

## **SUPPORTING INFORMATION (SI)**

### **Mercury isotopes as tracers of ecology and metabolism in two sympatric shark species**

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## **Additional discussion**

### **Hg bioaccumulation and biomagnification**

1 At the intraspecific level, muscle THg increased with both length and body mass in the two  
2 species, highlighting the well-documented bioaccumulation of MeHg in sharks (Kiszka et al.,  
3 2015; Le Bourg et al., 2019; McKinney et al., 2016). Although tiger sharks were larger than bull  
4 sharks (SI Table S2), their size would correspond to a mean age of 5 years (Meyer et al., 2014),  
5 compared to more than 15 years for bull sharks (Natanson et al., 2014). This longer  
6 accumulation time represents a first explanation for the higher THg levels in bull sharks.  
7 Within a same species, muscle THg concentration was correlated neither to muscle  $\delta^{15}\text{N}$  nor  
8  $\delta^{13}\text{C}$  values. Although this absence of correlation may be due to the narrow ranges in  $\delta^{15}\text{N}$  and  
9  $\delta^{13}\text{C}$  values, the same result was previously observed for these species (Matulik et al., 2017;  
10 Rumbold et al., 2014). However, at the interspecific level, Hg concentration generally  
11 increases with trophic position in sharks due to biomagnification (Le Bourg et al., 2019;  
12 McKinney et al., 2016). Since both  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  values also increase with trophic position  
13 (Hussey et al., 2015), the higher mercury concentration found in bull sharks compared to tiger  
14 sharks can be explained by a higher trophic position, highlighted by higher  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$   
15 values (SI Table S2). This is in agreement with a previous study on stomach contents that  
16 showed resource partitioning between these two sympatric species in the area (Le Croizier et  
17 al., 2019; Trystram et al., 2016). Alternatively, the higher  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  found in bull sharks  
18 compared to tiger sharks could reflect a higher trophic position or a more coastal habitat use,  
19 since  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  can increase with both the trophic position and coastal habits (Le Croizier  
20 et al., 2016). In a previous study in the southwestern Indian Ocean, higher Hg concentrations  
21 were found in pelagic and deep-sea species compared to coastal species (Le Bourg et al.,

22 2019), suggesting higher mercury bioavailability due to its enhanced methylation in oceanic  
23 deep waters (Blum et al., 2013). In the present study, bull sharks showed higher Hg  
24 concentrations than tiger sharks, despite a more inshore and shallower habitat use (Afonso  
25 and Hazin, 2015; Carlson et al., 2010; Domingo et al., 2016; Werry et al., 2012). Overall, our  
26 results suggest that the higher THg concentrations found in bull sharks are likely to reflect  
27 bioaccumulation and/or biomagnification processes, respectively due to higher age and  
28 trophic position.

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**Table S1:** Summary (mean  $\pm$  2SD) of  $\delta^{202}\text{Hg}$  and  $\Delta^{199}\text{Hg}$  values measured in secondary and certified reference materials (CRM). References:

- (1) Blum, J. D.; Popp, B. N.; Drazen, J. C.; Anela Choy, C.; Johnson, M. W. Methylmercury Production below the Mixed Layer in the North Pacific Ocean. *Nature Geosci* **2013**, *6* (10), 879–884. <https://doi.org/10.1038/ngeo1918>.
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CRM	n	$\delta^{202}\text{Hg} (\text{\textperthousand})$	$\Delta^{199}\text{Hg} (\text{\textperthousand})$	Reference
UM-Almadén	13	-0.64 $\pm$ 0.16	-0.02 $\pm$ 0.08	This study
		-0.57 $\pm$ 0.05	-0.02 $\pm$ 0.03	Blum et al., 2013 <sup>1</sup>
ETH-Fluka	12	-1.37 $\pm$ 0.18	0.11 $\pm$ 0.08	This study
		-1.44 $\pm$ 0.12	0.07 $\pm$ 0.05	Jiskra et al., 2017 <sup>2</sup>
TORT 3	4	0.05 $\pm$ 0.08	0.66 $\pm$ 0.04	This study
		0.13 $\pm$ 0.12	0.69 $\pm$ 0.10	Li et al., 2016 <sup>3</sup>
BCR 464	4	0.71 $\pm$ 0.10	2.28 $\pm$ 0.06	This study
		0.69 $\pm$ 0.06	2.40 $\pm$ 0.06	Blum et al., 2013 <sup>1</sup>
IAEA 436	2	0.67 $\pm$ 0.08	1.48 $\pm$ 0.02	This study

**Table S2:** Summary (mean  $\pm$  standard deviation) of the different variables measured in the two shark species (n=20 for each species). “Length” refers to the total length. All chemical analyzes were performed on muscle tissue. Significant differences between the two species are indicated by \*\* p < 0.01, \*\*\* p < 0.001.

Species	Mass (kg)	Length (cm)	$\delta^{13}\text{C} (\text{\textperthousand})$	$\delta^{15}\text{N} (\text{\textperthousand})$	THg ( $\text{ng}\cdot\text{g}^{-1}$ )	$\delta^{202}\text{Hg} (\text{\textperthousand})$	$\Delta^{199}\text{Hg} (\text{\textperthousand})$	$\Delta^{200}\text{Hg} (\text{\textperthousand})$	$\Delta^{201}\text{Hg} (\text{\textperthousand})$
Bull shark	149 $\pm$ 72	254 $\pm$ 48	-16.00 $\pm$ 0.58***	14.22 $\pm$ 0.47***	4148 $\pm$ 3069**	1.91 $\pm$ 0.52***	2.08 $\pm$ 0.16***	0.08 $\pm$ 0.04	1.67 $\pm$ 0.15***
Tiger shark	234 $\pm$ 132	323 $\pm$ 57***	-17.01 $\pm$ 0.49	13.27 $\pm$ 0.54	3186 $\pm$ 1252	1.23 $\pm$ 0.20	1.76 $\pm$ 0.14	0.06 $\pm$ 0.04	1.43 $\pm$ 0.14

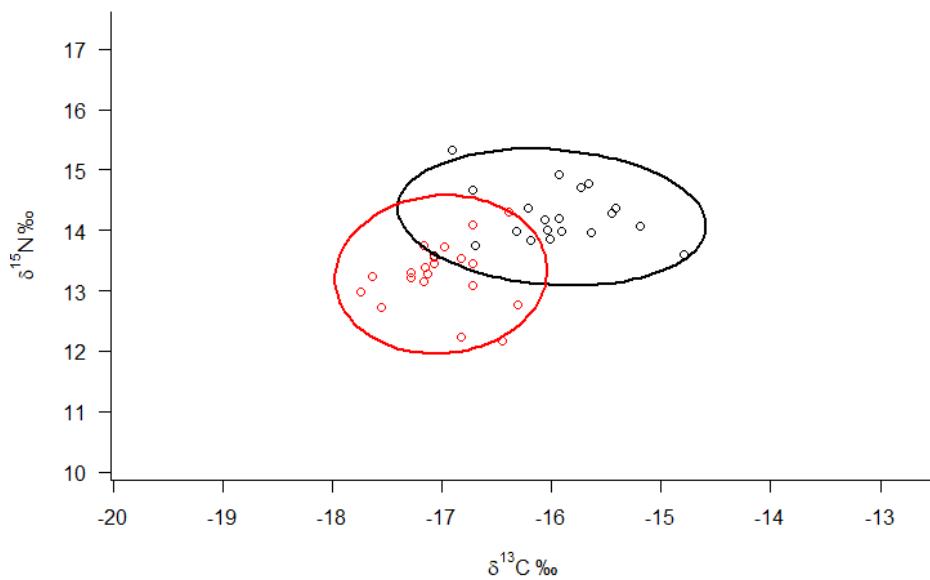
**Table S3:** Summary (mean ± standard deviation) of  $\delta^{202}\text{Hg}$  and  $\Delta^{199}\text{Hg}$  values measured in the muscle of some prey species. Different letters indicate significant differences between species ( $\delta^{202}\text{Hg}$ : ANOVA,  $\Delta^{199}\text{Hg}$ : KW). Habitat and diet are also reported according to previous studies.

- (1) Sepulveda, C. A.; Aalbers, S. A.; Ortega-Garcia, S.; Wegner, N. C.; Bernal, D. Depth Distribution and Temperature Preferences of Wahoo (*Acanthocybium Solandri*) off Baja California Sur, Mexico. *Mar Biol* 2011, **158** (4), 917–926. <https://doi.org/10.1007/s00227-010-1618-y>.
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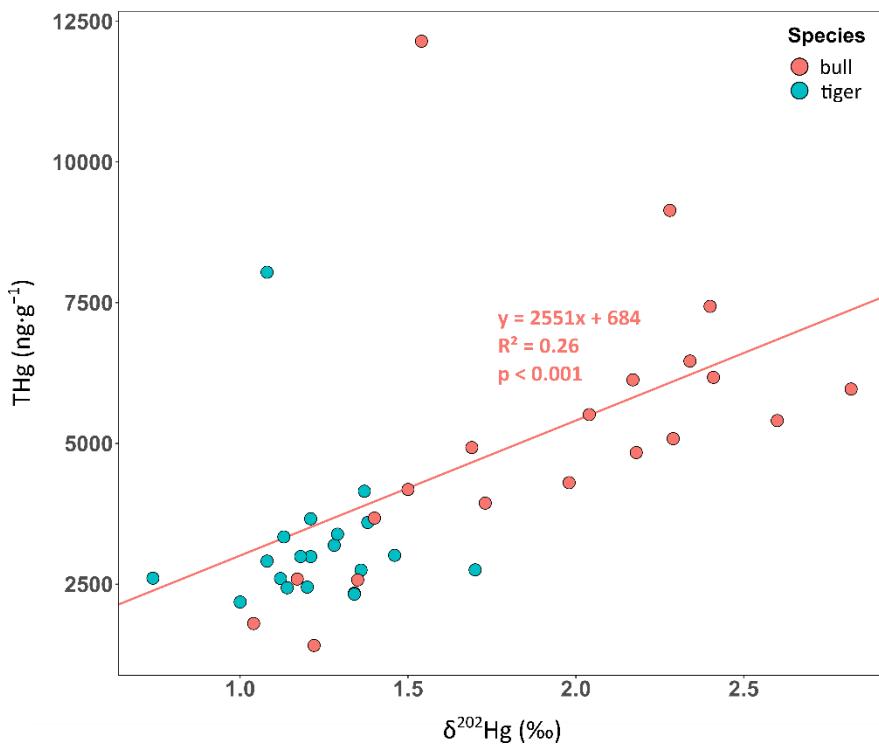
Common name	Scientific name	n	$\delta^{202}\text{Hg} (\text{\textperthousand})$	$\Delta^{199}\text{Hg} (\text{\textperthousand})$	Habitat (vertical / horizontal)	Diet	Reference
Wahoo	<i>Acanthocybium solandri</i>	3	0.74 ± 0.16 <sup>ab</sup>	2.60 ± 0.40 <sup>b</sup>	Epipelagic / Nearshore	juveniles of reef species	1, 2
Giant trevally	<i>Caranx ignobilis</i>	5	0.55 ± 0.12 <sup>b</sup>	2.14 ± 0.18 <sup>ab</sup>	Epipelagic / Nearshore	coastal fish	2, 3
Lantern fish	<i>Ceratoscopelus warmingii</i>	9	0.54 ± 0.16 <sup>b</sup>	1.93 ± 0.07 <sup>ac</sup>	Mesopelagic / Offshore	pelagic crustaceans	4
Deepwater snapper	<i>Etelis coruscans</i>	3	0.17 ± 0.04 <sup>c</sup>	1.77 ± 0.13 <sup>c</sup>	Mesopelagic / Nearshore	myctophids	2, 3
Skipjack tuna	<i>Katsuwonus pelamis</i>	3	0.50 ± 0.09 <sup>bc</sup>	2.34 ± 0.40 <sup>b</sup>	Epipelagic / Offshore	epipelagic crustaceans and squids	2, 5
Great barracuda	<i>Sphyraena barracuda</i>	3	1.05 ± 0.13 <sup>a</sup>	2.46 ± 0.12 <sup>b</sup>	Epipelagic / Nearshore	coastal and epipelagic species	2, 6
Yellowfin tuna	<i>Thunnus albacares</i>	3	0.20 ± 0.09 <sup>c</sup>	2.27 ± 0.23 <sup>b</sup>	Epipelagic / Offshore	epipelagic crustaceans and squids	2, 5
Lyretail grouper	<i>Variola louti</i>	3	1.03 ± 0.12 <sup>a</sup>	2.72 ± 0.04 <sup>b</sup>	Epipelagic / Nearshore	reef fish	7

**Table S4:** Summary (mean  $\pm$  standard deviation) of  $\delta^{202}\text{Hg}$  and  $\Delta^{199}\text{Hg}$  values measured in some prey species. Significant differences between two studies for a same species are indicated by \*  $p < 0.01$ , \*\*  $p < 0.001$ .

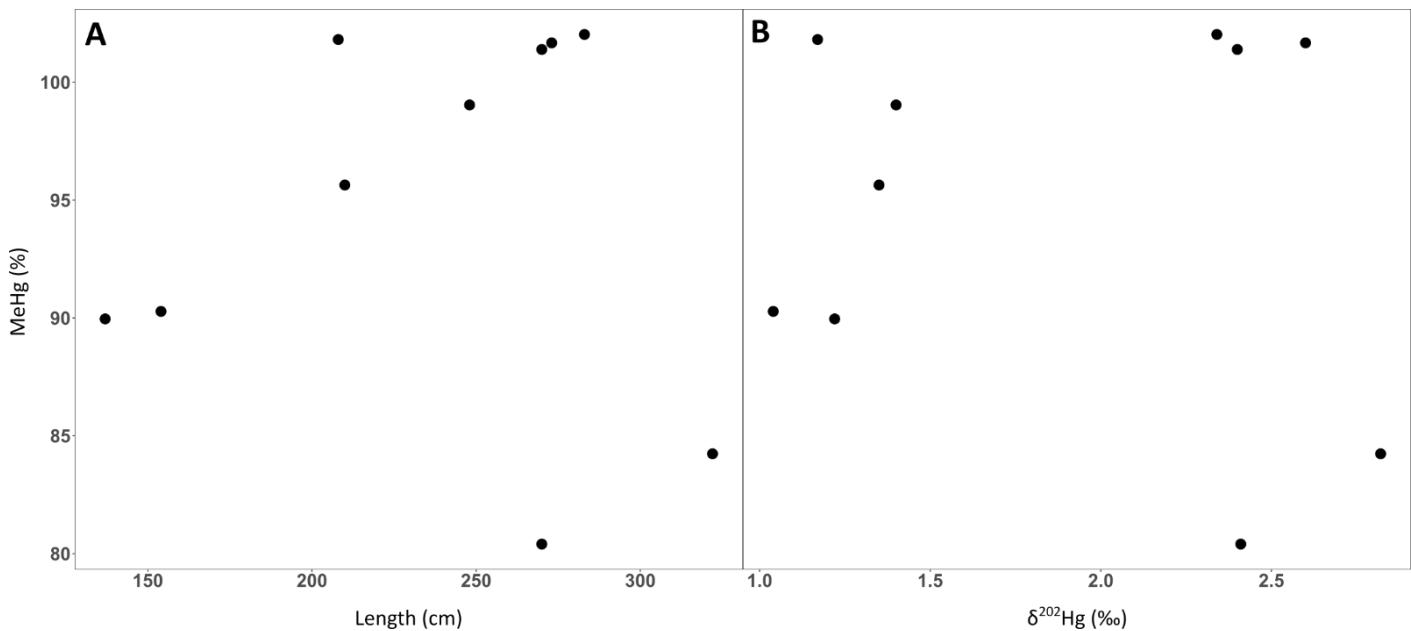
Species	$\delta^{202}\text{Hg} (\text{\textperthousand})$	$\Delta^{199}\text{Hg} (\text{\textperthousand})$	Reference
<i>Caranx ignobilis</i>	$0.55 \pm 0.12$	$2.14 \pm 0.17$	This study
	$-0.03 \pm 0.62$	$1.65 \pm 0.67$	Sackett <i>et al.</i> , 2017
<i>Etelis coruscans</i>	$0.08 \pm 0.17$	$1.66 \pm 0.24$	This study
	$0.52 \pm 0.14^*$	$1.94 \pm 0.15$	Sackett <i>et al.</i> , 2017
<i>Katsuwonus pelamis</i>	$0.50 \pm 0.09$	$2.34 \pm 0.39$	This study
	$0.62 \pm 0.04$	$2.71 \pm 0.08$	Blum <i>et al.</i> , 2013
<i>Thunnus albacares</i>	$0.20 \pm 0.09$	$2.27 \pm 0.22$	This study
	$0.87 \pm 0.11^{**}$	$2.76 \pm 0.03^*$	Blum <i>et al.</i> , 2013



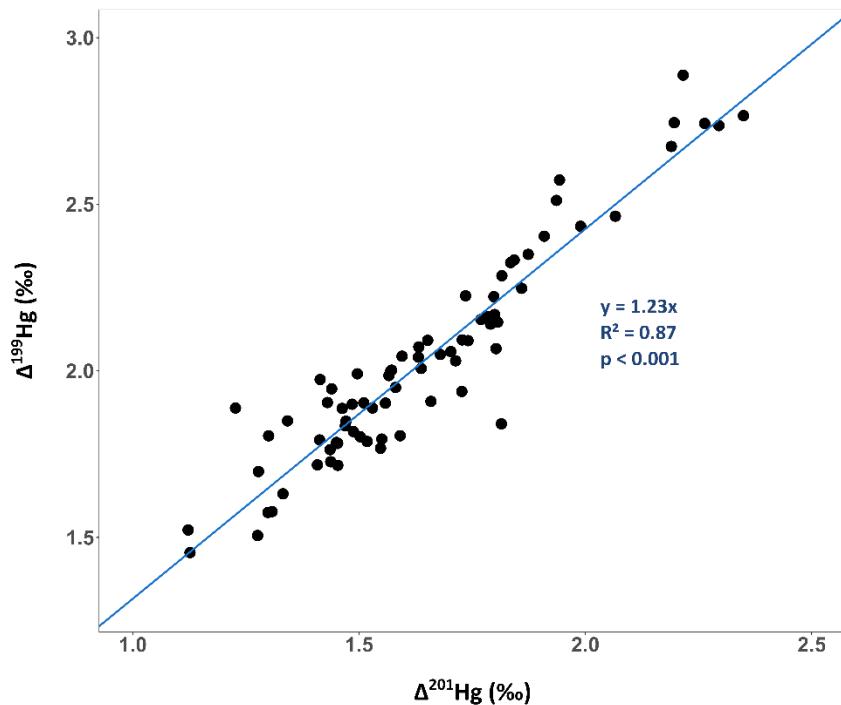
**Figure S1:** Trophic niche area based on carbon ( $\delta^{13}\text{C}$ ) and nitrogen ( $\delta^{15}\text{N}$ ) isotopes. Isotopic niche area was  $5.23\text{\textperthousand}^2$  for the bull shark (dark ellipse) and  $4.19\text{\textperthousand}^2$  for the tiger shark (red ellipse) (SEA<sub>B</sub> with 95% credible interval). The area overlap between species was 18%.



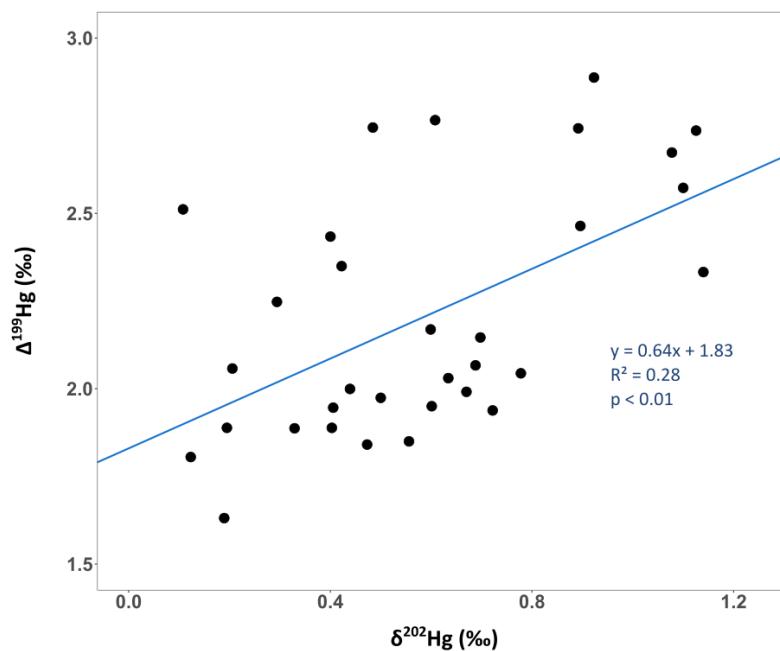
**Figure S2:** Total mercury concentrations (THg) versus  $\delta^{202}\text{Hg}$  values for the bull shark (black dots) and the tiger shark (red dots). THg was correlated with  $\delta^{202}\text{Hg}$  only for the bull shark.



**Figure S3:** MeHg fraction versus body length (A) and  $\delta^{202}\text{Hg}$  values (B) in a subset of 10 bull sharks. No correlation was found between Hg speciation and shark length or MDF signature.



**Figure S4:**  $\Delta^{199}\text{Hg}$  versus  $\Delta^{201}\text{Hg}$  in all the organisms (sharks and prey) from the coastal ecosystem of Reunion Island. Data fits a linear regression and the line represents the  $\Delta^{199}\text{Hg}/\Delta^{201}\text{Hg}$  slope of the samples (taking intercept at origin).



**Figure S5:** Muscle  $\Delta^{199}\text{Hg}$  versus  $\delta^{202}\text{Hg}$  values of individual teleost prey. Sharks are not included since they are characterized by high MDF values due to Hg demethylation. Data fits a linear regression.

## Global dataset

Nº	Species	Sex	Weight (Kg)	Total length (cm)	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)	THg (ng·g <sup>-1</sup> )	MeHg (%)	$\delta^{202}\text{Hg}$ (‰)	$\Delta^{199}\text{Hg}$ (‰)	$\Delta^{200}\text{Hg}$ (‰)	$\Delta^{201}\text{Hg}$ (‰)	$\delta^{13}\text{C}$ corrected (‰)	$\delta^{15}\text{N}$ corrected (‰)
228	<i>Acanthocybium solandri</i>		8.2	115	-16.75	13.03	538		0.61	2.77	0.01	2.35		
283	<i>Acanthocybium solandri</i>		6.3	102	-17.56	12.66	1128		0.70	2.15	0.13	1.81		
290	<i>Acanthocybium solandri</i>		7	106	-15.59	11.95	3291		0.92	2.89	0.04	2.22		
32	<i>Caranx ignobilis</i>		8		-16.69	12.73	2858		0.69	2.07	0.05	1.80		
34	<i>Caranx ignobilis</i>		15		-16.48	12.52	2842		0.63	2.03	0.02	1.71		
51	<i>Caranx ignobilis</i>		15		-14.66	12.47	1356		0.60	2.17	-0.07	1.80		
93	<i>Caranx ignobilis</i>			65	-16.53	12.38	752		0.40	2.43	-0.01	1.99		
119	<i>Caranx ignobilis</i>				-15.78	12.54	3341		0.44	2.00	0.06	1.57		
115	<i>Carcharhinus leucas</i>	M	31.72	154	-15.22	13.11	1801	90	1,04	1.99	0.02	1.57	-15.42	14.37
85	<i>Carcharhinus leucas</i>	M	68.81	208	-15.84	12.50	2591	102	1,17	1.81	0.05	1.30	-16.01	13.86
116	<i>Carcharhinus leucas</i>	F	20.32	137	-15.61	12.60	1411	90	1,22	2.09	0.13	1.73	-15.91	13.98
101	<i>Carcharhinus leucas</i>	M	75.51	210	-16.02	12.46	2575	96	1,35	1.90	0.10	1.48	-16.19	13.82
118	<i>Carcharhinus leucas</i>	M	128.83	248	-14.99	12.75	3675	99	1,40	1.79	0.03	1.52	-15.20	14.07
126	<i>Carcharhinus leucas</i>	M	90.19	225	-16.55	12.35	4183		1,50	2.07	0.04	1.63	-16.70	13.74
102	<i>Carcharhinus leucas</i>	M	134.82	250	-16.17	12.65	12146		1,54	2.00	0.06	1.57	-16.33	13.99
133	<i>Carcharhinus leucas</i>	F	162.6	270	-15.77	12.84	4928		1,69	2.15	0.11	1.77	-16.07	14.18
90	<i>Carcharhinus leucas</i>	F	110.39	235	-14.48	12.14	3940		1,73	2.40	0.05	1.91	-14.79	13.60
108	<i>Carcharhinus leucas</i>	F	162.6	270	-16.62	14.23	4303		1,98	2.04	0.08	1.63	-16.91	15.32
122	<i>Carcharhinus leucas</i>	F	256.9	309	-15.15	12.98	5513		2,04	2.16	0.07	1.78	-15.46	14.29
134	<i>Carcharhinus leucas</i>	F	214.6	291	-15.34	12.58	6130		2,17	1.90	0.11	1.56	-15.64	13.96
106	<i>Carcharhinus leucas</i>	M	136.9	264	-15.54	13.51	4838		2,18	2.09	0.03	1.74	-15.73	14.70
131	<i>Carcharhinus leucas</i>	F	200.74	291	-15.64	13.76	9140		2,28	2.29	0.14	1.82	-15.94	14.93
111	<i>Carcharhinus leucas</i>	F	244.34	307	-15.74	12.64	5084		2,29	2.01	0.10	1.64	-16.04	14.01
128	<i>Carcharhinus leucas</i>	F	187.4	283	-16.43	13.43	6463	102	2,34	2.14	0.11	1.79	-16.72	14.66
129	<i>Carcharhinus leucas</i>	M	156.1	270	-15.76	12.90	7435	101	2,40	2.23	0.21	1.74	-15.93	14.20
132	<i>Carcharhinus leucas</i>	M	156.1	270	-15.47	13.59	6174	80	2,41	2.22	0.07	1.80	-15.66	14.77
103	<i>Carcharhinus leucas</i>	M	151.68	273	-16.06	13.10	5406	102	2,60	2.32	0.08	1.83	-16.22	14.36
124	<i>Carcharhinus leucas</i>	F	293.82	322	-16.79	12.12	5966	84	2,82	2.05	0.08	1.68	-17.08	13.58
M3	<i>Ceratoscopelus warmingii</i>			6.8	-20.12	9.97	65		0.47	1.84	-0.06	1.81		
M5	<i>Ceratoscopelus warmingii</i>			6.2	-19.59	8.96	80	95	0.40	1.89	0.02	1.23		
M6	<i>Ceratoscopelus warmingii</i>			7.1	-19.68	10.11	76	8	0.78	2.04	-0.07	1.59		
R1	<i>Ceratoscopelus warmingii</i>			6.9	-18.88	8.11	80		0.67	1.99	0.16	1.50		
R10	<i>Ceratoscopelus warmingii</i>			6.6	-18.92	8.82	67		0.50	1.97	-0.01	1.41		
R2	<i>Ceratoscopelus warmingii</i>			6.5	-18.91	9.09	58		0.56	1.85	-0.02	1.34		
R3	<i>Ceratoscopelus warmingii</i>			6.5	-19.21	9.56	129		0.72	1.94	0.09	1.73		
R6	<i>Ceratoscopelus warmingii</i>			6.3	-19.01	8.74	79		0.33	1.89	-0.10	1.46		
R7	<i>Ceratoscopelus warmingii</i>			6.4	-18.89	9.14	75		0.41	1.95	-0.08	1.44		
317	<i>Etelis coruscans</i>			54	-18.07	12.28	291		0.12	1.81	0.05	1.59		

318	<i>Etelis coruscans</i>			54	-18.12	12.38	295		0.19	1.63	0.06	1.33		
322	<i>Etelis coruscans</i>			33	-18.04	12.13	305		0.19	1.89	0.07	1.53		
88	<i>Galeocerdo cuvier</i>	M	125.3	297	-16.92	11.88	2989		1.21	1.76	0.01	1.44	-17.13	13.29
91	<i>Galeocerdo cuvier</i>	F	203.6	320	-16.27	11.18	2338		1.34	1.52	0.00	1.12	-16.46	12.16
92	<i>Galeocerdo cuvier</i>	F	469	406	-17.33	12.23	3596		1.38	1.72	0.01	1.45	-17.29	13.29
93	<i>Galeocerdo cuvier</i>	F	375.4	364	-16.74	11.24	2754		1.70	2.09	0.12	1.65	-16.83	12.22
96	<i>Galeocerdo cuvier</i>	F	214.2	320	-16.07	11.74	3190		1.28	1.77	0.09	1.55	-16.30	12.76
97	<i>Galeocerdo cuvier</i>	F	239.3	341	-17.05	12.48	2750		1.36	1.73	0.07	1.44	-17.07	13.56
98	<i>Galeocerdo cuvier</i>	M	116.3	288	-17.71	11.34	2606		0.74	1.45	0.07	1.13	-17.74	12.98
104	<i>Galeocerdo cuvier</i>	M	41.9	197	-16.95	12.07	2909		1.08	1.58	0.08	1.31	-17.15	13.39
105	<i>Galeocerdo cuvier</i>	F	72.5	237	-17.66	11.69	2183		1.00	1.91	0.07	1.66	-17.55	12.72
107	<i>Galeocerdo cuvier</i>	M	163.7	310	-16.39	13.31	2991		1.18	1.78	0.17	1.45	-16.72	14.08
109	<i>Galeocerdo cuvier</i>	F	383.4	379	-16.61	12.38	3340		1.13	1.80	0.11	1.55	-16.73	13.45
110	<i>Galeocerdo cuvier</i>	F	104.7	260	-16.60	12.04	2448		1.20	1.82	0.06	1.49	-16.72	13.09
113	<i>Galeocerdo cuvier</i>	M	104.5	277	-16.96	11.64	8041		1.08	1.72	0.07	1.41	-17.16	13.15
114	<i>Galeocerdo cuvier</i>	F	440.18	400	-16.74	12.46	3662		1.21	1.79	0.03	1.45	-16.83	13.53
117	<i>Galeocerdo cuvier</i>	M	362.1	380	-16.85	12.17	4151		1.37	1.83	0.01	1.47	-17.07	13.45
119	<i>Galeocerdo cuvier</i>	M	325.5	367	-16.97	12.72	3012		1.46	1.78	0.02	1.45	-17.16	13.75
121	<i>Galeocerdo cuvier</i>	F	216.9	325	-17.77	12.17	2322		1.34	1.80	0.07	1.50	-17.63	13.23
123	<i>Galeocerdo cuvier</i>	M	409.5	397	-16.73	12.68	3388		1.29	1.90	0.03	1.43	-16.98	13.73
127	<i>Galeocerdo cuvier</i>	F	171.7	309	-17.32	12.16	2601		1.12	1.70	0.07	1.28	-17.28	13.22
130	<i>Galeocerdo cuvier</i>	F	156.1	294	-16.18	13.18	2436		1.14	1.79	0.05	1.41	-16.39	14.31
318	<i>Katsuwonus pelamis</i>		18	47	-18.08	11.22	1068		0.42	2.35	0.01	1.87		
416	<i>Katsuwonus pelamis</i>		13	39	-17.43	10.47	762		0.48	2.74	0.06	2.20		
450	<i>Katsuwonus pelamis</i>		17	49	-16.65	11.16	625		0.60	1.95	0.03	1.58		
285	<i>Sphyraena barracuda</i>		19	138	-16.27	11.64	3299		1.10	2.57	-0.03	1.94		
300	<i>Sphyraena barracuda</i>		11.64	115	-17.01	10.62	4110		1.14	2.33	0.04	1.84		
311	<i>Sphyraena barracuda</i>		8.67	105	-16.96	11.64	3826		0.90	2.46	0.03	2.07		
41	<i>Thunnus albacares</i>		24	103	-16.80	12.40	882		0.29	2.25	0.02	1.86		
234	<i>Thunnus albacares</i>		13	89	-16.90	12.80	431		0.11	2.51	0.03	1.94		
357	<i>Thunnus albacares</i>		30	128	-17.10	11.10	861		0.21	2.06	0.13	1.70		
197	<i>Variola louti</i>		19	54	-16.78	10.22	656		1.13	2.74	0.13	2.29		
227	<i>Variola louti</i>		12	42	-16.89	10.90	771		1.08	2.67	0.01	2.19		
337	<i>Variola louti</i>		26	52	-16.76	10.78	607		0.89	2.74	0.06	2.26		