





DATA NOTE

The genome sequence of the Straw-barred Pearl moth,

Pyrausta despicata Scopoli, 1763

[version 1; peer review: awaiting peer review]

Gavin R. Broad ¹, David C. Lees ¹, Douglas Boyes²⁺,
Natural History Museum Genome Acquisition Lab,
University of Oxford and Wytham Woods Genome Acquisition Lab,
Darwin Tree of Life Barcoding collective,
Wellcome Sanger Institute Tree of Life Management, Samples and Laboratory
team,
Wellcome Sanger Institute Scientific Operations: Sequencing Operations,
Wellcome Sanger Institute Tree of Life Core Informatics team,
Tree of Life Core Informatics collective, Darwin Tree of Life Consortium

¹Natural History Museum, London, England, UK²UK Centre for Ecology & Hydrology, Wallingford, England, UK

+ Deceased author

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Abstract

We present a genome assembly from a male specimen of *Pyrausta despicata* (Straw-barred Pearl; Arthropoda; Insecta; Lepidoptera; Crambidae). The genome sequence has a total length of 481.83 megabases. Most of the assembly (99.61%) is scaffolded into 31 chromosomal pseudomolecules, including the Z sex chromosome. The mitochondrial genome has also been assembled, with a length of 15.29 kilobases.

Keywords

Pyrausta despicata, Straw-barred Pearl, genome sequence, chromosomal, Lepidoptera



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

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Approval Status Awaiting Peer Review

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

Corresponding author: Darwin Tree of Life Consortium (mark.blaxter@sanger.ac.uk)

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Species taxonomy

Eukaryota; Opisthokonta; Metazoa; Eumetazoa; Bilateria; Protostomia; Ecdysozoa; Panarthropoda; Arthropoda; Mandibulata; Pancrustacea; Hexapoda; Insecta; Dicondylia; Pterygota; Neoptera; Endopterygota; Amphimesenoptera; Lepidoptera; Glossata; Neolepidoptera; Heteroneura; Ditrysia; Obtectomera; Pyraloidea; Crambidae; Pyraustinae; *Pyrausta*; *Pyrausta despicata* Scopoli, 1763 (NCBI:txid1281420)

Background

Found throughout Britain, *Pyrausta despicata* is a rather unassuming crambid moth, basically greyish-brown with obscure cross-lines and stigmata. The vernacular, Straw-barred Pearl, references the sharply defined cream-coloured lines across the hind wing. Coastal specimens, especially from the Isles of Scilly and the Shetlands, are more contrastingly patterned on the fore wings, apparently more closely resembling southern European specimens (Beirne, 1954). In the older literature, this species was usually called *Pyrausta cespitalis*.

As with other *Pyrausta* species, *P. despicata* is partly diurnal, flying in the sunshine, and comes to light at night. While the more strikingly patterned *Pyrausta* species feed on Lamiaceae, particularly mints and thymes, *P. despicata* larvae feed gregariously on the leaves of plantains, *Plantago* (Plantaginaceae). They stay within silken galleries near the base of the plant by day, feeding on the undersides of leaves by night (Beirne, 1954; Goater *et al.*, 1986). *Pyrausta despicata* requires open areas but accepts a wide variety of such habitats, including coastal shingle, grasslands and heath. Davis (2012) classified this as a common species in Britain, upgrading it from its previous categorisation as ‘Local’. Adults are bivoltine, the first generation on the wing in the spring until early June and the second generation from July to September (Sterling *et al.*, 2023). The geographic range of *P. despicata* is extensive, from North Africa to Scandinavia and from Portugal to the Russian Far East, according to records on the GBIF.

The mitogenome of *P. despicata* has been sequenced (Wu *et al.*, 2023), but this is only the second full genome for a *Pyrausta* species, following that of *P. nigrata* (Lees *et al.*, 2024), with others in production. Comparative genomics of these closely related species will help in understanding why some *Pyrausta* species are thriving and others declining (Parsons & Clancy, 2023) and help resolve the increasingly well-sampled Crambidae phylogenetic tree (Léger *et al.*, 2021).

Genome sequence report

Sequencing data

The genome of a specimen of *Pyrausta despicata* (Figure 1) was sequenced using Pacific Biosciences single-molecule HiFi long reads, generating 78.73 Gb (gigabases) from 7.48 million reads. GenomeScope analysis of the PacBio HiFi data estimated the haploid genome size at 479.08 Mb, with a heterozygosity of 1.97% and repeat content of 31.24%. These values provide an initial assessment of genome complexity and the challenges anticipated during assembly. Based on this estimated genome size, the sequencing data provided approximately



Figure 1. Photograph of the *Pyrausta despicata* (ilPyrDesp2) specimen used for genome sequencing.

159.0x coverage of the genome. Chromosome conformation Hi-C sequencing produced 121.30 Gb from 803.29 million reads. 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 14 misjoins or missing joins and removed four haplotypic duplications. These interventions increased the scaffold count by 1.49%. The final assembly has a total length of 481.83 Mb in 67 scaffolds, with 29 gaps, and a scaffold N50 of 16.62 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 (99.62%) was assigned to 31 chromosomal-level scaffolds, representing 30 autosomes and the Z sex chromosome. These chromosome-level scaffolds, confirmed by Hi-C data, are named according to size (Figure 5; Table 3). During curation, chromosome Z was assigned based on synteny to the genome of *Pyrausta aurata* (GCA_963584085.1).

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.

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

Table 1. Specimen and sequencing data for *Pyrausta despicata*.

Project information			
Study title	Pyrausta despicata (straw-barred pearl)		
Umbrella BioProject	PRJEB65670		
Species	<i>Pyrausta despicata</i>		
BioSpecimen	SAMEA111458735		
NCBI taxonomy ID	1281420		
Specimen information			
Technology	ToLID	BioSample accession	Organism part
PacBio long read sequencing	ilPyrDesp2	SAMEA111458806	whole organism
Hi-C sequencing	ilPyrDesp1	SAMEA8603673	whole organism
Sequencing information			
Platform	Run accession	Read count	Base count (Gb)
Hi-C Illumina NovaSeq 6000	ERR12035243	8.03e+08	121.3
PacBio Revio	ERR12015734	7.48e+06	78.73

Table 2. Genome assembly data for *Pyrausta despicata*.

Genome assembly		
Assembly name	ilPyrDesp2.1	
Assembly accession	GCA_963921415.1	
Alternate haplotype accession	GCA_963921445.1	
Assembly level for primary assembly	chromosome	
Span (Mb)	481.83	
Number of contigs	96	
Number of scaffolds	67	
Longest scaffold (Mb)	19.84	
Assembly metric	Measure	Benchmark
Contig N50 length	11.59 Mb	≥ 1 Mb
Scaffold N50 length	16.62 Mb	= chromosome N50
Consensus quality (QV)	Primary: 67.3; alternate: 68.0; combined 67.6	≥ 40
k-mer completeness	Primary: 68.87%; alternate: 67.39%; combined: 99.79%	≥ 95%
BUSCO*	C:98.7%[S:98.5%,D:0.2%], F:0.2%,M:1.0%,n:5,286	S > 90%; D < 5%
Percentage of assembly mapped to chromosomes	99.62%	≥ 90%
Sex chromosomes	Z	localised homologous pairs
Organelles	Mitochondrial genome: 15.29 kb	complete single alleles

* BUSCO scores based on the lepidoptera_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.

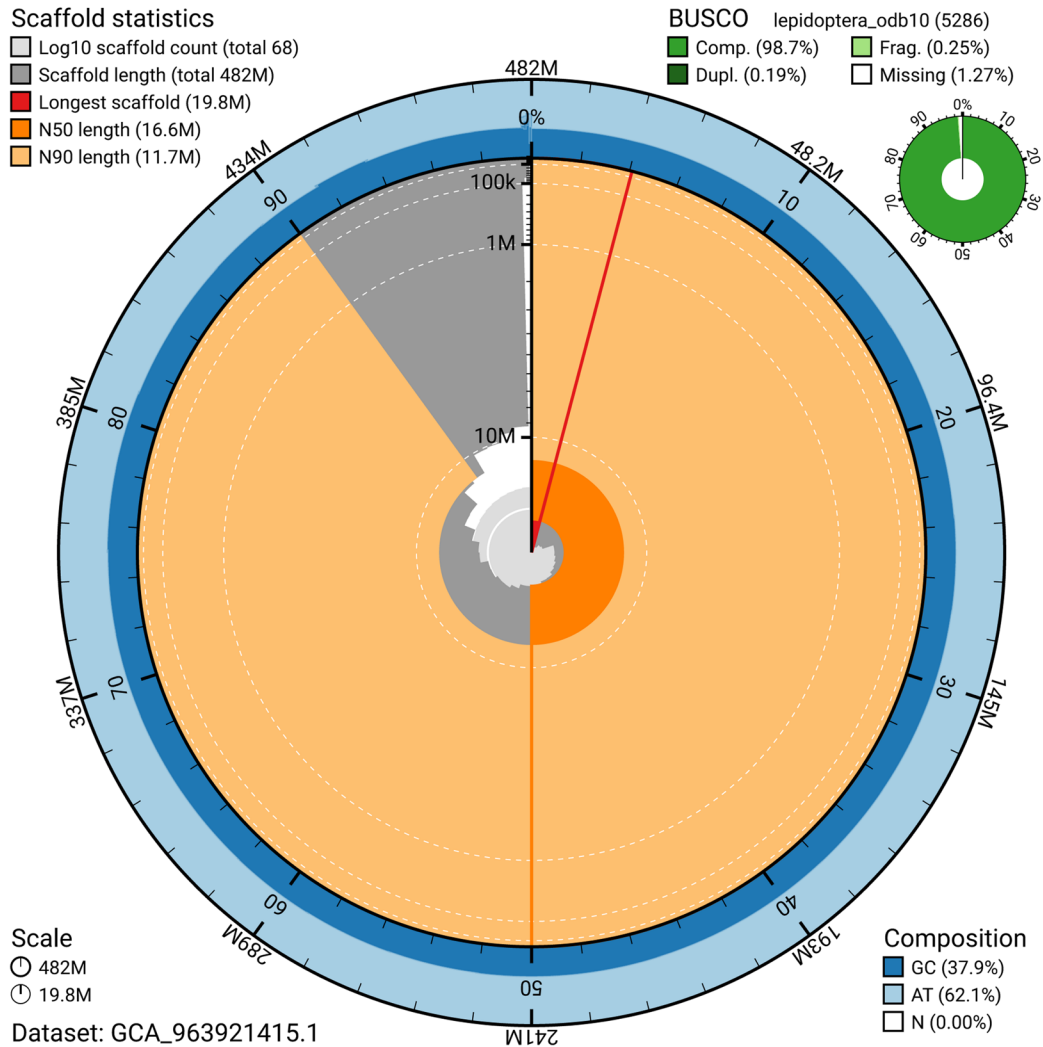


Figure 2. Genome assembly of *Pyrausta despicata*, ilPyrDesp2.1: metrics. The BlobToolKit snail plot provides an overview of assembly metrics and BUSCO gene completeness. The circumference represents the length of the whole genome sequence, and the main plot is divided into 1,000 bins around the circumference. 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 lepidoptera_odb10 set is presented at the top right. An interactive version of this figure is available at https://blobtoolkit.genomehubs.org/view/GCA_963921415.1/dataset/GCA_963921415.1/snail.

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 67.6. The k -mer recovery for the primary haplotype is 68.87%, and for the alternate haplotype 67.39%; the combined primary and alternate assemblies have a k -mer recovery of 99.79%. BUSCO analysis using the lepidoptera_odb10 reference set ($n = 5,286$) identified 98.7% of the expected gene set (single = 98.5%, duplicated = 0.2%).

Table 2 provides assembly metric benchmarks adapted from Rhie *et al.* (2021) and the Earth BioGenome Project (EBP) Report on Assembly Standards September 2024. The assembly achieves the EBP reference standard of 7.C.Q67.

Methods

Sample acquisition and DNA barcoding

An adult male *Pyrausta despicata* (specimen ID NHMUK 014425859, ToLID ilPyrDesp2) was collected from Tonbridge, England, United Kingdom (latitude 51.18, longitude 0.29) on 2021-09-01 by actinic light. The specimen was collected by

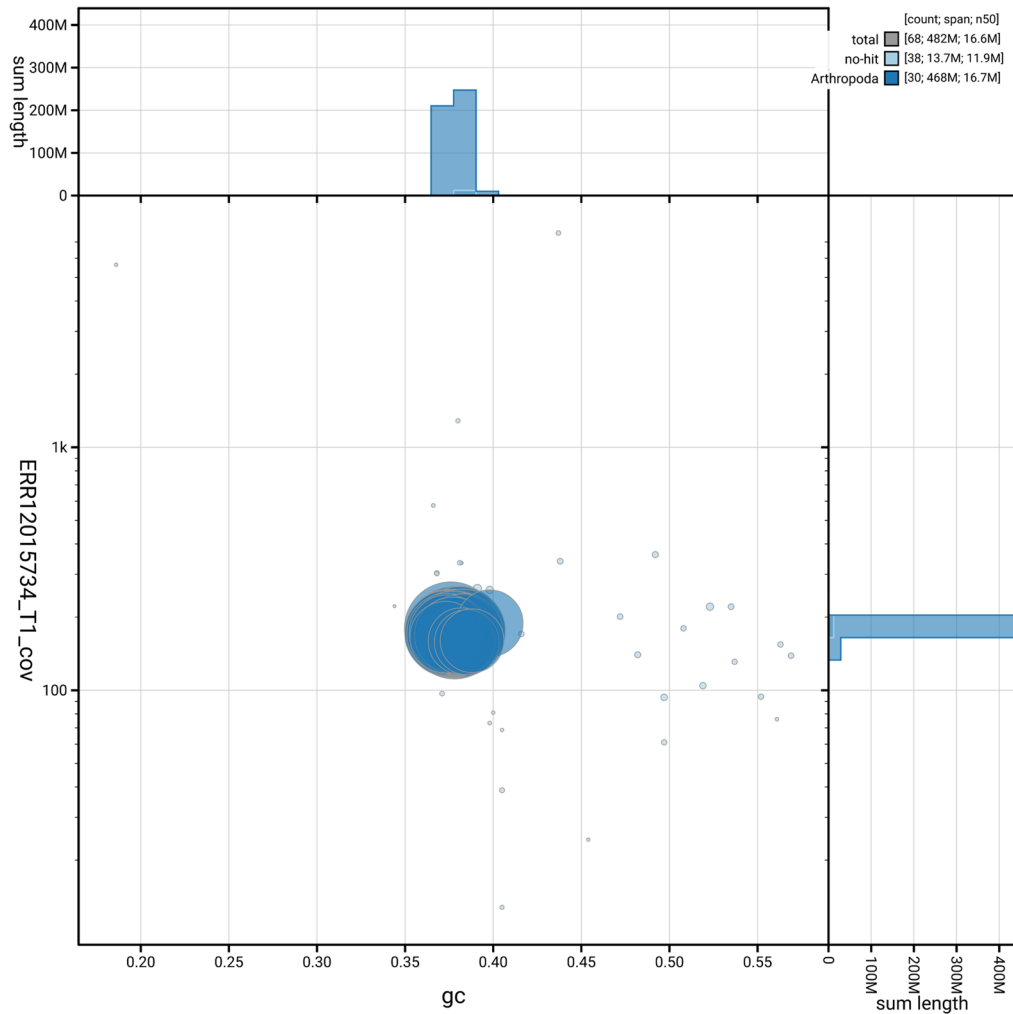


Figure 3. Genome assembly of *Pyrausta despicata*, ilPyrDesp2.1: BlobToolKit GC-coverage plot. Blob plot showing sequence coverage (vertical axis) and GC content (horizontal axis). The circles represent scaffolds, with the size proportional to scaffold length and the colour representing phylum membership. 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_963921415.1/dataset/GCA_963921415.1/blob.

Gavin Broad (Natural History Museum), identified by David Lees (Natural History Museum) and preserved by dry freezing (-80°C).

The specimen used for Hi-C sequencing (specimen ID Ox000962, ToLID ilPyrDesp1) was collected from Wytham Woods, Oxfordshire, United Kingdom (latitude 51.77, longitude -1.34) on 2020-09-08 using a light trap. The specimen was collected and identified by Douglas Boyes (University of Oxford) and preserved on dry ice.

The initial identification by Morphology 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

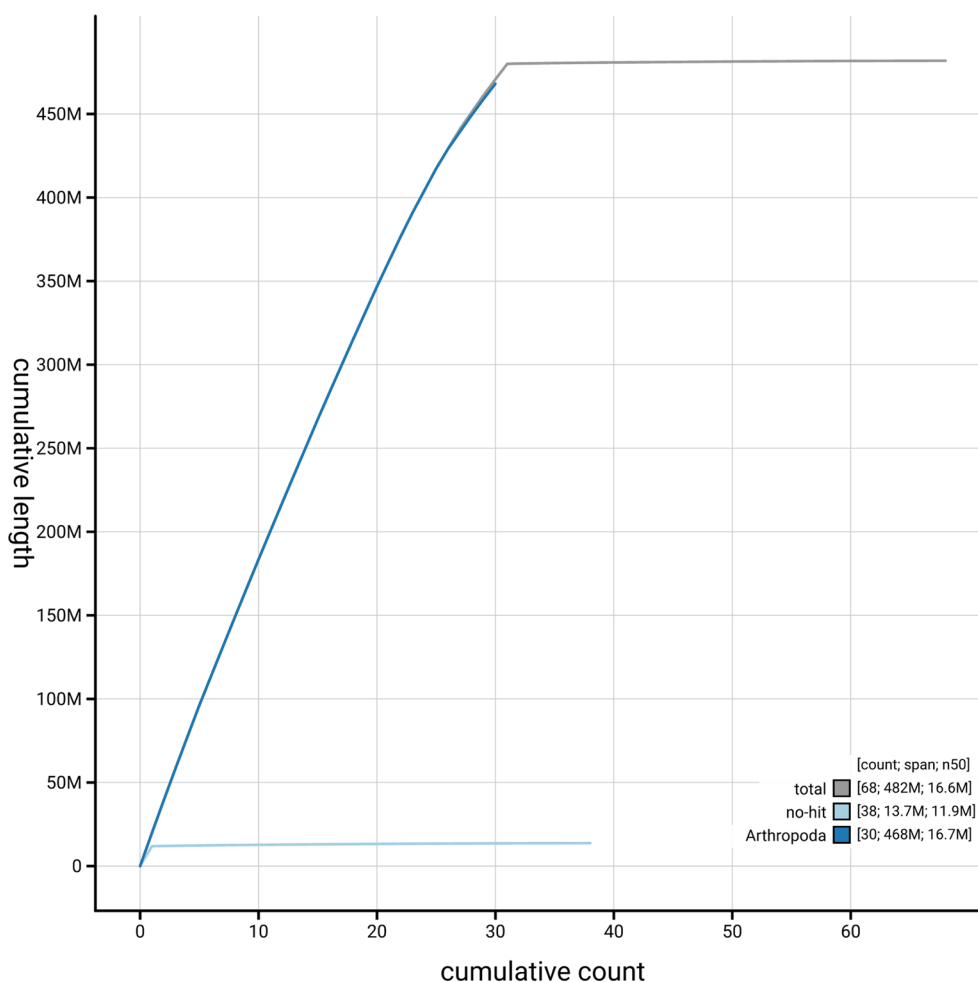


Figure 4. Genome assembly of *Pyrausta despicata*, ilPyrDesp2.1: BlobToolKit cumulative sequence plot. The grey line shows cumulative length for all scaffolds. Coloured lines show cumulative lengths of scaffolds assigned to each phylum using the *buscogenes* taxruler. An interactive version of this figure is available at https://blobtoolkit.genomehubs.org/view/GCA_963921415.1/dataset/GCA_963921415.1/cumulative.

purification. Detailed protocols are available on protocols.io (Denton *et al.*, 2023b). The ilPyrDesp2 sample was prepared for DNA extraction by weighing and dissecting it on dry ice (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.

Hi-C sample preparation

Tissue from the whole organism of the ilPyrDesp1 sample was processed for Hi-C sequencing at the WSI Scientific Operations core, using the Arima-HiC v2 kit. In brief, 20–50 mg of frozen tissue (stored at -80°C) was fixed, and the DNA crosslinked using a TC buffer with 22% formaldehyde concentration. After crosslinking, the tissue was homogenised using the Diagnocine Power Masher-II and BioMasher-II tubes and pestles. Following the Arima-HiC v2 kit manufacturer's instructions, crosslinked DNA was digested using a restriction enzyme master mix. The 5'-overhangs were filled in and labelled with biotinylated nucleotides and proximally ligated. An overnight incubation was carried out for enzymes to digest remaining proteins and for crosslinks to reverse. A clean up was performed with SPRIselect beads prior to library preparation. Additionally, the biotinylation percentage was estimated using the Qubit Fluorometer v4.0 (Thermo Fisher Scientific) and Qubit HS Assay Kit and Arima-HiC v2 QC beads.

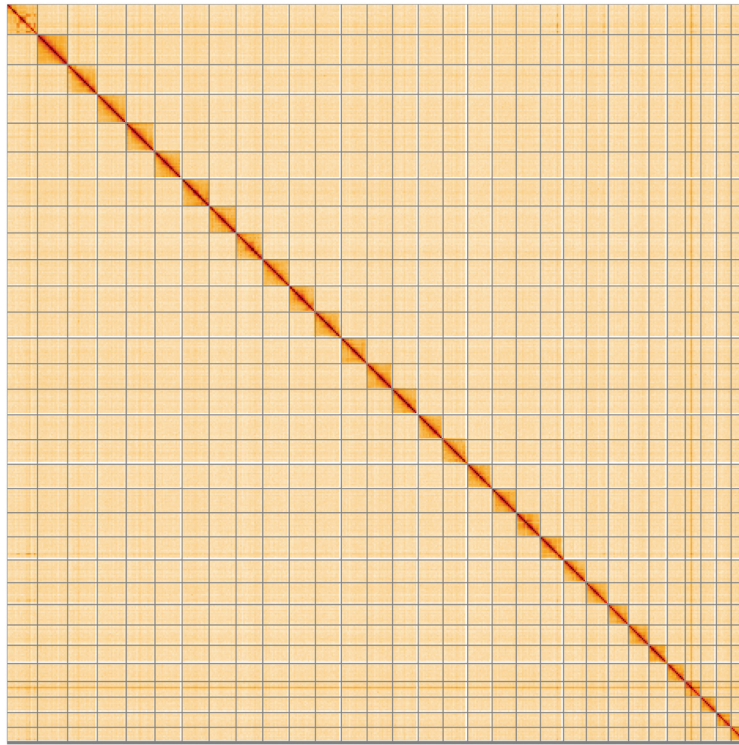


Figure 5. Genome assembly of *Pyrausta despicata*: Hi-C contact map of the iIPyrDesp2.1 assembly, visualised using HiGlass. Chromosomes are shown in order of size from left to right and top to bottom. An interactive version of this figure may be viewed at <https://genome-note-higlass.tol.sanger.ac.uk/I/?d=Rgrq5JISRYWvdUb5eRNsLg>.

Table 3. Chromosomal pseudomolecules in the genome assembly of *Pyrausta despicata*, iIPyrDesp2.

INSDC accession	Name	Length (Mb)	GC%
OY992844.1	1	19.84	37.5
OY992846.1	2	19.21	38
OY992847.1	3	18.84	37.5
OY992848.1	4	18.71	38
OY992849.1	5	17.62	37.5
OY992850.1	6	17.62	38
OY992851.1	7	17.5	37.5
OY992852.1	8	17.34	37.5
OY992853.1	9	17.22	38
OY992854.1	10	16.97	37.5
OY992855.1	11	16.93	37.5
OY992856.1	12	16.7	38
OY992857.1	13	16.62	38
OY992858.1	14	16.61	37.5
OY992859.1	15	16.23	38

INSDC accession	Name	Length (Mb)	GC%
OY992860.1	16	15.94	38.5
OY992861.1	17	15.92	38
OY992862.1	18	15.71	38
OY992863.1	19	15.67	37.5
OY992864.1	20	14.98	37.5
OY992865.1	21	14.89	38
OY992866.1	22	14.24	38.5
OY992867.1	23	13.28	37.5
OY992868.1	24	13.24	37.5
OY992869.1	25	11.88	38
OY992870.1	26	11.67	37.5
OY992871.1	27	10.16	40
OY992872.1	28	10.09	38
OY992873.1	29	9.55	38.5
OY992874.1	30	9.23	39
OY992845.1	Z	19.59	38
OY992875.1	MT	0.02	19

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 to proceed to the low input SMRTbell Prep Kit 3.0 protocol (Pacific Biosciences, California, USA), depending on genome size and sequencing depth required. Libraries were prepared using the SMRTbell Prep Kit 3.0 (Pacific Biosciences, California, USA) 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. Following the manufacturer's instructions, size selection and clean up was carried out using diluted AMPure PB beads (Pacific Biosciences, California, USA). DNA concentration was quantified using the Qubit Fluorometer v4.0 (Thermo Fisher 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 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 µL was used for making complexes. Primers were annealed and polymerases were hybridised 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, DNA was fragmented using the Covaris E220 sonicator (Covaris) and size selected using SPRISelect beads to 400 to 600 bp. The DNA was then enriched using the Arima-HiC v2 kit Enrichment beads. Using the NEBNext Ultra II DNA Library Prep Kit (New England Biolabs) for end repair, A-tailing, and adapter ligation. This uses a custom protocol which resembles the standard NEBNext Ultra II DNA Library Prep protocol but where library preparation occurs while DNA is bound to the Enrichment beads. For library amplification, 10 to 16 PCR cycles were required, determined by the sample biotinylation percentage. The Hi-C sequencing was performed using paired-end sequencing with a read length of 150 bp on an Illumina NovaSeq 6000 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 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 **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. The sex chromosome was identified by synteny analysis. The curation process is documented at <https://gitlab.com/wtsi-grit/rapid-curation>.

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 ($k = 31$) that were computed prior to genome assembly. The analysis outputs included assembly QV scores and completeness statistics.

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 contact map was visualised in **HiGlass** (Kerpedjiev *et al.*, 2018).

The **blobtoolkit** pipeline is a Nextflow port of the previous **Snakemake Blobtoolkit** pipeline (Challis *et al.*, 2020). It aligns the PacBio reads in **SAMtools** 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

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

Table 4. Software tools: versions and sources.

Software tool	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/e2lab/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
FastK	666652151335353eef2fcd58880bcef5bc2928e1	https://github.com/thegenemyers/FASTK
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/chhylyp123/hifiasm
HiGlass	44086069ee7d4d3f6f3f0012569789ec138f42b84aa44357826c0b6753eb28de	https://github.com/higlass/higlass
MercuryFK	d00d98157618f4e8d1a9190026b19b471055b22e	https://github.com/thegenemyers/MERURY.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	-	https://github.com/sanger-tol/ascc
sanger-tol/blobtoolkit	0.5.1	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

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.

Data availability

European Nucleotide Archive: *Pyrausta despicata* (straw-barred pearl). Accession number PRJEB65670; <https://identifiers.org/ena.embl/PRJEB65670>. The genome sequence is released openly for reuse. The *Pyrausta despicata* genome sequencing initiative is part of the Darwin Tree of Life (DToL) project

(PRJEB40665) 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 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 University of Oxford and Wytham Woods Genome Acquisition Lab are listed here: <https://doi.org/10.5281/zenodo.12157525>.

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.14870789>.

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>.

References

- Abdennur N, Mirny LA: **Cooler: scalable storage for Hi-C data and other genomically labeled arrays**. *Bioinformatics*. 2020; **36**(1): 311–316. [PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Allio R, Schomaker-Bastos A, Romiguiet J, et al.: **MitoFinder: efficient automated large-scale extraction of mitogenomic data in target enrichment phylogenomics**. *Mol Ecol Resour*. 2020; **20**(4): 892–905. [PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Altschul SF, Gish W, Miller W, et al.: **Basic Local Alignment Search Tool**. *J Mol Biol*. 1990; **215**(3): 403–410. [PubMed Abstract](#) | [Publisher Full Text](#)
- Bateman A, Martin MJ, Orchard S, et al.: **UniProt: the universal protein knowledgebase in 2023**. *Nucleic Acids Res*. 2023; **51**(D1): D523–D531. [PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Bates A, Clayton-Lucey I, Howard C: **Sanger Tree of Life HMW DNA fragmentation: diagenode Megaruptor[®]3 for LI PacBio**. *protocols.io*. 2023. [Publisher Full Text](#)
- Beasley J, Uhl R, Forrest LL, et al.: **DNA barcoding SOPs for the Darwin Tree of Life project**. *protocols.io*. 2023; [Accessed 25 June 2024]. [Publisher Full Text](#)
- Beirne BP: **British pyralid and plume moths**. London: Warne and Co, 1954.
- Buchfink B, Reuter K, Drost HG: **Sensitive protein alignments at Tree-of-Life scale using DIAMOND**. *Nat Methods*. 2021; **18**(4): 366–368. [PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Challis R, Kumar S, Sotero-Caio C, et al.: **Genomes on a Tree (GoAT): a versatile, scalable search engine for genomic and sequencing project metadata across the eukaryotic Tree of Life [version 1; peer review: 2 approved]**. *Wellcome Open Res*. 2023; **8**: 24. [PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Challis R, Richards E, Rajan J, et al.: **BlobToolKit – interactive quality assessment of genome assemblies**. *G3 (Bethesda)*. 2020; **10**(4): 1361–1374. [PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Cheng H, Concepcion GT, Feng X, et al.: **Haplotype-resolved *de novo* assembly using phased assembly graphs with hifiasm**. *Nat Methods*. 2021; **18**(2): 170–175. [PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Crowley L, Allen H, Barnes I, et al.: **A sampling strategy for genome sequencing the British terrestrial arthropod fauna [version 1; peer review:**

- 2 approved]. *Wellcome Open Res.* 2023; **8**: 123.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- da Veiga Leprevost F, Grüning BA, Alves Aflitos S, *et al.*: **BioContainers: an open-source and community-driven framework for software standardization.** *Bioinformatics.* 2017; **33**(16): 2580–2582.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Danecek P, Bonfield JK, Liddle J, *et al.*: **Twelve years of SAMtools and BCFtools.** *GigaScience.* 2021; **10**(2): giab008.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Davis T: **A review of the status of microlepidoptera in Britain. Butterfly conservation Report No. S12-02.** East Lulworth, Wareham, Dorset: Butterfly Conservation, 2012.
[Reference Source](#)
- Denton A, Oatley G, Cornwell C, *et al.*: **Sanger Tree of Life sample homogenisation: PowerMash.** *protocols.io.* 2023a.
[Publisher Full Text](#)
- Denton A, Yatsenko H, Jay J, *et al.*: **Sanger Tree of Life wet laboratory protocol collection V.1.** *protocols.io.* 2023b.
[Publisher Full Text](#)
- Diesch C, Stevens GJ, Xie P, *et al.*: **JBrowse 2: a modular genome browser with views of synteny and structural variation.** *Genome Biol.* 2023; **24**(1): 74.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Ewels P, Magnusson M, Lundin S, *et al.*: **MultiQC: summarize analysis results for multiple tools and samples in a single report.** *Bioinformatics.* 2016; **32**(19): 3047–3048.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Ewels PA, Peltzer A, Fillinger S, *et al.*: **The nf-core framework for community-curated bioinformatics pipelines.** *Nat Biotechnol.* 2020; **38**(3): 276–278.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Formenti G, Abueg L, Brajuka A, *et al.*: **Gfastats: conversion, evaluation and manipulation of genome sequences using assembly graphs.** *Bioinformatics.* 2022; **38**(17): 4214–4216.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Goater B, Senior G, Dyke R: **British Pyralid Moths.** Colchester: Harley Books, 1986.
[Reference Source](#)
- Grüning B, Dale R, Sjödin A, *et al.*: **Bioconda: sustainable and comprehensive software distribution for the life sciences.** *Nat Methods.* 2018; **15**(7): 475–476.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Guan D, McCarthy SA, Wood J, *et al.*: **Identifying and removing haplotypic duplication in primary genome assemblies.** *Bioinformatics.* 2020; **36**(9): 2896–2898.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Harry E: **PretextView (Paired REad TEXTure Viewer): a desktop application for viewing pretext contact maps.** 2022.
[Reference Source](#)
- Howe K, Chow W, Collins J, *et al.*: **Significantly improving the quality of genome assemblies through curation.** *GigaScience.* 2021; **10**(1): gjaa153.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Jay J, Yatsenko H, Narváez-Gómez JP, *et al.*: **Sanger Tree of Life sample preparation: triage and dissection.** *protocols.io.* 2023.
[Publisher Full Text](#)
- Kerpedjiev P, Abdennur N, Lekschas F, *et al.*: **HiGlass: web-based visual exploration and analysis of genome interaction maps.** *Genome Biol.* 2018; **19**(1): 125.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Kurtzer GM, Sochat V, Bauer MW: **Singularity: scientific containers for mobility of compute.** *PLoS One.* 2017; **12**(5): e0177459.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Lawniczak MKN, Davey RP, Rajan J, *et al.*: **Specimen and sample metadata standards for biodiversity genomics: a proposal from the Darwin Tree of Life project [version 1; peer review: 2 approved with reservations].** *Wellcome Open Res.* 2022; **7**: 187.
[Publisher Full Text](#)
- Lees DC, Sims I, Natural History Museum Genome Acquisition Lab, *et al.*: **The genome sequence of the Wavy-barred Sable moth, *Pyrausta nigrata* (Scopoli, 1763) [version 1; peer review: awaiting peer review].** *Wellcome Open Res.* 2024; **9**: 278.
[Publisher Full Text](#)
- Léger T, Mally R, Neinhuis C, *et al.*: **Refining the phylogeny of Crambidae with complete sampling of subfamilies (Lepidoptera, Pyraloidea).** *Zool Scr.* 2021; **50**(1):84–99.
[Publisher Full Text](#)
- Li H: **Minimap2: pairwise alignment for nucleotide sequences.** *Bioinformatics.* 2018; **34**(18): 3094–3100.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Manni M, Berkeley MR, Seppey M, *et al.*: **BUSCO update: novel and streamlined workflows along with broader and deeper phylogenetic coverage for scoring of eukaryotic, prokaryotic, and viral genomes.** *Mol Biol Evol.* 2021; **38**(10): 4647–4654.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Merkel D: **Docker: lightweight Linux containers for consistent development and deployment.** *Linux J.* 2014; **2014**(239): 2, [Accessed 2 April 2024].
[Reference Source](#)
- Oatley G, Denton A, Howard C: **Sanger Tree of Life HMW DNA extraction: automated MagAttract v.2.** *protocols.io.* 2023.
[Publisher Full Text](#)
- Parsons M, Clancy S: **A guide to the pyralid and crambid moths of Britain and Ireland.** Helston, Cornwall: Atropos Publishing, 2023.
- Pereira L, Sivell O, Sivess L, *et al.*: **DTOL Taxon-specific standard operating procedure for the terrestrial and freshwater arthropods working group.** 2022.
[Publisher Full Text](#)
- Pointon DL, Eagles W, Sims Y, *et al.*: **sanger-tol/treeval v1.0.0 – Ancient Atlantis.** 2023.
[Publisher Full Text](#)
- Quinlan AR, Hall IM: **BEDTools: a flexible suite of utilities for comparing genomic features.** *Bioinformatics.* 2010; **26**(6): 841–842.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Ranallo-Benavidez TR, Jaron KS, Schatz MC: **GenomeScope 2.0 and Smudgeplot for reference-free profiling of polyploid genomes.** *Nat Commun.* 2020; **11**(1): 1432.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Rao SSP, Huntley MH, Durand NC, *et al.*: **A 3D map of the human genome at kilobase resolution reveals principles of chromatin looping.** *Cell.* 2014; **159**(7): 1665–1680.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Rhie A, McCarthy SA, Fedrigo O, *et al.*: **Towards complete and error-free genome assemblies of all vertebrate species.** *Nature.* 2021; **592**(7856): 737–746.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Rhie A, Walenz BP, Koren S, *et al.*: **Merqury: reference-free quality, completeness, and phasing assessment for genome assemblies.** *Genome Biol.* 2020; **21**(1): 245.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Sterling P, Parsons M, Lewington R: **Field guide to the micro-moths of Great Britain and Ireland, Second Edition.** London: Bloomsbury Publishing, 2023.
[Reference Source](#)
- Strickland M, Cornwell C, Howard C: **Sanger Tree of Life fragmented DNA clean up: manual SPRI.** *protocols.io.* 2023.
[Publisher Full Text](#)
- Twyford AD, Beasley J, Barnes I, *et al.*: **A DNA barcoding framework for taxonomic verification in the Darwin Tree of Life project [version 1; peer review: 2 approved].** *Wellcome Open Res.* 2024; **9**: 339.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Uliano-Silva M, Ferreira JGRN, Krasheninnikova K, *et al.*: **MitoHiFi: a python pipeline for mitochondrial genome assembly from PacBio high fidelity reads.** *BMC Bioinformatics.* 2023; **24**(1): 288.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
- Vasimuddin M, Misra S, Li H, *et al.*: **Efficient architecture-aware acceleration of BWA-MEM for multicore systems.** In: *2019 IEEE International Parallel and Distributed Processing Symposium (IPDPS).* IEEE, 2019; 314–324.
[Publisher Full Text](#)
- Wu Y, Liu X, Zhang X, *et al.*: **Characterization of four mitochondrial genomes of Crambidae (Lepidoptera, Pyraloidea) and phylogenetic implications.** *Arch Insect Biochem Physiol.* 2023; **112**(1): e21914.
[PubMed Abstract](#) | [Publisher Full Text](#)
- Zhou C, McCarthy SA, Durbin R: **YaHS: yet another Hi-C scaffolding tool.** *Bioinformatics.* 2023; **39**(1): btac808.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)