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R E S E A R C H   P A P E R

Feeding Ecology of Four Demersal Shark Species (*Etmopterus spinax, Galeus melastomus, Scyliorhinus canicula* and *Squalus blainville*) from the Eastern Aegean Sea

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Abstract

In this study, the diet composition and trophic ecology of four demersal chondrichthyan species; *Etmopterus spinax*, *Galeus melastomus*, *Scyliorhinus canicula* and *Squalus blainville* were studied in the eastern Aegean Sea. In the stomachs of the samples which mostly consisted of juvenile individuals, a total of 97 prey taxa were identified. Teleost fishes were the most important prey group. The diversity of stomach content ranged between 15 species in *E. spinax*. and 70 species in *S. canicula*. The dietary breadth of *G. melastomus* and *S. canicula* were found to be narrower than the other two species examined. In addition, high niche overlap scores were detected amongst the species. All of the examined species had trophic levels higher than 4; with the highest trophic level being 4.20 and belonging to *E. spinax*. Comparisons among calculated trophic levels by global methods and a regional weighted method, which is proposed in this study, showed that the regional method offers remarkable advantages that can be used to reduce the uncertainty of the estimations.

Introduction

The effects of changes in either one or more components within a food web may propagate through the entire system, resulting in changes in the abundance and web connectivity of other species (Bornatowski, Wosnick, do Carmo, Corrêa, & Abilhoa, 2014). Considering the ongoing changes in many elasmobranch populations worldwide, and the potential impacts on their prey and communities, developing an understanding of trophic relationships of elasmobranches is important to comprehend how marine systems are functioning (Vaudo, 2011).

It is known that about 88 cartilaginous species inhabit the Mediterranean Sea (FAOa, 2018; FAOb, 2018), and that 75% of these species are also found in the Aegean Sea (Eronat & Bizsel, 2015). This group contributes a considerable part of by-catch in Turkish

trawl fisheries (Gurbet, Akyol, Yalçın, & Özaydın, 2013; Soykan, Akgül, & Kınacıgil, 2016). According to Eronat and Özaydın (2011) at least 15 chondrichthyan species are regularly presented in the fishery of study area. Four shark species investigated in this study: velvet belly lantern shark *Etmopterus spinax* (Linneus, 1758), blackmouth catshark *Galeus melastomus* Rafinesque, 1810, lesser spotted dogfish *Scyliorhinus canicula* (Linneus, 1758), and longnose spurdog *Squalus blainville* (Risso, 1826), are among frequently caught elasmobranches in the Aegean Sea (Maravelias, Tserpes, Pantazi, & Peristeraki, 2012).

Sharks are mostly large predatory fishes and their trophic levels range from 3.1 to 4.7 based on the global data sets (Ebert & Bizzaro, 2007; Cortés, 1999). Their diet composition reveals important differences among European seas according to available studies. For instance, *S. canicula* consumes mainly crustaceans in the
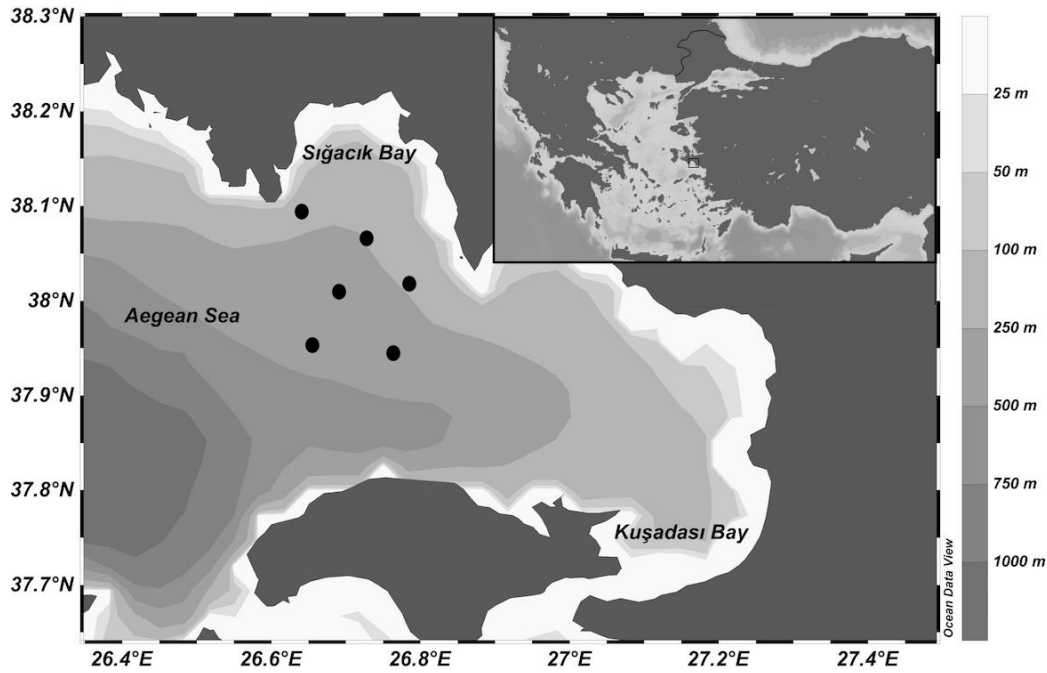
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Figure 1. Location of study area in the Aegean Sea, Eastern Mediterranean; black circles represent haul locations.

northern Atlantic (Ellis, Pawson, & Shackley, 1996), while in the western Mediterranean it also consumes polychaetas, teleosts and euphosid (Valls, Quetglas, Ordines, & Moranta, 2011). In general terms these same groups are also among the important food items for *E. spinax* (Bello, 1998; Fanelli, Rey, Torres, & Gil De Sola, 2009; Valls *et al.,* 2011) and *G. melastomus* (Anastasopoulou *et al.,* 2013; Valls *et al.,* 2011; Fanelli *et al.,* 2009; Özütemiz, Kaya, & Özaydın, 2009; Serena *et al.,* 2009 and references therein). To better understand sharks' trophic ecology, further studies are required from different parts of the Mediterranean Sea and its adjacent waters (Stergiou & Karpouzi, 2001; Neves, Figueiredo, Moura, Assis, & Gordo, 2007; Albo-Puigserver *et al*., 2015; Kousteni, Karachle, & Megalofonou, 2017a).

The aim of this study was to develop an understanding on the trophic ecology of four demersal elasmobranch species: *E. spinax*, *G. melastomus*, *S. canicula* and *S. blainville*. For this purpose, diet composition was identified, niche breadth, diet overlaps and trophic levels were estimated, and their ecological significances were subsequently compared and discussed for each species.

Materials and Methods

Samples belonging to four elasmobranch species: *E. spinax*, *G. melastomus*, *S. canicula* and *S. blainville* were collected with a commercial bottom trawler. A total of 15 hauls were performed in 2008 (April, May, June, September and November) and 3 hauls were performed in 2014 (April), at depths between 150 and 550 m, off Sigacik Bay (26.569° N - 37.939° E and 26.879°

N - 38.180° E), located in the eastern Aegean Sea (the eastern Mediterranean Sea) (Figure 1). A total of 2,174 specimens were collected and transported to the laboratory in a 4-8% formaldehyde solution. Total length and weights of individuals were measured and weighted to the nearest 0.1 cm and 0.01g using a measuring tape and electronic scale, respectively.

All prey items found in the stomach contents were identified to the lowest possible taxonomic level. Each prey item was weighed and recorded to the nearest 0.01 g using an electronic scale.

Analyses on diet comparisons were made between species. To evaluate the importance of each prey item, the percentage by number (N%), percentage by weight (W%), frequency of occurrence (FO%) and percentage index of relative importance (IRI%) were calculated (Hyslop, 1980) (see Eq. 1-5). For each species, percentage of empty stomachs were calculated from the ratio between the number of stomachs without prey items, and the total individuals examined.

$$N_i\% = \frac{N_i}{\sum_{i=1}^{n} N_i} 100 \qquad (1)$$

$$W_i\% = \frac{W_i}{\sum_{i=1}^{n} W_i} 100 \qquad (2)$$

$$FO_i\% = \frac{F_i}{S} 100 \qquad (3)$$

$$IRI_i = F_i\%(W_i\% + N_i\%) \qquad (4)$$

$$IRI_i\% = \frac{IRI_i}{\sum_{i=1}^{n} IRI_i} 100 \qquad (5)$$

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Where N_i is number of individual in prey category i; W_i is weight of prey category i; F_i is number of stomachs containing prey items in category i; S is the total number of full stomachs; IRI_i is index of relative importance of prey category i; n is the number of prey categories.

Cluster analysis was applied on the stomach contents of each examined species by using Euclidean distance with single linkage in order to evaluate similarities among feeding strategies.

Smith's (1982) index (Eq. 6) was chosen to assess the niche breadth for two main reasons. Firstly, this method takes into account the availability of prey groups, and secondly it is less sensitive to the selectivity of lesser important prey groups (Krebs, 2009).

$$FT = \Sigma(\sqrt{a_i p_i}) \tag{6}$$

Where FT is Smith's (1982) index; p_i is the proportion of individuals using prey category i; a_i is IRI% of prey category i to the total prey composition.

Levins' measure of niche breadth (Eq. 7) and Levins' standardized niche breadth were (Eq. 8) also calculated (Krebs, 2009).

$$\hat{B} = {}^1\!/_{\Sigma p_i^2} \tag{7}$$

$$\hat{B}_A = {}^{(\hat{B} - 1)}\!/_{(n - 1)} \tag{8}$$

Where $\hat{B}$ is Levins' measure of niche breadth; $\hat{B}_A$ is Levins' standardized niche breadth; and n is the number of prey categories.

Due to the various sample sizes, Morisita index was chosen to calculate niche overlap among the investigated species by using main taxa to avoid bias as suggested by Krebs (2009) (Eq. 9).

$$C = \frac{2 \Sigma_i^n p_{ij} p_{ik}}{\Sigma_i^n p_{ij} \left[{}^{(n_{ij}-1)}\!/_{(N_j-1)} \right] + \Sigma_i^n p_{ik} \left[{}^{(n_{ik}-1)}\!/_{(N_k-1)} \right]} \tag{9}$$

Where C is Morisita's index of niche overlap between species j and k; p_{ij} is the proportion of individuals using prey category i. to total prey composition used by species j; p_{ik} is the proportion of individuals using prey category i to the total prey composition used by species k; n_{ij} is the number of individuals of species j that used prey category i; n_{ik} is number of individuals of species k that used prey category i; N_j and N_k are the total number of species j and k, respectively.

According to the contribution of each taxon from different groups, trophic levels of the prey groups were adapted based on local prey composition. When available, the trophic level of each identified species was taken from FishBase (http://www.fishbase.org, Froese & Pauly, 2016) or SeaLifeBase (http://www.sealifebase.org, Palomares & Pauly, 2017). When the trophic levels were not available, group values were used from Pauly *et al.,* (2000) or from Ebert and Bizzarro (2007) and the references therein. Following

Table 1. Standardized diet compositions and their trophic levels for the Aegean Sea

Group code	Description	Trophic level
AMPH	Amphipods and isopods	3.18
CEPH	Octopi, cuttlefishes and unidentified cephalopods	3.21
CHOND	Chondrichthyan fishes	3.78
DECA	Decapods	2.93
EUPH	Euphausiids and mysids	2.25
FISH	Teleost and agnathan fishes	3.26
INVERT	Other invertebrates and unidentified invertebrates	2.17
MOLL	Mollusk (excluding cephalopods) and unidentified mollusks	2.10
OCRUST	Other crustaceans and unidentified crustaceans	2.60
POLY	Polychaetes and other marine worms	2.30
SQUID	Squids	4.06

Table 2. Total number of sampled specimens from each shark species (N), number of stomachs with food (N of FS), percentage of empty stomachs (PES), range and averages of length (L), and average of weight (W) measurements

Species	N	N of FS	PES	Range L (mm)	Average L (mm)	Average W (g)
Etmopterus spinax	129	97	25%	86-317	171	28
Galeus melastomus	441	336	24%	89-450	152	17
Scyliorhinus canicula	1296	241	81%	44-512	198	54
Squalus blainvillei	308	166	46%	162-705	267	131

Table 3. Standardized diet compositions and percentage index of relative importance for each shark species

Group	Taxon	Etmopterus spinax					Galeus melastomus					Scyliorhinus canicula					Squalus blainville				
		Fo%	N %	W %	IRI	IRI %	Fo%	N %	W %	IRI	IRI %	Fo%	N %	W %	IRI	IRI %	Fo%	N %	W %	IRI	IRI %
	Amphipoda	1.05	0.81	0.05	0.91	0.02	0.28	0.23	0.01	0.07											
AMPH	Isopoda						1.13	0.93	0.20	1.28	0.04	0.26	0.21	0.08	0.07	0.01					
	AMPH	1.05	0.81	0.05	0.91	0.02	1.41	1.17	0.22	1.95	0.03	0.26	0.21	0.08	0.07	0.00					
	Cephalopoda	36.84	45.53	38.95	3112.37	74.41	5.63	4.44	3.71	45.93	1.59	10.26	9.17	4.91	144.34	10.07	6.02	6.04	17.97	144.66	6.58
	Octopodidae						0.28	0.23	0.02	0.07		0.26	0.21		0.05						
	Pteroctopus tetracirrhus											0.26	0.21	2.93	0.80	0.06					
	Sepia officinalis											0.26	0.21	0.13	0.09	0.01					
	Sepia orbignyana											0.26	0.21	0.04	0.06						
CEPH	*Sepia sp.*											0.51	0.42	0.39	0.41	0.03					
	Sepietta neglecta											0.26	0.21	0.33	0.14	0.01					
	Sepietta oweniana											0.26	0.42	0.84	0.32	0.02					
	Sepietta sp.	1.05	0.81	2.35	3.33	0.08						1.79	1.46	1.90	6.03	0.42	1.20	1.65	1.30	3.55	0.16
	Sepiolidae						1.41	1.17	1.05	3.12	0.11	0.77	1.04	1.71	2.11	0.15	1.20	1.10	1.50	3.13	0.14
	CEPH	37.89	46.34	41.30	3321.22	58.73	7.32	5.84	4.78	77.82	1.12	14.87	13.54	13.18	397.34	8.62	8.43	8.79	20.77	249.28	5.59
	Chondrichthyes																0.60	0.55	0.76	0.79	0.04
CHOND	*Raja sp.*						0.28	0.23	0.08	0.09											
	Scyliorhinus canicula											0.51	0.42	0.81	0.63	0.04					
	CHOND						0.28	0.23	0.08	0.09	0.00	0.51	0.42	0.81	0.63	0.01	0.60	0.55	0.76	0.79	0.02
	Alpheus sp.											0.77	0.63	0.67	1.00	0.07	1.20	1.10	0.28	1.66	0.08
	Aristeomorpha folicea						0.56	0.47	0.15	0.35	0.01	0.51	0.42	1.14	0.80	0.06					
	Brachyura											0.26	0.21	0.14	0.09	0.01	0.60	0.55		0.33	0.02
	Chlorotoccus crassicornis											0.26	0.21	0.01	0.06						
	Galathea sp.											0.26	0.21	0.01	0.06						
	Goneplax rhomboides											0.26	0.42	0.19	0.16	0.01					
	Macropipus sp.											0.26	0.21	0.03	0.06						
	Munida rutllanti											0.77	0.63	0.86	1.14	0.08	0.60	0.55	0.12	0.40	0.02
	Natantia						2.82	2.34	1.21	10.00	0.35										
	Pagurus sp.											0.26	0.21	0.06	0.07		0.60	0.55	0.15	0.42	0.02
	Panaediae						0.28	0.23	0.07	0.09											
	Pandalus sp.						0.28	0.23	0.22	0.13											
DECA	*Parapenaeus longirostris*						0.56	0.47	0.29	0.43	0.01	8.46	9.58	11.86	181.48	12.66	5.42	5.49	5.70	60.71	2.76
	Parthenope massana											0.26	0.21	0.16	0.09	0.01					
	Pasiphea sivado	6.32	5.69	2.44	51.37	1.23															
	Penaeus kerathurus											2.05	2.08	2.26	8.91	0.62					
	Philocheras sp.						0.28	0.23	0.04	0.08											
	Plesionika edwarsi											3.59	3.33	1.15	16.09	1.12					
	Plesionika martia	1.05	0.81	1.61	2.55	0.06															
	Plesionika sp						5.35	4.91	4.37	49.65	1.72	2.05	2.08	2.02	8.43	0.59	2.41	3.30	1.00	10.36	0.47
	Pontocaris sp.						0.28	0.23	0.05	0.08		0.26	0.21	0.08	0.07	0.01	0.60	0.55	0.11	0.40	0.02
	Processa sp.	1.05	0.81	0.31	1.18	0.03						1.03	0.83	0.41	1.27	0.09	1.20	1.10	0.19	1.56	0.07
	Solenocera membranacea						0.85	1.17	0.39	1.31	0.05	0.26	0.21	0.11	0.08	0.01	0.60	0.55	0.28	0.50	0.02
	Synalpheus sp.											0.51	0.42	0.11	0.27	0.02	0.60	0.55	0.05	0.36	0.02
	DECA	8.42	7.32	4.37	98.38	1.74	11.27	10.28	6.80	192.45	2.77	22.05	22.08	21.27	955.93	20.74	13.86	14.29	7.89	307.26	6.89
	Euphasiacea											0.26	0.21	0.06	0.07						
EUPH	Mysidacea	3.16	6.50	0.22	21.24	0.51	9.58	21.50	1.14	216.77	7.51	7.18	15.63	0.94	118.96	8.30	2.41	3.85	0.06	9.42	0.43
	EUPH	3.16	6.50	0.22	21.24	0.38	9.58	21.50	1.14	216.77	3.13	7.44	15.83	1.01	125.21	2.72	2.41	3.85	0.06	9.42	0.21
	Antonogadus megalokynodon																1.20	1.10	0.77	2.25	0.10
	Argentina sphyraena						0.28	0.23	0.11	0.10		2.31	1.25	3.11	10.05	0.70	1.81	1.65	1.74	6.13	0.28
	Arnoglossus laterna											0.51	0.42	1.75	1.11	0.08					
FISH	*Arnoglossus rueppelli*											0.51	0.42	0.21	0.32	0.02					
	Buglossidium luteum											0.26	0.21	1.22	0.37	0.03					
	Capros aper											0.26	0.21	0.09	0.08	0.01					
	Chlorophtalmus agassizi						1.41	1.17	1.34	3.54	0.12										
	Citharus linguatula											0.26	0.21	2.57	0.71	0.05					
	Clupeidae											0.26	0.21	0.02	0.06						

Table 3. Continued

	Diplodus annularis										0.26	0.42	2.50	0.75	0.05					
	Engraulis encrasicholus										0.51	0.42	1.46	0.96	0.07					
	Gadella maraldi						0.56	0.47	0.47	0.53	0.02										
	Gadiculus argenteus						0.28	0.23	0.20	0.12		3.08	3.54	6.81	31.86	2.22	4.22	3.85	4.45	34.98	1.59
	Glossanodon leioglossus																1.20	1.10	3.86	5.97	0.27
	Gnathophis mystax											0.51	0.42	1.94	1.21	0.08	0.60	0.55	6.06	3.98	0.18
	Gobius niger											0.26	0.21	1.22	0.37	0.03					
	Gonostoma denudatum	1.05	0.81	0.70	1.59	0.04															
	Hymenocephalus italicus						0.85	0.70	0.58	1.09	0.04	1.03	0.83	1.75	2.65	0.19	1.20	1.10	0.29	1.67	0.08
	Lampanyctus crocodilus						0.85	0.70	1.30	1.69	0.06	0.26	0.21	0.18	0.10	0.01					
	Lesueurigobius friesii											0.51	0.42	0.47	0.46	0.03					
	Macrouridae						2.54	2.10	37.97	101.59	3.52						1.20	1.10	0.57	2.01	0.09
	Merluccius merluccius											0.51	0.42	2.05	1.26	0.09	0.60	0.55	3.30	2.32	0.11
	Myctophidae	11.58	8.94	16.47	294.30	7.04	7.32	7.01	9.82	123.29	4.27	1.54	1.67	4.26	9.12	0.64	0.60	0.55	0.22	0.47	0.02
	Nezumia sp.						0.56	0.47	1.02	0.84	0.03										
	Phycis blennoides											0.26	0.21	0.03	0.06		1.20	1.10	4.04	6.20	0.28
	Phycis phycis						0.28	0.23	0.71	0.27	0.01										
	Pleuronectiformes																0.6	0.55	0.53	0.65	0.03
	Scorpaenidae											0.26	0.21	0.71	0.23	0.02					
	Teleostei (Unidentified)	16.84	13.01	13.90	453.22	10.84	34.08	30.14	26.30	1923.80	66.67	21.03	18.75	19.07	795.17	55.49	21.08	19.23	17.46	773.53	35.20
	Trachinus draco											0.26	0.21		0.05						
	Vinciguerria attenuata	2.11	2.44	6.44	18.70	0.45															
	Zeus faber											0.26	0.21	0.15	0.09	0.01					
	FISH	31.58	25.20	37.52	1980.77	35.02	49.01	43.46	79.84	6043.33	87.12	34.87	31.04	51.57	2880.86	62.50	35.54	32.42	43.29	2690.78	60.34
	Actinaria (Unidentified)											0.26	0.21	0.61	0.21	0.01					
	Anthozoa (Unidentified)																1.20	3.30	0.24	4.27	0.19
	Antodon mediterranea											0.51	0.42	0.03	0.23	0.02	1.20	1.10	0.59	2.04	0.09
INVERT	Echinodermata											0.26	0.21	0.05	0.07						
	Hexacorallia						0.28	0.23	0.02	0.07											
	Siphonophora											0.26	0.21	0.30	0.13	0.01					
	Tunicata	1.05	0.81	5.07	6.19	0.15						0.26	0.42	1.22	0.42	0.03					
	INVERT	1.05	0.81	5.07	6.19	0.11	0.28	0.23	0.02	0.07	0.00	1.54	1.46	2.22	5.65	0.12	2.41	4.40	0.84	12.61	0.28
MOLL	Bivalvia (Unidentified)											0.26	0.21	0.01	0.05						
	MOLL											0.26	0.21	0.01	0.05	0.00					
	Crustacea (Unidentified)	14.74	11.38	2.49	204.50	4.89	18.87	15.65	5.41	397.46	13.77	6.15	5.00	1.37	39.18	2.73	28.31	27.47	10.31	1069.71	48.67
	Cumacean											0.51	0.63	0.01	0.32	0.02					
OCRUST	*Squilla mantis*											3.33	2.71	2.89	18.65	1.30					
	Squilla massavensis											0.77	0.63	1.61	1.72	0.12					
	Squilla sp.											2.05	1.88	0.65	5.19	0.36					
	OCRUST	14.74	11.38	2.49	204.50	3.62	18.87	15.65	5.41	397.46	5.73	12.82	10.83	6.53	222.56	4.83	28.31	27.47	10.31	1069.71	23.99
POLY	Polycheata (Unidentified)											3.08	2.50	1.65	12.78	0.89	3.01	3.30	1.98	15.89	0.72
	POLY											3.08	2.50	1.65	12.78	0.28	3.01	3.30	1.98	15.89	0.36
	Abralia veranyi						0.56	0.47	0.17	0.36	0.01	0.77	0.63	0.12	0.57	0.04					
	Allouteuthis media						0.28	0.23	0.05	0.08											
	Allouteuthis sp.																0.60	0.55	0.58	0.68	0.03
	Allouteuthis subulata											0.26	0.21	0.45	0.17	0.01					
SQUID	*Heteroteuthis dispar*						0.56	0.47	0.54	0.57	0.02										
	Illex coindetii	1.05	0.81	2.81	3.81	0.09						0.26	0.21	0.08	0.07	0.01	2.41	2.20	5.50	18.54	0.84
	Loligo vulgaris											1.03	0.83	1.05	1.93	0.13	1.20	1.10	2.42	4.24	0.19
	Pyroteuthis margoritifera						0.56	0.47	0.95	0.80	0.03										
	Todarodes sagittatus	1	1	6	7	0											1	1	4	3	0
	Todaropsis eblanea																0.6	0.55	1.36	1.15	0.05
	SQUID	2.11	1.63	8.98	22.32	0.39	1.97	1.64	1.72	6.62	0.10	2.31	1.88	1.69	8.23	0.18	5.42	4.95	14.11	103.30	2.32

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from this stage, all taxa found in the examined stomachs were then classed as the prey categories of Ebert and Bizzarro (2007). IRI% was used to calculate the proportional contribution of each taxon within a group. The contribution of each taxon and their trophic levels were then used to calculate the weighted average trophic level of each standardized prey group (Table 1). Afterwards, trophic levels of examined species (TL) were calculated by using Equation 10.

$$TL = 1 + (\sum_{j=1}^{n}(IRI\%)_j * TL_j) \qquad (10)$$

Where TL_j is the trophic level of each prey category j; $IRI\%_j$ is the percentage in index of relative importance per prey category j.

In addition, the trophic level values of each species were also estimated by using the methods of Ebert and Bizzarro (2007) as well as Cortés (1999) in order to compare both local and global trophic levels.

Results

Stomach Contents

During the study period a total of 2,174 specimens belonging to four investigated species were sampled. The total number of full stomachs were 97, 166, 241 and 336 for E. spinax, *S. blainvillei, S. canicula*, and *G. melastomus*, respectively. The lowest empty stomach percentage (24%) was observed in *G. melastomus*, with the highest percentage (81%) found in *S. canicula* (Table 2). The number of examined stomachs and descriptive statistics of the length measurements of each shark species are also given in Table 2. A total of 97 prey taxa were identified in the study, and all identified taxa were classified under 11 categories according to their trophic levels. The diet diversity was discovered to be highest in *S. canicula* with a total of 70 taxa (in 11 groups), and lowest in *E. spinax* with 15 taxa (in 8 groups). *G. melastomus* and *S. blainville* fed on 33 and 36 taxa belonging to 9 groups, respectively (Table 3).

The main prey taxa consumed by *E. spinax* were cephalopods and teleost fishes. More specifically, Myctopids and unidentified crustaceans were found to constitute an important part of the diet. The contributions of species from other prey groups were considered to be negligible (Table 3).

Teleost fishes were also identified as being dominant in the diet of *G. melastomus*. While unidentified teleost species showed the highest contribution to IRI%, the contributions of Macrouridae and Myctophidae were also relatively higher than other groups. Unidentified crustaceans and mysids (in the euphasiid group) were other important food items, with IRI% more than 5% (Table 3).

Although the stomach contents of *S. canicula* and *S. blainville* showed similar compositions (Figure 2), the IRI ratios of main prey taxa showed differences between the species. Results from *S. canicula* displayed unidentified teleost species as their dominant prey. On the other hand, this same group of teleost species were also the second dominant prey group for *S. blainville.* Relatively important prey species was a teleost, *Gadiculus argentatus,* along with *Parapenaus longirostris,* Unidentified Crustaceans and Cephalopods in the diets of both species (Table 3).

Niche Breath, Niche Overlap and Trophic Level

Smith's (1982) index for niche breadth indicated that the dietary breadths of *G. melastomus* and *S. canicula* were narrower than the other two examined
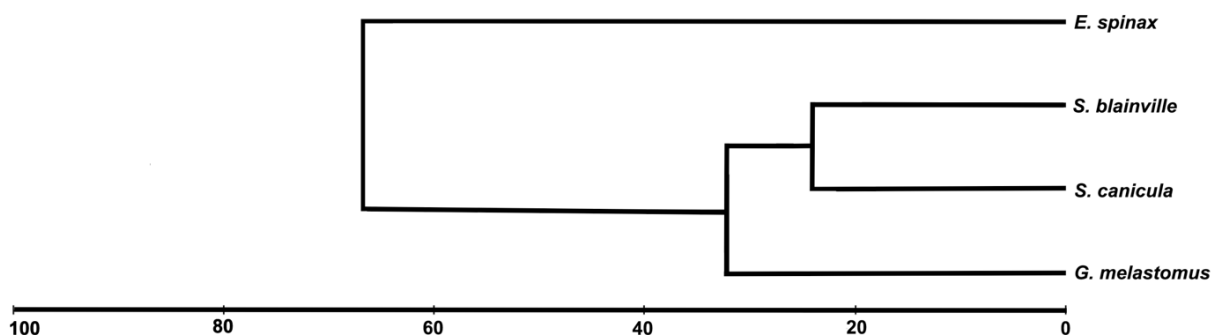

Figure 2. Cluster analysis dendogram of the examined species based on Euclidian similarity distance of their diets.

Table 4. Morisita's niche overlap values among the species. All values show significant overlap between the species

	E. spinax	G. melastomus	S. canicula	S. blainville
E. spinax				
G. melastomus	0.79			
S. canicula	0.87	0.93		
S. blainville	0.80	0.96	0.95	

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species, with the highest index score was seen in *S. blainville*. In addition to Smith's (1982) index, Levins' measures of niche breadth ranged from 3.65 (*E. spinax*) to 4.63 (*S. canicula*) and Levins's standardized niche breadth ranged from 0.29 (*G. melastomus*) to 0.40 (*S. blainville*) (Figure 3).

The results of Morisita's niche overlap analysis amongst the species showed that the maximum level in overlap was observed between *S. blainville* and *G. melastomus* (0.96). Niche overlaps between *S. blainville* – *S. canicula*, and *S. canicula* – *G. melastomus* were determined at

0.95 and 0.93, respectively (Table 4). The least overlap was detected between *E. spinax* and the other examined species. The overlap analyses indicated that the food tendencies of four species were considerably similar to each other.

The trophic levels of examined species ranged from 4.08 (*S. blainville*) to 4.20 (*E. spinax*) with an average value of 4.15±0.6 (±standard deviation, n=4). Trophic level calculations using global methods were given in Table 5.

Discussion

Stomach Content

In accordance with previous studies in adjacent regions from Mediterranean Sea, teleosts fishes, crustaceans and cephalopods are the main prey groups for the four shark species examined here, though their relative importance changes (Bello, 1998; Kabasakal, 2002; Olaso *et al.,* 2005; Neves *et al.,* 2007; Fanelli *et al.,* 2009; Özütemiz *et al.,* 2009; Serena *et al.,* 2009 and references therein; Valls *et al.,* 2011; Anastasopoulou *et al.,* 2013; Kousteni *et al.,* 2017a; Kousteni *et al.,* 2017b;). Parallel to our findings on main prey groups, the distribution of length measurements of samples collected in this study showed narrow ranges. Information on size at maturity of each species from previous studies also supports most of the examined specimens (85% of *S. canicula* individuals were immature, lowest among other species) were juveniles (see Metochis, Carmona-Antoñanzas, Kousteni, Damalas, & Megalofonou, 2016 for *G. melastomus*; Porcu *et al*., 2014 for *E. spinax*; Kousteni, Kontopoulou, & Megalofonou, 2010 for *S. canicula*; Kousteni &
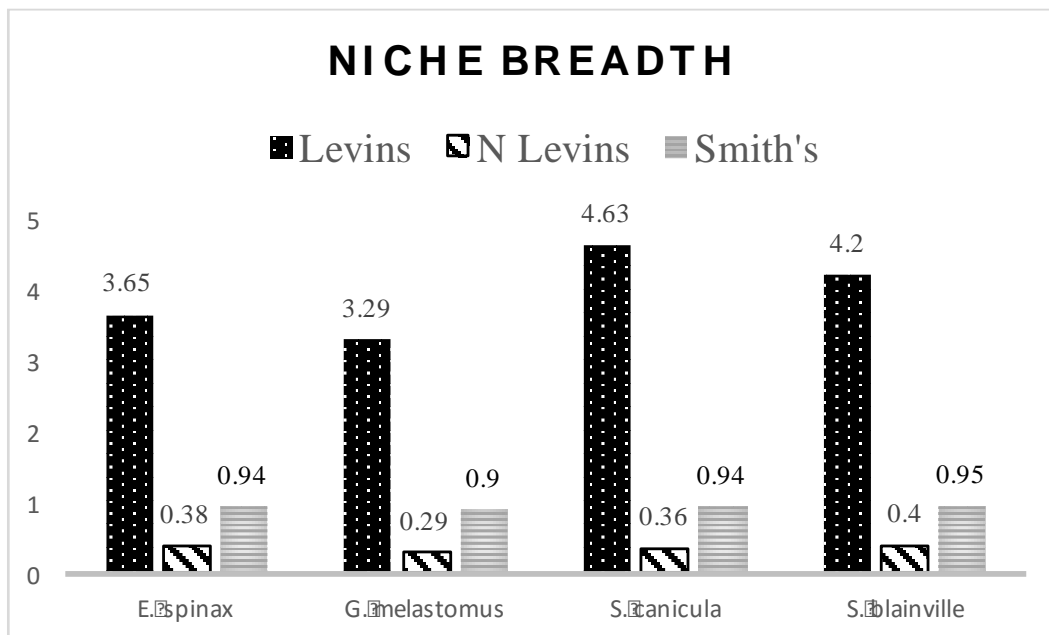
Figure 3. Scores of niche breadth from different approaches for each investigated species.

Table 5. Trophic levels of the species with weighted average of diet compositions and table values of Ebert and Bizzaro (2007) and Cortés (1999).

	Trophic Level		
	This study	Approach of Ebert and Bizzaro (2007)	Approach of Cortés (1999)
Etmopterus spinax	4.2	4.17	4.19
Galeus melastomus	4.18	4.14	3.64
Scyliorhinus canicula	4.12	4.02	4.11
Squalus blainville	4.08	3.98	3.98

Megalofonou, 2015 for *S. blainville*). Thus, our results may represent characteristics of mostly juveniles of these species.

Niche Breath, Niche Overlap, Trophic Level

Even though results on niche breadth indicated differences between the regions and studies (Ellis *et al.,* 1996; Fanelli *et al.,* 2009; Valls *et al.,* 2011), these findings should be interpreted with caution since these values may be biased due to the use of different approaches between studies. In this regard, Krebs (2009) suggested that the usage of the number of individuals gives a better result by comparisons to the usage of proportional prey items whilst calculating p values. Unlike the present study, the mentioned studies on niche breadth and overlap used proportions, and therefore this methodological deviation may explain the inconsistencies amongst studies.

Niche overlap had high values amongst the four shark species examined parallel to the results of stomach content analysis. Most similar stomach contents were observed between *S. blainville* and *S. canicula,* with these two species also showing one of the highest niche overlap. A similar significant overlap was observed between these species in the coasts of Portugal, in the Atlantic Ocean (Martinho *et al.,* 2012).

The results of present study showed that the trophic levels of shark species examined in this study are between 4.08 and 4.2 and these values indicated that the species under consideration are tertiary consumers based on the definition of Cortés (1999). In addition, the examined species can also be classified under groups of carnivores, which Stergiou and Karpouzi (2001) defined the group as potential top carnivores in the Mediterranean Sea. As the diet compositions of these demersal elasmobranch species showed a dominancy of benthic organisms, these sharks can be considered as representative of high level consumers in the demersal community of the Aegean Sea.

Findings from our trophic level comparisons showed that the trophic levels of the examined species are mostly higher in the eastern, rather than the western Mediterranean Sea (Table 6). This tendency can be an indicator of trophic structure in benthic ecosystems. Danovaro, Dinet, Duineveld, and Tselepides, (1999) reported eastern basin is subject to more limiting trophic conditions, and so may have had a higher efficiency in exploiting the particulate organic fluxes, opposed to the western Mediterranean Sea; with the higher trophic input in the western Mediterranean Sea being partially balanced by the higher trophic efficiency of the deep eastern Mediterranean Sea. Thus, results show clear differences in the trophic characteristics of the two environments.

Cortés (1999) points out that using weighted averages of diet compositions by different data-sets is a more accurate approach for trophic level calculation. Furthermore, using a standardized diet composition of sharks is practical for comparison purposes and better understanding their roles in ecosystems. Nonetheless, using standardized groups as a generalization brings some inherent bias for the estimation of trophic levels per examined species. For instance, the trophic level estimation of *G. melastomus* was found as 4.18 with locally weighted trophic level values of standardized diet composition, whilst it was found as 3.64 by using the method of Cortés (1999) and as 4.08 by using the method of Ebert and Bizzarro (2007). In contrast, some species, such as *E. spinax,* showed insignificant

Table 6. Trophic levels of the examined species from different studies and present study.

Species	Reference	Study area	Trophic level
Etmopterus spinax	Cortés (1999)		3.8
	Froese and Pauly (2016)		3.6
	Stergiou and Karpouzi (2002)	W Mediterranean	3.80-4.33 (Min-Max)
	Albo-Puigserver *et al*. (2013)	W Mediterranean	3.71
	This study	E Aegean	4.20
Galeus melastomus	Froese and Pauly (2016)		3.8
	Cortés (1999)		3.7
	Stergiou and Karpouzi (2002)	W Mediterranean	3.70-4.26 (Min-Max)
	Neves et al. (2007)	S Portagual	4.02
	Albo-Puigserver *et al.* (2013)	W Mediterranean	4.17
	This study	E Aegean	4.18
Scyliorhinus canicula	Froese and Pauly (2016)		3.6
	Cortés 1999		3.6
	Stergiou and Karpouzi (2002)	W Mediterranean	3.8
	Pinnegar *et al.* (2002)	Celtic Sea	4.29
	Kousteni *et al.* (2017)	C and W Aegean Sea	4.22
	This study	E Aegean	4.12
Squalus blainville	Froese and Pauly (2016)		3.6
	Cortés (1999)		4
	This study	E Aegean	4.08

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differences among the trophic level calculation methods (Table 5). Regional trophic level values for each standardized diet group should be used for reducing the uncertainty of the estimations, as faunal characteristics in a specific region can determine the species compositions of each standardized diet group. The relative contribution of each prey in the same group may significantly affect the value of trophic level. Global databases such as "FishBase" and "SeaLifeBase" make available trophic level values for most species. Simultaneously, most studies on stomach contents give information about prey items on species or genus level. When considering this availability, collections from studies in a certain region, such as the Aegean Sea or the eastern Mediterranean Sea, using local studies for calculation of weighted trophic level of each standardized diet composition, will give more accurate and comparable information either within a region or among regions.

Acknowledgement

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