

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	No software was used to collect data in this study
Data analysis	Custom code was used to process RNAseq data and run bioinformatic programs for transcriptome assembly and downstream analyses including phylogenomic tree reconstruction, Ancestral State Reconstruction (ASR), identification of orthologous genes and tests of positive selection. Support scripts for the data analyses will be made available on Github and the Dryad Digital Repository.

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](#)

Raw RNA sequencing data used for these analyses are publicly available through NCBI's Sequence Read Archive (SRA) database (BioProject ID: XXXXXXXX). The

tissue-specific eye assemblies, associated metadata, and support scripts for the data analyses are available on the Dryad Digital Repository: <https://doi.org/XXXXXXX>. Putative opsin sequences are also available on GenBank (accessions: XXXXX-XXXXX).

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

Reporting on race, ethnicity, or other socially relevant groupings

Population characteristics

Recruitment

Ethics oversight

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

Ecological, evolutionary & environmental sciences study design

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

Study description	This study involved the collection and sequencing of RNA from the eyes of various deep-sea shrimp belonging to the superfamily Oplophoroidea. Eye transcriptomes were subsequently assembled and analyzed using phylogenomic and bioinformatic methods to characterize visual opsin (protein) diversity and look for signatures of adaptive visual evolution among shrimp species that have evolved dual modes of bioluminescence (secretory and light organs) and/or vertically migrate to shallower, photic depths, where abiotic light is also present and exerts additional selective pressures on visual evolution relative to primarily deep species that are only exposed to point sources of biotic light (bioluminescence).
Research sample	Replicates of ten species belonging to the superfamily Oplophoroidea were used to explore visual diversity among these bioluminescent shrimps. Species were chosen to include representatives of as many different genera within the superfamily as possible, and to include species within each of the two major families that had diverse migration and bioluminescent strategies.
Sampling strategy	Samples were opportunistically collected from the water column using a tucker trawl or mocness net, with an insulated cod end. Species were sorted under red/dim light and preserved for RNAseq. As many biological replicates as possible were collected from the deep-sea. Only samples with high quality RNA were used for, sequencing and downstream analyses with the same number of replicates sequenced for the majority of samples (n=3).
Data collection	The authors collected, preserved and identified the samples. RNA was extracted and processed for sequencing by D. DeLeo, which was sequenced on an Illumina HiSeq4000 to obtain 150 bp paired-end reads at the GENEWIZ Core Facility (South Plainfield, NJ).
Timing and spatial scale	Specimens were collected from the Gulf of Mexico and Florida Straits aboard the RV Point Sur (2015-2018), RV PISCES and RV Walton Smith (2016, 2017), respectively. Collections were done by a 9-meter ² Tucker trawl fitted with a light-tight, thermally insulated cod-end or a multiple opening/closing net and environmental sensing system (MOC-10). Samples were immediately sorted and preserved in RNAlater to preserve RNA integrity.
Data exclusions	No data were excluded from the analyses.
Reproducibility	Raw RNAseq data and transcriptome assemblies will be made public, in addition to a description of all programs and versions used to analyze the data to ensure reproducibility.
Randomization	Replicates of each species were randomly chosen based on overall quality and concentration of RNA obtained from the eyes of each species.
Blinding	Blinding is not relevant to this study as each species was discretely assembled, analyzed and compared.

Did the study involve field work? ☒ Yes ☐ No

Field work, collection and transport

Field conditions	Specimens were collected from the Gulf of Mexico and Florida Straits aboard the RV Point Sur (2015-2018), RV PISCES and RV Walton Smith (2016, 2017), respectively. Collections were done by a 9-meter ² Tucker trawl fitted with a light-tight, thermally insulated cod-end or a multiple opening/closing net and environmental sensing system (MOC-10). Samples were immediately sorted in red/dim light and preserved in RNAlater. Samples were collected from the deep-sea.
Location	Gulf of Mexico and Florida Straits between 100-1000 meters.
Access & import/export	No permits were needed for this work.
Disturbance	All samples were collected from mid-water, with minimal impact, slowly towing high quality nets so that larger fauna were not captured.

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	n/a
Wild animals	n/a
Reporting on sex	n/a
Field-collected samples	Specimens were collected from the Gulf of Mexico and Florida Straits aboard the RV Point Sur (2015-2018), RV Sonne and RV Walton Smith (2016, 2017), respectively. Collections were done by a 9-meter ² Tucker trawl fitted with a light-tight, thermally insulated cod-end (Frank and Widder 1999) or a multiple opening/closing net and environmental sensing system (MOC-10). All animals were sorted shipboard under dim red light or low light to avoid damaging photosensitive tissues. Samples were preserved in RNAlater, frozen at -20°C before being transported to Florida International University and stored at -80°C.
Ethics oversight	n/a

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

Plants

Seed stocks

n/a

Novel plant genotypes

n/a

Authentication

n/a