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Biology of the thornback ray (*Raja clavata* Linnaeus, 1758) in the North Aegean Sea

by

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Abstract

The study deals with aspects of the population dynamics in the thornback ray (Raja clavata L., 1758), one of the most abundant cartilaginous fish caught in the North Aegean Sea. Females accounted for 73.08% and males 26.92% of all individuals. Total length of females and males ranged between 50.2 and 89.9 cm (disc width: 33.4-62.0 cm), and between 43.1 cm and 82.7 cm (disc width: 30.7-64.2 cm), respectively. Relationships between total length (TL) and total weight (TW), and between disc width (DW) and total weight (TW) were described by the equations: $TW = 0.0041 TL^{3.10}$ and $TW = 0.0178 DW^{3.03}$, respectively. Age data derived from vertebrae readings were used to estimate growth parameters using the von Bertalanffy function: L = 101.71 cm, K = 0.18 y⁻¹, t0 = -0.07y for males and $L_1 = 106.54$ cm, K = 0.16 y⁻¹, t0 = -0.28 y for females. The maximum age was 8 years for males and females. Total length at first maturity of males and females was 70.9 cm and 81.2 cm, respectively. Based on the gonadosomatic index and gonadal macroscopic observations, it was determined that the spawning period lasted throughout the year. Stomach content analysis showed that crustaceans (53.03% IRI) and teleosts (14.70% IRI) were the most preferred prey.

Key words: *Raja clavata*, age and growth, reproduction, feeding, North Aegean Sea

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1. Introduction

Elasmobranchs are among the top predators in the marine environment, thus they affect populations of both fish and invertebrates at lower trophic levels (Ellis et al. 1996). The thornback ray *Raja clavata* (Linnaeus 1758) is a common skate in the eastern Atlantic, distributed from Norway to South Africa, including the Mediterranean and the Black Sea (Stehmann & Burkel 1984). Although the reported depth ranges vary from 10 to 300 m, the species is most common in coastal waters at a depth of 10–60 m (Stehmann & Burkel 1984; Wheeler 1978). It is usually found on muddy, sandy and gravelly bottoms, rarely on rougher bottoms. Due to its wide distribution and abundance, thornback ray is one of the most studied skate species. However, little information is available on its biology.

R. clavata has low growth, maturity and fecundity rates compared to most teleostei. The thornback ray is oviparous and its reproductive activity occurs throughout the year (Jardas 1973). The reproduction peak occurs in June-July. Average production is estimated at 140 eggs year⁻¹ (Holden 1975), but more recent studies indicate lower fecundity, ranging from 48 to 74 eggs (Ellis & Shackley 1995). Egg cases hatch after about 4-5 months, and pups are about 11-13 cm in total length (Pawson & Ellis 2005). Embryos feed exclusively on yolk (Dulvy & Reynolds 1997). Raja clavata is a predator species in the marine environment. Thus, it plays an important role in marine ecosystems. R. clavata feeds mainly on crustaceans, teleosts and cephalopods (Saglam & Bascinar 2008; Yigin & Ismen 2010; Kadri et al. 2014b), and occasionally on gastropods and polychaetes (Kadri et al. 2014b).

The thornback ray is one of the most abundant elasmobranch species in Turkish coastal waters. Over the past ten years, catches of thornback ray have decreased from 668 t in 2010 to 82.6 t in 2018, which corresponds to a decrease of 87% (Tüik 2018). In Turkish waters of the Black Sea and the North Aegean Sea, sharks and rays are caught as bycatch by otter trawls, purse seines, bottom longlines and gillnets. As reported in a related circular, there has been a recent increase in sensitivity to shark and ray fishing in Turkey. Thornback rays are prohibited to be caught according to Fisheries Regulation Notice No. 2016/35 in all Turkish waters. An important factor in the decline of thornback rays is their longer life span, slower growth rate, lower fecundity and late sexual maturity. These factors make them susceptible to changes in the ecosystem. In this context, this study provides important information on age, growth, reproduction and feeding strategy of thornback rays in the North Aegean Sea.

2. Materials and methods

Samples were collected between January 2016 and March 2017 by commercial fishing vessels operating in the North Aegean Sea. A total of 104 specimens were brought to laboratory cold storage conditions. For each specimen, total length (TL, cm), disc width (DW, cm), clasper length (CL, cm), total weight (TW, g) and ovarium weight (OW, g) were determined to the nearest 0.1 cm and 0.1 g, respectively. The TL–W and DW–W relationships were determined using allometric equations (Sparre & Venema 1992), $W = a TL^b$ and $W = a DW^b$, respectively.

A vertebral segment containing at least five vertebrae, between the 10th and 20th vertebral segments, was extracted. The section was then thawed and three vertebrae were dissected, carefully separated and excess connective tissue, as well as neural and hemal arches were removed and placed in a 5-25% solution of sodium hypochlorite. Vertebrae were left in this solution for 5-30 min depending on their size and then scrubbed to remove any residual tissue (Kusher et al. 1992). The vertebrae were then embedded in epoxy resin to enable cutting. To prepare the epoxy resin, Araldite epoxy GY502 and hardener HY 956 were mixed at a ratio of 5:1 and poured over the vertebrae in silicone molds and allowed to dry for 24 h (Campana 2014). An IsoMet low-speed diamond bladed saw was used to prepare vertebral sections. Growth bands in vertebral sections were counted using an Olympus SZX7 stereomicroscope. The age of each specimen was determined from the number of opague and hyaline bands deposited on vertebral centra.

Growth was modeled using von Bertalanffy's (1938) growth equation:

 $L_{t} = L_{\infty} (1 - e^{-K(t-t0)})$

where L ∞ is the asymptotic TL, L_t is the TL at age t, K is the growth curvature parameter, and t_o is the theoretical age at which fish would have 0 total length. The VBGE was calculated using FISHPARM, a computer program for parameter estimation of nonlinear models with Marquardt's (1963) algorithm for least-square estimation of nonlinear parameters (Prager et al. 1987). Sex ratios were compared to the 1:1 ratio using the chi-square (χ^2) test.

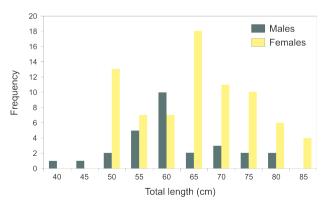
Maturity of males was determined based on the state of mixopterygia (claspers) development in relation to the edge of the pelvic fin. Maturity of females was determined by internal examination (Holden & Raitt 1974; Paesch & Oddone 2008).

To study the food intake, stomachs of the sampled fish were immediately dissected and stored in a 4% formaldehyde solution. In the laboratory, the identification of ingested prey was carried out to the order level. Identification of ingested prey was carried out systematically (Yığın & Ismen 2010). The following references were used in the identification of food items: Murduchay-Boltouski (1969), Fischer (1973), Whitehead et al. (1986a,b), Fischer et al. (1987a,b). Stomach contents were analyzed using the numerical percentage (N%) and frequency of occurrence (F%), percentage by weight (W%; Hyslop 1980) and the Index of Relative Importance (IRI; Pinkas et al. 1971). The index of relative importance (IRI) is used to determine important food items in the diet of fish. The formula used was IRI = F% (N% + W%) (Pinkas et al. 1971; Windell & Bowen 1978).

3. Results

3.1. Length distribution

Sex ratios of specimens were determined as M:F = 1:2.71 and the difference between sexes was statistically significant (χ^2 =18.1; df =1, p < 0.05; SPSS 21.0). Minimum total lengths, disc widths, and weights of the specimens were 43.1 cm, 30.7 cm, and 588.69 g, while the maximum values were 89.9 cm, 64.2 cm, and 5192.82 g, respectively. A total of 104 specimens were measured, of which 76 were females (73.1%) and 28 males (26.9%). TL of females ranged from 50.2 to 89.9 cm (DW 33.4–62.0 cm) and TL of males ranged from 43.1 to 82.7 cm (DW 30.7–64.2 cm; Fig. 1). The independent samples t-test revealed that the relationships between TL–W and DW–W did not change with sex (p > 0.05; SPSS/21).





The TL–W and DW–W relationships were assessed separately for females and males and are presented as $W = 0.003 \text{ TL}^{3.18}$ (95% Cl for b: 3.01–3.35) and W = 0.0187 DW^{3.01} (95% Cl for b: 2.84–3.18), and $W = 0.0223 \text{ TL}^{2.68}$ (95% Cl for b: 2.29–3.07) and $W = 0.0091 \text{ DW}^{3.21}$ (95% Cl for b: 2.85–3.57), respectively. A negative allometric growth (b < 0.05) was determined for males and a positive one (b > 0.05) for female individuals.

3.2. Age and growth

The age of 78.8% of the sampled 104 thornback rays was determined for the purpose of the study. A total of 82 individuals, ranging in age from 4 to 8 years, were examined; 76% were females and 24% were males. Each species had similar band morphology, with opaque bands appearing as relatively thick, regularly spaced, dark-colored bands along each sagittal section axis, separated by narrower lighter-colored translucent bands (Fig. 2).





As shown in Table 1, the age composition varied from 4 to 8 years for both sexes. According to the percentage of occurrence, age group 5 was the dominant one for both sexes. The rays with the most common length of 45–55 cm were mostly 4 years old.

The von Bertalanffy growth equation was used on the data set of 82 individuals to estimate growth parameters (Table 2) for each sex.

Asymptotic length (L_{ω}) values were 101.71 cm for males and 106.54 cm for females. When the mean length was observed at each age compared with the theoretical growth curve, no significant differences

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Table 1

Age frequency distribution for R. clavata from the North Aegean Sea

Age	Ma	les	Fem	ales	Combined		
Groups	n	%	n	%	n	%	
4	5	25	12	19.3	17	20.7	
5	6	30	20	32.3	26	31.7	
6	2	10	12	19.4	14	17.1	
7	3	15	11	17.7	14	17.1	
8	4	20	7	11.3	11	13.4	

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Von Bertalanffy growth parameters for R. clavata

	Growth parameters							
Sex	L_ (cm)	K (year⁻¹)	t _o (year)					
	(95% Cl)	(95% Cl)	(95% CI)					
Males	101.71	0.18	-0.07					
	(53.67–149.76)	(-0.060 to 0.410)	(-2.910 to 2.779)					
Females	106.54	0.16	-0.28					
	(73.18–139.89)	(0.029–0.286)	(-1.981 to 1.427)					
CI – confiden	CI – confidence interval							

were observed between these values. The differences observed between the lengths of the sexes for each age group can be explained by the fact that females reach longer lengths than males of the same age (Fig. 3).

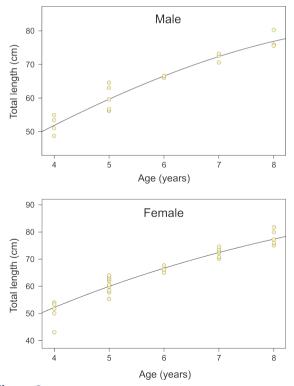


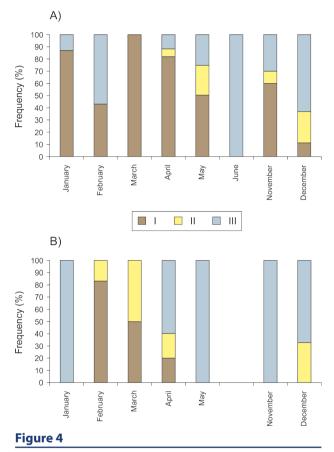
Figure 3

von Bertalanffy length growth curves for R. clavata

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3.3. Reproduction of males and females

Figure 4 shows the analysis of seasonal frequency distributions for both females and males by maturity stage. It was determined that 55.4% of the female individuals were in stage I, 10.8% in stage II and 33.8% in stage III. On the other hand, it was determined that 29.2% of the male individuals were in stage I, 20.8% in stage II and 50.0% in stage III.



Seasonal frequency distribution of R. clavata for females (A) and males (B) at maturity stages

The length and width of claspers and testicles increased gradually in animals with a TL of < 60 cm, followed by rapid growth until a TL of 80 cm, which according to several indicators is the TL for the beginning of sexual maturity (Fig. 5). The minimum and maximum total lengths of males in this research range between 43.1 cm and 82.7 cm. The examined claspers measured from 5.1 cm to 20 cm. The length of claspers in adult individuals (TL 70-82.7 cm) ranged from 16.5 cm to 19.3 cm. The length and width of testes increased with total length of fish. No statistically significant difference was found between the length and width of testes on the right and

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left sides of *R. clavata* (p > 0.05). The minimum and maximum length of testes were determined as 2.15 cm and 9.64 cm and the minimum and maximum width – as 1.20 cm and 4.35 cm, respectively.

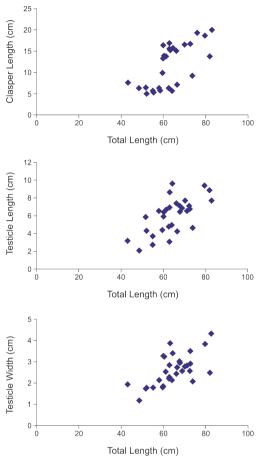
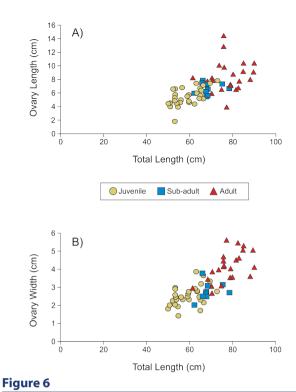


Figure 5

Relationship between total length and clasper length, testicle length, testicle width for males of *R. clavata*

The ovarian development of R. clavata individuals was also monitored throughout the study. In general, the length and width of ovaries in R. clavata females increased with total body length as they reached maturity. The length of ovaries ranged from 1.77 cm to 14.4 cm and their width ranged from 1.42 cm to 5.61 cm (Fig. 6a,b). The width of ovaries in relation to total length increased rapidly from a TL of about 70 cm, above which all specimens are mature, varying from a minimum of 2.67 cm to a maximum of 5.61 cm in width. Following this rapid growth, the ovary continues to grow in width, but at a slower rate (Fig. 6b). No statistically significant difference was found between the length and width of ovaries symmetrically located on both sides of the body cavity (p > 0.05).

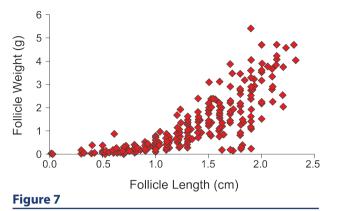


Relationship between total length and ovary length (A), ovary width (B) for females of *R. clavata*

The number of egg capsules ranged from one to two in each uterus, and of the 25 litters examined, 11 had the same number of egg capsules in each uterus. The length and width of 25 egg capsules ranged from a minimum of 6.3×3.9 cm to a maximum of 11.2×4.2 cm, respectively. Females carrying egg capsules were observed throughout the year, but the majority of them were captured in November, December, January, February, April, May and June. A total of 565 follicles were counted in *R. clavata* females. The minimum and maximum diameters of these follicles were calculated as 0.01 and 2.32 cm and the minimum and maximum weights as 0.01 and 5.40 g, respectively. A correlation of $r^2 = 0.81$ was observed between the follicle diameter and weight.

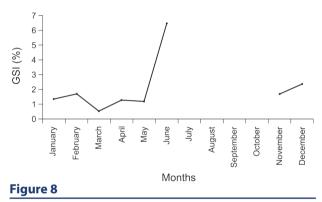
No statistically significant correlation was observed between the total length and follicle diameter in thornback rays (p > 0.05; Fig. 7).

Gonadosomatic index (GSI) values were calculated from monthly samples to determine the reproductive period. A statistically significant difference was found between gonadosomatic indices for males and females (independent samples t-test analysis; p < 0.05, t-value: 3.14, df: 100; SPSS 21). Although no sample was obtained in July, August, September and October, it was found that the breeding continued almost throughout the year and reached its highest



Relationship between follicle length (cm) and follicle weight (g) for females of *R. clavata*

level in spring and winter months. GSI values increased in males in January (1.36%) and November (1.23%), while the highest values for females were recorded in June (6.45%), December (2.82%), January (1.39%) and February (3.26%; Fig. 8).



Gonadosomatic index of thornback ray

The analysis of gonadosomatic values for *R. clavata* males and females showed that GSI values of male individuals were high in November, January, and May, while the maximum values for females were recorded in January, February, June, and December. The analysis of maturity frequencies showed that the most mature individuals of males were observed in January, May, and November, while of females – in February, June, and December. Based on the GSI values and maturity stages, it can be concluded that the reproductive cycle occurs throughout the year.

3.4. Diet composition

Of the 93 stomachs analyzed, 83 (89.2%) contained prey items. There were no significant differences between the sexes in terms of the relative significance index (IRI%; p > 0.05). The analysis of stomach contents in *R. clavata* individuals showed that crustaceans are their primary and most important food group (53.03% Index of Relative Importance IRI), followed by teleosts (14.7% IRI) and cephalopods (1.89% IRI). Their most preferred crustaceans are shrimps (31.42% IRI) and crabs (18.51% IRI), while Teleostei are represented mostly by horse mackerels *Trachurus trachurus* (2.35% IRI) and red mullets *Mullus barbatus* (1.46% IRI). As for Cephalopoda, squids are the most preyed upon (1.88% IRI; Table 3).

The percentage frequency of occurrence of the main prey categories within the length groups is presented in Table 4. Table 4 shows that crustaceans were consumed in each size group. As the length of the thornback ray increased, the composition of the diet changed – the importance of Crustacea decreased, while Teleostei became more dominant.

4. Discussion

The sex ratios and length-frequency distribution of 104 R. clavata individuals, obtained from commercial fishermen in the North Aegean Sea, were investigated in this study. The total length of the analyzed thornback rays ranged from 43.1 to 89.9 cm. The analysis of the length-frequency distribution revealed that females with a length of 65 cm dominated in 23.7%, while males with a length of 60 cm dominated in 35.7%. The least frequent lengths of females and males were 85 cm (5.3%), and 40 and 45 cm (3.6%), respectively. Similarly, Sağlam and Başçınar (2008) reported from the southeastern Black Sea, based on the length-frequency distribution, that female individuals with a length of 75 cm were dominant (30%), while the least frequently observed length was 15 cm (15%). The lengths of the highest and lowest frequencies for males were calculated as 60 cm and 30 cm, accounting for 25% and 15% of all males, respectively. Sağlam and Ak (2012) reported from the southern Black Sea that male and female individuals with a length of 75 cm dominated (80%) and the least frequently observed lengths of these two sexes were 95 cm and 15 cm, respectively. Šantić et al. (2012) reported length-frequency values of 35-45 cm (> 90%) and > 65 cm (50%) in the Adriatic Sea. Kadri et al. (2014a) determined the most frequent length of R. clavata females and males to be 70 cm, accounting for 20% and 25%, respectively. Length-frequency differences between individuals may vary depending on the landing size, fishing gear selectivity, and fishing location.

Studies on the length–weight and disc width– weight relationships for *R. clavata* are listed in Table 5.

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Parameter "b" is an exponent of the arithmetic form of the relationship between length and weight, which for fish varies between 2.5 and 3.5, respectively (Carlander 1969). Carlander (1977) reported that parameter "b" for fish sampled in a small number was b < 2.5 or b > 3.5. Table 5 shows that "b" values for *R. clavata* vary with high or low length and disc width values. Differences in the length–weight relationship parameters can be accounted for by annual changes in seasons, locations, sampling time, population size, and environmental conditions.

Table 6 compares other studies on the growth parameters of R. clavata. The analysis of the previous studies suggests that differences between growth parameters can be accounted for by several factors, such as photoperiod, temperature, food availability and mineral intake (Torres et al. 2005; Kadri et al. 2014a). According to the results of these studies, males grow faster than females and reach the L value at lower lengths and therefore live longer. This holds true for all studies on R. clavata populations (Lesser 1967; Cannizzaro et al. 1995; Serra-Pereira et al. 2005; Whittamore & McCarthy 2005). In addition, although age assignment methods minimize errors, differences in growth parameters may occur. Sex-related growth differences in cartilaginous fish and generally larger size of females are reported in many studies (Casey et al. 1985; Tanaka et al. 1990; Skomal & Natanson 2003). As with other cartilaginous fish, protective fishing management targeted at these species should be implemented as they show slower growth and are overfished.

It was observed that the length of claspers (CL, cm) increased in the direct proportion to the total length of fish (TL, cm). Similar results were reported in other studies on this species (Gallagher et al. 2005; Capapé et al. 2007; Serra-Pereira et al. 2011; Whittamore & McCarthy 2005; Sağlam & Ak 2012). A three-phase sigmoidal curve of clasper length and total length is generally observed for some species of the family Rajidae and the middle phase of the curve refers to maturing individuals (Capapé & Quignard 1974; Templeman 1987; Oddone & Velasco 2004; Sağlam & Ak 2012). Compared with other studies, the total

Table 4

Frequency of prey groups for each size class of thornback ray									
N	Size class (cm)	Annelida	Cephalopoda	Crustacea	Teleostei	Other			
2	40–50	0	0	0.15	0	0.23			
29	50–60	0.001	1.71	4.62	9.23	2.8			
35	60–70	0.004	2.03	11.91	18.58	4.99			
27	70–80	0.002	0.97	6.48	20.56	3.69			
11	80–90	0.004	0.28	3.76	7.18	0.84			

N – Number of fish

Table 3

Diet composition of *R. clavata* presented as percentage numerical composition (N%), percentage weight of prey items (W%), percentage frequency of occurrence (F%) and percentage index of relative importance (IRI%)

Prey	N%	W%	F%	IRI%
Nematoda				
Unidentified nematode	3.37	0.01	5.00	0.80
Cephalopoda				
<i>Loligo</i> sp.	2.25	5.64	5.00	1.88
Octopus sp.	0.16	0.08	0.45	0.01
Crustacea				
Decapoda				
Aegaeon cataphractus	0.16	0.11	0.45	0.01
Natantia				
Caridea	24.88	12.41	17.73	31.42
Parapenaeus longirostris	4.82	2.63	2.73	0.97
Solenocera membranacea	0.80	0.29	0.45	0.02
Stomatopoda				
Squilla mantis	4.17	7.14	3.64	1.96
Erugosquilla massavensis	0.16	0.02	0.45	0.00
Brachyura				
unidentified Brachyura	29.53	7.71	10.45	18.51
Medorippe lanata	0.96	0.79	1.36	0.11
Gonoplax rhomboides	0.32	0.19	0.45	0.01
Anapagurus petiti	0.48	0.07	0.45	0.01
Athelecyclus roduntatus	0.16	0.19	0.45	0.01
Teleostei				
Boops boops	0.48	3.47	0.45	0.09
Merluccius merluccius	0.48	1.27	0.91	0.08
Gobius sp.	0.32	0.70	0.45	0.02
Trachurus trachurus	1.77	9.12	4.55	2.35
Micromesistius poutassou	0.64	4.29	0.91	0.21
Mullus barbatus	0.96	10.27	2.73	1.46
Mullus surmuletus	0.16	1.37	0.45	0.03
Sardina pilchardus	0.32	2.23	0.91	0.11
Spicara smaris	0.32	3.18	0.45	0.08
Trachurus mediterraneus	0.16	0.30	0.45	0.01
Engraulis encrasicolus	1.12	1.28	0.45	0.05
Gadiculus argenteus	0.16	0.22	0.45	0.01
Triglidae	0.48	1.87	0.45	0.05
Serranus hepatus	0.16	0.62	0.45	0.02
Unidentified fish	8.19	9.20	12.27	10.14
Other	12.04	13.32	24.55	29.59

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Study	Study Region	Sex	N	Length(cm)		Weight (g)		Relationship Parameters		
Study		Sex		Min.	Max	Min.	Max	а	b	r²
Filiz & Mater 2002	North Aegean Sea	M + F	31	20.5	99.0	28.9	2614.3	0.0016	3.29	0.93
Borges et al. 2003	Southern Portugal	M + F	13	13.7	54.0	7.8	758.2	0.0014	3.36	0.99
Mendes et al. 2004	Coast of Portugal	M + F	63	30.6	86.2	110	4590	0.0025	3.23	0.96
Demirhan et al. 2005	Southeast Black Sea	Μ	23	48.0	95.0	620	5000	0.0005	3.02	0.96
Demiman et al. 2005	Southeast Black Sea	F	29	34.3	88.2	170	5450	0.0003	3.70	0.94
Rosa et al. 2006	Azores Archipelago	M + F	404	37.0	89.0	270.0	5110.0	0.0058	3.02	0.93
İşmen et al. 2007	Gulf of Saros	M + F	112	6.0	60.0	5	4610	0.01300	3.12	0.99
İlkyaz et al. 2008	Aegean Sea	M + F	24	29.0	51.3	_	-	0.0335	2.89	0.96
Yeldan et al. 2008	Northeast Mediterranean	M + F	90	29.3	64.6	110	1120	0.0034	3.10	0.99
		Μ	98	11.0	76.0	6.30	2900	0.00146	3.34	0.99
Yığın & İşmen 2009	North Aegean Sea	F	128	10.0	88.0	5.00 4622	0.00181	3.31	0.99	
		M + F	226	10.0	88.0	5.00	4622	0.0016 3.29 0.0014 3.36 0.0025 3.23 0.0005 3.02 0.0003 3.70 0.0058 3.02 0.01300 3.12 0.00335 2.89 0.0034 3.10 0.00146 3.34	0.99	
Bök et al. 2011	Marmara Sea	M + F	24	12.2	70	0.015	2628	0.00001	2.87	0.89
		M + F	66	6.1	42.6	2	1387	0.0100	3.16	0.99
Yemişken 2017	Gulf of Iskenderun	Μ	27	6.8	33.8	4	648	0.110	a b 0.0016 3.29 0.0014 3.36 0.0025 3.23 0.0005 3.02 0.0003 3.70 0.0003 3.70 0.0003 3.70 0.0033 3.70 0.0034 3.10 0.00146 3.34 0.00143 3.31 0.00143 3.32 0.00143 3.31 0.00163 3.32 0.00163 3.12 0.00163 3.12 0.00164 3.14 0.00163 3.12 0.00163 3.12 0.00163 3.12 0.00163 3.16 0.0100 3.16 0.0110 3.12 0.00127 3.13 0.0023 2.68	0.99
		F	39	6.1	42.6	2	1387	0.0096	3.17	0.99
	Around Gökçeada	M + F	19	6.8	46.3	6	2650	0.0127	3.13	0.80
		M + F	104	43.1	89.9	588.7	5192.8	0.0041	3.10	0.93
In this study	North Aegean Sea	М	28	43.1	82.7	588.7	3492	0.0223	2.68	0.88
		F	76	50.2	89.9	654.1	5192.8	0.003	3.18	0.95

Length-weight relationship parameters of thornback ray by region

M – Male; F – Female; N – Number of samples; Min. – Minimum; Max – Maximum; a and b – Length–weight relationship parameters, r² – Coefficient of correlation.

length-clasper length relationship in the present study is a sigmoid curve (Oddone & Vooren 2005; Kadri et al. 2014a). Similar to other species of the family Rajidae, oviparous reproduction was observed in R. clavata individuals in this study. In this reproduction type, each egg capsule contains a single embryo, which is attached to the oviduct. A total of 25 egg capsules in this study ranged in their length and width from a minimum of 6.3×3.9 cm to a maximum of 11.2×4.2 cm, respectively. Compared to other studies, Janez & Sueiro (2009) reported more than one embryo in each case of some Rajidae species, for example, Sympterygia bonapartii contained two embryos per capsule. In their research carried out in the Black Sea, Sağlam & Ak (2012) found an egg capsule 15.0 cm long, 5.0 cm wide and weighing 12 g. In their study on R. clavata conducted in Portugal, Maia et al. (2015) reported a capsule length and width of 6.5 cm and 4.8 cm, respectively.

For 565 follicles obtained from thornback rays, the relationships between total lengths and follicle diameters were not statistically significant. Similar results were reported for *Raja rhina* by Ebert et al.

(2008) and for *Raja clavata* by Sağlam & Ak (2012). Ebert (2005) determined a relationship between the number of mature follicles and the total length of *Bathyraja* individuals collected from the Bering Sea. The number of oocytes in *B. aleutica* increased up to a L_r of 145 cm and then decreased in individuals with greater length. As regards *Psammobatis extenta* in the Southwest Atlantic and *Dipturus laevis* in the western North Atlantic, Braccini & Chiaramonte (2002) reported that follicle diameters increase with increasing total lengths.

Holden (1975) stated that thornback rays are serially spawning species as are other oviparous species. According to Hamlett and Koob (1999), the majority of adult females in most oviparous shark and skate species tend to spawn throughout the year. Based on the previous studies (Du Buit 1976; Braccini & Chiaramonte 2002; Sağlam & Ak 2012) and considering GSI values for males and females and maturity stages, it was concluded that males and females of some species from the family Rajidae and of *R. clavata* in the present study do not exhibit seasonal variations and can spawn throughout the year. This is supported by

the fact that a total of 25 egg capsules were obtained in November, December, January, February, April, May and June. Sağlam and Ak (2012) obtained egg capsules from *R. clavata* individuals in July and October. Holden et al. (1971) claim that egg deposition in *R. clavata*, *R. brachyura* and *R. montagui* is closely associated with water temperature and the deposition increases in warmer temperatures. Holden (1975) observed that *R. clavata* individuals in the British waters have a more extended spawning period, lasting from February to September and peaking in June. A similar egg-laying period (between May and September) was observed for *R. clavata* by Ryland and Ajayi (1984). Capapé et al. (2007) conducted research in the Northern Mediterranean Sea, which showed that vitellogenic activity of *R. clavata* takes place throughout the year but slows down in April and August. In their research conducted in the Gulf of Gabès, Kadri et al. (2014a) showed, based on the analysis of different parameters, that *R. clavata* do not spawn continuously. Similar results were obtained in Portugal by Serra-Pereira et al. (2011). In the Black Sea, Demirhan et al. (2005) found egg capsules in females mostly in spring and autumn. Pawson and Vince (1999) also found egg capsules of *R. clavata* throughout the year, but more frequently in summer. Moreover, *R. clavata* was found to spawn off the Languedoc coast throughout the year (Capapé et al. 2007).

Table 6

Comparison of von Bertalan				1	+ ()(00r)	Maximum aga (upar)	C+u.dv
Region	N	Sex	L (cm)	K (year-1)	t _o (year)	Maximum age (year)	Study
UK waters	-	M	88.3	0.22	-1.30	-	Taylor & Holden 1964
		F	127.3	0.10	-2.50 -0.60	-	
Irish Sea and Bristol Channel	-	M F	85.6 107.0	0.21	-0.60	-	Holden 1972
Carmarthen Bay, United Kingdom	_	г M + F	139.2	0.13	-2.63	- 12	Ryland & Ajayi 1984
Irish Sea, Northeast coast	_	M + F	105.0	0.22	0.45	-	Brander & Palmer 1985
	_	M	96.8	0.19	-1.36	_	brander & raimer 1985
Irish Sea, southeast coast	_	F	107.8	0.15	-1.01	_	Fahy 1991
	200	M	116.7	0.106	-0.41	_	
Italy, Sicily Channel	200	F	126.5	0.098	-0.51	_	Cannizzaro et al. 1995
	_	M	98.0	0.17	-0.43	_	
North Sea	_	F	118.0	0.14	-0.88	-	Walker 1999
	258	M	106.5	0.14	-1.74	8	
Irish Sea		F	139.5	0.093	-1.84	8	Gallagher et al. 2005
Caernarfon Bay, North of Wales	54	М	100.9	0.18	-0.95	7	
	135	F	117.6	0.16	-0.70	9	Whittamore & McCarthy 200
	85	М	121.5	0.11	-0.11	-	
Portugal	115	F	130.5	0.10	-0.13	-	Serra-Pereira et al. 2005
	200	M + F	130.5	0.10	-0.14	10	
	53	М	61.9	0.22	-1.79	-	
North-East Mediterranean	37	F	95.7	0.92	-2.60	-	Yeldan et al. 2008
	90	M + F	79.7	0.13	-2.70	8	
	98	М	79.99	0.23	-1.09	7	
North Aegean Sea, Saros Bay	128	F	95.00	0.15	-1.10	8	Yığın 2010
	226	M + F	99.99	0.15	-1.08	8	
Gulf of Gabes, Tunisia	530	М	100.8	0.14	1.13	12	
	750	F	114.6	0.11	1.23	15	Kadri et al. 2014a
Wast Algorian coast	166	М	100.0	0.35	-0.68	-	Adda Uanifi at al 2017
West Algerian coast	397	F	105.0	0.46	-0.20	-	Adda-Hanifi et al. 2017
North Aggaan Saa	28	Μ	101.71	0.18	-0.07	8	this study
North Aegean Sea	76	F	106.54	0.16	-0.28	8	this study

M – Male; F – Female; N – Number of samples; L – Asymptotic total length (cm); K – Growth curvature parameter (year¹); t_o – theoretical age at which fish at "0" total length (year).

In this study, the most important prey categories in the diet of R. clavata were Crustacea (53.03% IRI), Teleostei (14.7% IRI) and Cephalopoda (1.89% IRI). On the other hand, Morato et al. (2003) reported that the diet of R. clavata in the North Atlantic consists mainly of Teleostei (IRI = 81.6%) and Crustacea (IRI = 17.4%). In the study carried out in the southeastern Black Sea by Demirhan et al. (2005), 35 (70%) of the examined stomachs were empty and 15 (30%) stomachs contained prey. The authors also emphasized that the primary diet of thornback rays consists of crustaceans (O = 52%) and demersal fish (O = 35%). Farias et al. (2006) found that thornback rays in Portugal feed mainly on shrimps. Similar to the results of the present study, the study conducted by Yeldan et al. (2008) in the Northeastern Mediterranean Sea reported a stomach fullness of 74.4% in R. clavata and listed its most important food groups as Crustacea, Teleostei, and Cephalopoda. Sağlam and Başçınar (2008) researched the feeding ecology of R. clavata in the southeastern Black Sea and reported that its main food groups are crustaceans (74.8% IRI), fish (18.6% IRI), nematodes (5.9% IRI), cephalopods (0.08% IRI), annelids (0.03% IRI) and mollusks (0.01% IRI). In their study on R. clavata conducted in the Aegean Sea, Karachle and Stergiou (2010) reported that the most important food group of this species are Natantia and Brachyura. A study performed in the Western Mediterranean Sea lists prey items of R. clavata as Teleostei (31% IRI) and Crustacea/Natantia (27% IRI; Valls et al. 2011). The most important prey items of this species in the Adriatic Sea are reported to be crustaceans/decapods (72.8% IRI) and teleosts (20.4% IRI; Šantić et al. 2012). In a study performed on R. clavata individuals in the Gulf of Gabès of the Mediterranean Sea, a stomach fullness of 83.98% was determined. In that study, Teleostei (56.6% IRI), Crustacea (41.1% IRI) and Cephalopoda (2.35% IRI) were found to be the most important food groups according to the index of relative importance (Kadri et al. 2014b). As noted by most researchers, thornback rays prey mainly on crustaceans and demersal/pelagic fish. The present research corroborates the results of the aforementioned studies. In the present study, it was found that thornback rays with a length of 60-70 cm fed mainly on Crustacea (11.91%). It was observed that as the length of thornback rays increased, they ate less crustaceans. Secondarily, Teleostei accounted for 18.58% and 20.56% of the diet composition of individuals measuring 60-70 cm and 70-80 cm, respectively. Some studies report that skates exhibit different feeding habits depending on their lengths and report that they feed on fewer crustaceans as their length increases (Smale & Cowley 1992; Ellis et al. 1996;

Ebeling 1988; Morato et al. 2003). Some authors state that larger predators prefer larger prey items (Smale & Cowley 1992), while others believe that the prey of skates occur from the benthic to benthopelagic zone (Ebeling 1988) or from the benthopelagic to benthic zone (Quiniou & Andriamirado 1979).

5. Conclusions

This study was carried out to determine the lengthweight relationship, age and growth parameters, maturity scales, reproductive periods and stomach contents of 104 thornback rays, R. clavata (Linnaeus, 1758), obtained from commercial fishermen in the North Aegean Sea. During the study, it was found that the length of thornback rays obtained from commercial fishermen ranged predominantly between 60 cm and 70 cm. Considering that thornback rays are long-lived species with low fecundity and slow growth rates, it can be concluded that the individuals in the current study make them susceptible to exploitation. Recently listed by the International Union for Conservation of Nature as "Near Threatened" in the near future, this species of economic value should be preserved and protected in a sustainable way. Due to the limited research in this field in Turkey, more studies should be conducted to investigate the biology, ecology, reproduction/spawning, growth and stock assessment of thornback rays. Overexploitation of this species is especially relevant as it is characterized by slow growth, long life span, and long period before individuals become reproductively mature. Such studies should be performed to locate their habitats and local fishing activities should be regulated based on their results.

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