The genome sequence of the dun-bar pinion, *Cosmia trapezina* (Linnaeus, 1758) [version 1; peer review: 2 approved]

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Abstract
We present a genome assembly from an individual male *Cosmia trapezina* (dun-bar pinion; Arthropoda; Insecta; Lepidoptera; Noctuidae). The genome sequence is 825 megabases in span. The majority of the assembly (99.87%) is scaffolded into 32 chromosomal pseudomolecules with the Z chromosome assembled. The complete mitochondrial genome was also assembled and is 15.4 kilobases in length.

Keywords
Cosmia trapezina, dun-bar pinion, genome sequence, chromosomal, Arthropoda

This article is included in the Tree of Life gateway.
Species taxonomy
Eukaryota; Metazoa; Ecdysozoa; Arthropoda; Hexapoda; Insecta; Pterygota; Neoptera; Endopterygota; Lepidoptera; Glossata; Ditrysia; Noctuoidea; Noctuidae; Ipimorphinae; Cosmia; Cosmia trapezina (Linneaus, 1758) (NCBI:txid116126).

Background
Cosmia trapezina, the dun-bar, is a medium-sized moth in the large family Noctuidae. It is found throughout most of the Palaearctic and in Britain is considered a common species. The forewings of C. trapezina are variable in colour, though the broad sharply-angled band running laterally, the ‘dun bar’, is characteristic, and this is an easy species to recognise.

To look at, the dun-bar is an unremarkable moth, but it has an unsavoury reputation. Lepidopterists learning to collect and rear caterpillars are taught to recognise dun-bar larvae and segregate them from other species and ideally rear them individually. Bucking the vegetarianism of most moths, the dun-bar has a carnivorous bent and while it will survive and grow on a diet of tree leaves it will eat the larvae of other moth species, notably Operophtera brumata (the Winter Moth), and will also cannibalise its own species. Newman in 1869 described C. trapezina larvae chasing prey and seizing them behind the head “with savage eagerness” (Newman, 1869). A few other species share this tendency, notably its North American relative C. calami (Donald Lafontaine & Troubridge, 2003) and there is a clade of Hawaiian pug moths, Eupithecia sp., that are obligate predators (Montgomery, 1983). Eating your competitors has been termed intraguild predation (Polis, 1981) and there is speculation that the dun-bar’s proclivities may affect insect community structure in European woodlands (Turcani & Patočka, 2011).

Genome sequence report
The genome was sequenced from a single male C. trapezina collected near Ant Hills, Wytham, UK (Figure 1). A total of 43-fold coverage in Pacific Biosciences single-molecule HiFi long reads and 42-fold coverage in 10X Genomics read clouds were generated. Primary assembly contigs were scaffolded with chromosome conformation Hi-C data. Manual assembly curation corrected 93 missing/misjoins and removed 19 haplotypic duplications, reducing the assembly size by 0.78% and the scaffold number by 51.56%, and increasing the scaffold N50 by 7.36%.

The final assembly has a total length of 825 Mb in 62 sequence scaffolds with a scaffold N50 of 28.02 Mb (Table 1). The majority, 99.87%, of the assembly sequence was assigned to 32 chromosomal-level scaffolds, representing 31 autosomes (numbered by sequence length) and the Z sex chromosome (Figure 2–Figure 5; Table 2).

The assembly has a BUSCO v5.2.2 (Manni et al., 2021) completeness of 98.8% (single 97.9%, duplicated 0.8%) using the lepidoptera_odb10 reference set (n=954). While not fully phased, the assembly deposited is of one haplotype. Contigs corresponding to the second haplotype have also been deposited.

Methods
Sample acquisition and nucleic acid extraction
A single male C. trapezina specimen (iCosTrap1) was collected using a light trap near Ant Hills, Wytham, UK (latitude 51.764, longitude -1.327) by Douglas Boyes (University of Oxford). The specimen was identified by Douglas Boyes and snap-frozen on dry ice.

DNA was extracted at the Tree of Life laboratory, Wellcome Sanger Institute. The iCosTrap1 sample was weighed and dissected on dry ice with tissue set aside for Hi-C sequencing. Thorax and abdomen tissue was disrupted using a Nippi Powermasher fitted with a BioMasher pestle. Fragment size analysis of 0.01–0.5 ng of DNA was then performed using an Agilent FemtoPulse. High molecular weight (HMW) DNA was

Figure 1. Image of the Cosmia trapezina specimen taken prior to preservation and processing.
Table 1. Genome data for *Cosmia trapezina*, ilCosTrap1.2.

| **Project accession data**                                      |                         |
|----------------------------------------------------------------|-------------------------|
| Assembly identifier                                           | ilCosTrap1.2            |
| Species                                                       | *Cosmia trapezina*      |
| Specimen                                                      | ilCosTrap1 (genome assembly; Hi-C) |
| NCBI taxonomy ID                                              | 116126                  |
| BioProject                                                    | PRJEB42119              |
| BioSample ID                                                  | SAMEA7519851            |
| Isolate information                                           | Male, abdomen/thorax tissue (genome assembly); head tissue (Hi-C) |
| **Raw data accessions**                                       |                         |
| PacificBiosciences SEQUEL II                                 | ERR6558181-ERR6558183   |
| 10X Genomics Illumina                                         | ERR6002608-ERR6002611   |
| Hi-C Illumina                                                 | ERR6002612-ERR6002614   |

**Genome assembly**

| Assembly accession                                           | GCA_905163495.2         |
| Accession of alternate haplotype                             | GCA_905163595.1         |
| Span (Mb)                                                     | 825                     |
| Number of contigs                                            | 266                     |
| Contig N50 length (Mb)                                       | 7.6                     |
| Number of scaffolds                                           | 62                      |
| Scaffold N50 length (Mb)                                     | 28.02                   |
| Longest scaffold (Mb)                                        | 35.87                   |
| BUSCO* genome score                                          | C:98.8%;[S:97.9%,D:0.8%]; F:0.2%; M:1.1%;n:5286  |

*BUSCO scores based on the lepidoptera_odb10 BUSCO set using v5.2.2. C= complete [S= single copy, D=duplicated], F=fragmented, M=missing, n=number of orthologues in comparison. A full set of BUSCO scores is available at https://blobtoolkit.genomehubs.org/view/ilCosTrap1.2/dataset/CAJHZR02/filters.*

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.

**Sequencing**

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) and Illumina HiSeq (10X) instruments. Hi-C data were generated in the Tree of Life laboratory from remaining head tissue of ilCosTrap1 using the Arima v1 kit and sequenced on an Illumina HiSeq (10X) instrument.

**Genome assembly**

Assembly was carried out with HiCanu (Nurk *et al.*, 2020); haplotypic duplication was identified and removed with purge_dups (Guan *et al.*, 2020). One round of polishing
Figure 2. Genome assembly of Cosmia trapezina, ilCosTrap1.2: metrics. The BlobToolKit Snailplot shows N50 metrics and BUSCO gene completeness. The main plot is divided into 1,000 size-ordered bins around the circumference with each bin representing 0.1% of the 825,187,588 bp assembly. The distribution of chromosome lengths is shown in dark grey with the plot radius scaled to the longest chromosome present in the assembly (37,523,413 bp, shown in red). Orange and pale-orange arcs show the N50 and N90 chromosome lengths (28,022,459 and 19,015,461 bp), respectively. The pale grey spiral shows the cumulative chromosome count on a log scale with white scale lines showing successive orders of magnitude. The blue and pale-blue area around the outside of the plot shows the distribution of GC, AT and N percentages in the same bins as the inner plot. A summary of complete, fragmented, duplicated and missing BUSCO genes in the lepidoptera_odb10 set is shown in the top right. An interactive version of this figure is available at https://blobtoolkit.genomehubs.org/view/ilCosTrap1.2/dataset/CAJHZR02/snail#Filters.

was performed by aligning 10X Genomics read data to the assembly with longranger align, calling variants with freebayes (Garrison & Marth, 2012). The assembly was then scaffolded with Hi-C data (Rao et al., 2014) using SALSAP2.
Figure 3. Genome assembly of *Cosmia trapezina*, iICosTrap1.2: 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/iICosTrap1.2/dataset/CAJHZR02/blob#Filter.

(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 (Howe et al., 2021) was performed using gEVAL, HiGlass (Kerpedjiev et al., 2018) and PreText. The mitochondrial genome was assembled using MitoHiFi (Uliano-Silva et al., 2021).
**Figure 4.** Genome assembly of *Cosmia trapezina*, ilCosTrap1.2: cumulative sequence. 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/ilCosTrap1.2/dataset/CAJHZR02/cumulative#Filters.

which performs annotation using MitoFinder (Allio *et al.*, 2020). The genome was analysed and BUSCO scores generated within the BlobToolKit environment (Challis *et al.*, 2020). Table 3 contains a list of all software tool versions used, where appropriate.

**Ethics/compliance issues**
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.
Figure 5. Genome assembly of *Cosmia trapezina*, ilCosTrap1.2: Hi-C contact map. Hi-C contact map of the ilCosTrap1.2 assembly, visualised in HiGlass. Chromosomes are arranged in size order from left to right and top to bottom. The interactive Hi-C map can be viewed at https://genome-note-higlass.tol.sanger.ac.uk/l/?d=DMmRE_dnRj2xSLEIhv538A.

Table 2. Chromosomal pseudomolecules in the genome assembly of *Cosmia trapezina*, ilCosTrap1.2.

| INSDC accession | Chromosome | Size (Mb) | GC% |
|-----------------|------------|-----------|-----|
| LR991020.1      | 1          | 35.87     | 37.7|
| LR991021.1      | 2          | 33.09     | 37.9|
| LR991022.1      | 3          | 31.67     | 38  |
| LR991023.1      | 4          | 30.74     | 38  |
| LR991024.1      | 5          | 30.25     | 37.7|
| LR991025.1      | 6          | 30.06     | 37.5|
| LR991026.1      | 7          | 29.98     | 37.8|
| LR991027.1      | 8          | 29.62     | 37.9|
| LR991028.1      | 9          | 29.4      | 38  |
| LR991029.1      | 10         | 28.86     | 37.6|
| LR991030.1      | 11         | 28.65     | 37.9|
| LR991031.1      | 12         | 28.12     | 37.7|
| LR991032.1      | 13         | 28.02     | 37.8|
| LR991033.1      | 14         | 27.94     | 37.6|
| LR991034.1      | 15         | 27.85     | 37.9|
| LR991035.1      | 16         | 27.24     | 38.1|
| LR991036.1      | 17         | 27        | 38  |
| LR991037.1      | 18         | 26.84     | 37.8|
| LR991038.1      | 19         | 26.61     | 38  |
| LR991039.1      | 20         | 26.03     | 38  |
| LR991040.1      | 21         | 24.38     | 38.1|
| LR991041.1      | 22         | 22.86     | 37.8|
| LR991042.1      | 23         | 22.81     | 37.9|
| LR991043.1      | 24         | 22.08     | 37.9|
| LR991044.1      | 25         | 19.21     | 38.3|
| LR991045.1      | 26         | 19.02     | 37.8|
| LR991046.1      | 27         | 17.32     | 38.1|
| LR991047.1      | 28         | 16.75     | 38.8|
| LR991048.1      | 29         | 15.86     | 38.3|
| LR991049.1      | 30         | 14.8      | 38.2|
| LR991050.1      | 31         | 7.69      | 38.4|
| LR991019.1      | Z          | 37.52     | 37.8|
| LR991051.2      | MT         | 0.02      | 19.9|
| -               | Unplaced   | 1.03      | 39.7|
### 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/advanced/other-pipelines |
| freebayes        | 1.3.1-17-gaa2ace8 | Garrison & Marth, 2012                                              |
| MitoHiFi         | 1.0     | Uliano-Silva et al., 2021                                             |
| HiGlass          | 1.11.6  | Kerpedjiev et al., 2018                                               |
| PretextView       | 0.2.x   | https://github.com/wtsi-hpag/PretextView                             |
| BlobToolKit      | 3.0.5   | Challis et al., 2020                                                  |

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 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: Cosmia trapezina (dun-bar pinion). Accession number PRJEB42119; https://identifiers.org/ena.embl/PRJEB42119.

The genome sequence is released openly for reuse. The *C. trapezina* genome sequencing initiative is part of the Darwin Tree of Life (DToL) project. All raw sequence data and the assembly have been deposited in INSDC databases. The genome will be annotated and presented through the Ensembl pipeline at the European Bioinformatics Institute. Raw data and assembly accession identifiers are reported in Table 1.

**Author information**

Members of the University of Oxford and Wytham Woods Genome Acquisition Lab are listed here: https://doi.org/10.5281/zenodo.6418202.

Members of the Darwin Tree of Life Barcoding collective are listed here: https://doi.org/10.5281/zenodo.6418156.

Members of the Wellcome Sanger Institute Tree of Life programme are listed here: https://doi.org/10.5281/zenodo.6418327.

Members of Wellcome Sanger Institute Scientific Operations: DNA Pipelines collective are listed here: https://doi.org/10.5281/zenodo.5746904.

Members of the Tree of Life Core Informatics collective are listed here: https://doi.org/10.5281/zenodo.6125046.

Members of the Darwin Tree of Life Consortium are listed here: https://doi.org/10.5281/zenodo.6418363.

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Camille Meslin
INRAE, CNRS, IRD, UPEC, University of Paris, Paris, France

This is a well-written data note on the genome of the dun-bar pinion Cosmia trapezina and a great resource given the high quality of the genome.

My only interrogation about this genome is whether some kind of annotation was performed on it? It would be a great addition for people interested in comparative genomics.

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: Chemosensory receptors, comparative genomics, evolution

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 06 October 2022

https://doi.org/10.21956/wellcomeopenres.19863.r52447
Amali Thrimawithana
Plant and Food Research Institute of New Zealand Ltd, Auckland, New Zealand

Well written data note on the dun-bar genome. Protocols for generating the assembly seem appropriate and have constructed high quality genome for the species. All versions and assembly methods are followable - one query: was it wasn't clear which tool was used for one round of polishing (though I understand longranger was used for aligning).

Another was, was the 10x data only used for correction - were no further scaffolding/assembly with it trialled?

I was also curious to know if the heterozygosity level etc was examined with kmer analysis of sort?

Again, great resource to have for the species.

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: Genomics, transcriptomics, metagenomics in plant insect and fish

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.