

Reporting Summary

Nature Portfolio wishes to improve the reproducibility of the work that we publish. This form provides structure for consistency and transparency in reporting. For further information on Nature Portfolio policies, see our [Editorial Policies](#) and the [Editorial Policy Checklist](#).

Statistics

For all statistical analyses, confirm that the following items are present in the figure legend, table legend, main text, or Methods section.

n/a	Confirmed
☐	☒ The exact sample size (<i>n</i>) for each experimental group/condition, given as a discrete number and unit of measurement
☐	☒ A statement on whether measurements were taken from distinct samples or whether the same sample was measured repeatedly
☐	☒ The statistical test(s) used AND whether they are one- or two-sided <i>Only common tests should be described solely by name; describe more complex techniques in the Methods section.</i>
☒	☐ A description of all covariates tested
☐	☒ A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons
☐	☒ A full description of the statistical parameters including central tendency (e.g. means) or other basic estimates (e.g. regression coefficient) AND variation (e.g. standard deviation) or associated estimates of uncertainty (e.g. confidence intervals)
☐	☒ For null hypothesis testing, the test statistic (e.g. <i>F</i> , <i>t</i> , <i>r</i>) with confidence intervals, effect sizes, degrees of freedom and <i>P</i> value noted <i>Give P values as exact values whenever suitable.</i>
☐	☒ For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings
☒	☐ For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
☒	☐ Estimates of effect sizes (e.g. Cohen's <i>d</i> , Pearson's <i>r</i>), indicating how they were calculated

Our web collection on [statistics for biologists](#) contains articles on many of the points above.

Software and code

Policy information about [availability of computer code](#)

Data collection	Pipelines for assembly of newly collected exon-capture data are public: https://github.com/lilychughes/FishLifeExonCapture This same pipeline can be used to mine exons from other types of data such as UCES.
Data analysis	R scripts and inputs needed to replicate analyses are available in a Dryad repository (doi: https://doi.org/10.5061/dryad.n2z34tn5r). The following public open source programs were used for analyses: Genomic assembly: Trimmomatic v.0.39; BWA v.0.7.17; SAMtools v.1.9; Velvet v.1.2.10; aTRAM 2.2; Trinity v.2.2; CD-HIT-EST v.4.8.1; Exonerate v.2.4.0; MACSE v.2.03 Phylogenomic inference: AMAS; IQ-TREE MPI multicore v.2.0; ASTRAL-II v.5.7.1 Divergence time estimation: PAML v.4.9h; MEGAX Phylogenetic comparative methods: BioGeoBEARS v.1.1.3; hisse v2.1.1; geomorph v.4.0.5; geiger v.2.0.11; BayesTraits V4; OUwie v.2.10

The BayesTraits executable can be downloaded here: <https://www.evolution.reading.ac.uk/BayesTraitsV4.0.1/BayesTraitsV4.0.1.html>

For manuscripts utilizing custom algorithms or software that are central to the research but not yet described in published literature, software must be made available to editors and reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Portfolio [guidelines for submitting code & software](#) for further information.

Data

Policy information about [availability of data](#)

All manuscripts must include a [data availability statement](#). This statement should provide the following information, where applicable:

- Accession codes, unique identifiers, or web links for publicly available datasets
- A description of any restrictions on data availability
- For clinical datasets or third party data, please ensure that the statement adheres to our [policy](#)

Data availability:

Raw sequence reads have been archived in National Center for Biotechnology Information (NCBI) Sequence Read Archive with the BioProject number PRJNA1074427. Any pre-existing sequence data used is given in Supplementary Tables S2 and S3 with accession codes. Aligned molecular data are available in the Dryad repository (<https://doi.org/10.5061/dryad.n2z34tn5r>). Phylograms, time-calibrated phylogenies and body shape dataset are available in the Dryad repository. All CT scans are publicly available on Morphosource (<https://www.morphosource.org/>) with media numbers in Supplementary Table S7.

Code availability:

Pipelines for genomic assembly are publically available: <https://github.com/lilychughes/FishLifeExonCapture>.

R scripts used to analyze data are available in a Dryad repository (doi: <https://doi.org/10.5061/dryad.n2z34tn5r>).

Research involving human participants, their data, or biological material

Policy information about studies with [human participants or human data](#). See also policy information about [sex, gender \(identity/presentation\), and sexual orientation](#) and [race, ethnicity and racism](#).

Reporting on sex and gender

NA

Reporting on race, ethnicity, or other socially relevant groupings

NA

Population characteristics

NA

Recruitment

NA

Ethics oversight

NA

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Field-specific reporting

Please select the one below that is the best fit for your research. If you are not sure, read the appropriate sections before making your selection.

☐ Life sciences ☐ Behavioural & social sciences ☒ Ecological, evolutionary & environmental sciences

For a reference copy of the document with all sections, see [nature.com/documents/nr-reporting-summary-flat.pdf](https://www.nature.com/documents/nr-reporting-summary-flat.pdf)

Ecological, evolutionary & environmental sciences study design

All studies must disclose on these points even when the disclosure is negative.

Study description

This study includes a phylogenomic analysis of the order Lophiiformes (anglerfishes). The phylogeny is time-calibrated using information from the fossil record (all fossil information is from published studies). Using these phylogenies, we performed phylogenetic comparative methods to infer the tempo and mode of evolution in this group.

Research sample

Phylogenomic data:

New data was collected from 152 individuals from 120 species of Lophiiformes. Target capture probes were based on a set of 1,105 single-copy nuclear exon markers designed for fish phylogenomics, plus an additional 19 nuclear legacy markers and mitochondrial DNA. Outgroup sampling included one holocentrid (representing the sister lineage to Percomorpha), one ophiidid (the earliest diverging member of Percomorpha), one pelagiarian, two syngnatharians, 18 tetraodontiforms and 15 additional eupercarians. Additionally, we successfully mined 5–357 exons for 93 individuals representing 48 species from published UCE data. An additional 1–5 legacy markers were downloaded from NCBI for 10 species. Our final taxonomic sampling when combining all data and remaining after all quality control steps was 132 ingroup species (37.8% of species and 78.1% of genera in Lophiiformes) and 20 of 21 families (all but the monotypic Lophichthyidae). Suborder-level sampling is as follows: 9 species of Lophioidei (32.1% of species and all four genera), 21 species of Ogcocephaloidei (28.7% of species and eight of ten genera), 40 species of Antennarioidei (62.5% of species

and 77.3% of genera [17 of 22 genera]), eight species of Chaunacoidei (50% of species and both genera), and 54 species of Ceratioidei (32.1% of species and 74.3% of genera [26 of 35 ceratioid genera]).

Phenotypic data:

Ten linear body shape measurements were taken from 331 individuals representing 113 species (after quality control). Our CT scan dataset contained 100 species of Lophiiformes (n=1 scan per species). Of these, 38 are new to this study, 33 were recently published in another study by the lead author, and 29 were downloaded from the online repositories MorphoSource (<https://www.morphosource.org/>) or Virtual Natural History Museum (<http://vnhm.de/VNHM/index.php>).

Sampling strategy

Sequence data from 1-3 individuals per species was collected depending on tissue availability and costs. CT scans were collected for one representative individual per species. Body shape measurements were collected from 1-9 individuals per species. All major lophiiform lineages (suborders and all but one family) were sampled for phylogenetic analysis. As many genera and species were sampled as possible given tissue availability. Outgroups were sampled to cover major acanthomorph lineages following Betancur-R et al. 2017.

Data collection

DNA was extracted from tissue samples and quantified by Elizabeth Miller in the MERLAB of the University of Washington (P.I. Luke Tornabene). New DNA sequencing was performed by Arbor Biosciences using FishLife target capture protocols (<https://www.fishtree.org/>). CT scans were obtained at the Karel F. Liem Bio-Imaging Center at the University of Washington Friday Harbor Laboratories as well as Rice University (P.I. Kory Evans). Body shape measurements were collected from museum specimens using calipers by Elizabeth Miller.

Timing and spatial scale

Data were collected between 2020–2022 as part of an NSF Postdoctoral Fellowship to Elizabeth Miller.

Data exclusions

For divergence time estimation and phylogenetic comparative methods, phylogenies were pruned to one representative individual per species as appropriate for these methods. Phylogenies including all individuals with genetic data can be viewed in the Supplementary Material (Appendix A1).

Reproducibility

Multiple individuals per species were included in phylogenetic inference to help confirm species identity. The all-individuals tree is available in Appendix A1. For body shape measurements, 1-9 individuals per species were measured to account for intraspecific variation. R code and inputs needed to replicate analyses can be found in the Dryad repository associated with this study.

Randomization

This is not relevant to our study.

Blinding

This is not relevant to our study.

Did the study involve field work? ☐ Yes ☒ No

Reporting for specific materials, systems and methods

We require information from authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, system or method listed is relevant to your study. If you are not sure if a list item applies to your research, read the appropriate section before selecting a response.

Materials & experimental systems

n/a	Involved in the study
☒	☐ Antibodies
☒	☐ Eukaryotic cell lines
☒	☐ Palaeontology and archaeology
☐	☒ Animals and other organisms
☒	☐ Clinical data
☒	☐ Dual use research of concern
☒	☐ Plants

Methods

n/a	Involved in the study
☒	☐ ChIP-seq
☒	☐ Flow cytometry
☒	☐ MRI-based neuroimaging

Animals and other research organisms

Policy information about [studies involving animals](#); [ARRIVE guidelines](#) recommended for reporting animal research, and [Sex and Gender in Research](#)

Laboratory animals

NA

Wild animals

NA

Reporting on sex

NA

Field-collected samples

Tissue samples of fishes used in this study were loaned from research museum collections listed in the Acknowledgments and Table S1. Contact collection managers for information about long-term storage specific to each collection's policy.

Ethics oversight

This study did not require IACUC approval because no live animals were used.

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Plants

Seed stocks

NA

Novel plant genotypes

NA

Authentication

NA