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Marine and Freshwater Research

Supplementary Material

Biological parameters and spatial segregation patterns in sharks from the North Aegean Sea, Greece

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Table S1. Maturity stages for viviparous elasmobranchs (adopted from MEDITS Working Group 2017).

Sex	Gonad aspect	Maturation state	Maturity	Stage
I	Sex not distinguished by naked eye.	Undetermined	Immature	0
M	Claspers flexible and shorter than pelvic fins. Testes small. Sperm ducts straight and thread-like.	Immature	Immature	1
F	Ovaries barely visible or small, whitish; undistinguishable ovarian follicles. Oviducal (nidamental) gland may be slightly visible. Uterus is thread-like and narrow.			
M	Claspers slightly more robust but still flexible. Claspers as long as or longer than pelvic fins. Testes enlarged; in sharks testes start to segment; Sperm ducts developing and beginning to coil (meander).	Developing	Immature	2
F	Ovaries enlarged with small follicles (oocytes) of different size. Some relatively larger yellow follicles may be present. Ovaries lack atretic follicles. Developing oviducal gland and uterus.			
M	Claspers fully formed, skeleton hardened, rigid and generally longer than pelvic fins. Testes greatly enlarged; in sharks testes are fully segmented. Sperm ducts tightly coiled and filled with sperm.	Spawning capable	Mature	3a
F	Large ovaries with enlarged yolk follicles all of about the same size so that they can be easily distinguished. Oviducal gland and uterus developed without yolky matter, embryos and not dilated.	Capable to re- produce		
M	Description similar to stage 3a, however with clasper glands dilated, often swollen and reddish (occasionally open). Sperm often present in clasper groove or glans. On pressure sperm is observed flowing out of the cloaca or in the sperm ducts.	Actively spawning	Mature	3b
F	Uteri well filled and rounded with yolk content (usually candle shape). In general segments cannot be distinguished and embryos cannot be observed.	Early pregnancy	Maternal	
F	Uteri well filled and rounded, often with visible segments. Embryos are always visible, small and with a relatively large yolk sac.	Mid pregnancy	Maternal	3c
F	Embryos fully formed, yolk sacs reduced or absent. Embryos can be easily measured and sexed.	Late pregnancy	Maternal	3d
M	Claspers fully formed, similar to stage 3. Testes and sperm ducts shrunken and flaccid.	Regressing	Mature	4
F	Ovaries shrunken without follicle development and with atretic (degenerating) follicles. The oviducal glands diameter may be reducing. Uterus appears much enlarged, collapsed, empty and reddish.	Regressing	Mature	4a
F	Ovary with small follicles in different stages of development with the presence of atretic ones. Uterus enlarged with flaccid walls. Oviducal gland distinguishable.	Regenerating (mature)	Mature	4b

Table S2. Maturity stages for oviparous elasmobranchs (adopted from MEDITS Working Group 2017).

Sex	Gonad aspect	Maturation state	Stage	MEDITS
I	Sex not distinguished by naked eye.	Undetermined	0	0
F	Ovary is barely discernible with small isodiametric eggs. Distal part of oviducts is thick-walled and whitish. The nidamental glands are less evident.	Immature or virgin	1	1
M	Claspers are small and flaccid and do not reach the posterior edge of the pelvic fins. Sperm ducts not differentiated. Testis small and narrow.			
F	Whitish or few yellow maturing eggs are visible in the ovary. The distal part of oviducts (uterus) is well developed but empty. The nidamental glands are small.	Maturing	2	2
M	Claspers are larger, but skeleton still flexible. They extend to the posterior edge of the pelvic fins. Sperm ducts well-developed eventually beginning to meander.			
F	Ovaries contain yellow eggs (large yolk eggs). The nidamental glands are enlarged and oviducts are distended.	Mature	3a	3
M	Claspers extends well beyond the posterior edge of the pelvic fin and their internal structure is generally hard and ossified. Testis greatly enlarged. Sperm ducts meandering over almost their entire length.			
F	Ovary walls transparent. Oocytes of different sizes, white or yellow. Nidamental glands large. Egg-cases more or less formed in the oviducts (Extruding Stage).	Mature or extruding–active	3b	
M	Clasper longer than tips of posterior pelvic fin lobes, skeleton hardened with axial cartilages hardened and pointed. Sperm ducts largely. Sperm flowing on pressure from cloaca (Active Stage).			
F	Ovary walls transparent. Oocytes of different sizes, white or yellow. Oviducts appear much enlarged, collapsed and empty. The nidamental glands diameter are reducing.	Resting	4a	4
M	Clasper longer than tips of posterior pelvic fin lobes, skeleton hardened with axial cartilages still hardened. Sperm ducts empty and flaccid.			
F	Ovaries full of small follicles similar to stage 2, enlarged oviducal glands and uterus	Regenerating	4b	

Table S3. Allometric growth parameters for female and male individuals for *Scyliorhinus canicula*, *Galeus melastomus*, *Squalus blainville*, and *Etmopterus spinax* in the North Aegean Sea.

Species	Sex	Growth	*T*	s.e.	*P*
Scyliorhinus canicula	Females	Positive	5.287	0.035	0
	Males	Isometric	0.295	0.034	0.76
Galeus melastomus	Females	Positive	6.221	0.037	0
	Males	Positive	10.341	0.033	0
Squalus blainville	Females	Positive	2.179	0.074	0.03
	Males	Isometric	-0.278	0.084	0.78
Etmopterus spinax	Females	Isometric	-0.306	0.040	0.75
	Males	Positive	2.216	0.032	0.02

Table S4. Analysis of covariance (ANCOVA) output for the total weight–total length relationship ($TW= aTL^{b}$).

Species	*F*	Pr(>*F*)
Scyliorhinus canicula	12.5	0.00041***
Galeus melastomus	5.68	0.017**
Squalus blainville	2.19	0.14
Etmopterus spinax	2.54	0.11

Significant differences are given by: ***, $P < 0.001$; **, $P < 0.01$.

Table S5. Sex Ratio for the most abundant shark species in the North Aegean Sea.

Species	Males	Females	Sex ratio	χ^2	*P*
Scyliorhinus canicula	800	785	0.98	0.14	0.706
Galeus melastomus	654	775	1.18	10.24	0.001
Squalus blainville	205	249	1.21	4.26	0.039
Etmopterus spinax	51	81	1.58	6.82	0.009

Table S6. Number of Individuals per Sampling year for *Scyliorhinus canicula*, *Galeus melastomus* and *Squalus blainville*.

	2014	2016	2018	2019	2020	2021
Scyliorhinus canicula	518	659	686	445	497	671
Galeus melastomus	647	548	789	521	148	399
Squalus blainville	36	80	150	171	154	171

Table S7. Model selection for the generalised additive model of sexual segregation according to sex for the most abundant species.

Sexual segregation	Model	d.f.	Sex	Year	s(D)	s(λ,φ)	s(D): Sex	s(λ,φ): Sex	AIC	DE(%)
Scyliorhinus canicula	**N~s(D)+s(λ,φ)+s(D: Sex)+s(λ,φ: Sex)+Sex+Year**	**20.89**	**1**	**5*** **	**2.69*** **	**6.62** **	**2.01**	**2.01**	**3696.57**	**28.8**
	N~s(λ,φ)+s(D)+s(D: Sex)+s(λ,φ: Sex)+Sex	15.78	1	-	2.59***	6.51***	2.01	1.01	3767.35	21.0
	N~s(λ,φ)+s(λ,φ: Sex)+Sex+Year	16.80	1	5***	-	6.55***	-	3.01	3934.69	14.2
	N~s(D)+s(D: Sex)+Sex+Year	11.82	1	5***	2.66***	-	1.00	-	3793.12	17.5
	N~s(D)+s(D: Sex)+Sex	6.82	1	-	2.67***	-	1.00	-	3844.77	10.9
	N~s(λ,φ)+s(λ,φ: Sex)+Sex	11.71	1	-	-	6.45***	-	3.01	3886.39	14.5
	N~Sex+Year	8.00	1	6***	-	-	-	-	3888.92	6.3
Galeus melastomus	**N~s(D)+s(λ,φ)+s(D: Sex)+s(λ,φ: Sex)+Sex+Year**	**20.69**	**1**	**5*** **	**2.73*** **	**6.01*** **	**1.00**	**3.00**	**1912.03**	**28.9**
	N~s(λ,φ)+s(D)+s(D: Sex)+s(λ,φ: Sex)+Sex	15.77	1	-	2.76***	6.18***	2.00	2.00	1935.86	21.3
	N~s(λ,φ)+s(λ,φ: Sex)+Sex+Year	16.64	1	5***	-	-	2.00	2.01	1929.32	23.2
	N~s(D)+s(D: Sex)+Sex+Year	11.99	1	5***	2.88***	-	1.00	-	1923.55	22.4
	N~s(D)+s(D: Sex)+Sex	6.95	1	-	3.96***	-	2.00	-	1953.52	12.7
	N~s(λ,φ)+s(λ,φ: Sex)+Sex	11.63	1	-	-	6.42***	-	2.00	1955.68	14.5
	N~Sex+Year	8.00	1	5***	-	-	-	-	1971.59	8.5
Squalus blainville	N~s(D)+s(λ,φ)+s(D: Sex)+s(λ,φ: Sex)+Sex+Year	24.69	1.00	5.00	2.21*	5.47**	1.85	5.96	911.02	36.2
	N~s(λ,φ)+s(D)+s(D: Sex)+s(λ,φ: Sex)+Sex	**19.59**	**1.00**	**-**	**2.25***	**5.27** **	**5.92**	**1.89**	**908.87**	**33.5**
	N~s(λ,φ)+s(λ,φ: Sex)+Sex+Year	20.54	1.00	5.00	-	5.58**	-	6.36	918.91	30.8
	N~s(D)+s(D: Sex)+Sex+Year	13.11	1.00	5.00	2.09	-	3.34***	-	944.57	15.7
	N~s(D)+s(D: Sex)+Sex	8.21	1.00	-	2.21	-	2.33	-	939.47	13.8
	N~s(λ,φ)+s(λ,φ: Sex)+Sex	15.44	1.00	-	-	5.34**	-	5.45	918.64	27.3
	N~Sex+Year	8.00	1.00	5.00	-	-	-	-	964.21	3.1

Estimated degrees of freedom for the Year, Sex and the two nonparametric terms are shown. Degrees of freedom (d.f.), AIC (Akaike information criterion), Deviance explained (DE, %). The best model is marked in bold. Significant differences are denoted by asterisks as ***, *P* < 0 .0001; **, *P* < 0.001; *, *P* < 0.01.

Table S8. Length at maturity (mm) and maximum total length (mm) across the Mediterranean Sea and the Atlantic Ocean for male and female *Scyliorhinus canicula*, *Galeus melastomus* and *Squalus blainville.*

Reference	Area	Maximum TL (mm)		Size at sexual maturity L_{50} (mm)	
		M	F	M	F
Galeus melastomus					
Present study	**North Aegean Sea**	525	550	401	463
Capapé and Zaouali (1977)	Mediterranean Tunisian coasts	420	470	>420	390–420
Compagno (1984b)	Eastern North Atlantic & Mediterranean	610	900	330–420	380–450
Que´ro (1984)	Eastern North Atlantic & Mediterranean	610	900	340–420	390–450
Tursi *et al.* (1993)	Ionian Sea	510	550	450	490
Costa *et al.* (2005)	Portuguese waters	640	670	490	560–590
Capapé *et al.* (2008)	France	>720	>720	430–460	450–500
Capapé *et al.* (2008)	Languedocian coasts	620	640	510–550	520–610
Ragonese *et al.* (2009)	Strait of Sicily	510	590	433	380
Marongiu *et al.* (2013)	Sardinia Seas	590	530	424	450
Metochis *et al.* (2016)	Gulf of Euboea & Corinth & North-Western Aegean	500	560	447	498
Scyliorhinus canicula					
Present study	**North Aegean Sea**	490	565	401	392
Ellis and Shackley (1997)	Bristol Channel	662	670	520	550
Ivory *et al.* (2005)	Irish waters	710	700	535	570
Lupi (2008)	Tyrrhenian Sea	505	490	402	405
Kousteni *et al.* (2010)	Aegean Sea	488	467	396	399
Finotto *et al.* (2015)	Strait of Sicily	495	480	385	368
Finotto *et al.* (2015)	Adriatic Sea	505	490	400	409
Squalus blainville					
Present study	**North Aegean Sea**	620	785	478	541
Cannizzaro *et al.* (1995)	Strait of Sicily	735	920	450–460	570–580
Sion *et al.* (2003)	Ionian Sea	664	785	–	601
Kousteni and Megalophonou (2011)	Aegean Sea	799	779	457	564
Marouani *et al.* (2012)	Gulf of Gabes	834	1000	523	625
Anastasopoulou *et al.* (2018)	Ionian Sea	1000	950	413	603
Marongiu *et al.* (2020)	Sardinia Island	615	840	491	604

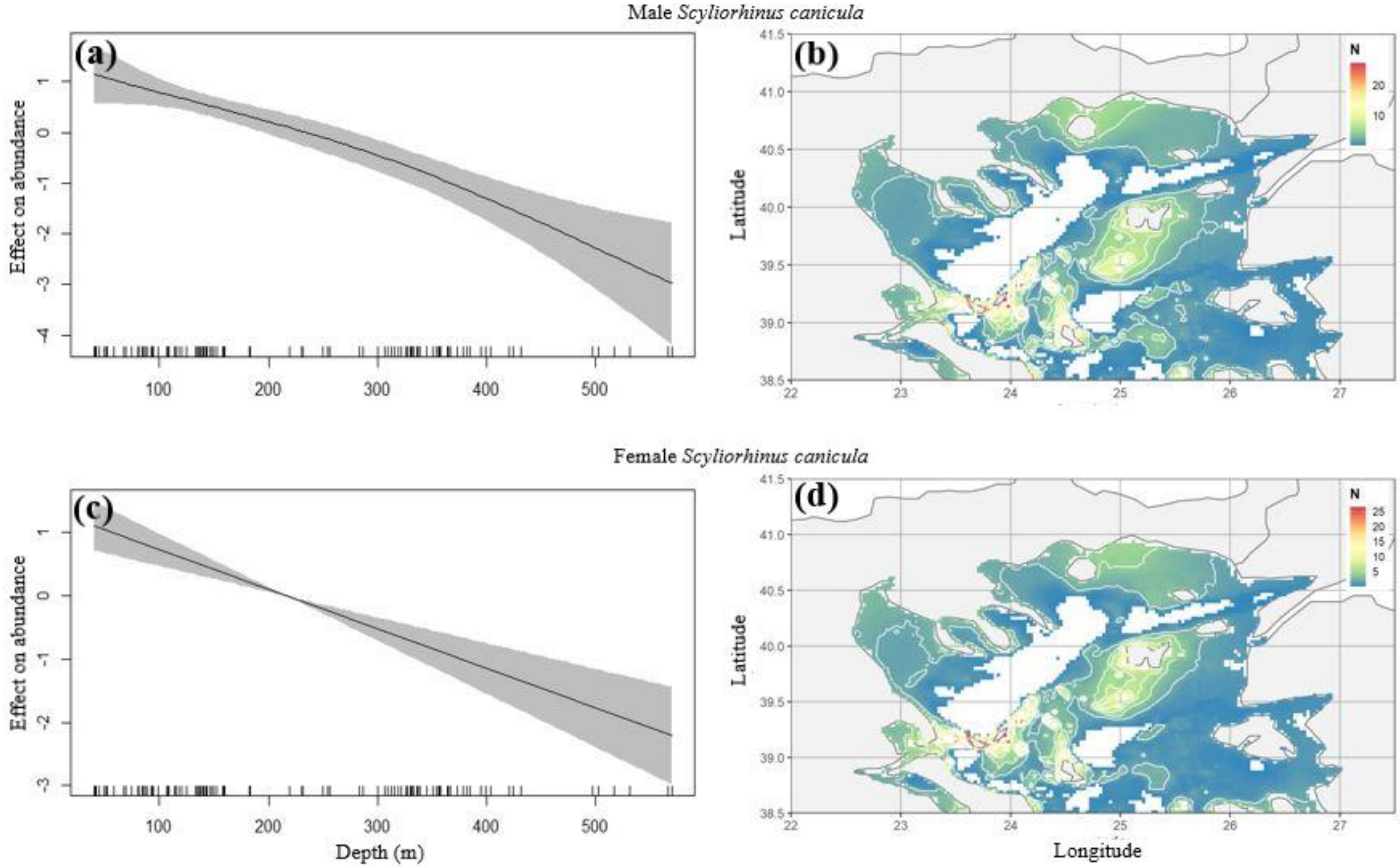

Figure S1. Effects of bottom depth and geographic position on *Scyliorhinus canicula* for male and female individuals, estimated from the generalised additive model. Shaded areas on the depth plots are 95% confidence intervals.

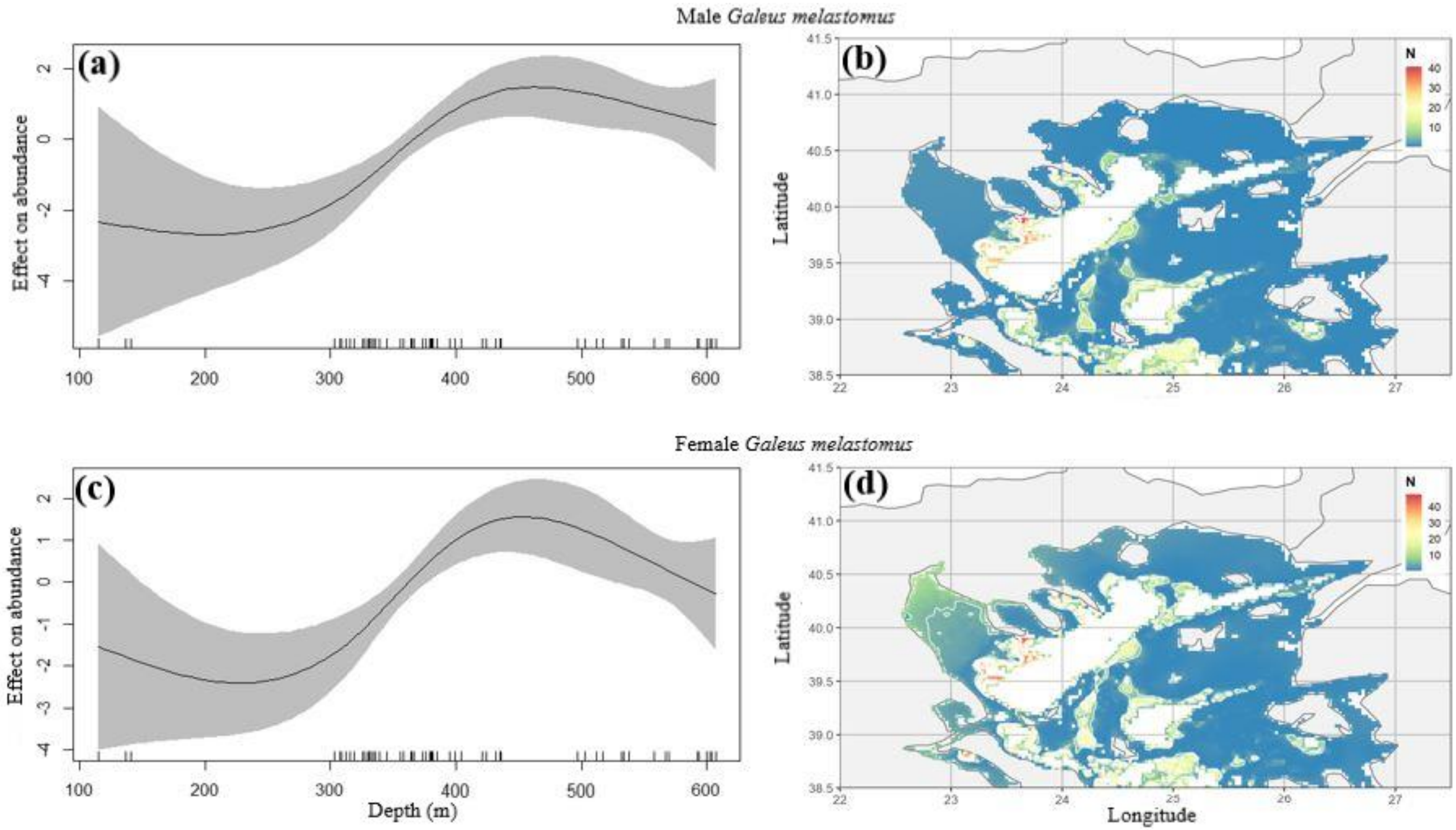

Figure S2. Effects of bottom depth and geographic position on *Galeus melastomus* for male and female individuals, estimated from the generalised additive model. Shaded areas on the depth plots are 95% confidence intervals.

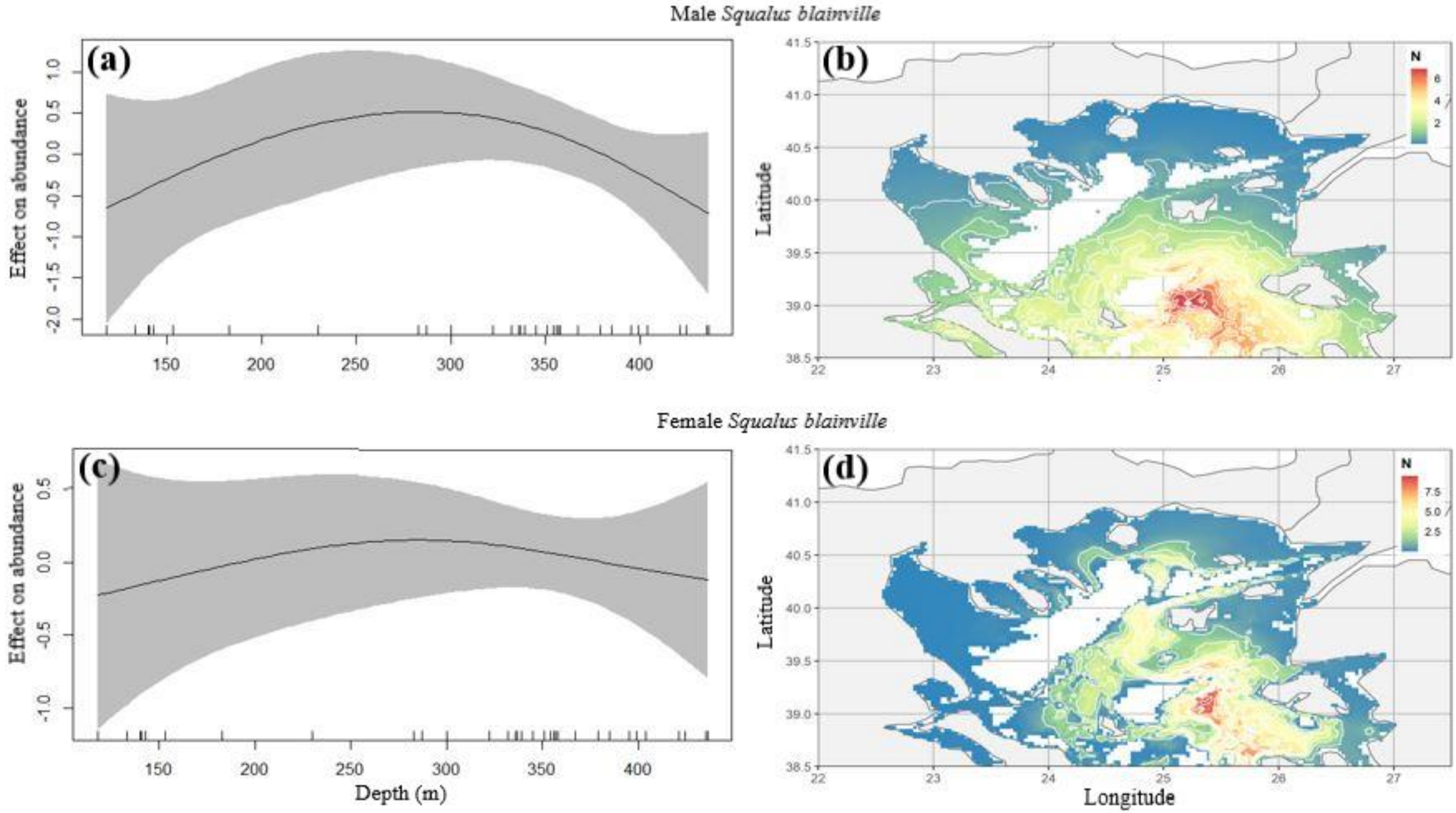

Figure S3. Effects of bottom depth and geographic position on *Squalus blainville* for male and female individuals, estimated from the generalised additive model. Shaded areas on the depth plots are 95% confidence intervals.

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