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# Historical contingency shapes adaptive radiation in Antarctic fishes

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# **Supplementary Materials for**

## **Historical contingency shapes adaptive radiation in Antarctic fishes**

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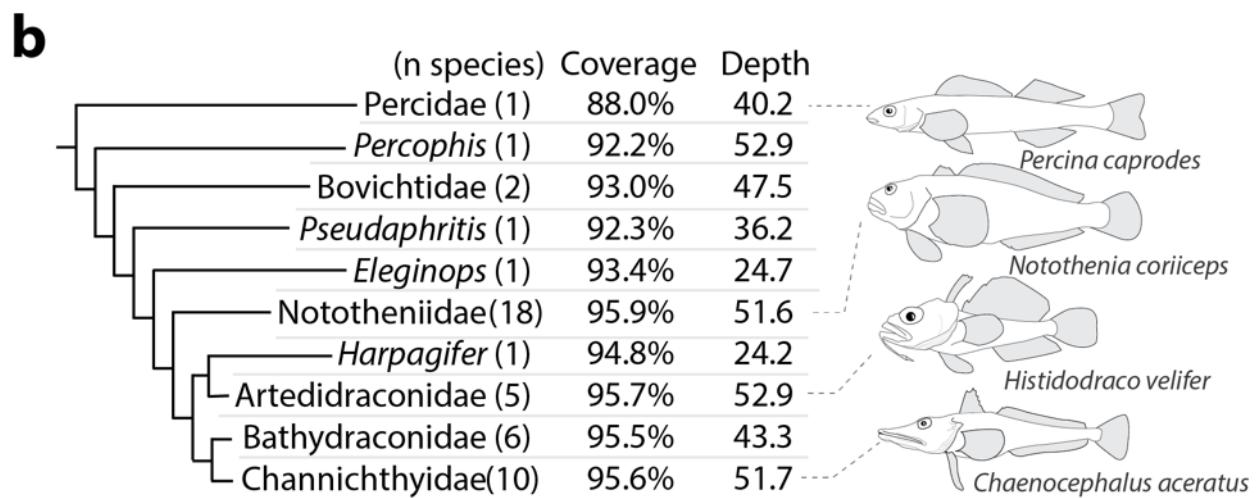
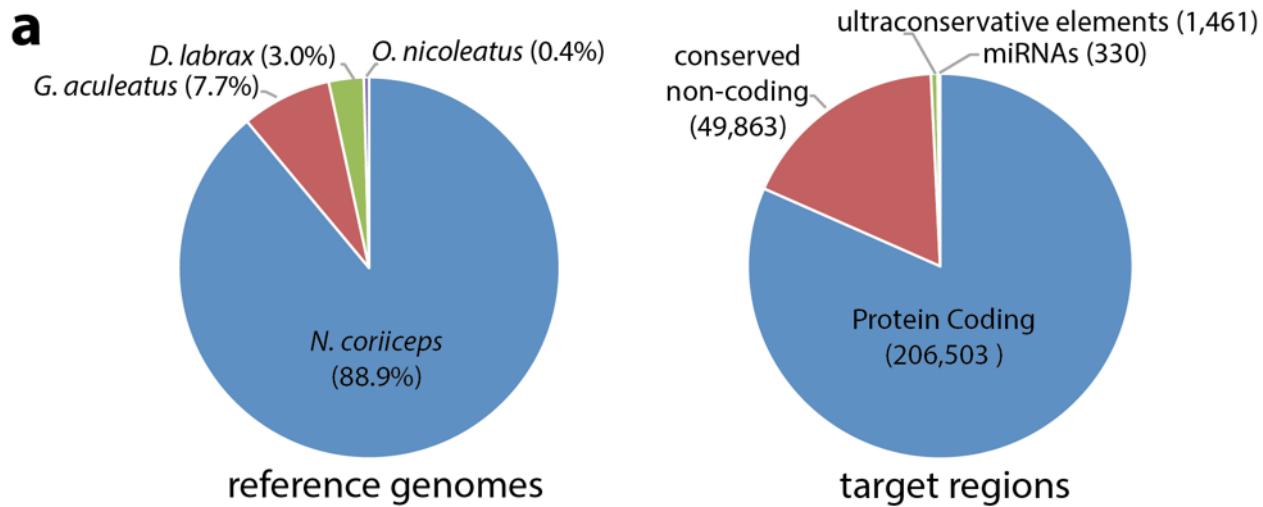
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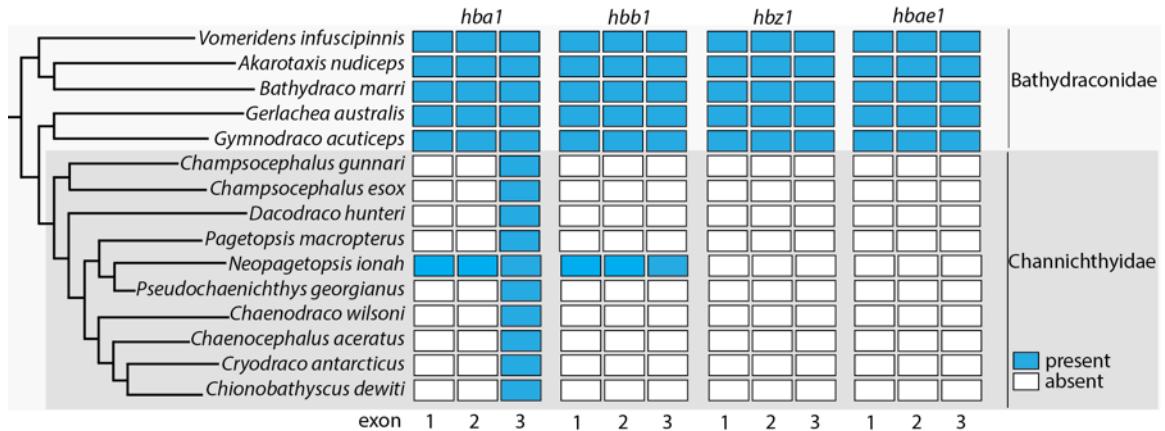
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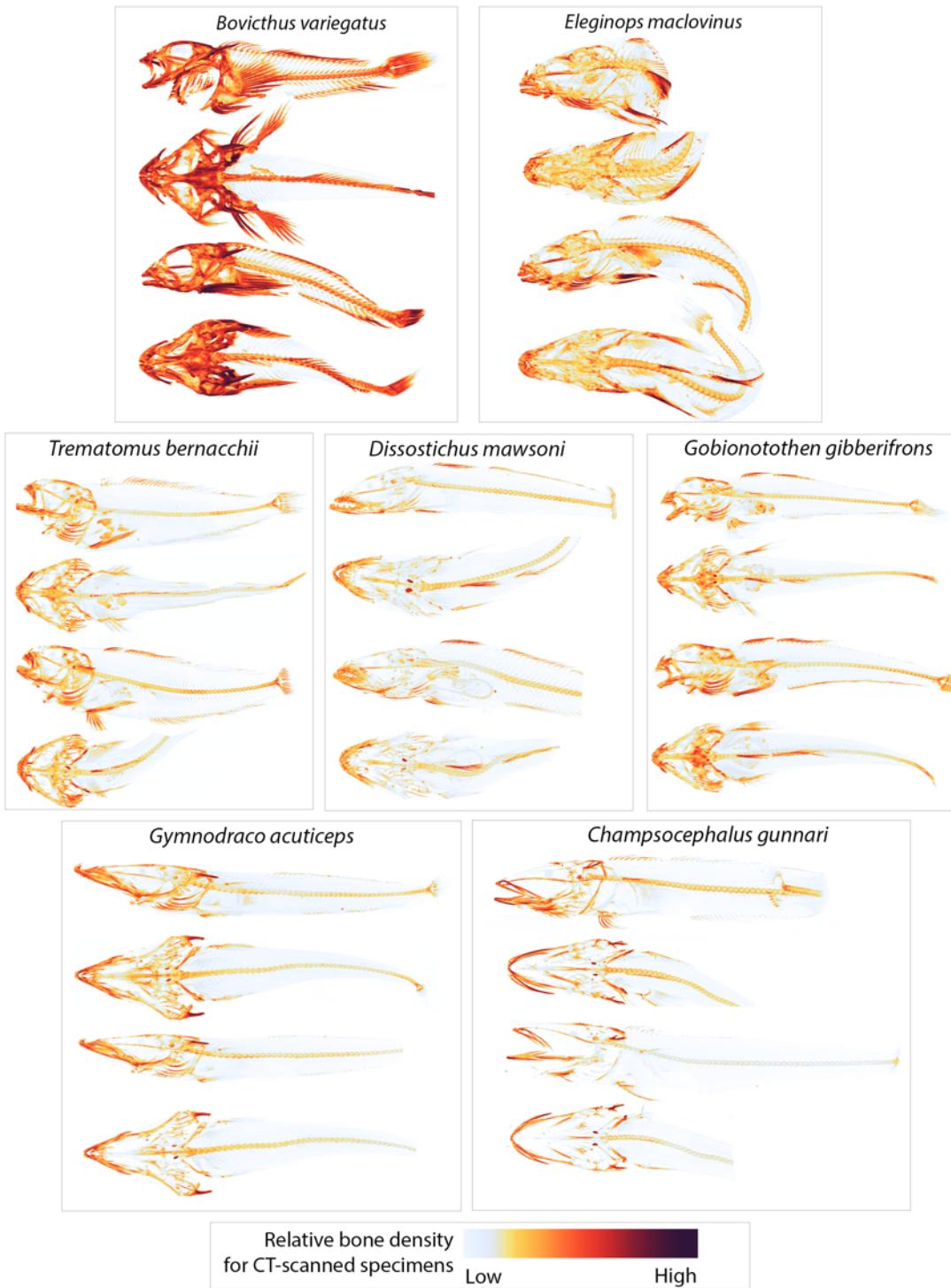
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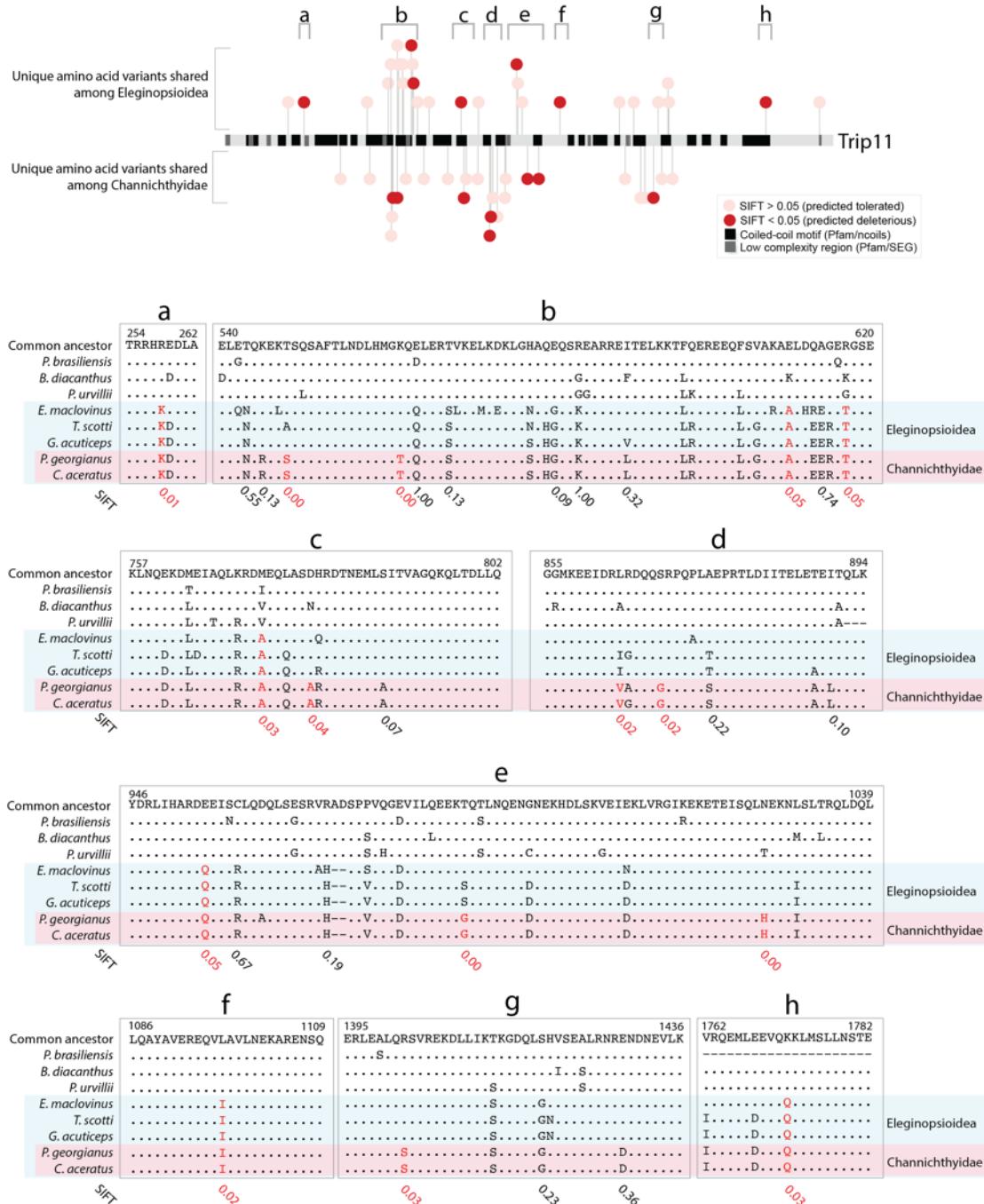
**Supplementary Figure 1. Evolutionary history of notothenioids revealed through targeted sequence enrichment.** (a) Designed ‘phylo-capture’ array demarcating regions targeted for cross-species sequence enrichment; total number of elements in parentheses. (b) Average coverage and sequencing read depth of the targeted regions for each major group within the notothenioids and close outgroups. The number of species within each group is shown in parenthesis.



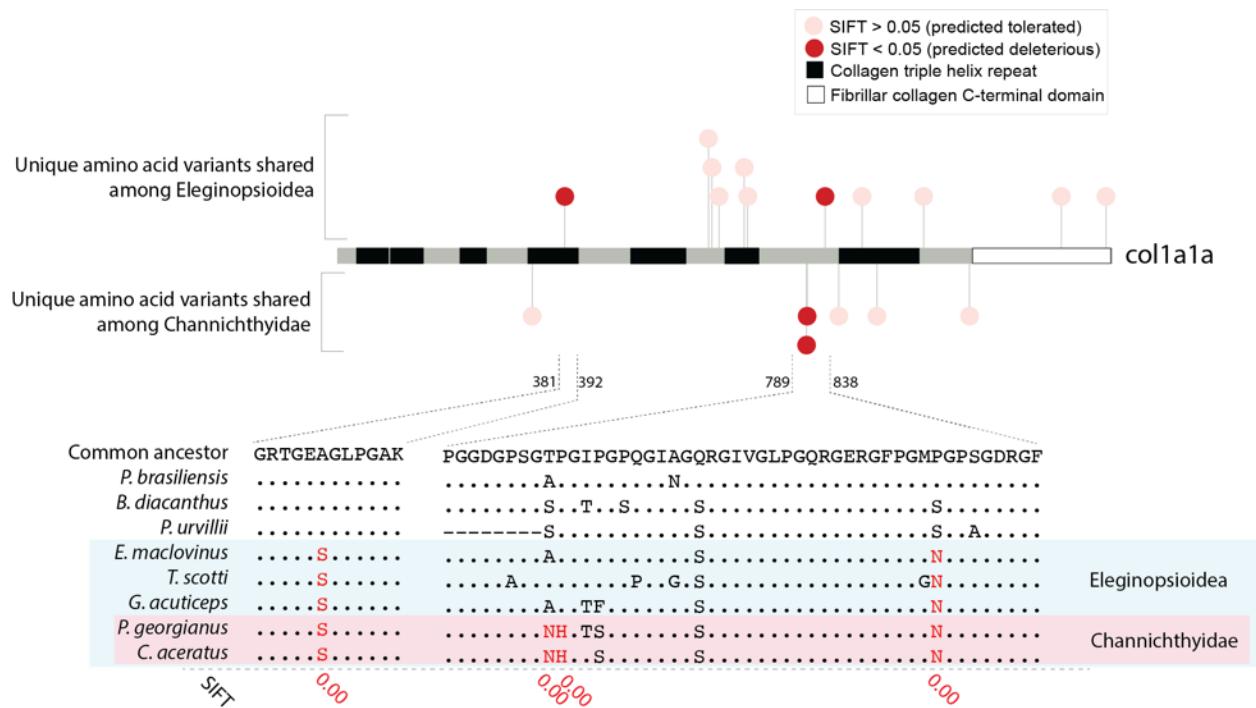
**Supplementary Figure 2. Sensitivity of targeted capture method in confirming and discovering variation in hemoglobin within Channichthyidae.** Presence and lack of detection of individual hemoglobin exons in Channichthyidae and Bathymyidae. Genomic analysis was sensitive in detecting loss of hemoglobins *hba1* and *hbb2* in the icefish clade as previously identified(16). The analysis was able to identify patterns of loss within two new hemoglobins *hbz1* and *hbae1* as well. Presence defined as > 25% coverage and > 5 sequencing reads per exon.



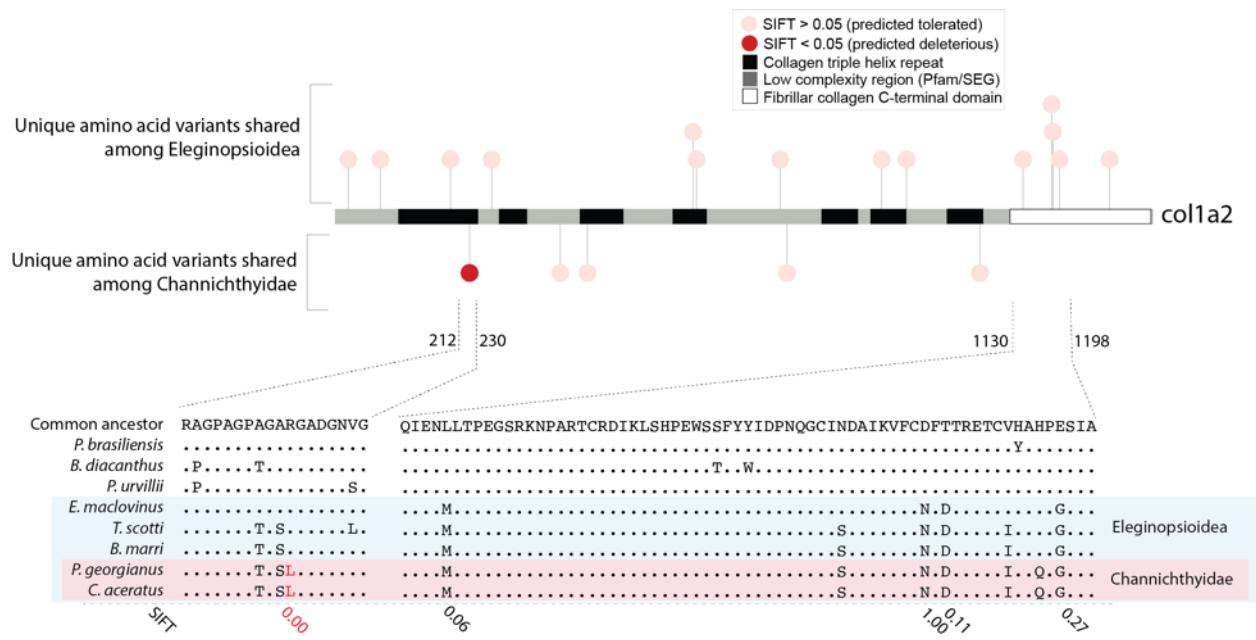
**Supplementary Figure 3. Heatmap of skeletal density in replicate notothenioid CT scans.**  
 Dorsal and ventral views from each adult notothenioid specimen. Voucher information in  
**Supplementary Table S11.**



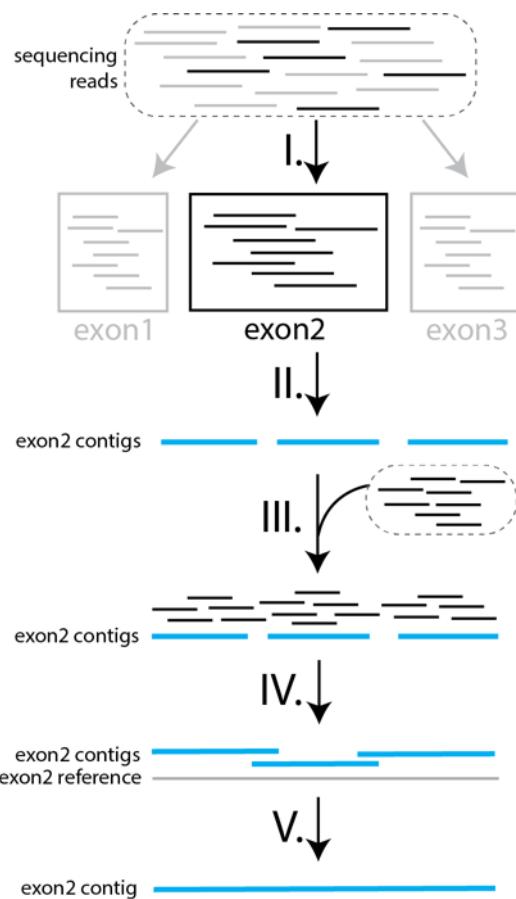
**Supplementary Figure 4. Missense mutations in *trip11* shared among the common ancestor to Eleginopsioidea and common ancestor to Channichthyidae.** Above, lollipop plot of missense mutations mapped onto the orthologous *Gasterosteus aculeatus* transcript (ENSGACT00000016209.3). Below, example multiple sequence alignments. Variants were classified as predicted deleterious (SIFT  $\leq 0.05$ ) or predicted neutral (SIFT  $> 0.05$ ) using SIFT 4G.



**Supplementary Figure 5. Missense mutations in *colla1a* shared among the common ancestor to Eleginopsioidea and common ancestor to Channichthyidae.** Above, lollipop plot of missense mutations mapped onto the *Notothenia coriiceps colla1a* transcript (XP\_010767913.1). Below, example multiple sequence alignments. Variants were classified as predicted deleterious (SIFT  $\leq 0.05$ ) or predicted neutral (SIFT  $> 0.05$ ) using SIFT 4G.



**Supplementary Figure 6. Missense mutations in *col1a2* shared among the common ancestor to Eleginopsioidea and common ancestor to Channichthyidae.** Above, lollipop plot of missense mutations mapped onto the *Notothenia coriiceps* *col1a2* transcript (XP\_010772950.1). Below, example multiple sequence alignments. Variants were classified as predicted deleterious (SIFT  $\leq 0.05$ ) or predicted neutral (SIFT  $> 0.05$ ) using SIFT 4G.



I. Bin sequencing reads by orthology to target regions using blastn and dc-megablast

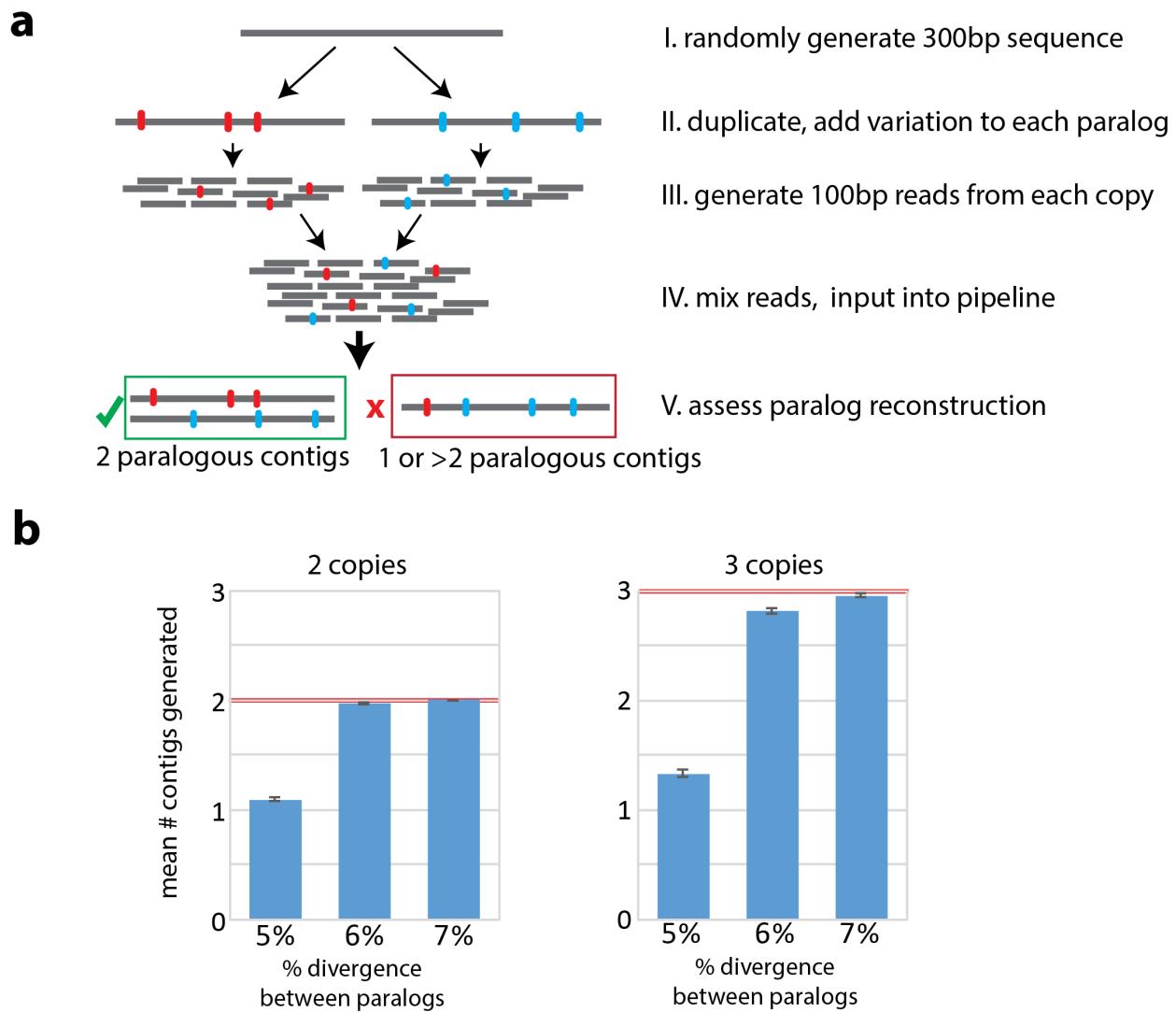
II. *de novo* contig assembly of target bins with CAP3 and UCLUST

III. Bin all reads to assembled contigs with NextGenMap to recruit previously un-binned reads

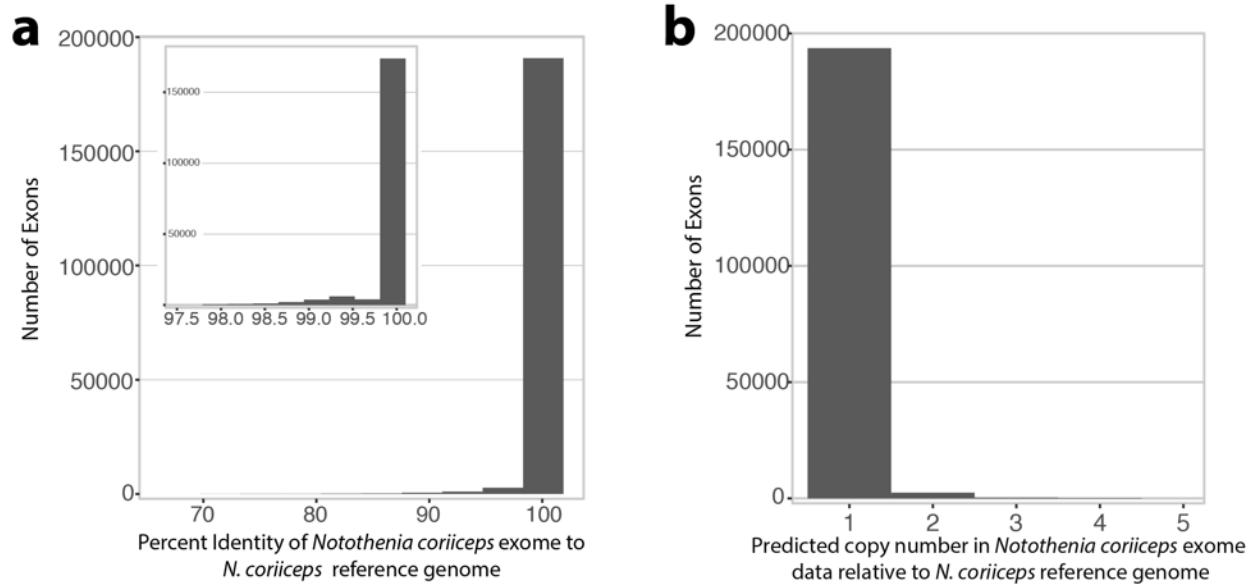
IV. Second round of *de novo* assembly with CAP3 to refine and extend edges of contigs

V. Merge multiple contigs (if necessary), using reference as a scaffold

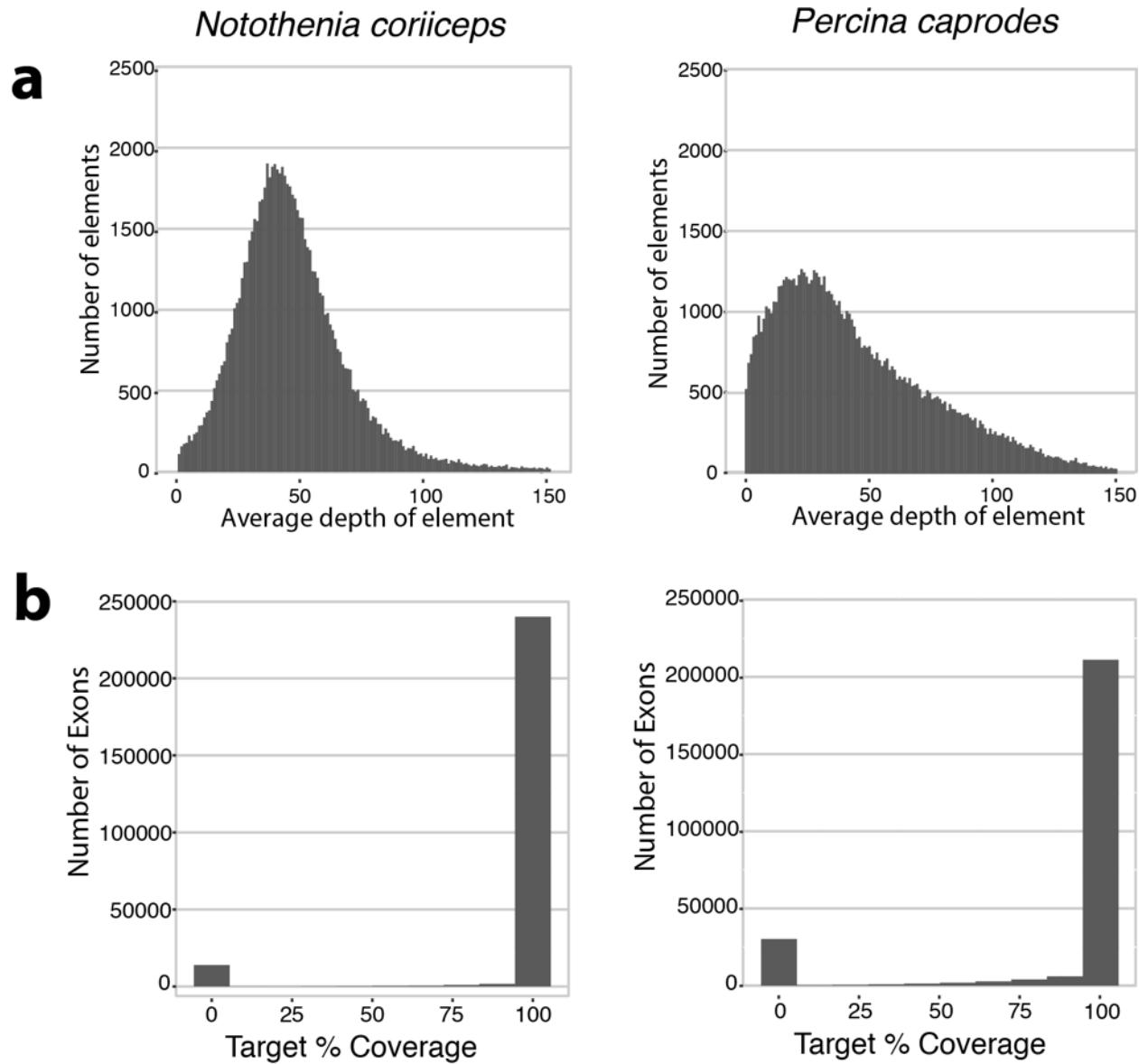
**Supplementary Figure 7. Overview of contig assembly**



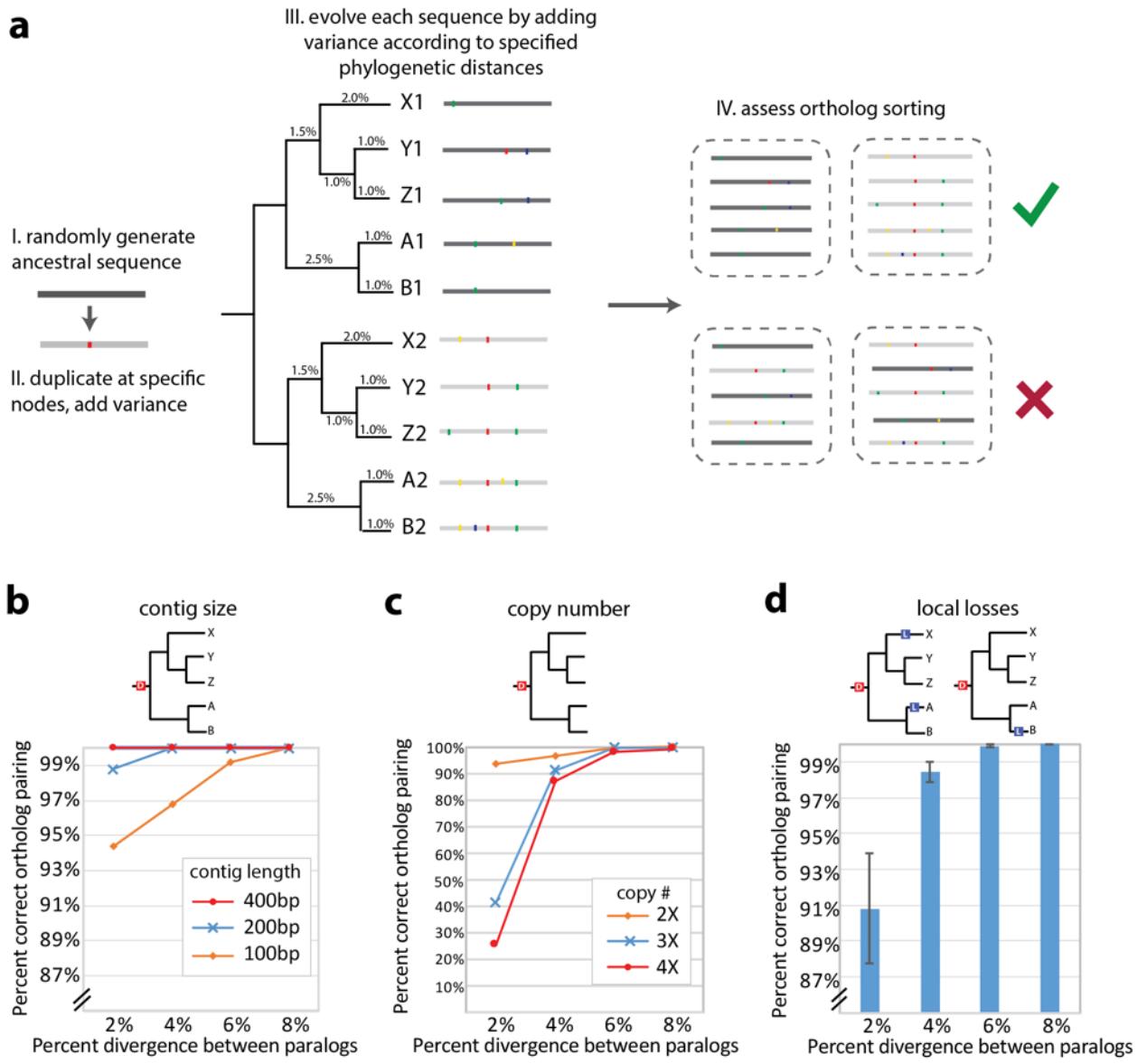
**Supplementary Figure 8. Detection of copy number variants through contig assembly.** (a) Simulation of the limits of the contig assembly pipeline to reconstruct duplicate sequences from mixed read populations. (b) The mean number of contigs generated by the pipeline when reads from multiple input contigs were placed into the simulation. Data from 250 simulations of random 300-bp DNA fragments. Red line indicates input number of contigs. Error bars represent  $\pm 1$  sem.



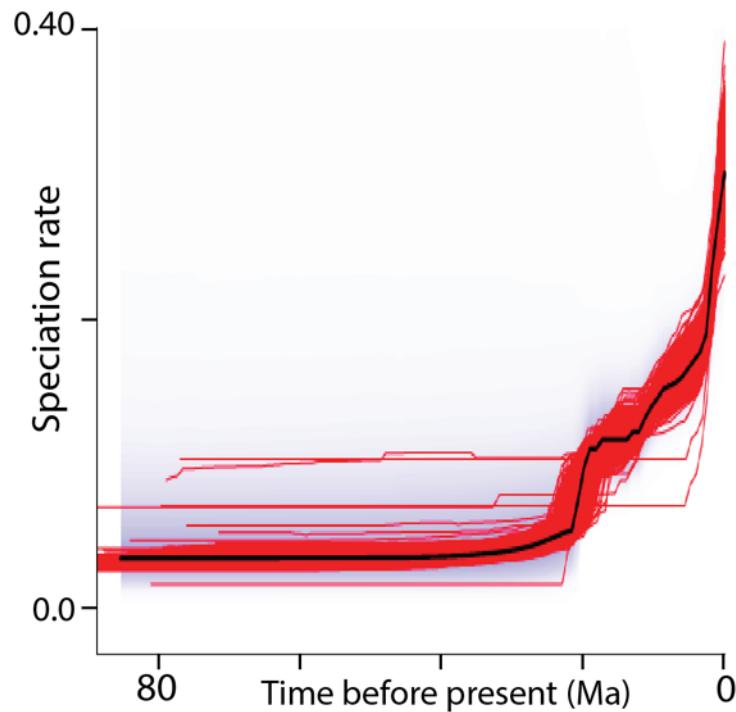
**Supplementary Figure 9. Comparison of reconstructed *Notothenia coriiceps* exome to published *N. coriiceps* genome.** (a) Histogram of percent identity of the reference target regions relative to the corresponding region on the previously assembled *N. coriiceps* genome<sup>30</sup>. (b) Histogram of predicted copy number for an exon in the exome reconstruction compared with the reference genome.



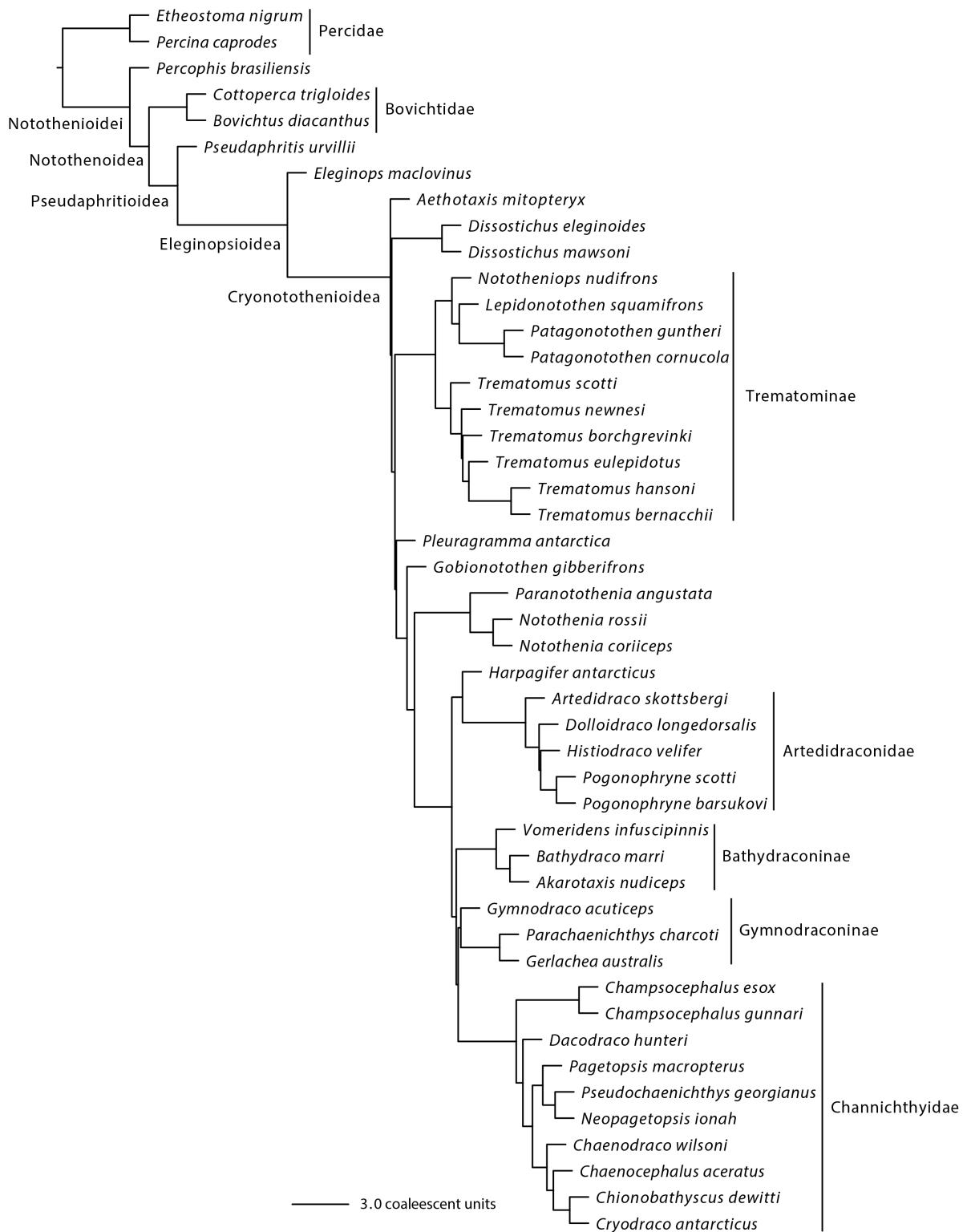
**Supplementary Figure 10. Distribution of coverage as a function of evolutionary distance from hybridization baits.** (a) Histogram of average depth per targeted region (exon, CNE, etc.), showing flattened distribution of read depth in *P. caprodes* suggesting that certain target regions in distantly related groups hybridize less efficiently. (b) Percent of bases covered with at least one read in both species, showing the majority of target regions were either highly covered or not covered at all.



**Supplementary Figure 11. Assessing robustness of pairing of duplicated elements for comparative genomics.** (a) Simulation to test the ability of this approach to properly identify orthologs among ancestrally duplicated sequences. As part of the pipeline for annotation, when there is more than one element per target region in the reference genome in at least two species, a gene tree is constructed with IQTree and reconciled with the species tree using Notung. Notung infers patterns of gain and loss throughout the phylogeny. Simulation was run 250 times. **b-d)** Effect of element length (**b**), copy number (**c**) and local losses within the phylogeny (**d**) on correct matching of duplicates relative to the percent divergence of the duplicated elements. Error bars represent  $\pm 1$  sem.



**Supplementary Figure 12.** Speciation rates through time estimated by BAMM across 500 random samples of the posterior distributions of timetrees. Blue color density shading corresponds to the 90% highest posterior density interval of rate estimates at a given time point across all trees. Red lines correspond with the mean speciation rates through time estimated from each tree of the posterior distribution. The black line corresponds to the mean speciation rate estimated from the maximum clade credibility tree that is also depicted in the primary text.



**Supplementary Figure 13. Notothenioid species tree.** Phylogenetic relationships of the sequenced species inferred as a species tree using ASTRAL and 11,627 gene trees. All nodes were supported with a quadpartition posterior probability of 1.00.

**Supplementary Table 1. Coverage at various read depth thresholds**

Species	Coverage at depth			
	>1X	>2X	>4X	>10X
<i>Percina caprodes</i>	88.0%	87.4%	84.5%	73.4%
<i>Percophis brasiliensis</i>	92.2%	92.0%	90.4%	83.7%
<i>Bovichtus diacanthus</i>	92.2%	91.9%	90.6%	82.8%
<i>Cottoperca trigloides</i>	93.9%	92.4%	88.6%	78.3%
<i>Pseudaphritis urvillii</i>	92.3%	91.8%	89.4%	79.1%
<i>Eleginops maclovinus</i>	93.4%	93.0%	90.9%	78.2%
<i>Pleurgramma antarctica</i>	95.9%	95.8%	95.3%	92.7%
<i>Aethotaxis mitopteryx</i>	96.4%	96.4%	96.1%	95.2%
<i>Dissostichus mawsoni</i>	95.6%	95.5%	94.7%	88.4%
<i>Dissostichus eleginoides</i>	95.9%	95.8%	95.4%	93.0%
<i>Nototheniops nudifrons</i>	95.9%	95.8%	95.4%	93.3%
<i>Lepidonotothen squamifrons</i>	95.9%	95.9%	95.5%	93.6%
<i>Patagonotothen squamiceps</i>	95.9%	95.9%	95.5%	93.6%
<i>Patagonotothen guntheri</i>	95.9%	95.8%	95.4%	93.3%
<i>Trematomus scotti</i>	95.7%	95.6%	95.1%	92.2%
<i>Trematomus newnesi</i>	95.8%	95.7%	95.2%	92.3%
<i>Trematomus eulepidotus</i>	95.9%	95.9%	95.5%	93.9%
<i>Trematomus bernacchii</i>	95.8%	95.6%	95.1%	91.6%
<i>Trematomus borchgrevinki</i>	95.7%	95.6%	95.1%	90.8%
<i>Trematomus hansonii</i>	95.6%	95.5%	95.1%	92.6%
<i>Gobionotothen gibberifrons</i>	96.0%	95.9%	95.4%	93.1%
<i>Notothenia coriiceps</i>	96.0%	95.9%	95.5%	93.3%
<i>Notothenia rossii</i>	95.7%	95.6%	95.1%	91.8%
<i>Paranotothenia angustata</i>	96.0%	95.9%	95.4%	92.2%
<i>Harpagifer antarcticus</i>	94.8%	94.6%	93.1%	81.9%
<i>Artedidraco skottsbergi</i>	95.3%	95.1%	94.2%	88.0%
<i>Histiadraco velifer</i>	95.8%	95.7%	95.3%	93.3%
<i>Dolloidraco longedorsalis</i>	95.8%	95.7%	95.4%	94.1%
<i>Pogonophryne scotti</i>	95.7%	95.6%	95.2%	92.8%
<i>Pogonophryne barsukovi</i>	95.8%	95.7%	95.3%	93.1%
<i>Gerlachea australis</i>	95.6%	95.4%	94.5%	88.9%
<i>Parachaenichthys charcoti</i>	94.2%	93.8%	92.0%	79.8%
<i>Vomeridens infuscipinnis</i>	95.8%	95.7%	95.1%	91.7%
<i>Akarotaxis nudiceps</i>	95.8%	95.7%	95.1%	91.9%
<i>Bathydraco marri</i>	95.7%	95.6%	95.2%	92.3%

<i>Gymnодraco acuticeps</i>	96.2%	96.1%	95.8%	94.4%
<i>Dacodraco hunteri</i>	95.4%	95.4%	94.9%	92.3%
<i>Champscephalus gunnari</i>	95.7%	95.5%	94.9%	92.0%
<i>Champscephalus esox</i>	95.4%	95.3%	94.7%	91.5%
<i>Pagetopsis macropterus</i>	95.4%	95.3%	94.7%	91.6%
<i>Neopagetopsis ionah</i>	95.5%	95.4%	94.8%	91.3%
<i>Pseudochaenichthys georgianus</i>	95.4%	95.3%	94.8%	92.1%
<i>Chaenodraco wilsoni</i>	95.5%	95.4%	95.0%	93.0%
<i>Chaenocephalus aceratus</i>	96.0%	95.9%	95.6%	93.9%
<i>Cryodraco antarcticus</i>	95.6%	95.5%	94.9%	91.5%
<i>Chionobathyscus dewitti</i>	95.2%	94.8%	93.7%	88.6%

**Supplementary Table 2. Coverage of targeted regions in cross-species targeted sequence enrichment**

species	total		<i>N. coriiceps</i> genome		non- <i>N. coriiceps</i> genome		CDS		CNE	
	cov.	depth	cov.	depth	cov.	depth	cov.	depth	cov.	depth
<i>Percina caprodes</i>	88.0%	40.2	87.6%	34.9	87.4%	60.3	86.6%	34.4	94.8%	64.7
<i>Percophis brasiliensis</i>	92.2%	52.9	92.3%	46.7	90.3%	76.3	91.3%	45.6	96.6%	84.7
<i>Bovichtus diacanthus</i>	92.2%	54.3	92.3%	47.5	89.9%	78.7	91.3%	46.1	96.1%	90.4
<i>Cottoperca trigloides</i>	93.9%	40.7	94.4%	35.8	89.7%	59.4	93.4%	35.0	95.9%	66.4
<i>Pseudaphritis urvillii</i>	92.3%	36.2	92.5%	32.2	88.7%	51.4	91.5%	31.4	96.1%	57.5
<i>Eleginops maclovinus</i>	93.4%	24.7	94.3%	23.4	85.4%	28.9	92.9%	23.0	95.4%	32.2
<i>Aethotaxis mitopteryx</i>	95.9%	48.5	97.2%	48.8	86.3%	43.7	95.8%	48.8	96.4%	47.1
<i>Dissostichus mawsoni</i>	96.4%	89.1	97.7%	89.6	87.9%	81.3	96.4%	89.3	96.7%	87.8
<i>Dissostichus eleginoides</i>	95.6%	29.9	97.0%	30.3	85.4%	26.9	95.5%	30.3	96.0%	27.8
<i>Nototheniops nudifrons</i>	95.9%	50.1	97.2%	50.2	86.5%	46.6	95.8%	50.2	96.4%	49.7
<i>Lepidonotothen squamifrons</i>	95.9%	52.1	97.2%	52.1	86.5%	48.9	95.8%	52.0	96.4%	52.6
<i>Patagonotothen squamiceps</i>	95.9%	53.0	97.2%	53.2	86.9%	49.6	95.9%	53.1	96.4%	52.6
<i>Patagonotothen guntheri</i>	95.9%	54.5	97.2%	54.8	86.9%	50.5	95.8%	55.1	96.4%	51.8
<i>Trematomus scotti</i>	95.9%	54.6	97.2%	54.7	86.6%	51.0	95.8%	54.8	96.4%	53.6
<i>Trematomus newnesi</i>	95.7%	44.4	97.0%	44.6	86.1%	41.2	95.6%	44.5	96.2%	43.9
<i>Trematomus eulepidotus</i>	95.8%	47.1	97.1%	47.5	86.1%	42.5	95.7%	47.5	96.3%	45.4
<i>Trematomus bernacchii</i>	95.9%	54.6	97.2%	55.4	86.5%	47.8	95.8%	55.5	96.4%	50.3
<i>Trematomus borchgrevinki</i>	95.8%	44.7	97.1%	44.8	86.1%	41.8	95.6%	44.8	96.3%	43.9
<i>Trematomus hansonii</i>	95.7%	41.6	97.1%	42.1	85.9%	37.9	95.6%	42.2	96.2%	38.9
<i>Pleuragramma antarctica</i>	95.6%	61.3	96.9%	61.1	86.4%	58.2	95.5%	60.9	96.1%	62.9
<i>Gobionotothen gibberifrons</i>	96.0%	51.0	97.3%	51.3	86.3%	46.5	95.8%	51.2	96.5%	49.9
<i>Notothenia coriiceps</i>	96.0%	54.3	97.5%	54.7	85.0%	49.3	95.8%	54.6	96.8%	53.0
<i>Notothenia rossii</i>	95.7%	54.9	97.2%	57.0	84.5%	43.5	95.5%	57.1	96.4%	44.8
<i>Paranotothenia angustata</i>	96.0%	43.1	97.4%	44.0	85.6%	36.9	95.9%	43.7	96.6%	40.6
<i>Harpagifer antarcticus</i>	94.8%	24.2	96.4%	24.5	83.3%	21.4	94.7%	24.4	95.4%	23.1
<i>Artedidraco skottsbergi</i>	95.3%	32.1	96.8%	32.4	84.5%	29.2	95.2%	32.3	96.0%	30.9
<i>Histiodraco velifer</i>	95.8%	55.3	97.2%	56.0	85.8%	49.1	95.7%	55.9	96.5%	52.7
<i>Dolloidraco longedorsalis</i>	95.8%	68.2	97.2%	69.2	85.6%	59.4	95.6%	69.1	96.5%	64.0
<i>Pogonophryne scotti</i>	95.7%	52.6	97.1%	53.1	85.6%	47.0	95.6%	53.0	96.5%	50.5
<i>Pogonophryne barsukovi</i>	95.8%	56.5	97.2%	57.2	85.8%	50.5	95.7%	57.1	96.5%	54.0
<i>Gerlachea australis</i>	95.6%	35.2	97.0%	35.4	84.9%	32.4	95.4%	35.3	96.2%	34.6
<i>Parachaenichthys charcotii</i>	94.2%	27.8	95.8%	28.5	82.0%	23.6	94.1%	28.5	94.6%	24.4
<i>Vomeridens infuscipinnis</i>	95.9%	63.2	97.3%	64.6	85.8%	53.9	95.8%	64.7	96.5%	56.6
<i>Akarotaxis nudiceps</i>	95.8%	44.2	97.2%	44.7	85.5%	39.8	95.6%	44.6	96.4%	42.4
<i>Bathydraco marri</i>	95.8%	45.7	97.2%	46.3	85.6%	40.8	95.7%	46.2	96.4%	43.4

<i>Gymnodraco acuticeps</i>	95.7%	43.4	97.2%	44.4	85.2%	37.1	95.6%	44.5	96.3%	38.5
<i>Champscephalus gunnari</i>	96.2%	74.8	97.5%	75.4	86.7%	67.5	96.0%	75.2	96.9%	72.6
<i>Champscephalus esox</i>	95.4%	50.3	96.9%	51.0	85.1%	43.3	95.3%	50.9	96.0%	47.1
<i>Dacodraco hunteri</i>	95.7%	47.8	97.1%	48.1	85.3%	43.3	95.6%	47.9	96.3%	47.1
<i>Pagetopsis macropterus</i>	95.4%	46.0	96.8%	46.7	84.9%	40.4	95.3%	46.5	96.0%	43.6
<i>Neopagetopsis ionah</i>	95.4%	48.0	96.8%	48.6	85.0%	42.5	95.3%	48.5	96.0%	45.7
<i>Pseudochaenichthys georgianus</i>	95.5%	44.6	96.9%	44.8	85.3%	41.5	95.4%	44.5	96.1%	45.3
<i>Chaenodraco wilsoni</i>	95.4%	48.3	96.8%	48.9	84.8%	43.0	95.2%	48.8	96.1%	45.9
<i>Chaenocephalus aceratus</i>	95.5%	53.6	96.9%	54.3	85.1%	47.3	95.4%	54.2	96.2%	50.8
<i>Cryodraco antarcticus</i>	96.0%	62.3	97.4%	63.2	86.3%	54.8	95.9%	63.1	96.6%	58.4
<i>Chionobathyscus dewitti</i>	95.6%	41.7	97.0%	42.3	85.3%	36.6	95.5%	42.3	96.1%	38.6

**Supplementary Table 3. Recovery of heterozygous and fixed variation**

Species	# Het SNPs	Het SNP/base	Percent of targets with Het	# Fixed species-specific SNPs	Fixed SNP/base	Percent of targets with fixed, species-specific SNP
<i>Percina caprodes</i>	48,777	0.0014	15.7%	300,691	0.0087	52.9%
<i>Percophis brasiliensis</i>	69,209	0.0019	19.7%	698,153	0.0187	77.4%
<i>Bovichtus diacanthus</i>	36,886	0.0010	11.6%	379,258	0.0096	54.1%
<i>Cottoperca trigloides</i>	30,261	0.0008	8.6%	428,614	0.0116	62.6%
<i>Pseudaphritis urvillii</i>	31,342	0.0009	10.2%	501,713	0.0137	69.9%
<i>Eleginops maclovinus</i>	53,559	0.0014	16.1%	599,422	0.0162	74.2%
<i>Pleuragramma antarctica</i>	154,174	0.0039	31.7%	223,284	0.0057	41.1%
<i>Aethotaxis mitopteryx</i>	77,172	0.0020	19.7%	69,895	0.0018	19.3%
<i>Dissostichus mawsoni</i>	42,867	0.0011	12.1%	40,827	0.0010	12.7%
<i>Dissostichus eleginoides</i>	53,849	0.0014	15.6%	44,805	0.0012	14.0%
<i>Nototheniops nudifrons</i>	58,203	0.0015	15.6%	41,278	0.0011	12.5%
<i>Lepidonotothen squamifrons</i>	62,976	0.0016	17.3%	26,827	0.0007	8.4%
<i>Patagonotothen squamiceps</i>	44,838	0.0011	12.4%	53,013	0.0014	16.0%
<i>Patagonotothen guntheri</i>	74,580	0.0019	19.7%	23,794	0.0006	7.5%
<i>Trematomus scotti</i>	66,169	0.0017	17.7%	25,783	0.0007	8.2%
<i>Trematomus newnesi</i>	53,985	0.0014	14.9%	39,385	0.0010	12.3%
<i>Trematomus eulepidotus</i>	47,855	0.0012	12.2%	25,282	0.0006	8.1%
<i>Trematomus bernacchii</i>	58,849	0.0015	16.1%	2,307	0.0001	0.4%
<i>Trematomus borchgrevinki</i>	37,388	0.0010	10.3%	33,673	0.0009	10.9%
<i>Trematomus hansonii</i>	61,373	0.0016	17.1%	2,041	0.0001	0.3%
<i>Gobionotothen gibberifrons</i>	64,191	0.0016	16.3%	111,759	0.0029	28.0%
<i>Notothenia coriiceps</i>	60,749	0.0015	16.2%	40,240	0.0010	12.6%
<i>Notothenia rossii</i>	66,220	0.0017	17.8%	45,190	0.0012	13.6%
<i>Paranotothenia angustata</i>	55,302	0.0014	15.5%	104,620	0.0027	27.7%
<i>Harpagifer antarcticus</i>	43,842	0.0012	13.1%	53,936	0.0014	16.4%
<i>Artedidraco skottsbergi</i>	64,811	0.0017	18.5%	11,638	0.0003	3.9%
<i>Histiodraco velifer</i>	42,299	0.0011	12.1%	8,164	0.0002	2.7%
<i>Dolloidraco longedorsalis</i>	52,428	0.0013	15.0%	5,526	0.0001	1.7%
<i>Pogonophryne scotti</i>	45,650	0.0012	13.2%	3,446	0.0001	1.0%
<i>Pogonophryne barsukovi</i>	44,453	0.0011	12.8%	3,061	0.0001	0.9%

<i>Gerlachea australis</i>	73,269	0.0019	20.5%	21,708	0.0006	7.0%
<i>Parachaenichthys charcoti</i>	30,350	0.0009	9.2%	46,682	0.0013	14.4%
<i>Vomeridens infuscipinnis</i>	41,820	0.0011	11.0%	36,750	0.0009	11.8%
<i>Akarotaxis nudiceps</i>	56,254	0.0014	15.6%	12,176	0.0003	4.0%
<i>Bathydraco marri</i>	48,433	0.0012	13.2%	19,462	0.0005	6.4%
<i>Gymnodraco acuticeps</i>	58,027	0.0015	16.5%	41,434	0.0011	12.9%
<i>Dacodraco hunteri</i>	52,739	0.0013	14.8%	23,605	0.0006	7.8%
<i>Champscephalus gunnari</i>	37,061	0.0010	10.9%	14,024	0.0004	4.5%
<i>Champscephalus esox</i>	35,548	0.0009	10.3%	40,568	0.0010	12.7%
<i>Pagetopsis macropterus</i>	41,887	0.0011	12.1%	20,670	0.0005	6.9%
<i>Neopagetopsis ionah</i>	58,368	0.0015	15.9%	17,087	0.0004	5.6%
<i>Pseudochaenichthys georgianus</i>	14,921	0.0004	3.7%	29,936	0.0008	9.8%
<i>Chaenodraco wilsoni</i>	108,985	0.0028	27.6%	4,751	0.0001	1.4%
<i>Chaenocephalus aceratus</i>	26,447	0.0007	7.1%	18,353	0.0005	6.2%
<i>Cryodraco antarcticus</i>	115,004	0.0029	26.9%	2,531	0.0001	0.4%
<i>Chionobathyscus dewitti</i>	44,558	0.0011	11.9%	8,258	0.0002	2.7%

**Supplementary Table 4. Substitution rates in notothenioids and outgroups**

Branch	Substitution Rate	95% HPD
<i>Percina caprodes</i>	0.00078	0.0005-0.0011
<i>Percophis brasiliensis</i>	0.00077	0.0005-0.0011
<i>Bovichtus diacanthus</i>	0.00077	0.0005-0.0011
<i>P. urvillii</i> + Eleginopsioidea	0.00098	0.0004-0.0023
<i>Pseudaphritis urvillii</i>	0.00079	0.0005-0.0011
Eleginopsioidea	0.00228	0.0010-0.0039
<i>Eleginops maclovinus</i>	0.00168	0.0009-0.0024
Cryonotothenioidea	0.00218	0.0008-0.0038
<i>Dissostichus mawsoni</i>	0.00042	0.0001-0.0009
<i>Trematomus scottii</i>	0.00061	0.0002-0.0010
<i>Gobionotothen gibberifrons</i>	0.00078	0.0004-0.0012
<i>Notothenia coriiceps</i>	0.00076	0.0005-0.0012
<i>Pogonophryne scotti</i>	0.00071	0.0004-0.0011
<i>Harpagifer antarcticus</i>	0.00076	0.0005-0.0012
<i>Chaenocephalus aceratus</i>	0.00092	0.0003-0.0022
<i>Bathydraco marri</i>	0.00069	0.0003-0.0011

**Supplementary Table 5. Positive selection in bone-associated<sup>†</sup> genes in Eleginopsioidea**

Ensembl Ortholog	Gene	P	P (Bonferroni)	LRT
ENSGACG00000014066	LRRK1	1.46E-03	2.93E-03	10.922
ENSGACG00000001740	prdm1a	5.90E-03	1.18E-02	8.170
ENSGACG00000014774	cahz	4.37E-02	8.73E-02	4.251
ENSGACG00000011819	col12a1b	4.44E-02	8.88E-02	4.218
ENSGACG00000010282	minpp1b	1.11E-02	2.21E-02	6.935
ENSGACG00000013609	tacr1a	3.90E-02	7.81E-02	4.469
ENSGACG00000016847	sparc	1.69E-03	3.39E-03	10.635
ENSGACG00000013381	GABBR1	2.69E-02	5.38E-02	5.192
ENSGACG00000014923	col5a2b	1.25E-05	2.49E-05	20.389
ENSGACG00000008814	tmem119a	2.14E-02	4.28E-02	5.640
ENSGACG00000007303	ctnnbip1	8.15E-03	1.63E-02	7.534
ENSGACG00000018120	IGFBP4	7.24E-05	1.45E-04	16.889
ENSGACG00000005830	prpsap2	2.48E-02	4.95E-02	5.354
ENSGACG00000012442	notch2	3.74E-02	7.47E-02	4.553
ENSGACG00000013102	mapk3	1.42E-03	2.83E-03	10.990
ENSGACG00000003838	si:ch211-285f17.1	8.50E-03	1.70E-02	7.451
ENSGACG00000005488	phex	1.17E-02	2.34E-02	6.819
ENSGACG00000004564	hoxd10a	2.44E-03	4.88E-03	9.911
ENSGACG00000007087	LIMD1	4.55E-03	9.11E-03	8.680
ENSGACG00000006673	rbp4	4.58E-04	9.17E-04	13.225
ENSGACG00000017924	ghrb	2.67E-02	5.34E-02	5.208
ENSGACG00000019885	slc8a1a	1.36E-02	2.72E-02	6.530
ENSGACG00000011433	sun1	8.17E-03	1.63E-02	7.529
ENSGACG00000007343	col9a2	2.79E-02	5.58E-02	5.120
ENSGACG00000006287	ulf1	1.88E-02	3.76E-02	5.892
ENSGACG00000008639	pdkcc	1.49E-02	2.99E-02	6.343
ENSGACG00000010609	ptk2bb	2.00E-02	4.00E-02	5.772
ENSGACG00000010218	gata1a	1.51E-03	3.03E-03	10.856
ENSGACG00000015542	LOCENSGACG00000015542	8.65E-03	1.73E-02	7.416
ENSGACG00000005143	col1a1a	5.37E-04	1.07E-03	12.912
ENSGACG00000018619	il6st	3.97E-02	7.94E-02	4.435
ENSGACG00000014732	penka	3.16E-02	6.33E-02	4.876
ENSGACG00000016426	LOCENSGACG00000016426	4.59E-02	9.18E-02	4.153
ENSGACG00000011376	plxnb1b	1.07E-03	2.15E-03	11.537
ENSGACG00000006690	TNFRSF11B	2.58E-02	5.15E-02	5.277

ENSGACG00000003940	ufd1l	3.28E-02	6.56E-02	4.807
ENSGACG00000009412	LOCENSGACG00000009412	1.94E-04	3.87E-04	14.935
ENSGACG00000016814	slc26a2	4.17E-03	8.34E-03	8.853
ENSGACG00000012223	trip11	1.35E-02	2.71E-02	6.537
ENSGACG00000008029	PCSK5	4.34E-02	8.68E-02	4.262
ENSGACG00000011806	LOCENSGACG00000011806	2.28E-02	4.57E-02	5.512
ENSGACG00000004355	EIF2AK2	6.96E-04	1.39E-03	12.397
ENSGACG00000006819	col2a1b	1.50E-02	3.00E-02	6.337
ENSGACG00000004632	KLF10	4.31E-06	8.63E-06	22.504
ENSGACG00000019023	actn3a	2.03E-02	4.06E-02	5.740
ENSGACG00000010790	six2a	4.38E-03	8.76E-03	8.757
ENSGACG00000014956	eng1b	2.25E-02	4.51E-02	5.539
ENSGACG00000018018	NPR2	9.96E-03	1.99E-02	7.139
ENSGACG00000000260	itgav	1.89E-02	3.79E-02	5.877
ENSGACG00000002837	sulf1	1.84E-02	3.69E-02	5.930
ENSGACG00000009713	matn3a	1.35E-02	2.69E-02	6.548
ENSGACG00000006853	usp1	2.92E-02	5.83E-02	5.035
ENSGACG00000006537	mcph1	1.49E-02	2.97E-02	6.353
ENSGACG00000017894	spp1	2.59E-02	5.19E-02	5.264
ENSGACG00000008948	itgb3a	1.01E-03	2.01E-03	11.666
ENSGACG00000014564	hdac4	3.97E-02	7.93E-02	4.437
ENSGACG00000016287	CSGALNACT1	3.48E-02	6.95E-02	4.694
ENSGACG00000012231	LOCENSGACG00000012231	4.13E-04	8.27E-04	13.430
ENSGACG00000017458	lef1	3.11E-02	6.22E-02	4.909
ENSGACG00000018036	slc34a1a	4.85E-02	9.71E-02	4.046
ENSGACG00000020663	rry1a	1.72E-02	3.45E-02	6.064
ENSGACG00000018814	ano6	1.44E-02	2.88E-02	6.416
ENSGACG00000016719	col11a1b	3.84E-03	7.69E-03	9.014
ENSGACG00000004416	eif2ak3	2.15E-02	4.30E-02	5.629
ENSGACG00000015182	NCAN (1 of many)	9.69E-03	1.94E-02	7.193
ENSGACG00000016060	ddx21	2.79E-02	5.59E-02	5.119
ENSGACG00000016773	FBN1	1.15E-02	2.30E-02	6.857
ENSGACG00000015511	jund	4.29E-02	8.59E-02	4.284
ENSGACG00000012007	mst1rb	1.62E-02	3.24E-02	6.184
ENSGACG00000004964	nf1b	3.70E-03	7.40E-03	9.090
ENSGACG00000020221	spns2	8.60E-03	1.72E-02	7.428
ENSGACG00000011567	LOCENSGACG00000011567	2.97E-02	5.94E-02	4.999
ENSGACG00000003402	foxp1a	4.78E-02	9.57E-02	4.074
ENSGACG00000008971	isg15	2.29E-07	4.58E-07	28.358
ENSGACG00000010238	igsf10	3.86E-02	7.71E-02	4.491

ENSGACG00000016670	mmp2	1.69E-02	3.38E-02	6.103
ENSGACG00000008267	nog2	1.56E-03	3.12E-03	10.795
ENSGACG00000006511	col1a2	7.06E-04	1.41E-03	12.367
ENSORLG00000002037	LOCENSORLG00000002037	5.35E-03	1.07E-02	8.363

+Select gene ontology IDs associated with bone development and mineralization (637 genes total)

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 GO:0060351,GO:0070168,GO:0072674,GO:0072675,GO:0090290,GO:0090291,GO:1900158  
 GO:1900159

**Supplementary Table 6. Accelerated sequence evolution in bone-associated<sup>†</sup> genes in Eleginopsioidea**

Ensembl Ortholog	Gene	P	P (Bonferroni)	LRT
ENSGACG00000014564	hdac4	0.00E+00	0.00E+00	45.267
ENSGACG00000002204	fasn	0.00E+00	0.00E+00	37.930
ENSGACG00000003893	shhb	0.00E+00	0.00E+00	28.550
ENSGACG00000003678	zic1	0.00E+00	0.00E+00	21.956
ENSGACG00000008942	deaf1	0.00E+00	0.00E+00	20.953
ENSGACG00000014956	eng1b	0.00E+00	0.00E+00	20.861
ENSGACG00000003838	si:ch211-285f17.1	0.00E+00	0.00E+00	18.399
ENSGACG00000014897	CHD7	0.00E+00	0.00E+00	17.048
ENSGACG00000011376	plxnb1b	0.00E+00	0.00E+00	15.932
ENSGACG00000017481	gtpbp4	0.00E+00	0.00E+00	15.515
ENSGACG00000003906	npr3	0.00E+00	0.00E+00	14.273
ENSGACG00000016083	notch1a	0.00E+00	0.00E+00	12.690
ENSGACG00000020196	wnt11r	0.00E+00	0.00E+00	11.440
ENSGACG00000020771	zbtb16a	1.00E-05	2.00E-05	9.754
ENSGACG00000005297	rarab	1.00E-05	2.00E-05	9.003
ENSGACG00000020663	ryr1a	1.00E-05	2.00E-05	8.860
ENSGACG00000016835	nipblb	5.00E-05	1.00E-04	7.520
ENSGACG00000011665	syncrip	5.00E-05	1.00E-04	7.517
ENSGACG00000007999	RARB	7.00E-05	1.40E-04	7.286
ENSORLG00000009503	chd	1.70E-04	3.40E-04	6.433
ENSGACG00000016814	slc26a2	1.70E-04	3.40E-04	6.426
ENSGACG00000005810	ACVR1	2.30E-04	4.60E-04	6.129
ENSGACG00000020010	tmem178	2.50E-04	5.00E-04	6.047
ENSGACG00000010046	id1	3.10E-04	6.20E-04	5.863
ENSGACG00000008814	tmem119a	4.00E-04	8.00E-04	5.626
ENSGACG00000016310	sh3pxd2aa	4.50E-04	9.00E-04	5.520
ENSGACG00000006853	usp1	4.60E-04	9.20E-04	5.488
ENSGACG00000016468	foxc1a	4.70E-04	9.40E-04	5.465
ENSGACG00000005279	gdf6a	4.80E-04	9.60E-04	5.445
ENSGACG00000008289	trim45	7.10E-04	1.42E-03	5.088
ENSGACG00000010282	minpp1b	8.50E-04	1.70E-03	4.924
ENSGACG00000006287	ufl1	9.50E-04	1.90E-03	4.820
ENSGACG00000020147	osr1	1.11E-03	2.22E-03	4.678
ENSGACG00000018575	hmwcs1	1.43E-03	2.86E-03	4.448
ENSGACG00000005508	sfrp1a	1.46E-03	2.92E-03	4.425

ENSGACG00000010238	igsf10	2.06E-03	4.12E-03	4.116
ENSGACG00000004632	KLF10	2.07E-03	4.14E-03	4.113
ENSGACG00000012411	SMOC1	2.15E-03	4.30E-03	4.076
ENSGACG00000020486	pafah1b1b	2.18E-03	4.36E-03	4.066
ENSGACG00000014186	fgf3	2.31E-03	4.62E-03	4.010
ENSGACG00000003402	foxp1a	2.45E-03	4.90E-03	3.960
ENSGACG00000009557	alx1	2.62E-03	5.24E-03	3.899
ENSGACG00000004931	dlx1a	2.82E-03	5.64E-03	3.831
ENSGACG00000004834	gli3	3.34E-03	6.68E-03	3.680
ENSGACG00000011493	si:dkey-42p8.3	3.39E-03	6.78E-03	3.666
ENSGACG00000014119	acp2	3.63E-03	7.26E-03	3.604
ENSGACG00000005488	phex	3.90E-03	7.80E-03	3.540
ENSGACG00000007912	smurf1	3.94E-03	7.88E-03	3.531
ENSGACG00000012407	ercc2	4.26E-03	8.52E-03	3.461
ENSGACG00000004764	DNAJC13	4.62E-03	9.24E-03	3.387
ENSGACG00000015083	acvr2ab	4.80E-03	9.60E-03	3.354
ENSGACG00000008948	itgb3a	4.96E-03	9.92E-03	3.325
ENSGACG00000009396	hoxc9a	7.28E-03	1.46E-02	2.984
ENSGACG00000007341	ireb2	7.42E-03	1.48E-02	2.968
ENSGACG00000009872	kat7b	7.75E-03	1.55E-02	2.930
ENSGACG00000013817	BMPR2	8.04E-03	1.61E-02	2.897
ENSGACG00000012967	MATN3	8.53E-03	1.71E-02	2.845
ENSGACG00000012822	LOCENSGACG00000012822	8.89E-03	1.78E-02	2.809
ENSGACG00000007795	inpl1a	9.54E-03	1.91E-02	2.747
ENSGACG00000004680	sema7a	9.61E-03	1.92E-02	2.741

†Select gene ontology IDs associated with bone development and mineralization (637 genes total)

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 GO:1900159

**Supplementary Table 7. Positive selection in bone-associated<sup>†</sup> genes in Channichthyidae**

Ensembl Ortholog	Gene	P	P (Bonferroni)	LRT
ENSGACG00000014066	LRRK1	0.00E+00	0.00E+00	101.842
ENSGACG00000010790	six2a	2.65E-10	5.29E-10	41.853
ENSGACG00000010565	tgfb3	7.14E-06	1.43E-05	21.500
ENSGACG00000016783	tbx3a	8.11E-06	1.62E-05	21.248
ENSGACG00000017894	spp1	2.50E-04	5.00E-04	14.429
ENSGACG00000011014	aak1a	4.19E-04	8.38E-04	13.403
ENSGACG00000005143	col1a1a	7.30E-04	1.46E-03	12.301
ENSGACG00000012822	LOCENSGACG00000012822	1.37E-03	2.75E-03	11.048
ENSGACG00000007087	LIMD1	1.85E-03	3.71E-03	10.455
ENSGACG00000010042	BCAP29	3.20E-03	6.40E-03	9.376
ENSGACG00000016860	mfge8a	4.62E-03	9.23E-03	8.654
ENSGACG00000017255	inpp4b	4.76E-03	9.52E-03	8.593
ENSGACG00000012007	mst1rb	5.49E-03	1.10E-02	8.310
ENSGACG00000016468	foxc1a	6.33E-03	1.27E-02	8.030
ENSGACG00000013533	tfa	8.34E-03	1.67E-02	7.487
ENSGACG00000000393	il19l	1.19E-02	2.39E-02	6.783
ENSGACG00000003946	gpm6bb	1.24E-02	2.47E-02	6.714
ENSGACG00000016773	FBN1	1.33E-02	2.65E-02	6.577
ENSGACG00000020771	zbtb16a	1.82E-02	3.64E-02	5.958
ENSGACG00000011694	axin2	1.95E-02	3.89E-02	5.824
ENSGACG00000008971	isg15	2.40E-02	4.80E-02	5.416
ENSGACG00000011781	ocstamp	3.01E-02	6.02E-02	4.975
ENSGACG00000009662	NA	3.13E-02	6.26E-02	4.898
ENSORLG00000002037	LOCENSORLG00000002037	3.85E-02	7.70E-02	4.495
ENSGACG00000004134	chad	4.95E-02	9.89E-02	4.009

<sup>†</sup>Select gene ontology IDs associated with bone development and mineralization (637 genes total)

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 GO:0030501, GO:0030502, GO:0031214, GO:0033687, GO:0033688, GO:0033689, GO:0033690  
 GO:0035630, GO:0036035, GO:0036179, GO:0043931, GO:0043932, GO:0044339, GO:0045667  
 GO:0045668, GO:0045669, GO:0045670, GO:0045671, GO:0045672, GO:0045778, GO:0048706  
 GO:0060351, GO:0070168, GO:0072674, GO:0072675, GO:0090290, GO:0090291, GO:1900158  
 GO:1900159

**Supplementary Table 8. Accelerated sequence evolution in bone-associated<sup>†</sup> genes in Channichthyidae**

Ensembl Ortholog	Gene	P	P (Bonferroni)	LRT
ENSGACG00000018025	gdf2	0.00E+00	0.00E+00	44.466
ENSGACG00000014066	LRRK1	0.00E+00	0.00E+00	18.742
ENSGACG00000010790	six2a	0.00E+00	0.00E+00	16.153
ENSGACG00000020111	BBX	0.00E+00	0.00E+00	16.044
ENSGACG00000012007	mst1rb	0.00E+00	0.00E+00	10.213
ENSGACG00000012223	trip11	1.00E-05	2.00E-05	9.635
ENSGACG00000005143	col1a1a	1.00E-05	2.00E-05	9.225
ENSGACG00000011839	FOXP1	1.00E-05	2.00E-05	8.772
ENSGACG00000003352	sec22c	2.00E-05	4.00E-05	8.542
ENSGACG00000018575	hmgcs1	4.00E-05	8.00E-05	7.720
ENSGACG00000014838	ptk2ba	6.00E-05	1.20E-04	7.335
ENSGACG00000018814	ano6	7.00E-05	1.40E-04	7.308
ENSGACG00000005830	prpsap2	1.40E-04	2.80E-04	6.617
ENSGACG00000016287	CSGALNACT1 (2 of 2)	1.60E-04	3.20E-04	6.447
ENSGACG00000014536	xpr1b	1.70E-04	3.40E-04	6.423
ENSGACG00000011843	cdh11	6.30E-04	1.26E-03	5.194
ENSGACG00000007087	LIMD1	6.50E-04	1.30E-03	5.165
ENSGACG00000014186	fgf3	8.90E-04	1.78E-03	4.878
ENSGACG00000018497	AXIN2 (1 of many)	1.16E-03	2.32E-03	4.640
ENSGACG00000013007	skia	1.82E-03	3.64E-03	4.227
ENSGACG00000016062	col27a1a	2.59E-03	5.18E-03	3.907
ENSGACG00000016083	notch1a	2.80E-03	5.60E-03	3.838
ENSGACG00000011349	src	3.32E-03	6.64E-03	3.684
ENSGACG00000019404	psmc2	3.69E-03	7.38E-03	3.589
ENSGACG00000001547	gli2a	3.96E-03	7.92E-03	3.525
ENSGACG00000014550	txlNg	4.58E-03	9.16E-03	3.395
ENSGACG00000007522	jag2b	6.46E-03	1.29E-02	3.090
ENSGACG00000016016	fam73b	6.48E-03	1.30E-02	3.087

<sup>†</sup>Select gene ontology IDs associated with bone development and mineralization (637 genes total)

GO:0001501, GO:0001503, GO:0001649, GO:0001957, GO:0001958, GO:0002051, GO:0002076  
 GO:0002158, GO:0003433, GO:0030278, GO:0030279, GO:0030282, GO:0030316, GO:0030500  
 GO:0030501, GO:0030502, GO:0031214, GO:0033687, GO:0033688, GO:0033689, GO:0033690  
 GO:0035630, GO:0036035, GO:0036179, GO:0043931, GO:0043932, GO:0044339, GO:0045667  
 GO:0045668, GO:0045669, GO:0045670, GO:0045671, GO:0045672, GO:0045778, GO:0048706  
 GO:0060351, GO:0070168, GO:0072674, GO:0072675, GO:0090290, GO:0090291, GO:1900158  
 GO:1900159

**Supplementary Table 9. Tissue samples used in this study**

Family	Species	Source	ID#
Artedidraconidae	<i>Artedidraco skottsbergi</i>	No voucher	YFTC 4131
Artedidraconidae	<i>Artedidraco skottsbergi</i>	No voucher	YFTC 4132
Artedidraconidae	<i>Artedidraco skottsbergi</i>	YPM ICH 016389	YFTC 7804
Artedidraconidae	<i>Artedidraco skottsbergi</i>	YPM ICH 016389	YFTC 7805
Artedidraconidae	<i>Artedidraco skottsbergi</i>	YPM ICH 022479	YFTC 15048
Artedidraconidae	<i>Dolloidraco longedorsalis</i>	Detrich lab	HWD 53
Artedidraconidae	<i>Dolloidraco longedorsalis</i>	Detrich lab	HWD 54
Artedidraconidae	<i>Dolloidraco longedorsalis</i>	Detrich lab	HWD 55
Artedidraconidae	<i>Dolloidraco longedorsalis</i>	Detrich lab	HWD 56
Artedidraconidae	<i>Dolloidraco longedorsalis</i>	Detrich lab	HWD 57
Artedidraconidae	<i>Histiodraco velifer</i>	No voucher	YFTC 2049
Artedidraconidae	<i>Histiodraco velifer</i>	No voucher	YFTC 4135
Artedidraconidae	<i>Histiodraco velifer</i>	No voucher	YFTC 4136
Artedidraconidae	<i>Histiodraco velifer</i>	No voucher	YFTC 13801
Artedidraconidae	<i>Histiodraco velifer</i>	No voucher	YFTC 14514
Artedidraconidae	<i>Pogonophryne barsukovi</i>	YPM ICH 022357	YFTC 15530
Artedidraconidae	<i>Pogonophryne barsukovi</i>	YPM ICH 022357	YFTC 15531
Artedidraconidae	<i>Pogonophryne barsukovi</i>	YPM ICH 022357	YFTC 15532
Artedidraconidae	<i>Pogonophryne barsukovi</i>	YPM ICH 022357	YFTC 15533
Artedidraconidae	<i>Pogonophryne barsukovi</i>	YPM ICH 022357	YFTC 15534
Artedidraconidae	<i>Pogonophryne scotti</i>	YPM ICH 022357	YFTC 15105
Artedidraconidae	<i>Pogonophryne scotti</i>	YPM ICH 022553	YFTC 15106
Artedidraconidae	<i>Pogonophryne scotti</i>	YPM ICH 022553	YFTC 15107
Artedidraconidae	<i>Pogonophryne scotti</i>	YPM ICH 022553	YFTC 15108
Artedidraconidae	<i>Pogonophryne scotti</i>	YPM ICH 022553	YFTC 15109
Bathydraconidae	<i>Akarotaxis nudiceps</i>	YPM ICH 024046	YFTC 20912
Bathydraconidae	<i>Akarotaxis nudiceps</i>	YPM ICH 024238	YFTC 20924
Bathydraconidae	<i>Akarotaxis nudiceps</i>	YPM ICH 024118	YFTC 20928
Bathydraconidae	<i>Akarotaxis nudiceps</i>	YPM ICH 024118	YFTC 20929
Bathydraconidae	<i>Akarotaxis nudiceps</i>	YPM ICH 024118	YFTC 20930
Bathydraconidae	<i>Bathydraco marri</i>	NMNZ 043393	YFTC 13884
Bathydraconidae	<i>Bathydraco marri</i>	NMNZ 043394	YFTC 13885
Bathydraconidae	<i>Bathydraco marri</i>	NMNZ 043553	YFTC 13886
Bathydraconidae	<i>Bathydraco marri</i>	NMNZ 043634	YFTC 13887
Bathydraconidae	<i>Bathydraco marri</i>	NMNZ 043635	YFTC 13888
Bathydraconidae	<i>Gerlachea australis</i>	NMNZ 043338	YFTC 13898

Bathyraconidae	<i>Gerlachea australis</i>	NMNZ 043495	YFTC 13899
Bathyraconidae	<i>Gerlachea australis</i>	NMNZ 043517	YFTC 13900
Bathyraconidae	<i>Gerlachea australis</i>	NMNZ 043518	YFTC 13901
Bathyraconidae	<i>Gerlachea australis</i>	NMNZ 043545	YFTC 13902
Bathyraconidae	<i>Gymnodraco acuticeps</i>	Detrich lab	HWD 6
Bathyraconidae	<i>Gymnodraco acuticeps</i>	Detrich lab	HWD 7
Bathyraconidae	<i>Gymnodraco acuticeps</i>	Detrich lab	HWD 8
Bathyraconidae	<i>Gymnodraco acuticeps</i>	Detrich lab	HWD 9
Bathyraconidae	<i>Gymnodraco acuticeps</i>	Detrich lab	HWD 10
Bathyraconidae	<i>Gymnodraco acuticeps</i>	Detrich lab	HWD 11
Bathyraconidae	<i>Parachaenichthys charcoti</i>	Detrich lab	HWD 24
Bathyraconidae	<i>Parachaenichthys charcoti</i>	Detrich lab	HWD 25
Bathyraconidae	<i>Parachaenichthys charcoti</i>	Detrich lab	HWD 26
Bathyraconidae	<i>Vomeridens infuscipinnis</i>	YPM ICH 020008	YFTC 12880
Bathyraconidae	<i>Vomeridens infuscipinnis</i>	YPM ICH 020009	YFTC 12881
Bathyraconidae	<i>Vomeridens infuscipinnis</i>	YPM ICH 020041	YFTC 12884
Bathyraconidae	<i>Vomeridens infuscipinnis</i>	YPM ICH 020025	YFTC 12882
Bathyraconidae	<i>Vomeridens infuscipinnis</i>	YPM ICH 020061	YFTC 12919
Bovichtidae	<i>Bovichtus diacanthus</i>	YPM ICH 021534	YFTC 3482
Bovichtidae	<i>Bovichtus diacanthus</i>	YPM ICH 021534	YFTC 3483
Bovichtidae	<i>Bovichtus diacanthus</i>	YPM ICH 021534	YFTC 3484
Bovichtidae	<i>Bovichtus diacanthus</i>	YPM ICH 021534	YFTC 3485
Bovichtidae	<i>Bovichtus diacanthus</i>	YPM ICH 021534	YFTC 3486
Bovichtidae	<i>Cottoperca trigloides</i>	Detrich lab	HWD 71
Bovichtidae	<i>Cottoperca trigloides</i>	Detrich lab	HWD 72
Bovichtidae	<i>Cottoperca trigloides</i>	Detrich lab	HWD 73
Bovichtidae	<i>Cottoperca trigloides</i>	Detrich lab	HWD 74
Bovichtidae	<i>Cottoperca trigloides</i>	Detrich lab	HWD 75
Channichthyidae	<i>Chaenocephalus aceratus</i>	Detrich lab	HWD 1
Channichthyidae	<i>Chaenocephalus aceratus</i>	Detrich lab	HWD 2
Channichthyidae	<i>Chaenocephalus aceratus</i>	Detrich lab	HWD 3
Channichthyidae	<i>Chaenocephalus aceratus</i>	Detrich lab	HWD 4
Channichthyidae	<i>Chaenocephalus aceratus</i>	Detrich lab	HWD 5
Channichthyidae	<i>Chaenodraco wilsoni</i>	Detrich lab	HWD 76
Channichthyidae	<i>Chaenodraco wilsoni</i>	Detrich lab	HWD 77
Channichthyidae	<i>Chaenodraco wilsoni</i>	Detrich lab	HWD 78
Channichthyidae	<i>Chaenodraco wilsoni</i>	Detrich lab	HWD 79
Channichthyidae	<i>Chaenodraco wilsoni</i>	Detrich lab	HWD 80
Channichthyidae	<i>Champscephalus esox</i>	Detrich lab	HWD 41
Channichthyidae	<i>Champscephalus esox</i>	Detrich lab	HWD 42

Channichthyidae	<i>Champscephalus gunnari</i>	YPM ICH 022626	YFTC 14750
Channichthyidae	<i>Champscephalus gunnari</i>	YPM ICH 022626	YFTC 14751
Channichthyidae	<i>Champscephalus gunnari</i>	YPM ICH 022626	YFTC 14752
Channichthyidae	<i>Champscephalus gunnari</i>	YPM ICH 022626	YFTC 14753
Channichthyidae	<i>Champscephalus gunnari</i>	YPM ICH 022626	YFTC 14754
Channichthyidae	<i>Chionobathyscus dewitti</i>	Detrich lab	HWD 32
Channichthyidae	<i>Chionobathyscus dewitti</i>	Detrich lab	HWD 33
Channichthyidae	<i>Chionobathyscus dewitti</i>	Detrich lab	HWD 34
Channichthyidae	<i>Chionobathyscus dewitti</i>	Detrich lab	HWD 35
Channichthyidae	<i>Cryodraco antarcticus</i>	No voucher	YFTC 11045
Channichthyidae	<i>Cryodraco antarcticus</i>	No voucher	YFTC 11046
Channichthyidae	<i>Cryodraco antarcticus</i>	No voucher	YFTC 11047
Channichthyidae	<i>Cryodraco antarcticus</i>	Detrich lab	HWD 81
Channichthyidae	<i>Cryodraco antarcticus</i>	Detrich lab	HWD 82
Channichthyidae	<i>Cryodraco antarcticus</i>	Detrich lab	HWD 83
Channichthyidae	<i>Dacodraco hunteri</i>	NMNZ 043399	YFTC 16112
Channichthyidae	<i>Dacodraco hunteri</i>	NMNZ 043430	YFTC 16113
Channichthyidae	<i>Dacodraco hunteri</i>	NMNZ 043450	YFTC 16114
Channichthyidae	<i>Dacodraco hunteri</i>	NMNZ 043496	YFTC 16115
Channichthyidae	<i>Neopagetopsis ionah</i>	NMNZ 043314	YFTC 16116
Channichthyidae	<i>Neopagetopsis ionah</i>	NMNZ 043315	YFTC 16117
Channichthyidae	<i>Neopagetopsis ionah</i>	NMNZ 043316	YFTC 16118
Channichthyidae	<i>Neopagetopsis ionah</i>	NMNZ 043358	YFTC 16119
Channichthyidae	<i>Neopagetopsis ionah</i>	NMNZ 043378	YFTC 16120
Channichthyidae	<i>Pagetopsis macropterus</i>	YPM ICH 016490	YFTC 7775
Channichthyidae	<i>Pagetopsis macropterus</i>	YPM ICH 016490	YFTC 7776
Channichthyidae	<i>Pagetopsis macropterus</i>	YPM ICH 016490	YFTC 7777
Channichthyidae	<i>Pagetopsis macropterus</i>	YPM ICH 016490	YFTC 7778
Channichthyidae	<i>Pagetopsis macropterus</i>	YPM ICH 016490	YFTC 7779
Channichthyidae	<i>Pseudochaenichthys georgianus</i>	Detrich lab	HWD 43
Channichthyidae	<i>Pseudochaenichthys georgianus</i>	Detrich lab	HWD 44
Channichthyidae	<i>Pseudochaenichthys georgianus</i>	Detrich lab	HWD 45
Channichthyidae	<i>Pseudochaenichthys georgianus</i>	Detrich lab	HWD 46
Channichthyidae	<i>Pseudochaenichthys georgianus</i>	Detrich lab	HWD 47
Eleginopsidae	<i>Eleginops maclovinus</i>	Detrich lab	HWD 58
Eleginopsidae	<i>Eleginops maclovinus</i>	Detrich lab	HWD 59
Eleginopsidae	<i>Eleginops maclovinus</i>	Detrich lab	HWD 60
Eleginopsidae	<i>Eleginops maclovinus</i>	Detrich lab	HWD 61
Eleginopsidae	<i>Eleginops maclovinus</i>	Detrich lab	HWD 62
Eleginopsidae	<i>Eleginops maclovinus</i>	Detrich lab	HWD 63

Harpagiferidae	<i>Harpagifer antarcticus</i>	YPM ICH 016640	YFTC 4399
Harpagiferidae	<i>Harpagifer antarcticus</i>	YPM ICH 016640	YFTC 4400
Harpagiferidae	<i>Harpagifer antarcticus</i>	YPM ICH 016640	YFTC 4401
Harpagiferidae	<i>Harpagifer antarcticus</i>	YPM ICH 016640	YFTC 4402
Harpagiferidae	<i>Harpagifer antarcticus</i>	YPM ICH 016640	YFTC 4403
Nototheniidae	<i>Aethotaxis mitopteryx</i>	YPM ICH 022552	YFTC 15291
Nototheniidae	<i>Aethotaxis mitopteryx</i>	YPM ICH 022552	YFTC 15292
Nototheniidae	<i>Aethotaxis mitopteryx</i>	YPM ICH 022552	YFTC 15293
Nototheniidae	<i>Aethotaxis mitopteryx</i>	YPM ICH 022552	YFTC 15294
Nototheniidae	<i>Aethotaxis mitopteryx</i>	YPM ICH 022552	YFTC 15295
Nototheniidae	<i>Dissostichus eleginoides</i>	Detrich lab	HWD 36
Nototheniidae	<i>Dissostichus eleginoides</i>	Detrich lab	HWD 37
Nototheniidae	<i>Dissostichus eleginoides</i>	Detrich lab	HWD 38
Nototheniidae	<i>Dissostichus eleginoides</i>	Detrich lab	HWD 39
Nototheniidae	<i>Dissostichus eleginoides</i>	Detrich lab	HWD 40
Nototheniidae	<i>Dissostichus mawsoni</i>	YPM ICH 022349	YFTC 15462
Nototheniidae	<i>Dissostichus mawsoni</i>	YPM ICH 022397	YFTC 15346
Nototheniidae	<i>Dissostichus mawsoni</i>	YPM ICH 022397	YFTC 15347
Nototheniidae	<i>Dissostichus mawsoni</i>	YPM ICH 022397	YFTC 15348
Nototheniidae	<i>Dissostichus mawsoni</i>	YPM ICH 022397	YFTC 15349
Nototheniidae	<i>Gobionotothen gibberifrons</i>	No voucher	YFTC 12759
Nototheniidae	<i>Gobionotothen gibberifrons</i>	No voucher	YFTC 12760
Nototheniidae	<i>Gobionotothen gibberifrons</i>	No voucher	YFTC 12761
Nototheniidae	<i>Gobionotothen gibberifrons</i>	No voucher	YFTC 12762
Nototheniidae	<i>Gobionotothen gibberifrons</i>	No voucher	YFTC 12763
Nototheniidae	<i>Nototheniops nudifrons</i>	YPM ICH 020081	YFTC 3809
Nototheniidae	<i>Nototheniops nudifrons</i>	YPM ICH 020081	YFTC 3810
Nototheniidae	<i>Nototheniops nudifrons</i>	YPM ICH 020081	YFTC 3811
Nototheniidae	<i>Nototheniops nudifrons</i>	YPM ICH 020081	YFTC 3812
Nototheniidae	<i>Nototheniops nudifrons</i>	YPM ICH 020081	YFTC 3813
Nototheniidae	<i>Lepidonotothen squamifrons</i>	YPM ICH 028210	YFTC 14785
Nototheniidae	<i>Lepidonotothen squamifrons</i>	YPM ICH 028210	YFTC 14786
Nototheniidae	<i>Lepidonotothen squamifrons</i>	YPM ICH 022548	YFTC 15065
Nototheniidae	<i>Lepidonotothen squamifrons</i>	YPM ICH 022548	YFTC 15066
Nototheniidae	<i>Lepidonotothen squamifrons</i>	YPM ICH 022548	YFTC 15067
Nototheniidae	<i>Paranotothenia angustata</i>	Detrich lab	HWD 12
Nototheniidae	<i>Paranotothenia angustata</i>	Detrich lab	HWD 13
Nototheniidae	<i>Paranotothenia angustata</i>	Detrich lab	HWD 14
Nototheniidae	<i>Paranotothenia angustata</i>	Detrich lab	HWD 15
Nototheniidae	<i>Paranotothenia angustata</i>	Detrich lab	HWD 16

Nototheniidae	<i>Notothenia coriiceps</i>	Detrich lab	HWD 27
Nototheniidae	<i>Notothenia coriiceps</i>	Detrich lab	HWD 28
Nototheniidae	<i>Notothenia coriiceps</i>	Detrich lab	HWD 29
Nototheniidae	<i>Notothenia coriiceps</i>	Detrich lab	HWD 30
Nototheniidae	<i>Notothenia coriiceps</i>	Detrich lab	HWD 31
Nototheniidae	<i>Notothenia rossii</i>	Detrich lab	HWD 64
Nototheniidae	<i>Notothenia rossii</i>	Detrich lab	HWD 65
Nototheniidae	<i>Notothenia rossii</i>	Detrich lab	HWD 66
Nototheniidae	<i>Notothenia rossii</i>	Detrich lab	HWD 67
Nototheniidae	<i>Notothenia rossii</i>	Detrich lab	HWD 68
Nototheniidae	<i>Notothenia rossii</i>	Detrich lab	HWD 69
Nototheniidae	<i>Notothenia rossii</i>	Detrich lab	HWD 70
Nototheniidae	<i>Trematomus borchgrevinki</i>	No voucher	YFTC 12070
Nototheniidae	<i>Trematomus borchgrevinki</i>	No voucher	YFTC 12071
Nototheniidae	<i>Trematomus borchgrevinki</i>	No voucher	YFTC 12072
Nototheniidae	<i>Trematomus borchgrevinki</i>	No voucher	YFTC 13638
Nototheniidae	<i>Patagonotothen squamiceps</i>	No voucher	YFTC 24046
Nototheniidae	<i>Patagonotothen squamiceps</i>	No voucher	YFTC 24047
Nototheniidae	<i>Patagonotothen squamiceps</i>	No voucher	YFTC 24048
Nototheniidae	<i>Patagonotothen squamiceps</i>	No voucher	YFTC 24049
Nototheniidae	<i>Patagonotothen squamiceps</i>	No voucher	YFTC 24050
Nototheniidae	<i>Patagonotothen guntheri</i>	No voucher	YFTC 24057
Nototheniidae	<i>Patagonotothen guntheri</i>	No voucher	YFTC 24058
Nototheniidae	<i>Patagonotothen guntheri</i>	No voucher	YFTC 24059
Nototheniidae	<i>Patagonotothen guntheri</i>	No voucher	YFTC 24060
Nototheniidae	<i>Patagonotothen guntheri</i>	No voucher	YFTC 24061
Nototheniidae	<i>Pleuragramma antarctica</i>	YPM ICH 022586	YFTC 15002
Nototheniidae	<i>Pleuragramma antarctica</i>	YPM ICH 022586	YFTC 15003
Nototheniidae	<i>Pleuragramma antarctica</i>	YPM ICH 022586	YFTC 15004
Nototheniidae	<i>Pleuragramma antarctica</i>	YPM ICH 022586	YFTC 15005
Nototheniidae	<i>Pleuragramma antarctica</i>	YPM ICH 022586	YFTC 15006
Nototheniidae	<i>Trematomus bernacchii</i>	Detrich lab	HWD 17
Nototheniidae	<i>Trematomus bernacchii</i>	Detrich lab	HWD 18
Nototheniidae	<i>Trematomus bernacchii</i>	Detrich lab	HWD 19
Nototheniidae	<i>Trematomus bernacchii</i>	Detrich lab	HWD 20
Nototheniidae	<i>Trematomus bernacchii</i>	Detrich lab	HWD 21
Nototheniidae	<i>Trematomus bernacchii</i>	Detrich lab	HWD 22
Nototheniidae	<i>Trematomus bernacchii</i>	Detrich lab	HWD 23
Nototheniidae	<i>Trematomus eulepidotus</i>	YPM ICH 022564	YFTC 15403
Nototheniidae	<i>Trematomus eulepidotus</i>	YPM ICH 022564	YFTC 15404

Nototheniidae	<i>Trematomus eulepidotus</i>	YPM ICH 022564	YFTC 15405
Nototheniidae	<i>Trematomus eulepidotus</i>	YPM ICH 022564	YFTC 15406
Nototheniidae	<i>Trematomus eulepidotus</i>	YPM ICH 022564	YFTC 15407
Nototheniidae	<i>Trematomus hansonii</i>	Detrich lab	HWD 48
Nototheniidae	<i>Trematomus hansonii</i>	Detrich lab	HWD 49
Nototheniidae	<i>Trematomus hansonii</i>	Detrich lab	HWD 50
Nototheniidae	<i>Trematomus hansonii</i>	Detrich lab	HWD 51
Nototheniidae	<i>Trematomus hansonii</i>	Detrich lab	HWD 52
Nototheniidae	<i>Trematomus newnesi</i>	YPM ICH 016464	YFTC 7708
Nototheniidae	<i>Trematomus newnesi</i>	YPM ICH 016464	YFTC 7709
Nototheniidae	<i>Trematomus newnesi</i>	YPM ICH 016464	YFTC 7710
Nototheniidae	<i>Trematomus newnesi</i>	YPM ICH 016464	YFTC 7711
Nototheniidae	<i>Trematomus newnesi</i>	YPM ICH 016464	YFTC 7712
Nototheniidae	<i>Trematomus scotti</i>	YPM ICH 024115	YFTC 20940
Nototheniidae	<i>Trematomus scotti</i>	YPM ICH 024115	YFTC 20941
Nototheniidae	<i>Trematomus scotti</i>	YPM ICH 024115	YFTC 20942
Nototheniidae	<i>Trematomus scotti</i>	YPM ICH 024115	YFTC 20943
Nototheniidae	<i>Trematomus scotti</i>	YPM ICH 024115	YFTC 20944
Percidae	<i>Percina caprodes</i>	YPM ICH 027375	YFTC 24610
Percidae	<i>Percina caprodes</i>	YPM ICH 027375	YFTC 24610
Percidae	<i>Percina caprodes</i>	YPM ICH 027375	YFTC 24611
Percidae	<i>Percina caprodes</i>	YPM ICH 027375	YFTC 24612
Percidae	<i>Percina caprodes</i>	YPM ICH 027375	YFTC 24613
Percophidae	<i>Percophis brasiliensis</i>	LBP 8668	35303
Percophidae	<i>Percophis brasiliensis</i>	LBP 8654	35304
Percophidae	<i>Percophis brasiliensis</i>	LBP 8654	35305
Percophidae	<i>Percophis brasiliensis</i>	LBP 8668	35307
Percophidae	<i>Percophis brasiliensis</i>	LBP 19527	73365
Percophidae	<i>Percophis brasiliensis</i>	LBP 19527	73366
Percophidae	<i>Percophis brasiliensis</i>	LBP 19527	73367
Pseudaphritidae	<i>Pseudaphritis urvillii</i>	CSIRO 6892-01	YFTC 16619
Pseudaphritidae	<i>Pseudaphritis urvillii</i>	CSIRO 6892-01	YFTC 16620
Pseudaphritidae	<i>Pseudaphritis urvillii</i>	CSIRO 7637-05	YFTC 16621

**Supplementary Table 10. Enrichment of genes with missing coverage in dataset**

GOTerm	Term Name	GO		# genes low cov. +	fold enrichment			FDR qvalue
		type	# genes		p-value			
GO:0043410	positive regulation of MAPK cascade	bp	92	15	4.02		4.22E-06	1.72E-03
GO:0030198	extracellular matrix organization	bp	209	30	3.54		1.89E-09	5.41E-06
GO:0016925	protein sumoylation	bp	111	15	3.33		4.27E-05	1.36E-02
GO:0006955	immune response	bp	268	29	2.67		1.70E-06	8.07E-04
GO:0006954	inflammatory response	bp	308	33	2.64		4.19E-07	2.99E-04
GO:0007166	cell surface receptor signaling pathway	bp	297	31	2.57		1.63E-06	8.07E-04
GO:0018108	peptidyl-tyrosine phosphorylation	bp	211	21	2.45		1.47E-04	3.81E-02
GO:0002376	immune system process	bp	251	24	2.36		9.61E-05	2.75E-02
GO:0016032	viral process	bp	377	35	2.29		5.23E-06	1.87E-03
GO:0007155	cell adhesion	bp	615	53	2.12		2.36E-07	2.25E-04
GO:0016504	peptidase activator activity	mf	13	5	9.48		1.07E-04	1.01E-02
GO:0005031	tumor necrosis factor-activated receptor activity	mf	23	6	6.43		2.45E-04	2.08E-02
GO:0005201	extracellular matrix structural constituent transmembrane	mf	86	21	6.02		2.27E-11	9.65E-09
GO:0004714	receptor protein tyrosine kinase activity	mf	89	13	3.60		6.03E-05	6.41E-03
GO:0005198	structural molecule activity	mf	230	21	2.25		4.76E-04	3.38E-02
GO:0003779	actin binding	mf	464	38	2.02		3.64E-05	4.43E-03
GO:0000800	lateral element	cc	13	4	7.58		1.44E-03	2.48E-02
GO:0005771	multivesicular body	cc	22	6	6.72		1.87E-04	5.02E-03
GO:0005640	nuclear outer membrane	cc	22	5	5.60		1.61E-03	2.70E-02
GO:0005581	collagen trimer	cc	111	18	4.00		5.16E-07	2.51E-05
GO:0005604	basement membrane	cc	92	14	3.75		1.98E-05	6.23E-04
GO:0030175	filopodium	cc	69	10	3.57		4.47E-04	9.58E-03
GO:0005578	proteinaceous extracellular matrix	cc	371	46	3.06		1.80E-11	2.41E-09
GO:0005814	centriole	cc	107	13	2.99		3.97E-04	8.87E-03
GO:0072562	blood microparticle	cc	91	11	2.98		1.13E-03	2.09E-02
GO:0009897	external side of plasma membrane	cc	166	20	2.97		1.40E-05	5.01E-04
GO:0005923	bicellular tight junction	cc	158	18	2.81		7.78E-05	2.32E-03

GO:0031225	anchored component of membrane	cc	88	10	2.80	2.98E-03	4.20E-02
GO:0043235	receptor complex	cc	116	13	2.76	8.64E-04	1.78E-02
GO:0005911	cell-cell junction	cc	193	19	2.43	3.43E-04	7.98E-03
GO:0031012	extracellular matrix	cc	322	31	2.37	8.63E-06	3.30E-04
GO:0005788	endoplasmic reticulum lumen	cc	194	17	2.16	2.45E-03	3.75E-02
GO:0009986	cell surface	cc	498	42	2.08	7.11E-06	2.93E-04
GO:0005874	microtubule	cc	357	29	2.00	3.35E-04	7.98E-03

bp – biological process, mf – molecular function, cc – cellular component

**Supplementary Table 11. Notothenioid specimens for CT scan**

Species	Catalog Number	Specimen #	STL (cm)
<i>Bovichtus variegatus</i>	YPM ICH 005976	5976	13.5
	MCZ ICH 25558	NA	13.0
<i>Eleginops maclovinus</i>	YPM ICH 009315	NA	32.0
	YPM ICH 009315	NA	25.0
<i>Dissostichus mawsoni</i>	YPM ICH 020040	12848	27.5
	MCZ ICH 195009	NA	35.0
<i>Trematomus bernacchii</i>	YPM ICH 020469	01-46	19.5
	YPM ICH 020469	01-45	15.0
<i>Gobionotothen gibberifrons</i>	YPM ICH 024029	20745	19.0
	YPM ICH 024029	20746	16.0
<i>Gymnodraco acuticeps</i>	YPM ICH 022337	15454	24.0
	YPM ICH 022337	15449	25.5
<i>Champscephalus gunnari</i>	YPM ICH 016478	07959	31.0
	YPM ICH 016478	07958	23.0