

DATA NOTE

REVISED The genome sequence of the John Dory, Zeus faber Linnaeus, 1758 [version 2; peer review: 2 approved, 1 not

approved]

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Abstract

We present a genome assembly from an individual Zeus faber (the John Dory; Chordata; Actinopteri; Zeiformes; Zeidae). The genome sequence is 804.7 megabases in span. Most of the assembly is scaffolded into 22 chromosomal pseudomolecules. The mitochondrial genome has also been assembled and is 16.72 kilobases in length.

Keywords

Zeus faber, John Dory, genome sequence, chromosomal, Zeiformes



article can be found at the end of the article.

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REVISED Amendments from Version 1

The Background section has been edited to be more concise and focused

Any further responses from the reviewers can be found at the end of the article

Species taxonomy

Eukaryota; Opisthokonta; Metazoa; Eumetazoa; Bilateria; Deuterostomia; Chordata; Craniata; Vertebrata; Gnathostomata; Teleostomi; Euteleostomi; Actinopterygii; Actinopteri; Neopterygii; Teleostei; Osteoglossocephalai; Clupeocephala; Euteleosteomorpha; Neoteleostei; Eurypterygia; Ctenosquamata; Acanthomorphata; Paracanthopterygii; Zeiogadaria; Zeariae; Zeiformes; Zeidae; Zeus; Zeus faber Linnaeus, 1758 (NCBI:txid64108).

Background

Zeus faber Linnaeus, 1758, known as John Dory or St Peter's fish, is a solitary, demersal marine fish with a laterally compressed, golden-brown body marked by a black spot on either side and long dorsal spines (Wheeler, 1978). It is widely distributed in the eastern Atlantic, Mediterranean, Pacific and Indian Oceans, and along the entire West African coast, occurring at depths of 0–200 m (Iwamoto, 2015; Maravelias *et al.*, 2007). It has recently been recorded for the first time in the Black Sea (Aydın & Karadurmuş, 2023). Its large, protrusible mouth and well-developed eyes enable it to prey on relatively large fish (Kim *et al.*, 2020; Stergiou & Fourtouni, 1991).

In the eastern Mediterranean, juveniles initially feed on zooplankton like mysids, then shift to small benthopelagic fishes as they grow, eventually preying on larger schooling pelagic species (Kim *et al.*, 2020; Stergiou & Fourtouni, 1991). In Korean coastal waters there is also varying diet composition with size and age (Kim *et al.*, 2020). Off the Portuguese coast, however, there is no prey switching from juvenile to adult life stages (Silva, 1999). *Z. faber* is considered an opportunistic feeder, switching prey depending on food availability and abundance which can vary seasonally and with life stage (Kim *et al.*, 2020).

Z. faber is known to make 'croaking' or 'barking' noises upon capture onboard (Radford et al., 2018). These vocalisations have since been documented in situ in Australia and were found to induce an escape response in conspecifics and heterospecifics such as the Australian Snapper (Pagurus auratus), suggesting they make sounds as a territorial display against competitors (Radford et al., 2018).

John Dory is commercially significant, valued for consumption, fish meal, and oil, and has a presence in the gamefish and aquarium trades (Iwamoto, 2015). It is a key species in mixed trawl fisheries in the British Isles and a common by-catch globally (Dunn, 2001; Iwamoto, 2015). The most recent stock

assessment in the British Isles occurred between 1994 and 1996, focusing on landings and biological data from the English Channel (Dunn, 2001). The English Channel appears to be a nursery ground, with seasonal peaks in landings correlating with recruitment during the third and fourth quarters, at approximately 23 cm TL (Dunn, 2001). Most landed individuals range from 23–29 cm TL, with a maximum observed TL of 59 cm.

The IUCN assessed the global conservation status of *Z. faber* as 'Data Deficient' in 2013 (Iwamoto, 2015), citing limited biological and historical data (Dunn, 2001), which contributes to uncertainties in stock status and fishing pressure.

Molecular investigation of this species has shown significant genetic differentiation (7.44%) between clades in the North Atlantic/Mediterranean region and Australasia, indicating potential speciation (Ward et al., 2008). The first genome of Z. faber was generated in 2016 for a study suggesting immunerelated genes play an important role in teleost evolution and speciation (Malmstrøm et al., 2016). This data note presents the second published genome of John Dory, collected and sequenced as part of the Darwin Tree of Life project (Blaxter et al., 2022). This dataset will be important for furthering our understanding of teleost pathology, immunology, evolution and phylogenetics (Malmstrøm et al., 2016; Ward et al., 2008).

Genome sequence report

The genome was sequenced from an individual *Zeus faber* (Figure 1) collected from Bigbury Bay, UK (50.27, -3.97). A total of 43-fold coverage in Pacific Biosciences single-molecule HiFi long reads was generated. Primary assembly contigs were scaffolded with chromosome conformation Hi-C data. Manual assembly curation corrected 9 missing joins or mis-joins, reducing the scaffold number by 1.04%.

The final assembly has a total length of 804.7 Mb in 190 sequence scaffolds with a scaffold N50 of 34.5 Mb (Table 1). The snail plot in Figure 2 provides a summary of the assembly statistics, while the distribution of assembly scaffolds on GC proportion and coverage is shown in Figure 3. The cumulative assembly plot in Figure 4 shows curves for subsets of scaffolds assigned to different phyla. Most (97.08%) of the assembly sequence was assigned to 22 chromosomal-level scaffolds. Chromosome-scale scaffolds confirmed by the Hi-C data are named in order of size (Figure 5; Table 2). While not fully phased, the assembly deposited is of one haplotype. Contigs corresponding to the second haplotype have also been deposited. The mitochondrial genome was also assembled and can be found as a contig within the multifasta file of the genome submission.

The estimated Quality Value (QV) of the final assembly is 52.6 with k-mer completeness of 99.98%, and the assembly has a BUSCO v5.3.2 completeness of 96.4% (single = 94.9%, duplicated = 1.4%), using the actinopterygii_odb10 reference set (n = 3.640).



Figure 1. Photograph of the *Zeus faber* (fZeuFab8) specimen used for genome sequencing.

Metadata for specimens, barcode results, spectra estimates, sequencing runs, contaminants and pre-curation assembly statistics are given at https://links.tol.sanger.ac.uk/species/64108.

Methods

Sample acquisition and nucleic acid extraction

A Zeus faber specimen (specimen ID MBA-211116-004A, ToLID fZeuFab8) was collected from Bigbury Bay, UK (latitude 50.27, longitude –3.97) on 2021-11-16 using an otter trawl deployed from the RV Sepia. The collectors were Patrick Adkins, Joanna Harley, Rachel Brittain, Kesella Scott-Somme (all Marine Biological Association) and identified by Rachel Brittain, and then preserved in liquid nitrogen. The fish died as part of a trawl attached to another project and was opportunistically taken and dissected by the DToL team who were also on board the Sepia that day.

Table 1. Genome data for Zeus faber, fZeuFab8.1.

Project accession data			
Assembly identifier	fZeuFab8.1		
Species	Zeus faber		
Specimen	fZeuFab8		
NCBI taxonomy ID	64108		
BioProject	PRJEB63619		
BioSample ID	SAMEA111562156		
Isolate information	fZeuFab8 (DNA, Hi-C and RNA sequencing)		
Assembly metrics*		Benchmark	
Consensus quality (QV)	52.6	≥ 50	
k-mer completeness	99.98%	≥ 95%	
BUSCO**	C:96.4%[S:94.9%,D:1.4%], F:1.4%,M:2.2%,n:3,640	<i>C</i> ≥ 95%	
Percentage of assembly mapped to chromosomes	97.08%	≥ 95%	
Sex chromosomes	None	localised homologous pairs	
Organelles	Mitochondrial genome: 16.72 kb	complete single alleles	
Raw data accessions			
PacificBiosciences SEQUEL II	ERR11641070, ERR11641069		
Hi-C Illumina	ERR11641144, ERR11641145		
PolyA RNA-Seq Illumina	ERR11641143		

Genome assembly		
Assembly accession	GCA_960531495.1	
Accession of alternate haplotype	GCA_960530785.1	
Span (Mb)	804.7	
Number of contigs	1,078	
Contig N50 length (Mb)	1.4	
Number of scaffolds	190	
Scaffold N50 length (Mb)	34.5	
Longest scaffold (Mb)	65.76	

^{*} Assembly metric benchmarks are adapted from column VGP-2020 of "Table 1: Proposed standards and metrics for defining genome assembly quality" from Rhie *et al.* (2021).

^{**} BUSCO scores based on the actinopterygii_odb10 BUSCO set using version 5.3.2. 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/fZeuFab8_1/dataset/fZeuFab8_1/busco.

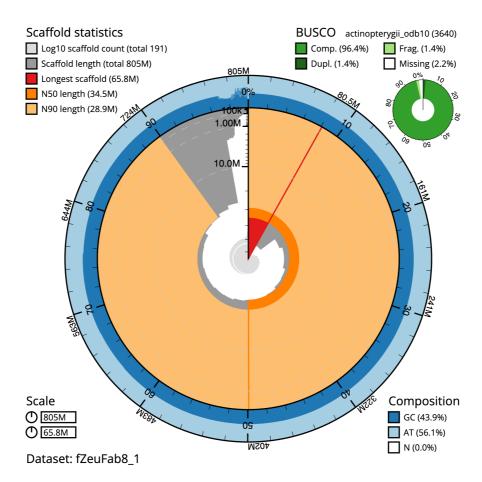


Figure 2. Genome assembly of *Zeus faber***, fZeuFab8.1: metrics.** The BlobToolKit snail plot shows N50 metrics and BUSCO gene completeness. The main plot is divided into 1,000 size-ordered bins around the circumference with each bin representing 0.1% of the 804,731,948 bp assembly. The distribution of scaffold lengths is shown in dark grey with the plot radius scaled to the longest scaffold present in the assembly (65,762,550 bp, shown in red). Orange and pale-orange arcs show the N50 and N90 scaffold lengths (34,476,449 and 28,869,016 bp), respectively. The pale grey spiral shows the cumulative scaffold 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 actinopterygii_odb10 set is shown in the top right. An interactive version of this figure is available at https://blobtoolkit.genomehubs.org/view/fZeuFab8_1/dataset/fZeuFab8_1/snail.

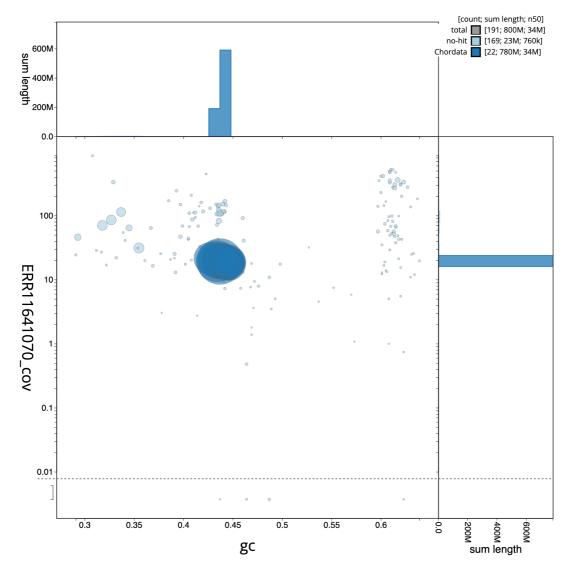


Figure 3. Genome assembly of *Zeus faber***, fZeuFab8.1: BlobToolKit GC-coverage plot.** Sequences are coloured by phylum. Circles are sized in proportion to sequence length. Histograms show the distribution of sequence length sum along each axis. An interactive version of this figure is available at https://blobtoolkit.genomehubs.org/view/fZeuFab8_1/dataset/fZeuFab8_1/blob.

The workflow for high molecular weight (HMW) DNA extraction at the Wellcome Sanger Institute (WSI) includes a sequence of core procedures: sample preparation; sample homogenisation, DNA extraction, fragmentation, and clean-up. In sample preparation, the fZeuFab8 sample was weighed and dissected on dry ice (Jay et al., 2023). Tissue 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 with speed setting 31 (Bates et al., 2023). Sheared DNA was purified by solid-phase reversible immobilisation (Strickland et al., 2023): in brief, the method employs a 1.8X ratio of AMPure PB beads to sample to eliminate

shorter fragments and concentrate the DNA. 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 tissue of fZeuFab8 in the Tree of Life Laboratory at the WSI using the RNA Extraction: Automated MagMaxTM *mir*Vana 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.

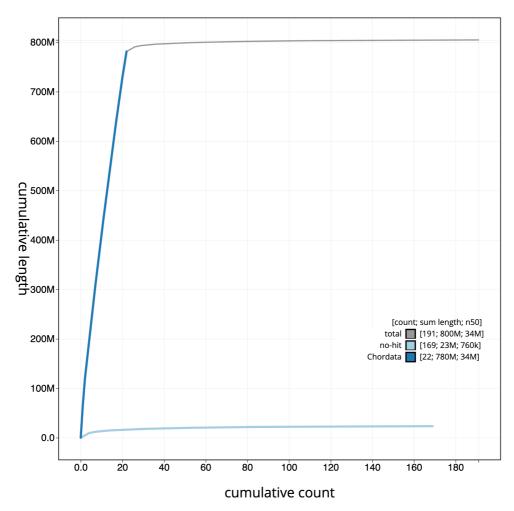


Figure 4. Genome assembly of *Zeus faber***, fZeuFab8.1: BlobToolKit cumulative sequence plot.** The grey line shows cumulative length for all sequences. Coloured lines show cumulative lengths of sequences assigned to each phylum using the buscogenes taxrule. An interactive version of this figure is available at https://blobtoolkit.genomehubs.org/view/fZeuFab8_1/dataset/fZeuFab8_1/cumulative.

Protocols developed by the WSI Tree of Life laboratory are publicly available on protocols.io (Denton *et al.*, 2023b).

Sequencing

Pacific Biosciences HiFi circular consensus DNA sequencing libraries were constructed according to the manufacturers' instructions. Poly(A) RNA-Seq libraries were constructed using the NEB Ultra II RNA Library Prep kit. DNA and RNA sequencing was performed by the Scientific Operations core at the WSI on Pacific Biosciences SEQUEL II (HiFi) and Illumina NovaSeq 6000 (RNA-Seq) instruments. Hi-C data were also generated from tissue of fZeuFab8 using the Arima2 kit and sequenced on the Illumina NovaSeq 6000, Illumina NovaSeq 6000 instrument.

Genome assembly, curation and evaluation

Assembly was carried out with Hifiasm (Cheng et al., 2021) and haplotypic duplication was identified and removed

with purge_dups (Guan et al., 2020). The assembly was then scaffolded with Hi-C data (Rao et al., 2014) using YaHS (Zhou et al., 2023). The assembly was checked for contamination and corrected as described previously (Howe et al., 2021). Manual curation was performed using HiGlass (Kerpedjiev et al., 2018) and PretextView (Harry, 2022). The mitochondrial genome was assembled using MitoHiFi (Uliano-Silva et al., 2023), which runs MitoFinder (Allio et al., 2020) or MITOS (Bernt et al., 2013) and uses these annotations to select the final mitochondrial contig and to ensure the general quality of the sequence.

A Hi-C map for the final assembly was produced using bwa-mem2 (Vasimuddin *et al.*, 2019) in the Cooler file format (Abdennur & Mirny, 2020). To assess the assembly metrics, the *k*-mer completeness and QV consensus quality values were calculated in Merqury (Rhie *et al.*, 2020). This work was done using Nextflow (Di Tommaso *et al.*, 2017) DSL2

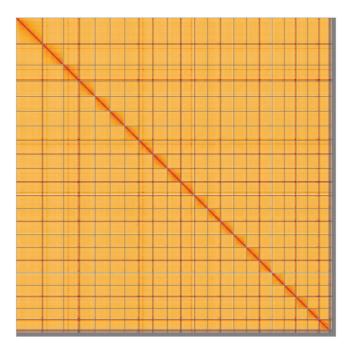


Figure 5. Genome assembly of *Zeus faber***, fZeuFab8.1: Hi-C contact map of the fZeuFab8.1 assembly, visualised using HiGlass.** Chromosomes are shown in order of size from left to right and top to bottom. An interactive version of this figure may be viewed at https://genome-note-higlass.tol.sanger.ac.uk/l/?d=ODHdK-fnRfy3tLmo69JGwQ.

Table 2. Chromosomal pseudomolecules in the genome assembly of *Zeus faber*, fZeuFab8.

INSDC accession	Chromosome	Length (Mb)	GC%
OY482845.1	1	65.76	43.5
OY482846.1	2	52.95	43.5
OY482847.1	3	40.01	43.5
OY482848.1	4	38.03	43.5
OY482849.1	5	37.98	44.0
OY482850.1	6	36.47	44.5
OY482851.1	7	36.1	44.0
OY482852.1	8	35.01	44.0
OY482853.1	9	34.68	44.5
OY482854.1	10	34.48	44.0
OY482855.1	11	33.96	43.5
OY482856.1	12	33.1	44.0
OY482857.1	13	32.89	44.5
OY482858.1	14	32.3	43.5
OY482859.1	15	32.12	44.5
OY482860.1	16	31.67	44.5

INSDC accession	Chromosome	Length (Mb)	GC%
OY482861.1	17	31.65	42.5
OY482862.1	18	31.31	44.5
OY482863.1	19	29.26	44.0
OY482864.1	20	28.87	44.5
OY482865.1	21	27.29	45.0
OY482866.1	22	25.41	44.0
OY482867.1	MT	0.02	42.5

pipelines "sanger-tol/readmapping" (Surana et al., 2023a) and "sanger-tol/genomenote" (Surana et al., 2023b). The genome was analysed within the BlobToolKit environment (Challis et al., 2020) and BUSCO scores (Manni et al., 2021; Simão et al., 2015) were calculated.

Table 3 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

Table 3. Software tools: versions and sources.

Software tool	Version	Source
BlobToolKit	4.1.7	https://github.com/blobtoolkit/blobtoolkit
BUSCO	5.3.2	https://gitlab.com/ezlab/busco
Hifiasm	0.19.5-r587	https://github.com/chhylp123/hifiasm
HiGlass	1.11.6	https://github.com/higlass/higlass
Merqury	MerquryFK	https://github.com/thegenemyers/MERQURY.FK
MitoHiFi	3	https://github.com/marcelauliano/MitoHiFi
PretextView	0.2	https://github.com/wtsi-hpag/PretextView
purge_dups	1.2.5	https://github.com/dfguan/purge_dups
sanger-tol/ genomenote	v1.0	https://github.com/sanger-tol/genomenote
sanger-tol/ readmapping	1.1.0	https://github.com/sanger-tol/readmapping/tree/1.1.0
YaHS	1.2a.2	https://github.com/c-zhou/yahs

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: Zeus faber (John dory). Accession number PRJEB63619; https://identifiers.org/ena.embl/PRJEB63619 (Wellcome Sanger Institute, 2023). The genome

sequence is released openly for reuse. The *Zeus faber* 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 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.

Author information

Members of the Marine Biological Association Genome Acquisition Lab are listed here: https://doi.org/10.5281/zenodo.8382513.

Members of the Darwin Tree of Life Barcoding collective are listed here: https://doi.org/10.5281/zenodo.4893703.

Members of the Wellcome Sanger Institute Tree of Life Management, Samples and Laboratory team are listed here: https://doi.org/10.5281/zenodo.10066175.

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Members of the Tree of Life Core Informatics collective are listed here: https://doi.org/10.5281/zenodo.5013541.

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Open Peer Review

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Version 2

Reviewer Report 25 October 2024

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Naiara Rodriguez-Ezpeleta 🗓

AZTI Basque Research and Technology Alliance, Bizkaia, Spain

The revised version is improved and ready for indexing.

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Population genomics, molecular evolution, high-throughput sequencing data analysis

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.

Version 1

Reviewer Report 08 August 2024

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Nastaran Mazloumi 🗓



University of Tasmania Institute for Marine and Antarctic Studies, Hobart, Tasmania, Australia

Title:

The chosen title "The genome sequence of the John Dory, Zeus faber Linnaeus, 1758" lacks clarity

and intention. You need to choose a better tile to define why you have done this study and what the genome sequestration is going to do!

Background:

In the background section, this paragraph below is disjointed and very hard to follow:

"John Dory is a commercially important species valued for human consumption, meal, and fish oil, as well as being important as a gamefish and in the aquarium trade (Iwamoto, 2015). It has been a prominent species in mixed species trawl fisheries in the British Isles and is a notable by-catch species for trawl gears globally (Dunn, 2001; Iwamoto, 2015). Despite being a highly regarded food fish, the only stock assessment for the British Isles, to our knowledge, was carried out between April 1994 and March 1996 (Dunn, 2001). Dunn describes landings from commercial fisheries in England and Wales alongside biological samples and catch data from the English Channel. John Dory was most abundant in the south and southwest of the British Isles (Dunn, 2001; Wheeler, 1969). Evidence suggests that the English Channel is a seasonal nursery ground for Z. faber: seasonal peaks in landings coincided with the period of recruitment, in this case during quarters three and four, when individuals were just over 1 year old at the transition between juvenile and adult life stages species (Dunn, 2001). Recruitment total length of the species was approximately 23 cm TL. Most commercial landings were in the range 23-29 cm TL with a maximum observed TL of 59 cm. Mean TL of first maturity was approximately 26 cm for males and 34.5 cm for females. The global conservation status of this species was last assessed in 2013, deemed 'Data Deficient' on the IUCN Red List (Iwamoto, 2015), which reflects the lack of historical data and biological information for this species (Dunn, 2001). Because of this, stock status and fishing pressure is uncertain for *Z. faber*".

This paragraph provides valuable information for reader but it significantly lacks clarity and confinement. For instance, human health issues with over the threshold elements in the flesh is one argument, data deficient stock assessment is another argument. Similarly, the genetically distinguished populations in different parts of the world that is discussed in the following paragraph, is another statement: "Molecular investigation of this species has shown significant genetic differentiation (7.44%) between clades in the North Atlantic/Mediterranean region and Australasia, indicating the possibility that they have speciated (Ward et al., 2008). The first genome of *Z. faber* was generated in 2016 for a study that suggests immune-related genes play an important role in teleost evolution and speciation (Malmstrøm et al., 2016). Here we present the second published genome of John Dory, collected and sequenced as part of the Darwin Tree of Life project (Blaxter et al., 2022). This dataset will be important for furthering our understanding of teleost pathology, immunology, evolution and phylogenetics (Malmstrøm et al., 2016; Ward et al., 2008)" – this confuse the reader as to why this study is important and why you made efforts to sequence genomes for John Dory.

Background section, requires substantial revision in term of clarity and confinement and more importantly the message the author want to communicate with the reader as to why they have done this study and what gaps this is going to fill and how it serves the intention of the study. It needs to be specifically clarified as to whether this is for seafood industry and human health matter, or conservation matter? Or even management of the stock? What is the main intention here, without this valuable bit, it is not recommended to jump into the methodology and analysis.

Summary:

Despite my encouragement for scientists to publish their work, I must admit that this particular research lacks clarity and conciseness. Therefore, I have no choice but to recommend rejection and suggest resubmission after thorough revisions, including expanding the research and writing in a more concise manner.

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

No

Are the protocols appropriate and is the work technically sound?

Partly

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

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

No

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Fisheries sciences, Biological Sciences, Climate Change, Fish Stock Assessment, influences of environmental and anthropogenic impacts on marine resources

I confirm that I have read this submission and believe that I have an appropriate level of expertise to state that I do not consider it to be of an acceptable scientific standard, for reasons outlined above.

Author Response 10 Oct 2024

Tree of Life Team Sanger

We respectfully disagree with the assertion that the title lacks clarity and intention. As this is a data note, the focus is specifically on presenting the genome sequence, which aligns with the journal's scope for such submissions. Regarding the request for "thorough revisions, including expanding the research and writing in a more concise manner," we believe there may be a misunderstanding of the purpose and structure of data notes. As stated in the journal's guidelines: "Data Notes are brief descriptions of scientific datasets that promote the potential reuse of research data and include details of why and how the data were created; they do not include any analyses or conclusions." Expanding the research or adding detailed analysis would be beyond the scope of this format. We have, however, made edits to the introduction to enhance its clarity. Additionally, we note that the reviewer's comments primarily address the introduction section and do not provide specific reasons for the assertion that the datasets are not presented effectively or that the work is technically unsound. We appreciate the feedback and have aimed to address the concerns raised within the boundaries of the data note format.

Competing Interests: No competing interests were disclosed.

Reviewer Report 14 June 2024

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Naiara Rodriguez-Ezpeleta 匝



AZTI Basque Research and Technology Alliance, Bizkaia, Spain

This manuscript presents the genome sequence of a fish species (John Dory) that is in general poorly studied and for which conservation strategies can not be appropriately defined. Considering that understanding the genetic connectivity is critical for defining management units, and that full genomes facilitate the analysis and interpretation of population genomic data, this study is considered important for improving the conservation of the John Dory. The methods are well described and the results are pertinent. I have only one comment that I think the authors can address easily:

- In the introduction they should better explain what is the knowledge of this species in terms of how many populations exist and potential subspecies. The data of "significant genetic differentiation (7.44%) between clades in the North Atlantic/Mediterranean region and Australasia" seem to be taken from the abstract of a cited paper, but there is not much information here to understand what is the main issue that could be resolved with the help of a full genome sequence.

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: Population genomics, molecular evolution, high-throughput seguencing data analysis

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 10 May 2024

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Shawn M. Burgess

National Human Genome Research Institute, Bethesda, USA

Adkins and colleagues have submitted a manuscript describing the genomic assembly of a John Dory fish (*Zeus faber*). The sequencing was performed using PacBio HiFi and Hi-C data and the assembly using hifiasm and manual curation. The resulting assembly exceeds general benchmarks for high-quality assemblies including scoring for BUSCO, k-mer completeness, and percentage of sequence mapped to chromosomes. All raw and analyzed datasets are deposited in publicly available repositories. The data are clear, the approaches adequately documented. I have no substantial objections to any of the data presented. It should be a very useful assembly for researchers who are interested in John Dory biology.

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: Zebrafish genetics and genomics. Fish genome assembly.

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.