



DATA NOTE

The genome sequence of the Ornate Brigadier fly, *Odontomyia*

ornata (Meigen, 1822)

[version 1; peer review: 3 approved]

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Abstract

We present a genome assembly from a male specimen of *Odontomyia ornata* (Ornate Brigadier; Arthropoda; Insecta; Diptera; Stratiomyidae). The genome sequence has a total length of 2,045.35 megabases. Most of the assembly (99.0%) is scaffolded into 7 chromosomal pseudomolecules, including the X and Y sex chromosomes. The mitochondrial genome has also been assembled, with a length of 16.12 kilobases.

Keywords

Odontomyia ornata, Ornate Brigadier, genome sequence, chromosomal, Diptera



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

Open Peer Review

Approval Status

	1	2	3
version 1			
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Species taxonomy

Eukaryota; Opisthokonta; Metazoa; Eumetazoa; Bilateria; Protostomia; Ecdysozoa; Panarthropoda; Arthropoda; Mandibulata; Pancrustacea; Hexapoda; Insecta; Dicondylia; Pterygota; Neoptera; Endopterygota; Diptera; Brachycera; Stratiomyomorpha; Stratiomyidae; Stratiomyinae; Stratiomyini; *Odontomyia*; *Odontomyia ornata* (Meigen, 1822) (NCBI:txid3037408)

Background

Odontomyia ornata is a soldierfly belonging to the family Stratiomyidae. It is a large fly, with a body length of between 12.5 mm and 14.5 mm, and with striking orange markings on the sides of its otherwise black tergites (Stubbs & Drake, 2014). It is strongly associated with ditches in coastal grazing marshes, where it is considered an indicator of good habitat quality, but does occur inland.

It is often easier to find the large elongate larvae than it is the adults. These larvae inhabit ditches, where they are either free-floating or crawling among aquatic vegetation near the surface. Ditches more than 1m wide are preferred, as are those with a rich and structurally-diverse cover of vegetation floating near the surface, rather than ditches that are choked with emerged vegetation (Stubbs & Drake, 2014). The adults, which fly between late May and early July, often seek nectar on the flowers of Hemlock Water-dropwort *Oenanthe crocata*, a tall white-flowered member of the Apiaceae.

The fly is found across the temperate zone of Europe, especially in England, Wales and the Netherlands, with occasional records in the Mediterranean zone (GBIF Secretariat, 2023, accessed 18 March 2025). In Britain it is considered Nationally Scarce, with strongholds in the Somerset Levels, Gwent Levels and Norfolk Broads (Harvey, 2018).

Stubbs and Drake (2014) provide identification keys and descriptions to both larvae and adults; Zeegers and Schulten (2022) covers the adult stage. Adults are similar in size and appearance to members of the genus *Stratiomys* (the 'Generals'), but have much shorter antennae, lacking an elongated first segment.

Here we present a chromosomally complete genome sequence for *Odontomyia ornata*, based on a specimen from Upton Broad and Marshes, England, United Kingdom (Figure 1).

Genome sequence report

Sequencing data

The genome of a specimen of *Odontomyia ornata* (Figure 1) was sequenced using Pacific Biosciences single-molecule HiFi long reads, generating 91.12 Gb from 7.94 million reads, which were used to assemble the genome. GenomeScope analysis estimated the haploid genome size at 1,739.21 Mb, with a heterozygosity of 2.19% and repeat content of 46.64%. These estimates guided expectations for the assembly. Based on the estimated genome size, the sequencing data provided approximately 50 coverage. Hi-C sequencing produced 92.79 Gb

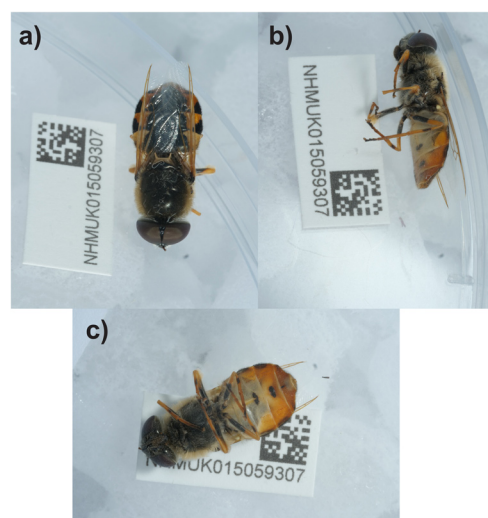


Figure 1. Photographs of the *Odontomyia ornata* (idOdoOrna1) specimen used for genome sequencing. **a)** Dorsal view; **b)** Lateral view; **c)** Central view.

from 614.49 million reads, and was used to scaffold the assembly. RNA sequencing data were also generated and are available in public sequence repositories. Table 1 summarises the specimen and sequencing details.

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 95 misjoins or missing joins and removed 3 haplotypic duplications. These interventions decreased the scaffold count by 14.64% and increased the scaffold N50 by 4.0%. The final assembly has a total length of 2,045.35 Mb in 203 scaffolds, with 614 gaps, and a scaffold N50 of 357.69 Mb (Table 2).

The snail plot in Figure 2 provides a summary of the assembly statistics, indicating the distribution of scaffold lengths and other assembly metrics. Figure 3 shows the distribution of scaffolds by GC proportion and coverage. Figure 4 presents a cumulative assembly plot, with separate curves representing different scaffold subsets assigned to various phyla, illustrating the completeness of the assembly.

Most of the assembly sequence (99.0%) was assigned to 7 chromosomal-level scaffolds, representing 5 autosomes and the X and Y sex chromosomes. These chromosome-level scaffolds, confirmed by Hi-C data, are named according to size (Figure 5; 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.

Table 1. Specimen and sequencing data for *Odontomyia ornata*.

Project information			
Study title	Odontomyia ornata		
Umbrella BioProject	PRJEB66027		
Species	Odontomyia ornata		
BioSpecimen	SAMEA112964298		
NCBI taxonomy ID	3037408		
Specimen information			
Technology	ToLID	BioSample accession	Organism part
PacBio long read sequencing	idOdoOrna1	SAMEA112975468	whole organism
Hi-C sequencing	idOdoOrna1	SAMEA112975468	whole organism
RNA sequencing	idOdoOrna1	SAMEA112975468	whole organism
Sequencing information			
Platform	Run accession	Read count	Base count (Gb)
Hi-C Illumina NovaSeq 6000	ERR12071236	6.14e+08	92.79
PacBio Revio	ERR12055565	7.94e+06	91.12
RNA Illumina NovaSeq X	ERR14792815	9.29e+07	14.02

Assembly quality metrics

The estimated Quality Value (QV) and *k*-mer completeness metrics, along with BUSCO completeness scores, were calculated for each haplotype and the combined assembly. The QV reflects the base-level accuracy of the assembly, while *k*-mer completeness indicates the proportion of expected *k*-mers identified in the assembly. BUSCO scores provide a measure of completeness based on benchmarking universal single-copy orthologues.

The combined primary and alternate assemblies achieve an estimated QV of 58.9. The *k*-mer recovery for the primary haplotype is 70.78%, and for the alternate haplotype 65.90%; the combined primary and alternate assemblies have a *k*-mer recovery of 98.82%. BUSCO v.5.5.0 analysis using the diptera_odb10 reference set (*n* = 3,285) identified 93.6% of the expected gene set (single = 92.5%, duplicated = 1.1%).

Table 2 provides assembly metric benchmarks adapted from Rhie *et al.* (2021) and the Earth BioGenome Project Report on Assembly Standards September 2024. The assembly achieves the EBP reference standard of 6.C.Q58.

Methods

Sample acquisition and DNA barcoding

The specimen used for genome sequencing was an adult male *Odontomyia ornata* (specimen ID NHMUK015059307, ToLID idOdoOrna1), collected from Upton Broad and Marshes, England, United Kingdom (latitude 52.67, longitude 1.52) on 2022-07-06 by handpicking. The specimen was collected by

Robert Wolton (Dipterists Forum) and identified by Martin Drake (Freshwater Biology Association) and preserved by dry freezing (−80 °C).

The initial identification was verified by an additional DNA barcoding process according to the framework developed by Twyford *et al.* (2024). A small sample was dissected from the specimen and stored in ethanol, while the remaining parts were shipped on dry ice to the Wellcome Sanger Institute (WSI) (Pereira *et al.*, 2022). The tissue was lysed, the COI marker region was amplified by PCR, and amplicons were sequenced and compared to the BOLD database, confirming the species identification (Crowley *et al.*, 2023). Following whole genome sequence generation, the relevant DNA barcode region was also used alongside the initial barcoding data for sample tracking at the WSI (Twyford *et al.*, 2024). The standard operating procedures for Darwin Tree of Life barcoding have been deposited on protocols.io (Beasley *et al.*, 2023).

Metadata collection for samples adhered to the Darwin Tree of Life project standards described by Lawniczak *et al.* (2022).

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 (Denton *et al.*, 2023b). The idOdoOrna1 sample was prepared for DNA extraction by weighing and dissecting it on dry ice

Table 2. Genome assembly data for *Odontomyia ornata*.

Genome assembly		
Assembly name	idOdoOrna1.1	
Assembly accession	GCA_963969285.1	
Alternate haplotype accession	GCA_963969255.1	
Assembly level for primary assembly	chromosome	
Span (Mb)	2,045.35	
Number of contigs	817	
Number of scaffolds	203	
Longest scaffold (Mb)	500.42	
Assembly metric	Measure	Benchmark
Contig N50 length	7.36 Mb	≥ 1 Mb
Scaffold N50 length	357.69 Mb	= chromosome N50
Consensus quality (QV)	Primary: 58.6; alternate: 59.1; combined: 58.9	≥ 40
k-mer completeness	Primary: 70.78%; alternate: 65.90%; combined: 98.82%	$\geq 95\%$
BUSCO*	C:93.6%[S:92.5%,D:1.1%], F:1.7%,M:4.7%,n:3,285	$S > 90\%$; $D < 5\%$
Percentage of assembly assigned to chromosomes	99.0%	$\geq 90\%$
Sex chromosomes	X and Y	localised homologous pairs
Organelles	Mitochondrial genome: 16.12 kb	complete single alleles

* BUSCO scores based on the diptera_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.

(Jay *et al.*, 2023). Tissue from the whole organism was homogenised using a PowerMasher II tissue disruptor (Denton *et al.*, 2023a). HMW DNA was extracted in the WSI Scientific Operations core using the Automated MagAttract v2 protocol (Oatley *et al.*, 2023). The DNA was sheared into an average fragment 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 (Strickland *et al.*, 2023). 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 extracted from whole organism tissue of idOdoOrna1 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.

Hi-C sample preparation and crosslinking

Hi-C data were generated from the whole organism of the idOdoOrna1 sample using the Arima-HiC v2 kit (Arima Genomics) with 20–50 mg of frozen tissue (stored at -80°C). As per manufacturer's instructions, tissue was fixed, and the DNA crosslinked using a TC buffer with 22% formaldehyde concentration, and a final formaldehyde concentration of 2%. The tissue was then homogenised using the Diagnocine Power Masher-II. The crosslinked DNA was digested using a restriction enzyme master mix, then biotinylated and ligated. A clean up was performed with SPRIselect beads prior to library preparation. DNA concentration was quantified using the Qubit Fluorometer v4.0 (Thermo Fisher Scientific) and Qubit HS Assay Kit, and sample biotinylation percentage was estimated using the Arima-HiC v2 QC beads.

Library preparation and sequencing

Library preparation and sequencing were performed at the WSI Scientific Operations core.

PacBio HiFi

At a minimum, samples were required to have an average fragment size exceeding 8 kb and a total mass over 400 ng

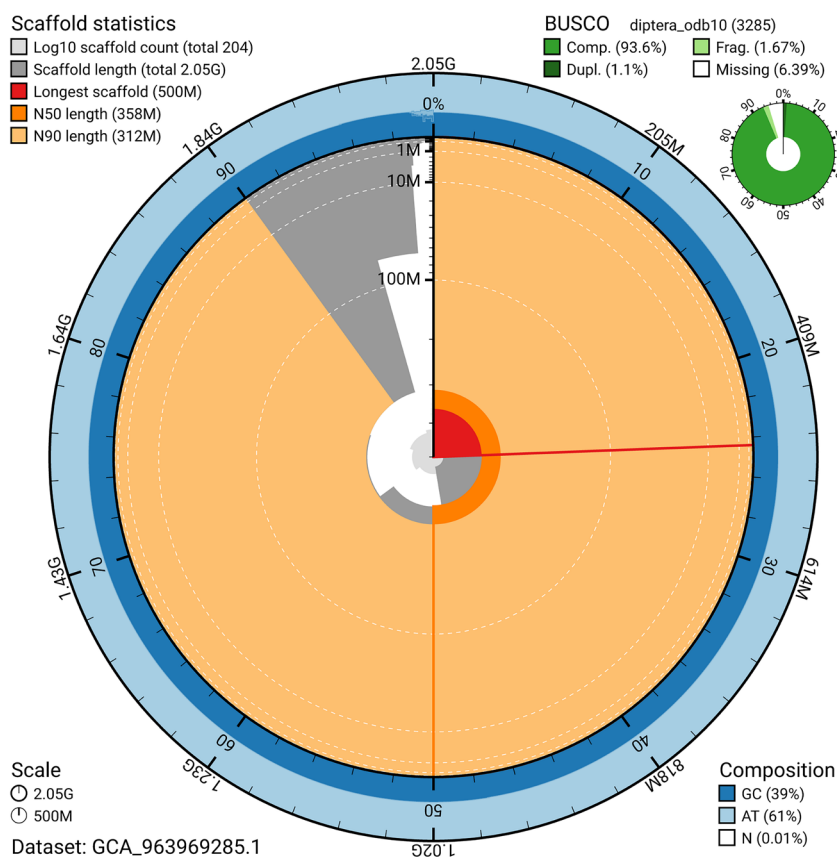


Figure 2. Genome assembly of *Odontomyia ornata*, idOdoOrna1.1: metrics. The BlobToolKit snail plot provides an overview of assembly metrics and BUSCO gene completeness. The circumference represents the length of the whole genome sequence, and the main plot is divided into 1,000 bins around the circumference. 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 diptera_odb10 set is presented at the top right. An interactive version of this figure is available at https://blobtoolkit.genomehubs.org/view/GCA_963969285.1/dataset/GCA_963969285.1/snail.

to proceed to the low-input SMRTbell Prep Kit 3.0 protocol (Pacific Biosciences), depending on genome size and sequencing depth required. Libraries were prepared using the SMRTbell Prep Kit 3.0 as per the manufacturer's instructions. The kit includes the reagents required for end repair/A-tailing, adapter ligation, post-ligation SMRTbell bead cleanup, and nuclease treatment. Size-selection and clean-up were carried out using diluted AMPure PB beads (Pacific Biosciences). DNA concentration was quantified using the Qubit Fluorometer v4.0 (ThermoFisher Scientific) with Qubit 1X dsDNA HS assay kit and the final library fragment size analysis was carried out using the Agilent Femto Pulse Automated Pulsed Field CE Instrument (Agilent Technologies) and the gDNA 55kb BAC analysis kit.

Samples were sequenced on a Revio instrument (Pacific Biosciences, California, USA). Prepared libraries were normalised to 2 nM, and 15 μ L was used for making complexes. Primers were annealed and polymerases were bound to create circularised complexes according to manufacturer's instructions.

The complexes were purified with the 1.2X clean up with SMRTbell beads. The purified complexes were then diluted to the Revio loading concentration (in the range 200–300 pM), and spiked with a Revio sequencing internal control. Samples were sequenced on Revio 25M SMRT cells (Pacific Biosciences, California, USA). The SMRT link software, a PacBio web-based end-to-end workflow manager, was used to set-up and monitor the run, as well as perform primary and secondary analysis of the data upon completion.

Hi-C

For Hi-C library preparation, the biotinylated DNA constructs were fragmented using a Covaris E220 sonicator and size-selected to 400–600 bp using SPRISelect beads. DNA was then enriched using Arima-HiC v2 Enrichment beads. The NEBNext Ultra II DNA Library Prep Kit (New England Biolabs) was used for end repair, A-tailing, and adapter ligation, following a modified protocol in which library preparation is carried out while the DNA remains bound to the enrichment

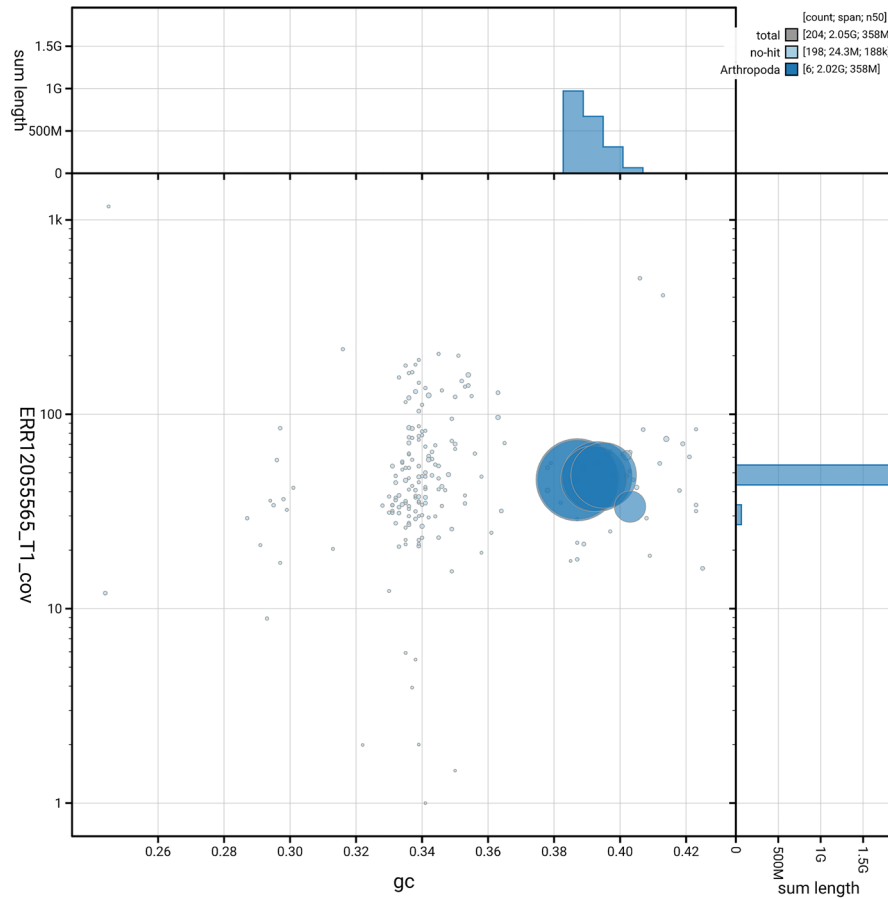


Figure 3. Genome assembly of *Odontomyia ornata*, idOdoOrna1.1: BlobToolKit GC-coverage plot. Blob plot showing sequence coverage (vertical axis) and GC content (horizontal axis). The circles represent scaffolds, with the size proportional to scaffold length and the colour representing phylum membership. 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_963969285.1/dataset/GCA_963969285.1/blob.

beads. PCR amplification was performed using KAPA HiFi HotStart mix and custom dual-indexed adapters (Integrated DNA Technologies) in a 96-well plate format. Depending on sample concentration and biotinylation percentage determined at the crosslinking stage, samples were amplified for 10–16 PCR cycles. Post-PCR clean-up was carried out using SPRIselect beads. The libraries were quantified using the Accuclear Ultra High Sensitivity dsDNA Standards Assay kit (Biotium) and normalised to 10 ng/μL before sequencing. Hi-C sequencing was performed on the Illumina NovaSeq 6000 instrument with 150 bp paired-end reads.

RNA

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

The HiFi reads were first 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 (Rao *et al.*, 2014) were mapped to the primary contigs using bwa-mem2 (Vasimuddin *et al.*, 2019), and the contigs were scaffolded using YaHS (Zhou *et al.*, 2023) using the --break option for handling potential misassemblies. The scaffolded assemblies were evaluated using

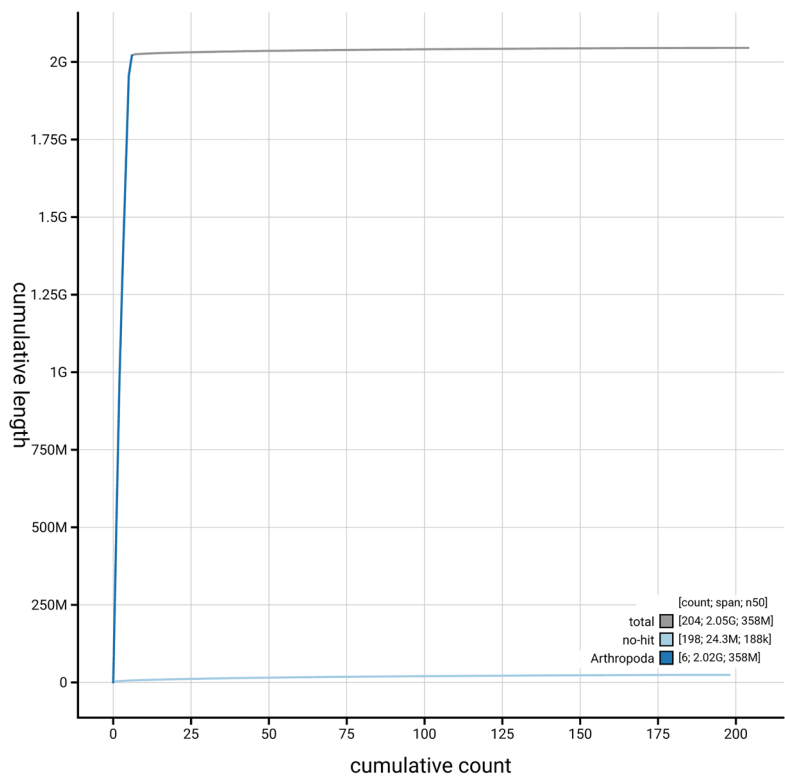


Figure 4. Genome assembly of *Odontomyia ornata*, idOdoOrna1.1: BlobToolKit cumulative sequence plot. The grey line shows cumulative length for all scaffolds. Coloured lines show cumulative lengths of scaffolds assigned to each phylum using the buscogenes taxrule. An interactive version of this figure is available at https://blobtoolkit.genomehubs.org/view/GCA_963969285.1/dataset/GCA_963969285.1/cumulative.

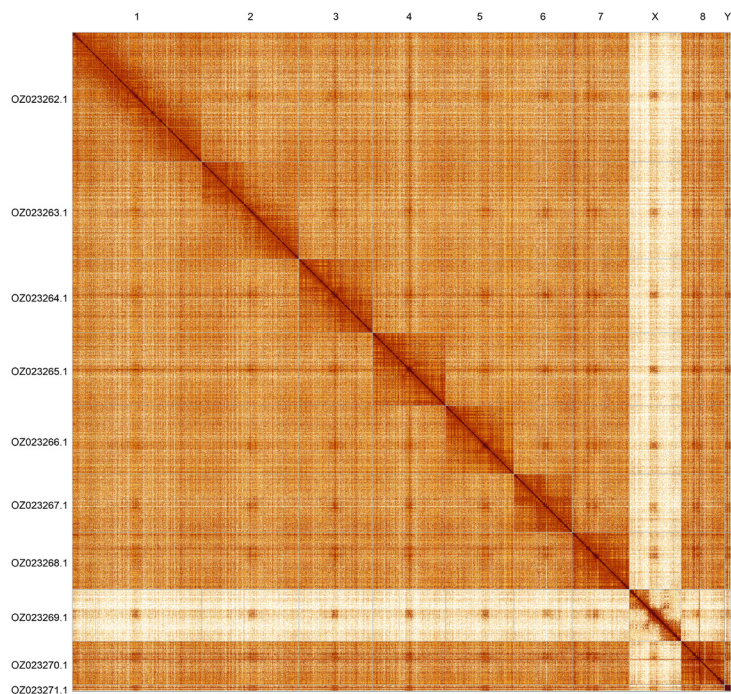


Figure 5. Hi-C contact map of the idOdoOrna1.1 assembly. The map was generated using PretextSnapshot. Chromosomes are shown in order of size and labelled with chromosome accession numbers (left) and names (top).

Table 3. Chromosomal pseudomolecules in the genome assembly of *Odontomyia ornata*, idOdoOrna1.

INSDC accession	Name	Length (Mb)	GC%
OZ017738.1	1	500.42	38.5
OZ017739.1	2	470.53	38.5
OZ017740.1	3	357.69	39.5
OZ017741.1	4	314.81	39
OZ017742.1	5	312.19	39.5
OZ017743.1	X	65.4	40.5
OZ017744.1	Y	3.79	40
OZ017745.1	MT	0.02	25

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. 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. Sex chromosomes were identified by read coverage analysis. The curation process is documented at <https://gitlab.com/wtsi-grit/rapid-curation>. PretextViewSnapshot 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 the primary and alternate 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 in the blobtoolkit pipeline, a Nextflow (Di Tommaso *et al.*, 2017) port of the previous Snake-make Blobtoolkit pipeline (Challis *et al.*, 2020). It aligns the PacBio reads in SAMtools (Danecek *et al.*, 2021) and minimap2 (Li, 2018) and generates coverage tracks for regions of fixed

size. In parallel, it queries the GoAT database (Challis *et al.*, 2023) to identify all matching BUSCO lineages to run BUSCO (Manni *et al.*, 2021). For the three domain-level BUSCO lineages, the pipeline aligns the BUSCO genes to the UniProt Reference Proteomes database (Bateman *et al.*, 2023) with DIAMOND blastp (Buchfink *et al.*, 2021). The genome is also 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 database using DIAMOND blastx. Genome sequences without a hit are chunked using seqtk and aligned to the NT database with blastn (Altschul *et al.*, 1990). The blobtools suite combines all these 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), relying on the Conda package manager, the Bioconda initiative (Grüning *et al.*, 2018), the Biocontainers infrastructure (da Veiga Leprevost *et al.*, 2017), as well as the Docker (Merkel, 2014) and Singularity (Kurtzer *et al.*, 2017) containerisation solutions.

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

Wellcome Sanger Institute – Legal and Governance

The materials that have contributed to this genome note have been supplied by a Darwin Tree of Life Partner. The submission of materials by a Darwin Tree of Life Partner is subject to the ‘Darwin Tree of Life Project Sampling Code of Practice’, which can be found in full on the Darwin Tree of Life website [here](#). By agreeing with and signing up to the Sampling Code of Practice, the Darwin Tree of Life Partner agrees they will meet the legal and ethical requirements and standards set out within this document in respect of all samples acquired for, and supplied to, the Darwin Tree of Life Project.

Further, 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 further undertaken according to a Research Collaboration Agreement or Material Transfer Agreement entered into by the Darwin Tree of Life Partner, Genome Research Limited (operating as the Wellcome Sanger Institute), and in some circumstances other Darwin Tree of Life collaborators.

Table 4. Software tools: versions and sources.

Software tool	Version	Source
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
DIAMOND	2.1.8	https://github.com/bbuchfink/diamond
fasta_windows	0.2.4	https://github.com/tolkit/fasta_windows
FastK	666652151335353eef2fcd58880bcef5bc2928e1	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
Goat CLI	0.2.5	https://github.com/genomehubs/goat-cli
Hifiasm	0.19.5-r587	https://github.com/chhyllp123/hifiasm
HiGlass	44086069ee7d4d3f6f3f0012569789ec138f42b84a a44357826c0b6753eb28de	https://github.com/higlass/higlass
MercuryFK	d00d98157618f4e8d1a9190026b19b471055b22e	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	23.04.1	https://github.com/nextflow-io/nextflow
PretextView	0.2.5	https://github.com/sanger-tol/PretextView
purge_dups	1.2.5	https://github.com/dfguan/purge_dups
samtools	1.19.2	https://github.com/samtools/samtools
sanger-tol/ascc	0.1.0	https://github.com/sanger-tol/ascc
sanger-tol/blobtoolkit	0.4.0	https://github.com/sanger-tol/blobtoolkit
Seqtk	1.3	https://github.com/lh3/seqtk
Singularity	3.9.0	https://github.com/sylabs/singularity
TreeVal	1.2.0	https://github.com/sanger-tol/treeval
YaHS	1.2a.2	https://github.com/c-zhou/yahs

Data availability

European Nucleotide Archive: *Odontomyia ornata*. Accession number PRJEB66027; <https://identifiers.org/ena.embl/PRJEB66027>. The genome sequence is released openly for reuse. The *Odontomyia ornata* genome sequencing initiative is part of the Darwin Tree of Life Project (PRJEB40665) and the Sanger Institute Tree of Life Programme (PRJEB43745). 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

Members of the Natural History Museum Genome Acquisition Lab are listed here: <https://doi.org/10.5281/zenodo.12159242>.

Members of the Darwin Tree of Life Barcoding collective are listed here: <https://doi.org/10.5281/zenodo.12158331>.

Members of the Wellcome Sanger Institute Tree of Life Management, Samples and Laboratory team are listed here: <https://doi.org/10.5281/zenodo.12162482>.

Members of Wellcome Sanger Institute Scientific Operations: Sequencing Operations are listed here: <https://doi.org/10.5281/zenodo.12165051>.

Members of the Wellcome Sanger Institute Tree of Life Core Informatics team are listed here: <https://doi.org/10.5281/zenodo.12160324>.

Members of the Tree of Life Core Informatics collective are listed here: <https://doi.org/10.5281/zenodo.12205391>.

Members of the Darwin Tree of Life Consortium are listed here: <https://doi.org/10.5281/zenodo.4783558>.

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Charles Plessy 

Okinawa Institute of Science and Technology, Okinawa, Japan

This data note follows the standard structure used in the Tree of Life gateway. All genome quality indicators appear satisfactory. The BUSCO scores fall within the expected range for assemblies of related fly species. As of today, the closest species with a publicly released chromosome-level assembly is *Odontomyia argentata* (GCA_965615855.1). However, it is evolutionarily distant (~85% nucleotide sequence identity based on my own computation), making genome alignments unsuitable for detecting potential inconsistencies.

I am assigning an “approved” status to this review.

Nevertheless, I would appreciate if the authors could comment on the signal strength of interactions between the Y chromosome and the rest of the genome. I would have expected this to be roughly half of the average, similar to the X chromosome, given that sex chromosomes are haploid. However, the plot does not seem to reflect this. Could this be due to the Y chromosome being so short that the resolution of the plot does not allow this feature to be visible?

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?

Yes

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Whole-genome alignments

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 24 December 2025

<https://doi.org/10.21956/wellcomeopenres.26770.r141431>

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Darren Obbard 

The University of Edinburgh, Edinburgh, Scotland, UK

This data note reports the sequencing and assembly of the genome of *Odontomyia ornata* as part of the “Darwin Tree of Life” programme. In common with other data notes from this research effort, the reporting is standardised and quite brief. As such, I have very few comments to make.

The approach is state-of-the-art, the raw data appear to be of a suitably high quality, and the assembly methods are appropriate. The public availability of raw data and genome assembly are appropriate. The resulting genome is likely to be of very high quality, and I have no doubt that it will be of great value to any researchers working on soldier flies and other diptera, or on the comparative or evolutionary genomics of insects more generally. I particularly applaud the efforts to identify sex chromosomes, and to report useful summary statistics such as heterozygosity

At this point I usually have a number of minor suggestions focused on broadening the referenced literature and biology – but on this occasion, I really do not think there is anything to add. It's great.

My one suggestion is that, in addition to the sequenced specimen itself, it would be nice to have an ‘in life’ photograph and/or perhaps photographs both sexes. CC-BY images seem to be available from Wikimedia Commons and iNaturalist.

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?

Yes

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Evolutionary genetics of invertebrates and their pathogens

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 19 August 2025

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Andrés López-Rubio 

Tecnológico de Antioquia, Medellín, Colombia

A great research paper, it will contribute to the knowledge of Stratiomyid. As mentioned before, this paper out cases a remarkable analysis workflow to produce a fully annotated genome of a Stratiomyid fly. Previous to this paper NCBI reports only *Odontomyia argentata* to be sequenced. Since all the results are available, it will improve the sequencing of more species in the family.

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?

Yes

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Bioinformatics, phylogenetics

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.
