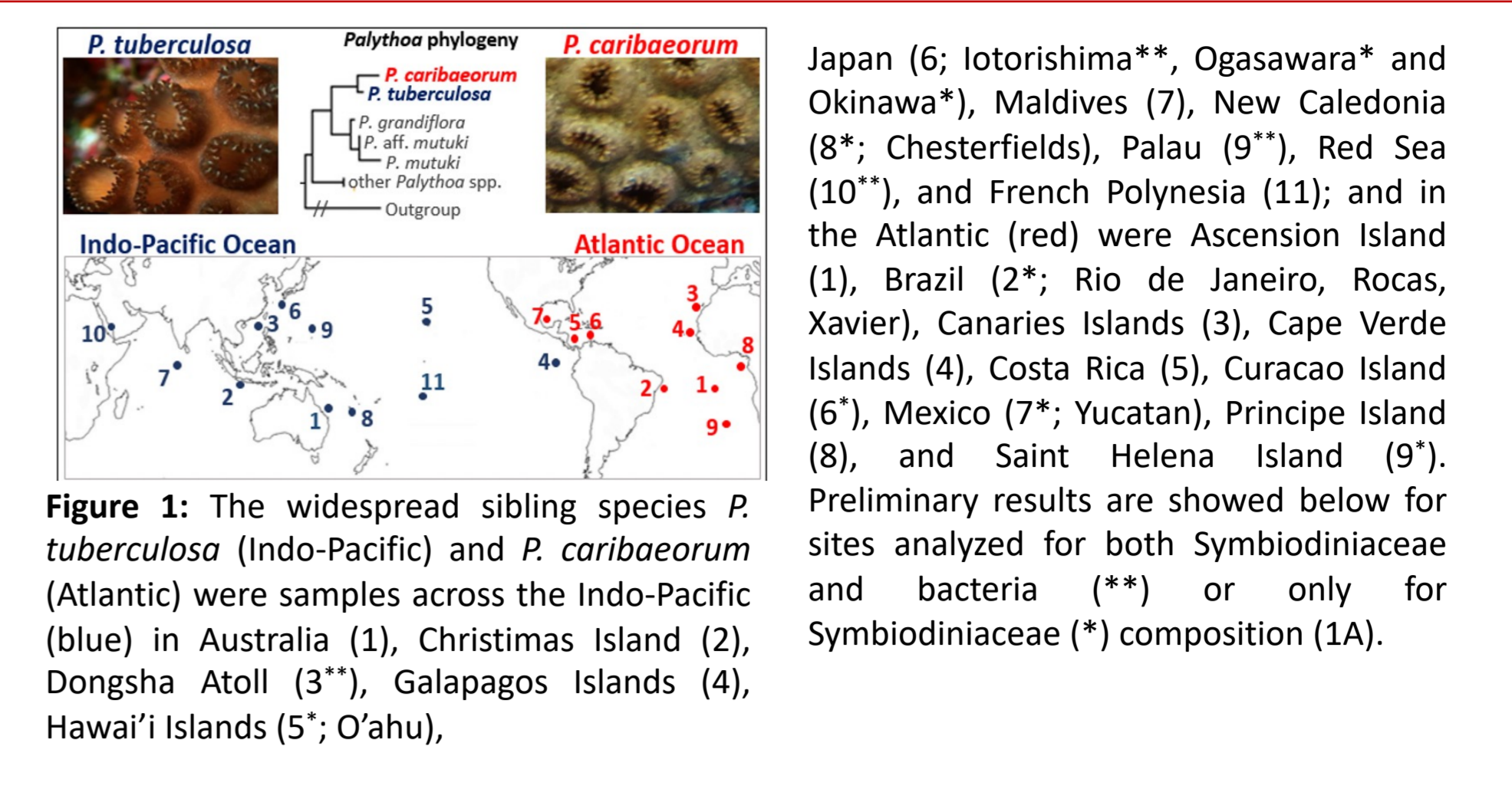


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Introduction

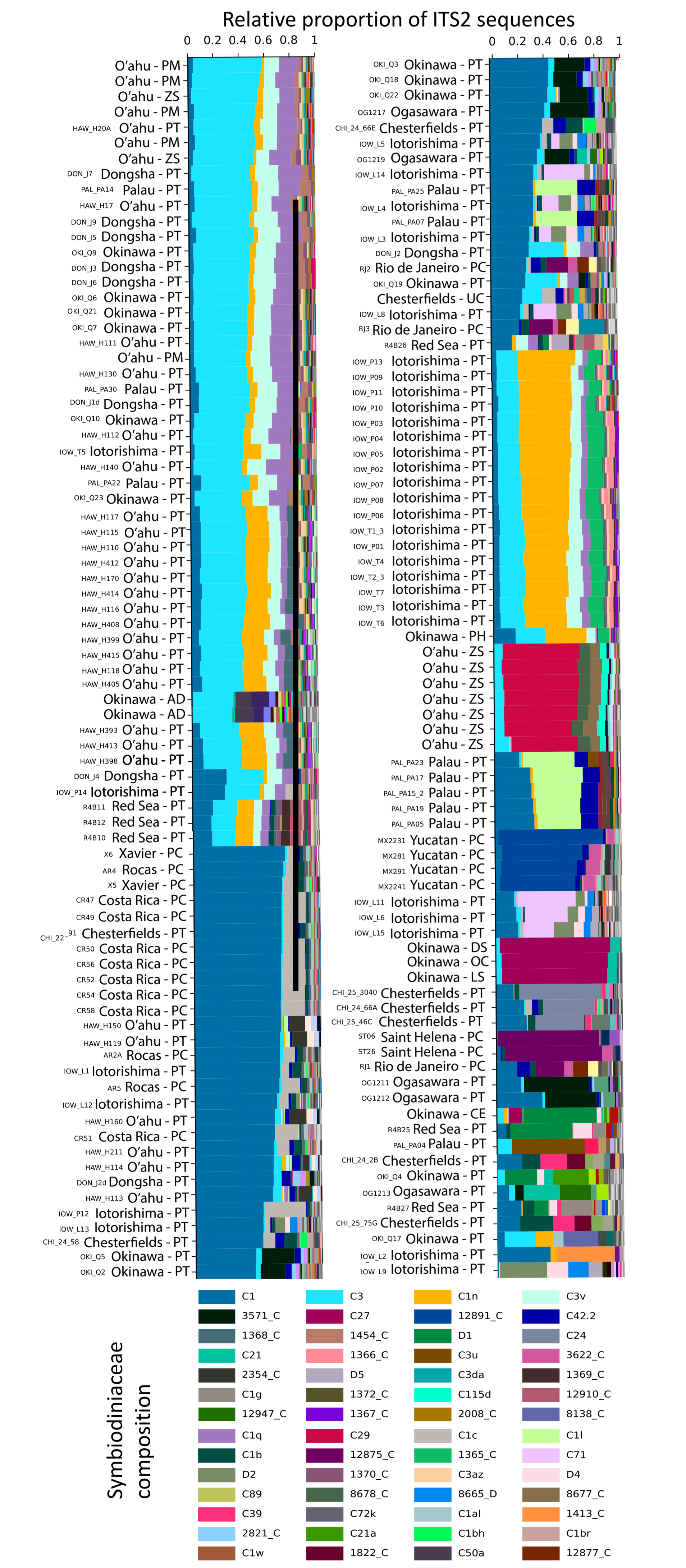
- Corals are heterotrophic animals of the phylum Cnidaria. They can associate with symbiotic photosynthetic dinoflagellates of the family Symbiodiniaceae, which exchange nutrients with their coral host¹
- Diverse environmental conditions, such as seawater temperature and pH, influence the composition of the coral microbiome community (e.g., Symbiodiniaceae and bacteria)
- We sampled the most widespread *Palythoa* sibling species² (Hexacorallia: Zoantharia) as a model system to examine global diversity of symbionts in the same coral host across regions with different seawater conditions (Fig. 1)



- How does the symbiont diversity differ across regions of the Atlantic and Indo-Pacific?
- How does the symbiont diversity from the region with the warmest seawater temperature (Red Sea³) and low pH (Iotorishima⁴) differ from other locations?

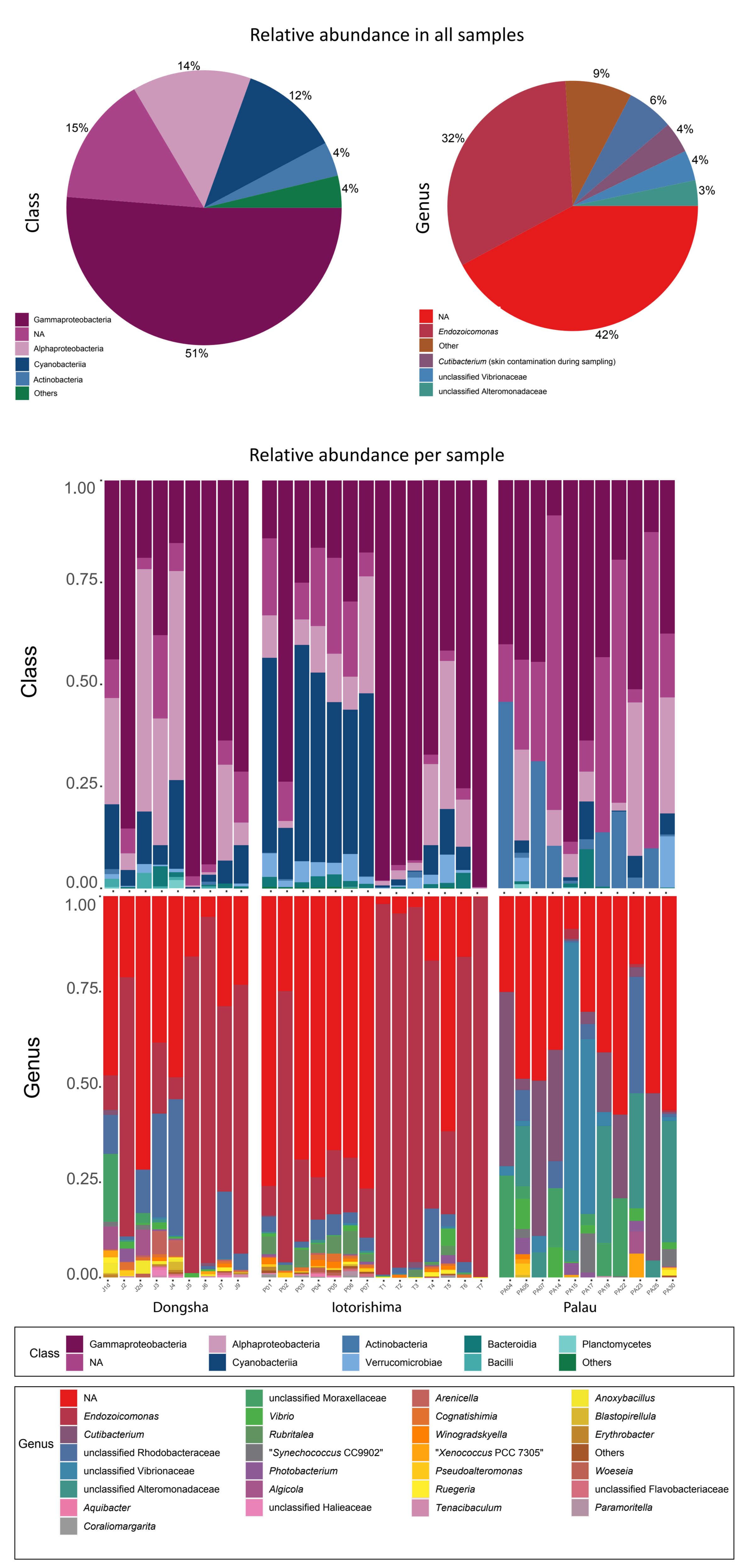
Symbiodiniaceae (Figs. 2 and 4A)

- Species of *Cladocopium*, *Durusdinium* and *Symbiodinium*
- Coral samples from Atlantic and Indo-Pacific have different Symbiodiniaceae composition (p<0.001)
- Coral samples from Red Sea and Iotorishima have different Symbiodiniaceae composition (p<0.001) from other regions



Bacteria (Figs. 3 and 4C)

- Endozoicomonas*: likely mutualistic bacterium of corals⁴
- Vibrio* spp.: opportunistic pathogens⁵
- Coral samples from the Indo-Pacific analyzed so far (Dongsha, Iotorishima and Palau) have different bacterial composition (p<0.001)
- Most samples from Iotorishima had lower diversity than Dongsha and Palau



Dendrogram of symbiont diversity across locations

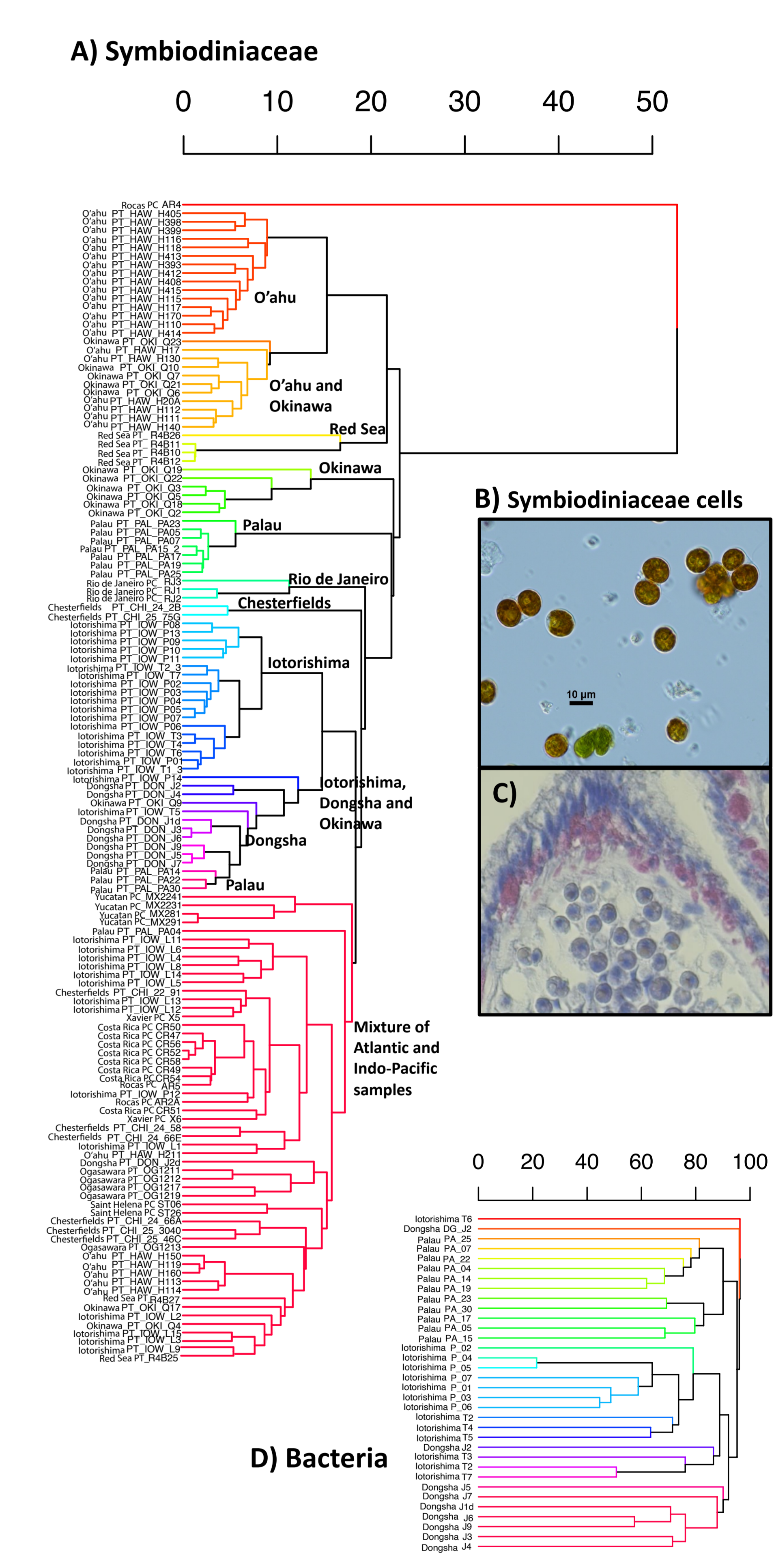


Figure 2: Relative proportion of ITS2 sequences of Symbiodiniaceae associated with each coral sample. Only the most abundant (>0.01% of all reads) sequences are displayed. Labels indicate the Symportal⁷ database ID number followed by the genus the sequence is from (C=*Cladocopium* and D=*Durusdinium*). Host *Palythoa* species codes are PT (*P. tuberculosa*) and PC (*P. caribbaeorum*), in addition to outgroups PM (*P. mutuki*), PH (*P. heliodiscus*), AD (*Acropora digitifera*), CE (*Ctenactis echinata*), DS (*Danafungia scaposa*), LS (*Lytrophyllon scabra*), OC (*Pobadacia crustacea*), UC (*Umimayanthus chanpuru*), ZS (*Zoanthus sansibaricus*). Symbiodiniaceae compositions was estimated using MiSeq sequencing of the ITS2 gene.

Figure 4: Cluster dendrograms of the SIMPROF-test with Bray-Curtis similarity measure (x-axis) and the coral samples (y-axis) for the microbial community of Symbiodiniaceae (A; ITS2) and bacteria (D; 16S rDNA). Colors indicate statically significant clusters (p<0.001). Images of Symbiodiniaceae cells (B-C). Credits 4B: MEA Santos and 4C: Y Masukagami.

Methods

- Molecular analyses:
 - DNA extraction of coral samples, PCR amplification (ITS2 for Symbiodiniaceae⁷ and 16SrRNA for bacteria⁸) and MiSeq sequencing
 - Bioinformatics: Symportal⁷ (ITS2) and Dadaist2⁹ (16S-rRNA) pipelines
- Significance statistics tests and clusters: Bray-Curtis, PERMANOVA, SIMPROF-test (999 permutations)

References

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