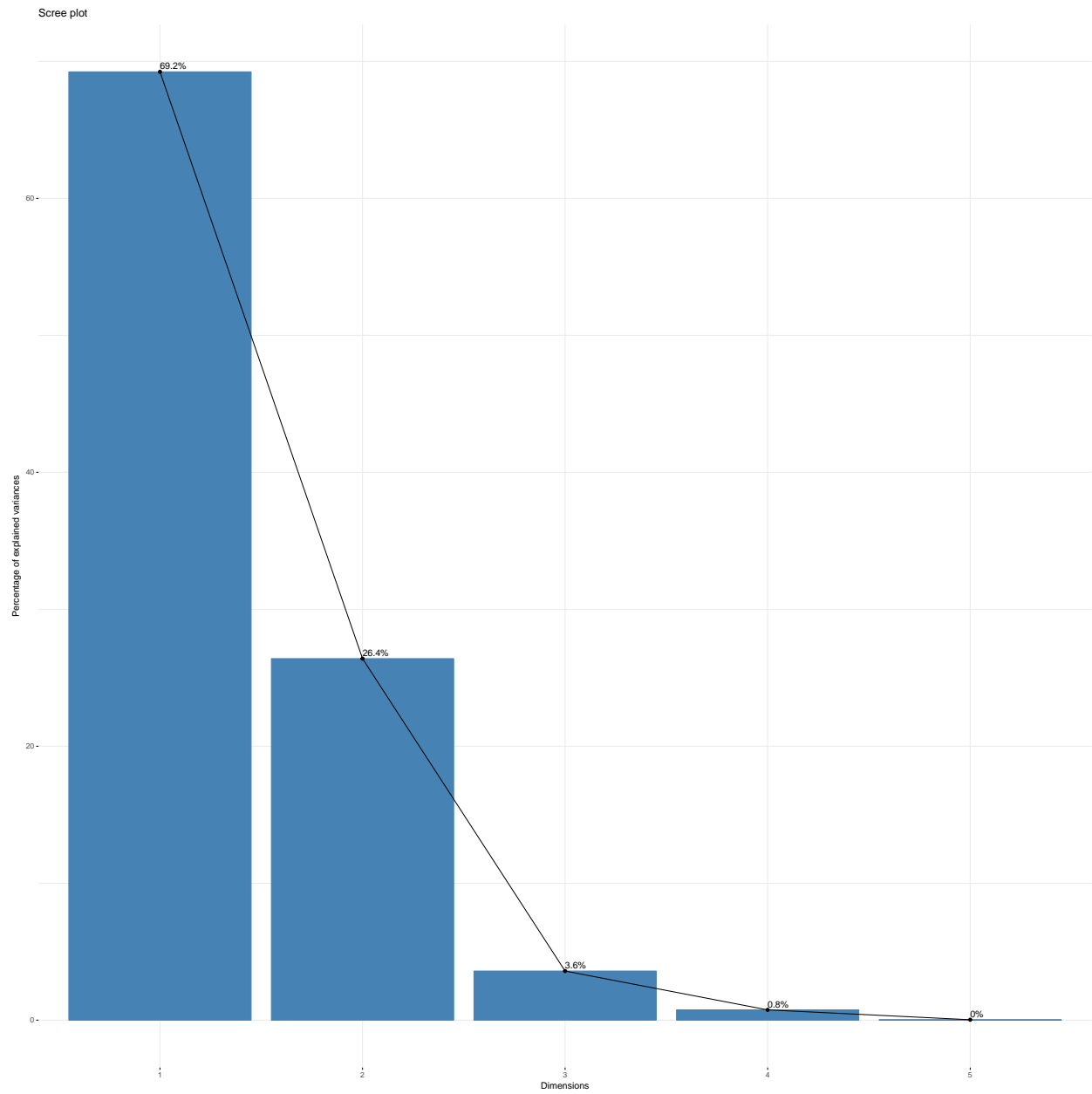
```
##
## Call:
## physignal(A = dpruned_data[, c(14, 15, 16, 18, 19)], phy = dpruned_tree)
##
##
## Observed Phylogenetic Signal (K): 0.713
##
## P-value: 0.041
##
## Based on 1000 random permutations
```

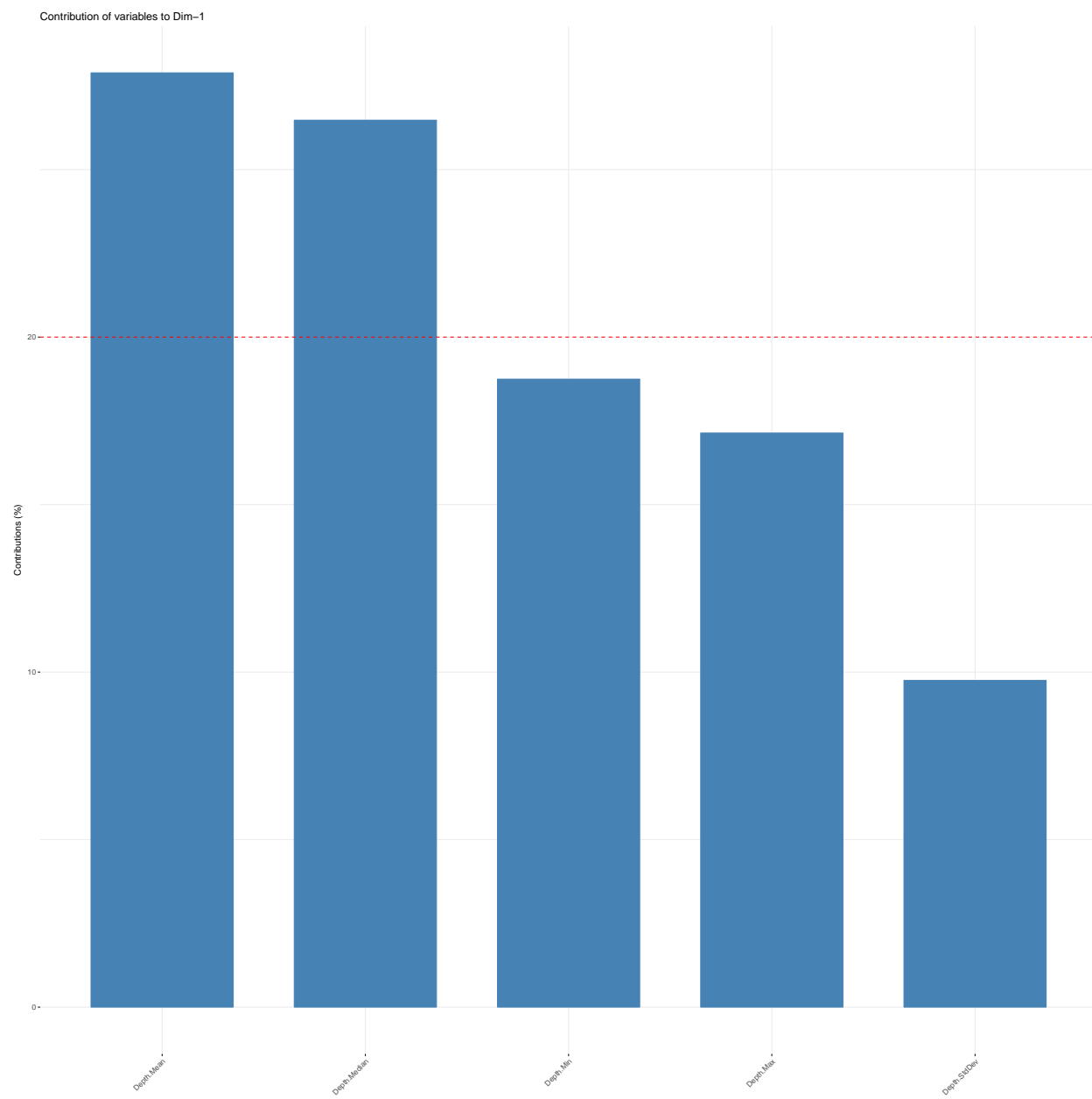
```
##           K PIC.variance.obs PIC.variance.rnd.mean
## Depth.Median 0.7102499      874496.0      1329862.7
## Depth.Mean   0.6810592      1019282.2      1479283.2
## Depth.StdDev 0.5889846      137874.8      171580.4
## Depth.Max    0.7249779      3266155.6      4851428.3
## Depth.Min    0.7287910      828844.8      1309962.6
## PIC.variance.P PIC.variance.Z
## Depth.Median      0.078      -1.0201344
## Depth.Mean        0.088      -1.0256350
## Depth.StdDev      0.195      -0.8584777
```

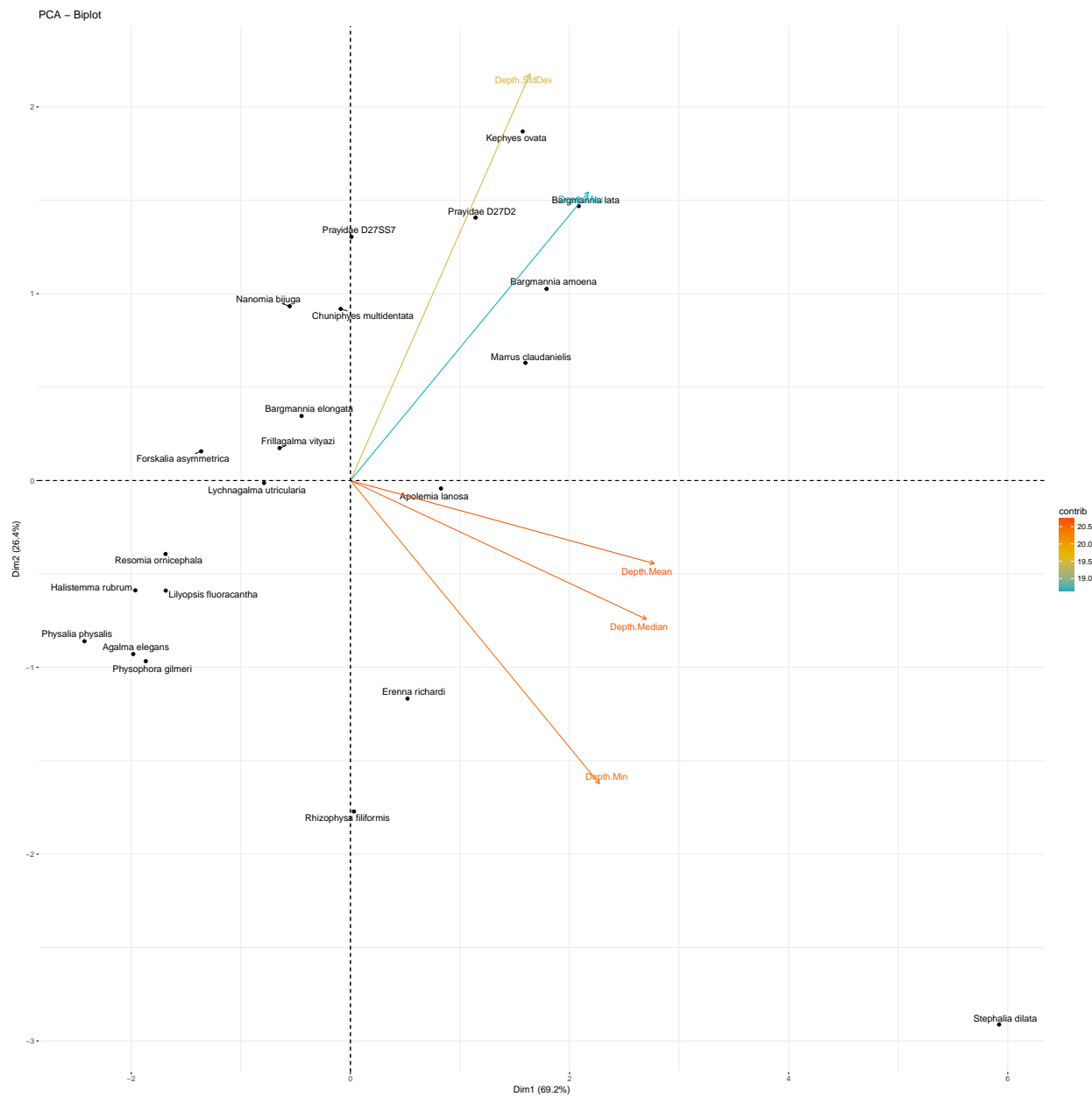
Depth.Max 0.049 -1.5430651
Depth.Min 0.124 -0.8662187

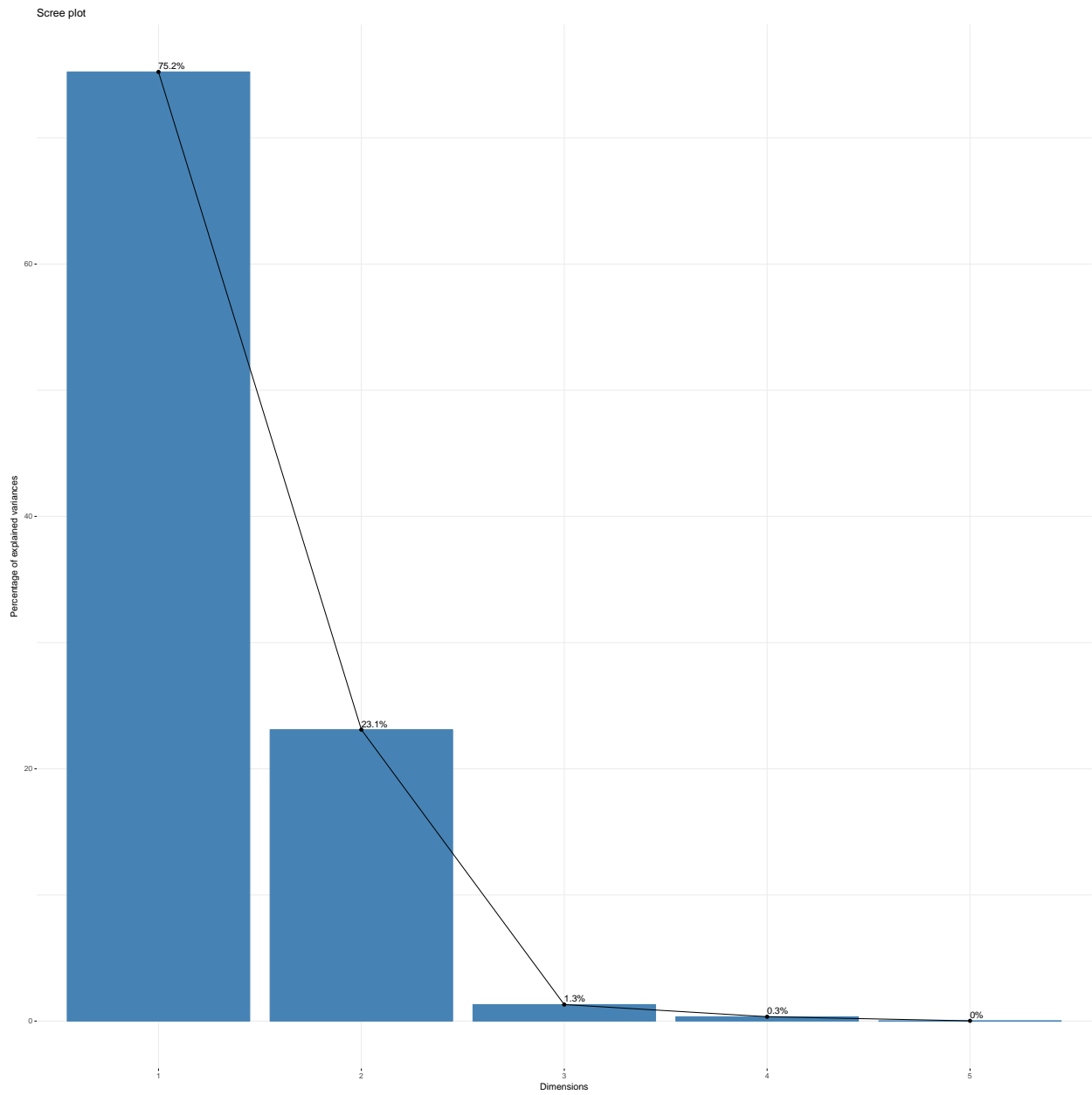


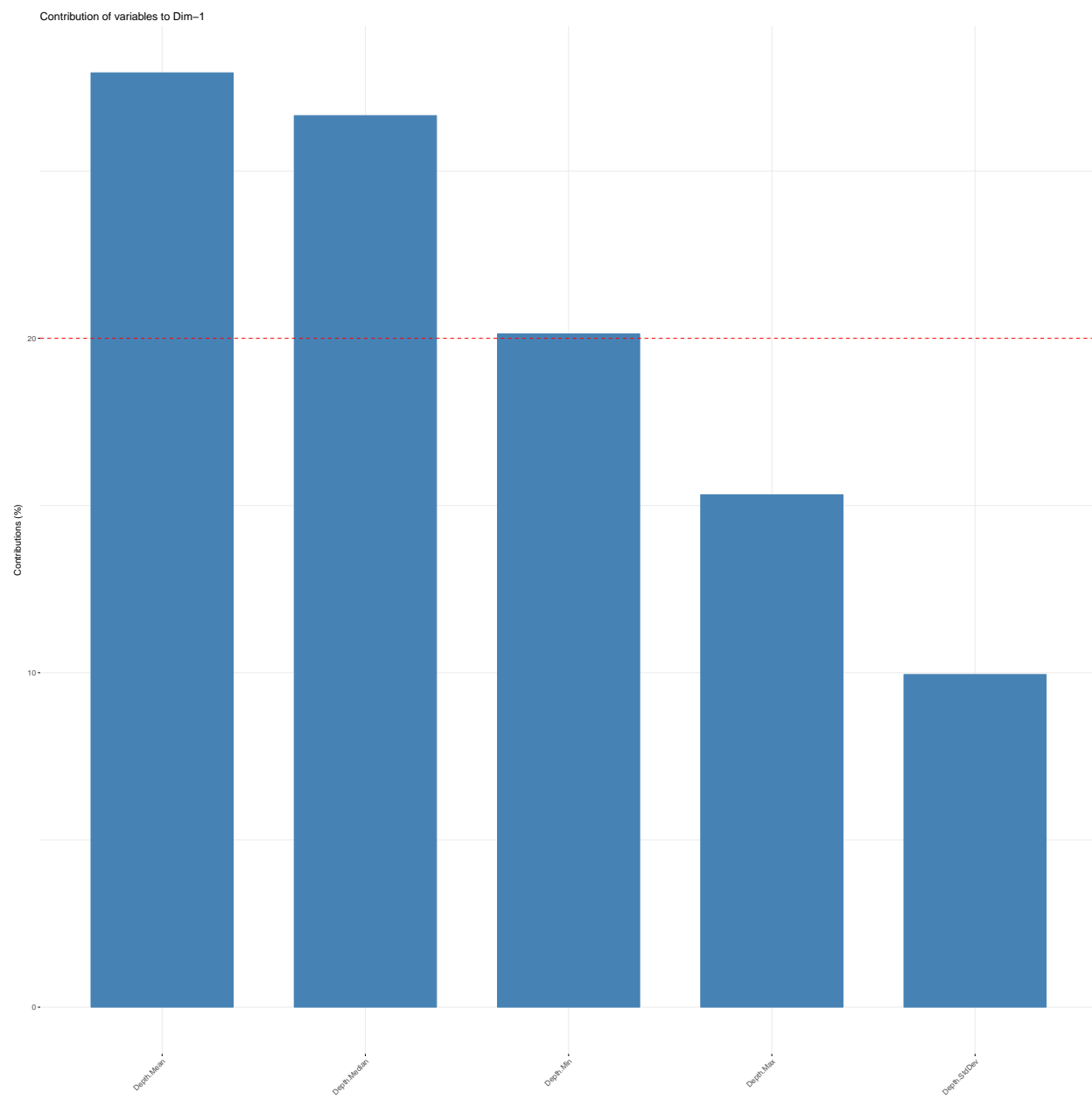
Phylogenetic PCA for bathymetrics

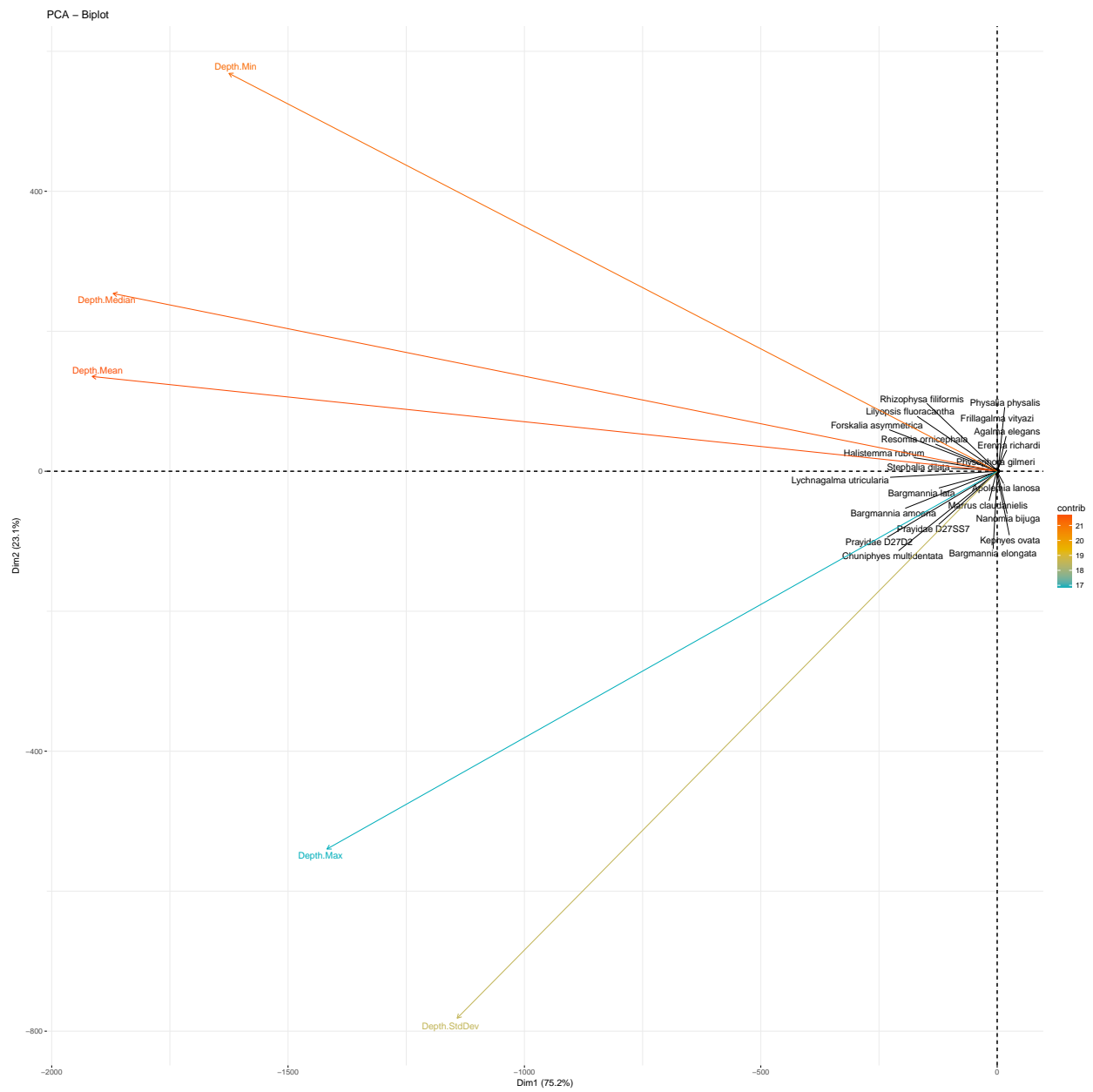


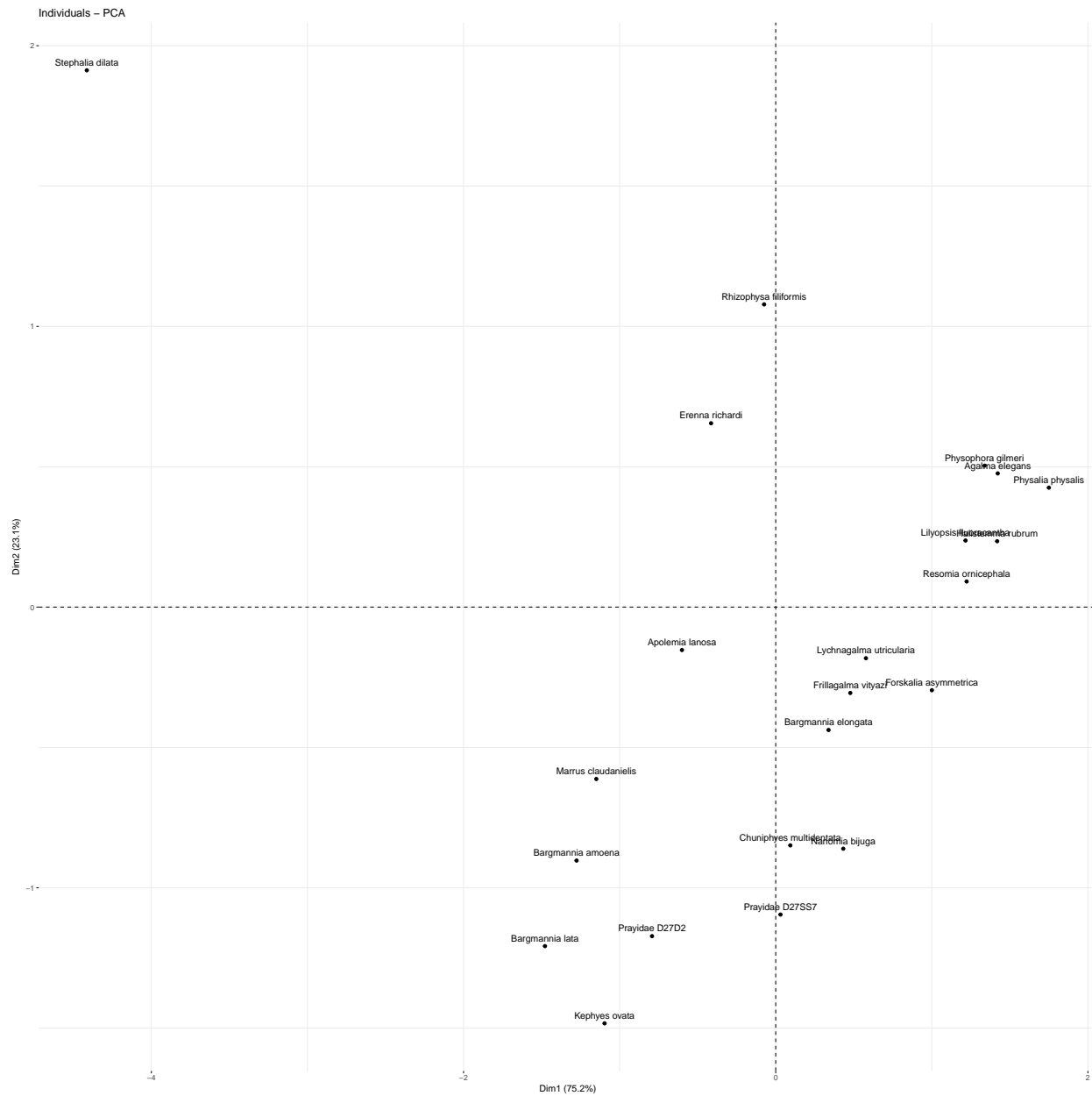












Phylogenetic ANOVA for depth

Each phylogenetic ANOVA compares variance of depth (continuous variable) within and between groups clustered by their categorical biological traits.

```
## [1] "Depth vs Sex Distribution"
```

```
## $F
```

```
## [1] 10.48139
```

```
##
```

```
## $Pf
```

```
## [1] 0.056
```

```
##
```

```
## $T
```

```
##          Dioecious Monoecious
```

```
## Dioecious 0.000000 3.237497
```

```

## Monoecious -3.237497  0.000000
##
## $method
## [1] "holm"
##
## $Pt
##           Dioecious Monoecious
## Dioecious      1.000      0.056
## Monoecious      0.056      1.000
## [1] "Depth vs Nectosome"
## $F
## [1] 0.2358588
##
## $Pf
## [1] 0.796
##
## $T
##           0          1
## 0 0.000000 -0.485653
## 1 0.485653  0.000000
##
## $method
## [1] "holm"
##
## $Pt
##           0          1
## 0 1.000 0.796
## 1 0.796 1.000
## [1] "Depth vs Nectophore Types and Number"
## $F
## [1] 0.156464
##
## $Pf
## [1] 0.98
##
## $T
##           Multiple of one type None One of one type
## Multiple of one type           NA  NA              NA
## None                        NA  NA              NA
## One of one type             NA  NA              NA
## Two of one type             NA  NA              NA
## Two of two types            NA  NA              NA
##           Two of one type Two of two types
## Multiple of one type      NA              NA
## None                      NA              NA
## One of one type           NA              NA
## Two of one type           NA              NA
## Two of two types          NA              NA
##
## $method
## [1] "holm"
##

```

```

## $Pt
##           Multiple of one type None One of one type
## Multiple of one type           NA  NA              NA
## None                        NA  NA              NA
## One of one type             NA  NA              NA
## Two of one type             NA  NA              NA
## Two of two types            NA  NA              NA
##           Two of one type Two of two types
## Multiple of one type           NA              NA
## None                        NA              NA
## One of one type             NA              NA
## Two of one type             NA              NA
## Two of two types            NA              NA

## [1] "Depth vs Nectosome Position"

## $F
## [1] 1.238642
##
## $Pf
## [1] 0.594
##
## $T
##           Dorsal      None      Ventral
## Dorsal  0.0000000 -0.4636838 -1.4922990
## None    0.4636838  0.0000000 -0.6838791
## Ventral 1.4922990  0.6838791  0.0000000
##
## $method
## [1] "holm"
##
## $Pt
##           Dorsal None Ventral
## Dorsal    1.000    1  0.831
## None      1.000    1  1.000
## Ventral   0.831    1  1.000

## [1] "Depth vs Palpons"

## $F
## [1] 0.05567054
##
## $Pf
## [1] 0.889
##
## $T
##           0      1
## 0  0.0000000 0.2359461
## 1 -0.2359461 0.0000000
##
## $method
## [1] "holm"
##
## $Pt
##           0      1
## 0  1.000 0.889

```



```

## 1 0.889 1.000
## [1] "Depth vs Tentilla"
## $F
## [1] 0.06009463
##
## $Pf
## [1] 0.873
##
## $T
##           0           1
## 0 0.0000000 -0.2451421
## 1 0.2451421  0.0000000
##
## $method
## [1] "holm"
##
## $Pt
##           0           1
## 0 1.000 0.873
## 1 0.873 1.000
## [1] "Depth vs Pneumatophore"
## $F
## [1] 0.1796993
##
## $Pf
## [1] 0.793
##
## $T
##           0           1
## 0 0.0000000 -0.4239096
## 1 0.4239096  0.0000000
##
## $method
## [1] "holm"
##
## $Pt
##           0           1
## 0 1.000 0.793
## 1 0.793 1.000
PGLS - Sex distribution
## -----
## | **Warning:                                     |
## |   User reports suggest that this method may frequently   |
## |   fail to find the ML solution. Please use with caution.   |
## |-----|
##
## Call:
## phylolm(formula = depth_median ~ dp_sex, data = as.data.frame(cbind(depth_median,
##   dp_sex)), phy = dpruned_tree, model = "BM", measurement_error = TRUE,
##   boot = 100)
##

```

```
##      AIC logLik
##    347.8 -169.9
##
## Parameter estimate(s) using ML:
## sigma2: 0.04396236
## sigma2_error: 299475.5
##
## Coefficients:
## (Intercept)      dp_sex
##    399.7538    805.7584

## Generalized least squares fit by REML
##   Model: dp_sex ~ depth_median
##   Data: NULL
##   Log-restricted-likelihood: -20.52555
##
## Coefficients:
## (Intercept) depth_median
## 0.0978229509 0.0004267556
##
## Degrees of freedom: 22 total; 20 residual
## Residual standard error: 0.4176998
```

Software versions

This manuscript was computed on Tue Oct 10 09:48:28 PM 2017 with the following R package versions.

```
R version 3.4.1 (2017-06-30)
Platform: x86_64-pc-linux-gnu (64-bit)
Running under: Debian GNU/Linux 9 (stretch)
```

```
Matrix products: default
BLAS/LAPACK: /usr/lib/libopenblas-r0.2.19.so
```

```
locale:
 [1] LC_CTYPE=en_US.UTF-8      LC_NUMERIC=C
 [3] LC_TIME=en_US.UTF-8      LC_COLLATE=en_US.UTF-8
 [5] LC_MONETARY=en_US.UTF-8  LC_MESSAGES=C
 [7] LC_PAPER=en_US.UTF-8     LC_NAME=C
 [9] LC_ADDRESS=C             LC_TELEPHONE=C
[11] LC_MEASUREMENT=en_US.UTF-8 LC_IDENTIFICATION=C
```

```
attached base packages:
[1] grid      parallel  stats      graphics  grDevices  utils      datasets
[8] methods   base
```

```
other attached packages:
 [1] bindrcpp_0.2      phylolm_2.5      geomorph_3.0.5    rgl_0.98.1
 [5] adephylo_1.1-10   ade4_1.7-8       phylobase_0.8.4   geiger_2.0.6
 [9] phangorn_2.2.0    phytools_0.6-20  picante_1.6-2     nlme_3.1-131
[13] vegan_2.4-4       lattice_0.20-35  permute_0.9-4     ape_4.1
[17] hutan_0.5.0       FactoMineR_1.38  factoextra_1.0.5  gridExtra_2.2.1
[21] seriation_1.2-2   fields_9.0       maps_3.2.0        spam_2.1-1
[25] dotCall64_0.9-04  ggtree_1.8.2     treeio_1.0.2      cowplot_0.8.0
```

[29]	xtable_1.8-2	jsonlite_1.5	knitr_1.16	digest_0.6.12
[33]	magrittr_1.5	forcats_0.2.0	stringr_1.2.0	dplyr_0.7.4
[37]	purrr_0.2.2.2	readr_1.1.1	tidyr_0.6.3	tibble_1.3.4
[41]	ggplot2_2.2.1	tidyverse_1.1.1		

loaded via a namespace (and not attached):

[1]	readxl_1.0.0	uuid_0.1-2
[3]	backports_1.1.0	fastmatch_1.1-0
[5]	plyr_1.8.4	igraph_1.1.2
[7]	lazyeval_0.2.0	sp_1.2-5
[9]	splines_3.4.1	rncl_0.8.2
[11]	foreach_1.4.3	htmltools_0.3.6
[13]	viridis_0.4.0	gdata_2.18.0
[15]	cluster_2.0.6	gclus_1.3.1
[17]	modelr_0.1.0	gmodels_2.16.2
[19]	prettyunits_1.0.2	jpeg_0.1-8
[21]	colorspace_1.3-2	rvest_0.3.2
[23]	ggrepel_0.7.0	haven_1.1.0
[25]	bindr_0.1	survival_2.41-3
[27]	iterators_1.0.8	glue_1.1.1
[29]	registry_0.3	gtable_0.2.0
[31]	seqinr_3.4-5	kernlab_0.9-25
[33]	prabclus_2.2-6	DEoptimR_1.0-8
[35]	scales_0.4.1	mvtnorm_1.0-6
[37]	Rcpp_0.12.13	plotrix_3.6-6
[39]	viridisLite_0.2.0	progress_1.1.2
[41]	spdep_0.6-15	flashClust_1.01-2
[43]	foreign_0.8-69	subplex_1.4-1
[45]	bold_0.5.0	mclust_5.3
[47]	deSolve_1.20	stats4_3.4.1
[49]	animation_2.5	htmlwidgets_0.9
[51]	httr_1.2.1	gplots_3.0.1
[53]	fpc_2.1-10	modeltools_0.2-21
[55]	pkgconfig_2.0.1	reshape_0.8.7
[57]	XML_3.98-1.9	flexmix_2.3-14
[59]	deldir_0.1-14	nnet_7.3-12
[61]	crul_0.4.0	labeling_0.3
[63]	rlang_0.1.2	reshape2_1.4.2
[65]	munSELL_0.4.3	cellranger_1.1.0
[67]	tools_3.4.1	broom_0.4.2
[69]	evaluate_0.10.1	yaml_2.1.14
[71]	robustbase_0.92-7	caTools_1.17.1
[73]	dendextend_1.5.2	mime_0.5
[75]	whisker_0.3-2	taxize_0.9.0
[77]	adeigenet_2.0.1	leaps_3.0
[79]	xml2_1.1.1	compiler_3.4.1
[81]	curl_2.8.1	clusterGeneration_1.3.4
[83]	RNeXML_2.0.7	stringi_1.1.5
[85]	highr_0.6	trimcluster_0.1-2
[87]	Matrix_1.2-10	psych_1.7.5
[89]	msm_1.6.4	LearnBayes_2.15
[91]	combinat_0.0-8	data.table_1.10.4
[93]	bitops_1.0-6	httpuv_1.3.5
[95]	R6_2.2.2	TSP_1.1-5

[97]	KernSmooth_2.23-15	codetools_0.2-15
[99]	boot_1.3-20	MASS_7.3-47
[101]	gtools_3.5.0	assertthat_0.2.0
[103]	rprojroot_1.2	mnormt_1.5-5
[105]	diptest_0.75-7	mgcv_1.8-17
[107]	expm_0.999-2	hms_0.3
[109]	quadprog_1.5-5	coda_0.19-1
[111]	class_7.3-14	rmarkdown_1.6
[113]	rvcheck_0.0.9	ggpubr_0.1.5
[115]	shiny_1.0.3	numDeriv_2016.8-1
[117]	scatterplot3d_0.3-40	lubridate_1.6.0

References

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- Dunn C.W., Howison M., Zapata F. 2013. Agalma: an automated phylogenomics workflow. *BMC Bioinformatics*. 14:330.
- Yu G., Smith D.K., Zhu H., Guan Y., Lam T.T.-Y. 2016. ggtree: an r package for visualization and annotation of phylogenetic trees with their covariates and other associated data. *Methods in Ecology and Evolution*.