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RESEARCH ARTICLE

Age, growth and reproduction of the rough ray, *Raja radula* (Delaroche, 1809) in Saros Bay (North Aegean Sea)

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Abstract

In this study age, growth and reproduction of rough ray (*Raja radula*) were investigated using 261 specimens from Saros Bay (North Aegean Sea) sampled between February 2005 and September 2008. Females made up 56.3% and males 43.7% of the total individuals studied. The total length of females and males ranged between 17 and 61 cm (Disc width; 11.1-39.8 cm), and between 19 and 54 cm (Disc width; 12.5-34.2 cm), respectively. The total length (TL)-weight (W) and disc width (DW)-weight relationships were described by the equations $W=0.0018*TL^{3.35}$ and $W=0.0081*DW^{3.32}$, respectively. Age data derived from vertebrae readings were used to estimate the growth parameters of von Bertalanffy equation: $L_\infty=74.70$ cm, $K=0.20y^{-1}$, $t_0=-0.22y$ for males, and $L_\infty=82.94$ cm, $K=0.16y^{-1}$, $t_0=-0.59y$ for females. The maximum age was 6 for males and 8 for females. Total length at first maturity of males and females was 40 and 46 cm, respectively.

Keywords: *Raja radula*, age, growth, reproduction, Saros Bay

Introduction

The rough ray *Raja radula* Delaroche, 1809 is a common skate occurring throughout the Mediterranean Sea, mainly in the western part, and absent from the Black Sea (Serena 2005). In Turkish waters, *R. radula* is relatively common along the Aegean and Mediterranean coasts (Mater *et al.* 2005), where it inhabits from coastal water to 350 m (Serena 2005). The rough ray is not a target species of the commercial fisheries as it caught as by-catch of demersal trawl, gillnet, trammel net and bottom longline fisheries (Serena 2005; Consalvo *et al.* 2010), and data on its biology are patchy. Further investigation are required to determine population size, biological parameters, interaction with fisheries and population trends of this species (Morey *et al.* 2006).

Published data are mainly limited to the North African population, where this skate is commonly caught and landed at fishing sites in Tunisian waters (Mejri *et al.* 2004) but it is also reported in the Aegean Sea and Ionian Sea (Kyrtatos 1982; Consalvo *et al.* 2010). Capapé (1974) studied maturity and reproductive cycle of *R. radula*, while Capapé and Quignard (1975) assessed the fecundity of this species. Related species of batoids have recently been studied in the Mediterranean Sea that refer to aspects of their age and growth (Ismen 2003; Ismen *et al*. 2007; Başusta *et al*. 2008; Yeldan *et al*. 2009; Demirhan *et al*. 2010; Başusta and Sulikowski 2012; Duman and Başusta 2013; Keskin and Karakulak, 2006).

The following is an examination of *R. radula* life history and ecology, including its age and growth development and reproduction. It provides important information on growth parameters, age and size at sexual maturity. This new information greatly improves the understanding of rough ray biology, and will be applicable to the stock assessment and management of this species.

Materials and Methods

Sampling

A total of 255 specimens of rough ray was collected monthly by using commercial bottom trawls between February 2005 and September 2008 in Saros Bay (40°30′00′′N-26°30′00′′E), in the North Aegean Sea (Figure 1). Hauls were carried out both at daytime and nighttime at depths ranging from 5 to 486 m. Individuals were mostly captured at depths between 5-50 m and 100-200 m. The trawl was equipped with a 44 mm stretched mesh size net at the cod-end. Trawling lasted 30 minutes. Trawling speed was 2.5 knots.

In the laboratory, the following measuremets were made to the nearest 1 cm and recorded for each specimen: Total length (TL, measured from the tip of the rostrum to the tip of the tail), disc width (DW, measured between the tips of the widest portion of the pectoral fins), clasper length (CL, measured from the points of insertion at the cloaca to the tip of the claspers) as well as total mass (W in g). Statistical comparison of length-weight and disc width-weight relationships between sexes was performed by applying the t-test (Zar 1999).

Age and Growth

Age was determined according to silver nitrate impregnation methods proposed by Caillet *et al.* (1983). To estimate age, a section of 15 cm of the vertebral column consisting of approximately five vertebrae from the area above the pelvic girdle was dissected from each specimen (Licandeo *et al.* 2007).

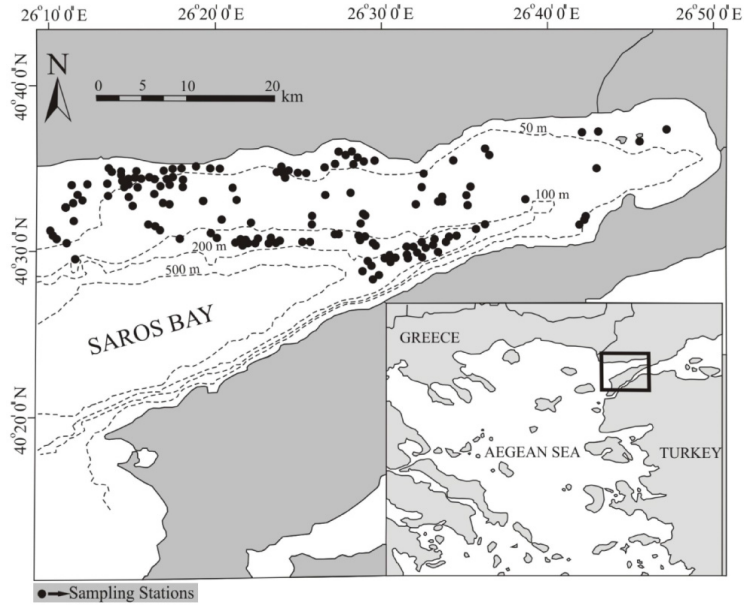


Figure 1. The sampling stations in Saros Bay

The section was then thawed and three vertebrae were dissected out, carefully separated, and excess connective tissue, including the neural and haemal arches, were removed by placing them in a 5-25% solution of sodium hypochlorite. Vertebrae were left in this solution for 5-30 minutes depending on their size, scrubbed to remove any remaining tissue. The centra surfaces were burned in 88% formic acid for 2-4 minutes and then washed in distilled water for 5 minute (Kusher *et al.* 1992). The centra were stained in 1% silver nitrate water solution for 10-15 minutes and then exposed to ultraviolet light for 15 minutes. Sections were then rinsed in distilled water and subsequently soaked in 5% isopropyl alcohol water solution for 1 minute to remove excess material (Schwartz 1983; Serena *et al.* 2005). Finally, all stained vertebrae were examined with an Olympus SZX16 microscope, illuminated from above.

The TL-W and DW-W relationships were determined using allometric equations (Sparre and Venema 1992), $W = aTL^b$ and $W = aDW^b$, respectively. Growth was modelled using the von Bertalanffy (1938) growth equation: $L_t = L_\infty (1\text{-}e^{-K(t\text{-}to)})$ where L_∞ is the asymptotic TL, L_t the TL at age t, K the growth curvature parameter and t_0 the theoretical age when fish would have been at 0 total length. The VBGE was calculated by 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).

Reproduction

The maturity of males was defined from the state of development of the mixopterygia (claspers) in relation to the edge of the pelvic fin. Maturity of females was determined by internal examination (Holden and Raitt 1974; Paesch and Oddone 2008). A logistic growth model was fitted to the relationship between the fraction of mature males or females as a function of TL: $P_{TL}=1/(1+e^{(a+bTL)})$, where P_{TL} is the fraction mature in length class TL, and a and b are model parameters. With this model, mean size at first maturity, TL_{50}, was estimated by –a/b, which represents the body size at which 50% of the fish are mature (White *et al.* 2001; Oddone and Vooren 2005).

Results

Morphological measurements

A total of 255 specimens were measured, of which 142 were females (55.7%) and 113 males (44.3%). Females TL ranged from 17 to 61 cm (11.1-39.8 cm DW) and males TL from 19 to 54 cm (12.5-34.2 cm DW) (Figure 2). Overall, the mean TL of females was greater than that of males (t-test=-2.01, df=252, P<0.05). Male and female TL-W and DW-W relationships (Figures 3a and 3b) were significantly different and were described by the equations: females $W=0.0019TL^{3.36}$ (F-test=4703.98, df=140, P<0.05, 95% CI for b: 3.26-3.45) and $W=0.0092DW^{3.28}$ (F-test=4847.00, df=141, P<0.05, 95% CI for b: 3.19-3.39); males $W=0.0022TL^{3.30}$ (F-test=3568.34, df=111, P<0.05, 95% CI for b: 3.19-3.40) and $W=0.0053DW^{3.46}$ (F-test=2855.58, df=111 P<0.05, 95% CI for b: 3.33-3.59).

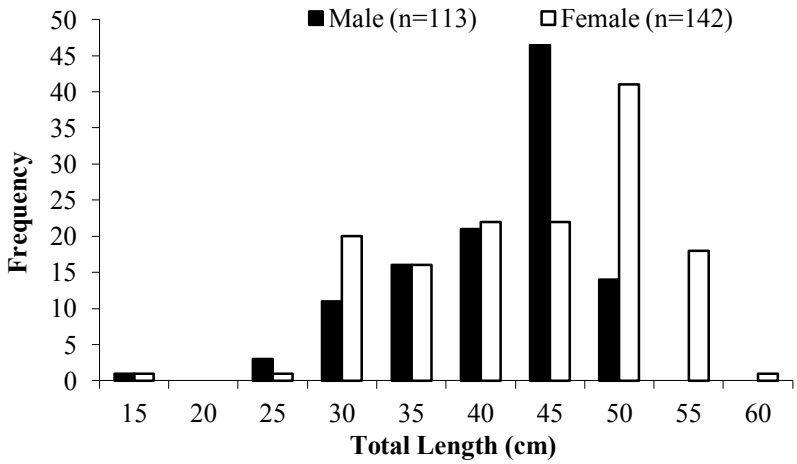


Figure 2. Length-frequency distribution by sex of rough ray

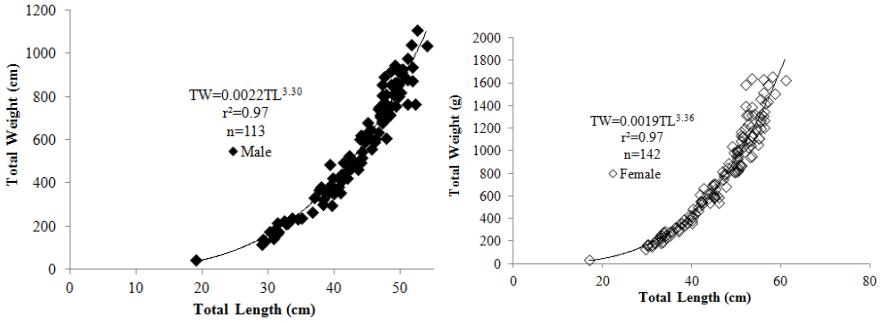


Figure 3a. Relationship between total length and weight for rough ray, *Raja radula*, from the North Aegean Sea

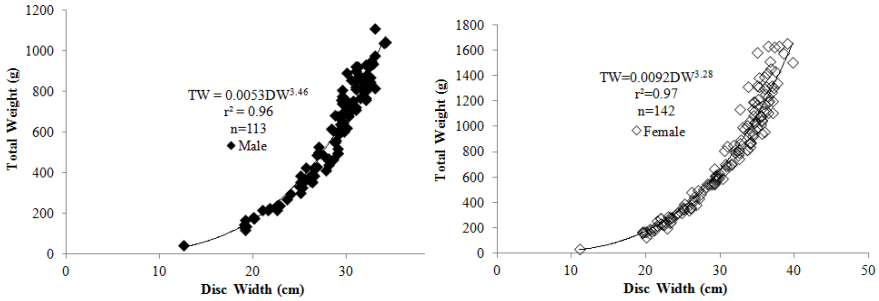


Figure 3b. Relationship between disc width and weight for rough ray. *Raja radula*, from the North Aegean Sea

Table 1. Age frequency distribution of *Raja radula* from Saros Bay, for each sex and combined sexes

Age Group	Males		Females		Combined	
	N	%	N	%	N	%
1	1	1.23	1	0.90	2	1.04
2	5	6.17	4	3.60	9	4.69
3	10	12.34	23	20.72	33	17.19
4	25	30.86	27	24.32	52	27.08
5	39	48.15	29	26.13	68	35.42
6	1	1.23	13	11.71	14	7.29
7	-	-	12	10.81	12	6.25
8	-	-	2	1.80	2	1.04
Total	81	100.00	111	100.00	192	100.00

Age and Growth

The samples consisted of individuals whose age ranging from 1 to 8. In this study, treatment by silver nitrate resulted in better visibility of the rings of 192 individuals. As shown in Table 1, the age composition varied from 1 to 6 for males and 1 to 8 for females. Age group 5 was dominant in males (48%) and but in females (26%) it was not as dominant as in males.

The von Bertalanffy growth equation was calculated on a data set of 192 individuals (Table 2) for each sex and combined data. Then mean length observed at each age was compared with that obtained from the theoretical growth curve (Figure 4). The agreement of these data sets suggests that the von Bertalanffy theoretical growth curve is consistent with the actual growth in the rough ray from Saros Bay. Our study suggests that females attain larger asymptotic length (82.94 cm) than males (74.40 cm). The von Bertalanffy growth model predicted an L_∞ of 107.75 cm for combined sexes that is larger than the L_∞ of 86.40 cm reported by Kadri *et al.* (2014).

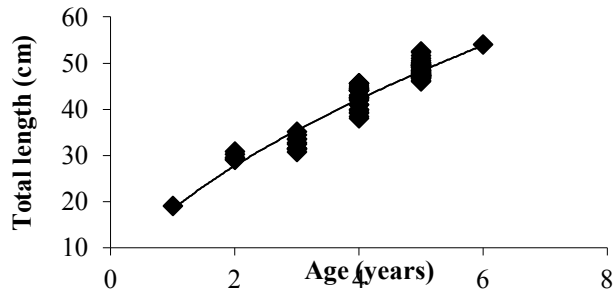
Table 2. Von Bertalanffy growth parameters for *Raja radula* from length data of males and females as well as both sexes combined, with in brackets are 95% confidence intervals for parameter estimates

Sex	L_∞ (cm)	K (year^{-1})	t_0 (year)	r^2
Males	74.70 [66.97-82.44]	0.20 [0.15-0.25]	-0.22 [-0.61-(-0.18)]	0.94
Females	82.94 [65.93-99.95]	0.16 [0.09-0.22]	-0.59 [-1.14-(-0.04)]	0.92
Combined	107.75 [82.02-133.48]	0.09 [0.06-0.13]	-1.40 [-1.88-(-0.92)]	0.92

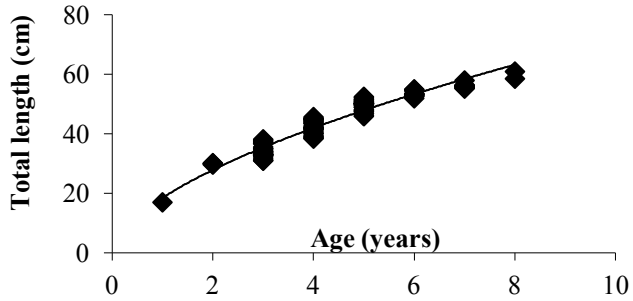
Reproduction

Females ranged from 30 to 58 cm of TL, with percentage of 47.2% immature and 52.8% mature specimens. The smallest mature female captured was 45.3 cm of TL, while the largest immature was 44.2 cm of TL. Examination of the male and female maturity stages indicated that the length at 50% maturity (L_{50}) were 40 cm TL (Standard error values for maturity estimate: 0.179) for males and 46 cm TL (Standard error values for maturity estimate: 0.140) for females (Figure 5). Of the total male samples, 34.9% were immature and 65.1% mature. The smallest mature male captured was 39.3 cm, whereas the largest immature one was 38.4 cm of TL. For the rough ray a very gradual increase in the size of claspers in males relative to body size was evident prior to maturity (Figure 6). All male specimens >39 cm were mature and had fully calcified claspers.

(a)



(b)



(c)

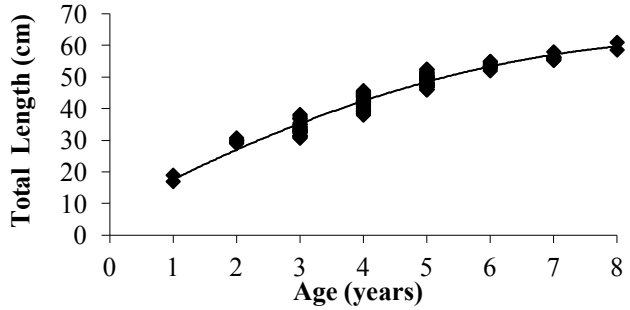


Figure 4. von Bertalanffy length growth curves for rough ray males (a), females (b), and combined sexes (c)

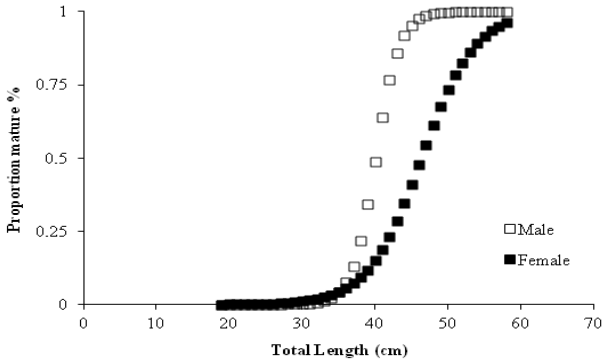


Figure 5. Length at first maturity of male (L_{50}: 40 (0.179)) and female (L_{50}: 46 (0.140)) *Raja radula*

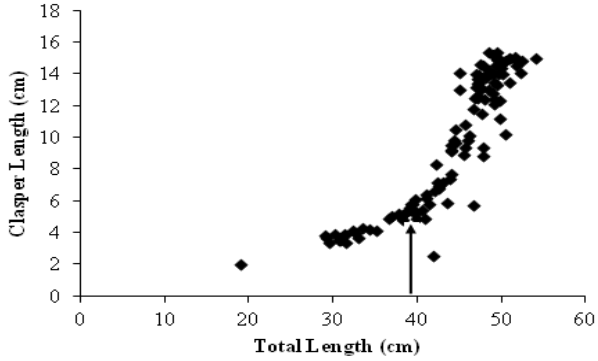


Figure 6. Relationship between total length (cm) and clasper length (cm) for *Raja radula.* Arrow indicates 39cm above which all males were mature.

Discussion

Stehmann (1990) and Karakulak *et al.* (2006) stated that *R. radula* reaches to 70 cm TL. Moutopoulos and Stergiou (2002) and Yeldan and Avşar (2007) noted that *R. radula* in the Aegean Sea reaches 68.2 and 68.1 TL, respectively. Deval *et al.* (2010) expressed that *R. radula* reaches 59.0 cm TL in the study made on length-weight relationship of *R. radula* in the eastern Mediterranean (Table 3). A number of factors are known to influence the length-weight relationship in fish, including growth phase, season, degree of stomach fullness, gonad maturity, sex, size range and general fish condition and preservation techniques (Tesch 1971).

For age determination, the silver nitrate impregnation technique (Schwartz 1983; Kusher *et al.* 1992) was used. In the present study, sufficient clarity over total vertebral surface was obtained with this technique. Yeldan (2005)

reported an age range of *R. radula* from 0 to 9 years and values of k, 0.036 for males and 0.031 for females in the northeastern Mediterranean. Kadri *et al.* (2013) expressed that oldest female was 12 years old and k values 0.14 y^{-1}, whereas the oldest male was 9 years old and k values 0.22 y^{-1} in the Gulf of Gabes, the central Mediterranean. Comparison of rough ray growth rates in the current study and the value in Kadri *et al.* (2013) suggests that *R. radula* is a slow-growing species.

Growth was estimated using the von Bertalanffy growth model, as determined by our study, suggesting that females attain larger asymptotic length (82.94 cm) than males (74.70 cm). The von Bertalanffy growth model predicted an L_∞ of 107.75 cm for combined sexes, which is smaller than the L_∞ of 197.13 cm reported by Yeldan (2005). The results of the present study was similar to Kadri *et al.* (2013) for *R. radula* in the Gulf of Gabes. They reported that females reached a larger maximum size (L_∞=97.94 cm) than did males (L_∞=76.36cm).

The observed differences in growth model parameters between these studies may be due to several factors; sampling methods, locations, the age classes included in the models (Neer and Thompson 2005; Yigin and Ismen 2012).

In this study, maturity stages of the males and females indicated that males matured at a smaller size (TL = 40 cm) than females (TL = 46 cm). Similar results were reported by Yeldan (2005), where it was TL = 37 cm for males and TL = 43 cm for females. It is common among elasmobranchs that females mature at larger sizes than males (Nottage and Perkins 1983; Francis *et al.* 2001; Demirhan *et al.* 2005; Kadri *et al.* 2013), as also demonstrated in our study.

Many elasmobranch populations are declining globally, and the lack of information about basic life-history characteristics are indicative of the need for more data about all populations (Hale and Lowe 2008).

This article is the first in a series aimed at providing life history data; age, growth parameters and reproductive biology for the management of rough rays, indigenous to Saros Bay.

Table 3. Length-weight relationships of *R. radula* in different regions

		Length (cm)		Weight (g)		Relationship parameters					
Sex	**n**	**min**	**max**	**min**	**Max**	**a**	**b**	**95% C.I. of b**	**r^2**	**Region**	**Source**
both	25	20.4	68.2	-	-	0.00515	3.07	0.08	0.98	Aegean Sea (Greece)	Moutopoulos and Stergiou (2002)
both	25	17.4	70.0	-	-	0.0030	3.21	0.17	0.94	Northern Aegean Sea, Gökçeada Island	Karakulak *et al.* (2006)
M	152	21.1	58.1	26.4	1102	0.0011	3.36	3.26-3.46	0.98	Northeastern Mediterranean	Yeldan and Avşar (2007)
F	144	22.6	68.1	25.6	1610	0.0013	3.35	3.26-3.44	0.99		
both	296	21.1	68.1	25.6	1610	0.0012	3.35	3.29-3.44	0.99		
M	22	18.9	53.4	35.5	1060	-	-	-	-	Eastern Mediterranean, Gulf of Antalya	Deval *et al.* (2010)
F	22	20.3	59.0	41.7	1600	-	-	-	-		
both	44	18.9	59.0	35.5	1600	0.00130	3.41	-	0.98		
M	113	19.0	54.0	46.0	1112	0.00220	3.30	3.19-3.40	0.97	North Aegean Sea, Saros Bay	This study
F	142	17.0	61.0	40.0	1661	0.00190	3.36	3.26-3.45	0.97		
both	255	17.0	61.0	40.0	1661	0.00180	3.35	3.28-3.43	0.97		

Acknowledgements

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Kuzey Ege Denizi, Saroz Körfezi'nde*Raja radula* (Delaroche, 1809)'nın Yaşı, Büyümesi ve Üremesi

Özet

Bu çalışmada, Şubat 2005-Eylül 2008 tarihleri arasında Kuzey Ege Denizi, Saros Körfezi'nden örneklenen 261 adet *Raja radula*'nın yaş, büyüme ve üremesi araştırılmıştır. Çalışılan toplam bireylerin %56,3'ü dişi, %43,7'si erkeklerden oluşmaktadır. Dişiler ve erkeklerde toplam boy (TB) sırasıyla, 17 ve 61 cm arasında (Disk genişliği (DG); 11,1-39,8 cm), ve 19 ve 54 cm arasında (Disk genişliği; 12,5-34,2 cm) olduğu tespit edilmiştir. Toplam boy-ağırlık ve disk genişliği-ağırlık ilişki denklemleri sırasıyla, $W=0,0018*TL^{3,35}$ ve $W=0,0081*DW^{3,32}$ olarak tanımlanmıştır. Yaş verileri omur okumalarından, von Bertalanffy büyüme denklemi kullanılarak hesaplanmıştır: Erkekler için, $L_\infty=74,70$ cm, $K=0,20$ y^{-1}, $t_0=-0,22$ y ve dişiler için $L_\infty=82,94$ cm, $K=0,16$ y^{-1}, $t_0=-0,59$ y. Maksimum yaş erkekler için 6, dişiler için 8'dir. Erkekler ve dişiler için ilk olgunluk toplam boyları sırasıyla, 40 ve 46 cm olarak hesaplanmıştır.

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