Additional file 1: analyses documentation

Table S1. List of species analysed.

Genus	**Species**	**GJPN tissue collection number**
Squalus	*acanthias*	GN6336
Cirrhigaleus	*asper*	GN3831
Centrophorus	*moluccensis*	GN8086
Deania	sp.	GN7456
Dalatias	*licha*	GN1006
Isistius	*brasiliensis*	GN3748
Squaliolus	*aliae*	GN6176
Euprotomicrus	*bispinatus*	GN3749
Somniosus	*microcephalus*	GN1156
Zameus	*squamulosus*	GN7131
Scymnodon	*ringens*	GN1703
Centroselachus	*crepidater*	GN7127
Centroscymnus	*coelolepis*	GN6626
Scymnodon	*plunketi*	GN5607
Oxynotus	*paradoxus*	GN1852
Aculeola	*nigra*	GN7450
Centroscyllium	*fabricii*	GN8195
Etmopterus	*splendidus*	GN10139
Etmopterus	*sheikoi*	GN12278
Etmopterus	cf. *molleri*	GN10014
Etmopterus	*spinax*	GN5162
Etmopterus	*gracilispinis*	GN7458
Trigonognathus	*kabeyai*	GN12280
Echinorhinus	*brucus*	GN1983
Pliotrema	*warreni*	GN7338
Pristiophorus	*japonicus*	GN1049
Squatina	*nebulosa*	GN10161
Hexanchus	*griseus*	GN2342

Table S2. Completeness statistics for captured data, raw dataset (cds = coding sequences; bp = basepairs).

Average number of genes captured:	1000	Out of 1265 possible
Completeness by cds:	74.57%	27,357 cds present out of 36,685
Completeness by position in alignment:	59.59 %	6,163,401 positions present out of 10,343,865
Average sequence length per specimen:	201,099 bp	Out of 352,605 possible

Search for phylogenetically informative loci using MARE

```
MARE OUTPUT
original multiple sequence alignment
-----------------------------------
information content of B     : 0.11
matrix saturation            : 0.746
nr of taxa                   : 29
nr of partitions             : 1264

Parameters
----------
weighting of inform. content : 3
weighting of taxa            : 1
number of evaluated quartets : 20000

reduced multiple sequence alignment
----------------------------------
information content of B'          : 0.357
nr of taxa                         : 29
nr of partitions                   : 174
ratio of reduced and complete matrix : 0.138
matrix saturation                  : 0.922
```

Figure S1: Overview of the MARE [33, 34] analysis. Rows show analysed taxa, columns different loci. Each locus is represented for each taxon by a cell. Cell colors code for the information content of loci: white cells = gene absent; red cells = treelikeness equals 0; different stages of blue to black cells = treelikeness is 0.1 – 1.

(A) Information content (treelikeness) for the full set of 1265 loci and 29 taxa.

(B) Reduced dataset after exclusion of uninformative loci (174 loci).

For a high resolution image please download Figure S1 directly from the Dryad data repository: http://datadryad.org/review?doi=doi:10.5061/dryad.n3581

PERL Script used to re-blast 174 loci resulting from the MARE analysis against the reference genome of *Callorhinchus milii* to check for multiple hits

```
#!/usr/bin/perl -w

#

# Name: reblast.pl

#

# This script is used to re-blast blastout sequences of each gene to the genome of designing baits and bin sequences into genes.

#

# Datafile: fasta file of the query and fasta files of the genes

#

# Input: the subject name and query name

#

# Output: blastn output file in format 6, sequences binned as genes.

#

# sample: ./reblast.pl -subject="Oreochromis_niloticus" -query="Gobioidei"

#

# Written by

#               Chenhong Li, Ting Kuang

#               Shanghai Ocean University, China.

#               Created on Sep 2014

#               Last modified on

use strict;

use warnings;

use Getopt::Long;   # include the module for input
```

```
my $subject; #genome sequence need make blast database
my $query; #folder containing all gene data file
my $opt = GetOptions( 'subject:s', \$subject,
                      'query:s', \$query); #set command line options

my $resultdir = $query . ".result";
`mkdir ../genebin/$resultdir`;#make dir for the output
my $resultlog = $query . ".log";
`mkdir ../genebin/$resultlog`;#make dir for the para log



#call outside program makeblastdb
#`makeblastdb -in ../subject/$subject.fas -out ../blastdb/$subject -dbtype nucl`;

my $que = "../query/$query";
opendir (DIR, $que) or die $!; #open folder containing all gene data file

while (my $file = readdir (DIR)) { #read all gene file under local alignment out folder
    next if ($file =~ /^\./); #skip files beginning with
    (my $gene) = $file =~ /(\S+)\.fas/;
    my @file = split (/\s/, $file); #split the names of gene
    chomp(@file);
    my @sortedfile = sort @file;



    foreach my $file (@sortedfile) {##call outside program blastn

        `blastn -query $que/$file -db ../blastdb/$subject -out ../blastout/$gene.blast.out.txt -word_size 7 -
gapopen 5 -gapextend 2 -penalty -1 -reward 1 -evalue 0.000001 -outfmt 6`;



        my ($BLAST_FILE, $FASTA_FILE, $RESULT_FILE, $RESULT_LOG);
        my $blastout = "../blastout";
```

```
	my $blast = "$blastout/$gene.blast.out.txt";


	#open 4 file: $new, $blast, $path and $log
	my $new = "../genebin/$resultdir/$gene.fas";
	open $RESULT_FILE, ">$new" or die ("Cannot open $new for writing ($!)");
	open $BLAST_FILE, "<$blast" or die ("Cannot open $blast for reading ($!)");
	my $log ="../genebin/$resultlog/$gene.fas";
	open $RESULT_LOG, ">$log" or die ("Cannot open $log for writing ($!)");
	my $path = "../query/$query/$file";
	open $FASTA_FILE, "<$path" or die ("Cannot open $path for reading ($!)");


	my $line = readline ($FASTA_FILE);#read line ">query$subject"
	chomp $line;
	print $RESULT_FILE "$line\n";#store line ">query$subject" to $new
	my $seqline = readline ($FASTA_FILE);#read sequence line of query$query
	chomp $seqline;
	print $RESULT_FILE "$seqline\n";##store sequence line of query$query to $new
	my %note_seq;
	while (my $newline = readline ($FASTA_FILE)) {
	    chomp $newline;
	    if ((my $speciesname) = $newline =~ /^>(\S+)/) {
	        my $newseqline = readline ($FASTA_FILE);
	        chomp $newseqline;
	        $note_seq{$speciesname} = $newseqline;
	    }
	}



	my $bline = readline ($BLAST_FILE);#read the first line "query$query" in file $blast
	chomp $bline;
	my ($species, $seqid, $pos1, $pos2) = $bline =~
/(\S+)\s+(\S+)\s+\S+\s+\S+\s+\S+\s+\S+\s+\S+\s+\S+\s+(\S+)\s+(\S+)\s+\S+\s+\S+/;
	($pos1, $pos2) = ($pos2, $pos1) if ($pos1 > $pos2);
```

```
    my $nspecieslag = "a";
    while (my $newline = readline ($BLAST_FILE)) {
      chomp $newline;
      my ($nspecies, $nseqid, $npos1, $npos2) = $newline =~ /(\S+)\s+(\S+)\s+\S+\s+\S+\s+\S+\s+\S+\s+\S+\s+\S+\s+(\S+)\s+(\S+)\s+\S+\s+\S+/;
      ($npos1, $npos2) = ($npos2, $npos1) if ($npos1 > $npos2);

      next if ($nspecies eq $nspecieslag);
      if ($nseqid ne $seqid) {
        print $RESULT_LOG ">$nspecies\n$note_seq{$nspecies}\n";
      } else {
        if ($npos2 <= $pos1) {
          print $RESULT_LOG ">$nspecies\n$note_seq{$nspecies}\n";
        } elsif ($npos1 >= $pos2){
          print $RESULT_LOG ">$nspecies\n$note_seq{$nspecies}\n";
        } else {
          print $RESULT_FILE ">$nspecies\n$note_seq{$nspecies}\n";
        }
      }
      $nspecieslag = $nspecies;
    }

    close ($BLAST_FILE) or die "Can't close $blast";
    close ($FASTA_FILE) or die "Can't close $path";
    close ($RESULT_FILE) or die "Can't close $new";
    close ($RESULT_LOG) or die "Cant't close $log";

  }
}

closedir (DIR);
```

Information on single loci used for phylogenetic and all subsequent analyses

Table S3A: Information on single alignments of 172 loci used for final analyses:

	# CDS	External name	# Sequences in alignment	Query Sequence Length (bp)	Max Sequence Length captured (bp)	Min Sequence Length captured (bp)
1	cd23	CDH23	28	453	450	111
2	cd28	UNC13C	28	1428	1413	495
3	cd30	PLCE1	28	681	681	243
4	cd55	DNAH1	25	189	189	99
5	cd66	SLC35B2	24	648	606	36
6	cd99	MED7	28	519	516	123
7	cd118	ALG11	27	252	252	117
8	cd120	MYCBP2	24	222	222	141
9	cd127	DYNC1H1	27	210	210	126
10	cd137	ABC1	20	213	213	93
11	cd153	CHTF18	21	210	207	123
12	cd170	ASB2	27	483	480	123
13	cd191	DNAH10	26	231	231	120
14	cd203	GUCD1	24	228	228	132
15	cd236	SMCR8	27	423	423	123
16	cd258	INTS2	26	222	222	90
17	cd267	ABCC3	26	300	300	117
18	cd272	CEP192	20	204	201	105
19	cd284	ACOX1	18	204	204	96
20	cd285	FLI1	25	204	201	93
21	cd295	BAIAP2	23	225	225	72
22	cd299	ZZEF1	15	258	258	168
23	cd300	DHX33	24	258	255	165
24	cd302	SUPT5H	25	198	198	111
25	cd324	AP4B1	25	226	223	1
26	cd329	RP2	28	474	471	123
27	cd330	TMEM60	26	378	375	69
28	cd334	TGFBRAP1	28	678	678	159
29	cd353	DYNC2H1	27	375	375	171
30	cd354	CTSC	27	591	498	186
31	cd357	DSTYK	27	375	375	135
32	cd362	EPRS	20	207	207	132
33	cd388	AMDHD1	28	276	273	129
34	cd393	MIEF1	26	219	216	69
35	cd404	ZDHHC23	26	204	204	120
36	cd405	IMPG2	26	207	207	93
37	cd426	VWA1	27	531	495	207
38	cd438	PANX2	28	1203	1200	396

	# CDS	External name	# Sequences in alignment	Query Sequence Length (bp)	Max Sequence Length captured (bp)	Min Sequence Length captured (bp)
39	cd441	VPS13D	28	696	693	120
40	cd447	PEX2	28	831	828	141
41	cd451	NAGLU	28	561	561	201
42	cd465	MON2	28	621	618	183
43	cd469	TMEM168	28	504	504	129
44	cd472	RAG1	28	1650	1650	168
45	cd473	YOD1	28	606	606	147
46	cd481	LRRC30	27	717	714	201
47	cd484	TGFBRAP1	27	336	336	162
48	cd485	NPC1	28	219	219	114
49	cd487	SS18	25	219	216	42
50	cd490	PRKDC	27	198	198	108
51	cd494	CYP7A1	28	546	546	162
52	cd506	BAMBI	26	285	282	147
53	cd514	TTN	27	270	270	144
54	cd520	MALRD1	24	231	231	69
55	cd521	C10orf112	27	366	363	120
56	cd522	OSGEPL1	28	381	381	126
57	cd550	POMGNT2	28	1728	1728	123
58	cd554	NPHP3	23	240	240	147
59	cd564	SCAP	27	319	319	97
60	cd567	MOCOS	27	654	642	123
61	cd568	EXOC3	28	750	675	147
62	cd570	ALG2	27	894	894	147
63	cd571	TDP2	21	237	237	147
64	cd599	KCNK2	27	309	309	180
65	cd604	THADA	28	660	660	123
66	cd608	ABCC10	23	237	228	117
67	cd609	ABCC10	25	480	471	114
68	cd616	CEBPZ	28	1122	1119	126
69	cd623	NUP13	26	222	222	120
70	cd624	GPER1	27	933	906	126
71	cd627	EPM2A	24	240	237	120
72	cd636	JMJD6	27	387	384	201
73	cd655	NSDHL	27	318	315	120
74	cd660	NUDT6	28	345	345	135
75	cd676	FRAS1	28	345	345	126
76	cd677	MOSPD1	25	219	216	120
77	cd687	ANKRD50	25	456	450	120
78	cd693	KIAA1239	28	591	591	150
79	cd695	FRAS1	25	201	201	126
80	cd698	PRDM8	26	228	228	111

	# CDS	External name	# Sequences in alignment	Query Sequence Length (bp)	Max Sequence Length captured (bp)	Min Sequence Length captured (bp)
81	cd699	FGF5	28	348	348	123
82	cd714	COPB1	25	225	225	78
83	cd722	RAG2	28	1419	1407	354
84	cd725	MYBPC3	27	258	258	108
85	cd727	DNAH10	27	420	417	144
86	cd758	PCNXL4	27	690	687	291
87	cd759	LRRC9	24	204	204	114
88	cd774	CDH23	13	210	204	147
89	cd805	DST	23	285	285	132
90	cd807	DTD1	28	234	234	123
91	cd814	FBXL4	28	498	498	129
92	cd817	RELN	26	231	228	87
93	cd834	TTN	28	300	300	105
94	cd845	RAB3GAP1	26	225	225	105
95	cd847	WDSUB1	28	387	381	132
96	cd850	PHKB	26	195	192	114
97	cd853	TSHZ3	26	255	255	126
98	cd856	TRIP4	26	198	198	105
99	cd880	SELRC1	27	399	396	126
100	cd888	FUT10	28	834	834	279
101	cd898	HMCN1	26	252	249	108
102	cd899	TRMT1L	27	300	252	150
103	cd905	IKBKAP	25	231	228	117
104	cd909	TRPC1	27	336	336	204
105	cd919	GPR171	28	870	870	177
106	cd927	TTN	28	273	273	126
107	cd932	TSPEAR	27	222	222	111
108	cd947	FYN	27	243	243	117
109	cd950	GLCE	25	231	231	129
110	cd953	ABCB7	27	210	210	90
111	cd960	CCDC127	26	636	636	126
112	cd963	NYX	27	915	915	270
113	cd971	DHTKD1	24	267	267	123
114	cd992	PDPR	27	399	399	126
115	cd998	FOCAD	26	234	234	117
116	cd1004	PIGM	26	255	255	105
117	cd1055	AHI1	26	186	183	120
118	cd1058	TNKS	26	237	237	57
119	cd1061	DPYD	25	213	213	123
120	cd1074	SLC35D3	28	435	432	114
121	cd1076	TRAPPC12	26	240	240	126
122	cd1078	RECQL5	18	468	468	174

	# CDS	External name	# Sequences in alignment	Query Sequence Length (bp)	Max Sequence Length captured (bp)	Min Sequence Length captured (bp)
123	cd1085	SORL1	23	243	243	171
124	cd1090	AGRN	25	213	210	120
125	cd1091	SLC22A23	20	291	288	135
126	cd1094	RAI2	26	315	303	192
127	cd1096	CLIP4	23	180	174	117
128	cd1099	LRP2	27	270	270	129
129	cd1108	GPR75	28	1503	1209	252
130	cd1110	SWAP70	25	225	225	117
131	cd1115	TUBGCP5	26	237	234	105
132	cd1123	NRDE2	25	312	312	123
133	cd1126	OXSM	28	855	855	219
134	cd1127	RAD50	14	195	192	117
135	cd1130	CCDC51	27	297	297	84
136	cd1133	ZNF598	26	219	219	123
137	cd1134	ASCC3	25	222	222	57
138	cd1145	HEPACAM	26	330	330	126
139	cd1150	HSPG2	25	264	264	105
140	cd1161	DNAH7	26	327	327	111
141	cd1171	DNAH5	27	321	318	159
142	cd1175	SLC35F6	28	372	372	123
143	cd1180	PID1	28	465	456	162
144	cd1195	EFHB	20	210	210	135
145	cd1201	VASH1	27	309	306	165
146	cd1207	KIAA1239	28	585	585	123
147	cd1217	ZFPM2	28	2142	1797	411
148	cd1234	SLC20A2	27	561	558	144
149	cd1239	USP19	25	207	207	126
150	cd1257	C8B	17	174	171	117
151	cd1258	KCTD4	28	726	726	123
152	cd1272	SRBD1	28	282	282	114
153	cd1279	MED23	28	246	246	114
154	cd1299	PDGFRL	27	234	231	120
155	cd1338	NBAS	23	228	228	117
156	cd1349	KLHL38	24	270	270	105
157	cd1351	KL	28	1116	1113	102
158	cd1352	KL	27	384	381	153
159	cd1354	VPS13C	21	387	387	90
160	cd1360	CPOX	22	216	216	111
161	cd1368	AIFM2	22	183	180	90
162	cd1371	KIAA1109	24	210	210	117
163	cd1374	SLC30A5	28	297	282	123
164	cd1379	LRIT3	23	255	255	120

	# CDS	External name	# Sequences in alignment	Query Sequence Length (bp)	Max Sequence Length captured (bp)	Min Sequence Length captured (bp)
165	cd1385	RAB30	26	237	237	126
166	cd1391	RGS9BP	28	543	543	264
167	cd1396	UBIAD1	21	411	408	78
168	cd1405	EOMES	28	582	561	171
169	cd1428	NARS	23	219	219	75
170	cd1429	DMRT3	28	837	837	291
171	cd1431	DAPK1	28	1518	1233	351
172	cd1438	GPR98	28	474	375	141

Table S3B: data missing per taxon in relation to the query sequence.

Genus	Species	Sequence length of concatenated 172 loci (linear unaligned, basepairs)	missing data
Query: ***Callorhinchus***	*milii*	71,234	0,00%
Squalus	*acanthias*	55,503	22,08%
Cirrhigaleus	*asper*	62,768	11,88%
Centrophorus	*moluccensis*	48,343	32,13%
Deania	sp.	22,732	68,09%
Dalatias	*licha*	45,206	36,54%
Isistius	*brasiliensis*	42,563	40,25%
Squaliolus	*aliae*	51,961	27,06%
Euprotomicrus	*bispinatus*	56,696	20,41%
Somniosus	*microcephalus*	44,741	37,19%
Zameus	*squamulosus*	49,333	30,75%
Scymnodon	*ringens*	43,818	38,49%
Centroselachus	*crepidater*	46,804	34,30%
Centroscymnus	*coelolepis*	52,123	26,83%

Scymnodon	*plunketi*	50,336	29,34%
Oxynotus	*paradoxus*	49,411	30,64%
Aculeola	*nigra*	51,910	27,13%
Centroscyllium	*fabricii*	59,249	16,82%
Etmopterus	*splendidus*	48,886	31,37%
Etmopterus	*sheikoi*	64,262	9,79%
Etmopterus	cf. *molleri*	57,800	18,86%
Etmopterus	*spinax*	50,153	29,59%
Etmopterus	*gracilispinis*	49,907	29,94%
Trigonognathus	*kabeyai*	61,048	14,30%
Echinorhinus	*brucus*	40,248	43,50%
Pliotrema	*warreni*	58,028	18,54%
Pristiophorus	*japonicus*	56,426	20,79%
Squatina	*nebulosa*	55,156	22,57%
Hexanchus	*griseus*	51,674	27,46%

Table S4: Overview of different phylogeentic analyses performed on the concatenated 172 loci dataset

Dataset	**Method**	**Tool**	**Substitution Model**	**Partition scheme**	**Outgroup**
Concatenated 1265 cds nucleotide loci	Maximum likelihood	RAxML	GTRCAT	none	*Hexanchus griseus*
Concatenated 1265 cds nucleotide loci	Maximum likelihood	RAxML	GTR+G	1^{st}+2^{nd} & 3^{rd} codon	*Hexanchus griseus*
Concatenated 1265 cds nucleotide loci	Maximum likelihood	RAxML	GTR+G	By gene	*Hexanchus griseus*
MARE reduced 174 loci amino acid matrix	Maximum likelihood	RAxML	JTT+G	By gene	*Hexanchus griseus*
MARE reduced 174 loci amino acid matrix	Maximum likelihood	RAxML	Blosum62	By gene	*Hexanchus griseus*
MARE reduced 174 loci amino acid matrix	Maximum likelihood	RAxML	see partition file*	Best partition scheme suggested by partitionfinderProtein vers. 1.1.1*	*Hexanchus griseus*
Concatenated 172 cds nucleotide loci	Maximum likelihood	RAxML	GTRCAT	none	*Hexanchus griseus*
Concatenated 172 cds nucleotide loci	Maximum likelihood	RAxML	GTR+G	1^{st}+2^{nd} & 3^{rd} codon	*no outgroup defined*
Concatenated 172 cds nucleotide loci	Maximum likelihood	RAxML	GTR+G	by gene and 1^{st}+2^{nd} & 3^{rd} codon	*Hexanchus griseus*
Concatenated 172 cds nucleotide loci	Maximum likelihood	RAxML	see partition file**	Best partition scheme suggested by partitionfinder vers. 1.1.1**	*Hexanchus griseus*
Concatenated 172 cds nucleotide loci	Maximum likelihood	RAxML	GTRCAT	none	no outgroup defined
Concatenated 172 cds nucleotide loci	Maximum likelihood	RAxML	GTRCAT	none	no outgroup defined, *Hexanchus griseus* deleted from alignment
Concatenated 172 amino acid loci	Bayesian	PhyloBayes	CAT	none	n.a.
Concatenated 172 cds nucleotide loci	Bayesian inference	BEAST	GTR+G	1^{st}+2^{nd} & 3^{rd} codon	*Hexanchus griseus*

*PartitionfinderProtein best partitioning scheme for RAxML:

```
branchlengths     : linked
models            : MTMAM+G+F, MTMAM+G, DAYHOFF+G, DCMUT+G+F, VT+G+F, DCMUT+G, JTT+G, LG+G+F, LG+G, MTREV+G,
MTREV+G+F, RTREV+G+F, WAG+G, CPREV+G, CPREV+G+F, WAG+G+F, JTT+G+F, DAYHOFF+G+F, RTREV+G, VT+G, BLOSUM62+G,
BLOSUM62+G+F
model_selection   : bic
search           : rcluster
weights           : rate = 1, base = 0, model = 0, alpha = 0
rcluster-percent  : 1.0


JTT, p1 = 1-285, 1188-1477, 2030-2206, 2803-3004, 3521-3606, 5044-5593, 6280-6495, 8078-8206, 11480-11556, 11997-12071, 12072-12195, 12818-
12898, 13178-13335, 15183-15256, 16167-16538, 16618-16726, 16727-16965, 17816-17930, 19078-19169, 20052-20135, 20743-20813, 20888-21129,
21744-21811, 22612-22748, 22749-22877, 23174-23254, 24016-24095, 24096-24166
JTT, p2 = 286-381, 382-686, 3170-3248, 6210-6279, 13336-13457
JTT, p3 = 687-1187, 11871-11996
JTT, p4 = 1478-1661
JTT, p5 = 1662-1756, 1869-2029, 3249-3338, 13701-13840, 23662-23740
JTT, p6 = 1757-1868, 14491-14685, 15969-16041, 18332-18404, 19423-19499, 21517-21743, 22401-22499, 22500-22611
JTT, p7 = 2207-2436, 6103-6209, 7642-7793, 8207-8920, 10152-10279, 11713-11797, 11798-11870, 18466-18535, 19170-19234, 19828-19902, 20269-
20413, 21423-21516
MTMAM, p8 = 2437-2536
JTT, p9 = 2537-2733, 4119-4416, 7300-7388, 13458-13527
JTT, p10 = 2734-2802, 10535-10638, 11625-11712
JTT, p11 = 3005-3095
JTT, p12 = 3096-3169, 3607-3826, 8001-8077, 9837-9913, 10435-10534, 11353-11479, 15423-15501, 15903-15968, 21130-21219
JTT, p13 = 3339-3520
JTT, p14 = 3827-3901
JTT, p15 = 3902-3967, 10280-10365, 20511-20742, 23417-23506
JTT, p16 = 3968-4118, 11267-11352, 12662-12817, 17931-18142
JTT, p17 = 4417-4496, 6035-6102, 12984-13177, 14058-14182, 16539-16617, 18227-18331, 19308-19422, 23255-23336, 23337-23416
JTT, p18 = 4497-4566, 11557-11624, 14260-14365, 14964-15037, 15257-15422, 16042-16166, 18143-18226, 21883-22159, 22291-22400, 23032-23097
JTT, p19 = 4567-4784, 14366-14428
JTT, p20 = 4785-4883, 5594-5967, 10082-10151, 12899-12983, 13528-13700, 14429-14490, 15111-15182
```

```
JTT, p21 = 4884-5043
JTT, p22 = 5968-6034, 7794-8000, 9979-10081, 16966-17471, 18612-18890, 18891-19077, 19750-19827, 19903-19978, 20414-20510, 20814-20887,
22218-22290, 23741-23937, 23938-24015
JTT, p23 = 6496-6971, 6972-7112, 8921-9053, 9914-9978, 12351-12661, 13841-13914, 14686-14963, 17640-17755, 17756-17815, 21320-21422,
22878-22952
JTT, p24 = 7113-7299, 9054-9131, 13990-14057, 18536-18611, 21220-21319
JTT, p25 = 7389-7483, 9132-9260, 9261-9836, 17472-17639, 19235-19307, 19500-19749, 20136-20268, 23507-23661, 24167-24235, 24236-24461
JTT, p26 = 7484-7641, 10726-11198
JTT, p27 = 10366-10434
JTT, p28 = 10639-10725, 14183-14259, 15038-15110, 23098-23173
JTT, p29 = 11199-11266, 12281-12350, 22160-22217
MTMAM, p30 = 12196-12280
JTT, p31 = 13915-13989, 15502-15902
JTT, p32 = 18405-18465
JTT, p33 = 19979-20051
MTREV, p34 = 21812-21882
JTT, p35 = 22953-23031
```

**Partitionfinder best partitioning scheme for RAxML:

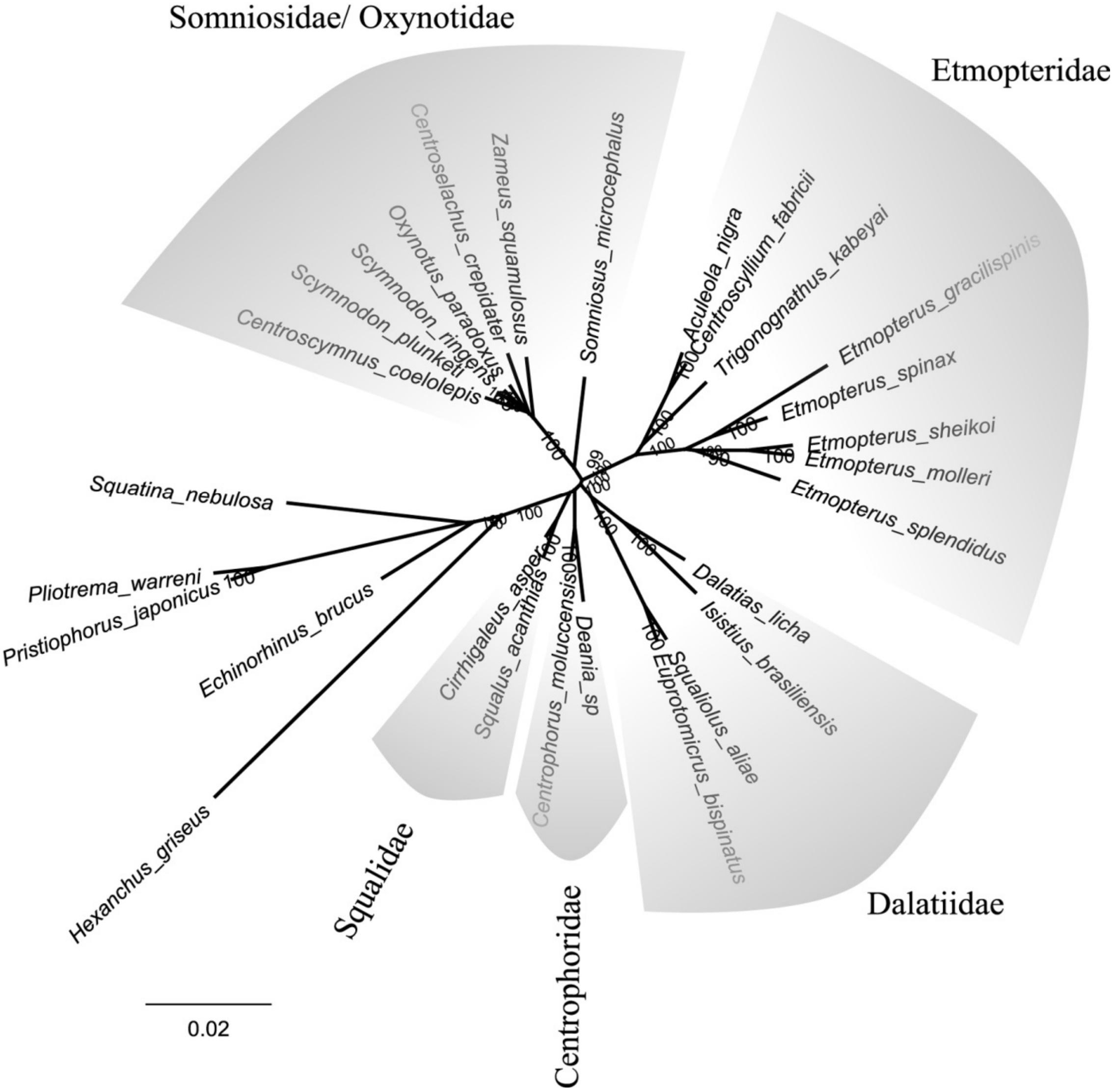
```
branchlengths     : linked
models            : GTR+I+G, GTR+G
model_selection   : bic
search            : rcluster
weights           : rate = 1, base = 0, model = 0, alpha = 0
rcluster-percent  : 0.1



DNA, p1 = 1-453, 6391-6612, 9164-9541, 32430-32669, 32670-33056, 43575-43826, 51471-51683, 51684-51974, 56583-56804, 56805-57134
DNA, p2 = 454-1881, 2563-2751, 3919-4170, 4603-4815, 5026-5508, 5740-5967, 5968-6390, 6913-7116, 7117-7320, 7525-7749, 8008-8265, 10595-11185,
11186-11560, 11768-12043, 12263-12466, 12674-13204, 13205-14407, 15935-16495, 16496-17116, 17621-19270, 19271-19876, 21368-21565, 21566-22111,
22112-22396, 22667-22897, 23264-23644, 25613-25931, 26586-27335, 27336-28229, 28230-28466, 28776-29435, 30153-31274, 31275-31496, 33057-33374,
33375-33719, 33720-34064, 34065-34283, 34284-34739, 34740-35330, 35331-35531, 35760-36107, 36333-37751, 38430-39119, 39324-39533, 39819-40052,
40782-41081, 41082-41306, 41694-41888, 41889-42143, 42144-42341, 42342-42740, 42741-43574, 43827-44126, 44127-44357, 44358-44693, 44694-45563,
```

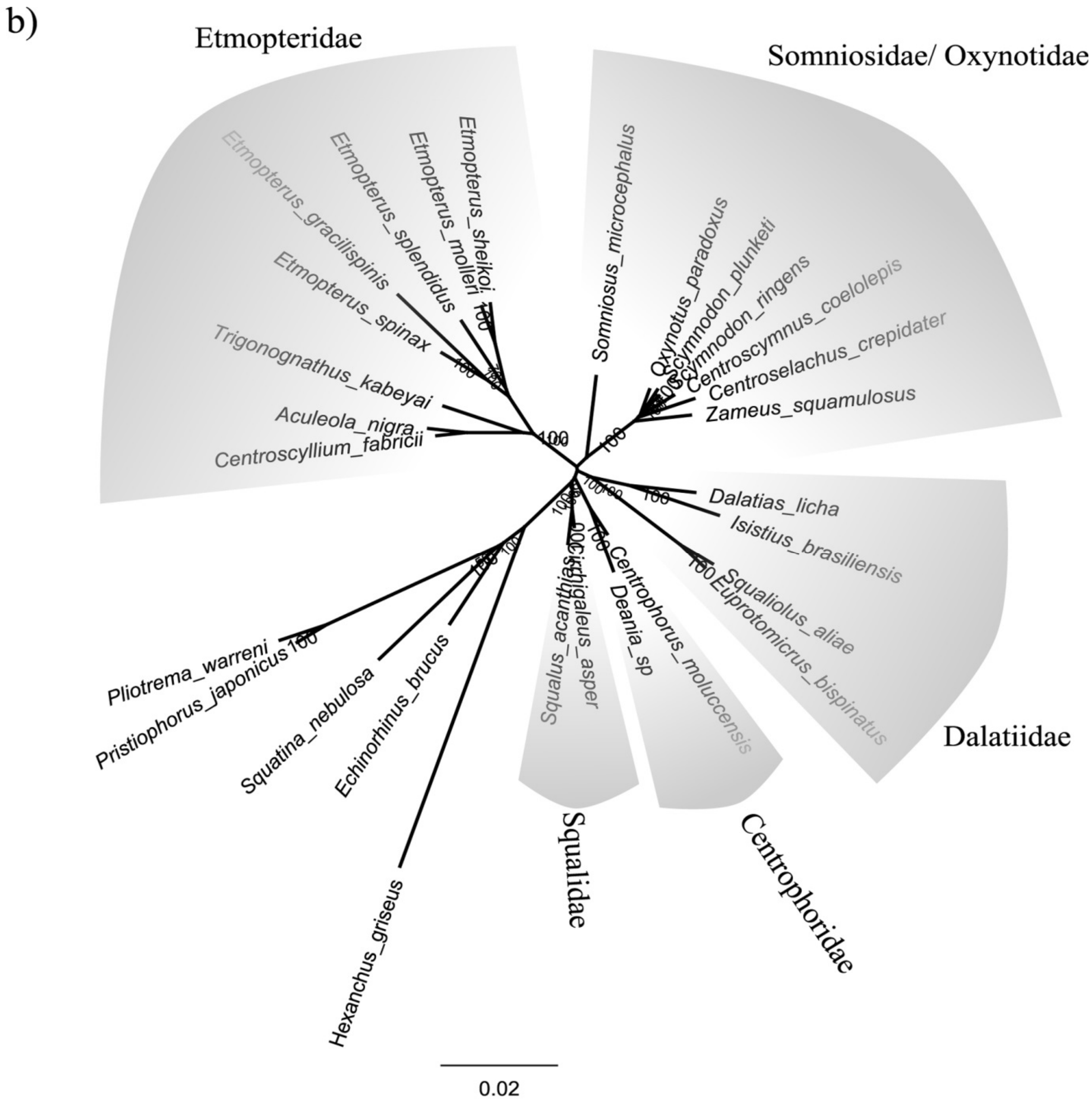
45837-46058, 46302-46532, 46533-46742, 46743-47378, 48960-49193, 49194-49448, 49449-49634, 49635-49871, 49872-50084, 50085-50519, 50520-50759, 52290-52469, 52470-52739, 54468-54704, 55017-55871, 55872-56066, 56067-56363, 56364-56582, 57726-58046, 58419-58883, 59094-59402, 59403-59987, 59988-62129, 62691-62897, 62898-63071, 63072-63797, 63798-64079, 64326-64559, 64560-64787, 64788-65057, 65058-66173, 66174-66557, 66945-67160, 67161-67343, 67344-67553, 67554-67850, 67851-68105, 69297-69878, 72453-72926
DNA, p3 = 1882-2562, 4171-4392, 4393-4602, 4816-5025, 5509-5739, 6613-6912, 8266-8463, 8464-8689, 8690-9163, 9542-10219, 11561-11767, 12467-12673, 14408-15103, 19877-20593, 20594-20929, 21149-21367, 22397-22666, 22898-23263, 23645-25372, 25373-25612, 25932-26585, 29436-29672, 29673-30152, 31497-32429, 36108-36332, 37752-38009, 38010-38429, 39534-39818, 40053-40550, 45564-45836, 46059-46301, 47379-48293, 48294-48560, 48561-48959, 50760-51227, 51228-51470, 54243-54467, 54705-55016, 57135-57398, 57399-57725, 58047-58418, 58884-59093, 62130-62690, 64080-64325, 66558-66944, 68886-69296
DNA, p4 = 2752-3399, 3400-3918, 7750-8007, 20930-21148, 52740-54242
DNA, p5 = 7321-7524, 10220-10594, 12044-12262, 15104-15934, 17117-17620, 28467-28775, 35532-35759, 39120-39323, 40551-40781, 41307-41693, 51975-52289, 68106-68342, 69879-70097, 70098-70934, 70935-72452
DNA, p6 = 68343-68885

Outgroup testing

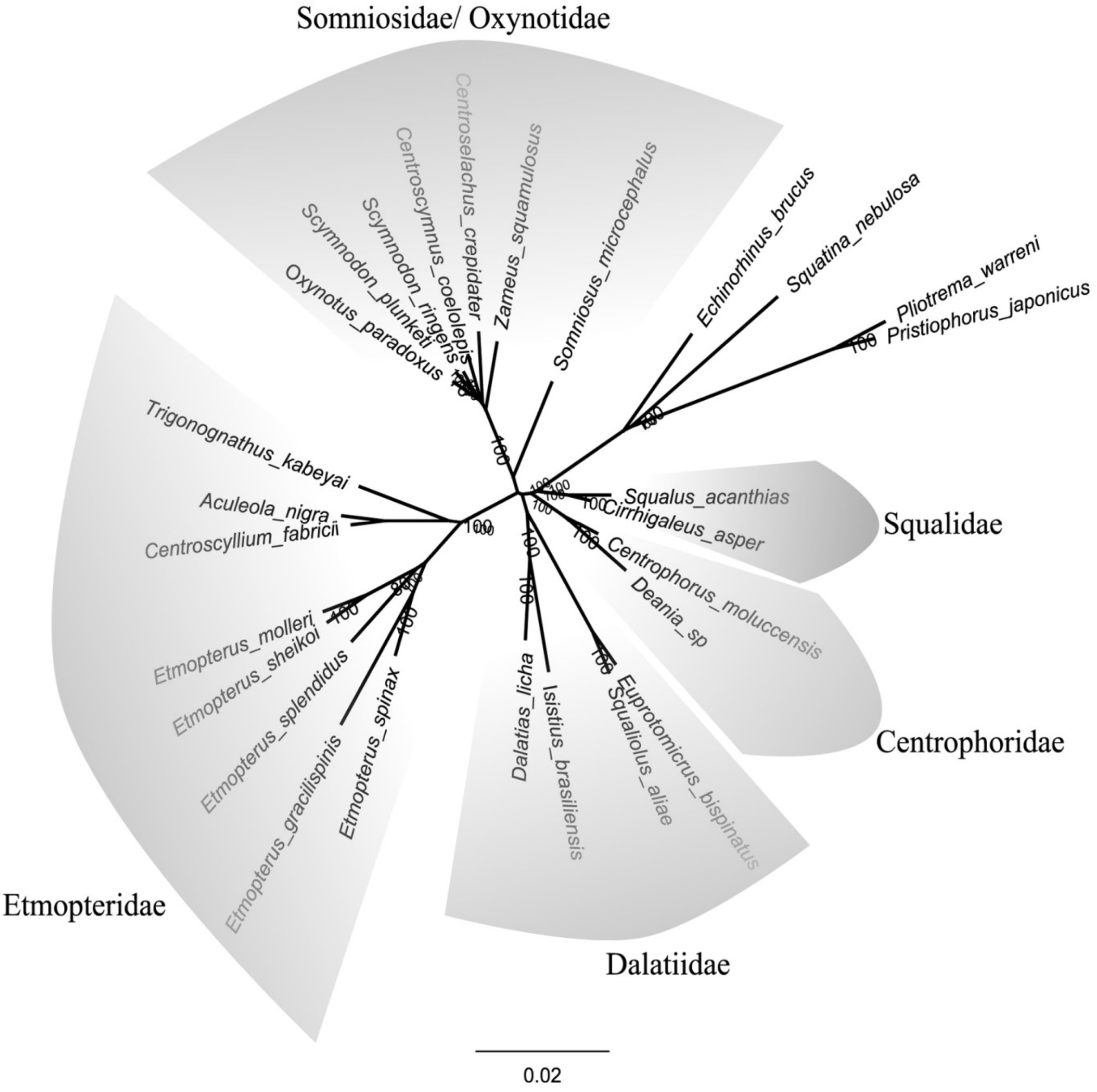
For testing the influence of different outgroups, we compared unrooted topologies (Maximum Likelihood, computed with RAxML under GTRCAT).

a)




b)
Etmopteridae
Somniosidae/ Oxynotidae
Dalatiidae
Squalidae
Centrophoridae
Etmopterus_gracilispinis
Etmopterus_splendidus
Etmopterus_molleri
Etmopterus_sheikoi
Etmopterus_spinax
Trigonognathus_kabeyai
Aculeola_nigra
Centroscyllium_fabricii
Somniosus_microcephalus
Oxynotus_paradoxus
Scymnodon_plunketi
Scymnodon_ringens
Centroscymnus_coelolepis
Centroselachus_crepidater
Zameus_squamulosus
Dalatias_licha
Isistius_brasiliensis
Squaliolus_aliae
Euprotomicrus_bispinatus
Centrophorus_moluccensis
Deania_sp
Cirrhigaleus_asper
Squalus_acanthias
Pliotrema_warreni
Pristiophorus_japonicus
Squatina_nebulosa
Echinorhinus_brucus
Hexanchus_griseus
100
0.02

c)


Somniosidae/ Oxynotidae
Centroselachus_crepidater
Zameus_squamulosus
Centroscymnus_coelolepis
Scymnodon_ringens
Scymnodon_plunketi
Oxynotus_paradoxus
Somniosus_microcephalus
Echinorhinus_brucus
Squatina_nebulosa
Pliotrema_warreni
Pristiophorus_japonicus
Trigonognathus_kabeyai
Aculeola_nigra
Centroscyllium_fabricii
Squalus_acanthias
Cirrhigaleus_asper
Squalidae
Centrophorus_moluccensis
Deania_sp
Centrophoridae
Etmopterus_molleri
Etmopterus_sheikoi
Etmopterus_splendidus
Etmopterus_gracilispinis
Etmopterus_spinax
Dalatias_licha
Isistius_brasiliensis
Squaliolus_aliae
Euprotomicrus_bispinatus
Etmopteridae
Dalatiidae
100
0.02

d)

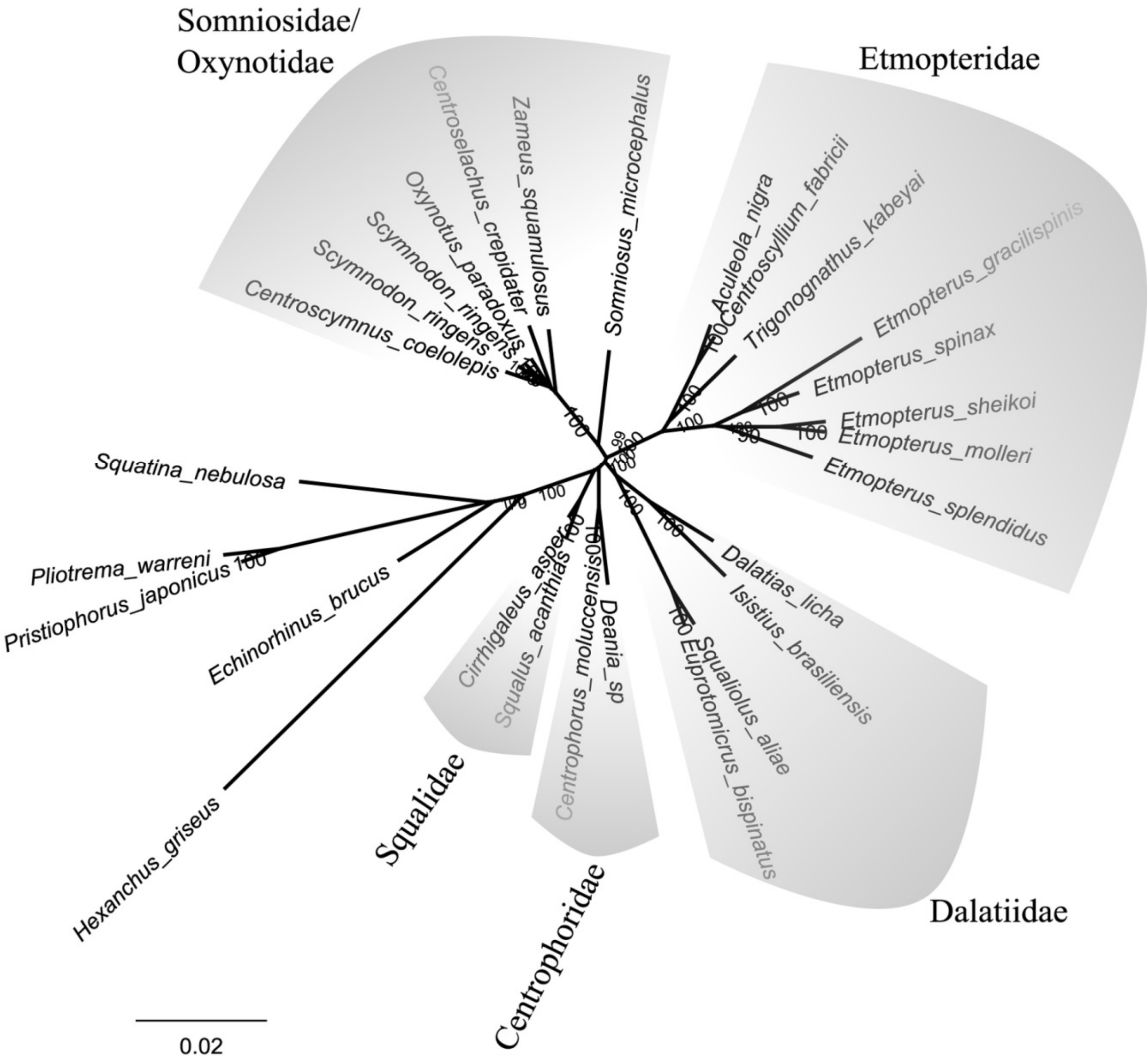


Figure S2: Maximum likelihood analyses comparing based on 172 nucleotide loci. Tree computed using RAxML [35]under GTRCAT.

a) *Hexanchus griseus* chosen as outgroup.

b) no outgroup defined

c) excluding *Hexanchus griseus* from the analysis, no outgroup defined

d) defining *Pristiophurus japonicus* and *Pliotrema warreni* as outgroup taxa.

Phylogenetic analysis:

Resulting phylogenetic estimates from analyses described in Table S4:

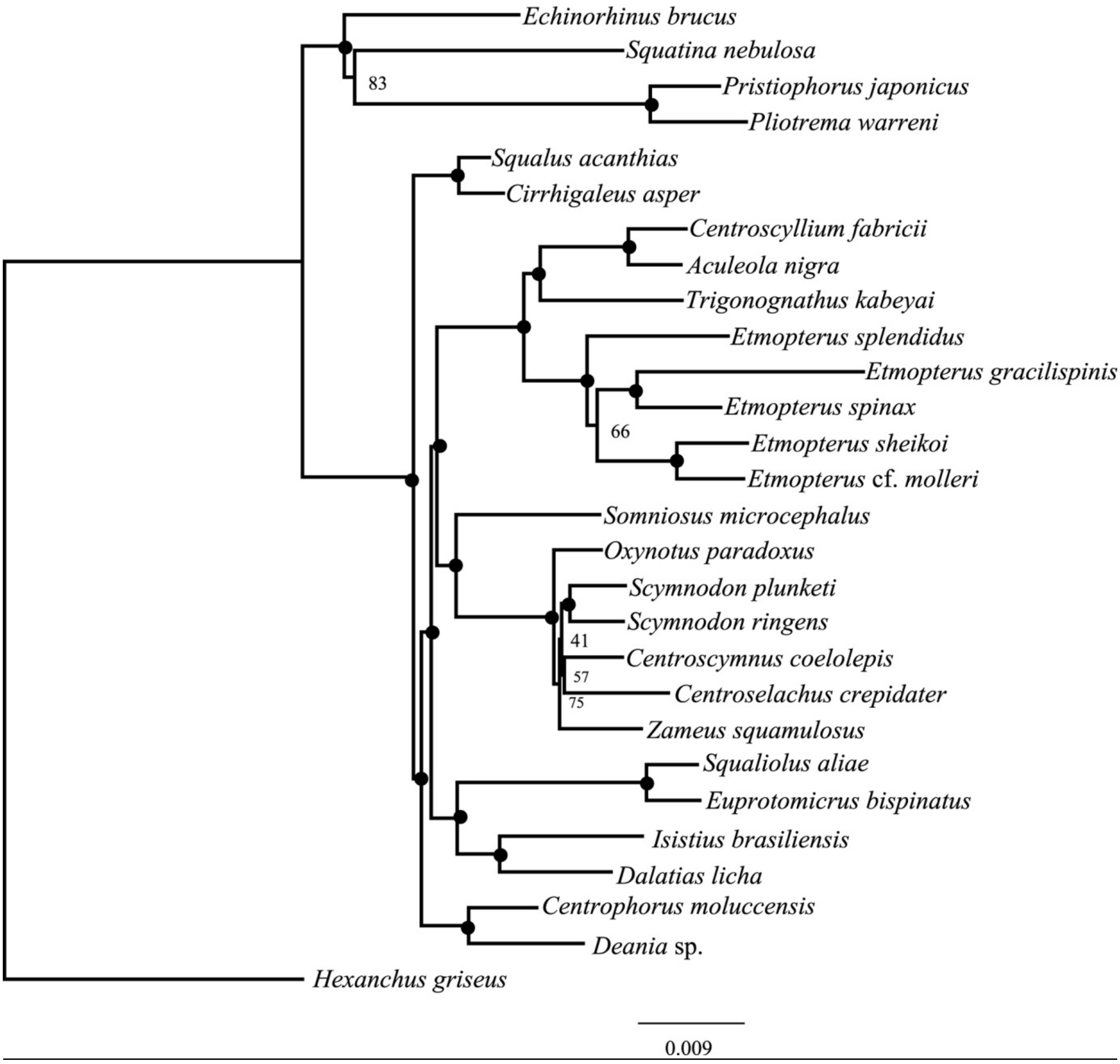


Figure S3: Maximum likelihood analysis using RAxML [35] under a GTRGAMMA model based on the full nucleotide dataset of 1265 loci including all sequence information. Data partitioned by 1st, 2nd and 3rd codon position. Black dots indicate 100% bootstrap support from 100 bootstrap replicates drawn. *Hexanchus griseus* was defined as outgroup.

Figure S4: Maximum likelihood analysis using RAxML [35] based on the 174 loci translated to amino acids resulting from the MARE [33, 34]analysis. Data partition and model applications are based on the result of Partitionfinder Protein [62, 63] applied to the data before the ML analysis (see Table 3 for details on the partitioning scheme). Black dots indicate 100% bootstrap support from 100 bootstrap replicates drawn. *Hexanchus griseus* was defined as outgroup.

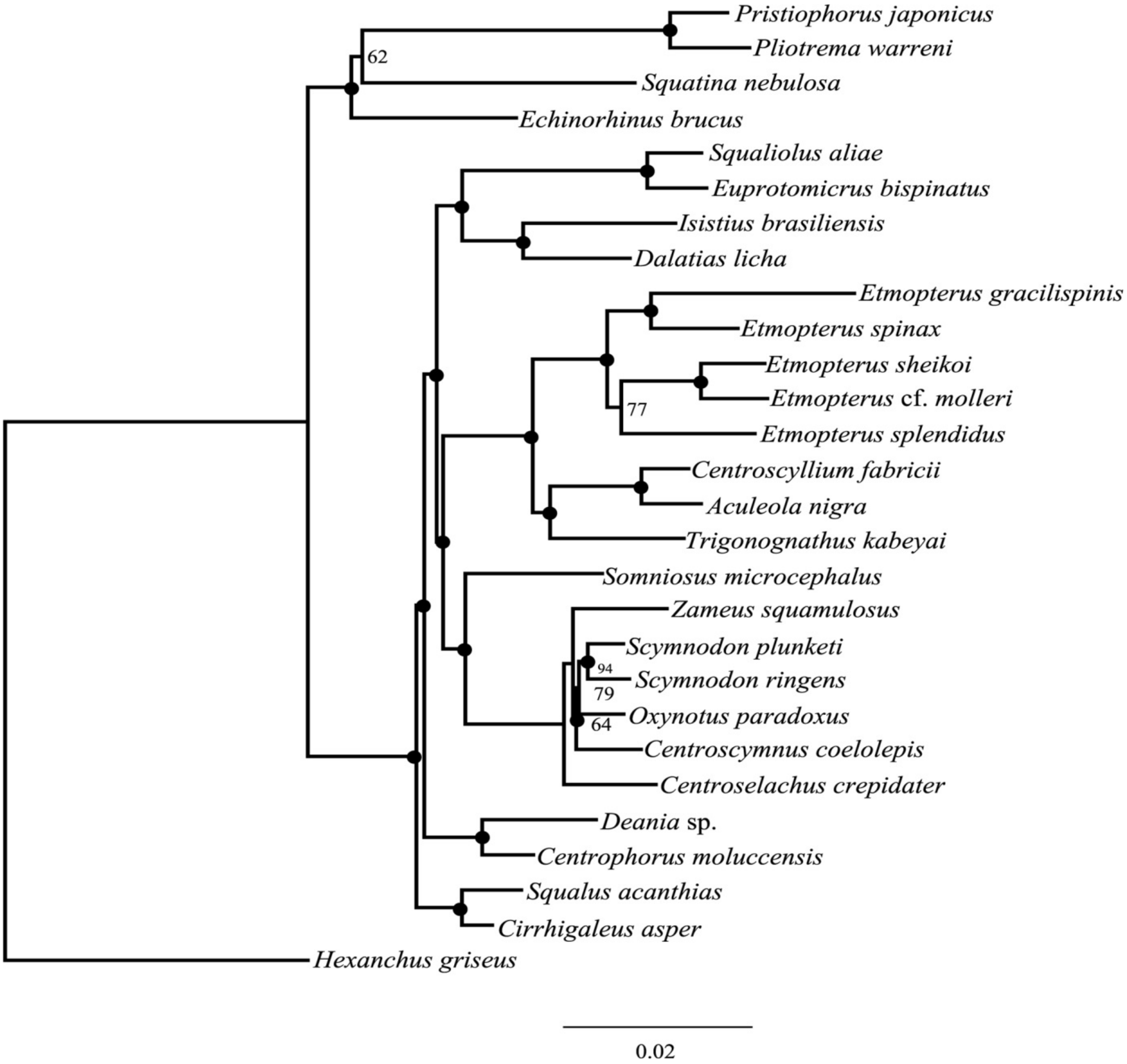


Figure S5: Maximum likelihood analysis using RAxML [35] based on the 172 loci translated to nucleotides resulting from the MARE [33, 34]and re-blast analysis. The data partition is based on the result of Partitionfinder [62, 63] applied to the data (see Table S4 for details on the partitioning scheme). Black dots indicate 100% bootstrap support from 100 bootstrap replicates drawn. *Hexanchus griseus* was defined as outgroup.

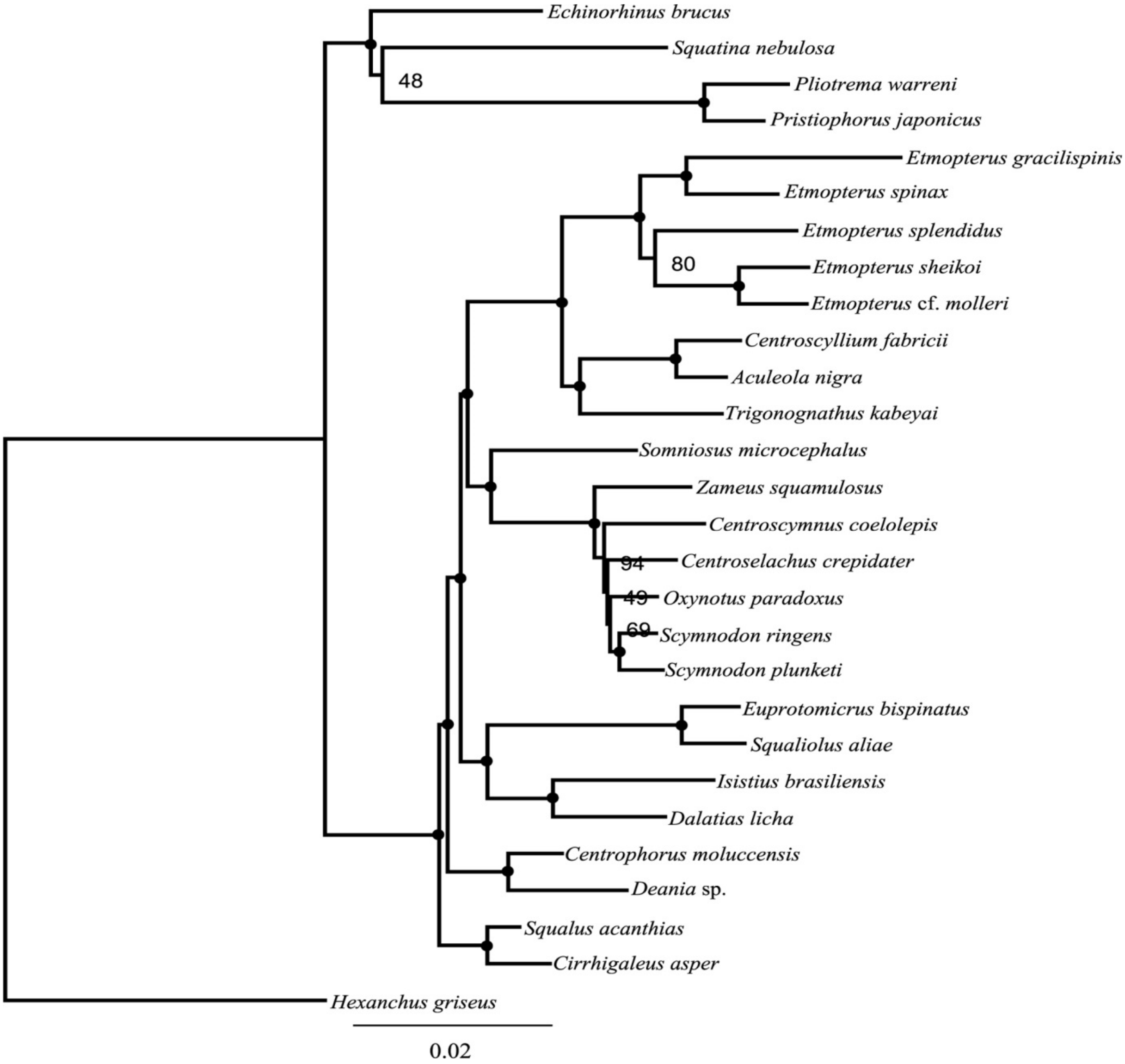


Figure S6: Maximum likelihood analysis using RAxML [35] based on the 172 loci translated to nucleotides resulting from the MARE and re-blast analysis. GTR GAMMA is used on unpartitioned data. Black dots indicate 100% bootstrap support from 100 bootstrap replicates drawn. *Hexanchus griseus* was defined as outgroup.

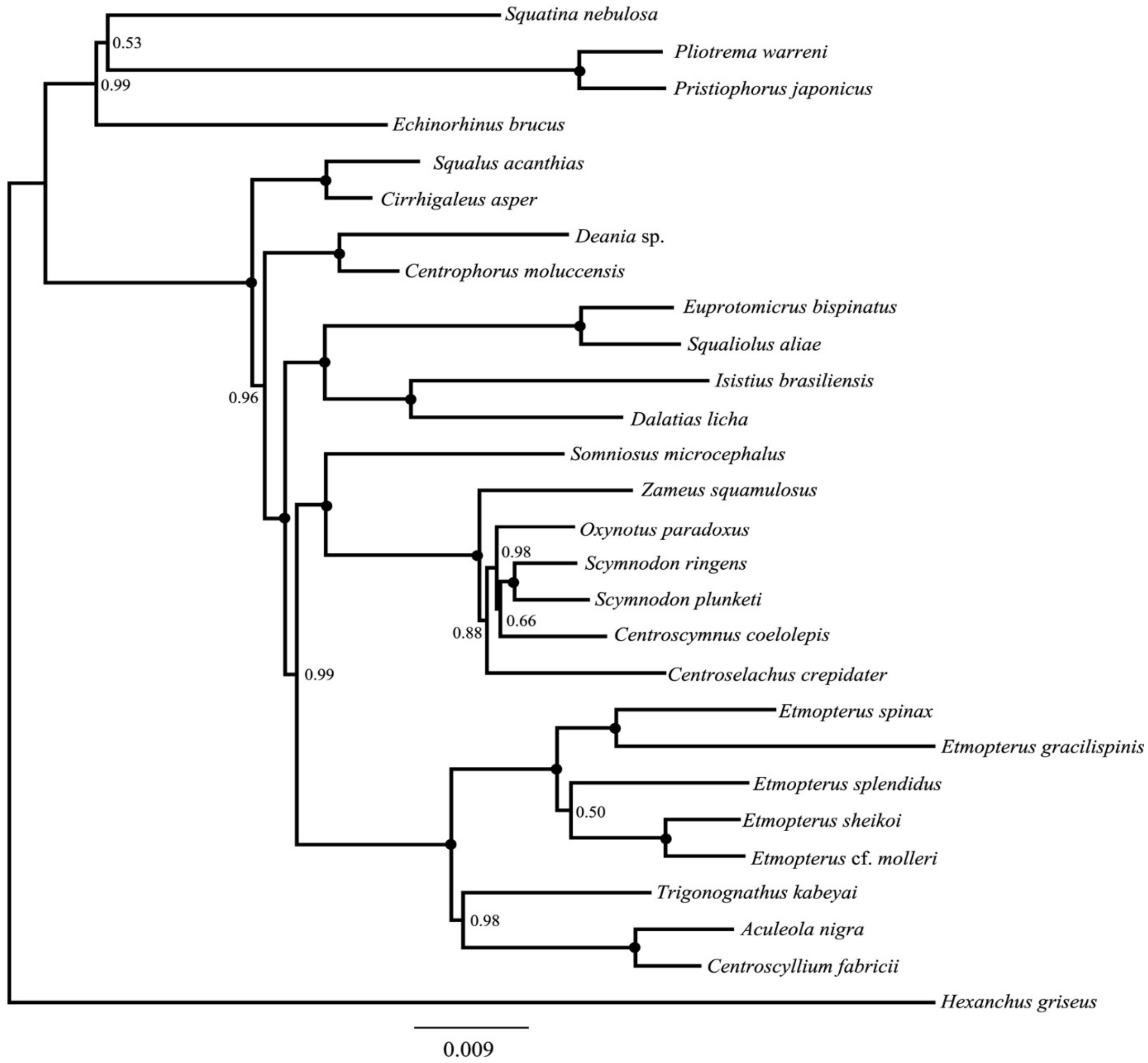


Figure S7: Bayesian inference using PhyloBayes 3.3f [36, 37] based on the 172 amino acid loci resulting from the MARE and re-blast analysis. The CAT model [64] is used on the unpartitioned dataset. Four chains were run in parallel. The tracefiles and treelists of all four chains were used to check for convergence . The analysis was stopped with a maximum difference of 0.16 and effective sample sizes exceeding 100, with the exception of the allocent statistic (see analysis protocol below). A majority rule consensus tree was computed from 12997 input trees from each chain (burnin = 1000 trees, analyzing every second tree of the pooled trees). Black dots indicate posterior probabilities of 1. Tree midpoint-rooted using Figtree 1.4.2 [69].

Analysis protocol for the Bayesian inference using Phylobayes 3.3f [36, 37]:

Phylobayes was called as follows:

```
pb -d "input alignmnet of 172 aminoacid loci" -cat -nchain 4 500 0.1 100.
```

tracecomp output after 4734362 generations:

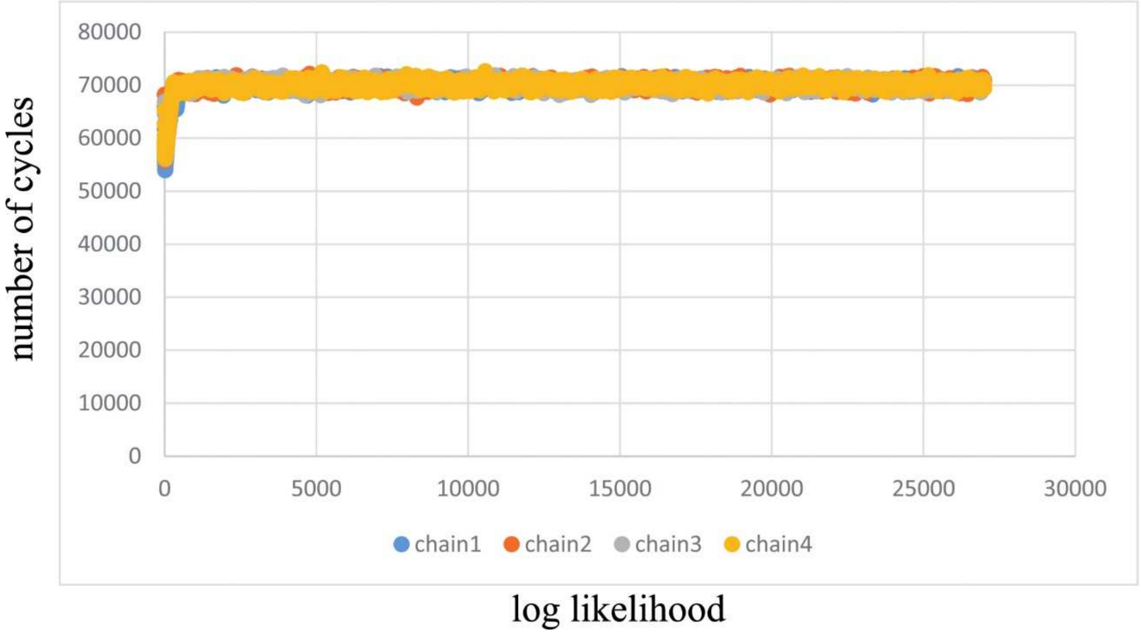
```
name              effsize   rel_diff
```

loglik	448	0.139033
length	3877	0.0473159
alpha	831	0.106427
nocc	410	0.163083
stat	457	0.111566
statalpha	19448	0.026536
kappa	881	0.103027
allocent	52	0.499115

bpcomp output after 4734362 generations:

maxdiff: 0.157806
meandiff: 0.00259295

Plot of log-likelihoods for 4734362 generations:



Node time estimation using BEAST

BEAUTi was used to create XML formatted input files for BEAST. Please see Table 2 in the main text for calibration points used. The full XML input file is available at http://datadryad.org/review?doi=doi:10.5061/dryad.n3581

MEDUSA [42, 43] analysis for estimating diversification rates and shifts

Table S5: species richness table

taxon	**Species richness values (deployed from [1])**
Aculeola	1
Centrophorus	12
Centroselachus	1
Centroscyllium	7
Centroscymnus	3
Cirrhigaleus	3
Dalatias	1
Deania	4
Echinorhinus	2
Etmopterus lucifer **clade**	11
Etmopterus gracilispinis **clade**	7
Etmopterus sheikoi	1
Etmopterus spinax **clade**	10
Etmopterus pusillus **clade**	8
Euprotomicrus	1
Hexanchus	2
Isistius	2
Oxynotus	5
Pliotrema	1
Pristiophorus	7
Scymnodon plunketi	1
Scymnodon	3
Somniosus	4
Squalus	25
Squaliolus	2
Squatina	22
Trigonognathus	1
Zameus	1

Note that *Etmopterus villosus* was not included in the analysis, as its *Etmopterus* subclade designation is unclear [23].

R protocol:

```
R version 3.0.3 (2014-03-06) -- "Warm Puppy"

Copyright (C) 2014 The R Foundation for Statistical Computing

Platform: x86_64-w64-mingw32/x64 (64-bit)

> library (laser)
Lade nötiges Paket: ape
Lade nötiges Paket: geiger

#loading timetree estimated with BEAST.
>  read.tree ("172.tre")->tree
> tree
```

```
Phylogenetic tree with 28 tips and 27 internal nodes.

Tip labels:
      Centrophorus_ moluccensis, Deania_sp, Etmopterus_gracilispinis, Etmopterus_spinax,
Etmopterus_molleri, Etmopterus_sheikoi, ...
Rooted; includes branch lengths.

#loading richness table including species richness information for each branch in the tree.
>  read.csv ("172richness_2.csv") ->richness
> richness
                      taxon ntaxa
1      Isistius_brasiliensis     2
2          Hexanchus_griseus     6
3             Dalatias_licha     1
4         Etmopterus_molleri    11
5  Centroselachus_crepidater     1
6     Trigonognathus_kabeyai     1
7    Centroscyllium_fabricii     7
8          Etmopterus_spinax    10
9         Etmopterus_sheikoi     1
10   Somniosus_microcephalus     4
11  Euprotomicrus_bispinatus     1
12          Pliotrema_warreni     1
13   Pristiophorus_japonicus     7
14         Cirrhigaleus_asper     3
15          Squalus_acanthias    25
16     Scymnodon_plunketi     1
17           Aculeola_nigra     1
18   Centroscymnus_coelolepis     3
19        Zameus_squamulosus     2
20          Squatina_nebulosa    22
21  Etmopterus_gracilispinis     7
22        Oxynotus_paradoxus     5
23          Scymnodon_ringens     3
24           Squaliolus_aliae     2
25   Centrophorus_moluccensis    12
26     Etmopterus_splendidus     8
27        Echinorhinus_brucus     2
28                  Deania_sp     4

#starting MEDUSA, using AICc as stopping criterion
> res1=medusa(tree, richness, warnings=FALSE)
Appropriate AICc threshold for tree of 28 tips is: 1.091845.

Step 1: lnLik=-187.6063; aicc=379.4434; model=bd
Step 2: lnLik=-183.8979; aicc=376.5959; shift at node 23; model=yule; cut=stem; # shifts=1
Step 3: lnLik=-181.5832; aicc=374.391; shift at node 35; model=yule; cut=stem; # shifts=2

No significant increase in aicc score. Disregarding subsequent piecewise models.

  Model.ID Shift.Node Cut.At Model Ln.Lik.part         r epsilon     r.low    r.high
1        1         29   node  yule   -85.80499 0.0198648      NA 0.0147561 0.0263547
2        2         23   stem  yule   -4.198604 0.0841583      NA 0.0471845 0.1585100
3        3         35   stem  yule   -91.57965 0.0410316      NA 0.0308829 0.0535196
> print(names(res1)) # output list elements
```

```
[1] "control"      "cache"        "model"        "summary"      "zSummary"      "medusaVersion"
[7] "richness"
> print(res1$summary) # show 'summary' object
  Model.ID Shift.Node Cut.At Model Ln.Lik.part        r epsilon      r.low     r.high
1        1         29   node  yule   -85.80499 0.0198648      NA 0.0147561 0.0263547
2        2         23   stem  yule   -4.198604 0.0841583      NA 0.0471845 0.1585100
3        3         35   stem  yule   -91.57965 0.0410316      NA 0.0308829 0.0535196
> summary(res1, criterion="aicc")
              Length Class      Mode
control         3    -none-     list
cache           2    -none-     list
model          11    -none-     list
summary         9    data.frame list
zSummary      756    -none-     numeric
medusaVersion   1    -none-     numeric
richness        2    data.frame list

#plotting results, plot breakpoints for best AIC model (see GEIGER package manual)
> plot(res1, cex=0.5,label.offset=1, edge.width=2)
```

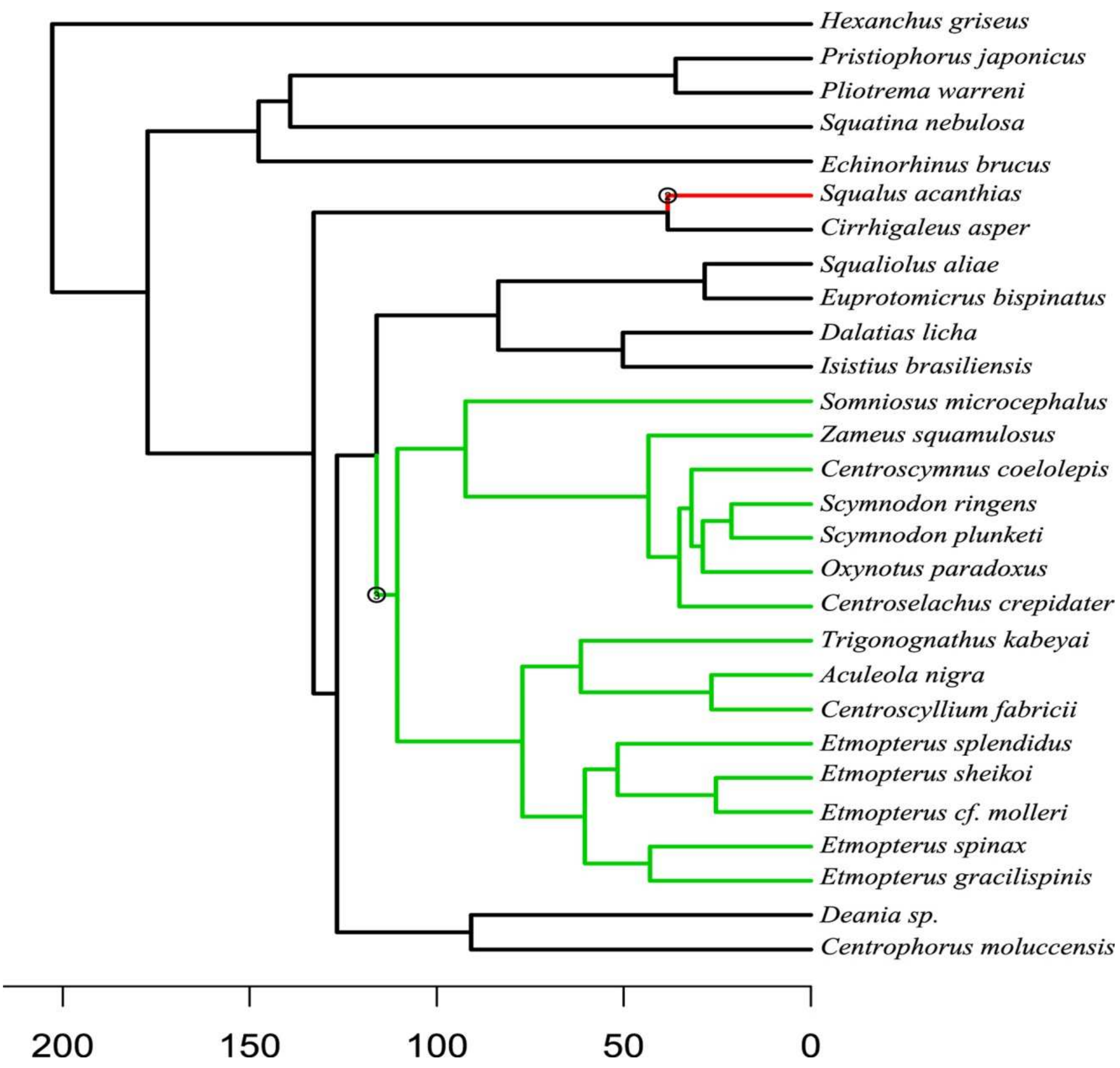


Figure S8: Output tree of MEDUSA [42, 43] analysis. Colored branches and dots mark significant increases in rate shifts, i.e. at the radiation of Etmopteridae and Somniosidae as well as the radiation of *Squalus*.

Species richness values were derived from the bibliographic database www.shark-references.com (accessed 18.04.2015) [1].

GEIGER Package [42] for ancestral character state reconstruction of occurrence of Bioluminescence

Table S6: Presence and absence of photophores in Squaliform sharks based on different information tested in the ancestral character state reconstructions.

taxon	**Presence of photophores (0=absent, 1=present; assuming Somniosidae as non-luminous)**	**Presence of photophores (0=absent, 1=present; based on observations in this study)**	**Presence of photophores (0=absent, 1=present; based on [8])**
Aculeola nigra	1	1	1
Centrophorus moluccensis	0	0	0
Centroselachus crepidater	0	0	1
Centroscyllium fabricii	1	1	1
Centroscymnus coelolepis	0	0	1
Cirrhigaleus asper	0	0	0
Dalatias licha	1	1	1
***Deania* sp.**	0	0	0
Echinorhinus brucus	0	0	0
Etmopterus* cf. *molleri	1	1	1
Etmopterus gracilispinis	1	1	1
Etmopterus sheikoi	1	1	1
Etmopterus spinax	1	1	1
Etmopterus splendidus	1	1	1
Euprotomicrus bispinatus	1	1	1
Hexanchus griseus	0	0	0
Isistius brasiliensis	1	1	1
Oxynotus paradoxus	0	0	0
Pliotrema warreni	0	0	0
Pristiophorus japonicus	0	0	0
Scymnodon plunketi	0	0	1
Scymnodon ringens	0	0	1
Somniosus microcephalus	0	0	0
Squalus acanthias	0	0	0
Squaliolus aliae	1	1	1
Squatina nebulosa	0	0	0

Trigonognathus kabeyai	1	1	1
Zameus squamulosus	0	1	1

R protocol:

```
R version 3.0.3 (2014-03-06) -- "Warm Puppy"
Copyright (C) 2014 The R Foundation for Statistical Computing
Platform: x86_64-w64-mingw32/x64 (64-bit)

> library (laser)

#loading different tables containing information on presence or absence of photphores in taxa.
#Lumi.csv= Table 4, column 1 as example.
> read.csv ("Lumi.csv")
                      taxon Lumi2
1      Isistius_brasiliensis     1    0
2         Hexanchus_griseus     0    0
3            Dalatias_licha     1    0
4        Etmopterus_molleri     1    0
5  Centroselachus_crepidater     0    0
6     Trigonognathus_kabeyai     1    0
7    Centroscyllium_fabricii     1    0
8         Etmopterus_spinax     1    0
9        Etmopterus_sheikoi     1    0
10   Somniosus_microcephalus     0    0
11  Euprotomicrus_bispinatus     1    0
12         Pliotrema_warreni     0    0
13   Pristiophorus_japonicus     0    0
14        Cirrhigaleus_asper     0    0
15         Squalus_acanthias     0    0
16       Scymnodon_plunketi     0    0
17            Aculeola_nigra     1    0
18      Centroscymnus_coelolepis     0    0
19        Zameus_squamulosus     0    0
20         Squatina_nebulosa     0    0
21  Etmopterus_gracilispinis     1    0
22        Oxynotus_paradoxus     0    0
23         Scymnodon_ringens     0    0
24          Squaliolus_aliae     1    0
25   Centrophorus_moluccensis     0    0
26     Etmopterus_splendidus     1    0
27       Echinorhinus_brucus     0    0
28                Deania_sp     0    0
> read.csv ("Lumi.csv") -> Lumi.dat
>   data.frame(Lumi.dat[,2:3]) -> LumiData
>  rownames(LumiData) <- Lumi.dat[,1]
> attach(LumiData)
> Lumi2
 [1] 1 0 1 1 0 1 1 1 1 0 1 0 0 0 0 0 1 0 0 0 1 0 0 1 0 1 0 0
> names(Lumi2) <- rownames(LumiData)
> name.check(172.tre, LumiData) -> LumiOverlap
> read.tree ("172.tre") -> tree1
> plot (tree1)
> name.check(tree1, LumiData) -> LumiOverlap
> plot (LumiOverlap)
> name.check(tree1, LumiData)
```

```
[1] "OK"
> Lumi2Label <- character(length(tree1$tip.label))
> names(Lumi2Label) <- names(Lumi2)
> Lumi2Label[Lumi2==0] <- "red"
> Lumi2Label[Lumi2==1] <- "blue"
> plot(tree1, cex=0.5, label.offset=2)
> points(rep(104, length(tree1$tip.label)), 1:length(tree1$tip.label), pch=22,
bg=Lumi2Label[tree1$tip.label], cex=0.5, lwd=0.25)
> ace(Lumi2, tree1, type="discrete") -> tree1Lumi2
> tree1Lumi2

    Ancestral Character Estimation

Call: ace(x = Lumi2, phy = tree1, type = "discrete")

    Log-likelihood: -8.745127

Rate index matrix:
  0 1
0 . 1
1 1 .

Parameter estimates:
 rate index estimate std-err
          1    0.001   6e-04

Scaled likelihoods at the root (type '...$lik.anc' to get them for all nodes):
          0           1
0.994056219 0.005943781

#plotting results on timetree estimated with BEAST
> plot(tree1, show.node.label=FALSE, label.offset=3, cex=0.4)
> points(rep(105, length(tree1$tip.label)), 1:length(tree1$tip.label), pch=22,
bg=Lumi2Label[tree1$tip.label], cex=0.5, lwd=0.25)
> nodelabels(pie=tree1Lumi2$lik.anc, cex=0.5)
```

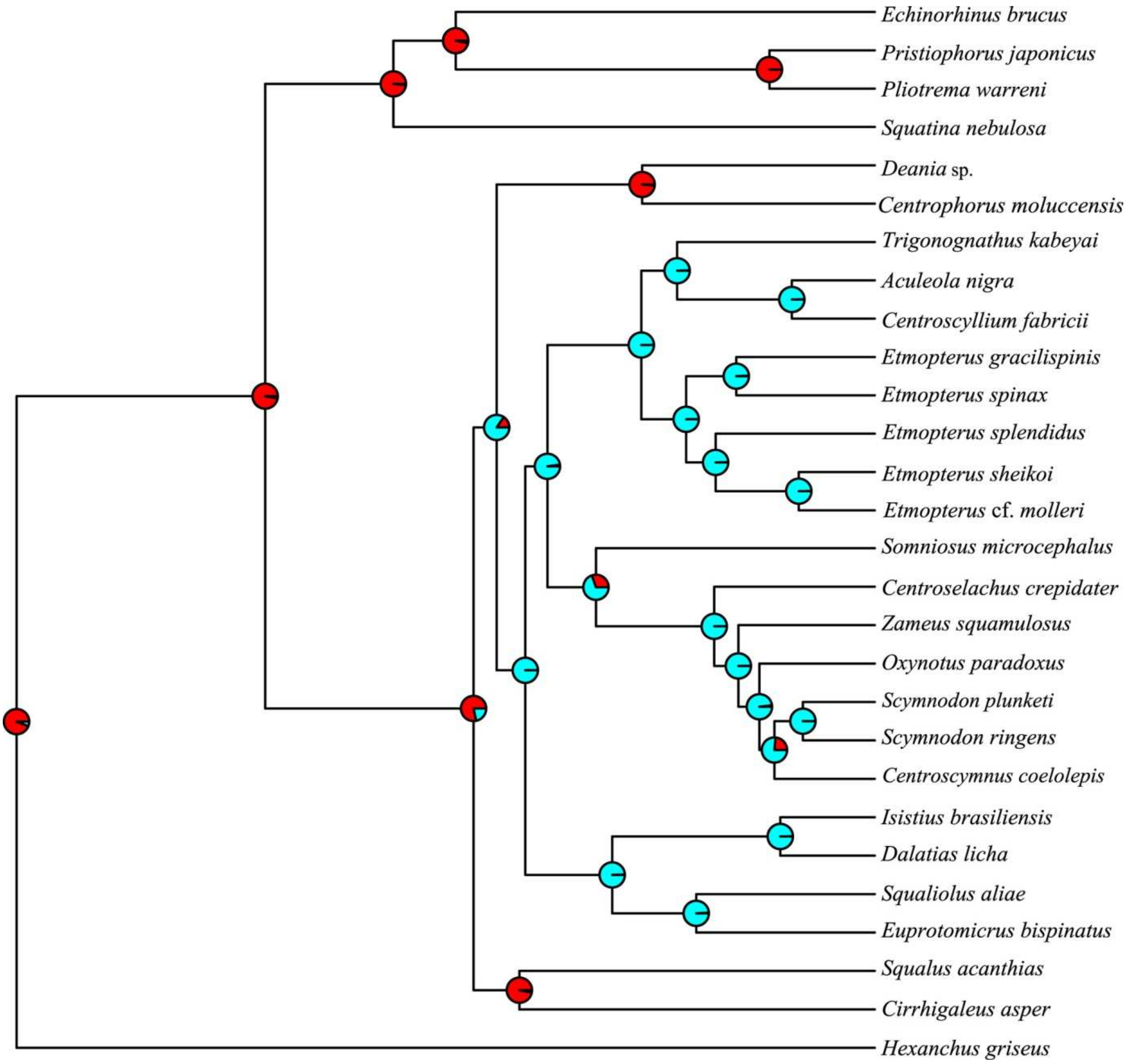


Figure S9: ancestral bioluminescence of Squaliformes. Pie charts indicate the probability that ancestral taxa are luminescent (blue) or not (red). Families Etmopteridae and Dalatiidae were coded as luminous as well as genera *Centroselachus*, *Centroscymnus*, *Scymnodon*, and *Zameus* within Somniosidae referring to [8].