



DATA NOTE

# The chromosomal genome sequence of the sponge *Phakellia ventilabrum* (Linnaeus, 1767) and its associated microbial metagenome sequences

[version 1; peer review: 2 approved]

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## Abstract

We present a genome assembly from a specimen of *Phakellia ventilabrum* (Porifera; Demospongiae; Bubarida; Bubaridae). The genome sequence has a total length of 211.92 megabases. Most of the assembly (99.97%) is scaffolded into 25 chromosomal pseudomolecules. The mitochondrial genome has also been assembled and is 24.36 kilobases in length. Gene annotation of this assembly by Ensembl identified 21 622 protein-coding genes. Thirty-three binned genomes were generated from the metagenome assembly, of which eight were classified as high-quality metagenome assembled genomes (MAGs) and of which four of the MAGs are fully circular. The MAGs were taxonomically assigned to Pseudomonadota (i.e. Candidatus Poriferihabitaceae), Nitrospirota, Nitrospina, and

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Approval Status

|                  | 1                    | 2                    |
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the archaeal Nitrosopumilus clade.

### Keywords

Phakellia ventilabrum, genome sequence, chromosomal, Bubarida, microbial metagenome



This article is included in the [Tree of Life gateway](#).

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## Species taxonomy

Eukaryota; Opisthokonta; Metazoa; Porifera; Demospongiae; Heteroscleromorpha; Bubarida; Bubaridae; *Phakellia*; *Phakellia ventilabrum* (Linnaeus, 1767) (NCBI:txid942649)

## Background

*Phakellia ventilabrum* (Linnaeus, 1767), is a deep-water demosponge, common on rock-sand habitats across the North Atlantic Ocean, although it has also been reported in the Western Mediterranean Sea (de Voogd *et al.*, 2024; Maldonado *et al.*, 2017; Sánchez *et al.*, 2009). This fan-shaped axinellid sponge has a wide bathymetric distribution from shallow waters of 10 m to depths reaching almost 1,900 m (Prado *et al.*, 2020). *Phakellia ventilabrum* can form dense aggregations and is considered as a Vulnerable Marine Ecosystem (VME) indicator species (Maldonado *et al.*, 2017).

*Phakellia ventilabrum* is an oviparous and gonochoristic (i.e. separate sexes) species, reproducing in May and September, with potentially lecithotrophic larva or direct development, given the relatively high amount of nutrients (mainly lipids) accumulated in the egg during vitellogenesis (Koutsouveli *et al.*, 2022). Its dispersal ability and the main oceanographic currents in the North Atlantic Ocean drive the genetic connectivity and genetic diversity patterns of this species. Two distinct genetic populations have been identified, one comprising individuals from the Cantabrian Sea to Roscoff, and another comprising sponges from the south of the British Islands up to the north of Norway (Taboada *et al.*, 2023). The genetic break detected in this genome-wide analysis is explained by oceanographic models and indicated a high degree of isolation. The low genetic diversity for the Cantabrian *P. ventilabrum* might result in compromised resilience for this population considering the current climate change scenario predictions for the southern areas (Taboada *et al.*, 2023).

*Phakellia ventilabrum* is an optimal natural sampler for environmental DNA (eDNA). Collections along the North Atlantic have identified unprecedented levels of metazoan diversity within this species, with more than 400 species detected by eDNA methods (Gallego *et al.*, 2024; Neave *et al.*, 2023). Because *P. ventilabrum* is a Low Microbial Abundance (LMA) species, its presumed high filtration rates may be favourable for accumulating a higher amount of eDNA (Weisz *et al.*, 2008). The microbiome of *P. ventilabrum* is dominated by Pseudomonadota and Nitrospirota and it displays sponge species-specificity in comparison to closely-related species from the same collection sites (Busch *et al.*, 2022; Taboada *et al.*, 2022).

The availability of the chromosome-level genome of *P. ventilabrum* will help to further explore population connectivity and adaptation patterns of this key-stone species, which will have direct implications for conservation purposes. The availability of another chromosome-level genome for *P. ventilabrum* (Mc Cartney *et al.*, 2024) now provides a wealth of data for comparative genomic analyses to address evolutionary and ecological questions with unprecedented detail. In addition, this genome will be particularly useful for investigating the evolution

of biological traits in Demospongiae as well as for clarifying the phylogenetic relationships within the family Bubaridae. This last case is especially relevant for the polyphyletic genus *Phakellia* (Morrow *et al.*, 2012; Redmond *et al.*, 2013; Taboada *et al.*, 2022) and with *P. ventilabrum* representing the type species. Finally, the microbial genomic data provided here will enable future studies on the microbial composition and functionality of this species across its geographic and phylogenetic distribution, (Hentschel *et al.*, 2012; Díez-Vives & Riesgo, 2024; Thomas *et al.*, 2016).

## Genome sequence report

### Sequencing data

The genome of a specimen of *Phakellia ventilabrum* (Figure 1) was sequenced using Pacific Biosciences single-molecule HiFi long reads, generating 26.14 Gb from 2.84 million reads. Based on the estimated genome size, the sequencing data provided approximately 93 coverage of the genome. Chromosome conformation Hi-C data produced 116.05 Gb from 768.51 million reads. RNA sequencing data were also generated and are available in public sequence repositories. Table 1 summarises the specimen and sequencing information.

### Assembly statistics

The primary haplotype was assembled, and contigs corresponding to an alternate haplotype were also deposited in INSDC databases. The assembly was improved by manual curation, which corrected 26 misjoins or missing joins and removed 6 haplotypic duplications. These interventions reduced the total assembly length by 4.22%, decreased the scaffold count by 83.33%, also decreasing the scaffold N50 by 0.58%. The final assembly has a total length of 211.92 Mb in 26 scaffolds, with 59 gaps, and a scaffold N50 of 8.44 Mb (Table 2).

The snail plot in Figure 2 provides a summary of the assembly statistics, indicating the distribution of scaffold lengths



**Figure 1. *Phakellia ventilabrum* specimen used for genome sequencing.** The image depicts the specimen immediately after collection onboard ship and prior to deep-freezing. The size of the sponge body is about 20–25 cm in diameter. Image taken by Kathrin Busch.

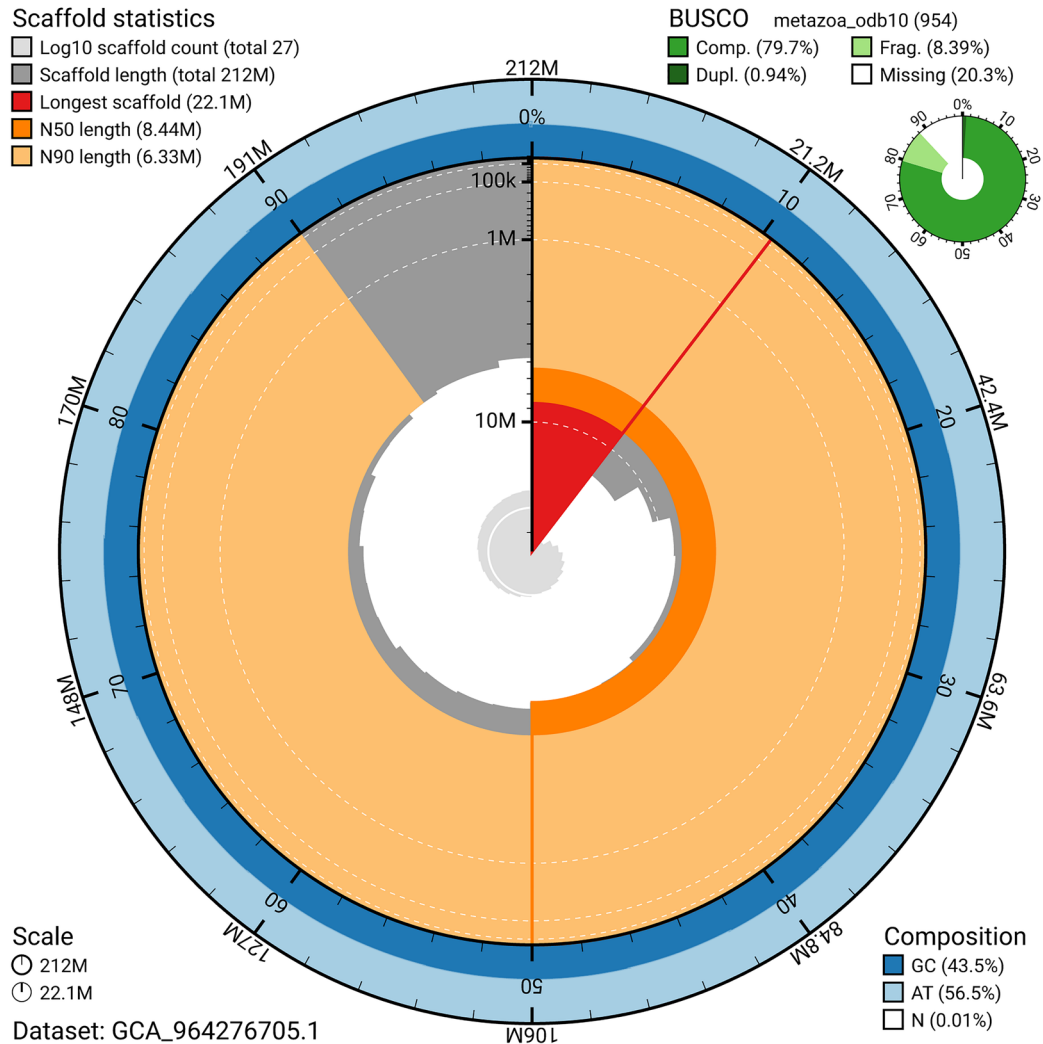
**Table 1. Specimen and sequencing data for *Phakellia ventilabrum*.**

| Project information         |                       |                     |                 |
|-----------------------------|-----------------------|---------------------|-----------------|
| Study title                 | Phakellia ventilabrum |                     |                 |
| Umbrella BioProject         | PRJEB78906            |                     |                 |
| Species                     | Phakellia ventilabrum |                     |                 |
| BioSpecimen                 | SAMEA115470870        |                     |                 |
| NCBI taxonomy ID            | 942649                |                     |                 |
| Specimen information        |                       |                     |                 |
| Technology                  | ToLID                 | BioSample accession | Organism part   |
| PacBio long read sequencing | odPhaVent3            | SAMEA115470889      | Somatic tissue  |
| Hi-C sequencing             | odPhaVent3            | SAMEA115470888      | Somatic tissue  |
| RNA sequencing              | odPhaVent3            | SAMEA115470887      | Somatic tissue  |
| Sequencing information      |                       |                     |                 |
| Platform                    | Run accession         | Read count          | Base count (Gb) |
| Hi-C Illumina NovaSeq X     | ERR13494022           | 7.69e+08            | 116.05          |
| PacBio Revio                | ERR13510315           | 2.84e+06            | 26.14           |
| RNA Illumina NovaSeq X      | ERR14711437           | 7.37e+07            | 11.13           |

**Table 2. Genome assembly data for *Phakellia ventilabrum*.**

| Genome assembly                                |                                               |
|------------------------------------------------|-----------------------------------------------|
| Assembly name                                  | odPhaVent3.1                                  |
| Assembly accession                             | GCA_964276705.1                               |
| Alternate haplotype accession                  | GCA_964276595.1                               |
| Assembly level for primary assembly            | chromosome                                    |
| Span (Mb)                                      | 211.92                                        |
| Number of contigs                              | 85                                            |
| Number of scaffolds                            | 26                                            |
| Longest scaffold (Mb)                          | 22.06                                         |
| Assembly metric                                | Measure                                       |
| Contig N50 length                              | 5.01 Mb                                       |
| Scaffold N50 length                            | 8.44 Mb                                       |
| Consensus quality (QV)                         | Primary: 63.4; alternate: 61.4; combined 62.3 |
| BUSCO*                                         | C:79.7%[S:78.7%,D:0.9%],F:8.4%,M:11.9%,n:954  |
| Percentage of assembly assigned to chromosomes | 99.98%                                        |
| Organelles                                     | Mitochondrial genome: 24.36 kb                |

\* BUSCO scores based on the metazoa\_odb10 BUSCO set using version 5.5.0. C = complete [S = single copy, D = duplicated], F = fragmented, M = missing, n = number of orthologues in comparison.



**Figure 2. Genome assembly of *Phakellia ventilabrum*, odPhaVent3.1: metrics.** The BlobToolKit snail plot provides an overview of assembly metrics and BUSCO gene completeness. The circumference represents the length of the whole genome sequence, and the main plot is divided into 1,000 bins around the circumference. The outermost blue tracks display the distribution of GC, AT, and N percentages across the bins. Scaffolds are arranged clockwise from longest to shortest and are depicted in dark grey. The longest scaffold is indicated by the red arc, and the deeper orange and pale orange arcs represent the N50 and N90 lengths. A light grey spiral at the centre shows the cumulative scaffold count on a logarithmic scale. A summary of complete, fragmented, duplicated, and missing BUSCO genes in the metazoa\_odb10 set is presented at the top right. An interactive version of this figure is available at [https://blobtoolkit.genomehubs.org/view/GCA\\_964276705.1/dataset/GCA\\_964276705.1/snail](https://blobtoolkit.genomehubs.org/view/GCA_964276705.1/dataset/GCA_964276705.1/snail).

and other assembly metrics. Figure 3 shows the distribution of scaffolds by GC proportion and coverage.

Most of the assembly sequence (99.98%) was assigned to 25 chromosomal-level scaffolds. These chromosome-level scaffolds, confirmed by Hi-C data, are named according to size (Figure 4; Table 3).

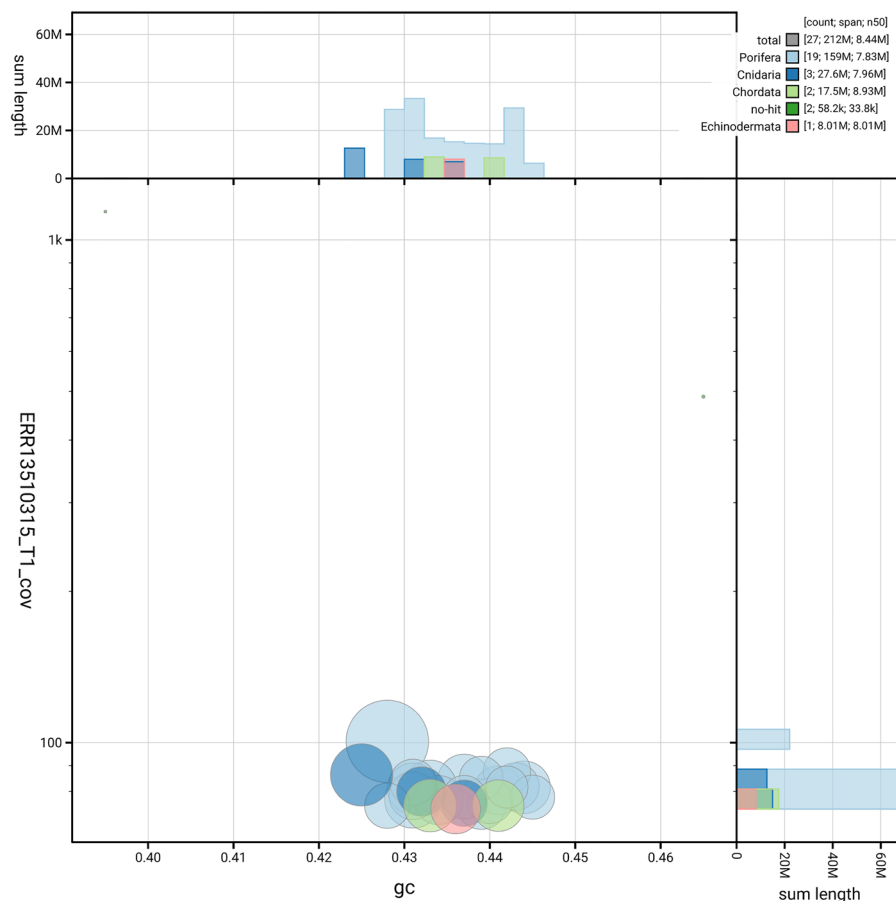
The mitochondrial genome was also assembled. This sequence is included as a contig in the multifasta file of the genome submission and as a standalone record in GenBank.

### Assembly quality metrics

The primary haplotype has a QV of 63.4, and the combined primary and alternate assemblies achieve an estimated QV of 62.3. BUSCO v.5.5.0 analysis using the metazoa\_odb10 reference set ( $n = 954$ ) indicated a completeness score of 79.7% (single = 78.7%, duplicated = 0.9%).

### Metagenome report

We recovered 33 bins from the metagenome assembly (Figure 5), of which eight met the criteria for MAGs, including four fully circularised genomes. The recovered bins represented



**Figure 3. Genome assembly of *Phakellia ventilabrum*, odPhaVent3.1: BlobToolKit GC-coverage plot.** Blob plot showing sequence coverage (vertical axis) and GC content (horizontal axis). The circles represent scaffolds, with the size proportional to scaffold length and the colour representing phylum membership. The histograms along the axes display the total length of sequences distributed across different levels of coverage and GC content. An interactive version of this figure is available at [https://blobtoolkit.genomehubs.org/view/GCA\\_964276705.1/dataset/GCA\\_964276705.1/blob](https://blobtoolkit.genomehubs.org/view/GCA_964276705.1/dataset/GCA_964276705.1/blob).

9 bacterial and archaeal phyla, with genome sizes ranging from 0.90 to 4.00 Mbp (mean:  $2.17 \pm 0.83$  Mbp). Mean completeness was 81.9% ( $\pm 15.1\%$ ) with 2.7% ( $\pm 2.8\%$ ) contamination. Figure 6 summarises the taxa and quality of the metagenome bins. The full per-bin table of taxa and quality metrics for the metagenome bins is available on [Zenodo](#).

### Genome annotation report

The *Phakellia ventilabrum* genome assembly (GCA\_964276705.1) was annotated by Ensembl at the European Bioinformatics Institute (EBI). This annotation includes 38 110 transcribed mRNAs from 21 622 protein-coding and 5 189 non-coding genes. The average transcript length is 7 147.00 bp, with an average of 1.42 coding transcripts per gene and 6.91 exons per transcript. For further information about the annotation, please refer to the [Ensembl annotation page](#).

## Methods

### Sample acquisition

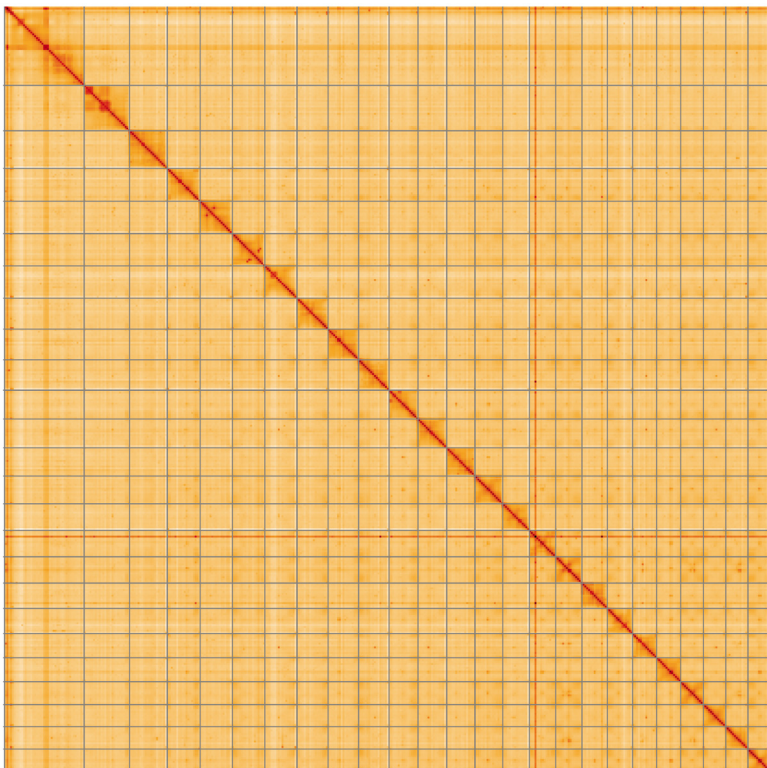
A specimen of *P. ventilabrum* (specimen ID GHC0000295, ToLID odPhaVent3; Figure 1) was collected on 2018-08-12 by

Remotely Operated Vehicle (ROV) *Ægir6000*, event GS2018108-70-ROV-50, during the GS2018108 cruise onboard research vessel *G.O. Sars*. It was collected in the Norwegian Sea at a water depth of 218 m in the Stjærnsund (latitude 70.2632°N, longitude 22.4744°E). The specimen was subsampled, flash-frozen and stored at  $-80^{\circ}\text{C}$  at GEOMAR until processing for genome sequencing. The specimen was collected by Kathrin Busch (Geomar) and identified by Hans Tore Rapp (UiB, Norway).

### Nucleic acid extraction

The workflow for high molecular weight (HMW) DNA extraction at the Wellcome Sanger Institute (WSI) Tree of Life Core Laboratory includes a sequence of procedures: sample preparation and homogenisation, DNA extraction, fragmentation and purification. Detailed protocols are available on [protocols.io](https://protocols.io) (Denton *et al.*, 2023). The odPhaVent3 sample was prepared for DNA extraction by weighing and dissecting it on dry ice (Jay *et al.*, 2023). Prior to DNA extraction, the sponge sample was bathed in “L buffer” (10 mM Tris, pH 7.6, 100 mM EDTA, 20 mM NaCl), minced into small pieces





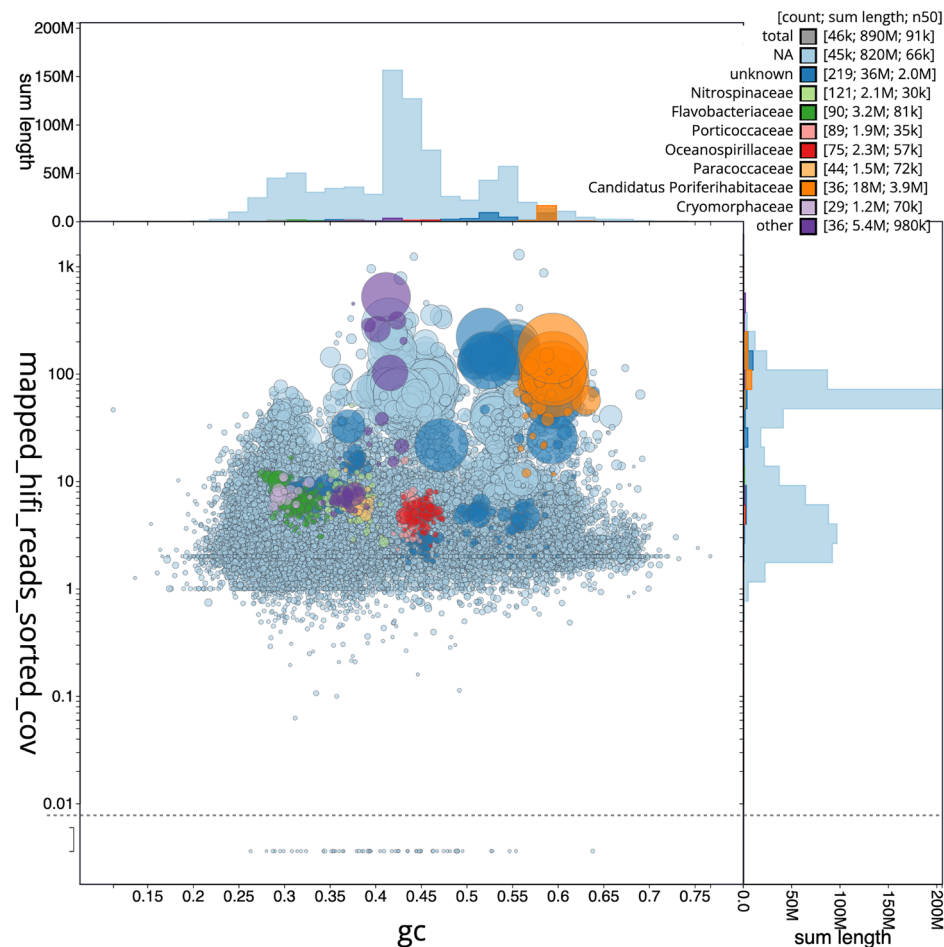
**Figure 4. Genome assembly of *Phakellia ventilabrum*: Hi-C contact map of the odPhaVent3.1 assembly, visualised using HiGlass.** Chromosomes are shown in order of size from left to right and top to bottom. An interactive version of this figure may be viewed at <https://genome-note-higlass.tol.sanger.ac.uk/l/?d=OdUSwO9xSWKqKrRfqJkbRg>.

**Table 3. Chromosomal pseudomolecules in the genome assembly of *Phakellia ventilabrum*, odPhaVent3.**

| INSDC accession | Name | Length (Mb) | GC%  |
|-----------------|------|-------------|------|
| OZ194358.1      | 1    | 22.06       | 43   |
| OZ194359.1      | 2    | 12.61       | 42.5 |
| OZ194360.1      | 3    | 10.43       | 43   |
| OZ194361.1      | 4    | 9.1         | 43.5 |
| OZ194362.1      | 5    | 8.98        | 43.5 |
| OZ194363.1      | 6    | 8.97        | 44.5 |
| OZ194364.1      | 7    | 8.93        | 43.5 |
| OZ194365.1      | 8    | 8.6         | 44   |
| OZ194366.1      | 9    | 8.49        | 43   |
| OZ194367.1      | 10   | 8.44        | 44   |
| OZ194368.1      | 11   | 8.01        | 43.5 |
| OZ194369.1      | 12   | 7.96        | 43   |
| OZ194370.1      | 13   | 7.83        | 43.5 |
| OZ194371.1      | 14   | 7.7         | 44   |

| INSDC accession | Name | Length (Mb) | GC%  |
|-----------------|------|-------------|------|
| OZ194372.1      | 15   | 7.37        | 43   |
| OZ194373.1      | 16   | 7.32        | 44.5 |
| OZ194374.1      | 17   | 7.29        | 44   |
| OZ194375.1      | 18   | 7.03        | 43   |
| OZ194376.1      | 19   | 6.99        | 43.5 |
| OZ194377.1      | 20   | 6.68        | 44   |
| OZ194378.1      | 21   | 6.66        | 43   |
| OZ194379.1      | 22   | 6.33        | 44.5 |
| OZ194380.1      | 23   | 6.17        | 44   |
| OZ194381.1      | 24   | 6.17        | 43.5 |
| OZ194382.1      | 25   | 5.77        | 44   |
| OZ194383.1      | MT   | 0.02        | 39.5 |

using a scalpel and the cellular interior separated from the mesohyl using forceps (Lopez, 2022). HMW DNA was extracted using the Automated MagAttract v2 protocol (Oatley *et al.*, 2023a). DNA was sheared into an average fragment



**Figure 5. Blob plot of base coverage mapped against GC proportion for sequences in the *Phakellia ventilabrum* metagenome.** Binned contigs are coloured by family. Circles are sized in proportion to sequence length on a square-root scale, ranging from 510 to 6 879 066. Histograms show the distribution of sequence length sum along each axis. An interactive version of this figure may be viewed [here](#).

size of 12–20 kb in a Megaruptor 3 system (Bates *et al.*, 2023). Sheared DNA was purified by solid-phase reversible immobilisation, using AMPure PB beads to eliminate shorter fragments and concentrate the DNA (Oatley *et al.*, 2023b). The concentration of the sheared and purified DNA was assessed using a Nanodrop spectrophotometer and Qubit Fluorometer using the Qubit dsDNA High Sensitivity Assay kit. Fragment size distribution was evaluated by running the sample on the FemtoPulse system.

RNA was also extracted from somatic tissue of odPhaVent3 in the Tree of Life Laboratory at the WSI using the RNA Extraction: Automated MagMax™ mirVana protocol (do Amaral *et al.*, 2023). The RNA concentration was assessed using a Nanodrop spectrophotometer and a Qubit Fluorometer using the Qubit RNA Broad-Range Assay kit. Analysis of the integrity of the RNA was done using the Agilent RNA 6000 Pico Kit and Eukaryotic Total RNA assay.

## Sequencing

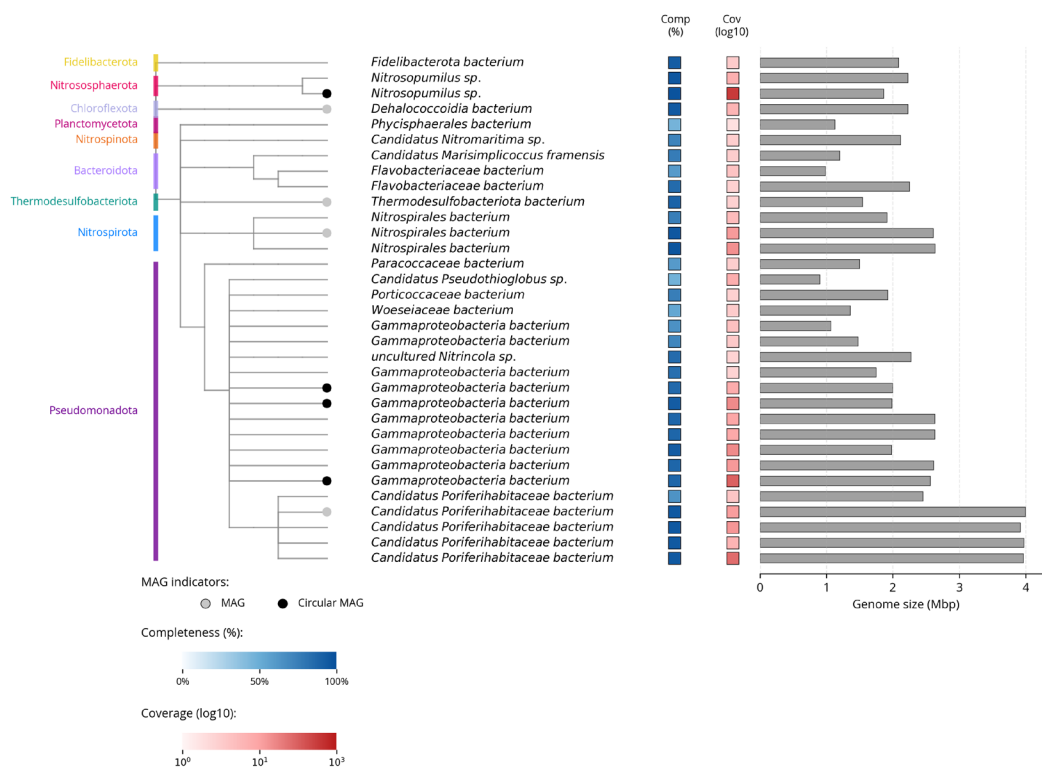
Pacific Biosciences HiFi circular consensus DNA sequencing libraries were constructed according to the manufacturers' instructions. DNA sequencing was performed by the Scientific Operations core at the WSI on Pacific Biosciences Revio. Tissue from the somatic tissue of the odPhaVent3 sample was processed for Hi-C sequencing at the WSI Scientific Operations core, using the Arima-HiC v2 kit and sequenced on the Illumina NovaSeq X instrument. Poly(A) RNA-Seq libraries were constructed using the NEB Ultra II RNA Library Prep kit, following the manufacturer's instructions. RNA sequencing was performed on the Illumina NovaSeq X instrument.

## Genome assembly, curation and evaluation

### Assembly

Prior to assembly of the PacBio HiFi reads, a database of  $k$ -mer counts ( $k = 31$ ) was generated from the filtered reads using FastK. GenomeScope2 (Ranallo-Benavidez *et al.*, 2020)





**Figure 6. Taxonomic tree based on taxonomic classifications of metagenome bins, constructed using ete3.** Colours indicate phylum-level taxonomy. Tracks show genome completeness (blue), sequencing coverage (red, log10), and genome size (grey bars, Mbp). High-quality MAGs are marked with grey circles; fully circularized MAGs in black.

was used to analyse the *k*-mer frequency distributions, providing estimates of genome size, heterozygosity, and repeat content.

The HiFi reads were assembled using Hifiasm (Cheng *et al.*, 2021) with the --primary option. Haplotypic duplications were identified and removed using purge\_dups (Guan *et al.*, 2020). The Hi-C reads were mapped to the primary contigs using bwa-mem2 (Vasimuddin *et al.*, 2019). The contigs were further scaffolded using the provided Hi-C data (Rao *et al.*, 2014) in YaHS (Zhou *et al.*, 2023) using the --break option for handling potential misassemblies. The scaffolded assemblies were evaluated using Gfastats (Formenti *et al.*, 2022), BUSCO (Manni *et al.*, 2021) and MERQUY.FK (Rhie *et al.*, 2020).

The mitochondrial genome was assembled using MitoHiFi (Uliano-Silva *et al.*, 2023), which runs MitoFinder (Allio *et al.*, 2020) and uses these annotations to select the final mitochondrial contig and to ensure the general quality of the sequence.

## Assembly curation

The assembly was decontaminated using the Assembly Screen for Cobionts and Contaminants (ASCC) pipeline. Flat files and maps used in curation were generated via the TreeVal pipeline (Pointon *et al.*, 2023). Manual curation was conducted primarily in PretextView (Harry, 2022) and HiGlass (Kerpedjiev *et al.*, 2018), with additional insights provided

by JBrowse2 (Diesh *et al.*, 2023). Scaffolds were visually inspected and corrected as described by Howe *et al.* (2021). Any identified contamination, missed joins, and mis-joins were amended, and duplicate sequences were tagged and removed. The curation process is documented at <https://gitlab.com/wtsi-grit/rapid-curation>.

## Taxonomic verification

Sequences of type material have not been available for taxonomic validation. Therefore, we reconstructed phylogenetic trees with the mitochondrial cytochrome oxidase subunit 1 (Folmer-region) and the C-region of the nuclear long ribosomal subunit (28S-C) for all sequences currently published for this genus. Sequences were aligned using MAFFT v7.450 (Kato & Standley, 2013), and Maximum Likelihood trees were reconstructed using RAXML 8 (GTR+GAMMA+ I model, Stamatakis, 2014). In both analyses the current sample was part of a monophyletic group of *P. ventilabrum*, comprising other, well-identified samples (e.g., HQ379409 (CO1, identical), or HQ379197 (28S, 1 bp different), both Morrow *et al.* (2012)). In addition, to validate the taxonomic identity of the sequenced specimen, the *cox1* gene sequence (animal DNA barcode) of a previously sequenced *P. ventilabrum* specimen spanning 658 bp was retrieved from NCBI (accession: HQ379409.1). This sequence was aligned against the scaffold identified as the mitochondrial genome of the specimen used for assembling the reference genome (NCBI accession: OZ194383.1), using

the software Geneious Prime v.2025.2.1 under default parameters. The barcode sequence was aligned from position 14,135 to position 14,793 of the new mitochondrial genome assembly. The alignment showed a 100% sequence identity, thereby providing molecular confirmation that indeed the assembled individual belonged to the species *P. ventilabrum*.

### Assembly quality assessment

The Merqury.FK tool (Rhie *et al.*, 2020), run in a Singularity container (Kurtzer *et al.*, 2017), was used to evaluate the assembly quality of the primary and alternate haplotypes using the *k*-mer databases (*k* = 31) that were computed prior to genome assembly.

A Hi-C contact map was produced for the final version of the assembly. The Hi-C reads were aligned using bwa-mem2 (Vasimuddin *et al.*, 2019) and the alignment files were combined using SAMtools (Danecek *et al.*, 2021). The Hi-C alignments were converted into a contact map using BEDTools (Quinlan & Hall, 2010) and the Cooler tool suite (Abdennur & Mirny, 2020).

The genome was analysed using the BlobToolKit pipeline, a Nextflow port of the earlier Snakemake version (Challis *et al.*, 2020). The pipeline aligns PacBio reads using SAMtools and minimap2 (Li, 2018) and generates coverage tracks for

regions of fixed size. It runs BUSCO (Manni *et al.*, 2021) using lineages identified from the NCBI taxonomy (Schoch *et al.*, 2020). For the three domain-level lineages, BUSCO genes are aligned to the UniProt Reference Proteomes (Bateman *et al.*, 2023) using DIAMOND blastp (Buchfink *et al.*, 2021). The genome is divided into chunks according to the density of the BUSCO genes from the closest taxonomic lineage, and each chunk is aligned to the UniProt Reference Proteomes using DIAMOND blastx. Genome sequences without a hit are split using seqtk and aligned to the NT database with blastn (Altschul *et al.*, 1990). The blobtools suite consolidates all outputs into a blobdir for visualisation. The BlobToolKit pipeline was developed using nf-core tooling (Ewels *et al.*, 2020) and MultiQC (Ewels *et al.*, 2016), with containerisation through Docker (Merkel, 2014) and Singularity (Kurtzer *et al.*, 2017).

Table 4 contains a list of relevant software tool versions and sources.

### Metagenome assembly

The metagenome assembly was generated using metaMDBG (Benoit *et al.*, 2024) and binned using MetaBAT2 (Kang *et al.*, 2019), MaxBin (Wu *et al.*, 2014), bin3C (DeMaere & Darling, 2019), and MetaTOR. The resulting bin sets of each binning algorithm were optimised and refined using

**Table 4. Software tools: versions and sources.**

| Software tool | Version                                                          | Source                                                                                                                |
|---------------|------------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------|
| BEDTools      | 2.30.0                                                           | <a href="https://github.com/arq5x/bedtools2">https://github.com/arq5x/bedtools2</a>                                   |
| bin3C         | 0.3.3                                                            | <a href="https://github.com/cerebis/bin3C">https://github.com/cerebis/bin3C</a>                                       |
| BLAST         | 2.14.0                                                           | <a href="ftp://ftp.ncbi.nlm.nih.gov/blast/executables/blast/">ftp://ftp.ncbi.nlm.nih.gov/blast/executables/blast/</a> |
| BlobToolKit   | 4.3.9                                                            | <a href="https://github.com/blobtoolkit/blobtoolkit">https://github.com/blobtoolkit/blobtoolkit</a>                   |
| BUSCO         | 5.5.0                                                            | <a href="https://gitlab.com/ezlab/busco">https://gitlab.com/ezlab/busco</a>                                           |
| bwa-mem2      | 2.2.1                                                            | <a href="https://github.com/bwa-mem2/bwa-mem2">https://github.com/bwa-mem2/bwa-mem2</a>                               |
| CheckM        | 1.2.1                                                            | <a href="https://github.com/ECogenomics/CheckM">https://github.com/ECogenomics/CheckM</a>                             |
| Cooler        | 0.8.11                                                           | <a href="https://github.com/open2c/cooler">https://github.com/open2c/cooler</a>                                       |
| DIAMOND       | 2.1.8                                                            | <a href="https://github.com/bbuchfink/diamond">https://github.com/bbuchfink/diamond</a>                               |
| dRep          | 3.4.0                                                            | <a href="https://github.com/MrOlm/drep">https://github.com/MrOlm/drep</a>                                             |
| fasta_windows | 0.2.4                                                            | <a href="https://github.com/tolkita/fasta_windows">https://github.com/tolkita/fasta_windows</a>                       |
| FastK         | 427104ea91c78c3b8b8b49f1a7d6bbeaa869ba1c                         | <a href="https://github.com/thegenemyers/FASTK">https://github.com/thegenemyers/FASTK</a>                             |
| Gfastats      | 1.3.6                                                            | <a href="https://github.com/vgl-hub/gfastats">https://github.com/vgl-hub/gfastats</a>                                 |
| GTDB-TK       | 2.3.2                                                            | <a href="https://github.com/ECogenomics/GTDBTk">https://github.com/ECogenomics/GTDBTk</a>                             |
| Hifiasm       | 0.19.8-r603                                                      | <a href="https://github.com/chhylp123/hifiasm">https://github.com/chhylp123/hifiasm</a>                               |
| HiGlass       | 44086069ee7d4d3f6f3f0012569789ec138f42b84aa44357826c0b6753eb28de | <a href="https://github.com/higlass/higlass">https://github.com/higlass/higlass</a>                                   |

| Software tool          | Version                                  | Source                                                                                                                    |
|------------------------|------------------------------------------|---------------------------------------------------------------------------------------------------------------------------|
| MAGScoT                | 1.0.0                                    | <a href="https://github.com/ikmb/MAGScoT">https://github.com/ikmb/MAGScoT</a>                                             |
| MaxBin                 | 2.7                                      | <a href="https://sourceforge.net/projects/maxbin/">https://sourceforge.net/projects/maxbin/</a>                           |
| MerquyFK               | d00d98157618f4e8d1a9190026b19b471055b22e | <a href="https://github.com/thegenemyers/MERQURY.FK">https://github.com/thegenemyers/MERQURY.FK</a>                       |
| MetaBat2               | 2.15-15-gd6ea400                         | <a href="https://bitbucket.org/berkeleylab/metabat/src/master/">https://bitbucket.org/berkeleylab/metabat/src/master/</a> |
| metaMDBG               | -                                        | <a href="https://github.com/GaetanBenoitDev/metaMDBG">https://github.com/GaetanBenoitDev/metaMDBG</a>                     |
| Minimap2               | 2.24-r1122                               | <a href="https://github.com/lh3/minimap2">https://github.com/lh3/minimap2</a>                                             |
| MitoHiFi               | 2                                        | <a href="https://github.com/marcelauliano/MitoHiFi">https://github.com/marcelauliano/MitoHiFi</a>                         |
| MultiQC                | 1.14, 1.17, and 1.18                     | <a href="https://github.com/MultiQC/MultiQC">https://github.com/MultiQC/MultiQC</a>                                       |
| NCBI Datasets          | 15.12.0                                  | <a href="https://github.com/ncbi/datasets">https://github.com/ncbi/datasets</a>                                           |
| Nextflow               | 23.10.0                                  | <a href="https://github.com/nextflow-io/nextflow">https://github.com/nextflow-io/nextflow</a>                             |
| PretextView            | 0.2                                      | <a href="https://github.com/sanger-tol/PretextView">https://github.com/sanger-tol/PretextView</a>                         |
| PROKKA                 | 1.14.5                                   | <a href="https://github.com/vdejaer/prokka">https://github.com/vdejaer/prokka</a>                                         |
| purge_dups             | 1.2.3                                    | <a href="https://github.com/dfguan/purge_dups">https://github.com/dfguan/purge_dups</a>                                   |
| samtools               | 1.19.2                                   | <a href="https://github.com/samtools/samtools">https://github.com/samtools/samtools</a>                                   |
| sanger-tol/ascc        | -                                        | <a href="https://github.com/sanger-tol/ascc">https://github.com/sanger-tol/ascc</a>                                       |
| sanger-tol/blobtoolkit | 0.6.0                                    | <a href="https://github.com/sanger-tol/blobtoolkit">https://github.com/sanger-tol/blobtoolkit</a>                         |
| Seqtk                  | 1.3                                      | <a href="https://github.com/lh3/seqtk">https://github.com/lh3/seqtk</a>                                                   |
| Singularity            | 3.9.0                                    | <a href="https://github.com/sylabs/singularity">https://github.com/sylabs/singularity</a>                                 |
| TreeVal                | 1.2.0                                    | <a href="https://github.com/sanger-tol/treeval">https://github.com/sanger-tol/treeval</a>                                 |
| YaHS                   | 1.1a.2                                   | <a href="https://github.com/c-zhou/yahs">https://github.com/c-zhou/yahs</a>                                               |

MAGScoT (Rühlemann *et al.*, 2022). PROKKA (Seemann, 2014) was used to identify tRNAs and rRNAs in each bin, CheckM (Parks *et al.*, 2015) (checkM\_DB release 2015-01-16) was used to assess bin completeness/contamination, and GTDB-TK (Chaumeil *et al.*, 2022) (GTDB release 214) was used to taxonomically classify bins. Taxonomic replicate bins were identified using dRep (Olm *et al.*, 2017) with default settings (95% ANI threshold). The final bin set was filtered for bacteria and archaea. All bins were assessed for quality and categorised as metagenome-assembled genomes (MAGs) if they met the following criteria: contamination  $\leq 5\%$ , presence of 5S, 16S, and 23S rRNA genes, at least 18 unique tRNAs, and either  $\geq 90\%$  completeness or  $\geq 50\%$  completeness with fully circularised chromosomes. Bins that did not meet these thresholds, or were identified as taxonomic replicates of MAGs, were retained as ‘binned metagenomes’ provided they had  $\geq 50\%$  completeness and  $\leq 10\%$  contamination. A

taxonomic tree of the bins was constructed from NCBI classifications using ete3 (Huerta-Cepas *et al.*, 2016) and visualised with matplotlib. Software tool versions and sources are given in Table 4.

### Wellcome Sanger Institute – Legal and Governance

The materials that have contributed to this genome note have been supplied by a Tree of Life collaborator. The Wellcome Sanger Institute employs a process whereby due diligence is carried out proportionate to the nature of the materials themselves, and the circumstances under which they have been/are to be collected and provided for use. The purpose of this is to address and mitigate any potential legal and/or ethical implications of receipt and use of the materials as part of the research project, and to ensure that in doing so we align with best practice wherever possible. The overarching areas of consideration are:

- Ethical review of provenance and sourcing of the material
- Legality of collection, transfer and use (national and international)

Each transfer of samples is undertaken according to a Research Collaboration Agreement or Material Transfer Agreement entered into by the Tree of Life collaborator, Genome Research Limited (operating as the Wellcome Sanger Institute) and in some circumstances other Tree of Life collaborators.

## Data availability

European Nucleotide Archive: *Phakellia ventilabrum*. Accession number PRJEB78906; <https://identifiers.org/ena.embl/PRJEB78906>. The genome sequence is released openly for reuse. The *Phakellia ventilabrum* genome sequencing initiative is part of the Aquatic Symbiosis Genomics (ASG) project (<https://www.ebi.ac.uk/ena/browser/view/PRJEB43743>). All raw sequence data and the assembly have been deposited in INSDC databases. The genome will be annotated using available

RNA-Seq data and presented through the [Ensembl](#) pipeline at the European Bioinformatics Institute. Raw data and assembly accession identifiers are reported in [Table 1](#) and [Table 2](#).

## Author information

Contributors are listed at the following links:

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- [Wellcome Sanger Institute Scientific Operations – Sequencing Operations](#)
- [Wellcome Sanger Institute Tree of Life Core Informatics team](#)
- [Tree of Life Core Informatics collective](#)
- [European Bioinformatics Institute ASG Data Portal team](#)
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# Open Peer Review

Current Peer Review Status:  

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**Arzu Karahan** 

Middle East Technical University, Erdemli-Mersin, Turkey

The research is well designed, thoroughly analyzed, and clearly presented. I have no criticisms regarding the analyses, writing, or data quality. However, it would have been beneficial to analyze eukaryotic metagenomes from species sharing the same environment (or living on or in it), such as Chordata, Cnidaria, and Echinodermata (Figure 3), and give at least the family- or genus-level taxa name.

Metagenome raw data should also be released.

**Is the rationale for creating the dataset(s) clearly described?**

Yes

**Are the protocols appropriate and is the work technically sound?**

Yes

**Are sufficient details of methods and materials provided to allow replication by others?**

Yes

**Are the datasets clearly presented in a useable and accessible format?**

Partly

**Competing Interests:** No competing interests were disclosed.

**Reviewer Expertise:** metagenomics, transcriptomics, regeneration, aging, metabolomics, metabarcoding

**I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.**

Reviewer Report 16 January 2026

<https://doi.org/10.21956/wellcomeopenres.28017.r144173>

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**Poppy Hesketh-Best** 

IGTP Institut Germans Trias i Pujol, Barcelona, Spain

This data note by Taboada and colleagues presents HiFi sequencing data for the marine sponge *Phakellia ventilabrum*. The authors generated a high-quality assembled and annotated sponge genome and further reconstructed bacterial metagenome-assembled genomes (MAGs) from the associated microbiome.

**Is the rationale for creating the dataset(s) clearly described?**

*P. ventilabrum* is a vulnerable marine ecosystem indicator species and is appropriately presented by the authors as a species of interest, particularly with respect to its potentially compromised resilience under climate change scenarios. In addition, its potential ability to accumulate environmental DNA (eDNA) broadens interest in research on genetic interconnectivity within this species and among surrounding taxa through eDNA-based studies. I am satisfied that the authors have presented a strong rationale for this dataset.

**Are the protocols appropriate and is the work technically sound?**

I cannot comment on the methodology used for the sponge genome assembly due to a lack of relevant expertise. However, the metagenome-assembled genome (MAG) protocol follows established practices, quality assignment for the MAGs, employing commonly used workflows and software that are widely adopted by the broader field.

It would be good for the authors to confirm their MAG QC using CheckM2 and the updated database, since the one used is 10 years old now. CheckM2 is more suited to MAGs with fewer representative species, which is likely the case with this deep-sea sponge species.

**Are sufficient details of methods and materials provided to allow replication by others?**

The methods are well described and rely on open-source software, software containers, and provided detailed version controls. Several analyses are implemented using established workflows managed with Nextflow, enhancing transparency and reproducibility.

**Are the datasets clearly presented in a useable and accessible format?**

Overall, the data are available in public repositories and were accessible as of 2026-01-15. However, none of the 33 metagenome-assembled genomes (MAGs) have been made publicly available. It is unusual that the sponge genome and its annotation are accessible, while the bacterial MAGs are not.

**Is the rationale for creating the dataset(s) clearly described?**

Yes

**Are the protocols appropriate and is the work technically sound?**

Yes

**Are sufficient details of methods and materials provided to allow replication by others?**

Yes

**Are the datasets clearly presented in a useable and accessible format?**

Partly

***Competing Interests:*** No competing interests were disclosed.

***Reviewer Expertise:*** Microbial genomics and bioinformatic

**I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.**

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