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DATA NOTE

The genome sequence of the Provence Hairstreak, *Tomares ballus* (Fabricius, 1787) (Lepidoptera: Lycaenidae)

[version 1; peer review: awaiting peer review]

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v1 **First published:** 16 Jan 2026, **11**:35
https://doi.org/10.12688/wellcomeopenres.25725.1
Latest published: 16 Jan 2026, **11**:35
https://doi.org/10.12688/wellcomeopenres.25725.1

Abstract

We present a genome assembly from a male specimen of *Tomares ballus* (Provence Hairstreak; Arthropoda; Insecta; Lepidoptera; Lycaenidae). The assembly contains two haplotypes with total lengths of 839.94 megabases and 831.10 megabases. Most of haplotype 1 (99.02%) is scaffolded into 23 chromosomal pseudomolecules, including the Z sex chromosome. Haplotype 2 was assembled to scaffold level. The mitochondrial genome has also been assembled, with a length of 15.43 kilobases. This work is part of Project Psyche, a collaborative programme generating genomes for European butterflies and moths.

Keywords

Tomares ballus; Provence Hairstreak; genome sequence; chromosomal; Lepidoptera

This article is included in the Tree of Life gateway.

Open Peer Review

Approval Status *AWAITING PEER REVIEW*

Any reports and responses or comments on the article can be found at the end of the article.

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Author roles: Menchetti M: Data Curation, Investigation, Resources, Writing – Original Draft Preparation; **Hinojosa JC**: Investigation, Resources; **Vila R**: Investigation, Resources, Supervision, Writing – Review & Editing; **Wright CJ**: Funding Acquisition, Project Administration, Supervision; **Meier JI**: Funding Acquisition, Project Administration, Supervision; **Blaxter ML**: Funding Acquisition, Project Administration, Supervision

Competing interests: No competing interests were disclosed.

Grant information: This work was supported by Wellcome through core funding to the Wellcome Sanger Institute (220540). R.V. is supported by grant PID2022-139689NB-I00 (MICIU/ AEI/ 10.13039/501100011033 and ERDF, EU) and by grant 2021-SGR-00420 (Departament de Recerca i Universitats, Generalitat de Catalunya).
The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

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How to cite this article: Menchetti M, Hinojosa JC, Vila R *et al.* **The genome sequence of the Provence Hairstreak, *Tomares ballus* (Fabricius, 1787) (Lepidoptera: Lycaenidae) [version 1; peer review: awaiting peer review]** Wellcome Open Research 2026, **11**:35 https://doi.org/10.12688/wellcomeopenres.25725.1

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Species taxonomy

Eukaryota; Opisthokonta; Metazoa; Eumetazoa; Bilateria; Protostomia; Ecdysozoa; Panarthropoda; Arthropoda; Mandibulata; Pancrustacea; Hexapoda; Insecta; Dicondylia; Pterygota; Neoptera; Endopterygota; Amphiesmenoptera; Lepidoptera; Glossata; Neolepidoptera; Heteroneura; Ditrysia; Obtectomera; Papilionoidea; Lycaenidae; Theclinae; Tomarini; *Tomares*; *Tomares ballus* (Fabricius, 1787) (NCBI:txid1107738)

Background

The Provence Hairstreak, *Tomares ballus* (Fabricius, 1787), is a small butterfly from the family Lycaenidae, characterised by the striking bright orange and green colouration on the underside of the wing. *T. ballus* is found in the western Mediterranean region (Spain, Morocco, Tunisia, Libya, and Egypt) (Krupitsky *et al.*, 2022). It inhabits warm and grassy areas, often those altered by human activities (García-Barros *et al.*, 2013; Krupitsky *et al.*, 2022). The species is polyphagous, and larvae feed on several genera in the Fabaceae. Adults are univoltine and fly between February and June, with a peak in March and April (Martín Cano *et al.*, 2009). It is a myrmecophilous species, and larvae have been found associated with the ants *Plagiolepis pygmaea* (Latreille, 1798) and *Temnothorax* spp. (Fiedler, 1991; Muñoz Sariot, 2011).

The genus *Tomares* is relatively small and is the sole representative of the tribe Tomarini within the subfamily Theclinae (Espeland *et al.*, 2018; Kawahara *et al.*, 2023). The split from the sister species *T. mauritanicus* probably occurred during the early–middle Pliocene. During the late Pleistocene, *T. ballus* dispersed across Africa and colonised the western Mediterranean region (Krupitsky *et al.*, 2022; Nazari & ten Hagen, 2020). The species shows rather low mitochondrial variation, with only some spatial structure across its range (Dapporto *et al.*, 2022). It is listed as Least Concern on the IUCN Red List of Europe (Van Swaay *et al.*, 2025), and populations near expanding urban areas are particularly vulnerable (García-Barros *et al.*, 2013).

The reference genome for *T. ballus* presented here provides a useful resource for butterfly phylogenomics, as it is the first representative genome for the tribe Tomarini within the Theclinae (Espeland *et al.*, 2018; Kawahara *et al.*, 2023). The sequence data were derived from a male specimen (Figure 1), collected in Cervià de les Garrigues, Garrigues, Catalonia, Spain.

Methods

Sample acquisition

The specimen used for genome sequencing was an adult male *Tomares ballus* (specimen ID SAN28000375, ToLID ilTomBall1; Figure 1), collected from Cervià de les Garrigues, Garrigues, Catalonia, Spain (latitude 41.4261, longitude 0.8322) on 2022-04-07. The specimen was collected by Roger Vila, Mattia Menchetti and Joan Carles Hinojosa (Institut de Biologia Evolutiva), and identified by Joan Carles Hinojosa and Roger Vila (Institut de Biologia Evolutiva).

Nucleic acid extraction

Protocols for high molecular weight (HMW) DNA extraction developed at the Wellcome Sanger Institute (WSI) Tree of Life Core Laboratory are available on protocols.io (Howard *et al.*, 2025). The ilTomBall1 sample was weighed and triaged to determine the appropriate extraction protocol. Tissue from the whole organism was homogenised by powermashing using a PowerMasher II tissue disruptor.

HMW DNA was extracted in the WSI Scientific Operations core using the Automated MagAttract v2 protocol. DNA was sheared into an average fragment size of 12–20 kb following the Megaruptor®3 for LI PacBio protocol. Sheared DNA was purified by automated SPRI (solid-phase reversible immobilisation). 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. For this sample, the final post-shearing DNA had a Qubit concentration of 62.61 ng/μL and a yield of 2 942.67 ng, with a fragment size of 15.4 kb.

PacBio HiFi library preparation and sequencing

Library preparation and sequencing were performed at the WSI Scientific Operations core. Libraries were prepared using the SMRTbell Prep Kit 3.0 (Pacific Biosciences, California, USA), according to the manufacturer's instructions. The kit

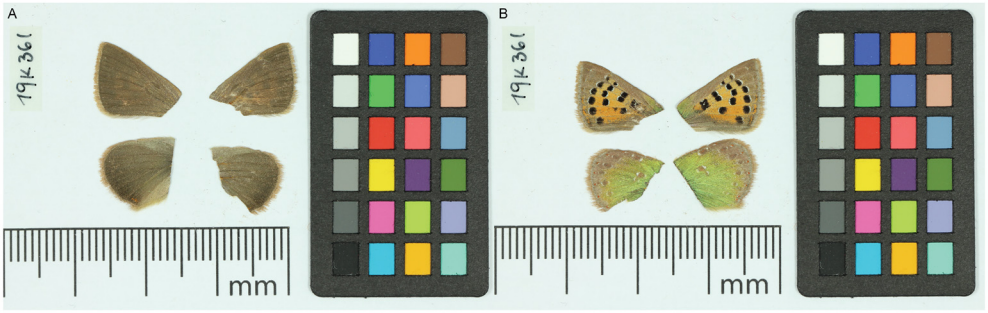

Figure 1. Voucher photographs of the *Tomares ballus* (ilTomBall1) specimen used for genome sequencing.

includes reagents for end repair/A-tailing, adapter ligation, post-ligation SMRTbell bead clean-up, and nuclease treatment. Size selection and clean-up were performed using diluted AMPure PB beads (Pacific Biosciences). DNA concentration was quantified using a Qubit Fluorometer v4.0 (ThermoFisher Scientific) and the Qubit 1X dsDNA HS assay kit. Final library fragment size was assessed with the Agilent Femto Pulse Automated Pulsed Field CE Instrument (Agilent Technologies) using the gDNA 55 kb BAC analysis kit.

The sample was sequenced on a Revio instrument (Pacific Biosciences). The prepared library was normalised to 2 nM, and 15 µL was used for making complexes. Primers were annealed and polymerases bound to generate circularised complexes, following the manufacturer's instructions. Complexes were purified using 1.2X SMRTbell beads, then diluted to the Revio loading concentration (200–300 pM) and spiked with a Revio sequencing internal control. The sample was sequenced on a Revio 25M SMRT cell. The SMRT Link software (Pacific Biosciences), a web-based workflow manager, was used to configure and monitor the run and to carry out primary and secondary data analysis.

Specimen details, sequencing platforms, and data yields are summarised in Table 1.

Hi-C

Sample preparation and crosslinking

The Hi-C sample was prepared from 20–50 mg of frozen tissue from the whole organism of the ilTomBall1 sample using the Arima-HiC v2 kit (Arima Genomics). Following the manufacturer's instructions, tissue was fixed and DNA crosslinked using TC buffer to a final formaldehyde concentration of 2%. The tissue was homogenised using the Diagnocine Power Masher-II. Crosslinked DNA was digested with a restriction enzyme master mix, biotinylated, and ligated. Clean-up was performed with SPRISelect beads before library preparation. DNA concentration was measured with the Qubit Fluorometer (Thermo Fisher Scientific) and Qubit HS Assay Kit. The biotinylation percentage was estimated using the Arima-HiC v2 QC beads.

Hi-C library preparation and sequencing

Biotinylated DNA constructs were fragmented using a Covaris E220 sonicator and size selected to 400–600 bp using SPRISelect beads. DNA was enriched with Arima-HiC v2 kit Enrichment beads. End repair, A-tailing, and adapter ligation were carried out with the NEBNext Ultra II DNA Library Prep Kit (New England Biolabs), following a modified protocol where library preparation occurs while DNA remains bound to the Enrichment beads. Library amplification was performed using KAPA HiFi HotStart mix and a custom Unique Dual Index (UDI) barcode set (Integrated DNA Technologies). Depending on sample concentration and biotinylation percentage determined at the crosslinking stage, libraries were amplified with 10–16 PCR cycles. Post-PCR clean-up was performed with SPRISelect beads. Libraries were quantified using the AccuClear Ultra High Sensitivity dsDNA Standards Assay Kit (Biotium) and a FLUOstar Omega plate reader (BMG Labtech).

Prior to sequencing, libraries were normalised to 10 ng/µL. Normalised libraries were quantified again to create equimolar and/or weighted 2.8 nM pools. Pool concentrations were checked using the Agilent 4200 TapeStation (Agilent) with High Sensitivity D500 reagents before sequencing. Sequencing was performed using paired-end 150 bp reads on the Illumina NovaSeq X.

Specimen details, sequencing platforms, and data yields are summarised in Table 1.

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

Table 1. Specimen and sequencing data for BioProject PRJEB85042.

Platform	PacBio HiFi	Hi-C
ToLID	ilTomBall1	ilTomBall1
Specimen ID	SAN28000375	SAN28000375
BioSample (source individual)	SAMEA115770361	SAMEA115770361
BioSample (tissue)	SAMEA115770381	SAMEA115770381
Tissue	whole organism	whole organism
Instrument	Revio	Illumina NovaSeq X
Run accessions	ERR14209152; ERR14209153	ERR14224629
Read count total	2.57 million	702.75 million
Base count total	27.36 Gb	106.12 Gb

The HiFi reads were assembled using Hifiasm in Hi-C phasing mode (Cheng *et al.*, 2021; Cheng *et al.*, 2022), producing two haplotypes. Hi-C reads (Rao *et al.*, 2014) were mapped to the primary contigs using bwa-mem2 (Vasimuddin *et al.*, 2019). Contigs were further scaffolded with Hi-C data 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 MERQURY.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. TreeVal was used to generate the flat files and maps for use in curation. Manual curation was conducted primarily in PretextView and HiGlass (Kerpedjiev *et al.*, 2018). Scaffolds were visually inspected and corrected as described by Howe *et al.* (2021). Manual corrections included five breaks and nine joins, which increased the scaffold N50 by 3.3%. The curation process is described at https://gitlab.com/wtsi-grit/rapid-curation. PretextSnapshot was used to generate a Hi-C contact map of the final assembly.

Assembly quality assessment

The Merqury.FK tool (Rhie *et al.*, 2020), run in a Singularity container (Kurtzer *et al.*, 2017), was used to evaluate *k*-mer completeness and assembly quality for both haplotypes using the *k*-mer databases (k = 31) computed prior to genome assembly. The analysis outputs included assembly QV scores and completeness statistics.

The genome was analysed using the BlobToolKit pipeline, a Nextflow (Di Tommaso *et al.*, 2017) implementation of the earlier Snakemake version (Challis *et al.*, 2020). The pipeline aligns PacBio reads using minimap2 (Li, 2018) and SAMtools (Danecek *et al.*, 2021) to generate coverage tracks. 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 database (Bateman *et al.*, 2023) using DIAMOND blastp (Buchfink *et al.*, 2021). The genome is divided into chunks based on the density of BUSCO genes from the closest taxonomic lineage, and each chunk is aligned to the UniProt Reference Proteomes database with DIAMOND blastx. Sequences without hits are chunked using seqtk and aligned to the NT database with blastn (Altschul *et al.*, 1990). The BlobToolKit 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).

We used lep_busco_painter to paint Merian elements along chromosomes (Wright *et al.*, 2024). Merian elements represent the 32 ancestral linkage groups in Lepidoptera. The painting process utilised BUSCO gene locations from the lepidoptera_odb10 set (Kriventseva *et al.*, 2019) and chromosome lengths from NCBI Datasets. Each complete BUSCO gene (both single-copy and duplicated) was assigned to a Merian element based on a reference database, then coloured and plotted along chromosomes drawn to scale.

Genome sequence report

Sequence data

PacBio sequencing of the *Tomares ballus* specimen generated 27.36 Gb (gigabases) from 2.57 million reads, which were used to assemble the genome. GenomeScope2.0 analysis estimated the haploid genome size at 798.21 Mb, with a heterozygosity of 1.40% and repeat content of 26.60% (Figure 2). These estimates guided expectations for the assembly. Based on the estimated genome size, the sequencing data provided approximately 33× coverage. Hi-C sequencing produced

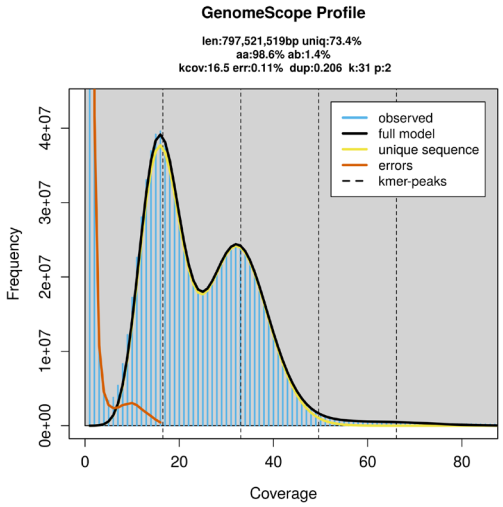

Figure 2. Frequency distribution of *k*-mers generated using GenomeScope2. The plot shows observed and modelled *k*-mer spectra, providing estimates of genome size, heterozygosity, and repeat content based on unassembled sequencing reads.

106.12 Gb from 702.75 million reads, which were used to scaffold the assembly. Table 1 summarises the specimen and sequencing details.

Assembly statistics

The genome was assembled into two haplotypes using Hi-C phasing. Haplotype 1 was curated to chromosome level, while haplotype 2 was assembled to scaffold level. The final assembly has a total length of 839.94 Mb in 125 scaffolds, with 196 gaps, and a scaffold N50 of 38.46 Mb (Table 2).

Most of the assembly sequence (99.02%) was assigned to 23 chromosomal-level scaffolds, representing 22 autosomes and the Z sex chromosome. These chromosome-level scaffolds, confirmed by Hi-C data, are named according to size (Figure 3; Table 3). Chromosome painting with Merian elements illustrates

Table 2. Genome assembly statistics.

Assembly name	ilTomBall1.hap1.1	ilTomBall1.hap2.1
Assembly accession	GCA_965153325.1	GCA_965153315.1
Assembly level	chromosome	scaffold
Span (Mb)	839.94	831.10
Number of chromosomes	23	scaffold-level
Number of contigs	321	290
Contig N50	7.07 Mb	6.72 Mb
Number of scaffolds	125	94
Scaffold N50	38.46 Mb	37.11 Mb
Longest scaffold length (Mb)	61.95	-
Sex chromosomes	Z	-
Organelles	Mitochondrion: 15.43 kb	-

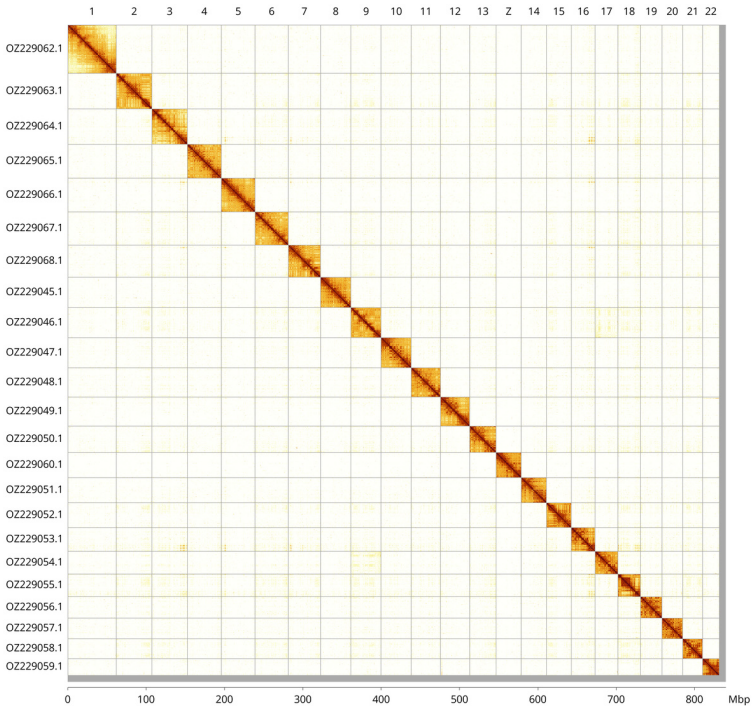

Figure 3. Hi-C contact map of the *Tomares ballus* genome assembly. Assembled chromosomes are shown in order of size and labelled along the axes, with a megabase scale shown below. The plot was generated using PretextSnapshot.

Table 3. Chromosomal pseudomolecules in the haplotype 1 genome assembly of *Tomares ballus* ilTomBall1.

INSDC accession	Molecule	Length (Mb)	GC%	Assigned Merian elements
OZ229062.1	1	61.95	37	M14;M26;M4
OZ229063.1	2	45.62	37	M8
OZ229064.1	3	45.20	37	M18;M30
OZ229065.1	4	43.22	37	M11;M23
OZ229066.1	5	43.15	36.50	M1;M19
OZ229067.1	6	42.40	37	M7
OZ229068.1	7	41.16	37	M17;M20
OZ229045.1	8	38.70	37	M12;M29
OZ229046.1	9	38.66	37.50	M22
OZ229047.1	10	38.46	37	M2
OZ229048.1	11	37.23	37	M9
OZ229049.1	12	37.20	36.50	M25;M5
OZ229050.1	13	33.67	37.50	M6
OZ229051.1	14	32.16	37	M3
OZ229052.1	15	31.68	37	M21
OZ229053.1	16	30.50	37	M13
OZ229054.1	17	29.06	37.50	M16
OZ229055.1	18	28.88	38	M27
OZ229056.1	19	27.32	37.50	M10
OZ229057.1	20	26.60	37.50	M15
OZ229058.1	21	25.21	38	M24
OZ229059.1	22	21.52	37	M28;M31
OZ229060.1	Z	32.19	35.50	MZ

the distribution of orthologues along chromosomes and highlights patterns of chromosomal evolution relative to Lepidopteran ancestral linkage groups (Figure 4). The Z chromosome was identified by Merian element painting.

The mitochondrial genome was also assembled (length 15.43 kb, OZ229061.1). This sequence is included as a contig in the multifasta file of the genome submission and as a standalone record.

Assembly quality metrics

For haplotype 1, the estimated QV is 61.6, and for haplotype 2, 61.8. When the two haplotypes are combined, the assembly achieves an estimated QV of 61.7. The *k*-mer completeness is 74.54% for haplotype 1, 74.25% for haplotype 2, and 99.48% for the combined haplotypes (Figure 5). BUSCO analysis using the lepidoptera_odb10 reference set (n = 5 286) identified 98.0% of the expected gene set (single = 97.4%, duplicated = 0.5%) in haplotype 1. For haplotype 2, BUSCO analysis identified 98.2% of the expected gene set (single = 97.7%, duplicated = 0.5%). The snail plot in Figure 6 summarises the scaffold length distribution and other assembly statistics for haplotype 1. The blob plot in Figure 7 shows the distribution of scaffolds by GC proportion and coverage for haplotype 1.

Table 4 lists the assembly metric benchmarks adapted from Rhie *et al.* (2021) the Earth BioGenome Project Report on Assembly Standards September 2024. The EBP metric,

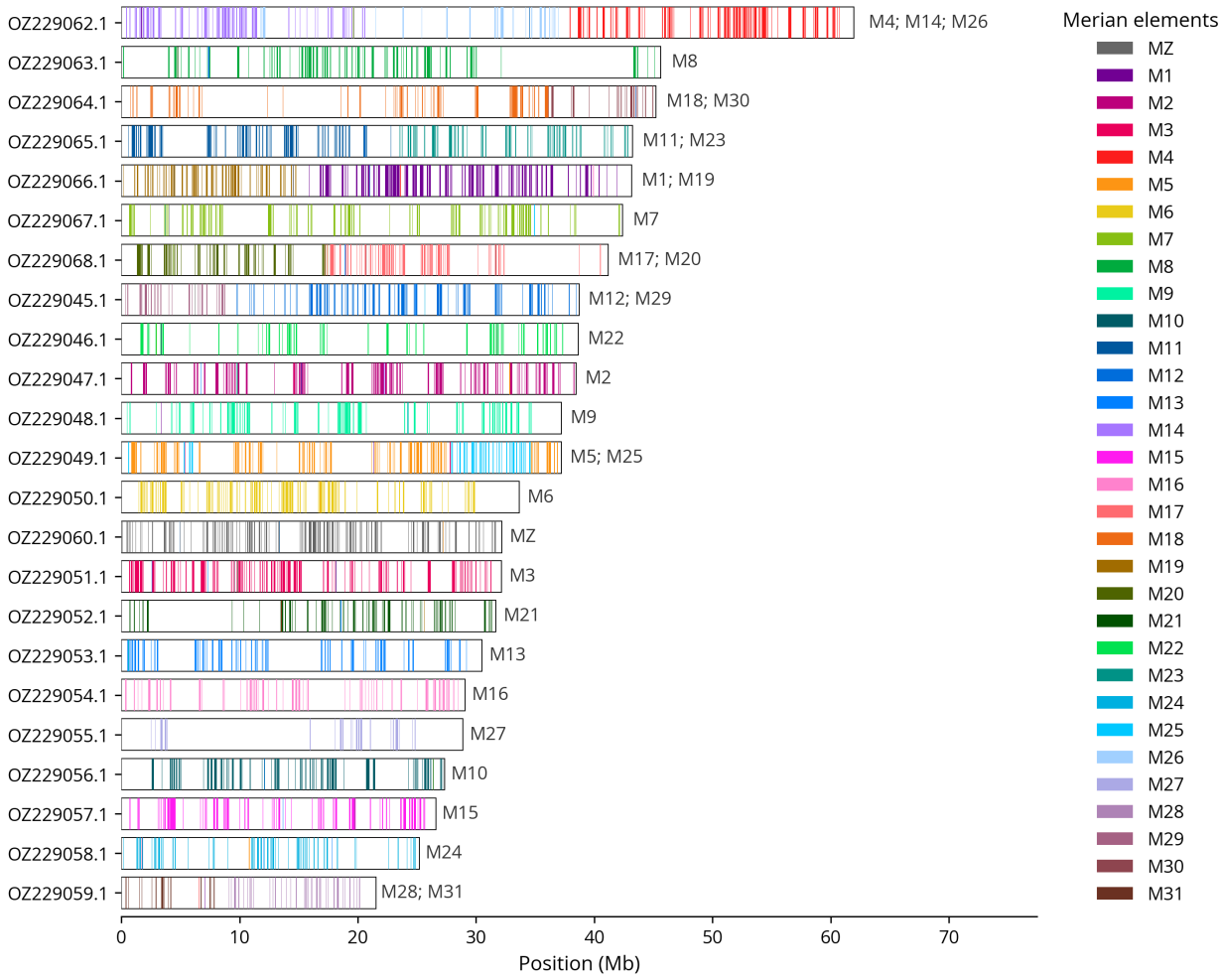

Figure 4. Merian elements painted across chromosomes in the ilTomBall1.hap1.1 assembly of *Tomares ballus*. Chromosomes are drawn to scale, with the positions of orthologues shown as coloured bars. Each orthologue is coloured by the Merian element that it belongs to. All orthologues which could be assigned to Merian elements are shown.

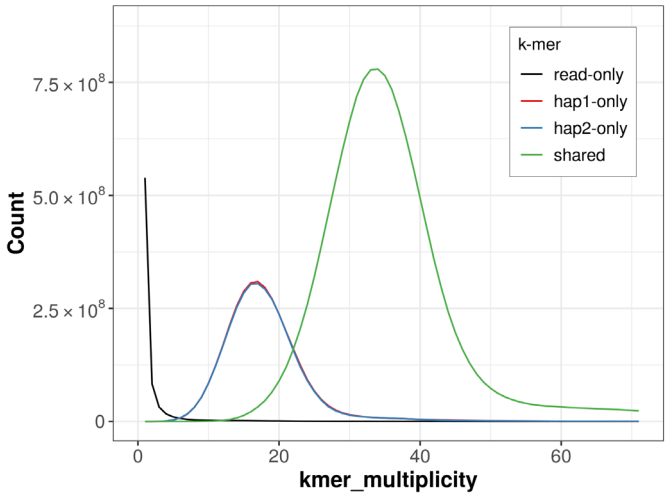

Figure 5. Evaluation of *k*-mer completeness using MerquryFK. This plot illustrates the recovery of *k*-mers from the original read data in the final assemblies. The horizontal axis represents *k*-mer multiplicity, and the vertical axis shows the number of *k*-mers. The black curve represents *k*-mers that appear in the reads but are not assembled. The green curve (the homozygous peak) corresponds to *k*-mers shared by both haplotypes and the red and blue curves (the heterozygous peaks) show *k*-mers found only in one of the haplotypes.

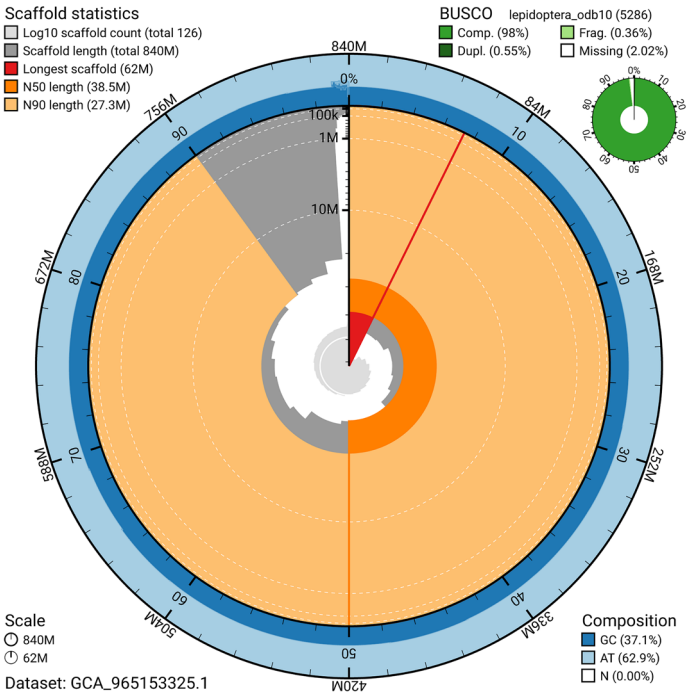

Figure 6. Assembly metrics for ilTomBall1.hap1.1. The BlobToolKit snail plot provides an overview of assembly metrics and BUSCO gene completeness. The circumference represents the length of the whole genome sequence, and the main plot is divided into 1,000 bins around the circumference. The outermost 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 set is presented at the top right. An interactive version of this figure can be accessed on the BlobToolKit viewer.

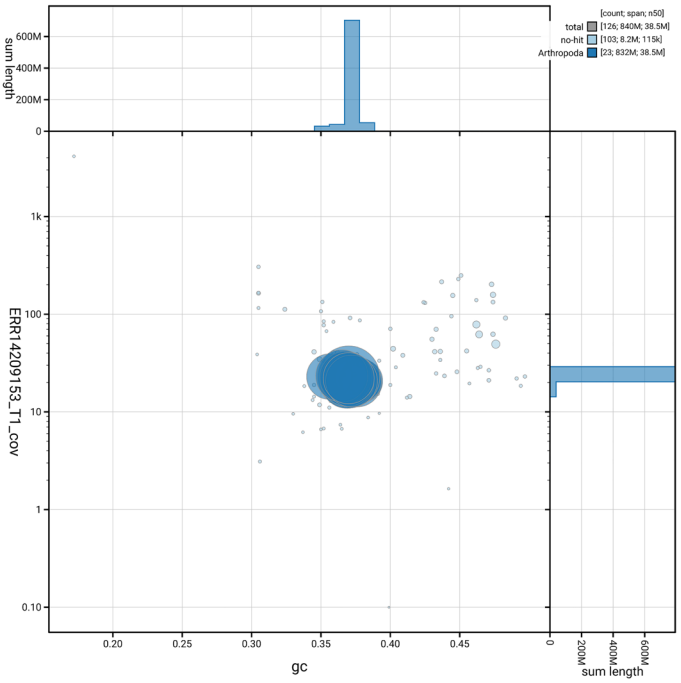

Figure 7. BlobToolKit GC-coverage plot for ilTomBall1.hap1.1. Blob plot showing sequence coverage (vertical axis) and GC content (horizontal axis). The circles represent scaffolds, with the size proportional to scaffold length and the colour representing phylum membership. The histograms along the axes display the total length of sequences distributed across different levels of coverage and GC content. An interactive version of this figure is available on the BlobToolKit viewer.

Table 4. Earth Biogenome Project summary metrics for the *Tomares ballus* assembly.

Measure	Value	Benchmark
EBP summary (haplotype 1)	6.C.Q61	6.C.Q40
Contig N50 length	7.07 Mb	≥ 1 Mb
Scaffold N50 length	38.46 Mb	= chromosome N50
Consensus quality (QV)	Haplotype 1: 61.6; haplotype 2: 61.8; combined: 61.7	≥ 40
k-mer completeness	Haplotype 1: 74.54%; Haplotype 2: 74.25%; combined: 99.48%	≥ 95%
BUSCO	C:98.0% [S:97.4%; D:0.5%]; F:0.4%; M:1.7%; n:5 286	S > 90%; D < 5%
Percentage of assembly assigned to chromosomes	99.02%	≥ 90%

calculated for the haplotype 1, is **6.C.Q61**, meeting the recommended reference standard.

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: Tomares ballus. Accession number PRJEB85042. The genome sequence is released openly for reuse. The *Tomares ballus* genome sequencing initiative is part of the Sanger Institute Tree of Life Programme (PRJEB43745) and Project Psyche (PRJEB71705). 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 Ensembl at the European Bioinformatics Institute. Raw data and assembly accession identifiers are reported in Table 1 and Table 2.

Pipelines used for genome assembly at the WSI Tree of Life are available at https://pipelines.tol.sanger.ac.uk/pipelines. Table 5 lists software versions used in this study.

Table 5. Software versions and sources.

Software	Version	Source
BEDTools	2.30.0	https://github.com/arq5x/bedtools2
BLAST	2.14.0	ftp://ftp.ncbi.nlm.nih.gov/blast/executables/blast+/
BlobToolKit	4.3.9	https://github.com/blobtoolkit/blobtoolkit
BUSCO	5.5.0	https://gitlab.com/ezlab/busco
bwa-mem2	2.2.1	https://github.com/bwa-mem2/bwa-mem2
Cooler	0.8.11	https://github.com/open2c/cooler
DIAMOND	2.1.8	https://github.com/bbuchfink/diamond
fasta_windows	0.2.4	https://github.com/tolkit/fasta_windows

Software	Version	Source
FastK	1.1	https://github.com/thegenemyers/FASTK
GenomeScope2.0	2.0.1	https://github.com/tbenavi1/genomescope2.0
Gfastats	1.3.6	https://github.com/vgl-hub/gfastats
Hifiasm	0.19.8-r603	https://github.com/chhylp123/hifiasm
HiGlass	1.13.4	https://github.com/higlass/higlass
lep_busco_painter	1.0.0	https://github.com/charlottewright/lep_busco_painter
MerquryFK	1.1.2	https://github.com/thegenemyers/MERQURY.FK
Minimap2	2.24-r1122	https://github.com/lh3/minimap2
MitoHiFi	3	https://github.com/marcelauliano/MitoHiFi
MultiQC	1.14; 1.17 and 1.18	https://github.com/MultiQC/MultiQC
Nextflow	23.10.0	https://github.com/nextflow-io/nextflow
PretextSnapshot	0.0.5	https://github.com/sanger-tol/PretextSnapshot
PretextView	1.0.3	https://github.com/sanger-tol/PretextView
samtools	1.19.2	https://github.com/samtools/samtools
sanger-tol/ascc	0.1.0	https://github.com/sanger-tol/ascc
sanger-tol/blobtoolkit	0.6.0	https://github.com/sanger-tol/blobtoolkit
sanger-tol/curationpretext	1.4.2	https://github.com/sanger-tol/curationpretext
Seqtk	1.3	https://github.com/lh3/seqtk
Singularity	3.9.0	https://github.com/sylabs/singularity
TreeVal	1.4.0	https://github.com/sanger-tol/treeval
YaHS	1.2.2	https://github.com/c-zhou/yahs

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