DATA NOTE

The genome sequence of the yellow-tail moth, *Euproctis similis* (Fuessly, 1775) [version 1; peer review: 1 approved, 2 approved with reservations]

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Abstract

We present a genome assembly from an individual male *Euproctis similis* (the yellow-tail; Arthropoda; Insecta; Lepidoptera; Lymantriidae). The genome sequence is 508 megabases in span. The majority of the assembly is scaffolded into 22 chromosomal pseudomolecules, with the Z sex chromosome assembled.

Keywords

Euproctis similis, yellow-tail, genome sequence, chromosomal

This article is included in the Tree of Life gateway.

Open Peer Review

Invited Reviewers

| Invited Reviewers | 1 | 2 | 3 |
|-------------------|---|---|---|
| version 1 | report | report | ✔ |
| 13 Sep 2021 | | | |

1. Steven M. Van Belleghem, University of Puerto Rico-Rio Piedras, San Juan, Puerto Rico
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Any reports and responses or comments on the article can be found at the end of the article.
Species taxonomy
Eukaryota; Metazoa; Ecdysozoa; Arthropoda; Hexapoda; Insecta; Pterygota; Neoptera; Endopterygota; Lepidoptera; Glossata; Ditrysia; Noctuoidea; Erebidae; Lymantriinae; Euproctis; Euproctis similis Fuessly 1775 (NCBI:txid987935).

Introduction
Larvae of Euproctis similis (yellow-tail) have long hairs that can cause skin irritation in humans, although the effects are rarely as serious as those caused by larvae of the closely related Euproctis chrysorrhoea (brown-tail). The genome of E. similis was sequenced as part of the Darwin Tree of Life Project, a collaborative effort to sequence all of the named eukaryotic species in the Atlantic Archipelago of Britain and Ireland. Here we present a chromosomally complete genome sequence for E. similis, based on one male specimen from Wytham Woods, Oxfordshire, UK.

Genome sequence report
The genome was sequenced from a single male E. similis collected from Wytham Woods, Oxfordshire, UK (latitude 51.772, longitude -1.338). A total of 70-fold coverage in Pacific Biosciences single-molecule long reads (N50 17 kb) and 78-fold coverage in 10X Genomics read clouds were generated. Primary assembly contigs were scaffolded with chromosome conformation Hi-C data. Manual assembly curation corrected 40 missing/missions and removed 3 haplotype duplications, reducing the assembly length by 0.10% and the scaffold number by 42.00%, and increasing the scaffold N50 by 14.24%. The final assembly has a total length of 508 Mb in 30 sequence scaffolds with a scaffold N50 of 24 Mb (Table 1). The majority, >99.9%, of assembly sequence was assigned to 22 chromosomal-level scaffolds, representing 21 autosomes (numbered by sequence length), and the Z sex chromosome (Figure 1–Figure 4; Table 2). The assembly has a BUSCO (Simão et al., 2015) v5.1.2 completeness of 98.6% using the lepidoptera_odb10 reference set. While not fully phased, the assembly deposited is of one haplotype. Contigs corresponding to the second haplotype have also been deposited.

Methods
A single male E. similis was collected from Wytham Woods, Oxfordshire, UK (latitude 51.772, longitude -1.338) by Douglas Boyes, University of Oxford, using a light trap. The specimens were snap-frozen in dry ice using a CoolRack before transferring to the Wellcome Sanger Institute (WSI).

DNA was extracted at the Tree of Life laboratory, WSI. The ilEupSimi1 sample was weighed and dissected on dry ice with tissue set aside for RNA extraction and Hi-C sequencing. Thorax/abdomen tissue was cryogenically disrupted to a fine powder using a Covaris cryoPREP Automated Dry Pulveriser, receiving multiple impacts. Fragment size analysis of 0.01-0.5 ng of DNA was then performed using an Agilent FemtoPulse. High molecular weight (HMW) DNA was extracted using the Qiagen MagAttract HMW DNA extraction kit.

Low molecular weight DNA was removed from a 200-ng aliquot of extracted DNA using 0.8X AMPure XP purification kit prior to 10X Chromium sequencing; a minimum of 50 ng DNA was submitted for 10X sequencing. HMW DNA was sheared into an average fragment size between 12-20 kb in a Megaruptor 3 system with speed setting 30. Sheared DNA was purified by solid-phase reversible immobilisation using AMPure PB beads with a 1.8X ratio of beads to sample to remove the shorter fragments and concentrate the DNA sample. The concentration of the sheared and purified DNA was assessed using a Nanodrop spectrophotometer and Qubit Fluorometer and Qubit dsDNA High Sensitivity Assay kit. Fragment size distribution was evaluated by running the sample on the FemtoPulse system.
RNA was extracted from thorax/abdomen tissue in the Tree of Life Laboratory at the WSI using TRIzol (Invitrogen), according to the manufacturer’s instructions. RNA was then eluted in 50 μl RNAse-free water and its concentration assessed using a Nanodrop spectrophotometer and Qubit Fluorometer using the Qubit RNA Broad-Range (BR) Assay kit. Analysis of the integrity of the RNA was done using Agilent RNA 6000 Pico Kit and Eukaryotic Total RNA assay.

Pacific Biosciences HiFi circular consensus and 10X Genomics Chromium read cloud sequencing libraries were constructed according to the manufacturers’ instructions. Sequencing was performed by the Scientific Operations core at the Wellcome Sanger Institute on Pacific Biosciences SEQUEL II (HiFi), Illumina HiSeq X (10X) and Illumina HiSeq 4000 (RNA-Seq) instruments. Hi-C data were generated from head tissue using the Qiagen EpiTect Hi-C kit and sequenced on HiSeq X.

Figure 1. Genome assembly of *Euproctis similis*, iEupSimi1.1: metrics. The BlobToolKit Snailplot shows N50 metrics and BUSCO gene completeness. An interactive version of this figure is available at https://blobtoolkit.genomehubs.org/view/iEupSimi1.1/dataset/CAJHUZ01/snail.
Assembly was carried out with HiCanu (Nurk et al., 2020); haplotypic duplication was identified and removed with purge_dups (Guan et al., 2020). The assembly was polished with the 10X Genomics Illumina data by aligning to the assembly with longranger align, calling variants with freebayes (Garrison & Marth, 2012). One round of the Illumina polishing was applied. Scaffolding with Hi-C data (Rao et al., 2014) was carried out with SALSA2 (Ghurye et al., 2019). The assembly was checked for contamination and corrected using the gEVAL system (Chow et al., 2016) as described previously (Howe et al., 2021). Manual curation was performed using gEVAL, HiGlass (Kerpedjiev et al., 2018) and Pretext. The mitochondrial genome was assembled using MitoHiFi (Uliano-Silva et al., 2021). The genome was analysed and BUSCO scores generated.

Figure 2. Genome assembly of *Euproctis similis*, iEupSimi1.1: GC coverage. BlobToolKit GC-coverage plot. Scaffolds are coloured by phylum. Circles are sized in proportion to scaffold length. Histograms show the distribution of scaffold length sum along each axis. An interactive version of this figure is available at https://blobtoolkit.genomehubs.org/view/iEupSimi1.1/dataset/CAJHUZ01/blob.
within the BlobToolKit environment (Challis et al., 2020). Table 3 contains a list of all software tool versions used, where appropriate.

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. 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. Each transfer of samples is further undertaken according to a Research Collaboration

**Figure 3.** Genome assembly of *Euproctis similis*, iEupSimi1.1: cumulative sequence. BlobToolKit cumulative sequence plot. The grey line shows cumulative length for all chromosomes. Coloured lines show cumulative lengths of chromosomes assigned to each phylum using the buscogenes taxrule. An interactive version of this figure is available at https://blobtoolkit.genomehubs.org/view/ilEupSimi1.1/dataset/CAJHUZ01/cumulative.
Table 2. Chromosomal pseudomolecules in the genome assembly of *Euproctis similis*, ilEupSimi1.1.

| INSDC accession | Chromosome | Size (Mb) | GC%  |
|-----------------|------------|-----------|------|
| LR990103.1      | 1          | 29.63     | 36.8 |
| LR990104.1      | 2          | 28.43     | 36.2 |
| LR990105.1      | 3          | 26.72     | 36.1 |
| LR990106.1      | 4          | 26.40     | 36.3 |
| LR990108.1      | 5          | 26.05     | 36.2 |
| LR990109.1      | 6          | 26.03     | 36.3 |
| LR990110.1      | 7          | 25.95     | 36.6 |
| LR990111.1      | 8          | 25.82     | 36.5 |
| LR990112.1      | 9          | 24.45     | 36.5 |
| LR990113.1      | 10         | 23.51     | 36.8 |
| LR990114.1      | 11         | 21.70     | 36.5 |
| LR990115.1      | 12         | 21.66     | 36.4 |
| LR990116.1      | 13         | 21.63     | 36.6 |
| LR990117.1      | 14         | 21.40     | 36.8 |
| LR990118.1      | 15         | 21.08     | 36.3 |
| LR990119.1      | 16         | 19.59     | 37   |
| LR990120.1      | 17         | 18.87     | 36.6 |
| LR990121.1      | 18         | 18.67     | 37.2 |
| LR990122.1      | 19         | 18.51     | 37   |
| LR990123.1      | 20         | 17.96     | 36.5 |
| LR990124.1      | 21         | 16.98     | 36.8 |
| LR990125.1      | MT         | 0.02      | 19.4 |
| -               | Unplaced   | 0.24      | 39.9 |

Figure 4. Genome assembly of *Euproctis similis*, ilEupSimi1.1: Hi-C contact map. Hi-C contact map of the ilEupSimi1.1 assembly, visualised in HiGlass.
### Table 3. Software tools used.

| Software tool     | Version | Source                                                                 |
|-------------------|---------|------------------------------------------------------------------------|
| HiCanu            | 2.1     | Nurk et al., 2020                                                     |
| purge_dups        | 1.2.3   | Guan et al., 2020                                                     |
| SALSA2            | 2.2     | Ghurye et al., 2019                                                   |
| longranger align  | 2.2.2   | https://support.10xgenomics.com/genome-exome/software/pipelines/latest/other-pipelines |
| freebayes         | 1.3-1-7-gaa2ace8 | Garrison & Marth, 2012                                           |
| MitoHiFi          | 1       | Uliano-Silva et al., 2021                                             |
| gEVAL             | N/A     | Chow et al., 2016                                                     |
| HiGlass           | 1.11.6  | Kerpedjiev et al., 2018                                               |
| PretextView       | 0.1.x   | https://github.com/wtsi-hpag/PretextView                                |
| BlobToolKit       | 2.6.2   | Challis et al., 2020                                                  |

Agreement or Material Transfer Agreement entered into by the Darwin Tree of Life Partner, Genome Research Limited (operating as the WSI), and in some circumstances other Darwin Tree of Life collaborators.

**Data availability**

European Nucleotide Archive: Euproctis similis (yellow-tail). Accession number PRJEB42127: https://identifiers.org/ena.embl: PRJEB42127

**Acknowledgements**

Members of the University of Oxford and Wytham Woods Genome Acquisition Lab are listed here: https://doi.org/10.5281/zenodo.4789929.

Members of the Darwin Tree of Life Barcoding collective are listed here: https://doi.org/10.5281/zenodo.4893704.

Members of the Wellcome Sanger Institute Tree of Life programme collective are listed here: https://doi.org/10.5281/zenodo.5377053.

Members of Wellcome Sanger Institute Scientific Operations: DNA Pipelines collective are listed here: https://doi.org/10.5281/zenodo.4790456.

Members of the Tree of Life Core Informatics collective are listed here: https://doi.org/10.5281/zenodo.5013542.

Members of the Darwin Tree of Life Consortium are listed here: https://doi.org/10.5281/zenodo.4783559.

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Uliano-Silva M, Nunes JGF, Krasheninnikova K, et al.: marcelauliano/MitoHiFi: mitohifi_v2.0. 2021. Publisher Full Text
Open Peer Review

Current Peer Review Status: ? ? ✓

Version 1

Reviewer Report 24 September 2021

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Thomas Blankers

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This data note presents the genome assembly of Euproctis similis as part of the Darwin Tree of Life Project. The information about the molecular lab methods is sufficient and clear. The appropriate, protocols have been used, following recommendations of the manufacturers and sequencing platforms. The quality of the genome is high, owing to the use of complementary sequencing technologies that allow for contiguous assemblies.

I only have three small suggestions for additional information, although I also see that none of these are commonly supplied in the notes coming from the Darwin Tree of Life project. First, as a biologist, I would be interested in knowing a little bit more about the organism. For example that it is a night-active moth, wide-spread across the Eurasian continent, that they're active from August to June and that they are associated with both urban and non-urban habitats and with several host plants. Second, in the presentation of the methods there are no details about the bioinformatic analyses beyond the programs that were used. It would be good to specify any deviation from default settings or even to have a brief summary of the commands used to perform the analyses. This could be done in a separate file archived along with the note or in a Table, possibly integrated in Table 3. Third, the data presentation can benefit from brief expansion of the results. The interactive figures are nice, because some explanation of what is shown can also be found at the corresponding blobtoolkit repository. However, there is no text accompanying these figures beyond a single sentence referencing the number of scaffolds and citing figures 1 through 4. Some expansion of the genome assembly statistics seems desirable. And figure 4 could use a legend as well as axis labels with the chromosome numbers. Again, I do see that other examples of notes on genomes coming from this project also do not necessarily contain these additional pieces of information, so I guess it is up to the authors to decide whether that continuity matters more or whether the details are simply not necessary.

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?
Partly

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

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

**Reviewer Expertise:** Evolutionary genomics, currently working on crickets, moths, and C. elegans

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 20 September 2021
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The manuscript by Boyes and Holland describes an effort to assemble both the nuclear and mitochondrial genome of a male yellow-tailed moth. The procedure includes multiple sequencing techniques allowing for both primary assembly of contigs from long-read libraries (PacBio), polishing with linked reads (10X) and scaffolding with chromosome interaction information (HiC). The highly contiguous (seven very short scaffolds, that only sum up to 0.24 Mb in total, that are not linked to chromosomes) final assembly contains 21 autosomes, the Z-chromosome and the mitochondrial genome. This will be a valuable resource for both population genetic analyses within the species/genus and comparative genomics studies in insects in general and in Lepidoptera in particular. The study sets high standards for the methodology of genome assembly by using and efficiently combining state-of-the-art sequencing technologies and novel bioinformatic pipelines.

**Minor comments:**

Abstract:
1. Maybe omit ‘majority of the’.

2. Information about that the mitochondrial genome is also assembled should be included.

3. Perhaps mention briefly that the number of assembled chromosomes is lower than many previously sequenced lepidopterans – perhaps indicating fusions in the *Euproctis* lineage?

**Introduction:**
1. A brief description of the abundance and distribution range of this species in the AABI area and globally would be very informative.

2. Perhaps it could be of interest to characterize species specific and general biological questions that can be addressed using this genomic resource.

**Genome sequence report + Methods:**
1. The first sentence in these two sections is redundant. Perhaps it is sufficient to describe the sampling location, collector, sample ID and date of sampling (missing) in the methods section and omit the redundant details in the GSR section.

**Genome sequence report:**
1. Omit ‘The majority,”?

2. Add information about that the mitochondrial genome was assembled (including assessment of completeness, e. g. exact length of the scaffold)?

3. The finding that scaffolds are anchored to only 21 autosomes is a bit surprising given that most lepidopterans sequenced so far have 31 chromosome pairs. This could be another example of chromosome rearrangements (here potentially fusions, similar to what has been observed in e.g. *Heliconius melpomene*)? This might be worth mentioning briefly.

4. Were RNAseq sequencing reads used in the downstream analysis?

5. Table 1. Unclear if the “Number of contigs” corresponds to the number of contigs before or after the manual correction of the Hi-C map?

**Methods:**
1. ‘specimen was’ instead of ‘specimens were’.

2. Second paragraph. The sample ID can be given in the first part of the section where sample location, collector, date of sampling etc. is stated.

3. 4th line, left column under Figure 1. ‘its concentration RNA assessed’ should perhaps be ‘the RNA concentration assessed’.

4. Information about RNAseq library preparation method, sequencing technique applied and data availability of RNAseq reads is missing.

**Reflection:**
1. An annotation with the RNAseq data generated here in combination with previously available lepidopteran gene models would add useful information for downstream analyses using this resource.

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

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

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

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

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

**Reviewer Expertise:** Comparative genomics, population genetics, molecular evolution.

We confirm that we have read this submission and believe that we have an appropriate level of expertise to confirm that it is of an acceptable scientific standard, however we have significant reservations, as outlined above.

Reviewer Report 13 September 2021

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Steven M. Van Belleghem

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Douglas Boyes and Peter Holland report a high-quality genome assembly for the yellow-tail moth from the UK. They do this using PacBio and 10X sequencing and scaffolded the initially obtained contigs using Hi-C data. The data and assembly seem of high quality and the methods used are rigorous. The assembly contains 30 scaffolds assigned to 22 chromosomes and has a 98.6 BUSCO completeness. I believe this chromosome level assembly will be a valuable tool for future studies.

**Abstract:**
- I suggest using a more precise alternative to the word ‘majority’.

**Introduction:**
- Is it possible to give more precise information on the geographic distribution of this
Results:
- I do not see any results on the RNA-seq data. Was a transcriptome assembly performed? I also do not see accession numbers to these data in Table 1.
- An assessment of the repetitiveness (TE content) of the genome could be useful.

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

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

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

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

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Genomics, functional genomics, insects

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, however I have significant reservations, as outlined above.