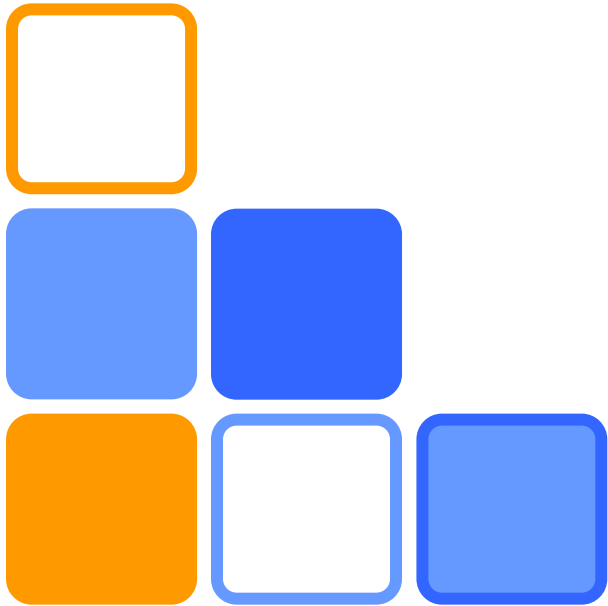


Centrohelida is still searching for a phylogenetic home

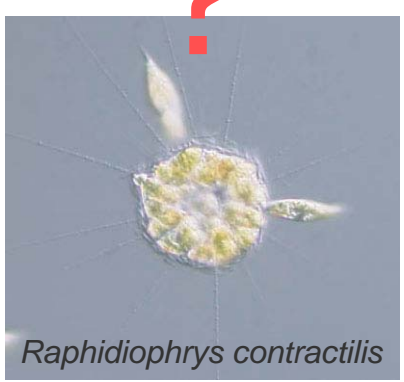
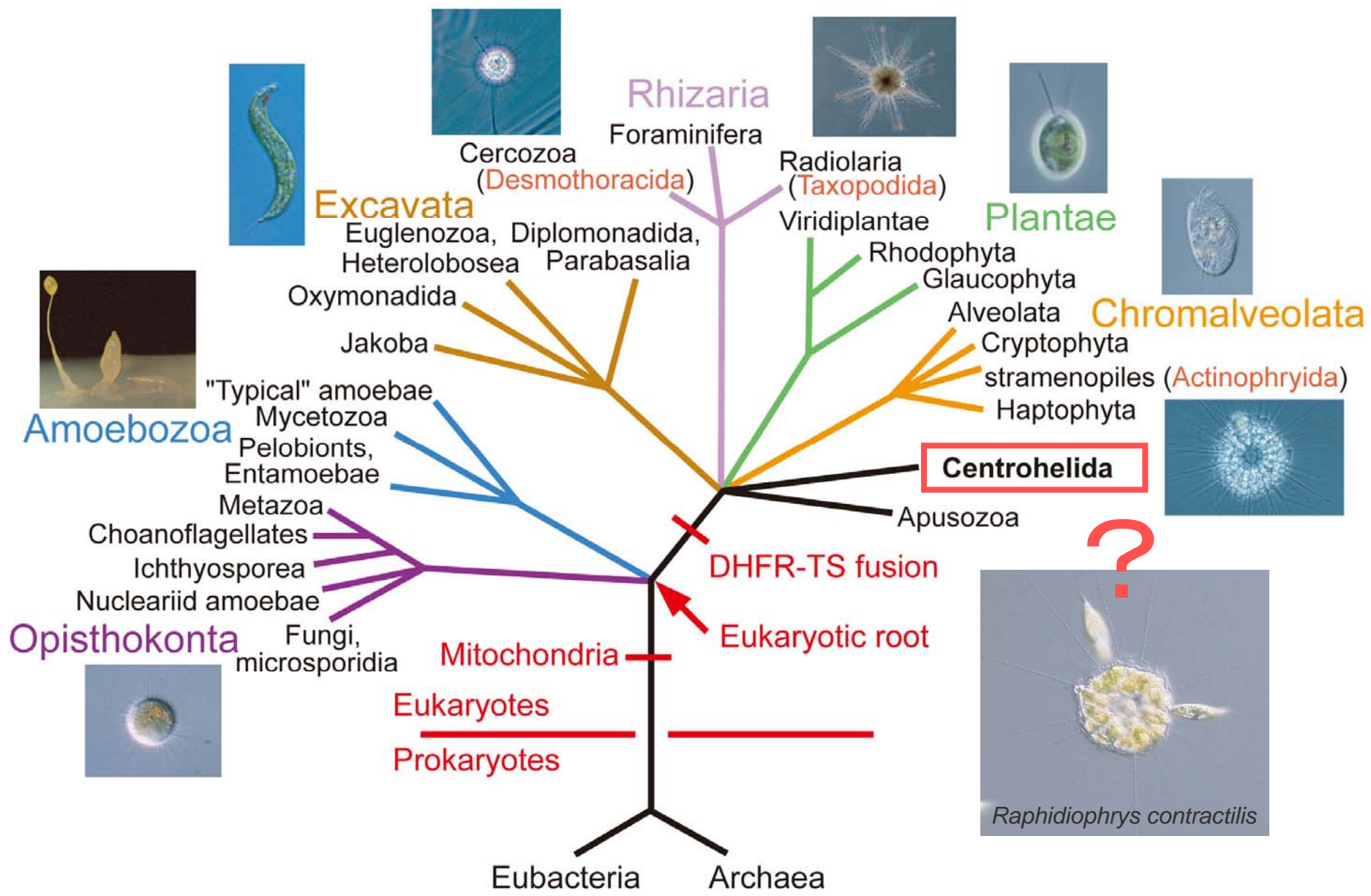
–analyses of seven *Raphidiophrys contractilis* genes–



Inst. Biol. Sci., Univ. Tsukuba
Miako Sakaguchi



Phylogeny of eukaryotes



Raphidiophrys contractilis



Classification of Heliozoa

Sarcodina (肉質虫類)

Rhizopoda (根足虫類)

- Lobosea
Amoeba, Naegleria, Entamoeba, etc.
- Acarpomyxea
- Acrasea
- Eumycetozoea
- Plasmodiophorea
- Filosea
Nuclearia, Gromia, etc.
- Granuloreticulosea
- Foraminiferida, etc.
- Xenophyophorea

Actinopoda (有軸仮足類)

- Acantharea
Astrolithium, etc.
- Polycystinea
Spongaster, etc.
- Phaeodarea
Aulosphaera, etc.
- Heliozoa (太陽虫類)
- Desmothoracida
- Taxopodida (*Sticholonche*)
- Actinophryida
- Centrohelida**

Radiolaria
(放射虫類)

Levine et al. (1980)



Amoebozoa (アメーボゾア)

- Tubulinea
Amoeba, Leptomyxa, Arcella, etc.
- Flabellineaea
Acanthamoeba, Entamoeba, Mastigamoeba, etc.
- Eumycetozoea

Opisthokonta (オピストコンタ)

- Mesomycetozoea
Nuclearia, etc.

Excavata (エクスカベート)

- Heterolobosea
Acrasis, etc.

Rhizaria (リザリア)

- Gromia*
- Foraminifera
- Cercozoa
 - Silicofilosea
 - Euglyphida, etc.
 - Nucleochelea**
 - Clathrulinidae, Gymnosphaerida
 - Phaeodarea
Aulosphaera, etc.
 - Radiolaria**
 - Acantharea
Astrolithium, etc.
 - Polycystinea
Spongaster, etc.
 - Sticholonche*

Chromalveolata (クロムアルベオラータ)

- Stramenopiles
Actinophryidae

Centrohelida ?

Adl et al. (2005)



Aim of this study

- To clarify the phylogenetic position of
- **Centrohelida...**



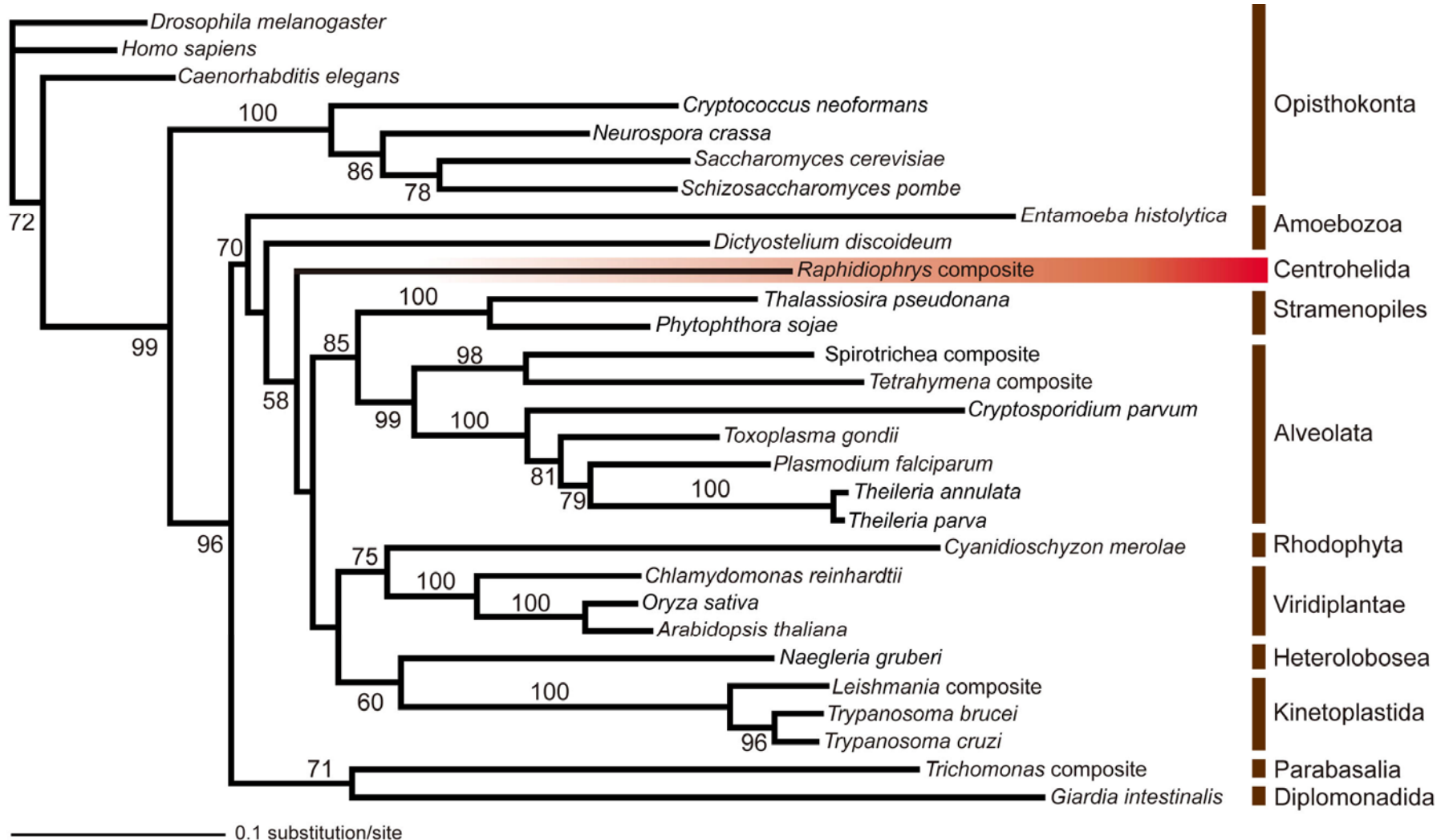
ML combined analyses of Centrohelida

- ◆ 7-gene data sets
(SSU rRNA, actin, α -tubulin, β -tubulin, EF2, HSP70, HSP90)
- ◆ ‘concatenate model’ & ‘separate model’



Multi-gene ML analysis -concatenate model-

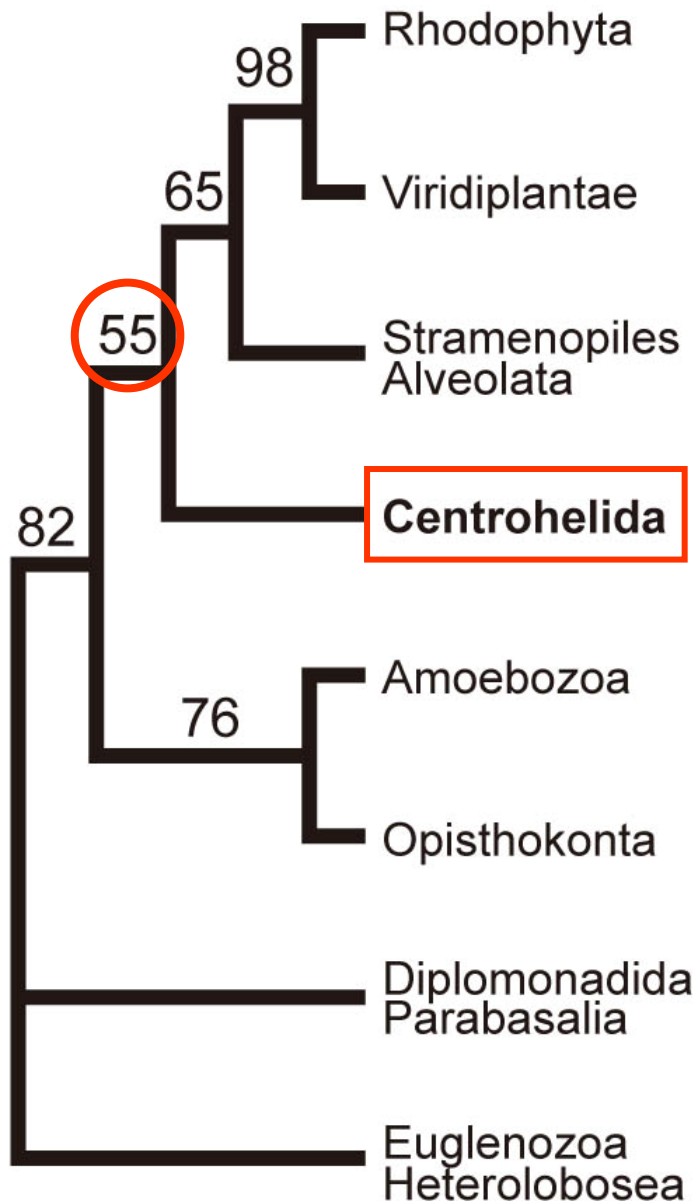
6-protein: α -tubulin, β -tubulin, actin, EF2, HSP70, HSP90



proml
2937aa sites



Multi-gene ML analyses –separate model–



7-gene

α -tubulin, β -tubulin, Actin, EF2
HSP70, HSP90, SSU rRNA



Centrohelida branched with...

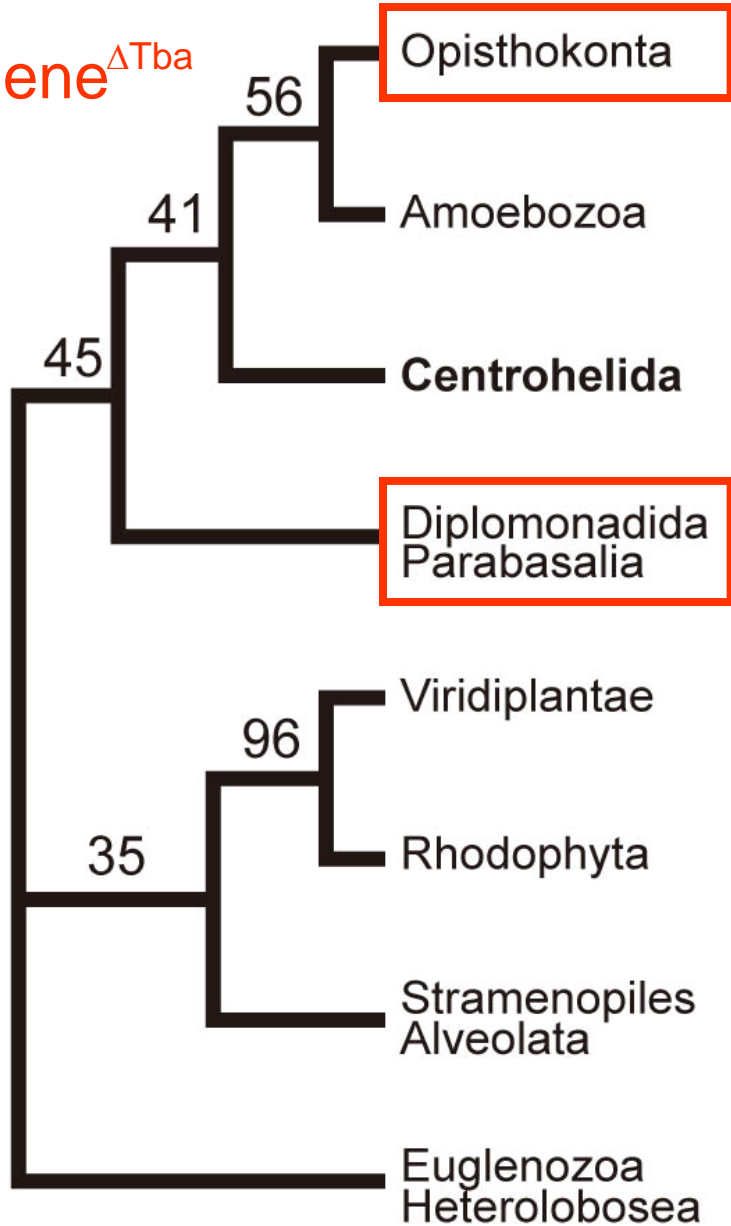
- Rhodophyta
- Viridiplantae
- Stramenopiles/Alveolata

But, the RELL BP support for this clade was **weak**



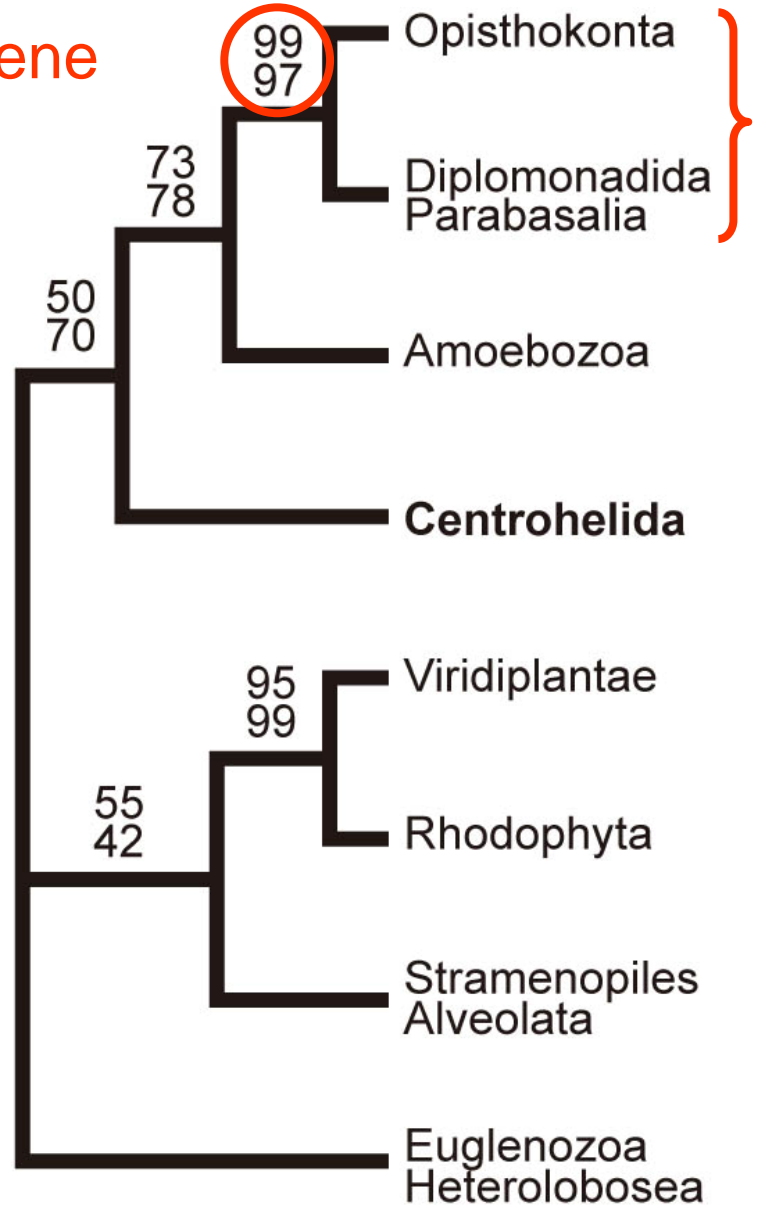
Gene-specific signals - α -tubulin-

5-gene ^{Δ Tba}



5-gene

Δ Tbb
 Δ Act

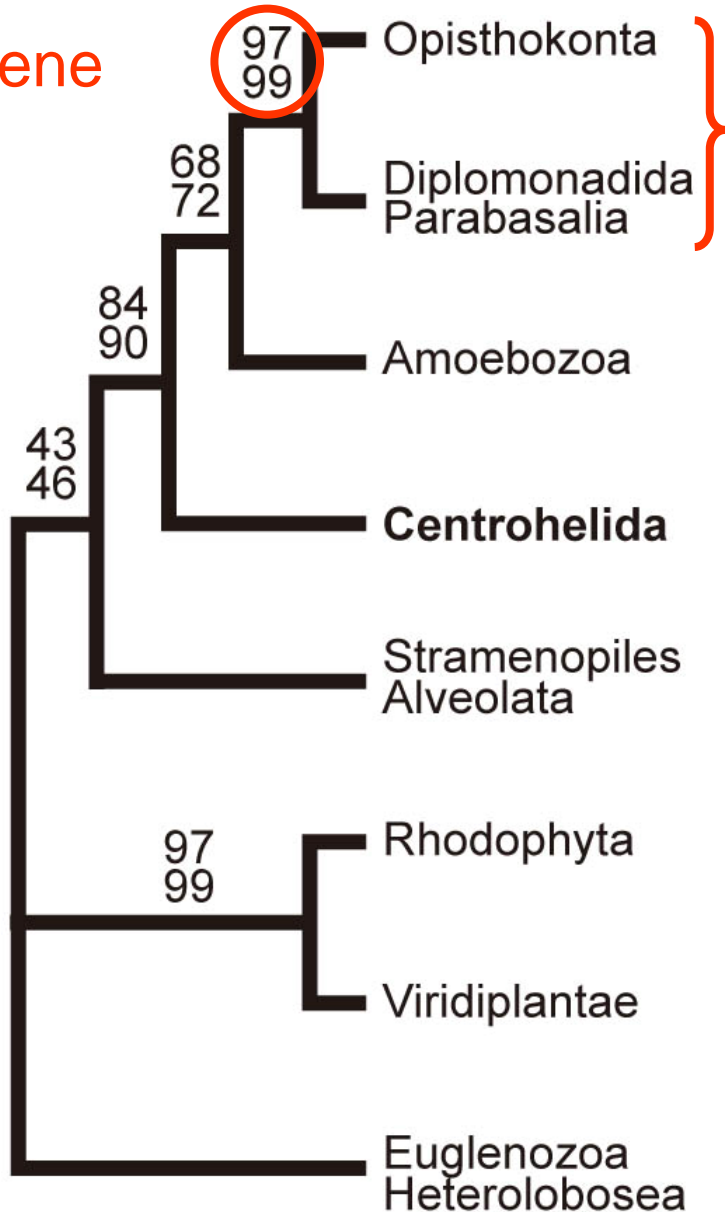




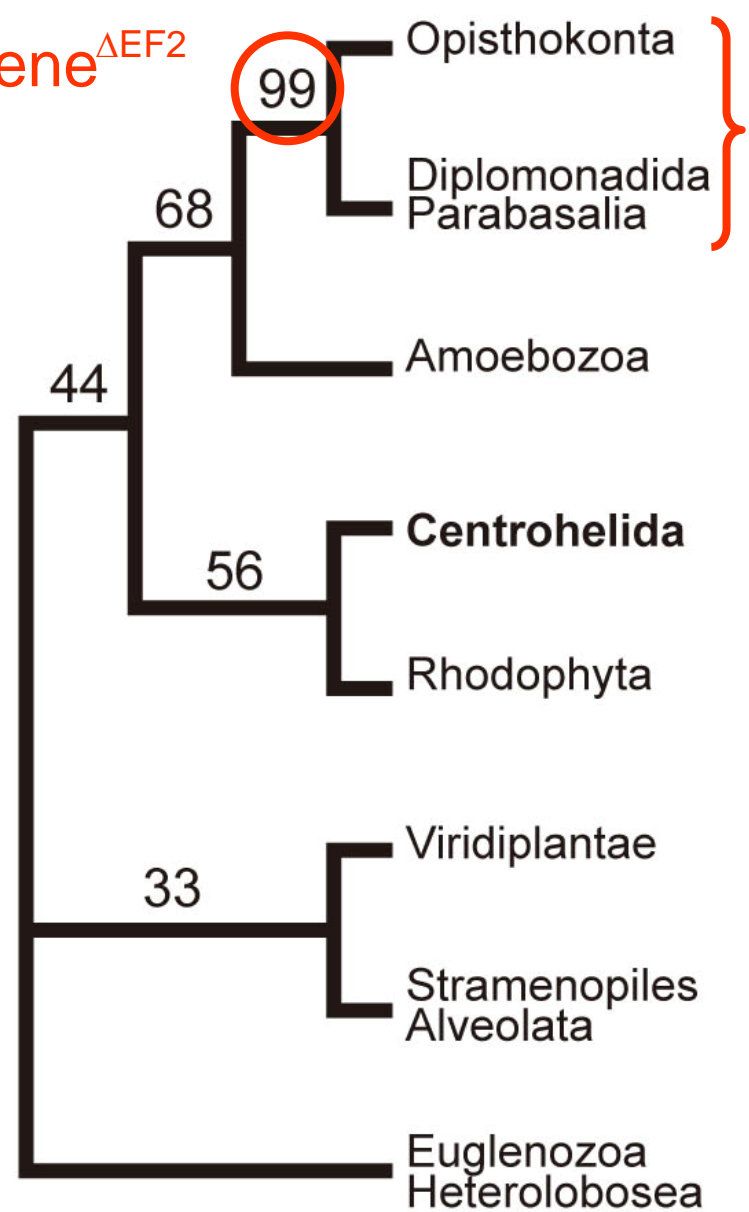
Gene-specific signals - α -tubulin-

5-gene

$\Delta 70$
 $\Delta 90$



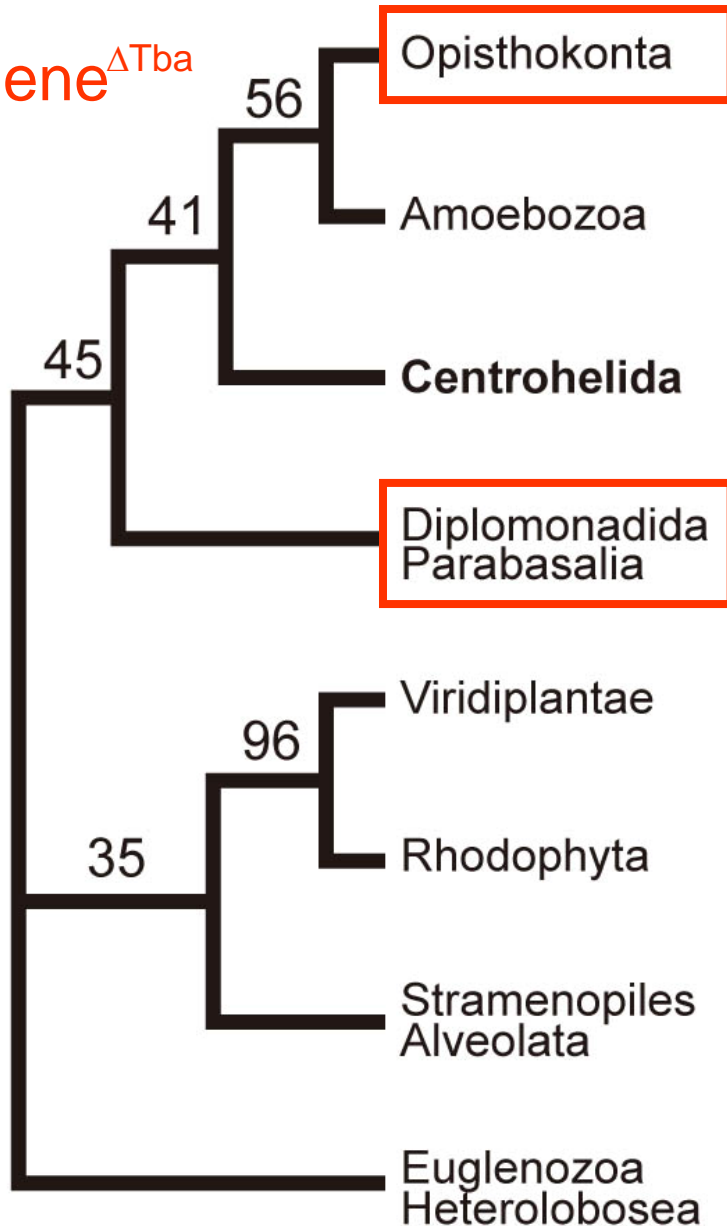
5-gene $\Delta EF2$





Gene-specific signals - α -tubulin-

5-gene ^{Δ Tba}



Multi-gene analyses...

→ including α -tubulin

The Dip/Par union formed a clade with the Opisthokonta with high RELL BP support

→ excluding α -tubulin

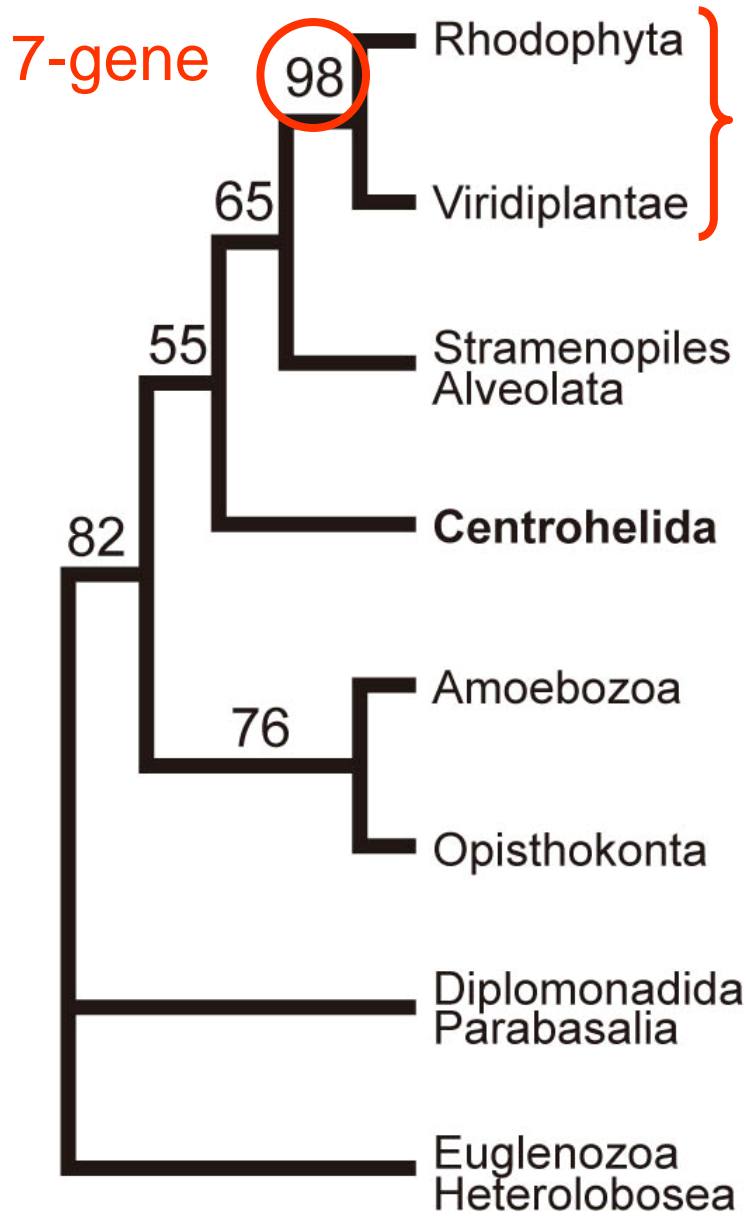
The Dip/Par union did not cluster with the Opisthokonta



The affinity between the Dip/Par union and the Opisthokonta is attributed solely to the α -tubulin signal



Gene-specific signals -EF2-



Multi-gene analyses...

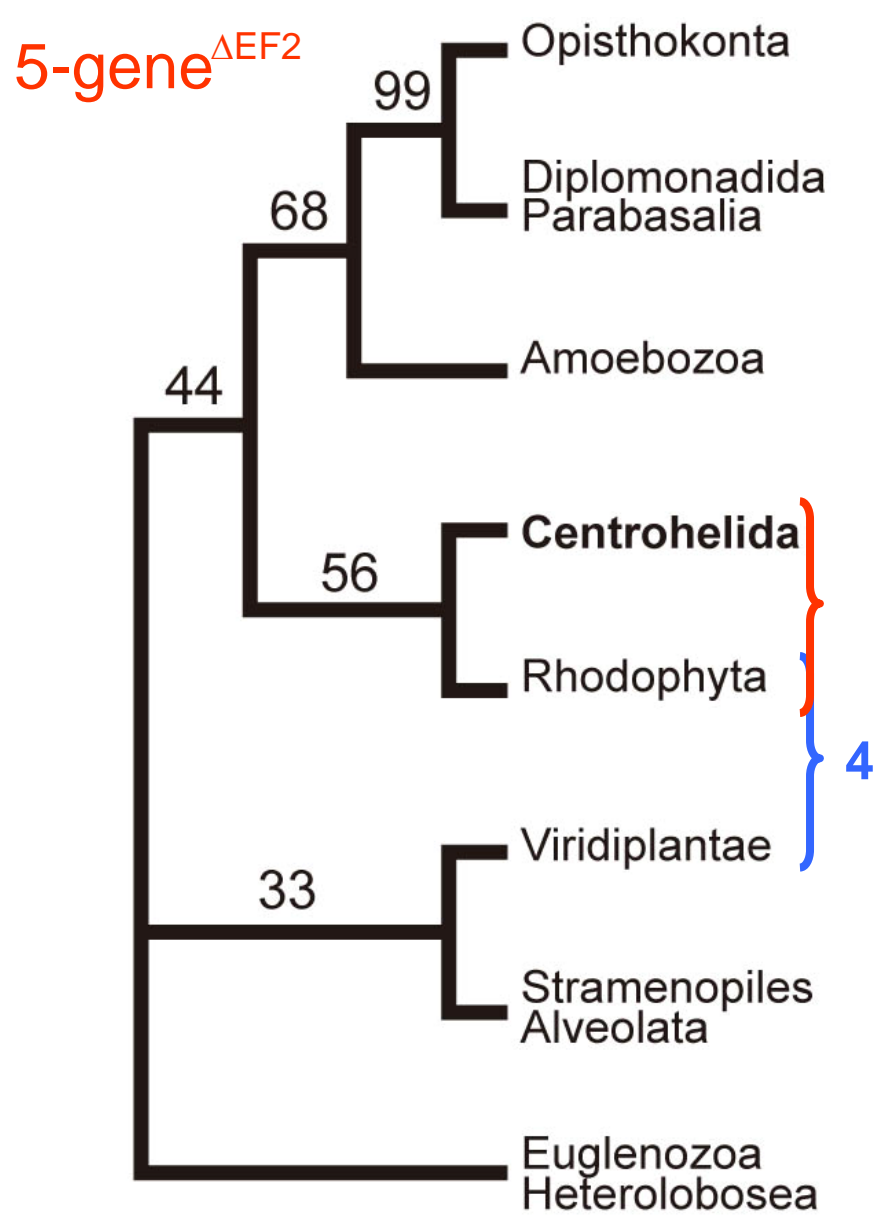
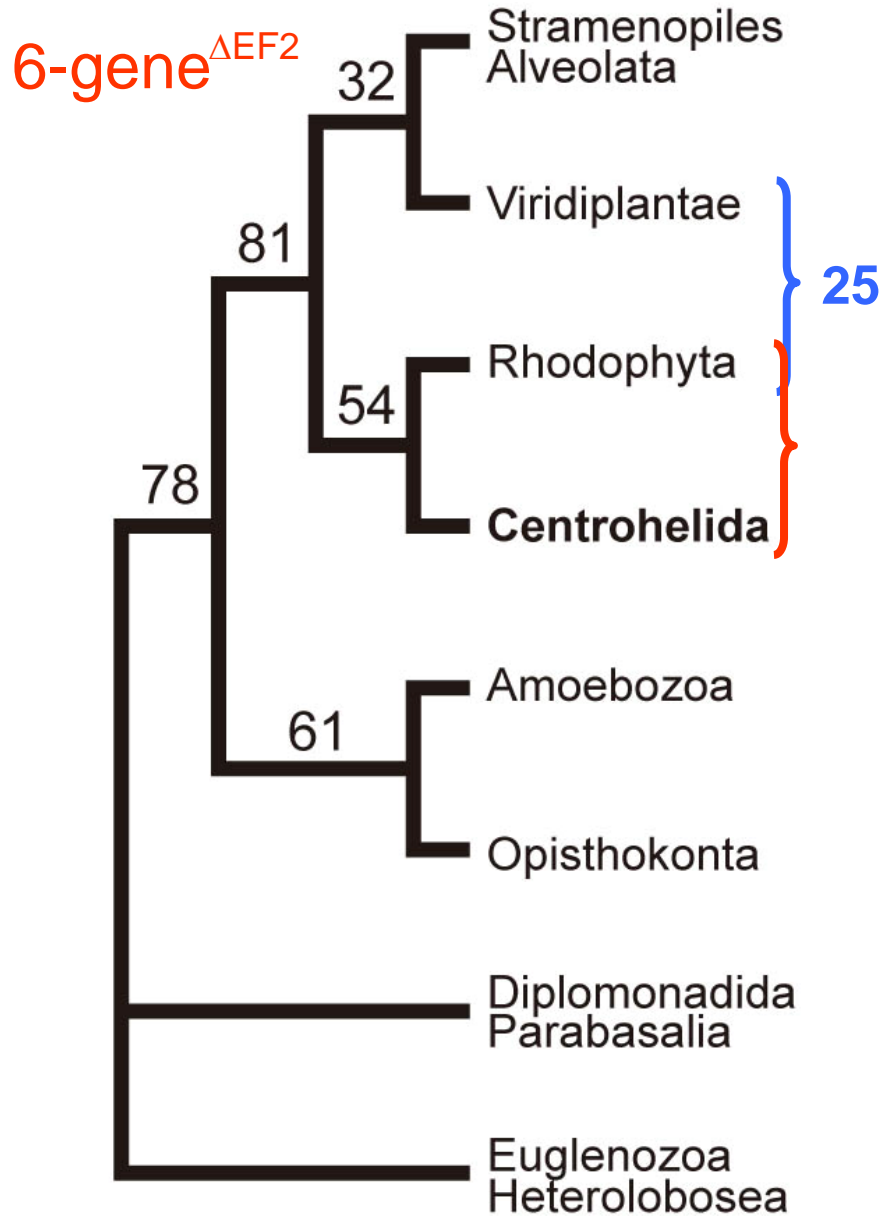
→ including EF2

Rhodophyta and Viridiplantae formed a monophyletic clade (Plantae) with high RELL BP support

The robust clade is attributed to the EF2 signal...?

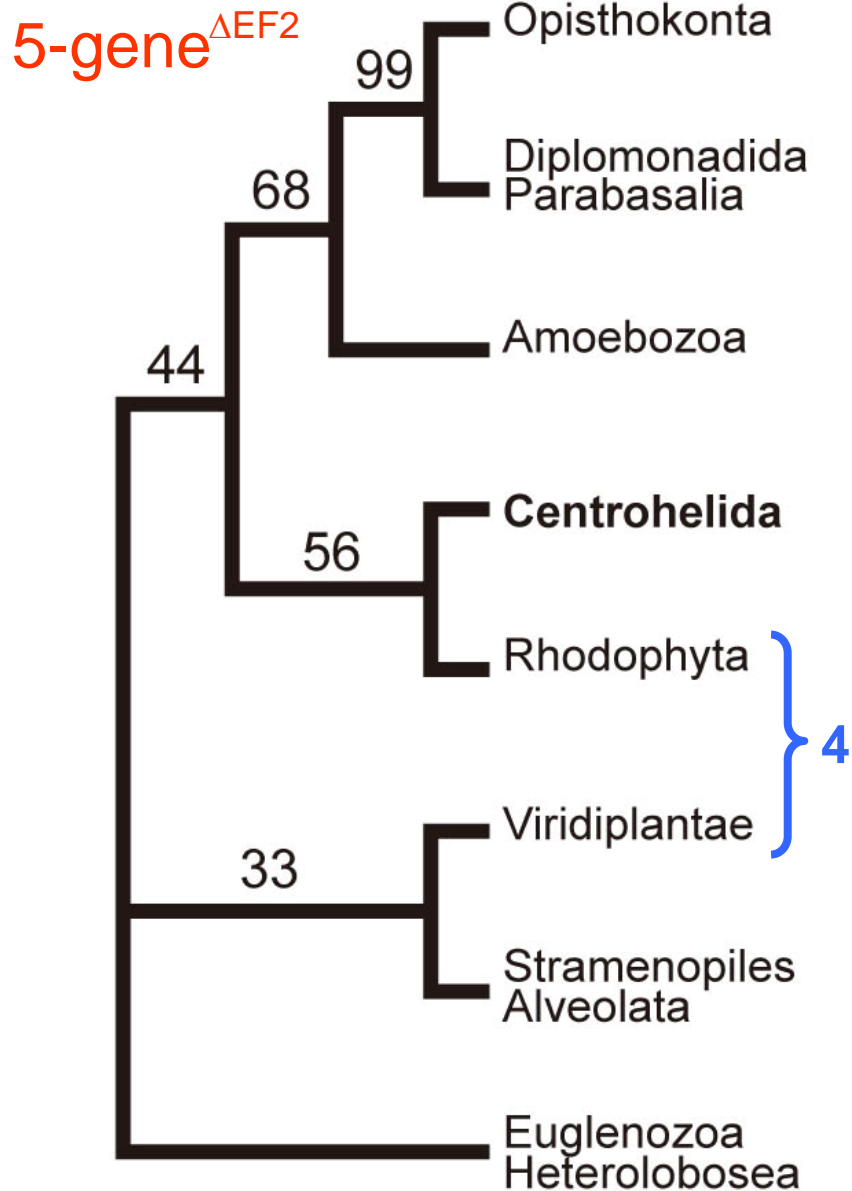


Gene-specific signals -EF2-





Gene-specific signals -EF2-



Multi-gene analyses...

→ including EF2

The Rhodophyta formed a clade with the Viridiplantae with high RELL BP support

→ excluding EF2

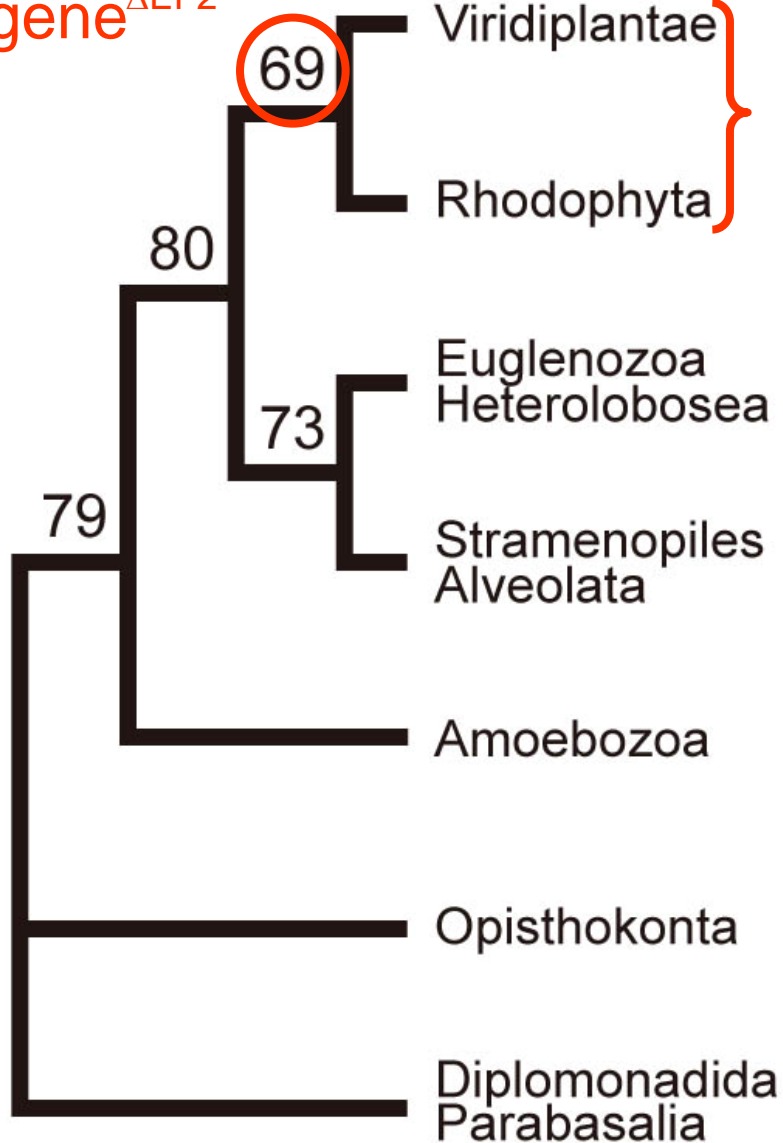
The Rhodophyta did not cluster with the Viridiplantae

Other genes do not support the monophyly...?



Gene-specific signals -EF2-

23-gene^{ΔEF2}



Multi-gene analyses...

→ excluding EF2

Rhodophyta formed a clade with Viridiplantae

Other genes support the monophyly of the Rhodophyta and Viridiplantae

Burger et al. (1999)

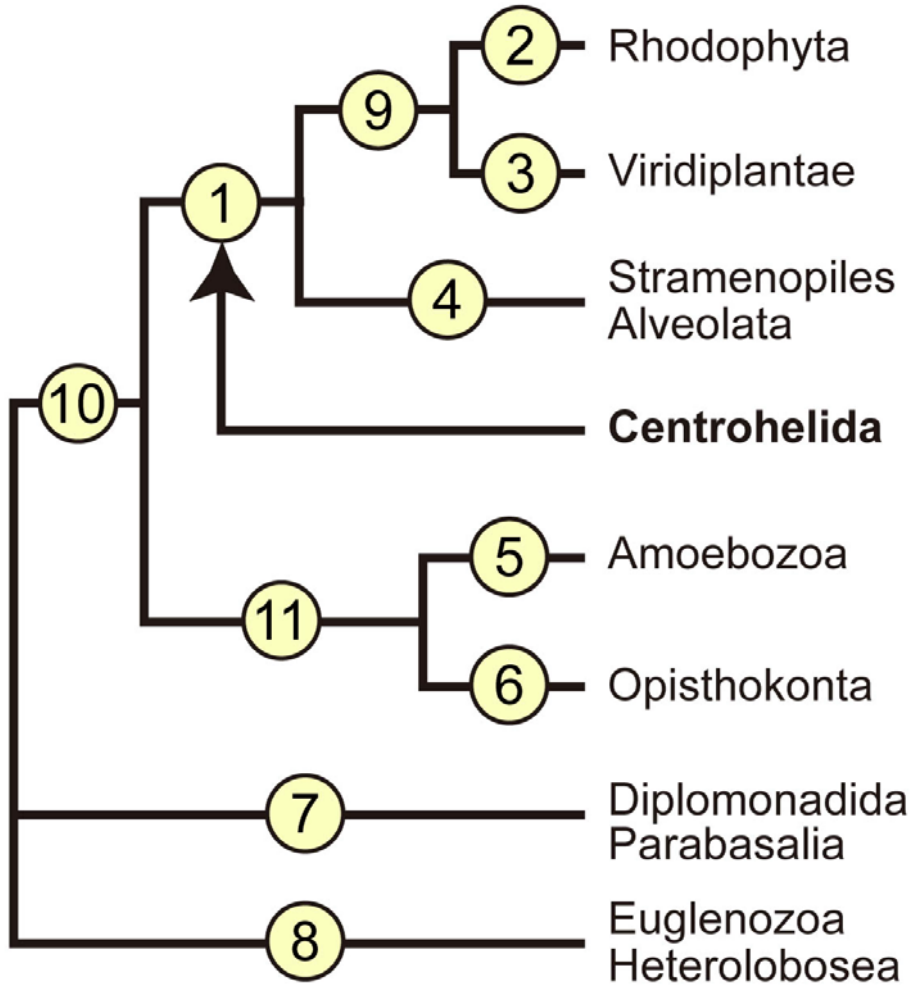
Rodríguez-Ezpeleta et al. (2005)



The phylogenetic signals in our data sets without EF2 are insufficient...



AU test



	7-gene	6-gene _{ASSU}	5-gene _{ΔTba}	5-gene _{ΔEFF2}	4-gene
1	★	○	○	○	○
2	●	●	●	★	★
3	●	●	●	○	○
4	○	○	○	○	○
5	●	●	●	●	●
6	○	○	○	○	○
7	●	●	○	●	●
8	●	○	○	○	○
9	○	○	○	●	○
10	●	○	○	○	○
11	○	★	★	○	○

★ best ○ $P \geq 0.05$ ● $0.01 \leq P < 0.05$ ● $P < 0.01$



Conclusion

- Multi-gene combined analyses of **Centrohelida**
 - concatenate model (Δ SSU rRNA)
 - separate model
 - AU test

the phylogenetic position of Centrohelida is **unclear**

- single-gene jackknifing

strong gene-specific signals are predominant in our multi-gene data

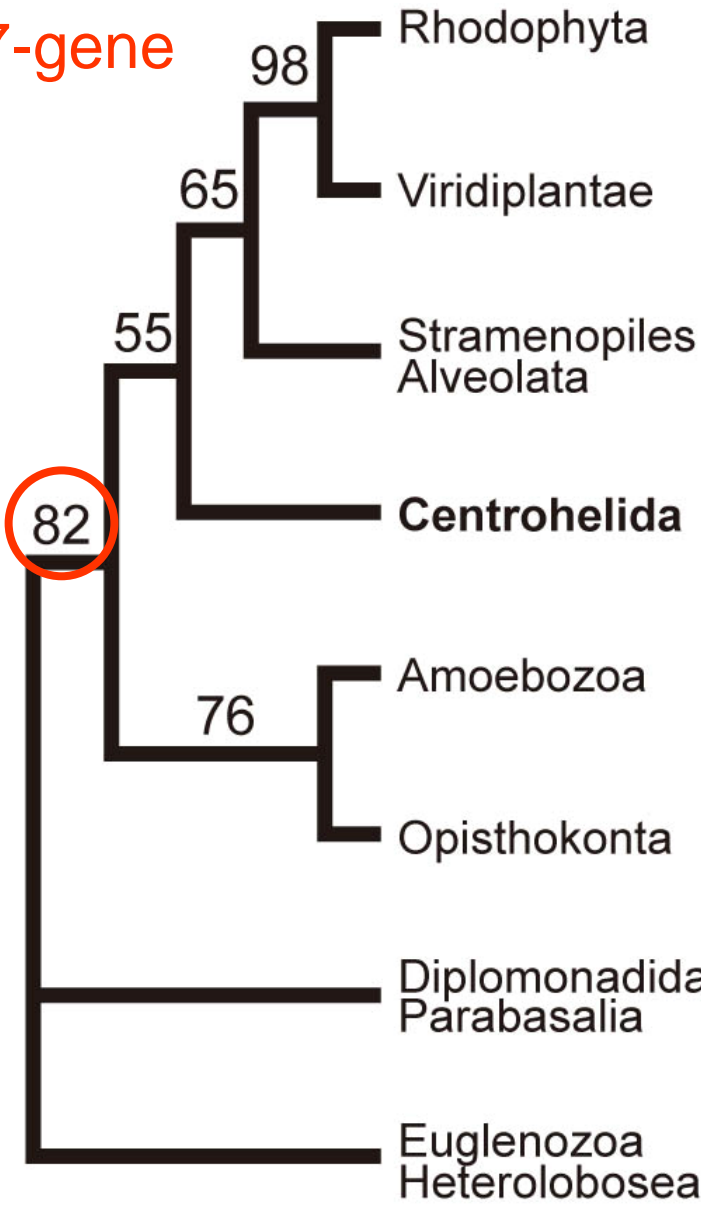


Larger scale multi-gene phylogenies are necessary to clarify the origin and evolution of Centrohelida

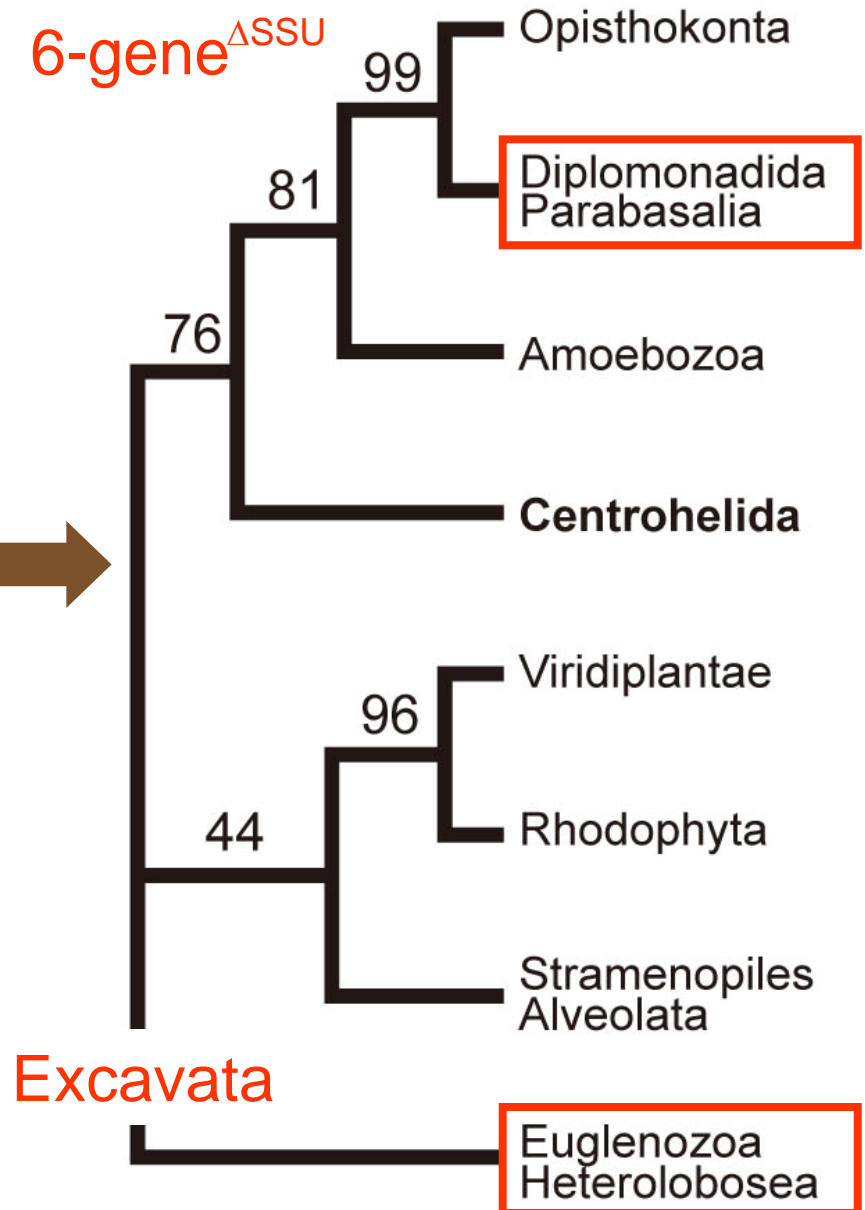


Gene-specific signals -SSU rRNA-

7-gene

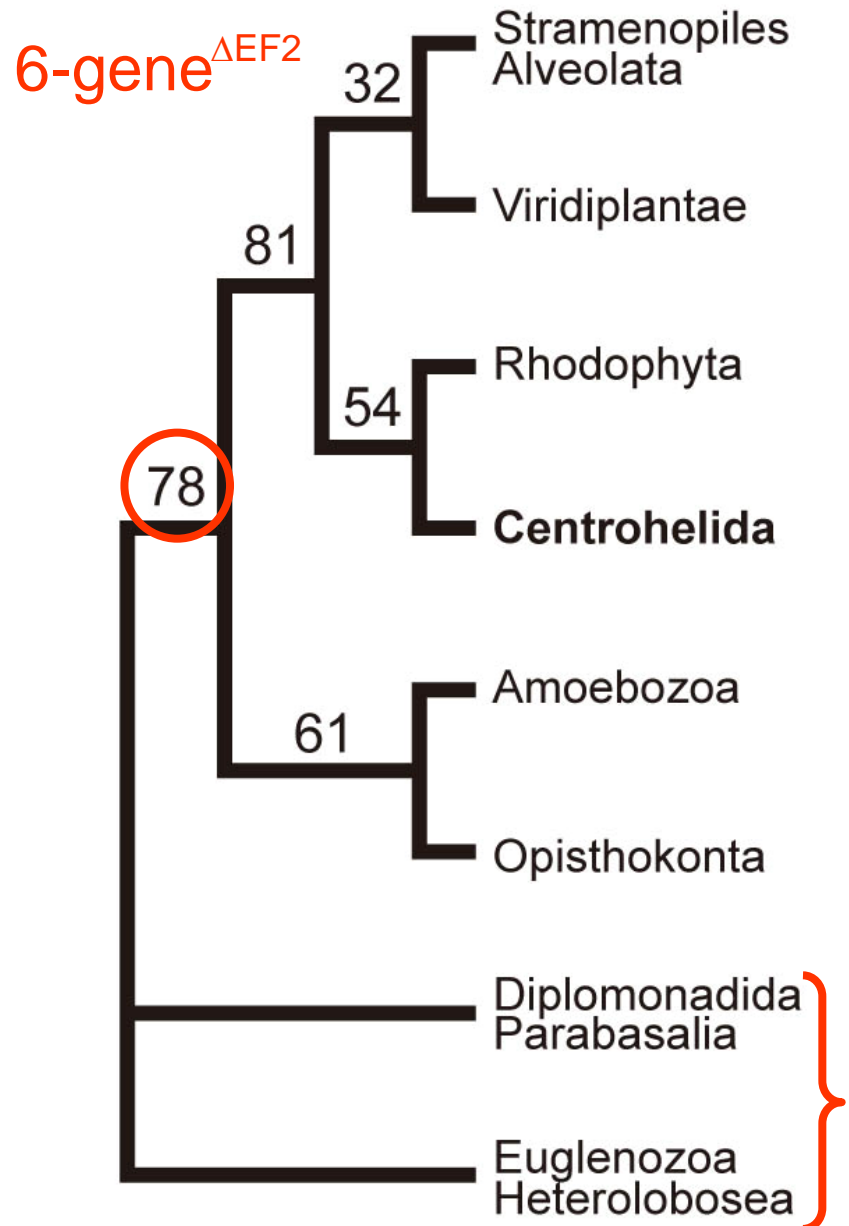
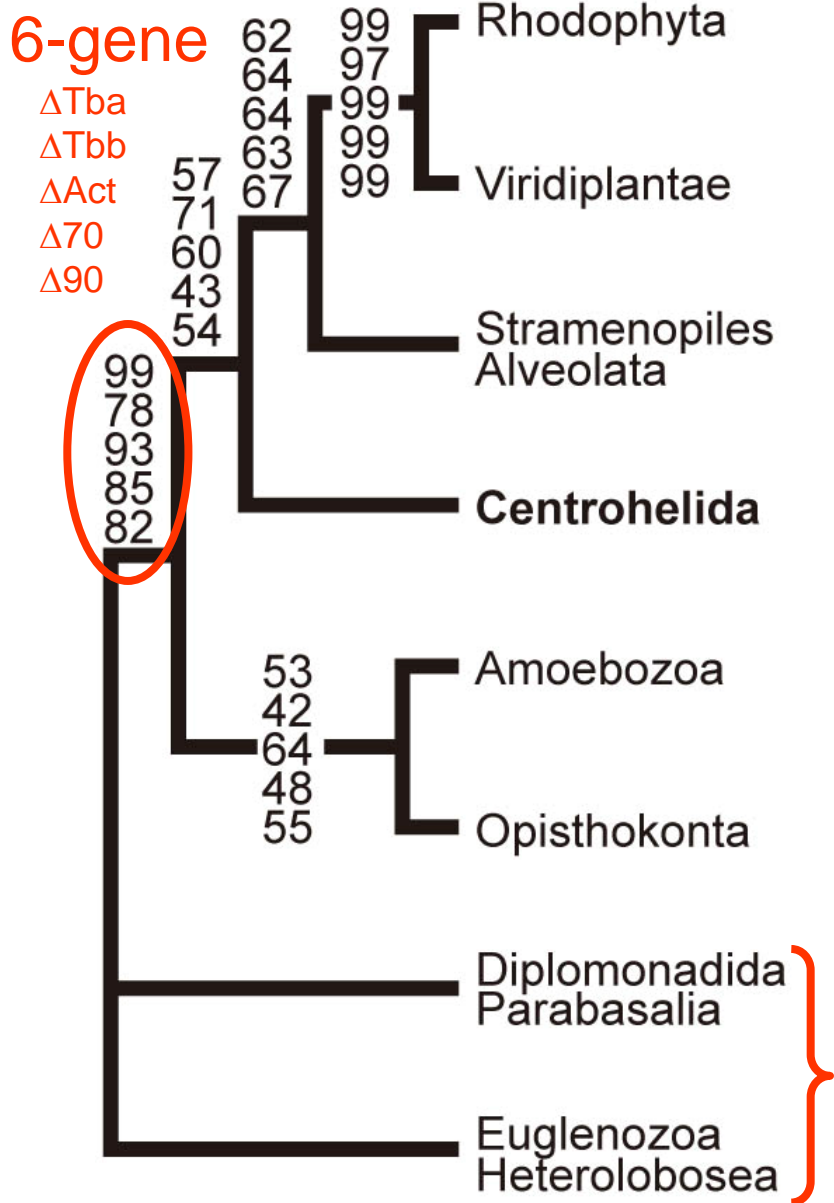


6-gene^{ΔSSU}



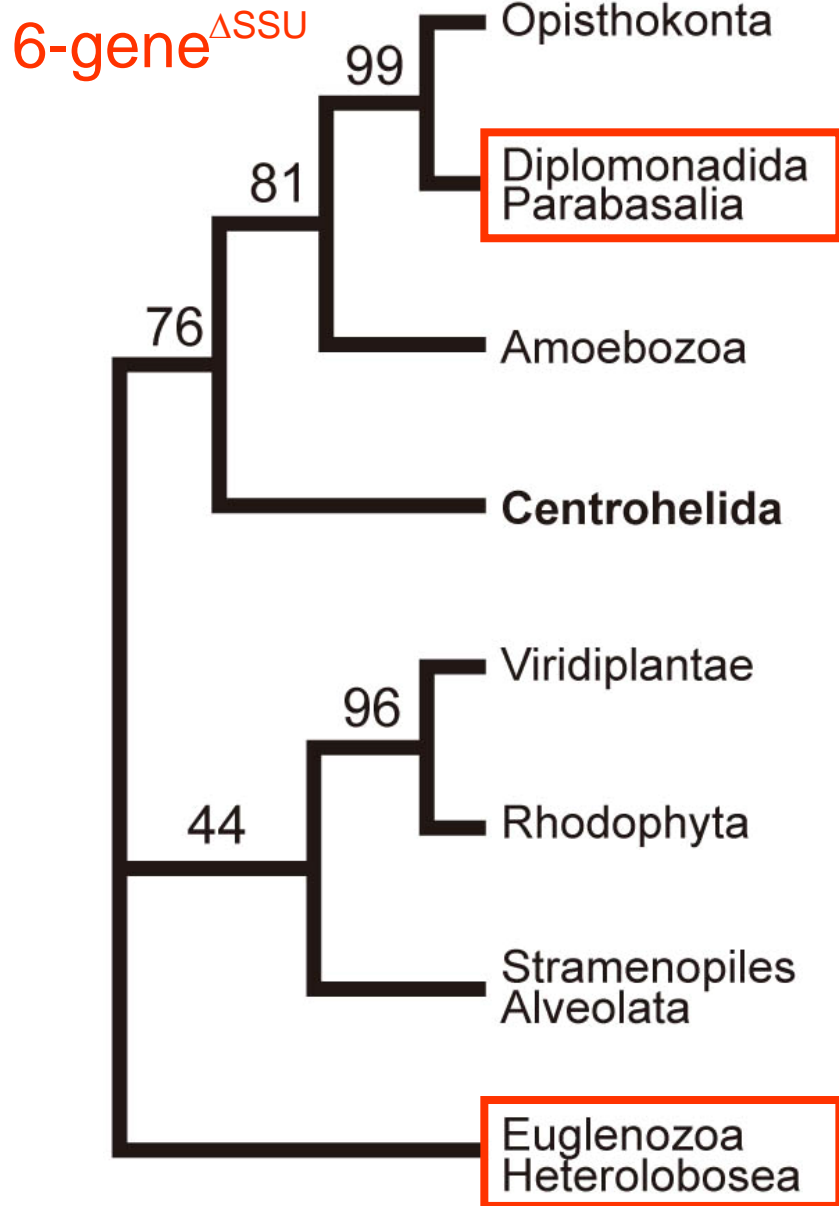


Gene-specific signals -SSU rRNA-





Gene-specific signals –SSU rRNA–



Multi-gene analyses...

→ including SSU rRNA

The Dip/Par union formed a clade with the Eug/Het union

→ excluding SSU rRNA

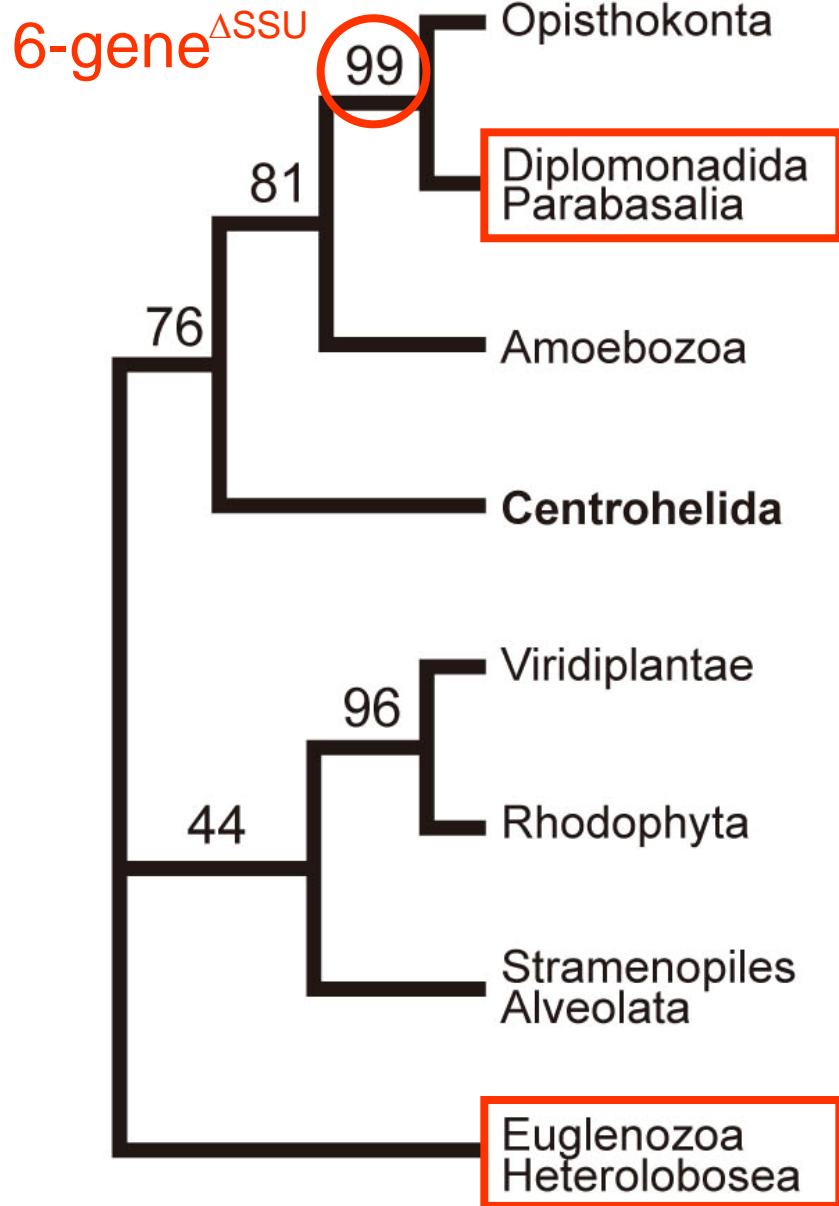
The Dip/Par union did not cluster with the Eug/Het union



The **Excavata** clade is supported only by SSU rRNA



Gene-specific signals - α -tubulin-



Multi-gene analyses...

→ excluding SSU rRNA

The Dip/Par union did not cluster with the Eug/Het union, but branched with the Opisthokonta

The Dip/Par union has a sister relationship with the Opisthokonta...?