



UNIVERSITÀ
POLITECNICA
DELLE MARCHE

**Effects of anthropogenic activities on life cycle and abundance of *Panaeus*
(*Melicertus*) *kerathurus* in the Adriatic Sea.**

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A dissertation submitted in partial
fulfillment of the requirements for the
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CURRICULUM: MARINE BIOLOGY AND ECOLOGY



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Thesis on the biology and ecology of caramote prawn in the Northern-Central Adriatic Sea in relation to human activities, with particular attention to reproductive biology, the effects of breakwaters in the biological cycle of the species and the main population parameters.

Luca Bolognini

Abstract

Effects of anthropogenic activities on life cycle and abundance of *Penaeus (Melicertus) kerathurus* in the Adriatic Sea.

Penaeus kerathurus is a widespread species on Mediterranean sandy-muddy seabeds; in the Adriatic Sea it was reported since 1863.

The data indicate that in the Adriatic Sea landing of this species increased from 167 t in 2004 to 550 tonnes in 2010. This work was performed in order to identify the causes that led such an increase of this stock.

The study of reproductive biology has shown that 53% of the specimens was represented by females. The size at first sexual maturity was 40.3 mm (LC) for females and 19.7 mm LC for males. The gonado-somatic index revealed a peak in July, while gonadal development was made up of 5 stages: previtellogenesis, early and late vitellogenesis, mature eggs and atresia. The profile of the gonadal lipid content and the GSI trends are well correlated.

The environmental characterization of a sea area protected by breakwaters showed the presence of juveniles *P. kerathurus* in the summer months, with a peak in August, showing high densities.

In addition, the lengths frequency distribution revealed a marked sexual dimorphism; the sizes of males showed a range between 17 and 38 mm while for females between 17 and 58 mm LC.

Therefore, the study area seem to represent a possible nursery area for various species, including *P. kerathurus*, which has been regularly surveyed during the summer period of three years of sampling. This suggests that the construction of breakwater barriers may have changed the habitat, forming a sort of man-made lagoon, with thermal/hydrodynamic characteristics ideal for prawn juveniles. These particular areas seem to positively act on the species growth.

The study also highlighted the high reproductive capacity of the species and the pronounced reproductive seasonality.

Effetti di attività antropogeniche sul ciclo vitale e abbondanza di *Penaeus (Melicertus) kerathurus* nel mare Adriatico.

Riassunto

Penaeus kerathurus è una specie diffusa su fondali sabbio-fangosi Mediterranei; in Adriatico fu segnalata a partire dal 1863.

I dati degli sbarcati indicano che in Adriatico questa specie è passata dalle 167 t del 2004 alle 550 t del 2010. Il presente lavoro è stato svolto al fine di identificare le cause che hanno portato all'aumento di questo stock.

Lo studio della biologia riproduttiva ha evidenziato che il 53% degli esemplari è rappresentato da femmine. La taglia di prima maturità sessuale è di 40,3 mm (LC) per le femmine e di 19,7 mm LC per i maschi. L'indice gonado-somatico ha rivelato un picco nel mese di luglio, mentre lo sviluppo gonadico è risultato composto da 5 stadi: previtellogenesi, vitellogenesi iniziale e avanzata, ovociti maturi ed atresia. Il profilo del contenuto lipidico delle gonadi e il trend dell'IGS sono risultati significativamente correlati.

La caratterizzazione ambientale di un tratto di mare protetto da frangiflutti ha dimostrato la presenza di esemplari giovanili di *P. kerathurus* nei mesi estivi, con un picco in agosto 2013, durante il quale la specie ha raggiunto elevate densità.

Inoltre la distribuzione di frequenza delle lunghezze ha evidenziato uno spiccato dimorfismo sessuale; i maschi hanno mostrato un range di taglie compreso tra 17 e 38 mm e compreso tra 17 e 58 mm LC le femmine.

Pertanto, l'area di studio sembra connotarsi come una possibile area di nursery per varie specie, tra cui *P. kerathurus*, che è stata regolarmente censita durante il periodo estivo dei tre anni di campionamento. Ciò lascia supporre che la costruzione di barriere frangiflutto possa aver modificato l'habitat, andando a formare una sorta di laguna artificiale, con caratteristiche termiche/idrodinamiche idonee ad essere sfruttate dalle fasi giovanili della mazzancolla, che in questo ambiente trovano le condizioni ambientali idonee per accrescersi.

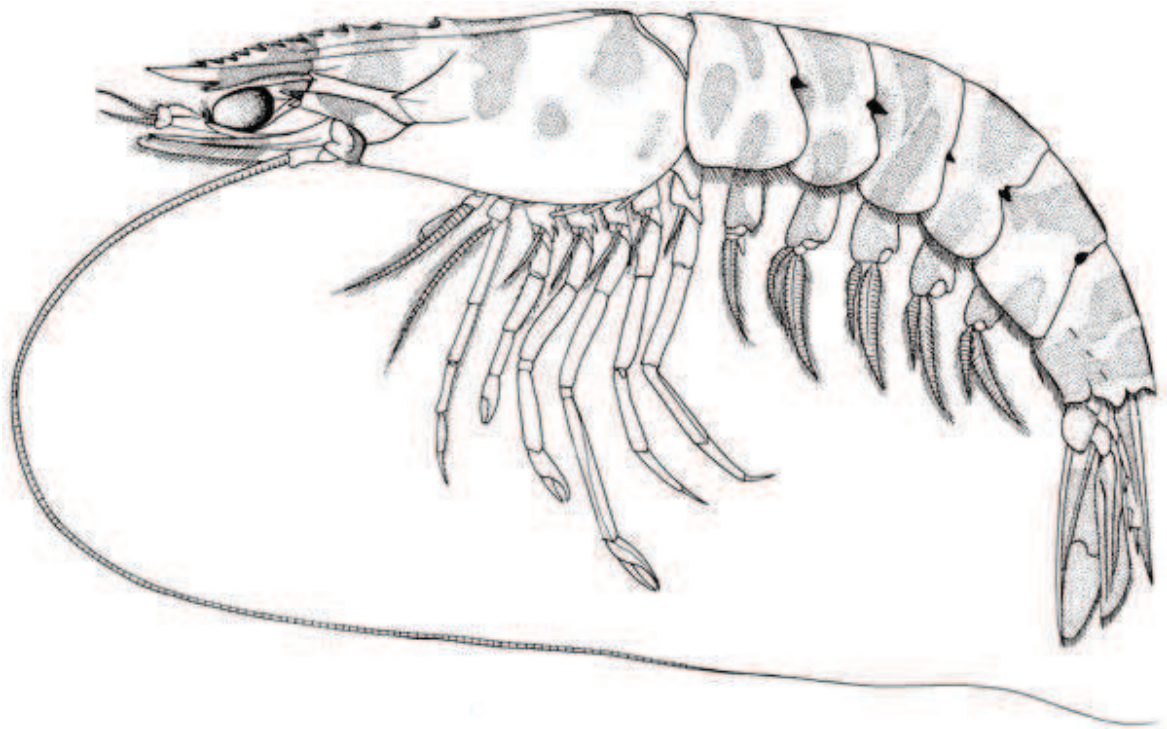
Lo studio ha inoltre evidenziato l'elevata capacità riproduttiva della specie e la netta stagionalità riproduttiva.

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Penaeus kerathurus (Forskål, 1775)



IT: mazzancolla; EN: caramote prawn; FR: caramote; ES: langostino

FAO-3alphacode: TGS
MEDITS code: PENAKER

(Bolognini 2015)

1. BIOLOGY OF THE CARAMOTE PRAWN

1.1 Morphology

Commonly known as caramote prawn, *Penaeus kerathurus* belongs to the Penaeidae family. It has a light coloration with transversal pink-light orange bands or spots (Fig. 1).

The rostrum is quite short and slightly exceeds the eyes; the lower edge shows a single tooth and this is a distinguishing mark of the species with respect to other penaeids, whilst it has a dozen teeth, reaching half carapace, on the upper edge. From this last point, another typical characteristics of the species can be observed: a deep and narrow furrow reaching the posterior edge, originated by a doubling of rostral carina. A crest runs parallel to this furrow and reaches the rostrum point, thus delimiting another furrow. The carapace is well calcified, with carinas and furrows between the supra-orbital and hepatic spine, in the anterior lateral half. A spine can be found on the coxopodite and basipodite of the Ist and IInd pair of pereio-pods. Posterior abdominal segments show a more marked dorsal carina, from the IVth segment, terminating in a spine in the VIth segment (Zariquiey Alvarez 1968).



Fig. 1. Lateral (above) and upper (below) view of *P. kerathurns*.

1.2 Ecology

1.2.1 Geographical and bathymetric distribution

The caramote prawn can be found in all Italian seas, in the Mediterranean and Eastern Atlantic, from England to Angola (Palomares & Pauly 2015), on soft bottoms of the continental shelf, typically at depths lower than 60 m (Frogliia et al. 2013). This crustacean, which is an indigenous species of the Mediterranean Sea, is reported in different sectors and areas with different names. However, the caramote prawn was unknown in the Adriatic sea till the second half of the last century and was reported for the first time only in 1863 (Heller 1863); Pesta in 1918 still expressed some doubts about its presence in the Northern Adriatic sea (Pesta 1918).

1.2.2 Habitat

P. kerathurus spends most of the day burried in the sediment where it lives; it gets out mainly during the night in order to feed and, during the reproduction season, to mate. It is a euryhaline species; during the breeding season it migrates in coastal waters and toward the river mouths. It can also be found in brackish water lagoons (Falciai & Minervini 1992). It is a typically resident species, and migrates towards and from the coast only to favor reproduction (Lumare et al.

1971). Adults can be found in typically marine salt waters, whilst during their post-larval and juvenile stages, the species favors low salinity environments, such as estuaries and lagoons, due to the abundance of preys. Its marked euryhalinity is due to an efficient osmoregulatory mechanism, which allows it to expel water in hypohaline environments and ions in hyperhaline environments (Palmeggiano 1983). This complicated osmoregulating system is negatively affected by temperature and determines winter migration towards the open sea, where temperature is more constant (Palmeggiano 1983).

1.2.3 Diet

This species has extremely varied feeding habits, mostly influenced by seasonal availability of benthic preys, mainly crustaceans, annelids and mollusks. Samples taken in the Central Adriatic sea (Prato et al. 2011), showed that amphipods represented the most abundant feeding resource, followed by mollusks and annelids. Among amphipods, the most represented genus, is *Ampelisca*, followed by *Corophium* and *Gammarus*. The same authors reported a prevalence of mollusks and amphipods in stomach contents, during summer and of annelids, mollusks and foraminifera during winter.

Several species, among which, nematodes and ophiurids are observed as occasional preys. Besides active predation, which is performed mainly during

the night, the caramote prawn does not disdain organic remains, which represent a necrophagous component of its diet. Juveniles (~60 mm CL) seem to feed mainly on amphipods and bivalves, as well as gasteropods and crustaceans (Prato et al. 2011), which are abundant in estuary and lagoon brackish waters.

1.3 Biology

1.3.1 Growth

Like most penaeids, also *P. kerathurus* shows a clear sexual dimorphism, due above all to the larger sizes of females which, in the Mediterranean, may reach a maximum total length of 225 mm, whilst in males the maximum size is of 180 mm (Holthuis 1980; Fig. 2). Anyway, the most common size in commercial landings is within a range of 110-140 mm TL for males and 130-170 mm TL for females (Holthuis 1980).

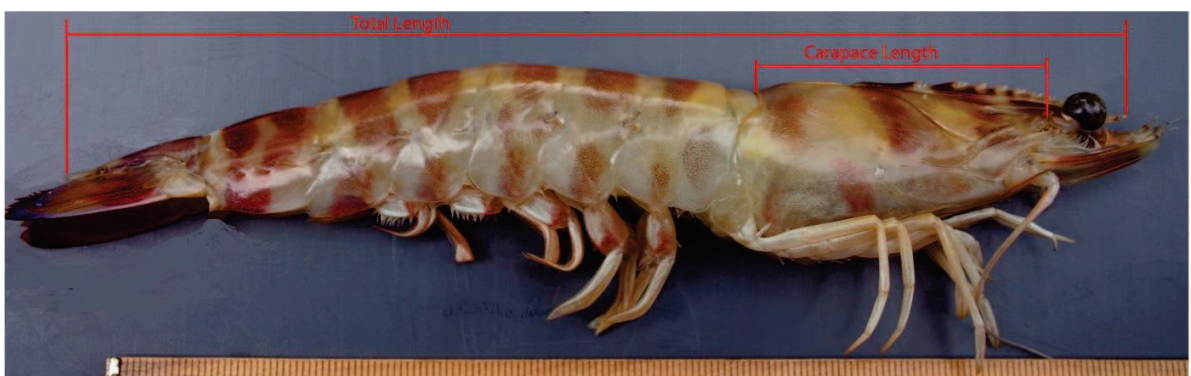


Fig. 2. Example of total length (TL), carapace length (CL) and tail length in a crustacean.

Growth is fast in the first months of life, and continues in the following months thanks also to good climatic conditions and abundance of food in coastal waters; seasonal differences, strictly associated with temperature can be observed. The average age is of about 20 months (Rodriguez 1987) and only a few specimens can reach their third year of life (Vitale et al. 2010; Froglija et al. 2013). In particular, in Italian seas, sizes of 63.3 mm CL for females and 52.7 mm CL for males were reported in western waters of the South of Sicily (Vitale et al. 2010) and 63 mm CL for females and 43 mm CL for males in Tuscani waters (Righini et al. 1998). Von Bertalanffy growth function parameters reported in literature for different Geographical sub-areas (GSA) of the Mediterranean Sea (Fig. 3) are shown in Tab. I.

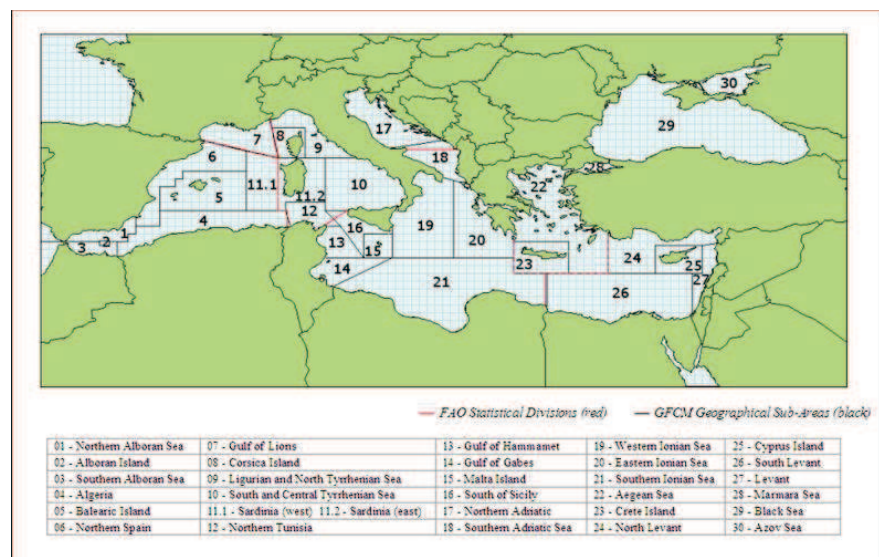


Fig. 3. Geographical sub-division of the Mediterranean Sea (FAO Statistical Division, red lines; GFCM Geographical Sub-Area, black lines)

The Geographical Sub-Areas (Fig. 3) were established by the *General Fisheries Commission for the Mediterranean* area division, through resolution GFCM/33/2009/2.

Tab. I. Von Bertalanffy growth function parameters.
GSA: Geographical Sub-Area.

GSA/Area	Method	Sex	L_{∞} (CL, mm)	k	t_0	Reference
GSA 9 Ligurian & North Tyrrhenian Sea	LFD	F	64.90	0.70	-	(Righini et al. 1998)
		M	46.00	0.90	-	
GSA 16 South of Sicily	LFD	C	72.00	0.78	-	(Vitale et al. 2010)
GSA 20 Eastern Ionian Sea	LFD	F	69.70	1.06	-	(Conides et al. 2006)
		C	59.69	1.05	-	
		M	62.70	1.25	-	
GSA 4 Algeria	LFD	F	64.14	0.80	-	(El Mekki 1994)
		M	45.50	1.00	-	
GSA 13 Gulf of Hammamet	LFD	F	54.25	0.60	-	(Ben Mariem 2004)
		M	37.46	0.78	-	

A study carried out by Vitale et al. (2010) off south-western coast of Sicily estimated a relationships between length/age (Tab. II), highlighting, through a length distribution analysis, that population was made up of two dominant modes, represented by one or two years old specimens, and only by a few three years old specimens. Another survey conducted along Tuscan coasts estimated size/age values smaller than the previous ones (Righini et al. 1998), as specified in Tab. II.

Tab. III shows parameters of the length-weight relationship $W = aL^b$ (Bolognini 2015).

Tab. II. *P. kerathurus* length at age.

GSA/Area	Sex	Age I	Age II	Age III	Reference
GSA 9 Ligurian & North Tyrrhenian Sea	F	32.7	48.90	57.00	(Righini et al. 1998)
	M	27.3	38.40	42.90	
GSA 16 South of Sicily	C	39.0	56.87	65.07	(Vitale et al. 2010)

Tab. III. *P. kerathurus* length-weight relationship parameters.

GSA/Area	Sex	a	b	Units	Reference
GSA 9 Ligurian & North Tyrrhenian Sea	F	0.0029	2.54	LC mm, W g	(Righini et al. 1998)
	M	0.0037	2.47	LC mm, W g	
	F	0.0036	2.48	LC mm, W g	(Carbonara et al. 2012)
	M	0.0015	2.71	LC mm, W g	
	C	0.0025	2.57	LC mm, W g	
GSA 10 South & Central Tyrrhenian Sea	C	0.0064	2.33	LC mm, W g	(Carbonara et al. 2012)
GSA 18 Southern Adriatic Sea	F	0.0027	2.55	LC mm, W g	(Carbonara et al. 2012)
	M	0.0017	2.69	LC mm, W g	
	C	0.0031	2.51	LC mm, W g	
GSA 19 Western Ionian Sea	F	0.0043	2.42	LC mm, W g	(Carbonara et al. 2012)
	M	0.0021	2.63	LC mm, W g	
	C	0.0053	2.36	LC mm, W g	
GSA 13 Gulf of Hammamet	F	0.0036	2.48	LC mm, W g	(Ben Mariem 1995)
	M	0.0047	2.41	LC mm, W g	
Gulf of Cádiz	F	$7.36 \cdot 10^{-6}$	3.00	LC mm, W g	(Rodriguez 1987)
	M	$3.28 \cdot 10^{-6}$	3.17	LC mm, W g	

1.3.2 Sexual maturity and reproduction

P. kerathurus sexual activity is strongly affected by water temperature (Holthuis 1980). Actually, gonadal maturation takes place during spring/summer (Lumare et al. 1971). June and July are the months with the highest percentage of mature females (about 85%): in this period all females have spermatophores adhering to their *thelycum*, as reported by Lumare et al. (2011) in south Adriatic. Usually,

mating occurs during the night, between a non-exuviating male and an exuviating female (Scovacricchi 1994). A male lays 2 winged spermatophores into female's *thelycum*. The spawning takes place during the night and usually has a short duration. The number of eggs varies according to animal size (Scovacricchi 1994). Upon spawning, eggs have an average diameter of about 0.2-0.3 mm. Hatching begins in around 16-30 hours (Heldt 1938) and, larval succession entails three different phases: nauplius, zoea and mysis (Fig. 4).

This species performs the typical larval succession of decapod penaeidae crustaceans. The first larval stage (nauplius) moves towards the surface where phytoplankton is more abundant. A nauplius exuviates several times, and grows up till full transformation in protozoea and, then, in zoea. The latter actively feeds on phytoplankton and it is strongly attracted by light and brackish water environments (estuaries and lagoons) where it can find extremely favorable trophic and thermal conditions, above all during spring-summer. Once it reaches the last larval stage of mysis, it starts feeding on zooplankton. Benthic post-larvae go on developing in brackish waters and coastal environments until they reach a juvenile stage. At the end of summer season juveniles leave coastal areas, and settle on infralittoral sandy bottoms, where waters are less affected by surface temperature variations (Scovacricchi 1994; Fig. 4).

According to Rodriguez (1987), size at first maturity of females is estimated to be around 120 mm TL, whilst Lumare et al. (2011) estimates such size in 31.4 mm CL for males and 45.5 mm CL for females (Tab. IV).

Righini et al. (1998) reported females/males ratio at 0.9 during spring, 1.2 during summer, 1.0 during autumn and 1.1 during winter, therefore slightly in favor of females, as observed by Rodriguez (1977) in the Gulf of Cádiz. Lumare et al. (2011) reported a males/females ratio in southern Adriatic sea of 1.44, with fluctuations comprised between 0.61 and 2.70.

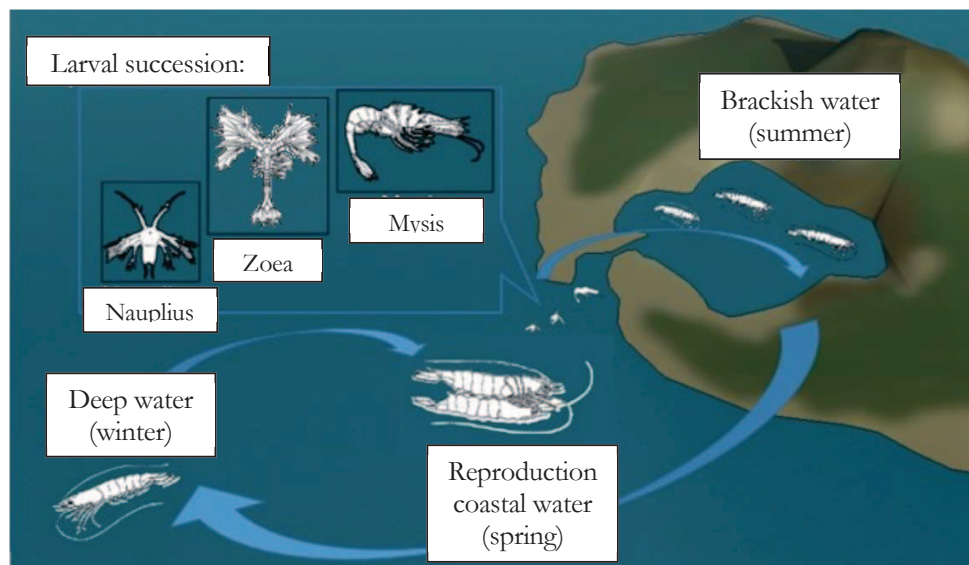


Fig. 4. Biological cycle of *P. kerathurus* (from Palmeggiano 1983, modified by Author, Bolognini 2015).

Tab. IV. *P. kerathurus* reproductive period and size at first maturity.

GSA/Area	Spawning period	Sex	Size at 50% maturity (CL, mm)	Reference
GSA 17 Central & Northern Adriatic Sea	Jun-Jul	F	45.56	(Lumare et al. 2011)
		M	31.42	

1.3.3 Recruitment

Lumare (2009) reports that caramote prawn reproduction takes place in coastal waters, close to the shore (June-July). Within one or two months at a weight around 0.25-1.50 g juveniles migrate into lagoons. At the end of summer (September-October), due to temperature decrease and a reduction of trophic resources, young caramote prawns migrate in the opposite direction, from lagoon to the open sea (Fig. 4).

1.4 Evaluation

Caramote prawn is extremely appreciated by consumers, with a high commercial value at the markets (Lumare & Scordella 2001). However, catches of this species were not important, from an economical point of view, until a few years ago, considering modest landings; indeed, it appeared in FAO statistics only in 2005. Although caramote prawn occurs in all Italian seas, it

does not represent a targeted species but it is a high-valuable by-catch of bottom trawl fishery. This species is caught also with passive nets, particularly during summer in southern Adriatic (Lumare et al. 2011) and during autumn in Tuscan waters (Righini et al. 1998).

In the Mediterranean Sea the assessment of demersal resources is carried out in the framework of “MEDITS project”. The MEDITS survey programme (International bottom trawl survey in the Mediterranean) intends to produce basic information on benthic and demersal species in terms of population distribution as well as demographic structure, on the continental shelves and along the upper slopes at a global scale in the Mediterranean Sea, through systematic bottom trawl surveys. However, density and biomass indexes obtained within MEDITS surveys are not indicative of the real abundance of such species because of the type of sampling gear and sampling protocol. In fact, data coming from the exploratory fishing surveys in the framework of SoleMon project (AdriaMed 2011) carried out with rapido trawl (a towed trawl net with a rigid frame with rakes at mouth) in the GSA 17, showed higher biomass and density indexes than those obtained during MEDITS survey, although fluctuating between 85.9 and 314.4 ind./km² (Fig. 5).

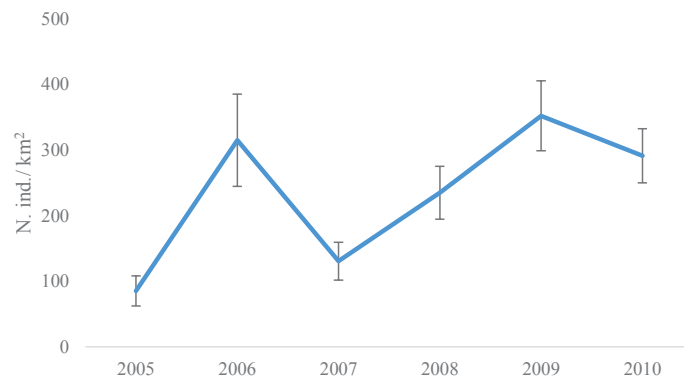


Fig. 5. *P. kerathurus*. Temporal evolution of the density index (No. ind./km²±standard deviation) in GSA 17 (SoleMon, late-fall surveys; AdriaMed, 2011, modified).

1.5 Exploitation

1.5.1 Fishery

Official commercial landing data for the period 2004-2013, highlighted the importance gained by this species in recent years, above all in the GSAs 9, 17 and 18. For example, in GSA 17 landings increased from 167 t in 2004 up to 550 t in 2010 (Tab. V; Bolognini 2015). In this latter area, a peak of catch is recorded in the last quarter of the year, ascribable to migration of the new generation born during summer, towards the open sea, when recruitment into fishery takes place (Frogliia et al. 2013). Anyway, the high price of caramote prawns, independently of landing quantitative values, makes this resource important for fishery in certain period (Frogliia et al. 2013; Righini et al. 1998).

Tab. V. *P. kerathurus* official landings (tons) in the Italian GSAs from 2004 to 2013 (source MIPAAF).

GSA/Area	2004	2005	2006	2007	2008	2009	2010	2011	2012	2013
GSA 9 Ligurian & North Tyrrhenian Sea	30.7	86.5	59.7	82.1	85.6	124.9	142.6	128.3	145.5	73.6
GSA 10 South Tyrrhenian Sea	2.7	7.3	21.8	25.6	13.9	17.8	15.7	22.9	22.7	69.4
GSA 16 South of Sicily	19.2	18.2	23.1	7.4	1.7	13.1	1.3	0.9	2.1	0.4
GSA 17 Northern Adriatic Sea	169.0	179.0	317.9	690.5	501.7	515.2	550.3	545.9	324.1	386.2
GSA 18 Southern Adriatic Sea	5.2	1.4	7.2	32.5	30.5	111.1	116.8	28.3	75.3	100.7
GSA 19 Western Ionian Sea	34.1	44.9	95.4	25.0	3.2	7.5	25.2	20.5	7.4	7.7

1.5.2 Exploitation pattern

In Italy *P. kerathurus* is mainly caught by bottom trawling (both otter- and rapido trawl), and to a marginal extent, by small-scale fishing gears and polyvalent passive gears. National statistics (MIPAAF: Ministero delle Politiche Agricole Alimentari e Forestali; IREPA: Istituto di Ricerche Economiche per la Pesca e l'acquacoltura) show a general increase of carapide prawn catches in the period 2004-2012. Comparing catches trends obtained by different gears, bottom trawl showed an increase during the time, although a slight decline was recorded in the last years; vice-versa, catches produced by small-scale fishery showed a decreasing trend in the investigated period; finally, catches obtained with polyvalent passive gears had a minor importance

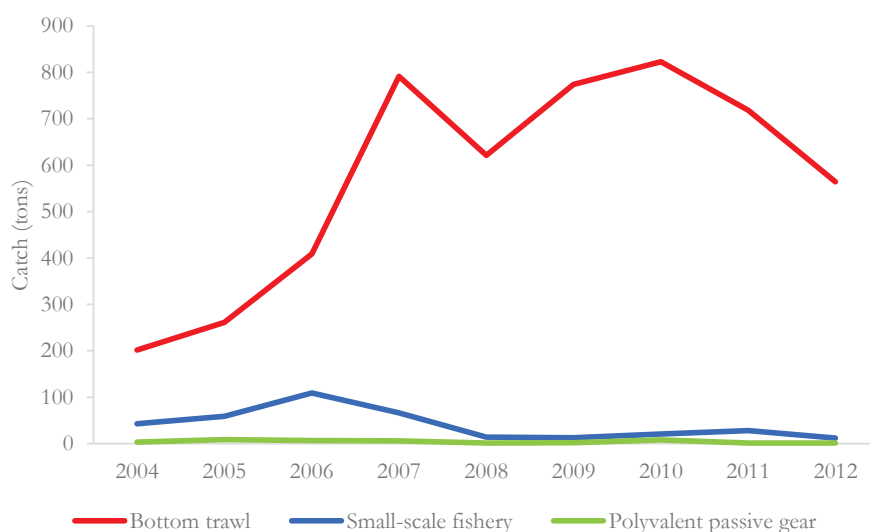


Fig. 6. *P. kerathurus* landing evolution (tons) in the period 2004-2012 for different gears in the Italian seas (source MIPAAF-IREPA).

(Bolognini 2015; Fig. 6). By way of example, Fig. 7 shows distribution according to size-frequency of *P. kerathurus* landings, obtained during SoleMon surveys, in GSA 17 in the period 2005-2010.

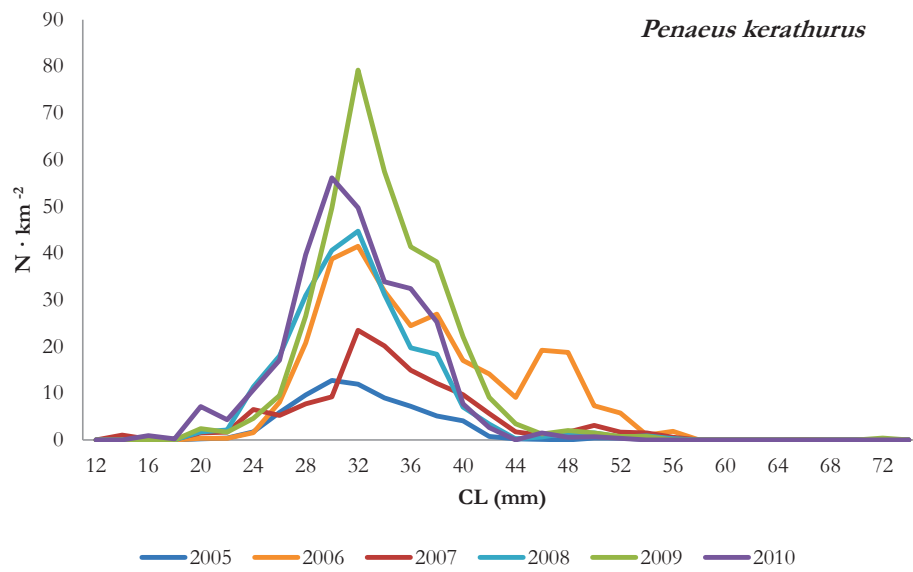


Fig. 7. *P. kerathurus* length-frequency distribution of the specimens in GSA 17 (AdriaMed, 2011, modified).

1.6 Captures trends of *P. kerathurus* at different spatial and time scale

1.6.1 Global capture

Due to its economic importance, *P. kerathurus* is caught in inshore water along all the Mediterranean coasts; instead, along the West African coast the species is of minor importance.

An overview of global catches of caramote prawn shows an increasing growth, more evident from 1970's until now (Fig. 8; FAO 2015).

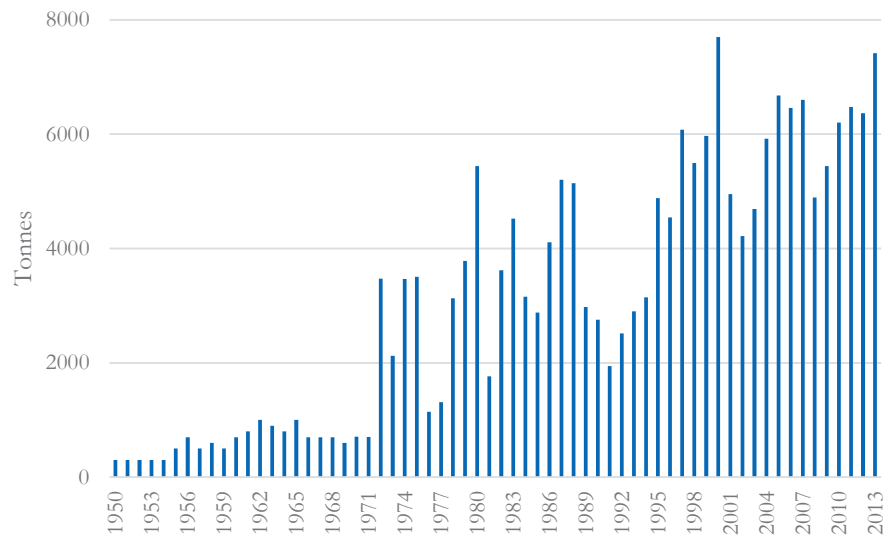


Fig. 8. Global capture production (tonnes) for *P. kerathurus* (1950-2013; FAO 2015).

1.6.2 National capture

Fishery statistics point out that catches of this species has become important in Italy only recently; in fact it appeared for the first time in 2005.

It is possible to remark a rapid growth, with maximum catches (over 850 tons) in 2007 and 2010 (Fig. 9; FAO, 2015).

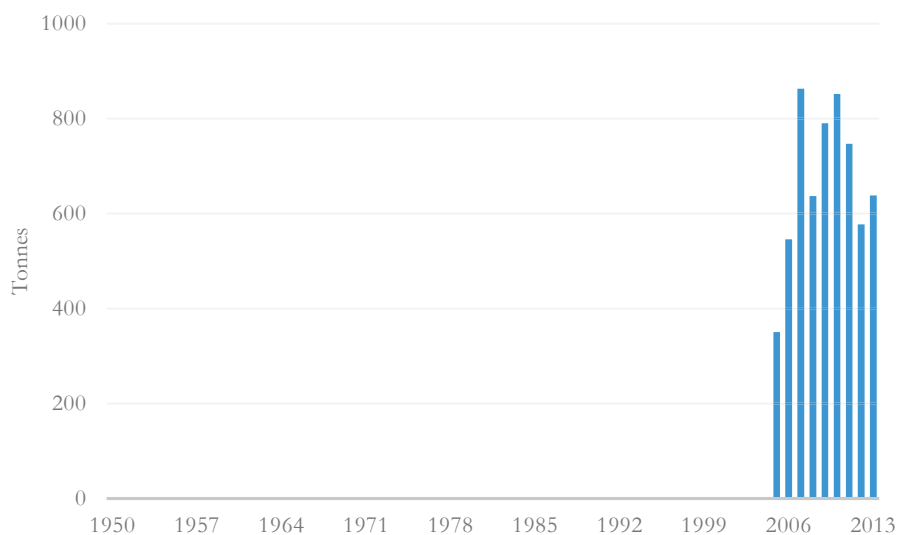


Fig. 9. Italian capture production (tonnes) for *Melicertus kerathurus* (1950-2013; FAO , 2015)

1.6.3 Capture in GSA 17: Northern Adriatic Sea

A careful analysis of catches, comparing 2004 with 2012, highlights a significant decrease in landings and revenues, for the whole Geographical sub-area 17 "Northern Adriatic" (Fig. 10).

This phenomenon reflects what has been observed on a large scale in the whole Mediterranean basin. In detail, in the Geographical sub-area 17 some species showed a decrease that exceeded 50% both in landings and in revenues (Fig. 11). Only the caramote prawn showed an opposite trend, with an increase of

91.7% in landings (155 tons) and of 41.8% in revenues (1683 thousands of euro;
 source: *Mipaaf – Irep*; Tab. VI).

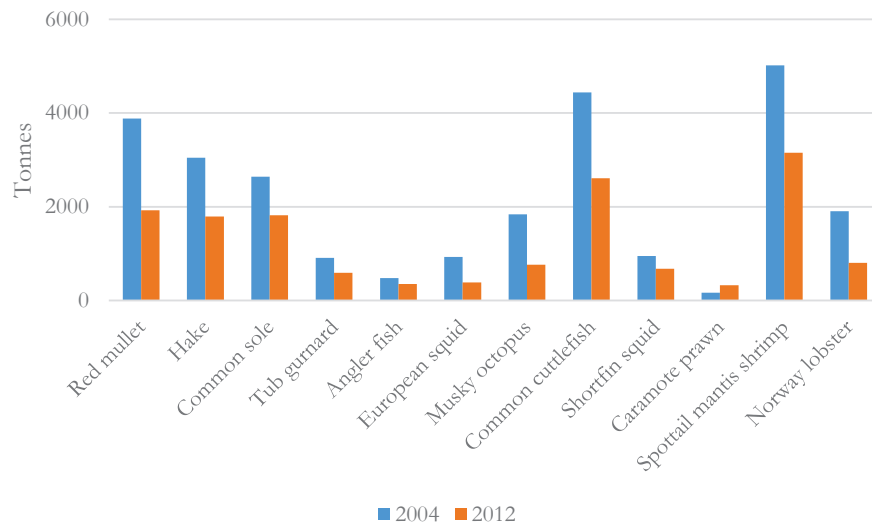


Fig. 10. Landings of some demersal stocks in GSA 17 – Northern Adriatic, in 2004 and 2012.

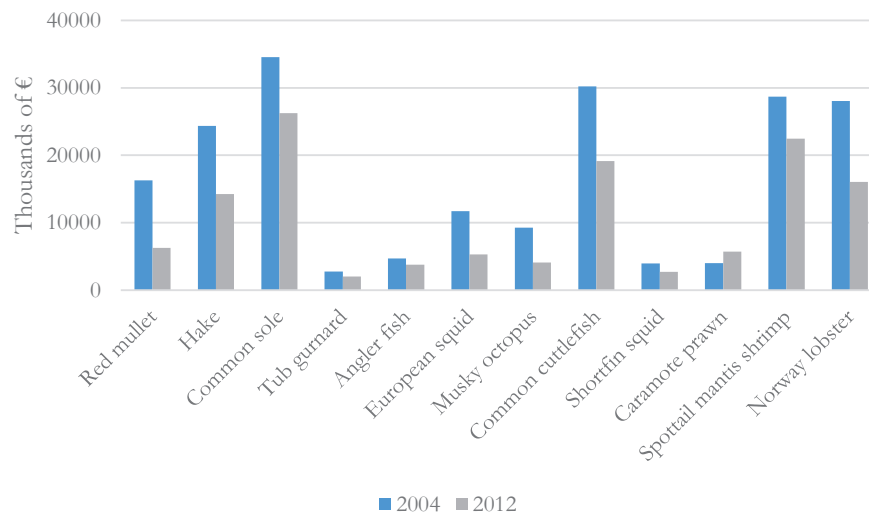


Fig. 11 Revenues (thousands of €) of some demersal stocks in GSA 17 – Northern Adriatic Sea, in 2004 and 2012.

Tab. VI. Comparison between years 2004 and 2012 of some demersal target species expressed in tonnes (t) and thousands of euro (000 €) in Geographical Sub-Area 17 "Northern Adriatic" (source Mipaaf - IREPA).

Geographical Sub-Area 17 "Northern Adriatic"									
Demersal target species		2004		2012		Variation		Variation	
		t	000 €	t	000 €	t	t %	000 €	000 € %
F i s h	<i>Red mullet</i>	3883	16262	1926	6251	-1342	-50.4%	-10011	-61.6%
	<i>Hake</i>	3045	24335	1794	14251	-658	-41.1%	-10084	-41.4%
	<i>Common sole</i>	2639	34531	1821	26264	-287	-31.1%	-8267	-23.9%
	<i>Tub gurnard</i>	908	2767	588	2008	-215	-35.2%	-759	-27.4%
	<i>Angler fish</i>	480	4691	350	3778	-152	-27.1%	-913	-19.5%
Tot. Fish		10955	82586	6479	52552	-2654	-40.9%	-30034	-36.4%
M o l l u s c	<i>European squid</i>	932	11717	382	5320	-211	-59.0%	-6397	-54.6%
	<i>Musky octopus</i>	1839	9285	766	4089	-451	-58.3%	-5196	-56.0%
	<i>Common cuttlefish</i>	4439	30217	2609	19131	-450	-41.2%	-11086	-36.7%
	<i>Shortfin squid</i>	951	3955	680	2711	-184	-28.5%	-1244	-31.5%
Tot. Mollusc		8161	55174	4437	31251	-1296	-45.6%	-23923	-43.4%
C r u s t a c e a n s	<i>Caramote prawn</i>	169	4028	324	5711	155	+91,7%	1683	+41,8%
	<i>Spottail mantis shrimp</i>	5013	28709	3150	22487	-1863	-37.2%	-6222	-21.7%
	<i>Norway lobster</i>	1904	28036	803	16037	-382	-57.8%	-1999	-42.8%
Tot. Crustaceans		7086	60773	4277	44235	-125	-39.6%	-16538	-27.2%
Grand Total		26202	198533	15193	128038	-4075	-42.0%	-70495	-35.5%
<i>Source: Mipaaf - IREPA</i>									

1.6.4 Regional capture in the Adriatic Sea

In the last few years (2004 - 2013), fishing data from the trawl fleets of the Adriatic regions, show significant regional differences in terms of fishing effort (expressed in kilowatts, KW), total catch (expressed in tonnes, t) and catch per unit of effort (KgKW^{-1} , CPUE). Also for prawn was just the same. Emilia Romagna was the most productive region in terms of catch per unit effort (8.4 kgKW^{-1} in 2007 and 4.7 kgKW^{-1} average of the period). Marche region was one with the highest catches totaled 298 t in 2010 and 177.5 t average of the period (Tab. VII).

Generally, since their appearance in the national statistics, catches of caramote prawn in the Adriatic Sea exhibited strong growth, with a peak that exceeded 700 tonnes in 2007. Although in recent years the catches of this species have remained at lower values, their trend appears evidently growing (Fig. 12), contrariwise from the trend observed for many groundfish species in this basin.

Tab. VII. Total effort (KW), total catches (t) and Total Catch per Unit Effort (kgKW⁻¹), and catches and Catch per Unit Effort of caramote prawn for the Italian Adriatic regions, for the Geographical sub-area 17 and for the whole Adriatic Sea (data from Puglia region of 2013 refer only to the Adriatic side; source: *Mipaaf-IREPA*).

Region			Year										Average period
			2004	2005	2006	2007	2008	2009	2010	2011	2012	2013	
F r i u l i	Tot.	KW	7656	7370	9047	8345	8344	8171	8101	7807	6521	3563	7492.5
		t	1013	1056	1564	1986	1253	1381	1074	834	1033	424	1161.8
		kg/KW	132	143	173	238	150	169	133	107	158	119	152
	Caramote prawn	t	1	1	4	34	2	4	1	1	1	1	5
		kg/KW	0.1	0.1	0.4	4.1	0.2	0.5	0.1	0.1	0.2	0.3	0.6
V e n e t o	Tot.	KW	47959	49186	45170	43025	41277	40260	41312	41230	38899	25334	41365.2
		t	8215	7314	7060	7857	6207	6221	4894	3917	4096	7136	6291.7
		kg/KW	171	149	156	183	150	155	118	95	105	282	156
	Caramote prawn	t	2	6	15	98	39	31	24	18	14	13	26.0
		kg/KW	0.0	0.1	0.3	2.3	0.9	0.8	0.6	0.4	0.4	0.5	0.6
E m i l i a	Tot.	KW	41513	32758	33599	31482	31754	32190	31209	34640	32925	28346	33041.6
		t	8963	6377	5683	6434	6084	5679	4737	4706	4326	2595	5558.4
		kg/KW	216	195	169	204	192	176	152	136	131	92	166
	Caramote prawn	t	83	76	164	264	190	189	164	233	117	47	152.7
		kg/KW	2.0	2.3	4.9	8.4	6.0	5.9	5.3	6.7	3.6	1.7	4.7
M a r c h e	Tot.	KW	61648	56412	48777	44410	44082	45361	44654	40345	41015	33827	46053.1
		t	9968	11457	11666	10763	9326	8902	8693	7138	5892	6073	8987.8
		kg/KW	162	203	239	242	212	196	195	177	144	180	195
	Caramote prawn	t	59	78	123	257	213	235	298	211	132	169	177.5
		kg/KW	1.0	1.4	2.5	5.8	4.8	5.2	6.7	5.2	3.2	5.0	4.1
A b r u z z o	Tot.	KW	39773	29106	23454	20410	20107	20800	22385	22055	22818	22326	24323.4
		t	7064	6121	5375	4078	3393	3873	3109	3001	3140	3186	4234.0
		kg/KW	178	210	229	200	169	186	139	136	138	143	173
	Caramote prawn	t	12	9	3	8	25	22	25	30	6	84	22.4
		kg/KW	0.3	0.3	0.1	0.4	1.2	1.1	1.1	1.4	0.3	3.8	1.0
M o l i s e	Tot.	KW		7692	7859	8762	9034	9190	9584	9277	9046	7263	8634.0
		t		1188	981	1377	1584	1508	1594	1573	1349	918	1341.3
		kg/KW		154	125	157	175	164	166	170	149	126	154
	Caramote prawn	t	12	10	9	28	32	34	38	52	54	53	32.2
		kg/KW	N.A.	1.3	1.1	3.2	3.5	3.7	4.0	5.6	6.0	7.3	4.0
G S A 1 7	Tot.	KW	198549	182524	167906	156434	154598	155972	157245	155354	151224	120659	160046.4
		t	35223	33513	32329	32495	27847	27564	24101	21169	19836	20332	27440.9
		kg/KW	177	184	193	208	180	177	153	136	131	169	171
	Caramote prawn	t	169	180	318	689	501	515	550	545	324	367	415.8
		kg/KW	0.9	1.0	1.9	4.4	3.2	3.3	3.5	3.5	2.1	3.0	2.7
P u g l i a	Tot.	KW	106512	103210	100761	95523	95865	96207	95633	84788	81982	68035	92851.6
		t	22.713	19.937	21.751	17.720	16.618	19.128	16.280	13.962	12.059	15.647	17581.5
		kg/KW	213	193	216	186	173	199	170	165	147	230	189
	Caramote prawn	t	5	1	7	32	30	111	117	28	76	101	50.8
		kg/KW	0.0	0.0	0.0	0.3	0.3	1.2	1.2	0.3	0.9	1.5	0.6
A d r i a t i c a	Tot.	KW	305061	285734	268667	251957	250463	252179	252878	240142	233206	188694	252898.1
		t	57936	53450	54080	50215	44465	46692	40381	35131	31895	35979	45022.4
		kg/KW	190	187	201	199	178	185	160	146	137	191	177
	Caramote prawn	t	174	181	325	721	531	626	667	573	400	468	466.6
		kg/KW	0.6	0.6	1.2	2.9	2.1	2.5	2.6	2.4	1.7	2.5	1.9

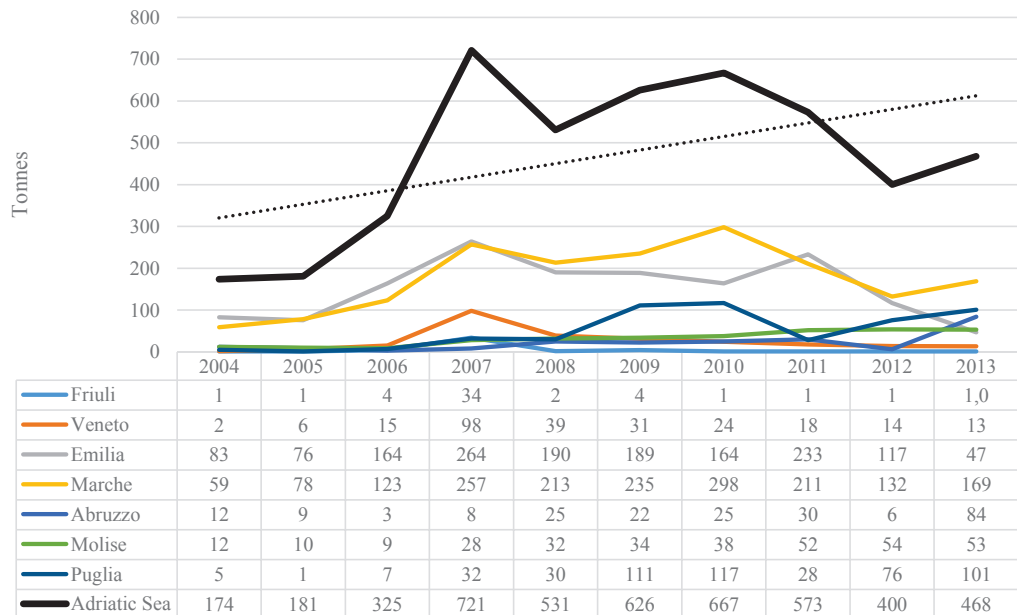


Fig. 12. Catches of caramote prawn (t) in the Adriatic regions (coloured lines), in the whole Adriatic Sea (black line) and its trend (dashed line) in the period 2004-2012.

1.6.5 Local capture of *P. kerathurus*

The statistics of *P. kerathurus* landings are not available prior to 1980 because at that time the common practice at the fish markets entailed the selling of few specimens in mixed boxes. Only at the end of 80's, when landings significantly increased, the caramote prawn catches were recorded separately in the statistics for product auctioned (Frogliata et al. 2013). Data available for the Northern Adriatic fishing harbor as Chioggia (Fig. 13; Clodia database, 2013) and Central

Adriatic as Ancona (Fig. 14; Froglija et al. 2013) point out a general increase in caramote prawn landings in the observed period.

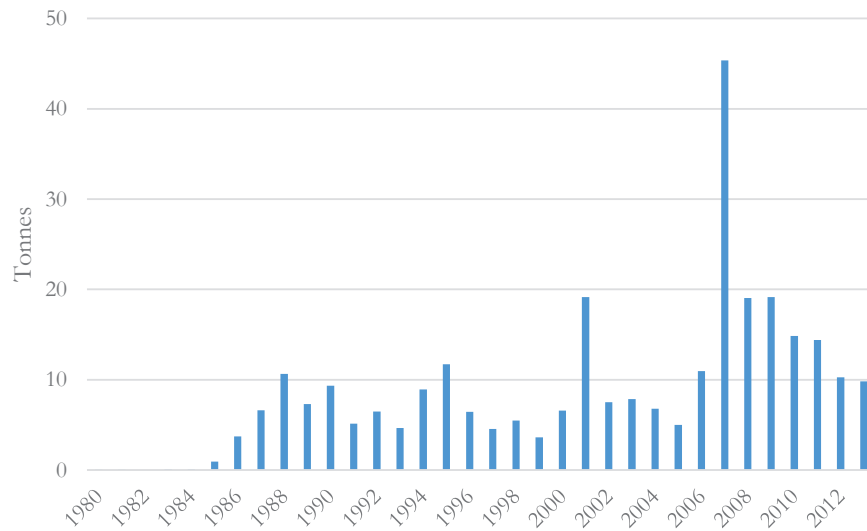


Fig. 13. *P. kerathurus* landings (t) of Chioggia fishing harbor (Northern Adriatic Sea) from 1980 to 2013 (Clodia database, 2013)

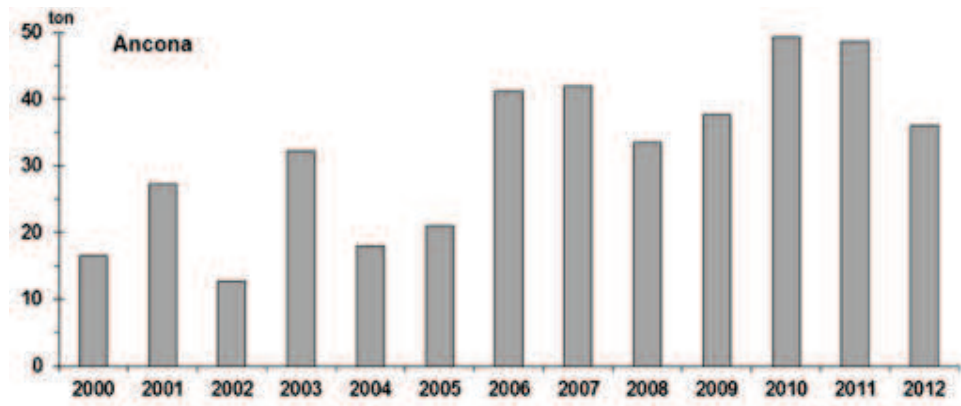


Fig. 14. Catch of *P. kerathurus* landings (t) auctioned by the trawlers fleet based in the Ancona fishing harbor (Central Adriatic Sea) from 2000 to 2012 (Frogliata et al. 2013).

2. HYPOTHESIS AND PURPOSE OF THE STUDY

The presence of *P. kerathurus* in the Adriatic Sea appears to be relatively recent, dating the first uncertain records on the second half of 800's - early 900's from the southern side. At the beginning, this species bypassed the fishery statistics, because of the marketing practices in the Adriatic markets (see Froglija et al. 2013). However, the increased catches in recent years have made this species an object of interest, with the remarkable retail assessment of 36 €kg⁻¹ (personal observation, 2015). In addition, the recent crisis of the fisheries sector and general decrease of fisheries resources leded a growing interest for this species. Froglija et al. (2013) reported that the increase of *P. kerathurus* in certain areas and periods was linked to warmer waters and to a general decrease of other crustaceans such as the norway lobster *Nephrops norvegicus*, typical of cold-temperate waters.

The same authors refer to the *P. kerathurus* stock increase as another example of "meridionalization", a northward expansion of Mediterranean species in the past restricted to the southern sectors as a consequence of the Mediterranean Sea warming (Axaopoulos P. 2010).

These authors hypothesized the synergical action of at least two factors:

- 1) The Italian annual trawling ban for 30-45 days at the end of the summer.

2) An extension of nursery ground due to the construction of breakwaters, which could enhance recruitment.

The first factor, in force since 1987, aims at reducing fishing effort exerted by towed gears in the recruitment period of several species having a commercial interest. *P. kerathurus* could have benefit to this effort reduction in coastal water. This species is characterized by fast growth, reproduction at the end of first year of life and short life-span (max. <3 years).

As concerns the breakwaters, they represent a man-made structures that have been built in the last 50 years at 100-200 m from shoreline, parallel to the shore, to reduce erosion of sandy beaches (Dugan J.E. et al. 2011), a problem that plagues the Western Adriatic coast.

The purpose of this study is to investigate the biology and ecology of the carapote prawn in order to understand possible causes of the stock growth in the Adriatic, in contrast with the decreasing trends observed for other stocks already exploited by the commercial fishery. Based on the information reported on literature for this species, we want to elucidate if and how the construction of breakwaters can influence the biological cycle of the selected species, the role of water temperature, as well as filling some gaps on its reproductive biology and on the main parameters of the population in the selected area. The ultimate goal is to define how the biotic, abiotic and anthropogenic factors can affect the stock of *P. kerathurus* in Northern and Central Adriatic Sea.

3. REPRODUCTIVE BIOLOGY

3.1 Introduction

To better formulating proper management practices for fishery science, one of the main issue is reproductive biology. To understand the dynamics of a stock is crucial the relation between size and reproduction event, such as mating and sexual maturity (Perdichizzi et al. 2012).

One of the most important factor determining the reproductive potential of a species is the reproductive pattern, therefore the importance to understand the reproductive biology (ovarian development), such as for future domestication of the species in local waters (Küçükdermenci & Lok 2011).

In addition, standardization criteria for evaluating sexual maturity in target species was strongly recommended by International Council for the Exploitation of the Sea (ICES) and General Fisheries Council for the Mediterranean (GFCM). An Expert Group of ICES, recently (2010) has highlighted the need for a common system for classification of maturity stages in Crustaceans, which also pointed out the importance of histology as a tool for obtaining the highest accuracy in these type of studies (Perdichizzi et al. 2012).

The reproductive cycle of penaeids are affected by many factors as seasonal rainfall, temperature regime, and depth, exhibiting a rather complex life history (Dall et al. 1990; Crocos et al. 2001). Typically, a bimodal seasonal spawning

patterns was exhibit by tropical and subtropical penaeids, while spawning becomes unimodal in penaeids that inhabit temperate latitudinal zones, characterized by only one well-defined recruitment period (Dall et al. 1990; Staples 1991; Crocos & van der Velde 1995).

During the reproductive cycle of crustaceans decapod, the ovarian development is accompanied by changes in color and size (Adiyodi & Subramonian 1983; Arculeo et al. 1995; Cavalli et al. 1997). In some species these changes in color are easily visualized and result from carotenoids content differences, which play an important role during embryogenesis (Goodwin 1951; Dall et al. 1995; Liñán-Cabello et al. 2002).

In recent years increased attention has been reported to the ovarian development of penaeid shrimp, and the macroscopic and histological characterization have been studied for various species, such as *Melicertus japonicas* (Yano 1988), *Melicertus vannamei* (Arcos et al. 2005), *Melicertus semisulcatus* (Badawi 1975), *Melicertus kerathururs* (Medina et al. 1996; Küçükdermenci & Lok 2011), *Melicertus monodon* (Tan-Fermin 1991; Qunitio et al. 1993), *Litopenaeus setiferus* (King 1948), *Penaeus indicus* (Subrahmanyam 1965), *Melicertus aztecus* (Caillouet 1972), *Fenneropenaeus merguensis* (Crocos & Kerr 1983), *Lucifer orientalis* (Oka & Shirata 1965) and *Melicertus plebejus* (Kelemec & Smith 1980).

As suggested by many works, a crucial role during natural and induced sexual maturation of various shrimp species is exerted by lipid deposition that occurs in the ovary (Gehring 1974; Teshima & Kanazawa 1983; Galois 1984; Teshima, Kanazawa, Horinouchi, et al. 1988; Teshima, Kanazawa, Koshio, et al. 1988; Jeckel et al. 1989; Teshima et al. 1989; Bray et al. 1990). Several authors have suggested that successful maturation is dependent on diet (Brown, A. et al. 1979; Lawrence et al. 1980; Cahu & Quazuguel 1989). Relatively lipid richness of ovaries and eggs compared to the adult shrimp is due to the limited capacity since juvenile for *de novo* phospholipid synthesis and inability to synthesis cholesterol (Teshima 1982; Teshima et al. 1986), then the quality of the dietary lipid source may be of great importance in maturation. There are very limited information on the quantitative changes that occur in the different lipid classes and their fatty acids during the maturation period.

The present study was focused on gametogenic cycle of caramote prawn in the northern and central Adriatic Sea. The current study aimed to contribute to characterizing reproduction in *P. kerathurus* using qualitative techniques such as histology of the ovaries and testis; quantitative parameters such as morphometry (carapace length CL, total length TL, body weight W, gender and gonado somatic index GSI) and combined technique (quali-quantitative) such as lipid composition profile of the ovary during maturation. An important role is played by lipids as sources of energy and as cell constituents in the processes

of spawning, embryogenesis, hatching and early development of crustacean larvae. The present study also aimed for the first time in Mediterranean Sea at explaining the variation in the lipid contents of the ovary during sexual maturation in wild *P. kerathurus*, based on the most recent analysis techniques.

3.2 Materials and Methods

3.2.1 Sampling

Specimens of wild *P. kerathurus* were monthly captured (when possible) from August 2013 to September 2015 from northern and central Adriatic Sea, Italy (Fig. 15). Specimens were collected from 39 sampling activities, 22 of them were scientific surveys and 17 of them were commercial fishery. Due to characteristic species migration, samples by commercial fishery were mainly obtained from trawlers in the period fall-winter and by artisanal fisheries in spring.

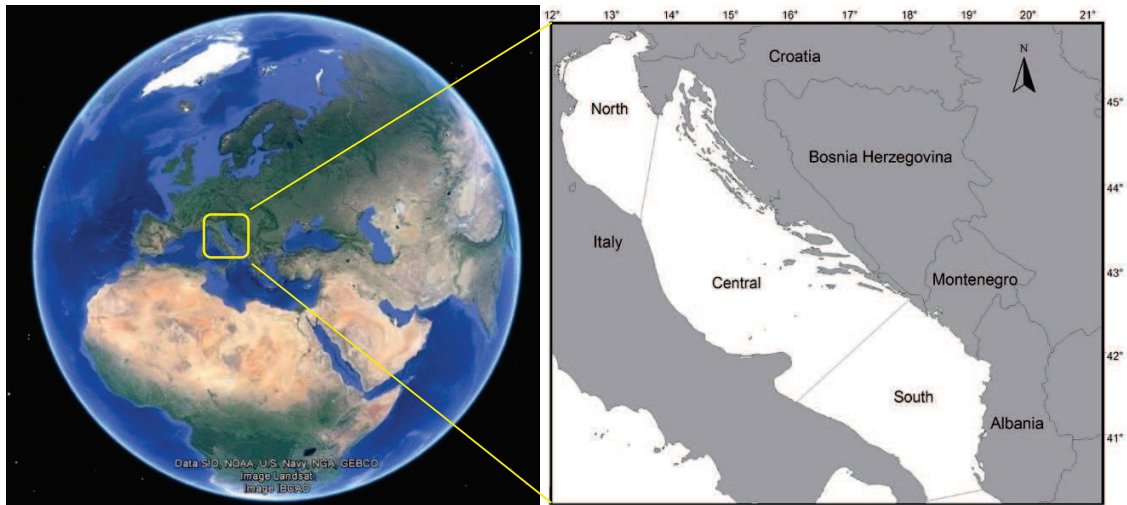


Fig. 15. *P. kerathurus* sampling area in northern and central Adriatic Sea, Italy.

3.2.2 Prawn Measurement

Using a Vernier calliper with an accuracy of 0.01 mm, the carapace length (CL) of each specimens collected was measured from the eye orbit to rear dorsal end of the carapace (Fig. 16). Body wet weight was measured to the nearest 0.1 g (electronic balance *Mettler PL 3000*). Females were dissected; ovaries were gently removed and weighted at 0.0001 g (electronic analytical balance *Mettler Toledo ML 204*).

GSI was expressed as the percentage of the ovarian weight in relation to the total body weight (Medina et al. 1996) and calculated as:

$$GSI = \frac{Weight_{gonad}}{Weight_{total} - Weight_{gonad}} \times 100$$

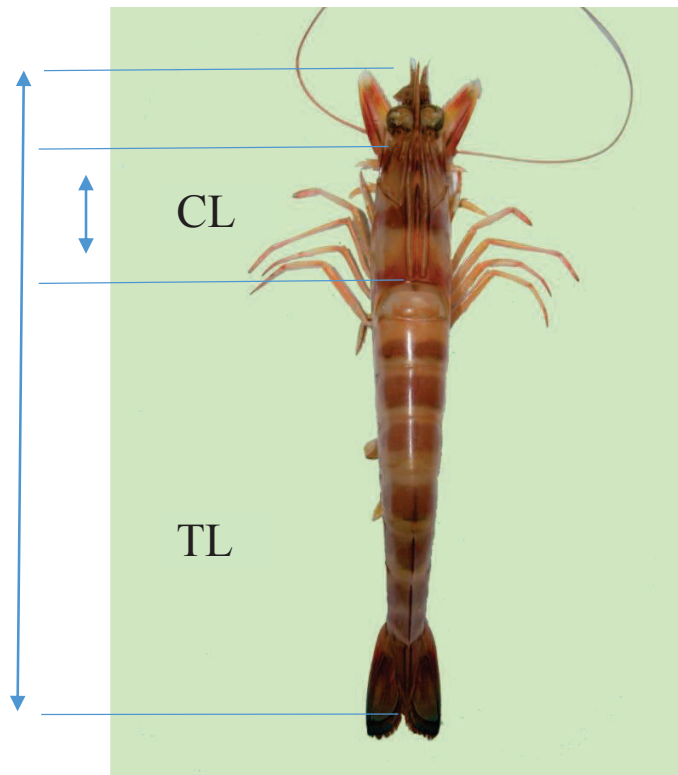


Fig. 16. Schematic representation of *P. kerathurus* measurements recorded: carapace length (CL) and total length (TL).

3.2.3 Histology

After removal, ovaries were preserved in Dietrich solution (900 ml of distilled water, 450 ml of 95% ethanol, 150 ml of 40% formaldehyde and 30 ml of acetic acid) for histological analyses. Histological analyses were carried out on 31 female samples to evaluate gonadal development throughout the reproductive season and to validate the macroscopic stage attribution.

All samples, conserved in the fixative for at least 20 days, were embedded in paraplast, cut in transverse serial sections (7 μm) and mounted on slides. An

increasing alcohol concentration protocol was adopted for dehydration. Slides were then stained with Harrys' hematoxylin and eosin (Pearse 1985).

Also male are dissected; deferent are gently removed and their content immediately examined under optical microscope (*Wild M20*; 40x) to check presence-absence of spermatozoids.

3.2.4 Lipid Profile

Sample Treatment - From February to September 2015, ovary of five specimens per month were collected in order to define lipid composition. Ovaries were homogenized and lyophilized (lyophilized by sublimation to -80° C in very high vacuum) until constant weight. Lipids were extracted through microwave extraction with petroleum ether and acetone in a ratio of 2:1 (v/v; Ramalhosa et al. 2012). Methyl esters of fatty acids (FAMES) were obtained from total lipid extraction adding 1% of sodium methylate in methanol (Carreau & Dubacq 1978).

FAMES Analysis – Methyl ester of fatty acids analysis were performed with gas chromatograph Agilent-6890 equipped with an Agilent-5973N quadrupole mass selective detector. Helium (He) has been used as carrier gas, at inlet temperature of 250° C; the injection volume was 1 µl; split ratio 1/50. The temperature program adopted was: 100° C (1 min.) to 150° C at 25° C/min., to 200° C at 5° C/min., to 230° C at 1° C/min.; transfer line 250° C. The mass

selective detector (MSD) parameters were: ion source at 230° C, quad MS at 150° C and SIM mode with 2 min. of solvent delay. Main ions fragment were recorded and for fatty acids identification a database of mass spectrum of reference has been used (NIST, National Institute of Standards and Technology, Gaithersburg, MD 2002).

3.3 Results

3.3.1 Sex-ratio

From August 2013 to September 2015 a total of 1482 *P. kerathurus* specimens were collected, 790 of these were female (53%) and 692 were male (47%; Fig. 17). The monthly analysis of sex-ratio shows an equilibrium condition with an average value of 1.05 (± 0.65) males/females, and a minimum value of 0.34 in February 2015 and a maximum value of 2.85 in July 2014 (Fig. 17).

3.3.2 Female LFD-Monthly Frequency of Spermatophores

On a total of 789 females examined, 285 were found to be inseminated (36.12%). Their size ranged between 29 and 58 mm CL with a mean (\pm sd) of 44.31 ± 5.28 while not inseminated showed a wider size range, with dominant smaller size classes with a mean of 37.90 ± 7.28 mm CL, and ranged between 21 and 60 mm CL (Fig. 18). Significant differences for medians and size frequency distribution between this two groups have been observed (Mann-Whitney test:

U=33569, $p < 0.001$; two-sample Kolmogorov-Smirnov test: $D=0.46007$, $p < 0.001$).

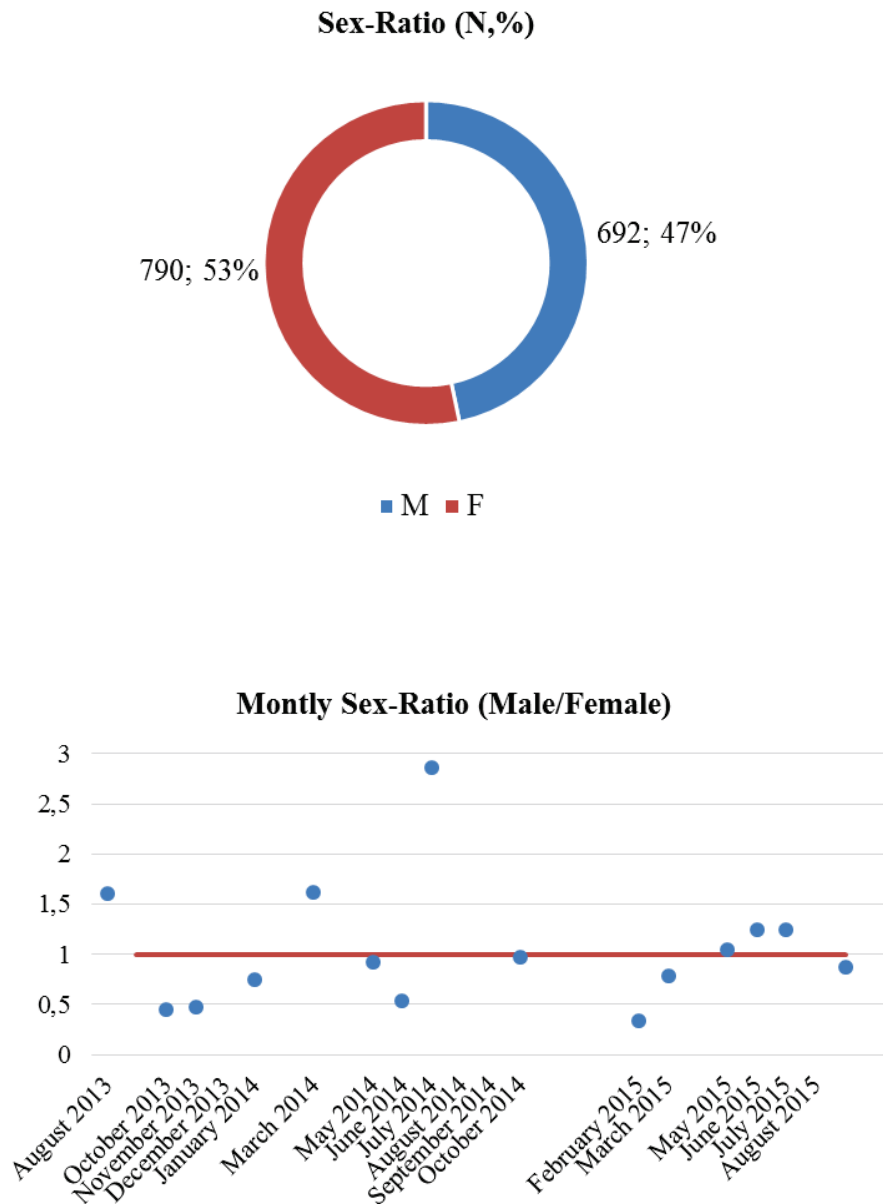


Fig. 17. Total and monthly sex-ratio of *P. kerathurus* (males/females) population, from August 2013 to September 2015.

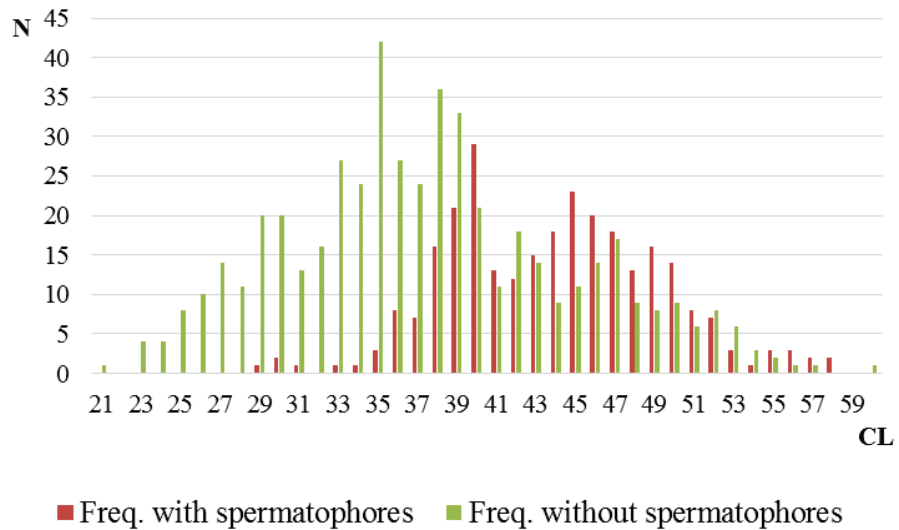


Fig. 18. Length-frequency distribution of *P. keratburus* females with and without spermatophores.

The monthly frequency of inseminated females shows highest percentage value during summer seasons, in particular June, July and August. These months exhibited values ranged from 87% in June 2014 to 100% in June and July 2015, while showed lower and variable values in October (44.8% in 2013 and 34.8% in 2014) and in May (62.5% in 2014 and 26.1% in 2015). Values under 26% of inseminated females were recorded in the remaining months (Fig. 19).

Values of August are not significant due to low number of specimens available because trawling ban is in place in that month.

3.3.2 Size at First Sexual Maturity

Size at first maturity based on the presence of spermatophores in female (CL_{50}^{Female}) and based on well-developed sperms in male (CL_{50}^{Male}) were estimated,

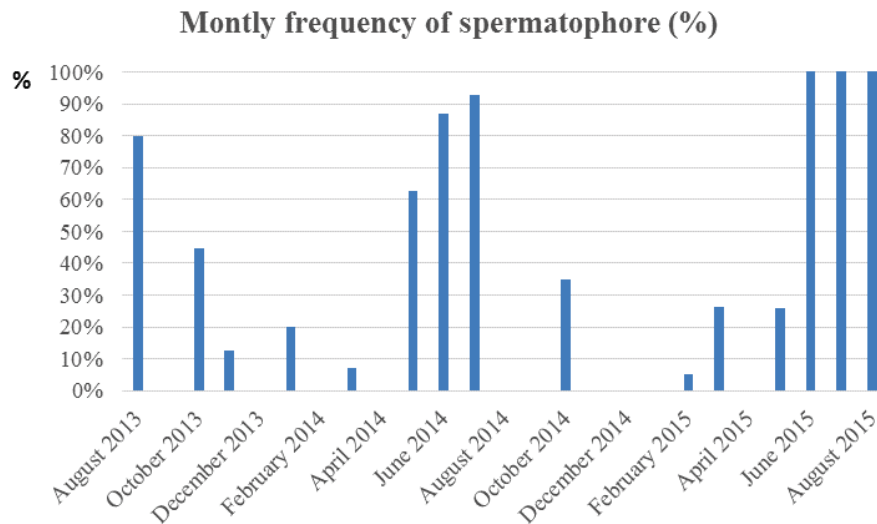


Fig. 19. Monthly frequency of spermatophores (%) in *P. kerathurus* females, from August 2013 to September 2015.

by logistic equation as described by King (1995), respectively in 40.3 mm CL for female (Fig. 21) and in 19.7 mm CL for male (Fig. 20).

3.3.4 Gonado Somatic Index (GSI)

Regarding variation of GSI for monthly-pooled data, a pronounced seasonality was exhibited. The minimum value was recorded on December (0.80 ± 0.34) and in general, under 1.6 in winter months; a very pronounced increased values started from May (2.93 ± 2.45) until July (11.24 ± 5.72), followed by a decrease until October (2.23 ± 2.70 ; Fig. 22).

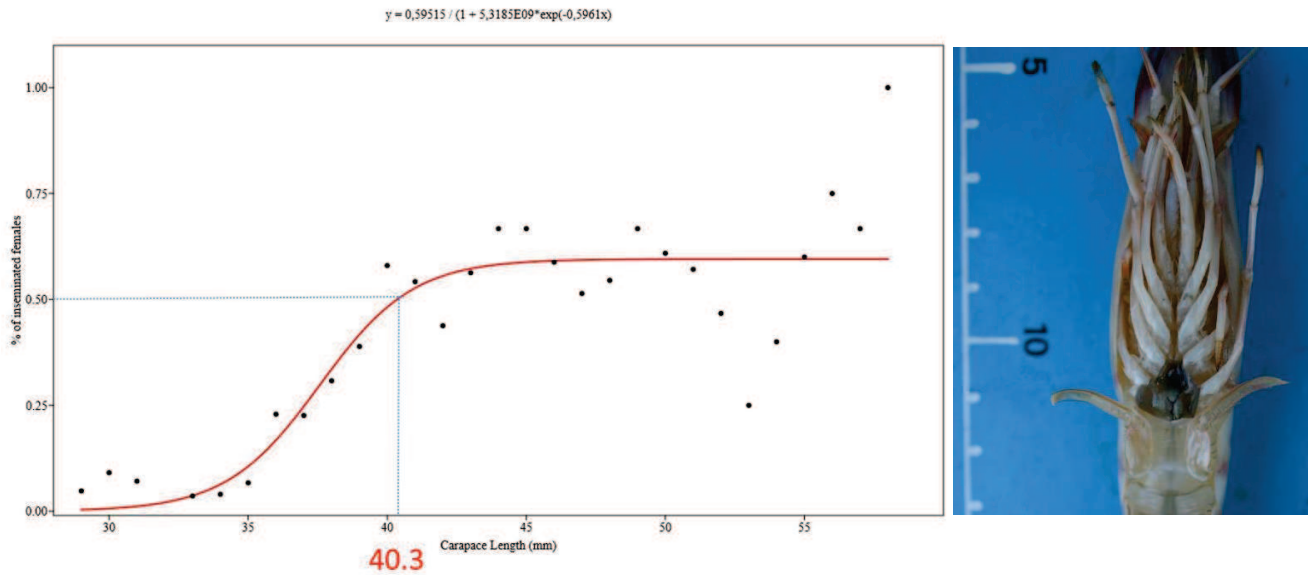


Fig. 21. Logistic curve for the estimation of length at first maturity (on the left) based on the occurrence of insemination in *P. kerathurus* females (on the right). Logistic equation: $y=0.59515/(1+5.3185E09*\exp(-0.5961x))$.

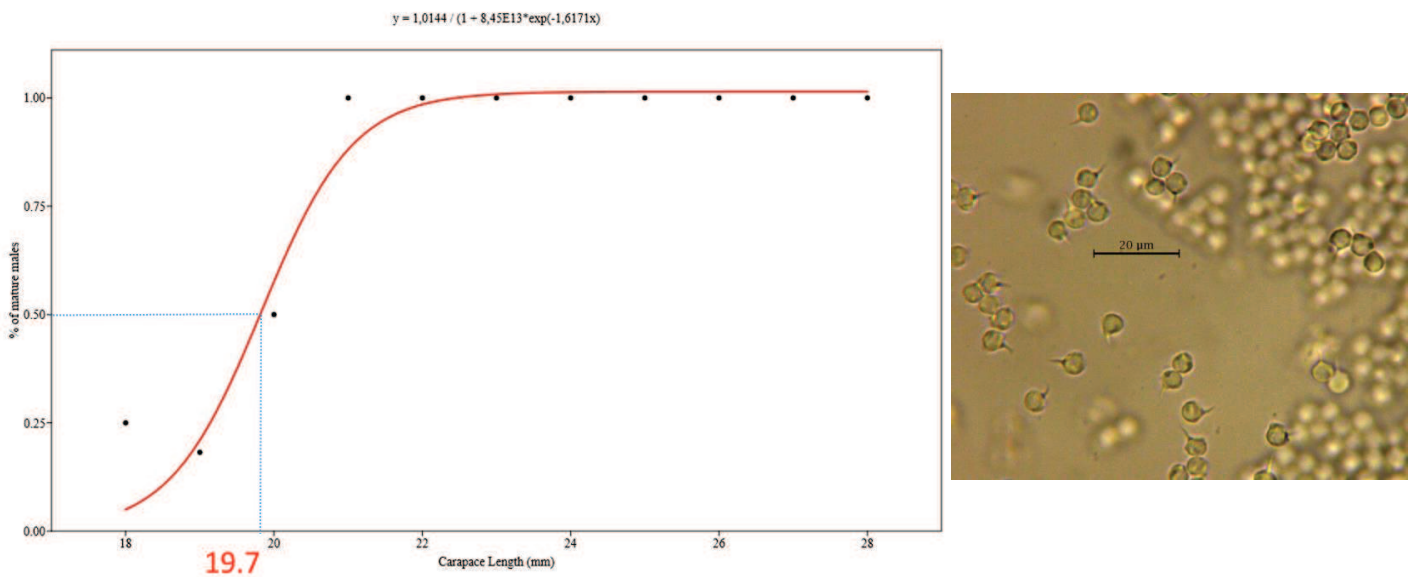


Fig. 20. Logistic curve for the estimation of length at first maturity (on the left) based on the occurrence of well-developed sperm in *P. kerathurus* males (on the right). Logistic equation: $y=1.0144/(1+8.45E13*\exp(-1.6171x))$.

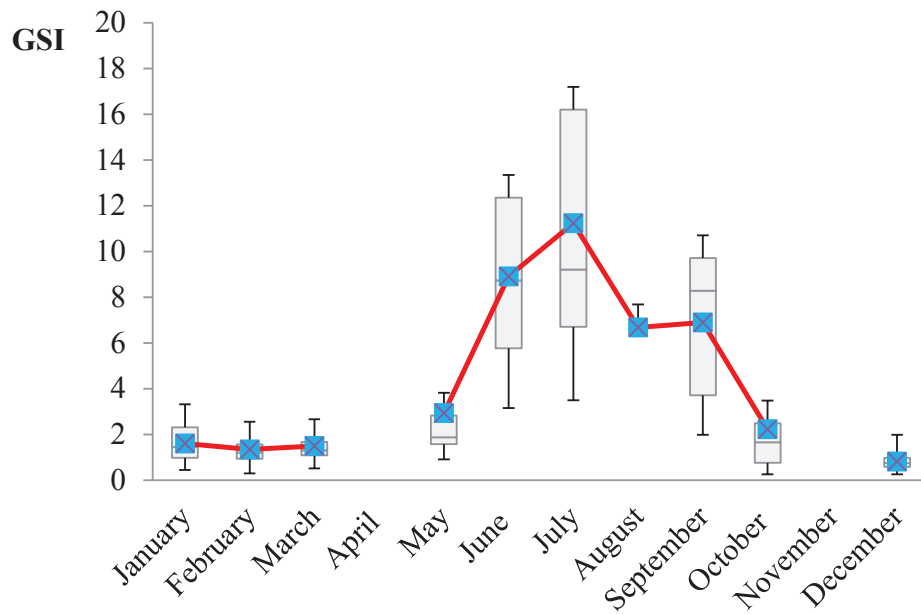


Fig. 22. Box and Whiskers plot of monthly variation of GSI of *P. kerathurus* females.

3.3.5 Histology

According to literature, five stages of ovarian development of *P. kerathurus* were identified, based on most advanced oocyte stage. Stages were:

STAGE I (PREVITELLOGENIC) – Macroscopically gonad are very small and not visible through the exoskeleton. The ovary contained mainly two categories of previtellogenic oocytes: the smaller oocytes (17-26 μm) at the center of the ovary and larger oocytes (26-65 μm) appear located more peripherally. Oogonia measured from 34 to 53 μm in diameter (Fig. 23, Fig. 24, Fig. 25).

STAGE II (EARLY VITELLOGENIC) – By naked eye, developing ovaries can be easily differentiated from other tissues. They are flaccid and white-ivory. Mainly represented by previtellogenic oocytes (80-100 μm in diameter) with presence of late vitellogenic oocytes. Cytoplasm increase in size and becomes

more basophilic. These oocytes, when are relatively small, are gradually surrounded by follicle cells. Central basophilic nucleus within cytoplasm with one or two nucleoli, that appear in perinucleus stage (Fig. 23, Fig. 24, Fig. 25).

STAGE III (LATE VITELLOGENIC) – Fresh ovaries are large and visible through the exoskeleton and yellow to pale-olive. Ovaries contain large oocytes (90-150 μm), yolky, acidophilic, whereas the nucleolus is basophilic (Fig. 23, Fig. 24, Fig. 25).

STAGE IV (MATURE) – Ripe stage is recognized by large and turgid ovaries visible through the exoskeleton, from yellow to green intense color. Mature oocytes exceeds 150-200 μm and a conspicuous cortical rods at the cell periphery represents a typically distinctive feature (Fig. 23, Fig. 24, Fig. 25).

STAGE V (SPENT) – Gonads were small, flaccid, and invisible through the exoskeleton. Ovaries are spent or had a resorbed appearance with presence of atretic (degenerating) oocytes, nucleus disappears and germinal vesicle breakdown; cortical rods elongate and extending radially inward (Fig. 23, Fig. 24, Fig. 25).



Fig. 23. Female gonad of *P. kerathurus* after (left) and before dissection (right).

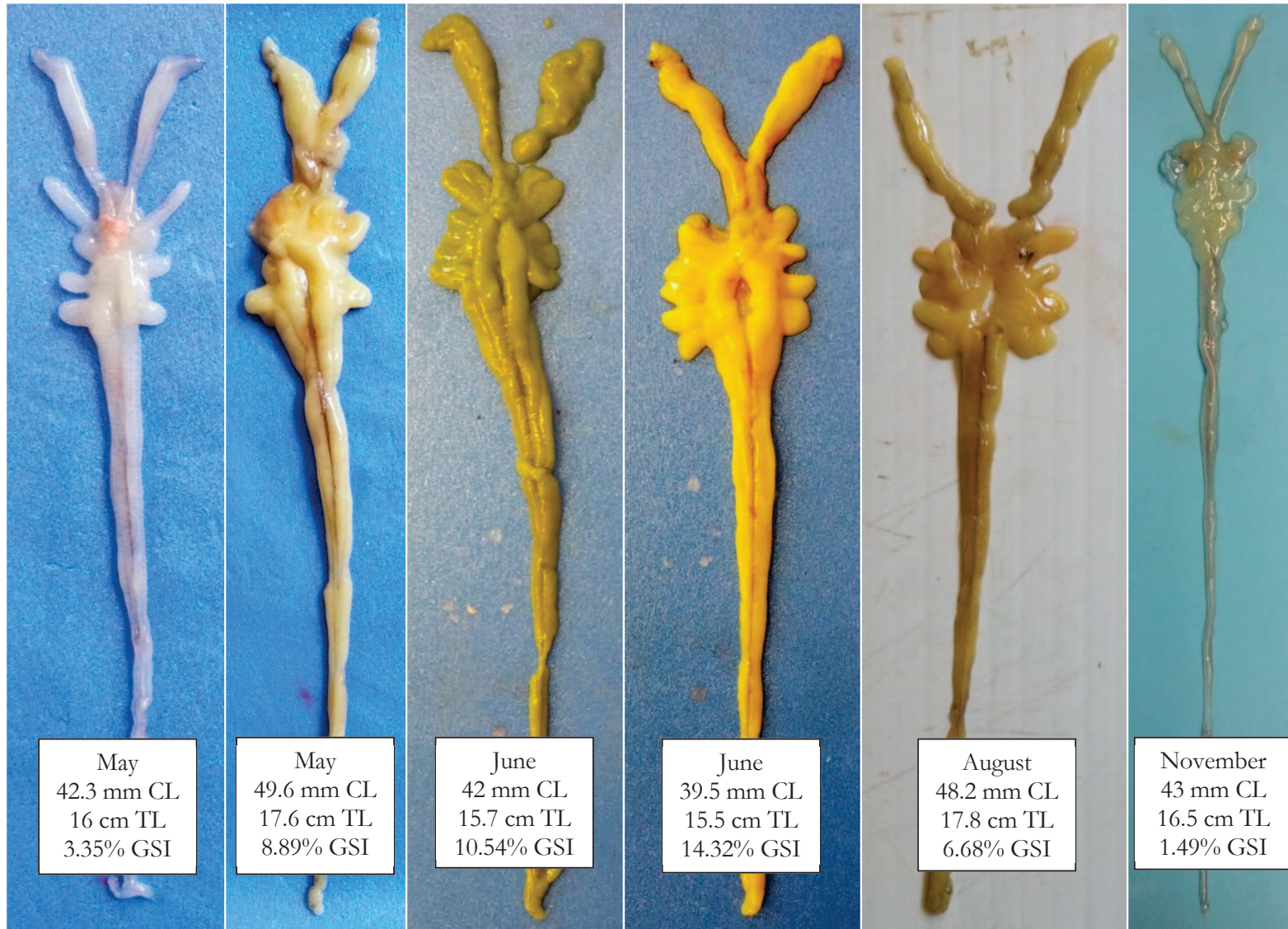


Fig. 24. Macroscopic temporal variation of female gonad of *P. kerathurus*, from May to November (CL= carapace length, TL= total length, GSI= gonado-somatic index).

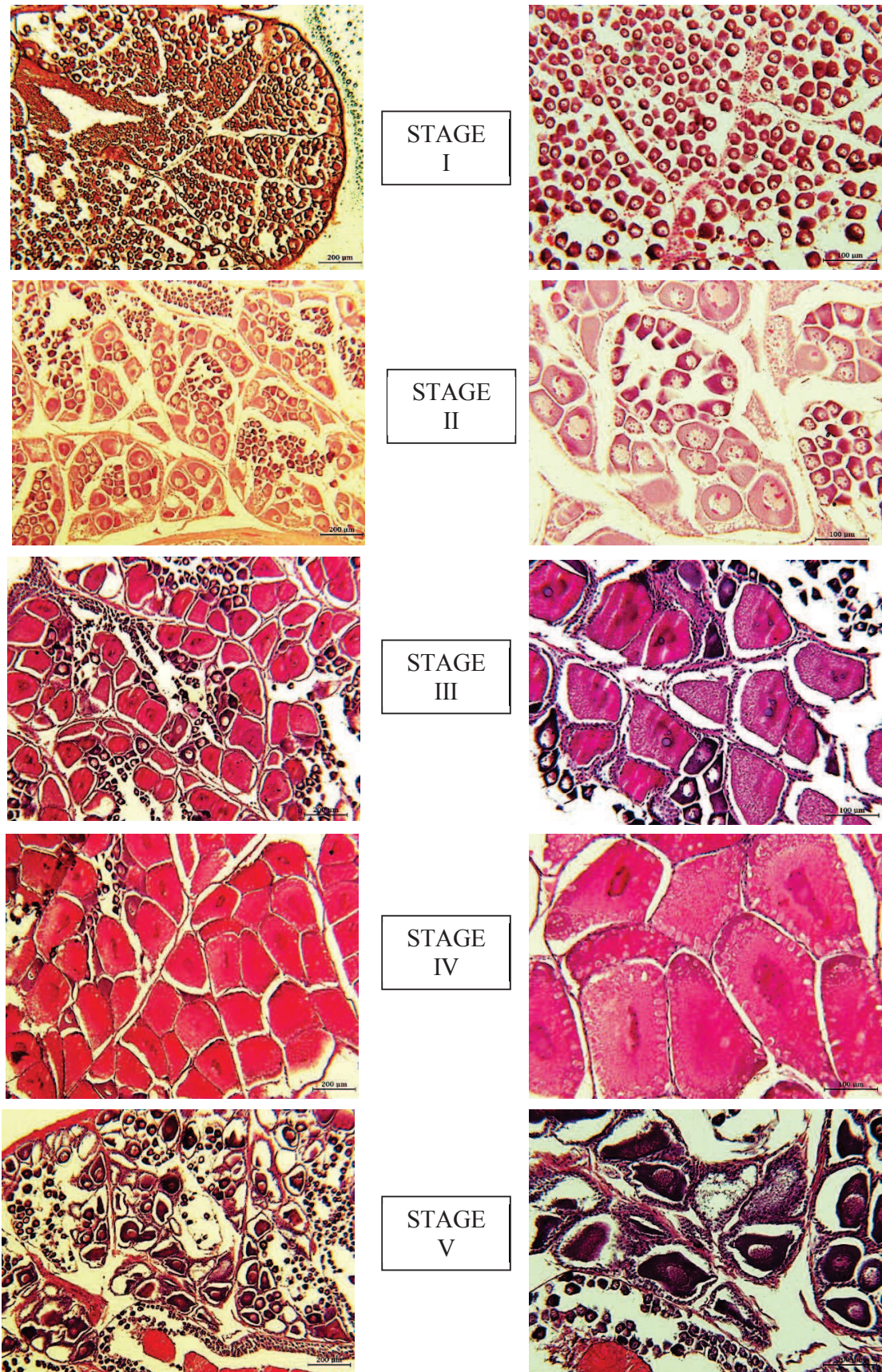


Fig. 25. Histological sections of five stages of female gonad maturation of *P. kerathurus* (scale bars: 200 µm on the left, 100 µm on the right).

3.3.6 Lipid Profile

From February to September 2015 (except for April when samples are not available) mean value of extract lipids was 16.01 ± 4.84 mg, corresponding to 7.1% of gonad sample dry weight. The minimum value was recorded in August (11.1 mg; 5.5%) and maximum in June (22.1; 8.8%).

During this study, polyunsaturated fatty acid (PUFA: ranged from 56.01% in February to 33.46% in September) showed higher values than saturated fatty acid (SFA: from 22.12% in March to 36.08% in July) and monounsaturated fatty acid (MUFA: from 20.26% in February to 30.86% in September), to exception for September, when SFA exceed PUFA value. In general, PUFA represented the main fraction in winter-spring season while it was comparable with SFA and MUFA in summer season. About essential fatty acids (EFAs), ratio between *n3/n6* ($\omega 3/\omega 6$) PUFAs shows values in favor of *n3*, and ranged from 2.28 in February to 5.81 in June. Statistical analysis revealed significance correlations between SFA and *n3/n6* PUFA ratio with GSI of sub-sample (respectively $p=0.019$ and $p=0.024$), as well as GSI of sub-sample of ovaries intended to lipid content showed high statistical significance with GSI of total females sample collected ($p=0.0013$; Tab. X; Fig. 26).

Tab. VIII. Total lipid contents in ovaries of *P. kerathurus* from February to September 2015 expressed as mg and percentage, mean \pm standard deviation.

Lipid	February	March	May	June	July	August	September	Mean	\pm sd
mg	13.8	12.5	14.0	22.1	15.0	11.1	23.6	16.0	4.8489
%	6.87	7.87	6.27	8.83	6.95	5.50	7.50	7.1	1.0854

Tab. IX. Percentage of saturated fatty acid (SFA), monounsaturated fatty acids (MUFA), polyunsaturated fatty acid (PUFA), and *n3/n6* ratio in ovarian of *P. kerathurus*, from February to September 2015.

	February	March	May	June	July	August	September
SFA	23,74	22,12	22,47	28,16	36,08	34,23	35,68
MUFA	20,26	23,38	20,44	23,3	25,06	26,79	30,86
PUFA	56,01	54,5	57,09	48,54	38,86	38,98	33,46
n6/n3	0,44	0,34	0,41	0,17	0,21	0,18	0,37
n3/n6	2,28	2,98	2,46	5,81	4,75	5,45	2,69

Tab. X. Correlation and linear regression parameters between GSI of sub-sample and saturated fatty acid (SFA), monounsaturated fatty acid (MUFA), polyunsaturated fatty acid (PUFA), *n3/n6* ratio and GSI of total sample (*= $p < 0.05$; **= $p < 0.01$).

Variable	Slope (a)	Intercept (b)	r	R ²	p	
SFA	1.1556	22.358	0.83393	0.69543	0.019699	*
MUFA	0.37389	22.174	0.45953	0.21117	0.29957	
PUFA	-1.5299	55.472	-0.72649	0.52779	0.064429	
n6/n3	-0.02122	0.42345	-0.85263	0.72699	0.014766	*
n3/n6	0.27002	2.2397	0.81713	0.6677	0.024824	*
GSI _{Tot}	1.1287	-0.68743	0.9444	0.89189	0.001356	**

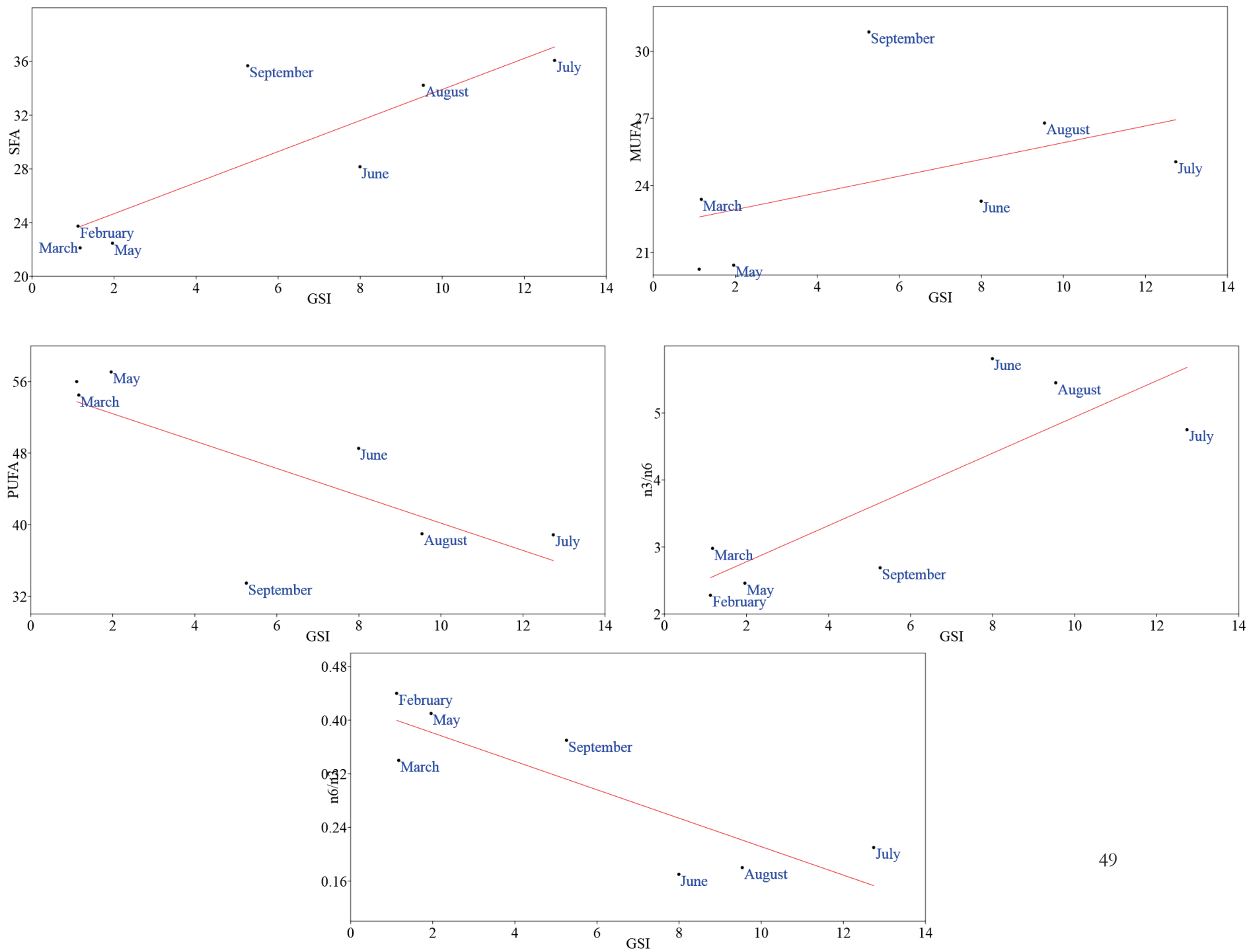


Fig. 26. Linear regression of GSI of sub-sample and saturated fatty acid (SFA), monounsaturated fatty acid (MUFA), polyunsaturated fatty acid (PUFA), $n3/n6$, $n6/n3$ and GSI of total sample.

Lipid composition reflect gonad maturation during reproductive season. In more detail, some SFAs as the *myristic acid* (C12), *palmitic acid* (C16), *heneicosylic acid* (C21) and MUFAs as *cis-vaccenic acid* (C18:1 n7), *erucic acid* (C22:1 n9), and PUFAs as *eicosapentaenoic acid* (C20:5 n3; EPA), *linoleic acid* (C18:2 n6) and *arachidonic acid* (C20:4 n6; AA), have shown a statistical significant correlation with GSI of sub-sample (Tab. XI; Tab. XII; Tab. XIII). The multivariate linear regression revealed a positive relation between *myristic, palmitic, heneicosylic, erucic acids* and GSI and negative relation between *cis-vaccenic, eicosapentaenoic (EPA), linoleic, arachidonic (AA) acids* and the same GSI (Tab. XIII; Fig 27). High statistical significance correlation have been demonstrate for two SFAs as *margaric acid* (C17; $p=0.0096$) and *arachidic acid* (C20; $p=0.0097$); negative linear relation have shown the *margaric acid* and positive linear relation the *arachidic acid* (Tab. XIII; Fig 27). These last SFAs exhibited a strong fluctuation particularly evident in reproductive months, with minimum value in July (1.15 ± 0.05) and maximum in February (2.42 ± 0.06) the first, and minimum in March – May (both 0.11 ± 0.01) and maximum in August (0.4 ± 0.01) the second (Tab. XII).

Tab. XI. Classification of fatty acids extracted in the sub-sample; saturated fatty acid (SFA), monounsaturated fatty acid (MUFA), polyunsaturated fatty acid (PUFA), essential fatty acid (EFA), *n3* ($\omega 3$), *n6* ($\omega 6$).

	Carbon Chain	Common Name	
SFA	C12	Lauric acid	
	C14	Myristic acid	
	C15	Pentadecylic acid	
	C16	Palmitic acid	
	C17	Margaric acid	
	C18	Stearic acid	
	C20	Arachidic acid	
	C21	Heneicosylic acid	
	C22	Behenic acid	
MUFA	C14:1n5	Myristoleic acid	
	C16:1n7	Palmitoleic acid	
	C17:1n7	Heptadecanoic acid	
	C18:1n7	<i>cis</i> -vaccenic acid	
	C18:1n9	Oleic acid	
	C20:1n9	Eicosenoic acid	
	C22:1n9	Erucic acid	
PUFA	C16:2n7		
	EFA (<i>n3</i>)	C18:3n3	α -linolenic acid (ALA)
		C20:3n3	Eicosatrienoic acid (ETE)
		C20:5n3	Eicosapentaenoic acid (EPA)
		C22:6n3	Docosahexaenoic acid (DHA)
		C18:2n6	Linoleic acid
	EFA (<i>n6</i>)	C18:3n6	γ -linolenic acid (GLA)
		C20:2n6	Eicosadienoic acid
	C20:3n6	Dihomo- γ -linolenic acid (DGLA)	
	C20:4n6	Arachidonic acid (AA)	

Tab. XII. Fatty acids content in *P. kerathurus* ovaries from February to September 2015, expressed as means \pm standard deviation of dry weight.

	February	March	May	June	July	August	September
C12	0,04 \pm 0	0,32 \pm 0,01	0,04 \pm 0	0,63 \pm 0,02	0,64 \pm 0,54	1,21 \pm 0,06	0,8 \pm 0,09
C14	0,67 \pm 0,02	1,65 \pm 0,05	0,66 \pm 0,02	2,91 \pm 0,09	4,9 \pm 0,24	6,12 \pm 0,27	4,66 \pm 0,49
C14:1n5	0,02 \pm 0	0,03 \pm 0	0,02 \pm 0	0,11 \pm 0	0,28 \pm 0,2	0,44 \pm 0,02	0,32 \pm 0,03
C15	0,91 \pm 0,03	1,08 \pm 0,03	0,95 \pm 0,02	0,99 \pm 0,03	1,18 \pm 0,01	1,35 \pm 0,07	1,71 \pm 0,17
C16	12,12 \pm 0,2	12,02 \pm 0,61	11,13 \pm 0,18	16,2 \pm 0,35	21,04 \pm 0,76	16,22 \pm 0,66	20,33 \pm 2,31
C16:1n7	5,25 \pm 0,17	7,86 \pm 0,36	5,23 \pm 0,09	8,18 \pm 0,22	12,31 \pm 0,45	9,8 \pm 0,46	12,71 \pm 1,26
C16:2n7	0,12 \pm 0,01	0,15 \pm 0	0,14 \pm 0	0,13 \pm 0	0,16 \pm 0,01	0,14 \pm 0,01	0,15 \pm 0,01
C17	2,42 \pm 0,06	1,83 \pm 0,03	2,37 \pm 0,04	1,17 \pm 0,02	1,15 \pm 0,05	1,39 \pm 0,08	1,66 \pm 0,14
C17:1n7	1,8 \pm 0,11	1,79 \pm 0,04	1,85 \pm 0,03	1,12 \pm 0,01	1,3 \pm 0,06	1,41 \pm 0,07	2,39 \pm 0,22
C18	7,17 \pm 0,13	4,94 \pm 0,07	6,55 \pm 0,11	5,76 \pm 0,03	6,55 \pm 0,25	7,26 \pm 0,26	5,95 \pm 0,41
C18:1n9	6,48 \pm 0,11	6,86 \pm 0,18	6,41 \pm 0,11	7,67 \pm 0,05	9,46 \pm 0,32	9,66 \pm 0,48	10,22 \pm 0,79
C18:1n7	4,65 \pm 0,07	5,17 \pm 0,12	4,81 \pm 0,08	4,37 \pm 0,03	0,05 \pm 0	3,85 \pm 0,21	3,65 \pm 0,27
C18:2n6	1,33 \pm 0,04	1,41 \pm 0	1,24 \pm 0,06	1,23 \pm 0,02	0,74 \pm 0,03	0,84 \pm 0,06	0,9 \pm 0,07
C18:3n6	0,03 \pm 0	0,08 \pm 0	0,04 \pm 0	0,13 \pm 0	0,1 \pm 0	0,1 \pm 0	0,09 \pm 0,01
C18:3n3	0,17 \pm 0	0,27 \pm 0,01	0,18 \pm 0,04	0,57 \pm 0,01	0,3 \pm 0,02	0,36 \pm 0,02	0,3 \pm 0,03
C20	0,13 \pm 0	0,11 \pm 0,01	0,11 \pm 0,01	0,32 \pm 0,02	0,33 \pm 0,01	0,4 \pm 0,01	0,32 \pm 0,03
C20:1n9	1,95 \pm 0,04	1,58 \pm 0,06	2,03 \pm 0,12	1,72 \pm 0,06	1,5 \pm 0,03	1,43 \pm 0,05	1,4 \pm 0,08
C20:2n6	1,19 \pm 0,04	2,42 \pm 0,07	1,2 \pm 0,36	1,48 \pm 0,03	1,32 \pm 0,03	1,22 \pm 0,05	1,33 \pm 0,09
C20:3n6	0,25 \pm 0,02	0,3 \pm 0,01	0,25 \pm 0,04	0,34 \pm 0,01	0,24 \pm 0	0,23 \pm 0,01	0,21 \pm 0,03
C21	0,04 \pm 0	0,04 \pm 0	0,02 \pm 0	0,05 \pm 0	0,07 \pm 0	0,07 \pm 0	0,06 \pm 0,01
C20:4n6	14,26 \pm 0,15	9,42 \pm 0,61	13,73 \pm 0,2	3,93 \pm 0,04	4,33 \pm 0,14	3,63 \pm 2,67	6,49 \pm 0,39
C20:3n3	0,17 \pm 0,02	0,17 \pm 0,01	0,16 \pm 0,01	0,34 \pm 0,01	0,23 \pm 0,01	0,23 \pm 0,01	0,17 \pm 0,01
C20:5n3	24,22 \pm 0,42	24,3 \pm 0,12	23,83 \pm 0,18	18,2 \pm 0,07	13,87 \pm 2,48	17,71 \pm 0,6	13,5 \pm 0,68
C22	0,11 \pm 0,02	0,04 \pm 0	0,1 \pm 0	0,1 \pm 0,01	0,15 \pm 0,01	0,14 \pm 0,02	0,12 \pm 0,03
C22:1n9	0,07 \pm 0,01	0,04 \pm 0,01	0,06 \pm 0	0,11 \pm 0,02	0,13 \pm 0,01	0,1 \pm 0,01	0,13 \pm 0,03
C22:6n3	14,27 \pm 0,32	15,98 \pm 1,31	16,32 \pm 0,42	22,18 \pm 0,75	17,58 \pm 0,33	14,51 \pm 0,37	10,32 \pm 7,59

Tab. XIII. Detailed correlation and linear regression parameters between GSI of sub-sample and recorded saturated fatty acids (SFAs), monounsaturated fatty acids (MUFAs), polyunsaturated fatty acids (PUFAs), *n3/n6* ratio and GSI of total sample (*= $p < 0.05$; **= $p < 0.01$).

	FAs	Slope (a)	Intercept (b)	r	R ²	p	
SFA	C12	0.068063	0.13891	0.73227	0.53622	0.061289	
	C14	0.39899	0.81394	0.83204	0.69229	0.020242	*
	C15	0.020207	1.0523	0.32615	0.10637	0.47528	
	C16	0.7094	11.548	0.80338	0.64542	0.029524	*
	C17	-0.10104	2.2871	-0.87634	0.76797	0.009653	**
	C18	0.044748	6.0571	0.24829	0.06165	0.59135	
	C20	0.023763	0.11067	0.87579	0.767	0.009758	**
	C21	0.003326	0.031099	0.8321	0.6924	0.020223	*
	C22	0.00578	0.075726	0.73763	0.54409	0.058454	
MUFA	C14:1n5	0.027868	0.01591	0.74416	0.55378	0.055092	
	C16:1n7	0.47325	6.0733	0.71262	0.50783	0.072316	
	C17:1n7	-0.05588	1.9833	-0.59911	0.35894	0.15515	
	C18:1n7	-0.30914	5.5497	-0.81457	0.66352	0.025666	*
	C18:1n9	0.26351	6.611	0.73756	0.54399	0.058489	
	C20:1n9	-0.0328	1.8449	-0.59755	0.35706	0.15652	
	C22:1n9	0.006218	0.056091	0.80414	0.64664	0.029252	*
PUFA	C16:2n7	0.001277	0.13417	0.43361	0.18802	0.33108	
	C18:3n3	0.016219	0.21497	0.55134	0.53267	0.19953	
	EFA (n3) C20:3n3	0.008665	0.16076	0.61319	0.376	0.14313	
	C20:5n3	-0.84974	24.205	-0.81363	0.66199	0.025977	*
	C22:6n3	0.23082	14.568	0.29212	0.085334	0.52497	
	C18:2n6	-0.04893	1.3766	-0.8421	0.70913	0.017444	*
	C18:3n6	0.005644	0.049356	0.72984	0.53267	0.062599	
	EFA (n6) C20:2n6	-0.0315	1.6304	-0.32775	0.10742	0.47299	
	C20:3n6	-0.00118	0.26673	-0.12091	0.014618	0.79624	
C20:4n6	-0.86109	12.864	-0.86023	0.74	0.012991	*	

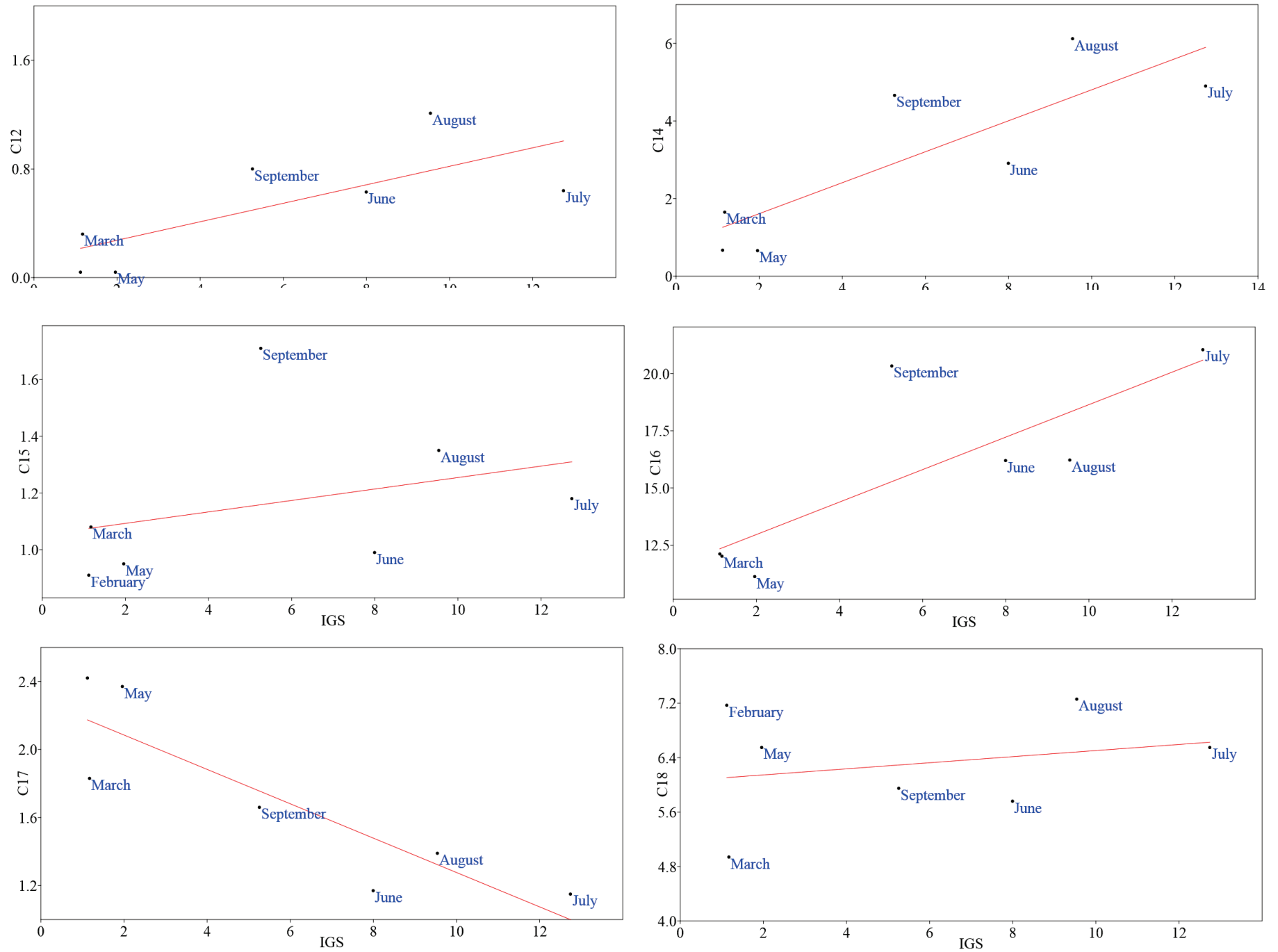


Fig. 27. Multivariate linear regressions between recorded fatty acids (FAs) and gonado somatic index (GSI) in the ovaries of *P. kerathurus* female from February to September.

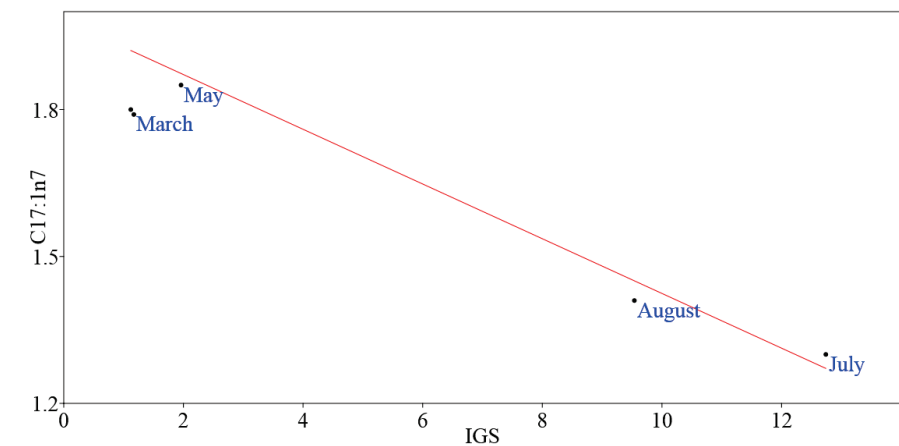
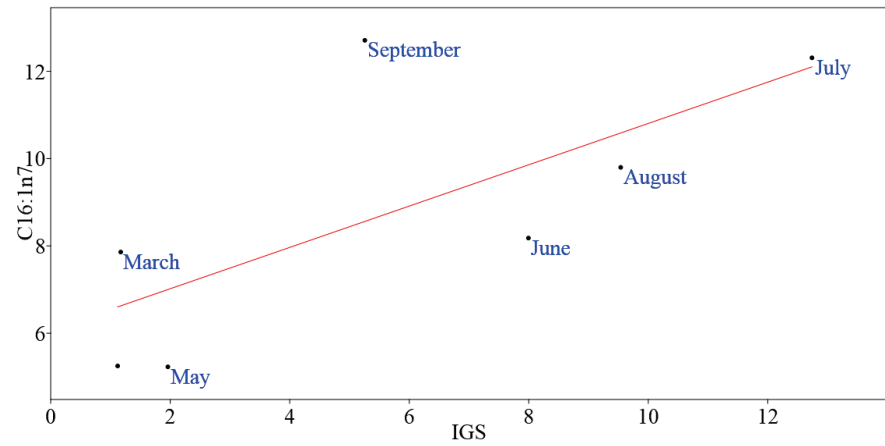
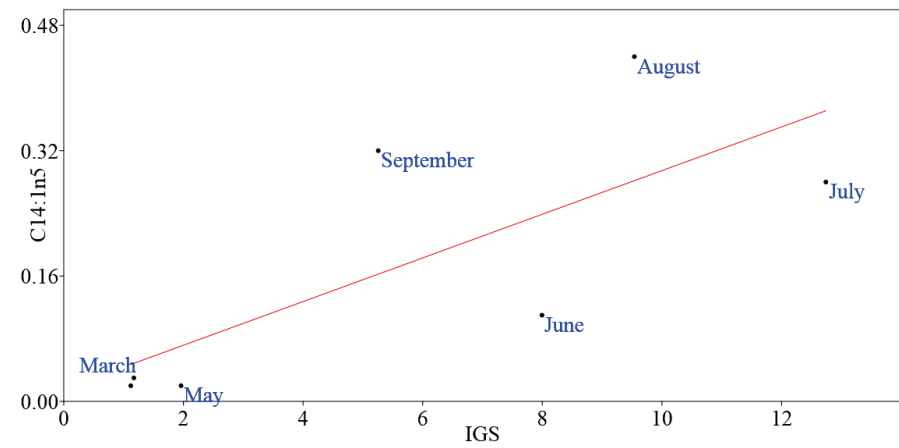
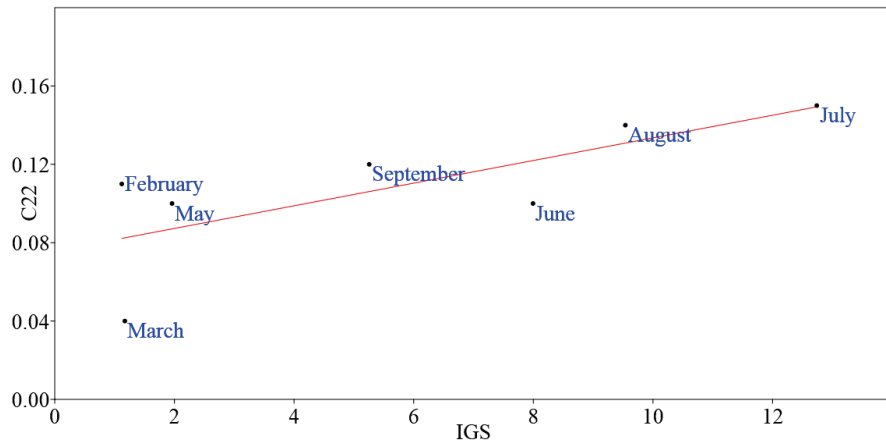
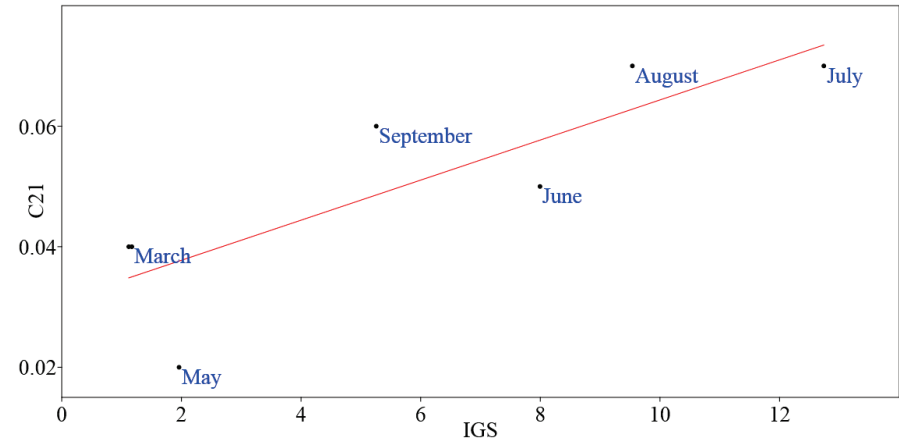
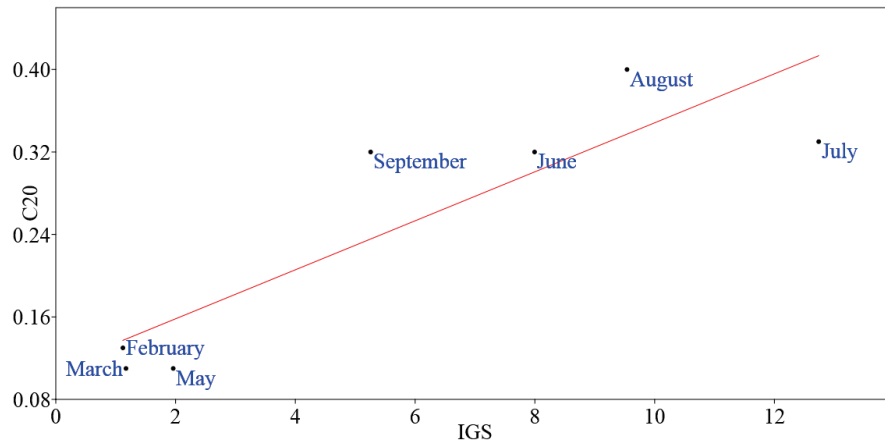


Fig. 28 following. Multivariate linear regressions between recorded fatty acids (FAs) and gonado somatic index (GSI) in the ovaries of *P. kerathurus* female from February to September.

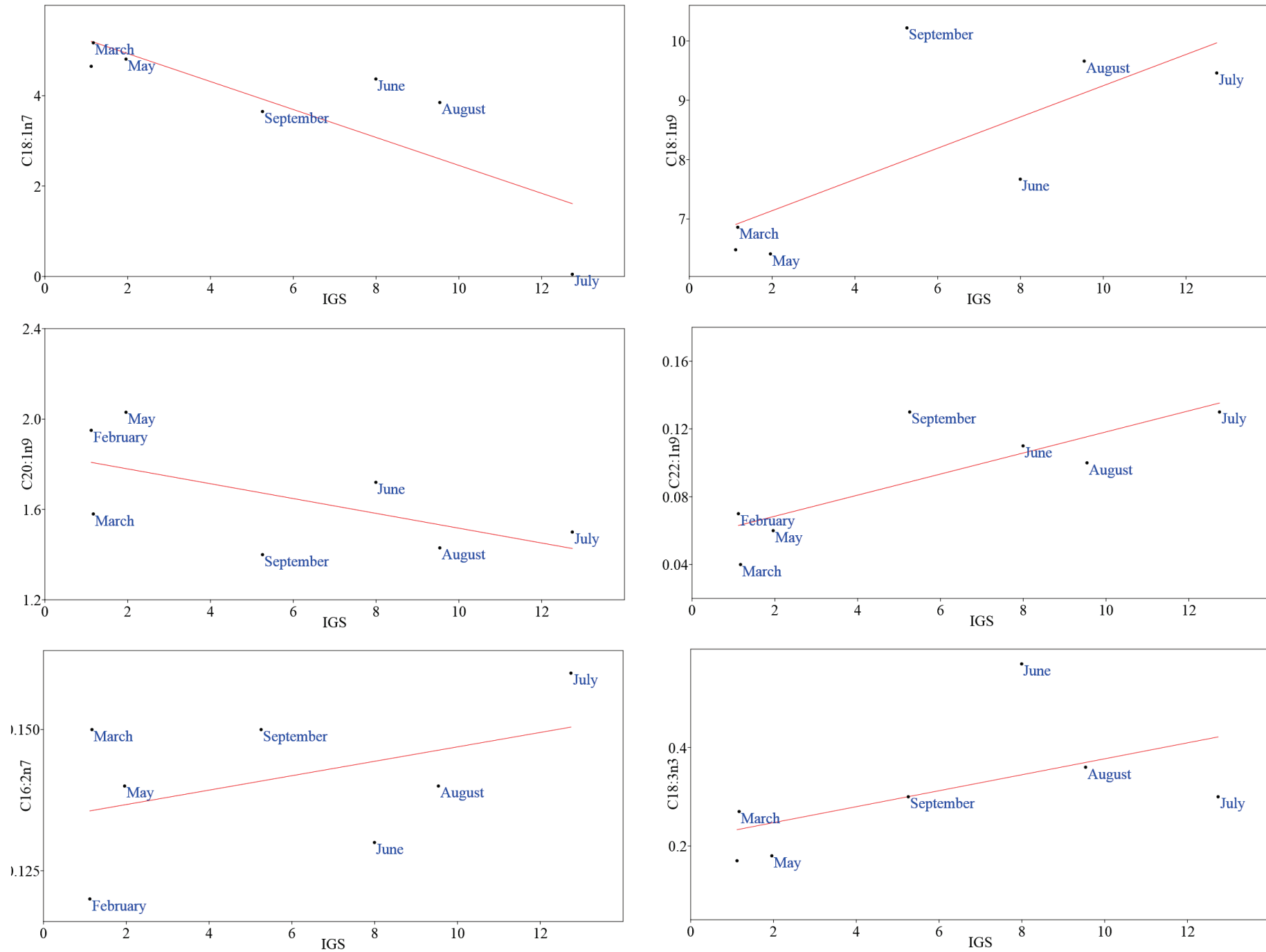


Fig. 29 following. Multivariate linear regressions between recorded fatty acids (FAs) and gonado somatic index (GSI) in the ovaries of *P. kerathurus* female from February to September.

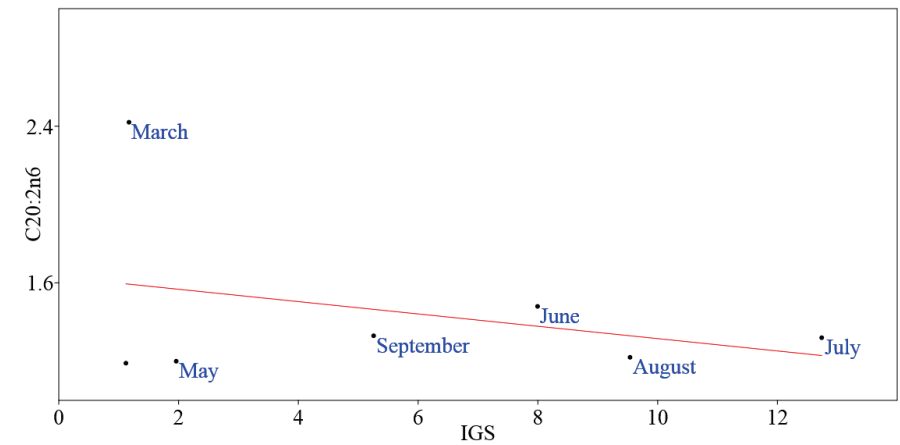
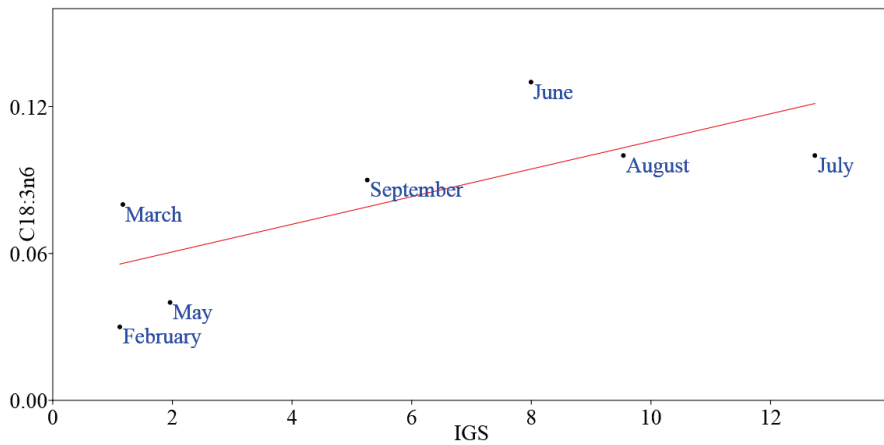
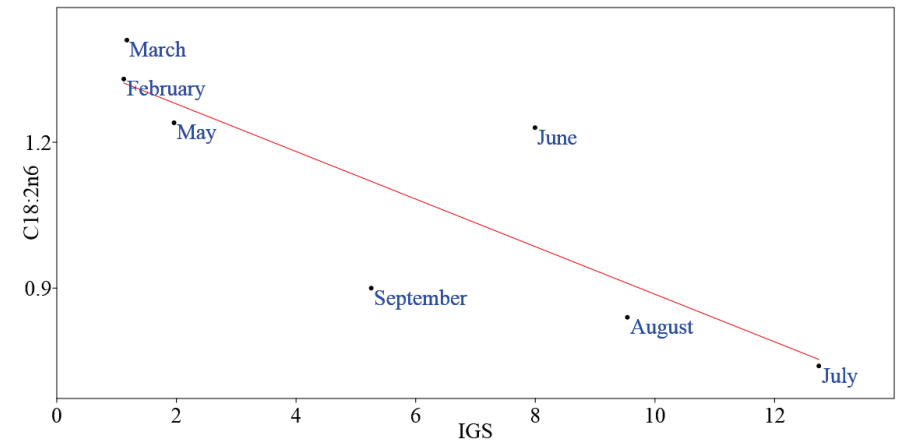
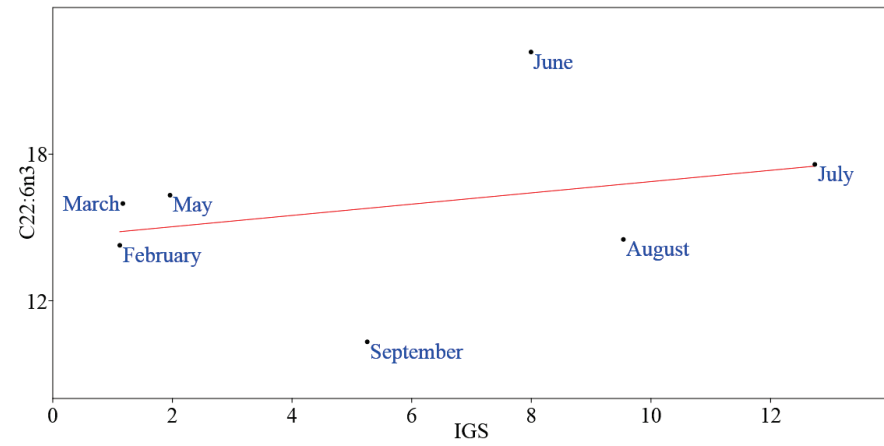
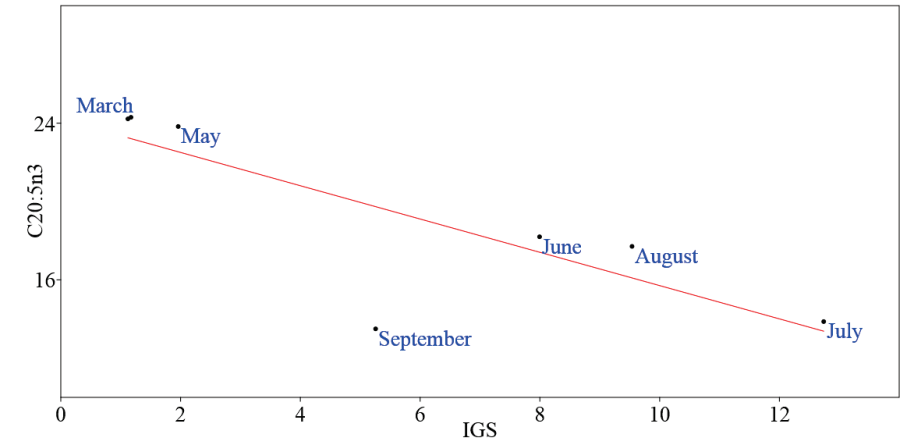
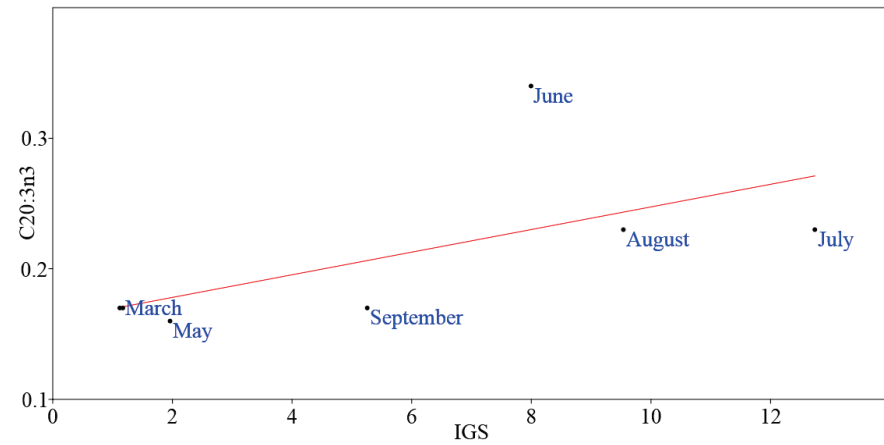


Fig. 30 following. Multivariate linear regressions between recorded fatty acids (FAs) and gonado somatic index (GSI) in the ovaries of *P. kerathurus* female from February to September.

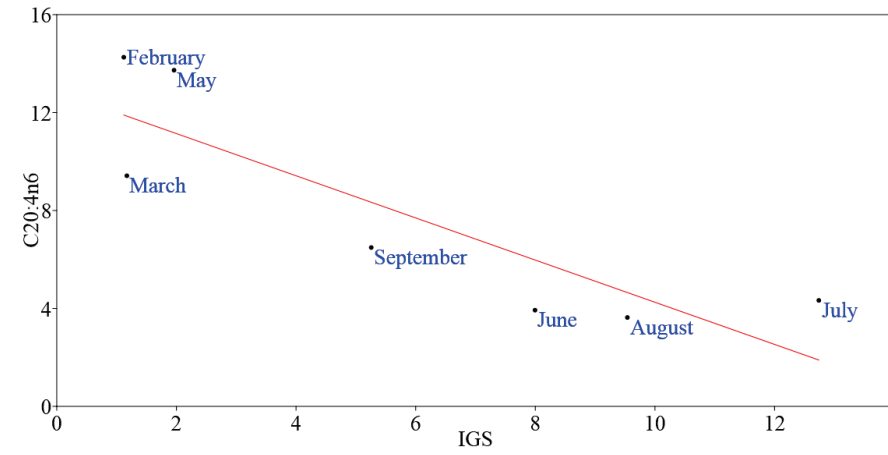
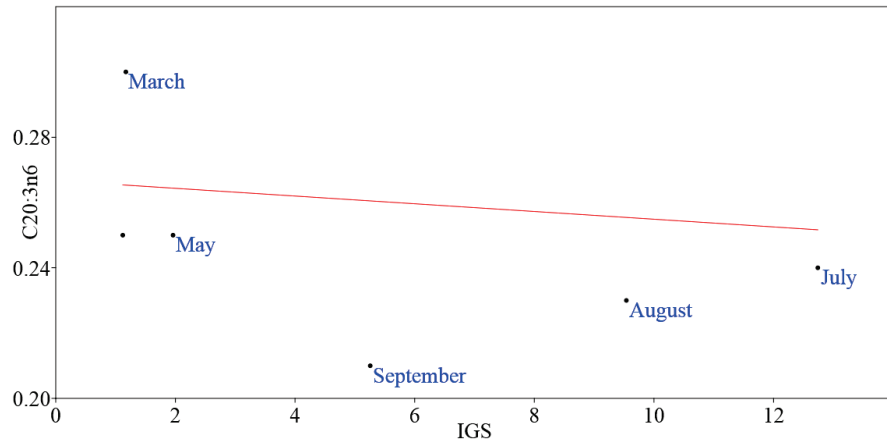


Fig. 31 following. Multivariate linear regressions between recorded fatty acids (FAs) and gonado somatic index (GSI) in the ovaries of *P. kerathurus* female from February to September.

3.4 Discussion

Within the present study valid information on reproductive biology of *P. kerathurus* were obtained. In more detail, a variable sex-ratio was observed during the study and also size at first sexual maturity, estimated in 40.3 mm of CL for female and 19.7 mm CL for male. While the female values were comparable with those reported in literature, the male values were well below to whatever so far reported. In more detail, 40.7 mm of CL for female and 39.2 mm of CL for male were the size at first sexual maturity estimated for *P. kerathurus* by Kevrekidis & Thessalou-Legaki (2012) in Thermaikos Gulf (N. Aegean Sea), instead, were estimated in 45.5 mm of CL for female and 31.4 mm of CL for male by Lumare et al. (2011) for the same species in South-Eastern coast of Italy. In addition, presence of spermatophore adhering to *telycum* in female have been strongly connected with seasonality and size of the specimen, indicating summer season as the best period for insemination, but, unlike what has been reported in Thermaikos Gulf by Kevrekidis & Thessalou-Legaki (2012), in the present study the percentage of inseminated female that exceed 50% were recorded only in summer months. The GSI ranged from 0.8 (December) to 11.2 (July), indicated July as a peak of the total reproductive season (summer, from May to September), as confirmed by histological analysis of ovarian maturation.

This approach was supported by the study of lipid content variation in ovaries of *P. kerathurus* during the annual cycle. This information was reported for the first time in the present study, in species of the Mediterranean Sea. A statistically significant correlation of some fatty acids with GSI of the same sample have been identified.

This work highlighted the ovary maturation processes under different point of view. Important physiological changes occur during reproductive season aimed to sustain the reproductive success.

Ovarian development was accompanied with changes in color (due to differences in carotenoids content) and size (Adiyodi & Subramonian 1983; Arculeo et al. 1995; Cavalli et al. 1997), due to the substances reserve storage needed for the future offspring. This carotenoids play an important role during embryogenesis, as suggested by Goodwin (1951), Dall et al. (1995), and Liñán-Cabello et al. (2002).

A strong changing in ovaries color was observed, accompanied with a deep coloring increase in reproductive months, strongly related to gonado somatic index.

During gametogenesis, in addition to the gonadal color changing, an increase in total lipid content corresponding to 8.8% was observed in June, one month before to the gonado somatic peak (July), equivalent to stage IV - mature oocyte.

Similar results on increasing in total ovarian lipid content have been reported by Mourente & Rodriguez (1991) for *P. kerathurus* and by Teshima & Kanazawa (1983) for *P. japonicus*.

In this work, in more detail, at this stage there was an increase of saturated fatty acids (SFAs) and mono unsaturated fatty acids (MUFAs) and a decrease of poly unsaturated fatty acids (PUFAs). Two saturated fatty acids (SFAs) such as *margaric acid* (C17) and *arachidic acid* (C20, also called *eicosanoic acid*) have shown trends high statistically significant correlated with GSI, highlighting negative linear relation the first and positive linear relation the second.

It is well known that successful of gonadal maturation is dependent on the diet (Brown, A. et al. 1979; Lawrence et al. 1980; Cahu & Quazuguel 1989), and in particular lipid deposition in the ovary is crucial for various shrimp species (Gehring 1974; Teshima & Kanazawa 1983; Galois 1984; Teshima, Kanazawa, Horinouchi, et al. 1988; Jeckel et al. 1989; S. Teshima et al. 1989; Bray et al. 1990). Also abundant and continuous feeding activities during maturation of *P. kerathurus* play a very important role in supplying lipids during the process of formation and maturation of ovaries (Sargent 1976). In consequence, this species could have a higher dietary lipid requirement during maturation. As illustrated by Bray et al. (1990) for *P. styliorostris*, the lipid component in the ovary increases during development, and the midgut gland has a limited storage capability.

The great importance of the quality of dietary lipid source is related to the inability to *de novo* phospholipid synthesis as cholesterol by the juveniles, thus the relatively lipid richness of ovaries and eggs compared to the adult shrimp (Teshima 1982; Teshima et al. 1986).

An increase in the lipid concentration in the ovaries during maturation and a concurrent decrease in the hepatopancreatic lipids has been noted by Teshima & Kanazawa (1983) for wild *P. japonicas* and for *P. kerathurus* by Mourente & Rodriguez (1991). Phospholipids were largely responsible for the increases in ovarian lipids in *P. kerathurus*, which conforms with data reported for developing ovaries of *P. monodon* (Millamena & Pascual 1990). The principal lipid components of haemolymph and tissues of crustaceans are phospholipids, except in the midgut gland where it is well established neutral lipids (particularly triacylglycerol) represent the bulk of total lipids (Chapelle 1977).

In *P. kerathurus* the majority of neutral lipids ingested by female are converted to polar lipids in the midgut gland and transported to the developing ovaries for use as constituent and reserve lipids during sexual maturation (Mourente & Rodriguez 1991).

In this species approximately 65% of fatty acids of the total ovarian lipids are conveyed to eggs during spawning; the fatty acids stored as *triacylglycerol* (TAG) in the midgut gland could play an important reserve role in the case of prolonged starvation before or during maturation or moulting (Dall 1981).

Middleditch et al. (1980) stated that increase in the concentration of PUFAs in the TAG fraction of both ovary and midgut gland would seem to reinforce the theory that long-chain fatty acids are necessary for vitellogenesis of penaeids.

4. IDENTIFICATION OF NURSERY AREAS OF *P. KERATHURUS* IN NORTHERN AND CENTRAL ADRIATIC SEA

4.1 Introduction

To better identify the nursery areas, it is important to know the reproductive characteristics of the species. As previously reported, gonadal maturation takes place during spring/summer (Lumare et al. 1971). The highest percentage of mature females (about 85%) were found in June and July. In this period all females have spermatophores adhering to their *thelycum*, as reported by Lumare et al. (2011) in south Adriatic. Hatching begins in around 16-30 hours (Heldt 1938) and, larval succession entails three different phases: nauplius, zoea and mysis.

This species performs the typical larval succession of decapod penaeidae crustaceans. The zoea actively feeds on phytoplankton and it is strongly attracted by light and brackish water environments (estuaries and lagoons), where it can find extremely favorable trophic and thermal conditions, above all during spring-summer. Once it reaches the last larval stage of mysis, it starts feeding on zooplankton. Benthic post-larvae go on developing in brackish waters and coastal environments until they reach a juvenile stage. At the end of summer season juveniles leave coastal areas, and settle on infralittoral sandy

bottoms, where waters are less affected by surface temperature variations (Scovacicchi 1994).

In the Adriatic basin, the main Italian lagoons are located in the northern side, between Grado and the Po Delta (Grado Lagoon, Marano Lagoon, Lagoon of Caorle, Venetian Lagoon and the Po Delta) and in the southern side, in the Gargano promontory (Lagoon of Lesina and Varano Lagoon). Sandy beaches are the usual landscape of this side of the Adriatic Sea, except for the littorals of Gargano, Conero and other smaller promontories. As a typical problem of sandy beach is erosion, it is reduced by coastal defence structures, that dissipate the approaching wave energy, especially by breaking (Lorenzoni et al. 2012). In particular, the 170 km of the Marche Region coast is protected by more than 100 km of defences (Istituto di Idraulica 2001). To solve erosion phenomena (that started from the first half of the last century) mainly two types of coastal structures were built and used for coastal defence plans: the emerged and the submerged rubble-mound breakwaters (Lorenzoni et al. 1987; Lorenzoni et al. 2012).

The aim of the present study was to verify if and how breakwaters can play a role in *P. kerathurus* biological cycle in the Adriatic Sea, focusing on the effects of this artificial structures as new feasible nursery areas.

Moreover, the present study had the objective of verifying if these artificial lagoons could have any similarities with the natural ones in *P. kerathurus*

biological cycle, and the role of water temperature on it, especially as regards the juveniles migration in and out the nursery areas.

4.2 Materials and Methods

4.2.1 Study Area

The study was conducted along the area located both 5 kilometers northward to Ancona harbor and 5 kilometers southward to the Esino river, in the central Adriatic Sea (Fig. 32), characterized by flat sandy substrate, exposed to wind of northern quadrants.

In the last 60 years the area was urbanized and protected by emerged rubble-mound breakwaters to prevent erosion; at the small scale level, the main vocation of area is tourism and the recreational activity connected and petrochemical.

To verify the presence of *P. kerathurus* juveniles, a limited area has been chosen for sampling at sea, between shoreline and the opposite breakwater construction (Fig. 32), with the depth ranging between 0 and about 3m.



Fig. 32. Study area, central Adriatic Sea.

4.2.2 Control Site

In order to verify if results obtained in the study area were comparable with other areas, exclusively for 2015, additional sampling at sea were carried out in August, with the aim of compare the presence of juveniles of *P. kerathurus* in the study area with control site.

Sampling protocol was applied for sampling in the study area and in another area protected by emerged rubble-mound breakwater located 80 km southern to it (Grottammare: control, Fig. 33).

4.2.3 Natural Lagoon

It is known that, as previously reported, at the end of summer season juveniles of *P. kerathurus* leave coastal areas, and settle on infralittoral sandy bottoms, where waters are less affected by surface temperature variations (Scovacicchi 1994). In order to check for any similarities between samples of caramote prawn juveniles from the artificial lagoons and the natural ones, samples were taken in September 2015 from commercial fishing activity of *P. kerathurus* made at Marano Lagunare (lagoon, Fig. 33).

This fishing is practiced by traps ("bertovello", Fig. 34, a similar gear to fyke net). At the end of summer season, specimens leave lagunar areas for settle on infralittoral sandy bottoms, along this path run into traps placed for their capture.



Fig. 33. Location of additional samplings carried out in summer 2015 in Study area, in control site (Grottammare) and in a natural lagoon located at Marano Lagunare (lagoon).



Fig. 34. The “bertovello”, a like fyke net gear used for commercial fishing of *P. kerathurus* in Marano Lagunare.

4.2.4 Temperature Monitoring in the Study Area

Temperature is considered as one of the main driving factors for benthic composition and seems to play an important role in *P. kerathurus* migrations, mostly evident in juveniles phases. In order to outline the thermal characteristics waters of the study area, a Mini-Log *STAR ODDI*[®] *Data Storage Tag DST centi-TD temperature depth recorder* (Fig. 35A) was placed in the middle part of that sea stretch (Fig. 36). This instrument is capable to record

water temperature and depth (pressure) at 1 hour intervals. To prevent data loss due to theft/loss or depletion of the charge reserve, the instrument has been replaced at intervals and promptly replaced with an identical model. Water temperatures were recorded in the study area for about one year, from 10 May 2013 to 3 April 2014, with replacement of the *DST centi-TD temperature depth recorder* at day 19 June 2013, 24 September 2013 and 4 December 2014. Temperature data were also collected in a control site placed in open sea at 1.2 nautical miles offshore Senigallia harbor ($43^{\circ}44.21'N$, $13^{\circ}13.13'E$; Fig. 36) at the observation station *TeleSenigallia* pylon owned by the CNR-ISMAR of Ancona (Paschini & Penna 2015), in order to assess any possible influence of breakwaters on this physical parameter. This detection station is equipped with different types of multiparameter probes, including the *Campbell Scientific*[®], *Inc. CS450 submersible Pressure Temperature Transducer* (Fig. 35B), fixed -2 m underwater and *Campbell Scientific*[®], *Inc. CS109 Temperature Probe* (Fig. 35C), fixed above sea level (for more instruments specifications see Appendix). All the data collected have been automatically real time transmitted.

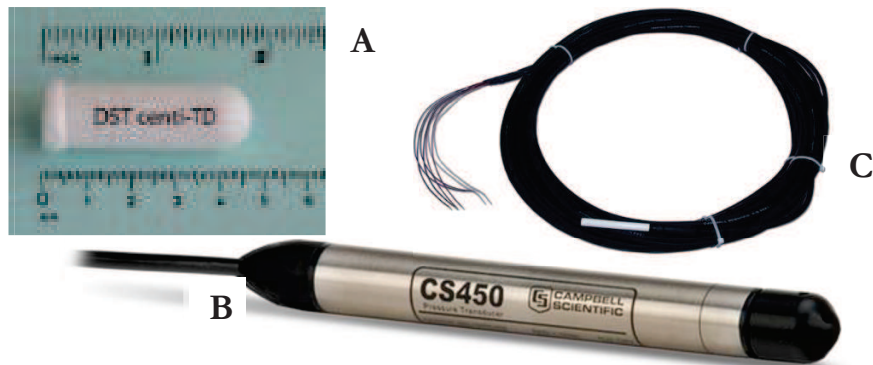


Fig. 35. Temperature recorder instruments: for water (STAR ODDI® Data Storage Tag DST centi-TD temperature depth recorder; 8A; Campbell Scientific©, Inc. CS450 submersible Pressure Temperature Transducer; 8B) and air (Campbell Scientific©, Inc. CS109 Temperature Probe; 8C).



Fig. 36. Location of temperature data recorder in offshore water (TeleSenigallia) and inshore study area (DST centi-TD).

4.2.5 Sampling of *P. kerathurus*

The monitoring of presence of *P. kerathurus* juveniles has been carried out on a monthly basis. From July 2013 to September 2014 a total of 14 samplings were carried out (including seven in 2013 and seven in 2014).

In August 2015 samplings were conducted simultaneously in the study area and in control site.

An experimental dredge was adopted, specifically designed for the purpose of this study. The gear consists of a metal frame 73 cm width and a net bag to collect the catch (stretched mesh size 2 mm, total length 300 cm). The frame was mounted on two sledge runners to prevent it from digging into the substratum.

A small chain was placed in front of the net to avoid losing contact with the bottom and to facilitate the entry of the organisms during the tow (Fig. 38, Fig. 39).

At each sampling the dredge was towed by means of a rope of known length (6 replicates). This enabled to perfectly assess the seabed surface investigated (Fig. 37). In this way, once ascertained the length of dredged, it was possible to standardize the sampled fauna as number for 100 m².

In order to limit "disturbance" generated during sampling operations, the gear has been lowered into the water with the aid of a small boat without mechanical propulsion. In addition, even to prevent the rope could alter the sampling approaching the bottom, it was decided to use a floating rope to tow the boat, pulled by hand from operators on beach (Fig. 37).



Fig. 37. Study area, site of sampling; each line represents one of the six replicates of dredged stretch towards breakwater – shoreline.

Given the ethological characteristics of *P. kerathurus*, the main species surveyed in this study, the samples were carried out only at night, when the animal shows a certain mobility for food purposes.

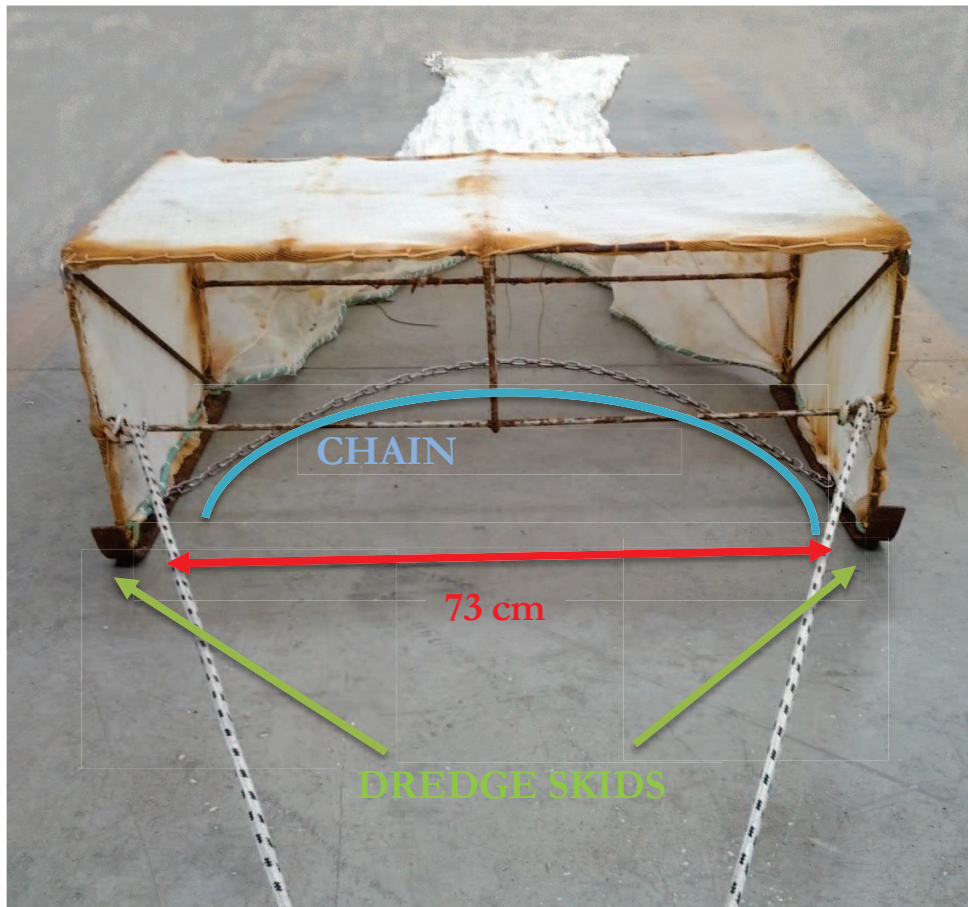


Fig. 38. Dredge adopted for sampling and some of its features (73 cm horizontal opening, in red; dredge skids, in green; chain, in lightblue).

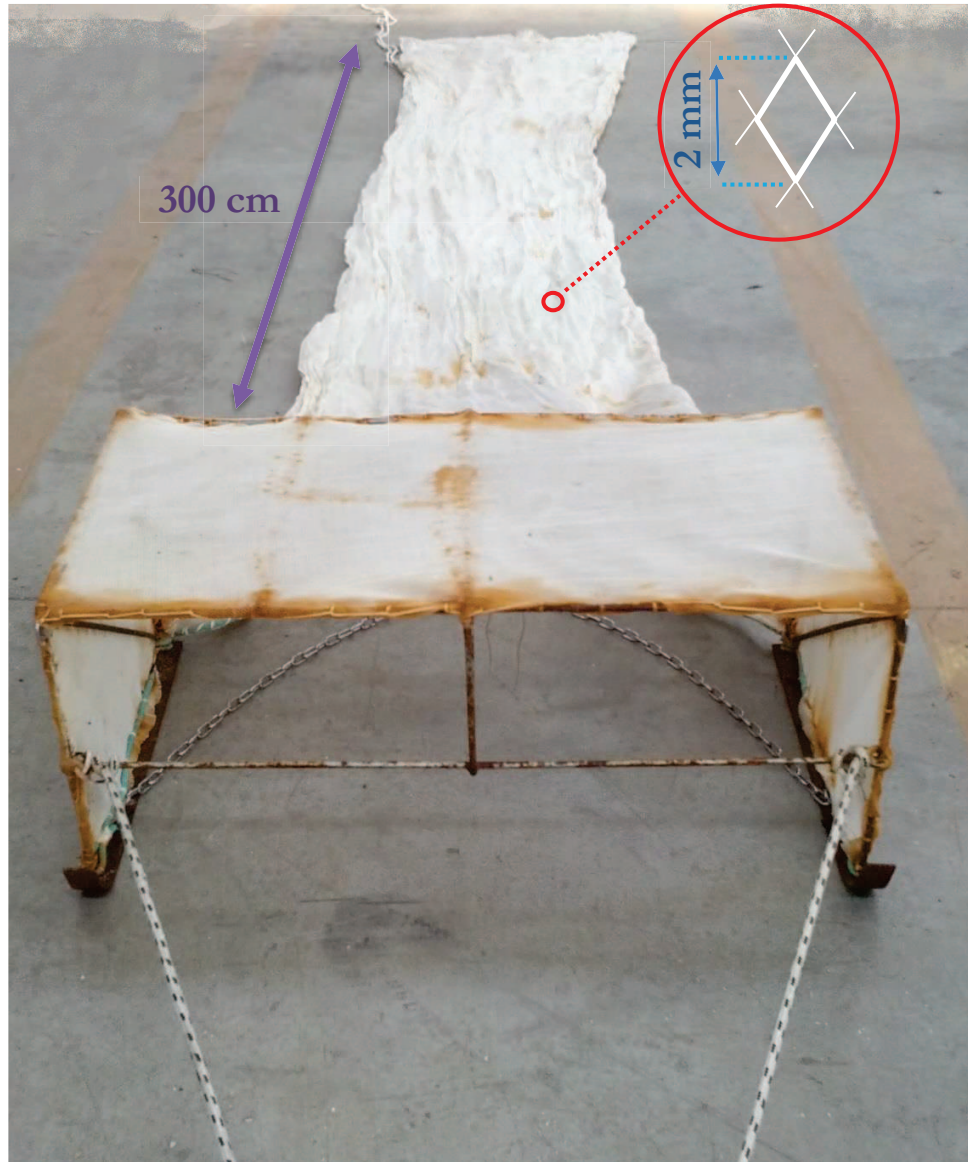


Fig. 39. Dredge adopted for sampling and other of its features (300 cm of total length of the net, in purple; 2 mm stretched mesh size, red circle).

4.2.6 Laboratory Analysis

The catch obtained at each replicates, all organisms retained were preserved in 5% buffered formalin, before being transported to the laboratory for subsequent identification to the lowest possible taxonomic level using standard nomenclature, and quantified. Benthic organism have been studied by means of a stereomicroscope *Leica MZ7.5* with a magnification range from 6.3x to 50x (Fig. 40).

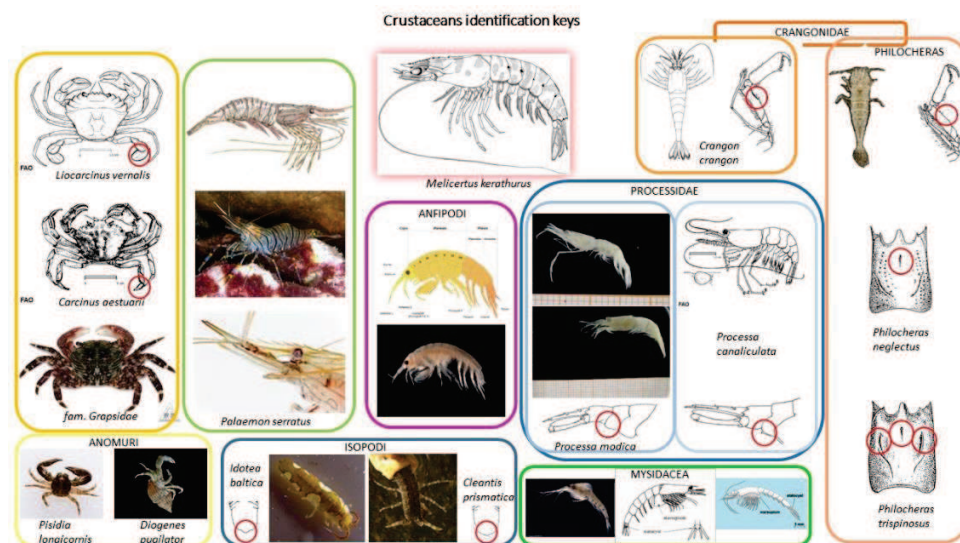


Fig. 40. Macroscopic table adopted for sorting operations.

In order to assess the sampler instrument functionality adopted, within 6 samplings (spring-autumn 2014), all anthropogenic materials ascribed as "marine litter" were recorded .

4.2.7 Data Analysis

Data from each replicate were analyzed according to degree width (73 cm) and length of towing. Then, the abundance and biomass of each species collected during samplings have been standardized as number per 100 m².

ComEcoPaC[®] Program (Community Ecology Parameter Calculator. Version 1; Drozd 2010) was then used to compute common parameters of community ecology samples. The parameters obtained were:

- Species richness, **S** (number of species)
- Abundance, **N** (number of specimens 100 m²)

Based on *Tischler's* scale:

- Number of eudominant species, **S_E**
- Number of dominant species, **S_D**
- Number of subdominant species, **S_{sd}**
- Number of recedent species, **S_R**
- Number of subrecedent species, **S_{sr}**
- Abundance for eudominant species, **N_E**
- Abundance for dominant species, **N_D**
- Abundance for subdominant species, **N_{sd}**
- Abundance for recedent species, **N_R**
- Abundance for subrecedent species, **N_{sr}**

Where dominance was calculated as a percentage of the individuals of a given species in the sample:

$$D_i = \frac{n_i}{N} \cdot 100\%$$

n_i = abundance of species i , N = total abundance in the sample

Tischler's scale for species dominance (Tischler 1949) refer to:

Eudominant, **E**: $10\% \leq D_i \leq 100\%$

Dominant, **D**: $5\% \leq D_i \leq 10\%$

Subdominant, **Sd**: $2\% \leq D_i \leq 5\%$

Recedent, **R**: $1\% \leq D_i \leq 2\%$

Subrecedent, **Sr**: $0\% \leq D_i \leq 1\%$

- *Shannon-Wiener* diversity index, **H'**

$$H' = \sum_{i=1}^s p_i \cdot \log_2 p_i$$

Where:

S = species richness (number of species)

$$p_i = \frac{n_i}{N}$$

p_i = proportion of species i ; n_i = abundance of species i

In addition, length-frequency distributions of *P. kerathurus* were recorded for all sites (CL; mm).

Based on the water temperature and abundance of *P. kerathurus* data availability (summer 2013), a Spearman test was used to detect linear correlation for abundance of *P. kerathurus* juveniles (ind. 100 m⁻²) and temperature (T° C).

Taking into account the total species density (n ind. 100 m⁻²) and total species richness (number of species) of samples from summer season (June, July, August) and all the other seasons, a one way ANOVA was used to detect significance differences for equal means, followed by *Kruskal-Wallis* test for equal medians and *Mann-Whitney* pairwise test.

Statistical differences in length-frequency distribution of samples from study area, control site and natural lagoon were tested by *Mann-Whitney* test for equal medians and *Kolmogorov-Smirnov* test for equal distributions.

4.3 Results

4.3.1 Study Area

4.3.1.1 Temperature Monitoring

During the observation period the air temperature showed the typical seasonal fluctuations, with an increasing trend in the first period (from May to August), followed by a lowering lasted until December, before rising again in the remaining period (December-April).

The highest value was recorded on July 2013 (32.6 ° C) and the minimum on December 2013 (2.5 ° C), with an average value for the period of 16.1 ° C (Tab. XIV, Fig. 41, Fig. 45).

Tab. XIV. Number of total records, minimum, maximum and mean temperature recorded for air and water (-2 m depth) at TeleSenigallia station, at study area (MiniLog station) and absolute maximum and minimum ΔT (°C) between *TeleSenigallia* and *MiniLog* stations, mean \pm standard deviation of mean (from the 10th of May 2013 to the 2nd of April 2014).

	N of records	T° C Min	T° C Max	T° C Mean	\pm Std. Dev. of Mean
Air	7865	2.5	32.6	16.1	6.48
<i>TeleSenigallia</i>	7865	7.6	28.6	17.6	6.32
<i>MiniLog</i>	7858	2.2	31.4	17.6	6.70
ΔT <i>TeleSenigallia-MiniLog</i>	7858	-8.4	6.9	-0.1	1.91

Water temperatures recorded at the *TeleSenigallia* station followed the trend of the air temperature above mentioned, although with much smaller fluctuations. The highest value was recorded on August 2013 (28.6 ° C) and the minimum on December 2013 (7.6 ° C), with an average value for the period of 17.6 ° C (Tab. XIV, Fig. 42, Fig. 45).

A similar temperature trend was also recorded in the study area within the sea protected by breakwaters, however peculiar characteristics have been showed in this area, such as temperature changes much more pronounced than the open sea (*TeleSenigallia*), also on a short time scale.

The highest value was registered on August 2013 (31.4 ° C) and the minimum on December 2013 (2.2 ° C), with an average value for the period of 17.6 ° C (Tab. XIV, Fig. 43, Fig. 45).

Analyzing the temperature differences (ΔT , °C) between open sea (*TeleSenigallia*) and study area protected by breakwaters, ΔT has hired tendentially negative values until the first half of June 2013, oscillating between positive and negative values for most of the summer. The following period, shows a temperature difference between the two areas examined, generally positive since the end of the summer and throughout the fall - early winter, around zero until the spring, and then assumed negative values the remaining period.

The major differences in temperature were recorded on days 17 and 19 May, both $-8.4\text{ }^{\circ}\text{C}$ and $+6.9\text{ }^{\circ}\text{C}$ on 17 December, with a mean value of entire period at $0.1\text{ }^{\circ}\text{C}$ (Tab. XIV, Fig. 44, Fig. 45). The ΔT values show how the air temperature influences the breakwaters area more than the open sea one.

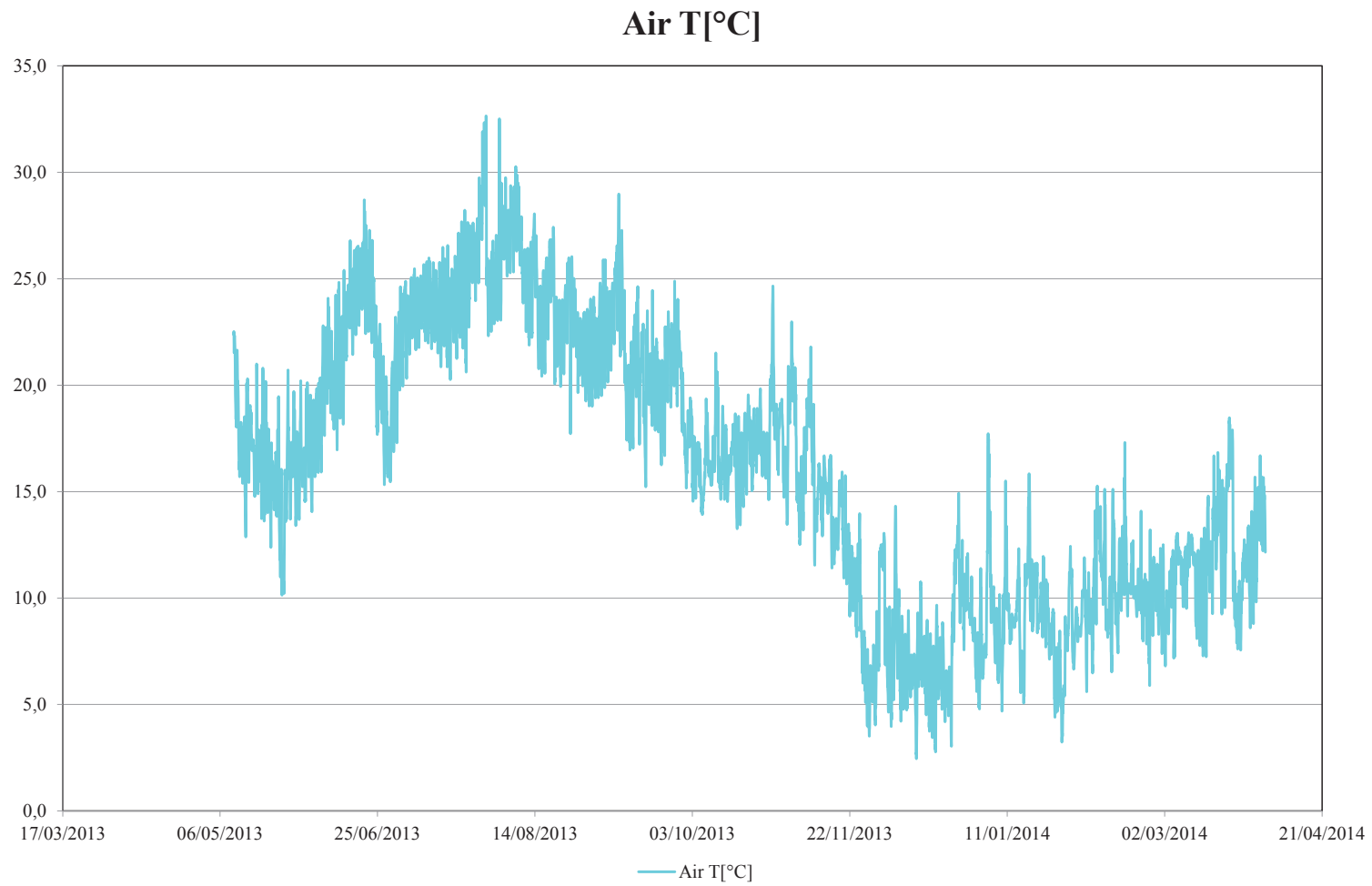


Fig. 41. Air temperature recorded from the 10th of May 2013 to the 2nd of April 2014 in *TeleSenigallia* station.

TeleSenigallia T [°C] (-2m depth)



Fig. 42. Water temperature recorded from the 10th of May 2013 to the 2nd of April 2014 in *TeleSenigallia* station at - 2 m depth.

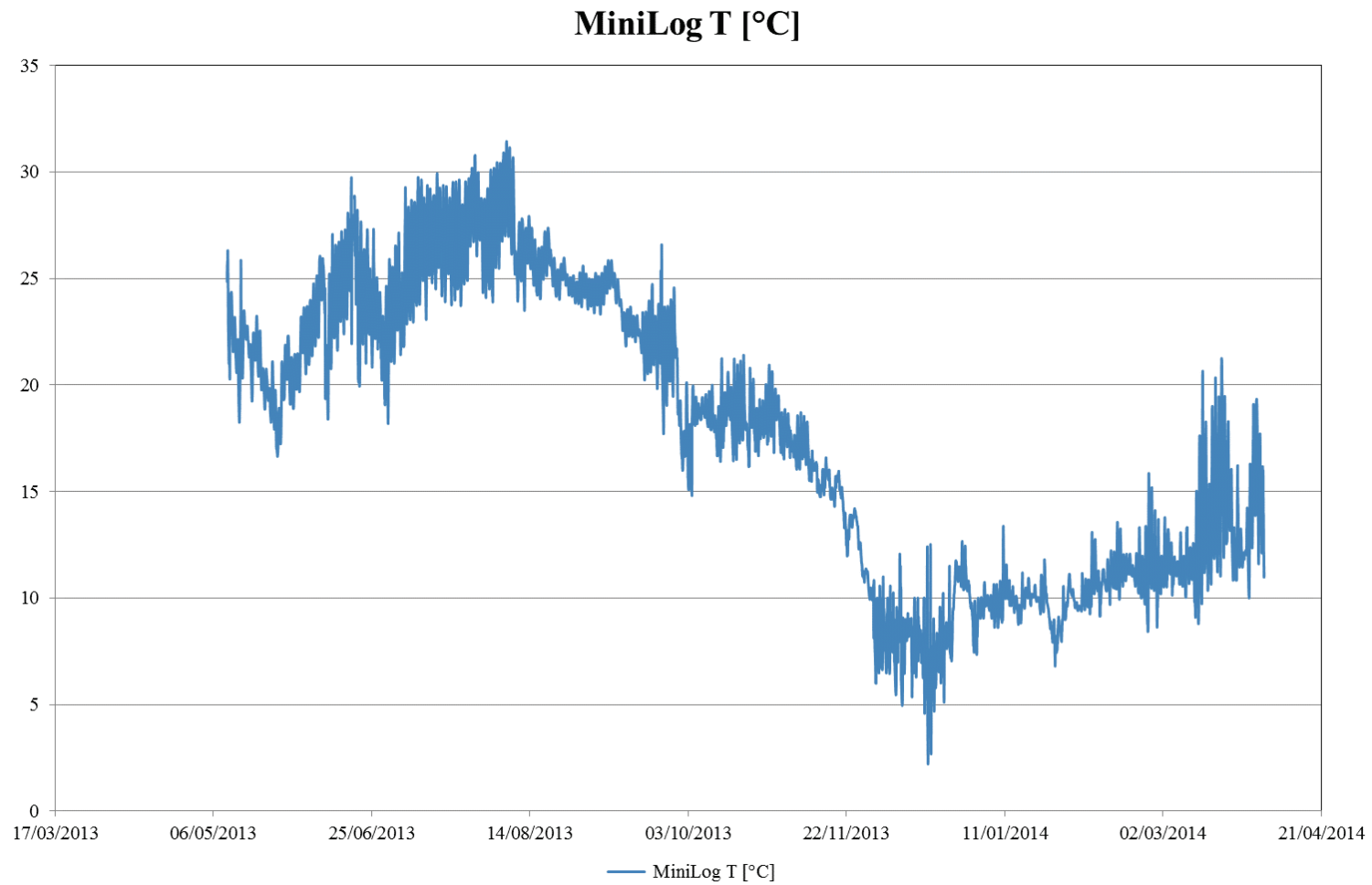


Fig. 43. Water temperature recorded from the 10th of May 2013 to the 2nd of April 2014 in the study area station.

ΔT TeleSenigallia - MiniLog [°C]

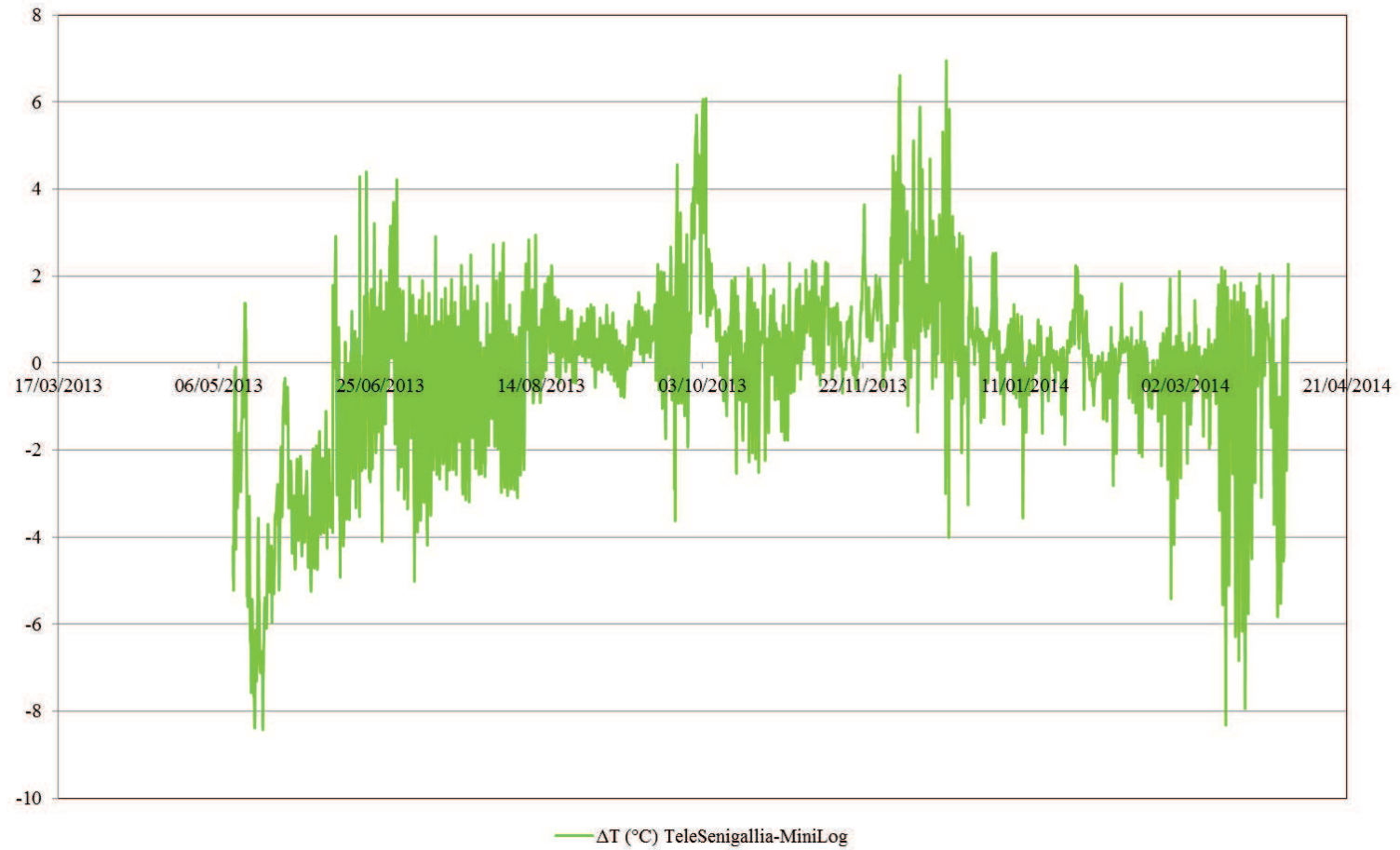


Fig. 44. Temperature differences (ΔT °C) between open sea (*TeleSenigallia* station) and study area (*MiniLog* station) recorded from the 10th of May 2013 to the 2nd of April 2014.

Air, Water, ΔT (*TeleSenigallia* - *MiniLog*) T [°C]

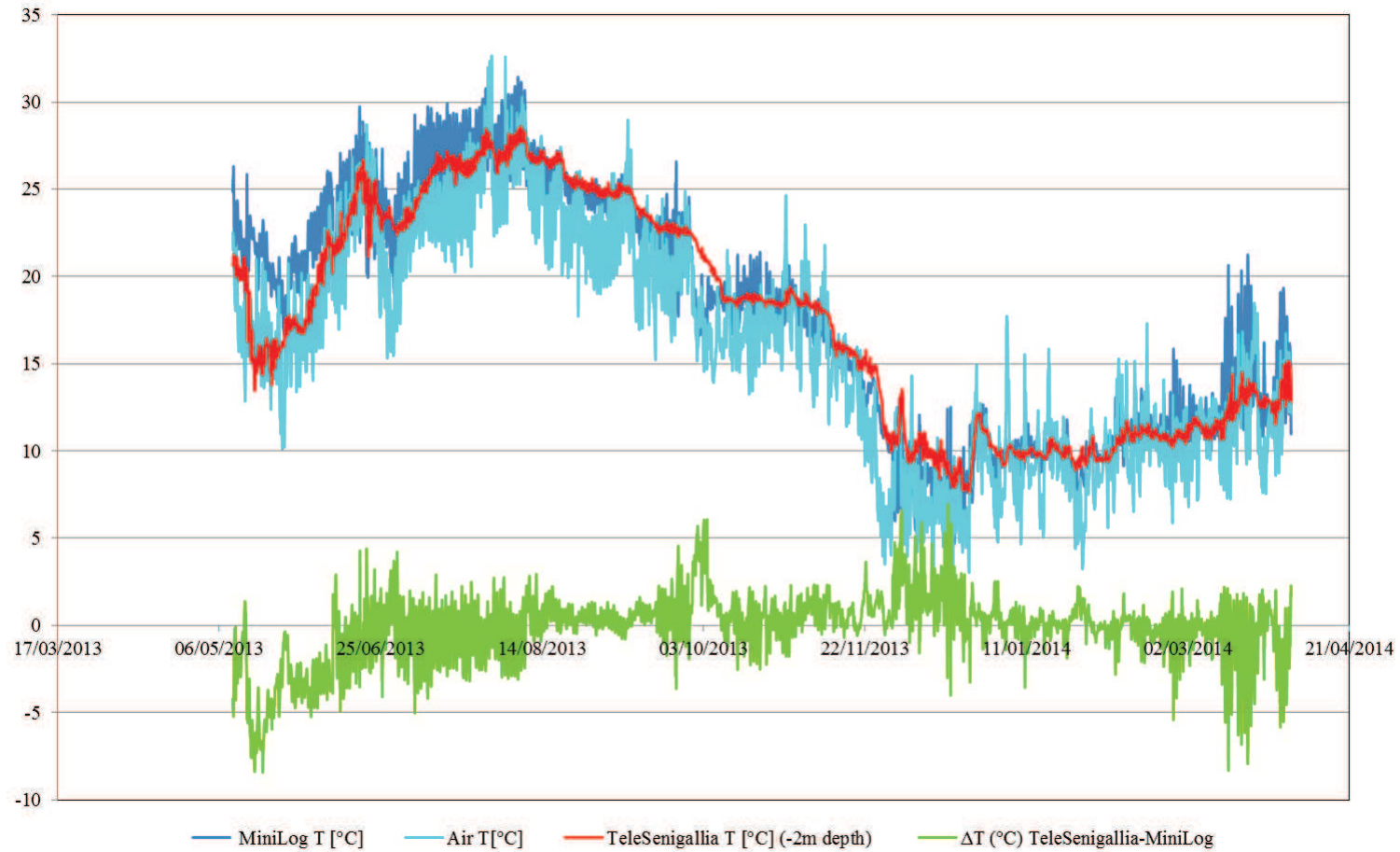


Fig. 45. Temperature recorded (T° C) of air (light blue line) and water (-2 m depth; red line) at *TeleSenigallia* station, on study area (*MiniLog* station, blue line) and ΔT (°C) between water temperature of *TeleSenigallia* and *MiniLog* stations (green line).

4.3.1.2 Abundance

During two years of investigation 27 species of crustaceans and 17 species of fishes have been collected in the study area.

The presence of Mollusks in the samples was always really low, therefore they were not considered in the following analysis.

Five different orders of Crustaceans have been identified. The most abundant was *Decapoda* with 16 species, followed by *Mysida* with 7 species, *Isopoda* with 2 species, then *Amphipoda* and *Tanaidacea*, both with 1 species each.

The most abundant among fishes was *Perciformes* with 9 species, followed by *Pleuronectiformes* with 5 species, then *Clupeiformes* and *Syngnathiformes* both with 1 species each, and finally a not identified fish *larvae* group (Tab. XV).

Crustaceans *Decapoda* showed the highest density, in particular *Phillocheras spp.* (333.7 ind. 100 m⁻² in the sample on July 1, 2014; Tab. XV, Fig. 50). Also *Mysidae NEI*, surveyed in each sampling, showed remarkable densities that reached 173,8 ind. 100 m⁻² on 19 March, 2014. The third species in terms of abundance was *P. kerathurus* (in particular juveniles), which recorded for the entire study period a total abundance value of 101.9 ind. 100 m⁻². The species showed a high seasonality being collected only during summer-autumn months, with peaks next to the middle of August (35.9 ± 6.44 and 28.7 ± 2.32 ind. 100 m⁻² respectively on 13 August, 2013 and 19 August 2014; Tab. XV; Tab. XVI, Fig. 46, Fig. 50).

The trend of sea temperature and the abundance of *P. kerathurus* in the study area seem to show a strong correlation. In fact, the highest abundance value registered followed the maximum temperature one (29,2 °C, 6 August 2013) followed highest abundance values (35,9 ind. 100 m⁻²; 13 August 2013). During the rest of the summer, a progressive lowering of water temperature was accompanied by an equally progressive lowering of abundance of caramote prawn juveniles (Fig. 46). In 2013 the last specimens were recorded on 10 October (1.3 ind. 100 m⁻²), when the water temperature was 18.0 °C (Tab. XV, Fig. 46).

Spearman test evidenced a statistically significant relationship ($R= 0.796$; $p=0.0153^*$) and linear correlation between abundance of *P. kerathurus* juveniles and water temperature (slope=0.291; intercept=18.814; Fig. 47).

Among fish, *Pisces larvae NEI* showed higher densities, both in summer 2014 and 2013 (respectively 10 ind. 100 m⁻² on 3 June 2014 and 5.4 ind. 100 m⁻² on 6 August 2013; Tab. XV, Tab. XVI, Fig. 51)

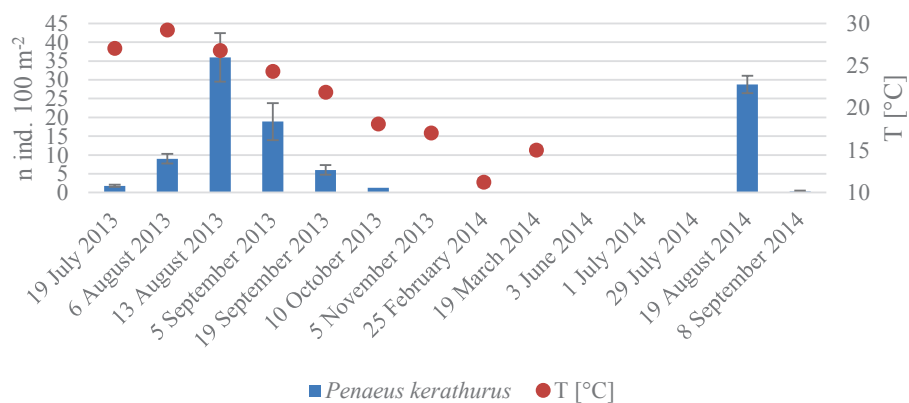


Fig. 46. Mean abundances of *P. kerathurus* (n ind. 100m⁻²; blu bars, \pm standard error) and water temperature (red line, \pm standard error, from *MiniLog*, see above) recorded in the study area from 19 July 2013 to 8 September 2014.

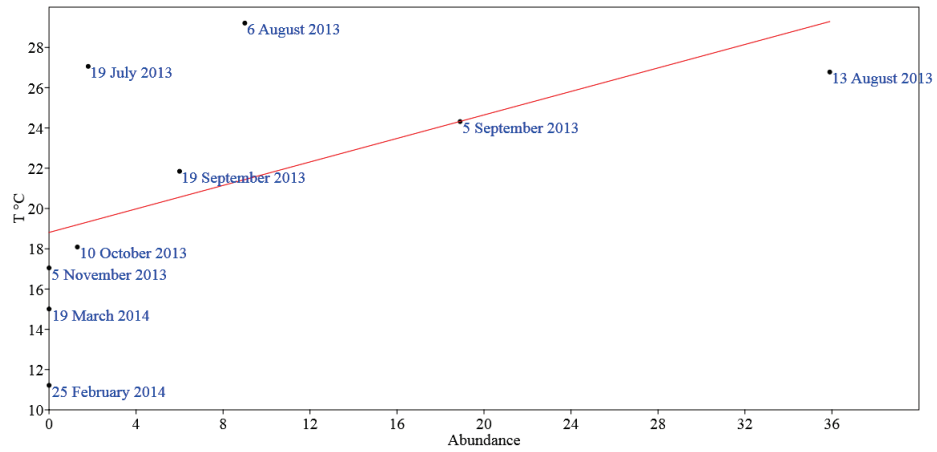


Fig. 47. Linear correlation between abundance (n. ind. 100 m⁻²) of *P. kerathurus* juveniles and water temperature collected in the study area from July 2013 to March 2014.

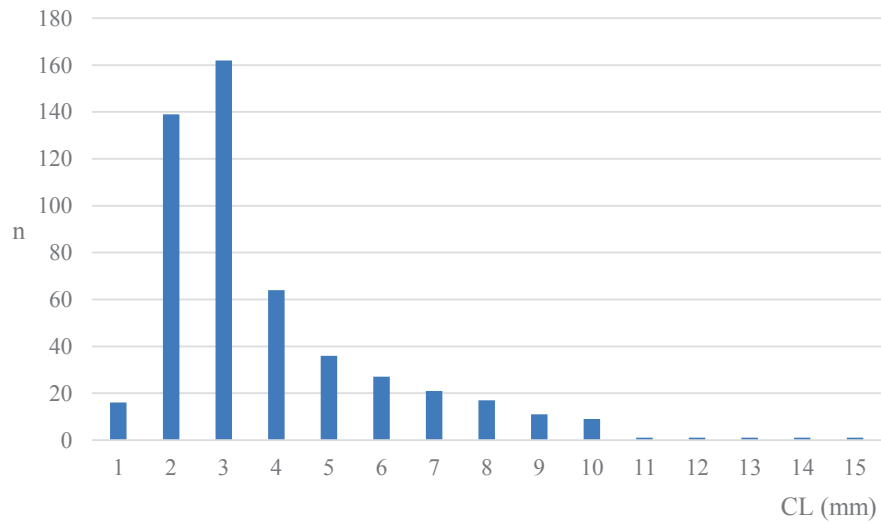


Fig. 48. Total length-frequency distribution of Carapace Length (CL) of *P. kerathurus* recorded in the study area from the 19 July 2013 to 8 September 2014.

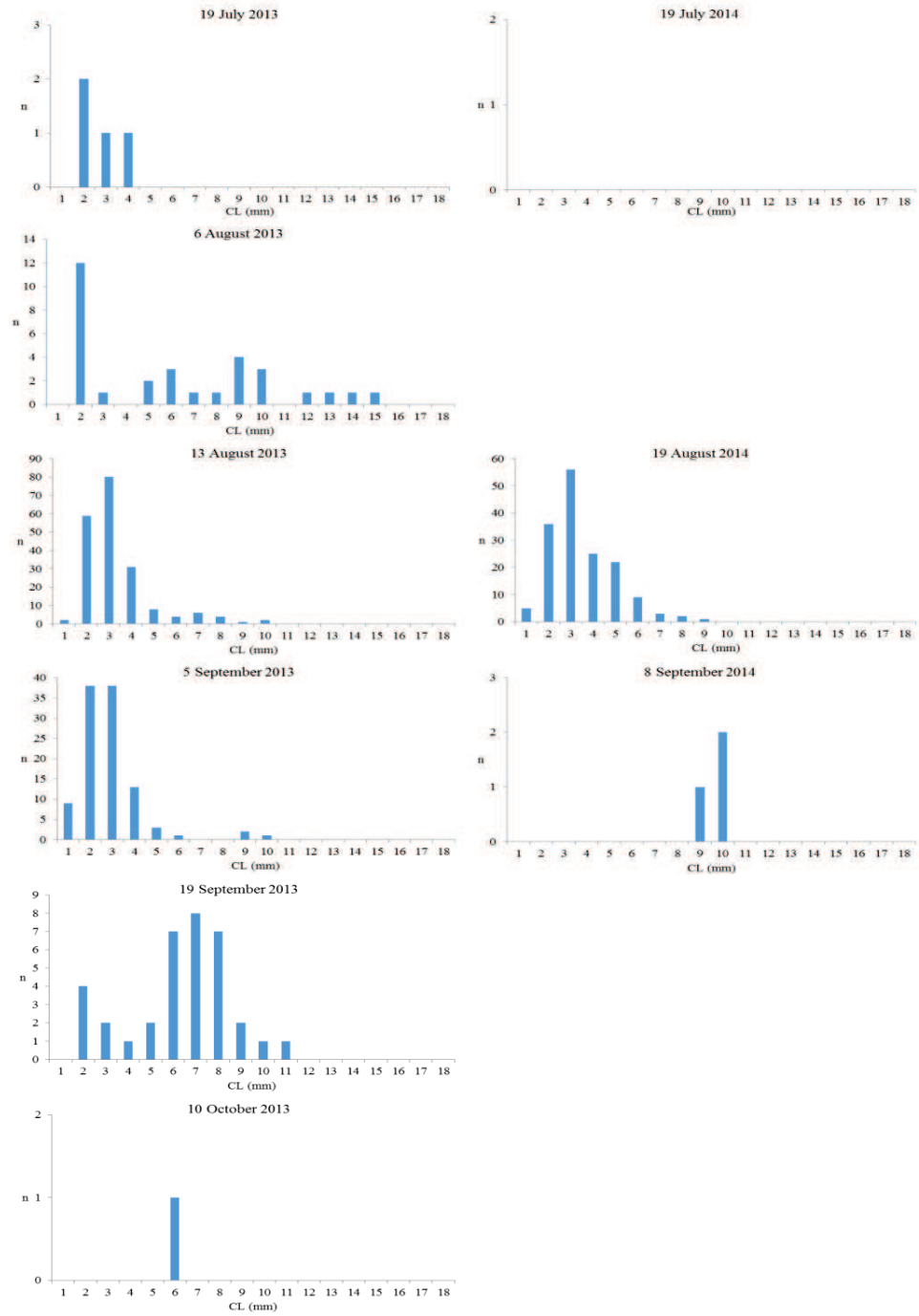


Fig. 49. Length-frequency distribution of Carapace Length (CL) of *P. kerathurus* recorded in each sampling in the study area from 19 July 2013 to 8 September 2014.

Tab. XV. Faunal list and mean abundances (n. of specimens 100 m⁻²) of the censed species in the study area in each samplings, from 19 July 2013 to 8 September 2014.

			2013						2014						Total			
			19 July	6 August	13 August	5 September	19 September	10 October	5 November	25 February	19 March	3 June	1 July	29 July		19 August	8 September	
Crustacea	Amphipoda	<i>Amphipoda</i> NEI	1,7						1,3		85,4	3,9	1,6	7,7	6,6	0,7	108,9	
	Decapoda	<i>Athanas nitescens</i>			1,1											0,2		1,3
		<i>Brachynotus gemmellari</i>														0,2		0,2
		<i>Carcinus aestuarii</i>										0,3	0,5	0,9		0,4	0,3	2,2
		<i>Crangon crangon</i>	2,1		1,4								3,0	1,7			1,0	9,2
		<i>Diogenes pugilator</i>								1,0			1,2	0,7		1,1	2,1	6,0
		Hippolytidae		1,6	2,1	1,0	1,0	1,1		1,4						0,4		8,5
		<i>Liocarcinus vernalis</i>								4,7	0,4			0,7	1,1	6,8	6,1	20,6
		<i>Palaemon adspersus</i>														1,1		1,1
		<i>Palaemon elegans</i>													2,2	1,1		5,6
		<i>Palaemon serratus</i>			4,3	1,0	1,4	1,8			0,7	1,9	3,1	1,4		0,9	0,8	17,2
		<i>Penaeus kerathurus</i>	1,8	9,0	35,9	18,9	6,0	1,3								28,7	0,3	101,9
		PHILOCHERAS spp	100,0	71,4	67,0	82,9	37,1	66,7		126,7	0,4	91,8	265,1	333,7	130,8	115,1	62,8	1551,6
		<i>Pisidia longimana</i>														2,0		2,0
		<i>Portunus latipes</i>												0,0		0,2		0,2
	<i>Processa macrophthalma</i>		1,6	1,3	2,0	1,2				0,1	1,4	0,2			0,7	2,4	11,1	
	<i>Processa modica</i>								1,4		3,6		0,2	4,4	2,7	2,9	15,3	
	Isopoda	<i>Cleantis prismatica</i>	1,5								1,2	1,9	0,9		1,8		7,2	
		<i>Idotea balthica</i>	1,3							1,2		0,9	0,7	1,1	5,2	0,3	10,6	
	Mysida	<i>Gastrosaccus</i> sp.										4,7		1,1	0,2		6,0	
		<i>Leptomysis</i> sp.										0,0			0,7		0,7	
		<i>Mesopodopsis slabberi</i>		15,1	8,0	3,3	8,3	1,0	4,5			0,4			0,5		41,2	
		<i>Misidae</i> NEI	14,6	53,8	47,5	146,6	124,3	34,3	72,4	0,1	173,8	17,4	18,9	8,8	72,5	63,2	848,2	
		<i>Paramysis (Longidentia) helleri</i>	1,0									1,6			2,9		5,5	
		<i>Schistomysis assimilis</i>	3,1									0,0			4,1		7,2	
	Tanaidacea	<i>Siriella jaltensis</i>	19,0									8,3		1,1	62,3		90,8	
		<i>Apseudes</i> sp										1,2					1,2	
Total Crustacea			146,2	152,5	168,7	255,8	179,3	106,2	214,7	1,9	359,4	316,5	361,2	158,2	318,4	142,7	2881,6	
Fish	Clupeiformes	<i>Sardina pilchardus</i>										1,2					1,2	
	Perciformes	Blennidae (juv.)											1,1					1,1
		<i>Callyonimus rissoi</i>		1,6			1,0	2,0									0,2	4,7
		<i>Diplodus sargus sargus</i>										0,3					1,1	1,4
		<i>Echiichthys vipera</i>						1,0				0,2	0,2					1,4
		<i>Gobius</i> sp.		1,6	1,1							0,5						3,2
		<i>Lithognathus mormyrus</i> (juv.)			6,3	3,4	1,4	1,3								0,4	0,2	13,0
		<i>Liza</i> sp.														0,5		0,5
		<i>Mullus barbatus barbatus</i>														1,1		1,1
	<i>Pomatoschistus</i> sp.	2,0	3,2	2,2	2,9	3,3	1,2	2,0	0,3			1,0	1,5	1,1	0,4		21,1	
	Pleuronectiformes	<i>Buglossidium luteum</i>			1,4	1,0	1,0	1,5	1,2			0,5	0,4					6,9
		<i>Pegusa</i> sp.	1,4		1,6			1,3					0,4	1,1	0,9	1,3		8,0
		<i>Scophthalmus maximus</i>										1,2						1,2
		<i>Scophthalmus rhombus</i>															1,1	1,1
		<i>Solea solea</i>										0,7						0,7
	Syngnathiformes	<i>Syngnathus</i> sp.									1,1		1,1					2,2
		<i>Pisces larvae</i> NEI		5,4	5,9	1,4	2,0	1,1		0,0		10,0	0,0	0,0	0,1	0,2		26,2
Total Fish			3,5	11,7	18,5	8,8	8,6	9,3	3,2	0,3	1,1	16,7	3,6	2,2	3,5	4,1	95,1	
TOTAL			149,6	164,2	187,2	264,6	187,9	115,5	217,9	2,1	360,5	333,2	364,9	160,4	321,8	146,7	2976,7	



Fig. 50. Example of all organisms obtained in a sampling replicate after sorting (A) and *P. kerathurus* juveniles (B,C and D).

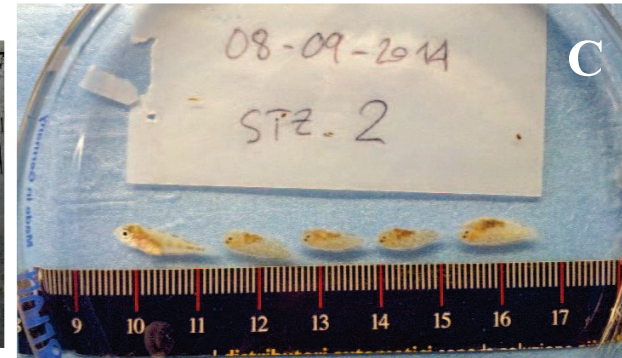
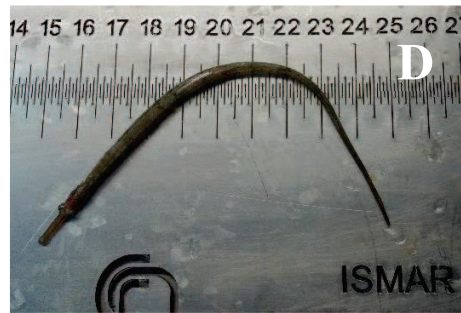
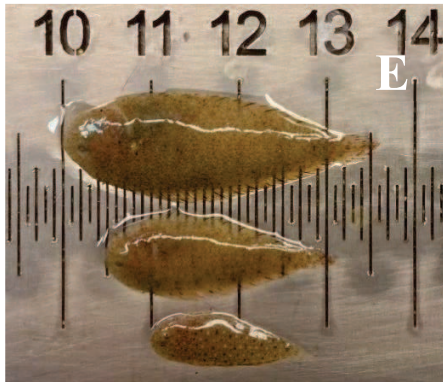
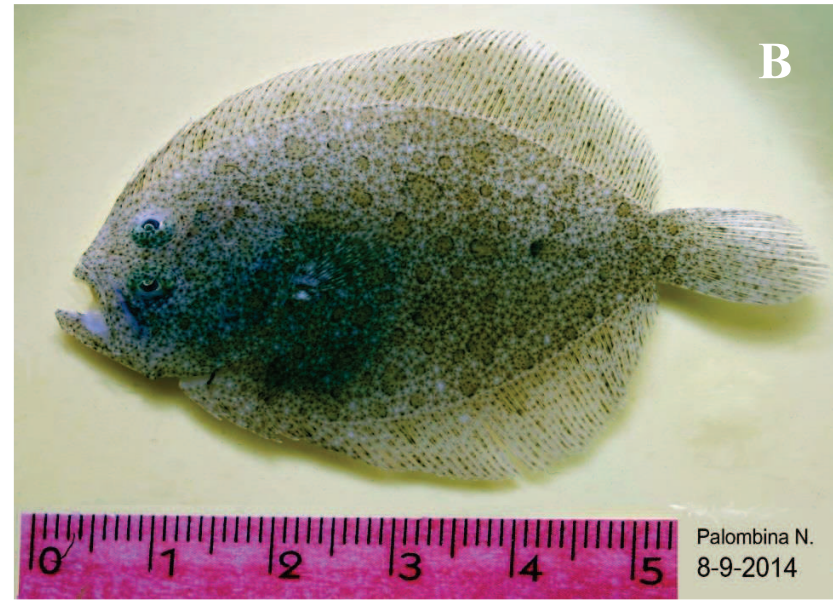
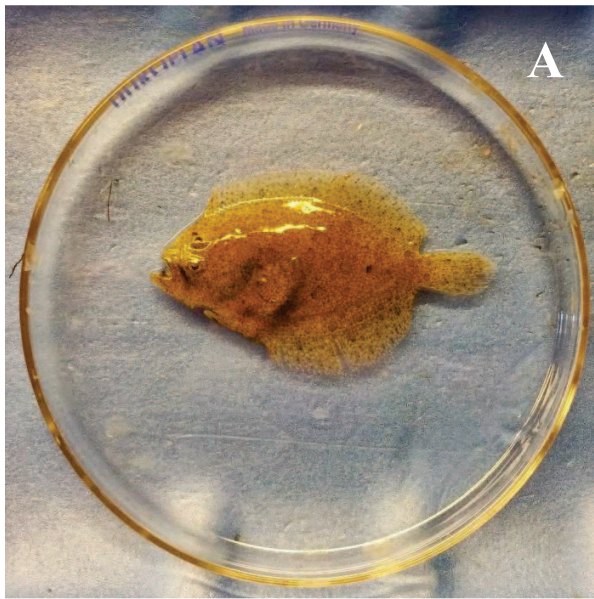


Fig. 51. Example of fish collected during a sampling replicate; *Pleuronectiformes* (A, B, E), *Pices larvae NEI* (C) and *Syngnathiformes* (*Syngnathus* sp.; D).

4.3.1.3 Dominance

The graphical representation of dominance shows a certain complexity of the populations in the study area. According to *Tischler's* scale for species dominance (Tischler 1949), in general, at each sampling a few species eudominant or dominant, and much more species belonging to lower classes of dominance were observed. The genus *Philocheras* was eudominant during all the year, while amphipods only in March 2014, with over 40% because they have been examined since the end of 2013 (Fig. 53).

The target species *P. kerathurus* was eudominant on 13 August 2013, with 21.0% and dominant on 6 August, on 5 September 2013 and on 19 August 2014, respectively with 5.6%, 9.8% and 8.46%. This species appeared subdominant on 19 September 2013 and subprecedent during the limits of their census, represented by July and October 2013 and September 2014 (respectively 0.6%, 0.1% and 0.2%; Fig. 53, Fig. 52, Tab. XVI).

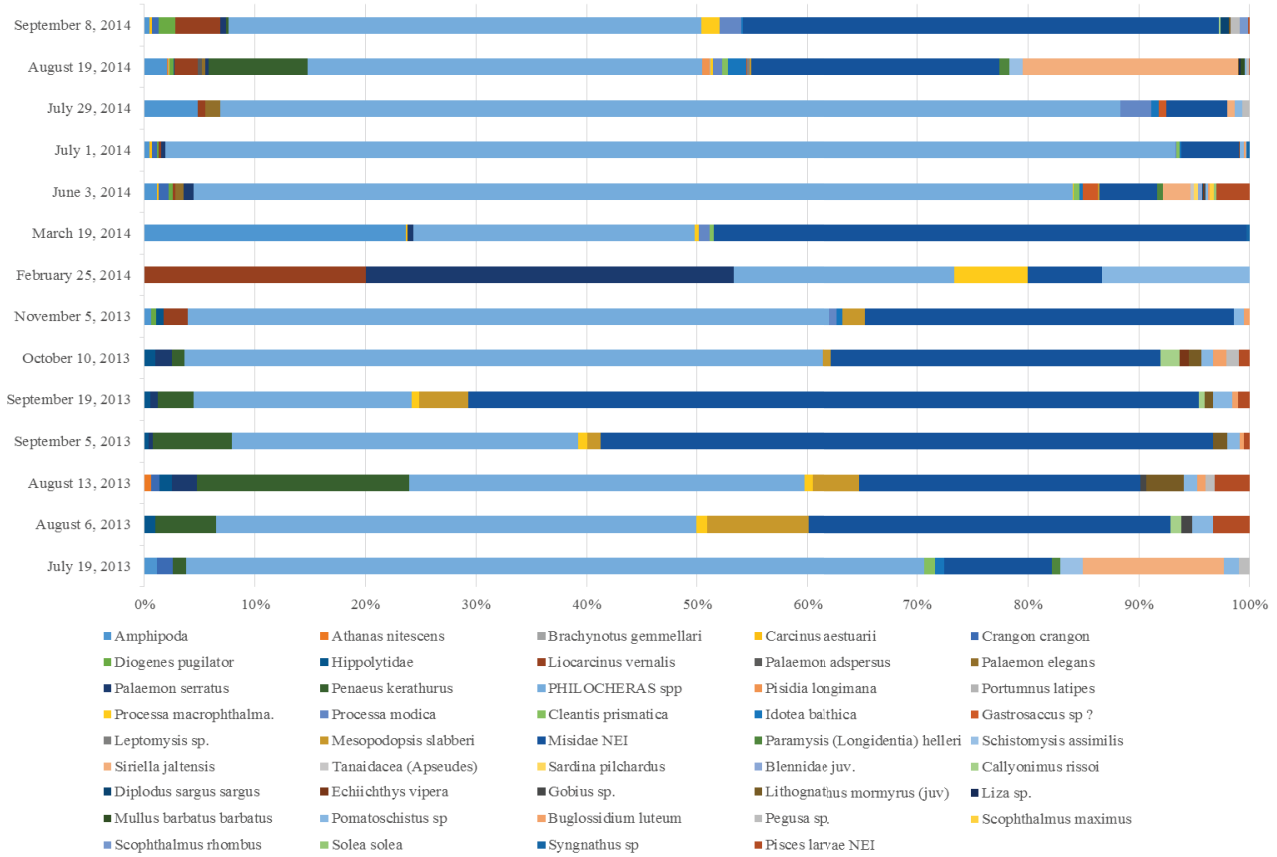


Fig. 53. Graphical representation of dominance (species %) recorded in the study area from 19 July 2013 to 8 September 2014.

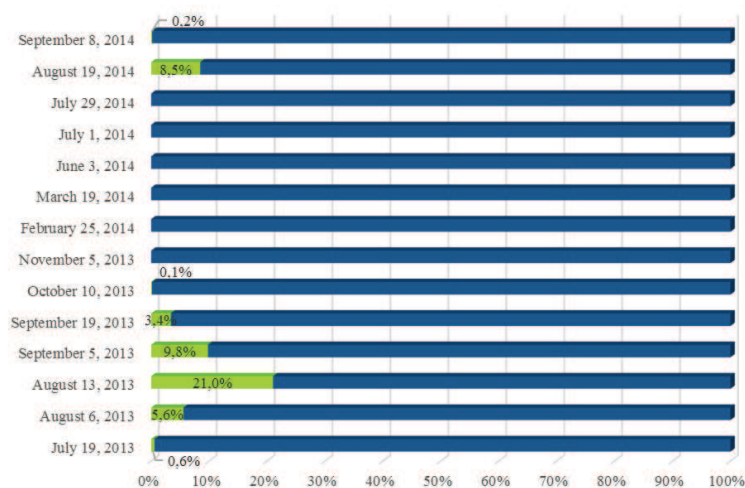


Fig. 52. Graphical representation of dominance (%) of *P. kerathurus* recorded in the study area from 19 July 2013 to 8 September 2014.

4.3.1.4 Total Species Richness

Total species richness was quite variable and mainly represented by crustaceans. In fact, crustaceans have shown always more species richness than fishes, to exception for sampling of 10 October 2013. For crustaceans the minimum number of species was recorded in February 2014 (5) and maximum in August of the same year.

Fishes species richness exhibited minimum value in February and March 2014 (1 species respectively) and maximum in June 2014 (10 species; Fig. 54, Tab. XVI).

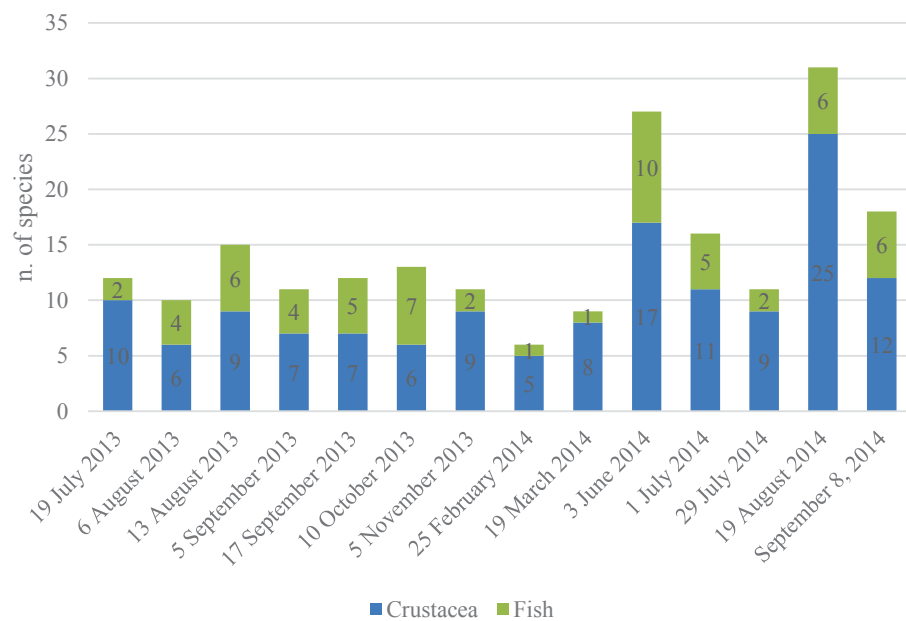


Fig. 54. Total species richness (S; total n of species; Blu bars: Crustacea; Green bars: Fish) recorded in the study area from 19 July 2013 to 8 September 2014.

4.3.1.5 Average Richness

Average species richness has been variable, especially comparing the two years. The highest values in 2013 were recorded in November (8.8 ± 0.7), while in 2014 were recorded in August (17.6 ± 1.5 ; Fig. 55). In the last year, average richness assumed higher values than previous, although with greater fluctuations between each sampling (Fig. 55).

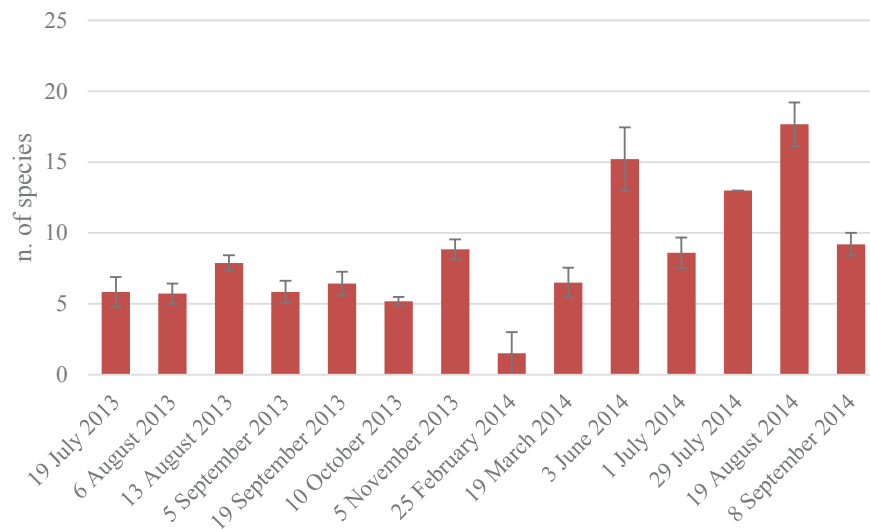


Fig. 55. Average species richness ($S_a \pm$ standard error) recorded in the study area from 19 July 2013 to 8 September 2014.

4.3.1.6 Shannon - Wiener's Diversity Index

The *Shannon - Wiener* specific diversity index was quite variable throughout the study period, with highest values recorded around the middle of the months of August, with values respectively of 2.51 and 2.58. For what concern the remaining period the specific diversity values were between 1 and 2.5, with the exception of 1 July 2014 when it reached the minimum of the entire observation period (0.62; Fig. 56, Tab. XVI).

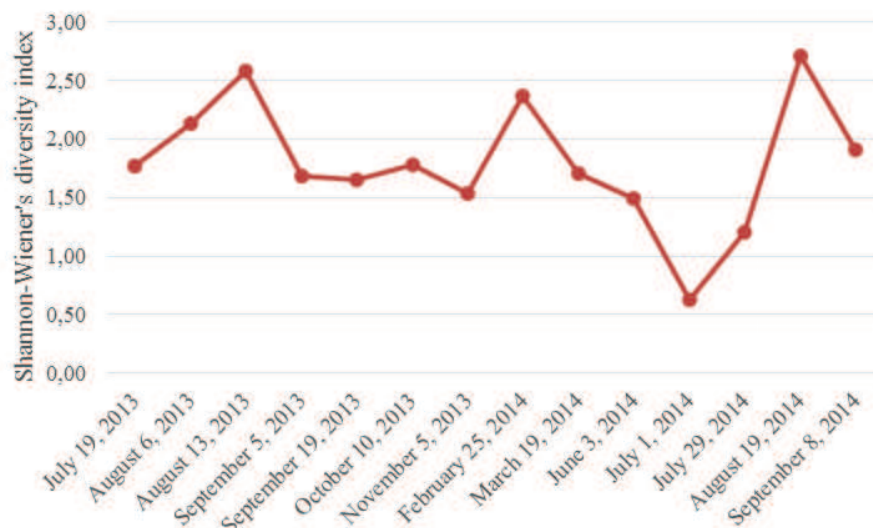


Fig. 56. Shannon - Wiener specific diversity index recorded in the study area from 19 July 2013 to 8 September 2014.

Analysis of variance and non-parametric statistical analysis, take into account species density (n ind. 100 m⁻²) of samples from summer season (June, July, August) and all the other seasons, pointed out no statistical significance for

equal means (one-way ANOVA, $p=0.1803$), as confirmed by *Kruskal-Wallis* test for equal medians ($H=2.077$; $p=0.1495$) and *Mann-Whitney* pairwise ($U=567$; $p=0.151$; Fig. 57). On the contrary, analysis of variance and non parametric statistical analysis revealed for species richness (number of species) a high statistical significance for these two groups (one-way ANOVA for equal means: $p \ll 0.001^{**}$; *Kruskal-Wallis* test for equal medians: $H=15.91$; $p \ll 0.001^{**}$, and *Mann-Whitney* pairwise: $U=361$; $p \ll 0.001^{**}$; Fig. 58).

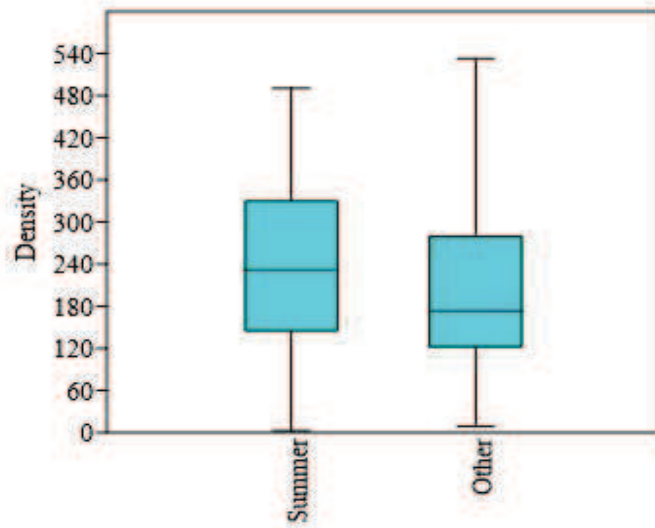


Fig. 57. Box and Whiskers plot of species density in the study area take into account summer and other seasons.

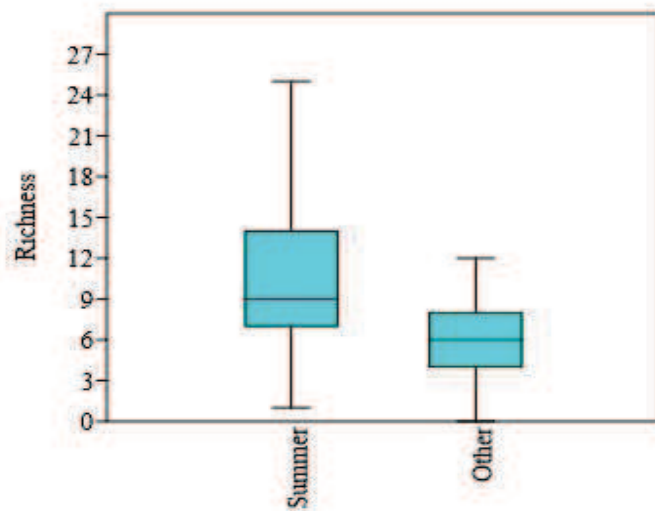


Fig. 58. Box and Whiskers plot of species richness in the study area take into account summer and other seasons.

Tab. XVI. Community ecology parameter recorded in the study area from July 19, 2013 to September 8, 2014 (species richness, abundance, number and abundance of eudominant, dominant, subdominant, recedent and subrecedent species based on Tischler's scale, Shannon – Wiener diversity index).

			2013							2014							
			19 July	6 August	13 August	5 September	19 September	10 October	5 November	25 February	19 March	3 June	1 July	29 July	19 August	8 September	
Species richness	S	number of species	12	10	15	11	12	13	11	6	9	27	16	11	31	18	
Abundance	N	number of specimens	149,6	164,2	187,2	264,6	187,9	115,5	217,9	15,0	360,5	333,2	364,9	160,4	321,8	146,7	
Number of eudominant species	SE	$D_t = \frac{n_i}{N} \cdot 100\%$	2	2	3	2	2	2	2	4	3	1	1	1	3	2	
Number of dominant species	SD		1	2		1				2		1	1	1	1		
Number of subdominant species	SSd		1	1	4		2		2			2		2	2	2	1
Number of recedent species	SR		5	1	2	3	2	7			1	2		1	2	2	3
Number of subrecedent species	SSr		3	4	6	5	6	4	7		5	21	14	6	23	12	
Abundance of eudominant species	NE		119,0	125,2	150,4	229,5	161,5	101,0	199,2	13,0	351,0	265,1	333,7	130,8	249,9	126,0	
Abundance of dominant species	ND		14,6	24,1		18,9				2,0		17,4	18,9	8,8	28,7		
Abundance of subdominant species	NSd		3,1	5,4	24,5		14,3		9,2			18,4		12,1	13,4	6,1	
Abundance of recedent species	NR		9,2	3,2	4,3	9,7	5,3	10,3			3,6	8,6		2,2	9,3	7,4	
Abundance of subrecedent species	NSr		3,7	6,4	7,9	6,5	6,9	4,2	9,5		5,9	23,7	12,3	6,6	20,5	7,3	
Shannon-Wiener diversity index	H'		$H' = \sum_{i=1}^S p_i \cdot \log_2 p_i$	1,77	2,13	2,58	1,69	1,65	1,78	1,54	2,37	1,70	1,49	0,62	1,20	2,71	1,91

4.3.2 Study area, control site and natural Marano Lagoon in summer 2015

Sampling conducted in the study area in August 2015 confirmed the presence of *P. kerathurus* juveniles. A total of 26 specimens were recorded with abundance of 9.03 ± 2.503 ind. 100 m^{-2} (mean \pm std. dev.). Length-frequency distribution showed size ranged from 2 to 10 mm CL, with mode at 3 mm CL (Fig. 59).

In the control site, sampled 80 km southward study area, a total of 14 specimens were recorded, with abundance of 7.99 ± 2.411 ind. 100 m^{-2} (mean \pm std. dev.). Their length-frequency distribution have been shown size ranged from 2 to 9 mm CL, with same mode to 3 mm CL (Fig. 60).

Different length-frequency distribution have been shown by samples provided by artisanal fisheries in lagoon; a total of 601 specimens were measured with size ranged from 11 to 23 mm CL and mode at 15 mm CL (Fig. 61).

Analysis of length-frequency distribution of samples obtained in summer 2015 confirmed that no significant differences have been shown between the study area and other site, whereas was evident between these last groups and sample from Marano lagoon (Fig. 62; Fig. 63), as confirmed by statistical analysis such as *Mann-Whitney* test for equal medians and *Kolmogorov-Smirnov* test for equal distributions (Tab. XVII).

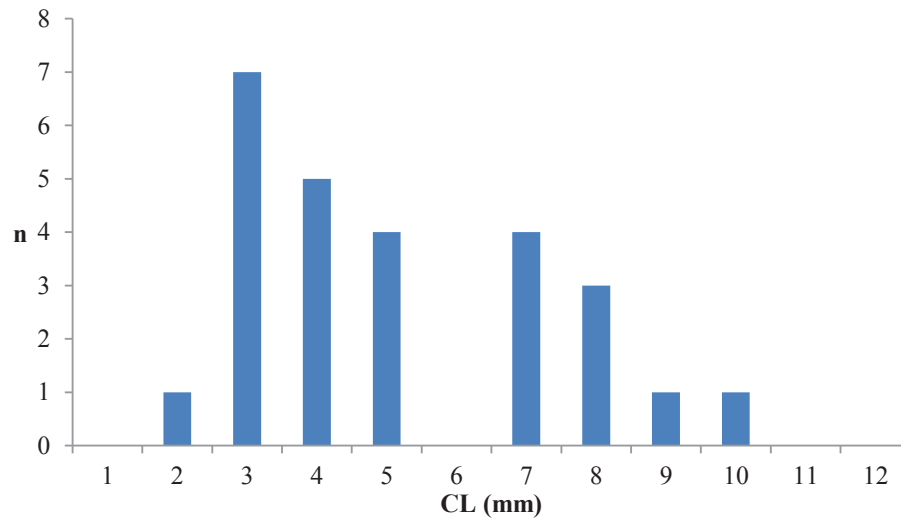


Fig. 59. Length-frequency distribution of *P. keraturnus* sampled in the study area during August 2015.

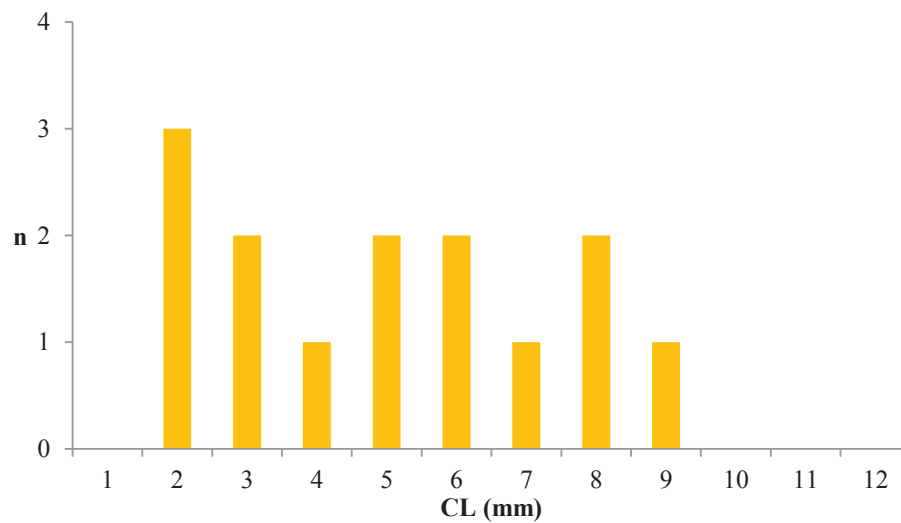


Fig. 60. Length-frequency distribution of *P. keraturnus* sampled 80 km southward the study area during August 2015.

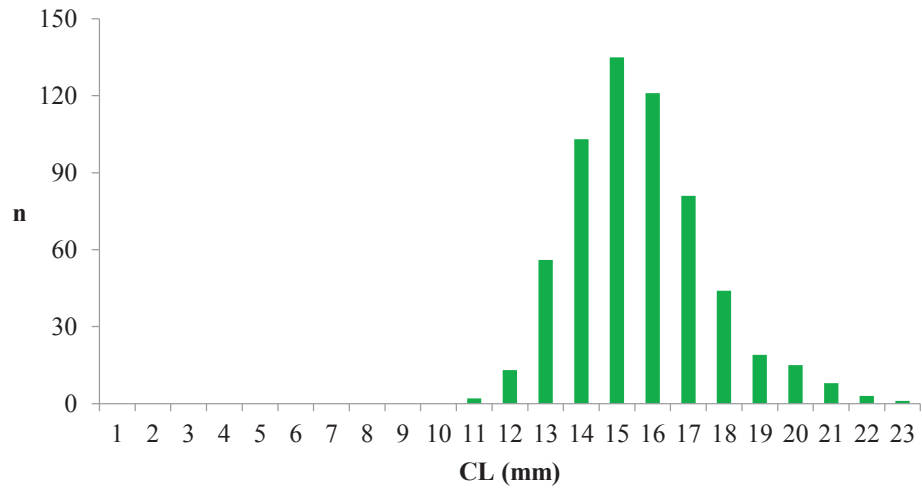


Fig. 61. Length-frequency distribution of *P. kerathurus* from Marano lagoon collected on September 2015.

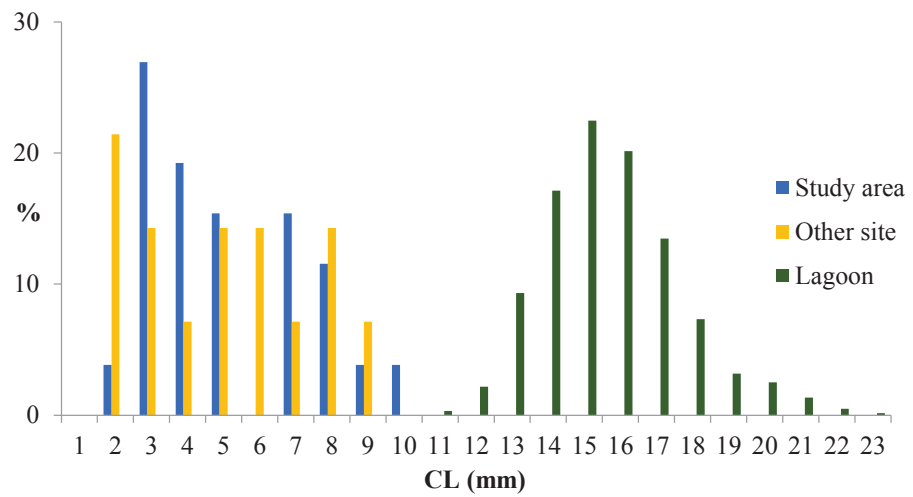


Fig. 62. Length-frequency distribution (percentage) of *P. kerathurus* from the study area (blue), from other site 80 km southward study area (yellow) collected in August and from Marano lagoon (green) collected on September 2015.

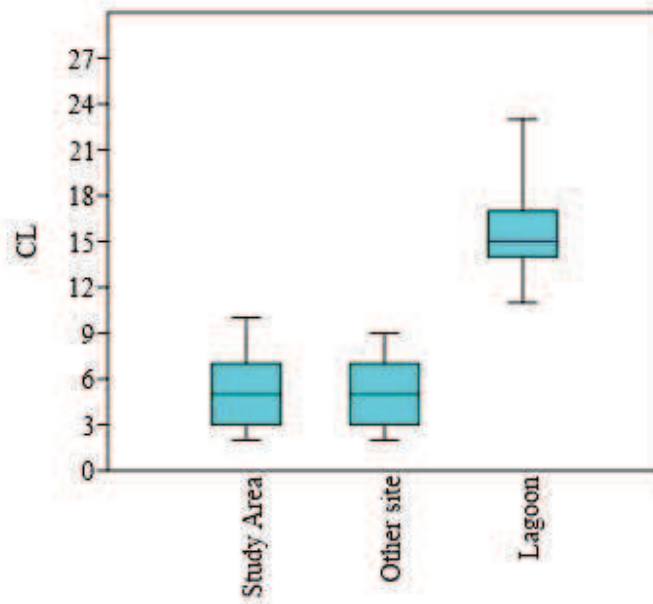


Fig. 63. Box and Whiskers plot of length-frequency distribution (CL, mm) of *P. kerathurus* from the study area, from other site 80 km southward study area collected in August and from Marano lagoon collected on September 2015.

Tab. XVII. Results of statistical tests for equal medians (Mann-Whitney) and for equal distribution (Kolmogorov-Smirnov) between study area, other site (southward study area) and Marano lagoon.

<i>Mann-Whitney</i>			
	Study area	Other site	Lagoon
Study area		p=0.818	P<<0.001**
Other site	U=173.5		P<<0.001**
Lagoon	U=0	U=0	
<i>Kolmogorov-Smirnov</i>			
	Study area	Other site	Lagoon
Study area		p=0.598	P<<0.001**
Other site	D=0.241		P<<0.001**
Lagoon	D=1	D=1	

4.4 Marine Litter

The preliminary results of capability to adopt a similar sampler gear and methodology for marine litter sampling, highlight that it could be a very useful technique for the purpose.

Samples collected in the study period, have shown a good performance of the sampler in terms of wide range of size and material (anthropogenic) collected (Fig. 64). However, data obtained in this study will not be covered in this context.



Fig. 64. Some marine litter samples obtained during sampling activity in the study area.

4.5 Discussion

The Adriatic coastline represents an important economic resource, especially for local communities. It is affected by erosion due to both natural and man-induced processes. The causes of this phenomenon includes hydrodynamical factors as waves, tides and induced currents, and human factors as river management and sand-gravel mining, which determines sediment loss. To avoid this, in the last 50 years a variety of coastal protection structures have been developed, including breakwaters. There are many examples, unfortunately, where this structure have produced adverse effects on the coastline (Colantoni et al. 1997; Lamberti & Mancinelli 1996).

Along European coasts in order to prevent and reduce shoreline erosion, the most common artificial constructions adopted are breakwaters, that interrupt wave action, modifying water circulation (Zyserman et al. 2005) and sediment transportation (Thomalla & Vincent 2003; Cuadrado et al. 2005).

Thus, changes in bottom topography, sediment grain size and organic content could happen (Agnew et al. 2009; Correggiari & Frascari 1992; Bull et al. 1998). Concurrently, biotic alterations of fauna from adjacent bottoms are displayed, as species abundance, distribution pattern and community structure.

Some studies reported to the impact of this hard substrata on the native soft bottom fauna (Davis et al. 1982; Ambrose & Anderson 1990; Correggiari &

Frascari 1992; Posey & Ambrose 1994; Barros et al. 2001; Fabi et al. 2002; Jaramillo et al. 2002; Barros et al. 2004; Martin et al. 2005).

Bertasi et al. (2007) evidence the positive influence on macrofaunal settlement due to modifications in hydraulic conditions, particularly on most sensitive species to hydrodynamic stress.

The establishment of assemblages that differ in species richness, composition or relative abundances from those associated with nearby exposed rocky habitats could be produced by reduction of water flow, turbidity and sediment abrasion (Bulleri & Chapman 2004; Clynick 2006; Vaselli et al. 2008).

Some key ecological processes may differ between natural and artificial structures, such as recruitment (Glasby 1999; Bulleri 2005), foraging (Bulleri et al. 2004), competition, predation (Kirk et al. 2007) and reproduction (Moreira et al. 2006), therefore this structures do not function as surrogates of natural habitats (Bulleri & Chapman 2010).

In addition, this human-made structure could contribute to the expansion of invasive species, such as the example of *Codium fragile* ssp. *tomentosoides* in north Adriatic Sea, that has benefited of provided hard substrata for its colonization (Bulleri & Airoidi 2005; Bulleri et al. 2006).

This study showed the particular thermal regime that happens in area protected by breakwater. Identical average water temperature were recorded during the study between the study area and an area of open sea (17.6 °C), instead a broad

temperature ranges were shown in the study area (respectively -8.4 °C and +6.9 °C the wider recorded temperature differences).

It has been demonstrate the presence of juveniles of *P. kerathurus* in the study area and in a control site 80 km southward the study area during the summer season, with very high concentration, particularly evident in the middle part of the months of August (35.9±6.44 and 28.7±2.32 ind. 100 m⁻² in the study area respectively in August 2013 and 2014). In addition, the presence of caramote prawn juveniles in a control site so far, may suggest that this “lagoon effect” exerted by breakwater constructions is not limited to the study area but it’s reasonable to assume the effect as generalized in a wider area. A correlation between water temperature and abundance of *P. kerathurus* juveniles in the study area was observed. The length-frequency distribution of juveniles collected during the period ranged from 1 to 15 mm of carapace length (CL), with mode at 3 mm CL. In addition, comparison of length-frequency distribution of juveniles of caramote prawn collected in study area, control site and natural lagoon in summer 2015, showed very similar length-frequency distribution for study area and control site, instead they showed a certain complementarity with those recorded for natural lagoon, as confirmed by statistical analysis. This can be ascribed to one month shift in the sampling and fact that juveniles, after reaching a certain size, move from the lagoon to offshore water.

In the summer season *P. kerathurus* has held some importance than the other species, reaching 21.0% of dominance in August 2013. Take into account species density (ind. 100 m⁻²) and species richness (number of species) for summer season (June, July and August) and the other seasons, no statistical relation have been found for density, while, high statistical significance relation have been found for species richness.

Finally, gear and methodology adopted for the study showed a certain functionality as marine litter sampler.

During this study many samples of marine litter were collected (Fig. 64). At first glance, sampler gear and sampling methodology can be useful to monitoring marine litter. In fact, this methodology is not only effective for this purpose, but allows the sampling in an area still never investigated for the purpose. This narrow strip of sea, as well as representing natural link between open sea and beach (both already considered for the marine litter monitoring) is subject to high anthropic impact. Therefore, it is reasonable suppose this method can open up new horizons in marine litter monitoring, providing accurate data useful to identify origin of this litter.

5. POPULATION PARAMETERS

5.1 Introduction

One of the most basic concepts of fisheries science is that fish are usually not seen, counted or weighed before they are caught, hence, from the very start, fisheries research was dependent on indirect inference (mathematical and statistical) for quantifying the vital statistics of fish populations required for fisheries management. Thus, some of the earliest applications of mathematics and statistics (outside of the physical sciences) are those of fisheries biologist (Gayanilo & Pauly 1997).

The weight-length relationship (LWR) is a useful tool in fish biology, physiology, ecology, and stock assessment. Actually, size is generally more relevant than age, mainly because several ecological and physiological factors are more size dependent than age-dependent (Santos et al. 2002). Indeed, in sampling programs, it is usually easier to measure only length, while weight cannot be easily recorded. The LWR of a species allows the interconversion of these two parameters. In biological studies LWRs allow the assessment of seasonal variations in fish growth and the calculation of condition indexes (Richter et al. 2000), which is frequently used in the analysis of ontogenic changes (Safran 1992), for between-region life-history comparisons (Weatherley & Gill 1987; Petrakis & Stergiou 1995) as well as to identify the spawning season (Olim & Borges 2006). In fisheries studies LWRs have many

different uses, including the estimation of weight from length (Beyer 1991; Froese 2006; Froese et al. 2011) and of weight-at age (Petrakis & Stergiou 1995), as well as the conversion of growth-in-length equations to growth in-weight (Pauly 1993).

Furthermore, LWRs allow life history and morphological comparisons between different fish species, or between fish populations from different habitats and/or regions (Gonçalves et al. 1997).

Another useful model can be used to estimate, directly or indirectly, the parameters of the Von Bertalanffy growth function, from length frequencies. The growth may be defined as the change over time of the body mass (weight) of an organism, being the net result of two processes with opposite tendencies, one the building-up of body substances (anabolism) and the other the breaking-down of these substances (catabolism; Gayanilo & Pauly 1997).

From these concepts arises the Von Bertalanffy growth function:

$$L_t = L_\infty [1 - e^{-k(t-t_0)}]$$

Where:

L_∞ is the asymptotic length, that is the mean length the organism of a given stock would reach if they were to grow indefinitely;

K is the rate (of dimension time^{-1}) at which L_∞ is approached; and

t_0 is the “age of the organism at zero length” if they had always grown in the manner described by the equation (t_0 is generally negative).

The Von Bertalanffy growth function can also be expressed in terms of weight, as:

$$W_t = W_\infty [1 - e^{-k(t-t_0)}]^3$$

Where:

W_∞ is the asymptotic weight and all other parameters are as defined above.

The exponent 3 is justified because, when weight growth is isometric:

$$L = (W)^{\frac{1}{3}}$$

And more generally, weights proportional to length by raising them to the inverse of the power of the length-weight relationship b :

$$L = (W)^{\frac{1}{b}}$$

Which can be used when weight growth is either isometric or allometric to obtain growth parameters (K , t_0) generally equivalent to those that would be obtained by fitting the corresponding length data (Gayaniilo & Pauly 1997).

The aim of this study was to report the weight-length relationship of caramote prawn caught in the northern and central Adriatic Sea and other morphometric relations that characterize the species, in juveniles, males and females. In addition, allometry and Von Bertalanffy growth function parameters were estimate in order to evaluate the growth of the species in the Adriatic Sea.

Beverthon-Holt method was adopted in order to estimate the total mortality rate Z for adult male and female.

Materials and Methods

Morphometric characteristics include carapace length (CL, mm), total length (TL, cm), weight (nearest 0.1 g) and 2° abdominal width and were recorded for *P. kerathurus* specimens sampled during this study, considering juvenile specimens from the study area (Palombina, Ancona), juvenile specimens from natural lagoon (Marano Lagoon, Udine; with exception of 2° abdominal width) and male and female adult specimens collected from scientific and commercial sampling. A total of 73 juvenile specimens were recorded from the study area, 601 juvenile specimens from Marano Lagoon, 814 male and 878 female specimens both from scientific and commercial samples. Length-frequency distribution for adult male and female were evaluated and compared; the Kolmogorov-Smirnov test was adopted in order to test equal distribution.

Correlation and linear regression analysis were conducted between carapace length (CL) – tail length, carapace length (CL) – total length (TL) and 2° abdominal width – carapace length (CL) for juveniles from study area (n=73) and natural lagoon (n=423) and adult specimens (n=690 male; n=797 female) when paired data were both available.

The relationship between weight and length was calculated using the expression: $W = aL^b$, where W is the weight (g), L the total length (mm), a the intercept (initial growth coefficient) and b the slope (growth coefficient). This

equation can also be expressed in its logarithmic form: $\ln W = \ln a + b \ln L$ (Le Cren 1951).

The parameters a and b of LWR were estimated by linear regression analysis (least-squares method) on log-transformed data. The allometry coefficient is expressed by the exponent b of the linear regression equation. In the relationships between different types of variables (linear and ponderal), WLR reflects an isometric growth when $b = 3$, i.e., relative growth of both variables is identical (Mayrat 1970; Ricker 1975; Quinn II & Deriso 1999). When b value is < 3 it can be said to have a negative allometric growth and is defined hypoallometry; instead when b value is > 3 it showed a positive allometric growth and is defined hyperallometry (Shingleton et al. 2009; Shingleton 2010). The b -value of each groups (juveniles from study area, from Marano Lagoon and male – female adults) was tested by a t -test (Sokal & Rohlf 2009) with a confidential level of 95% in order to confirm if it was significantly different from the isometric value ($H_0: b = 3$).

The comparison between obtained values of t -test and the respective critical values allowed the determination of the ‘ b ’ values statistically significant and their inclusion in the isometric range ($b=3$) or allometric range (negative allometric; $b<3$). The Pauly’s seasonal model of Von Bertalanffy growth function (VBGF) was used to estimate growth parameters for adult male and female, and, due to their exclusively summer availability, non-seasonal model

was adopted for juveniles from the study area. The estimation was conducted using the electronic length frequency analysis I (ELEFAN I) routine incorporated into the software, Length-Frequency Distribution Analysis (LFDA; Kirkwood et al. 2001). The overall population value of total mortality (Z) was estimated as a functional relationship of the mean length of shrimp in the catch using the Beverton & Holt(1965) equations.

5.2 Results

5.2.1 Morphometry

Length-frequency distribution analysis highlighted a distribution ranged from 21 to 42 mm CL with mode at 30 mm CL for male, while ranged from 21 to 60 mm CL with mode at 36 mm CL for female (Fig. 66, Fig. 65).

The Kolmogorov-Smirnov test for equal distribution confirmed high statistical differences between these length-frequency distributions ($D=0.534$; $p \ll 0.001^{**}$).

Linear regressions between tail length (total length – carapace length) and carapace length (CL) and between total length (TL) and carapace length, taking into account juveniles from study area, from Marano Lagoon and from adult male and female, showed R^2 values ranged from 0.870 to 0.961 for tail length-CL and from 0.927 and 0.982 for TL-CL, respectively for juveniles from Marano Lagoon and for adult male (Fig. 67).

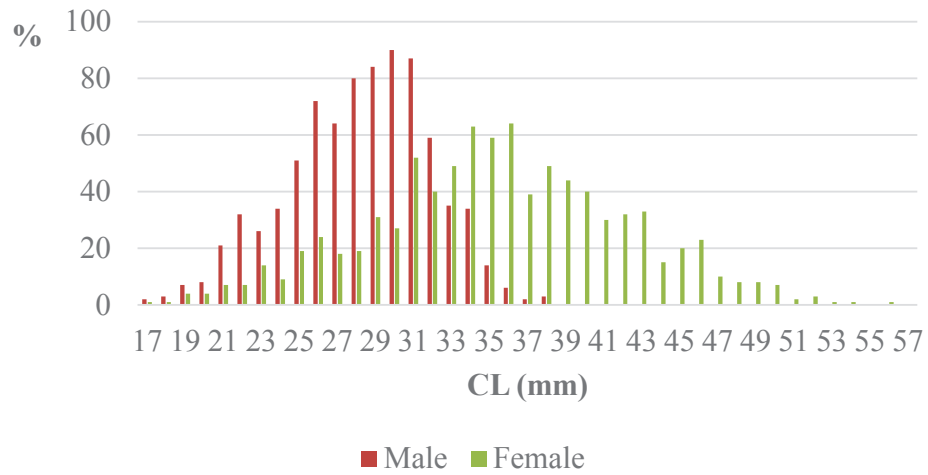


Fig. 66. Length-frequency distribution for male (n=814) and female (n=878) of *P. kerathurus*; CL: carapace length (mm).

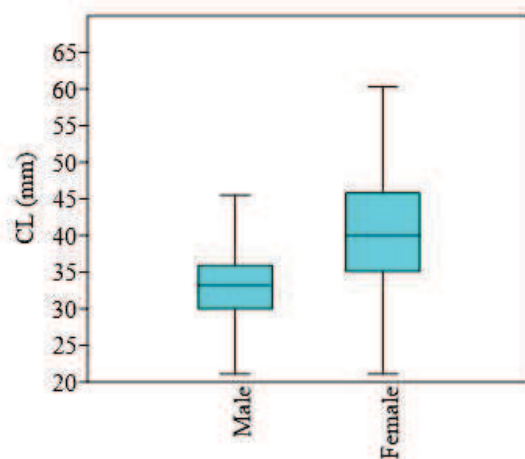


Fig. 65. Box and Whisker plot of carapace length (CL; mm) for male and female of *P. kerathurus*.

All morphometric relations taken into consideration have shown high significant statistical correlations ($p < 0.001^{**}$; Fig. 67).

Linear regressions between width of the 2° abdominal segment and carapace length (CL), taking into account juveniles from Marano Lagoon and from adult

male and female, showed R^2 values ranged from 0.86 to 0.95, respectively for juveniles from Marano Lagoon and for adult male (Fig. 68).

Also these morphometric relations taken into consideration have shown high significant statistical correlations ($p < 0.001^{**}$; Fig. 68).

5.2.2 Length-Weight Relationship of *P. kerathurus*

The exponential and log-transformed correlation between carapace length (LC, mm) and weight (g) have highlighted that all regression were highly significant ($p < 0.01^{**}$) and the coefficient R^2 values ranged from 0.863 for juveniles from Marano Lagoon to 0.986 for adult male and female (Fig. 69).

The highest value of parameter b was 3.03 (± 0.131 SE) for juveniles from the study area and the lowest one was 2.62 (± 0.064 SE) for juveniles from Marano Lagoon (Tab. XVIII).

In terms of growth type, juveniles from study area showed positive allometric growth ($b > 3$) and negative allometric growth ($b < 3$) juveniles from Marano Lagoon and adult male and female.

However, comparison between obtained values of *t*-test and the respective critical values evidenced no statistically significant differences with isometric range ($b = 3$) for juveniles from study area, instead evidenced high statistically significant differences with isometric range for the all the other, indicating an allometric range (negative allometric; $b < 3$; Tab. XVIII).

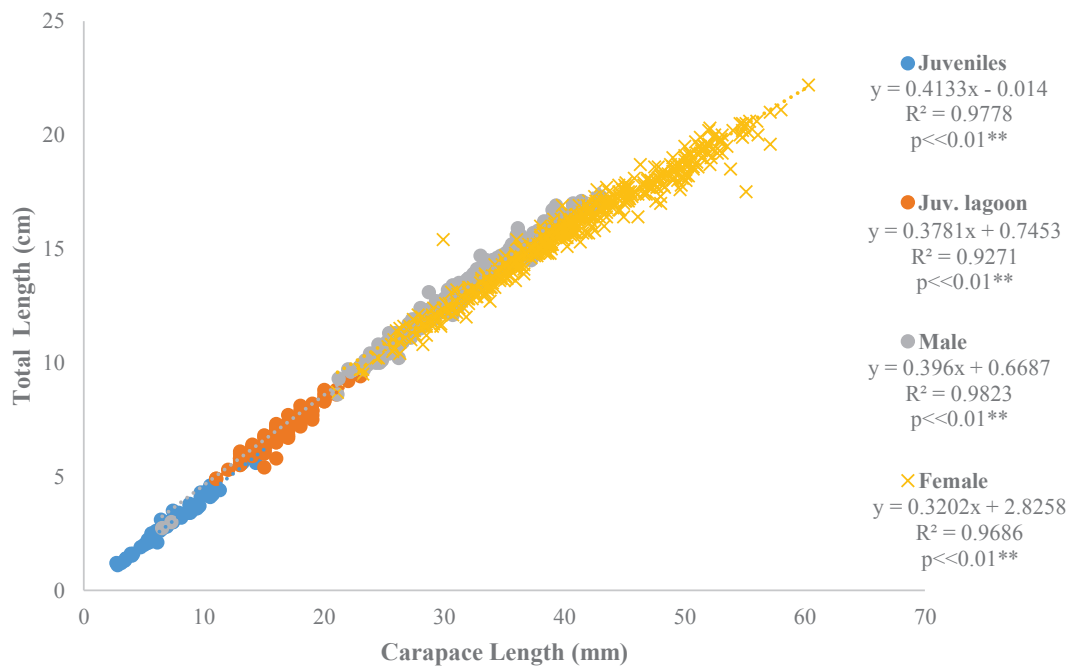
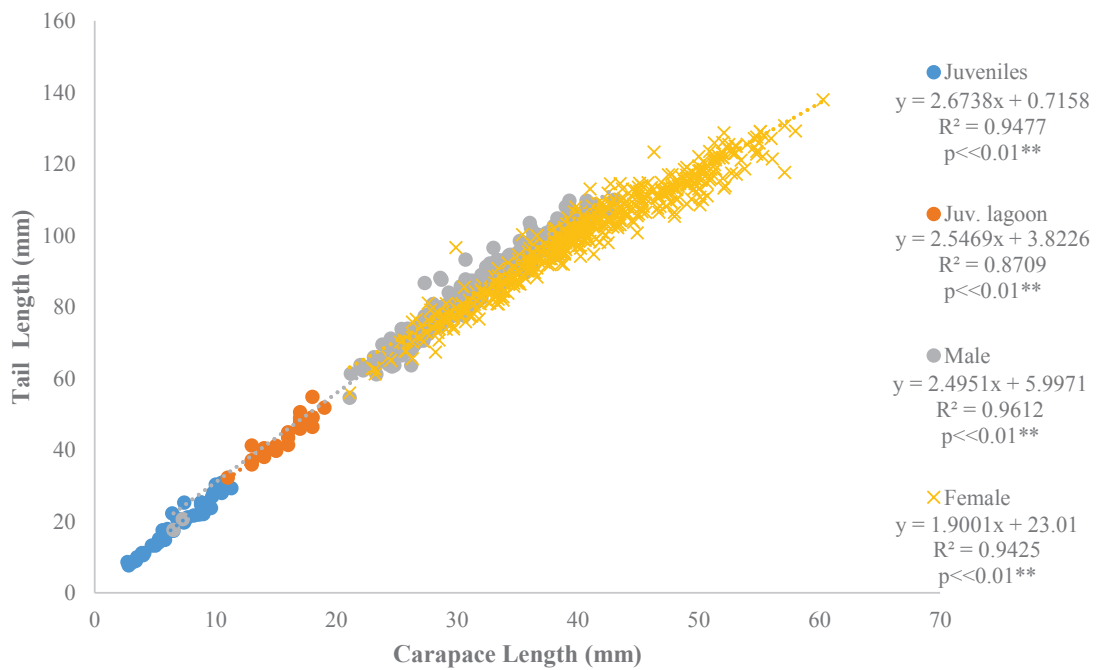


Fig. 67. Correlation and linear regression between carapace length (CL) and tail length (above) and between carapace length (CL) and total length (TL) for juveniles from the study area, from Marano Lagoon and for adult male and female of *P. kerathurus*, and their function, R^2 and p value.

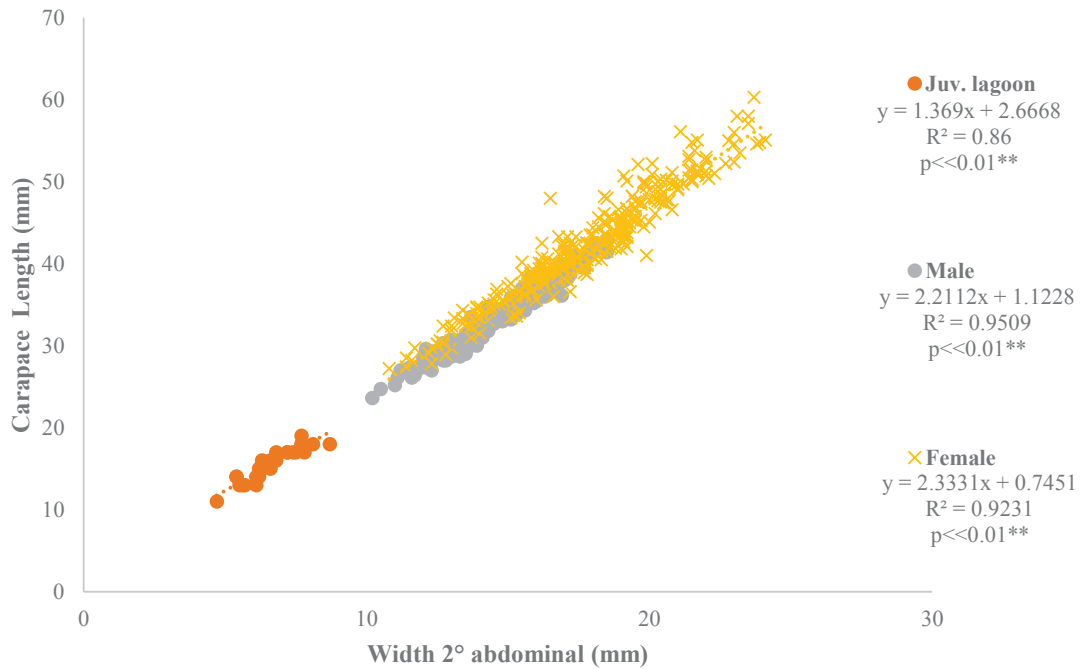


Fig. 68. Correlation and linear regression between width of 2° abdominal segment and carapace length (CL) for juveniles from Marano Lagoon and for adult male and female of *P. kerathurus*, and their function, R^2 and p value.

Also exponential and log-transformed correlation between total length (TL, cm) and weight (g) have highlighted that all regression were highly significant ($p < 0.01^{**}$) and the coefficient R^2 values ranged from 0.987 for male to 0.936 for juveniles from Marano Lagoon (Fig. 70).

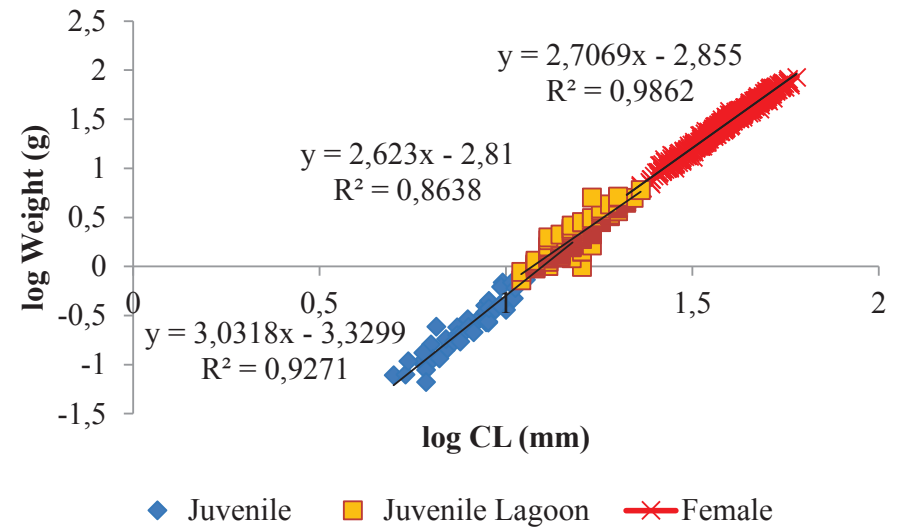
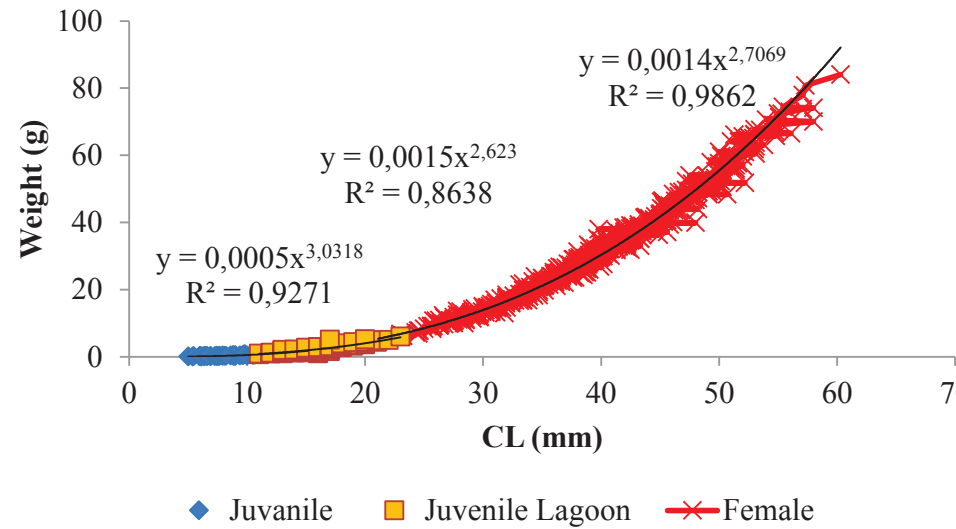
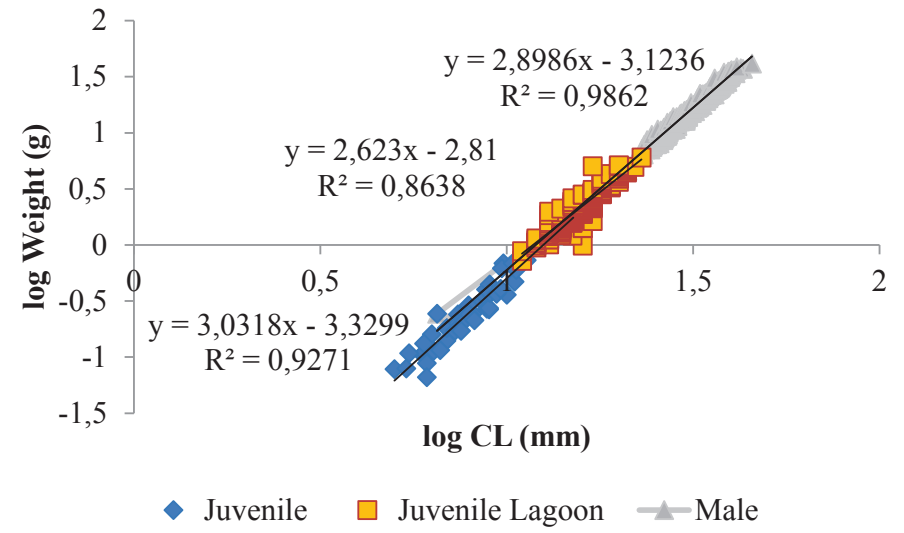
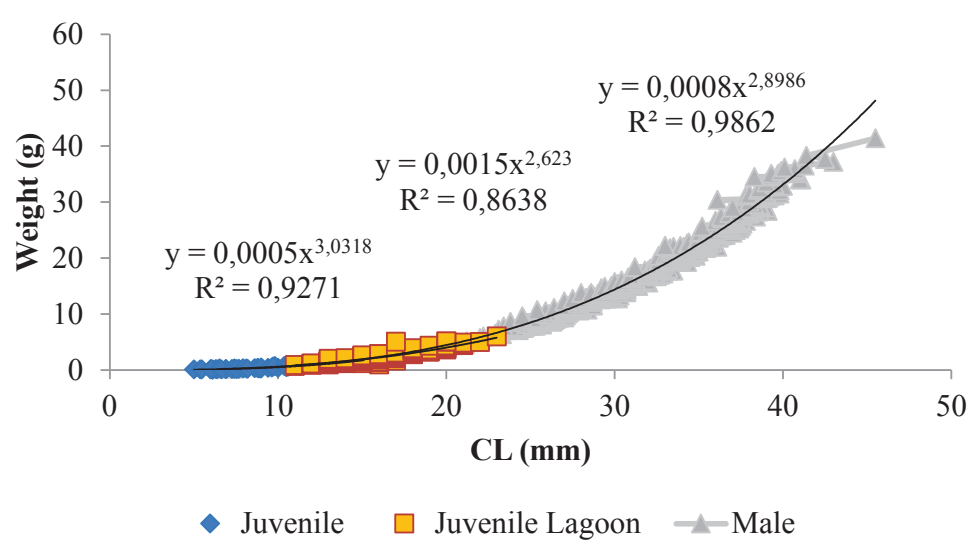


Fig. 69. Correlations between carapace length (CL, mm) and weight (g) and exponential relations (left) and linear relation (right) of log-transformed data, for juveniles from the study area, juveniles from Marano Lagoon, from adult male and female of *P. kerathurus*.

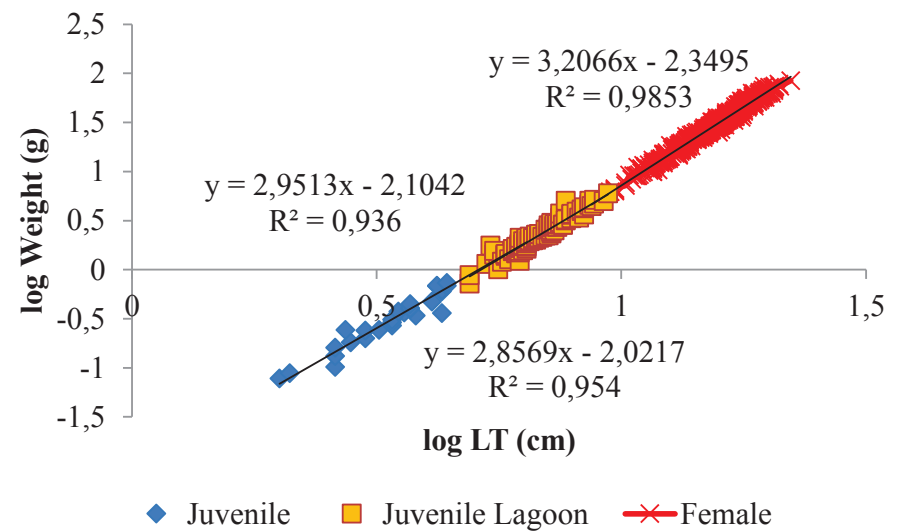
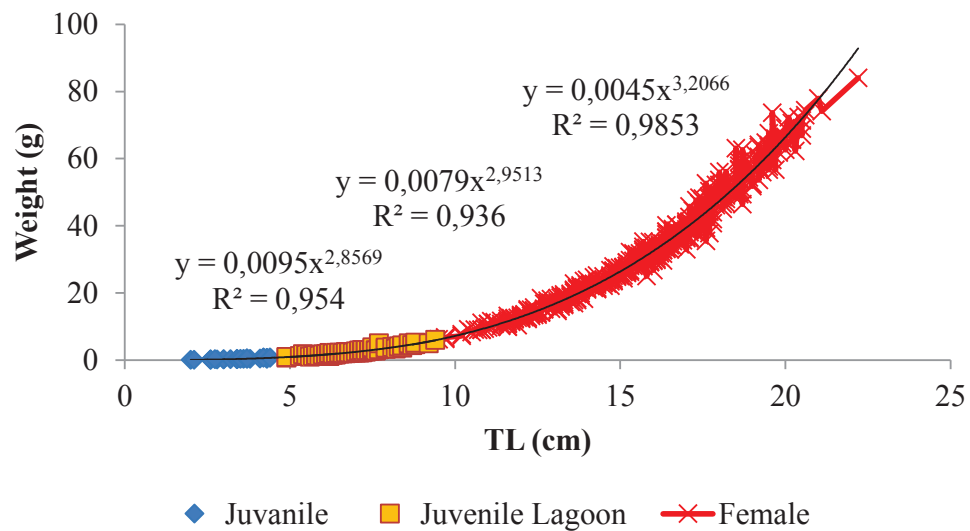
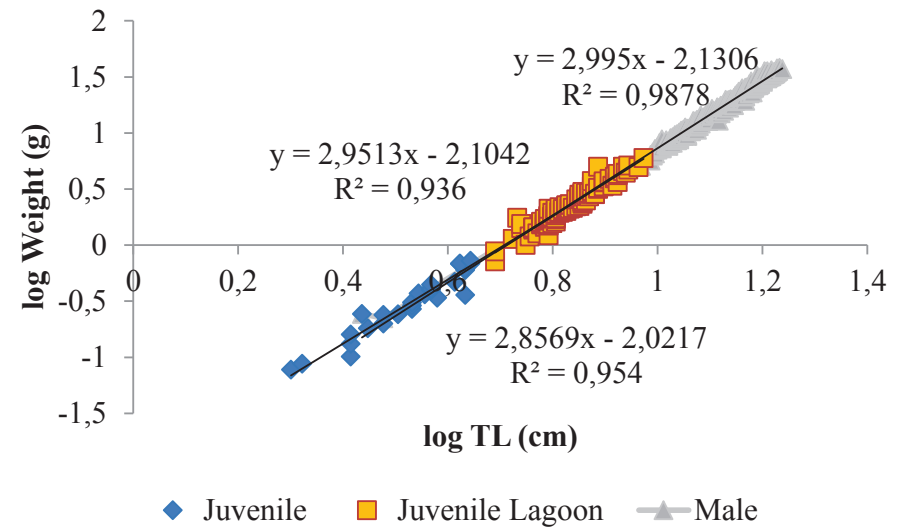
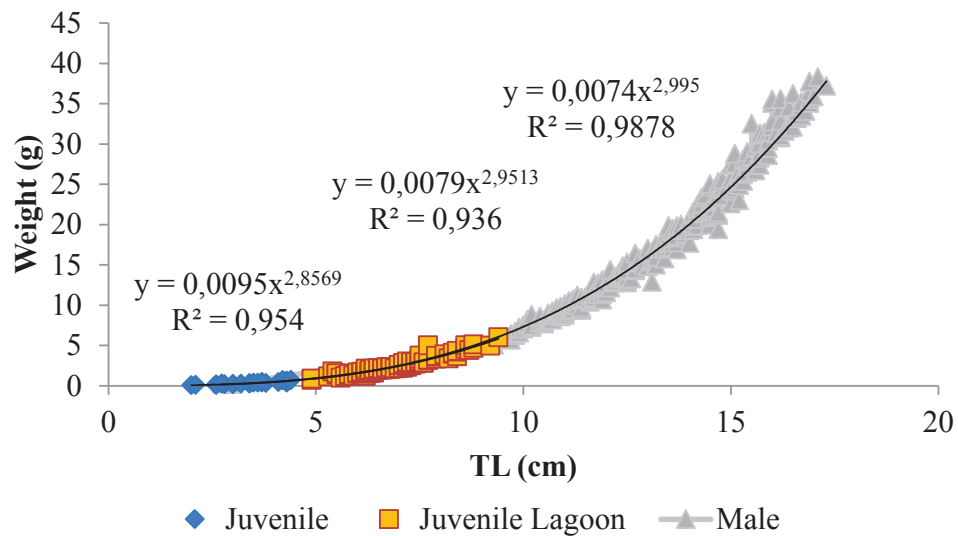


Fig. 70. Correlations between total length (TL, mm) and weight (g) and exponential relations (left) and linear relation (right) of log-transformed data, for juveniles from the study area, juveniles from Marano Lagoon, from adult male and female of *P. keraturnus*.

Tab. XVIII. Obtained b -value \pm standard error (SE), t -test value, degree of freedom, critical value (α) 95% of confidence interval and 99% of confidence interval and p -value (ns: not significant; **= high significant), for juveniles from study area, Marano Lagoon and adult male and female of *P. kerathurus*.

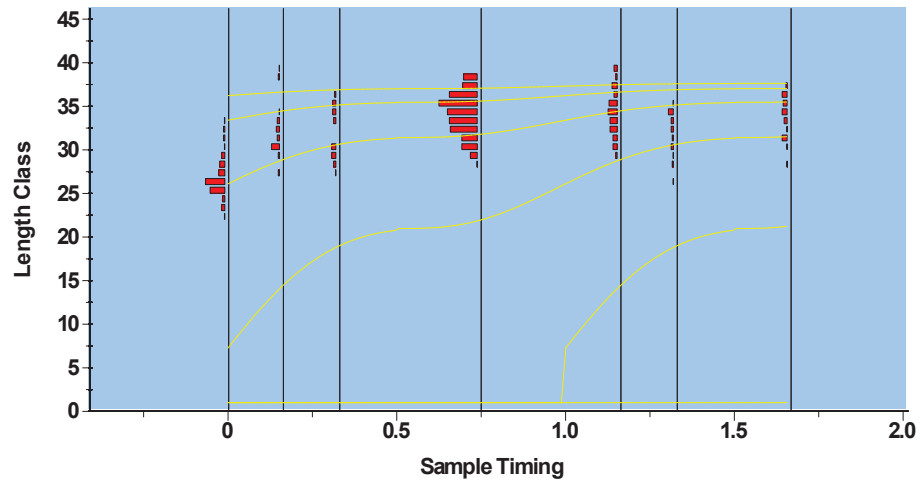
	b	\pm SE	t -test	df	Critical value α		p -value	
					0.05	0.01		
Juvenile	3.031	0.131	0.242	42	2014	2.69	$p > 0.05$	ns
Juvenile Lagoon	2.623	0.064	-5.858	262	1.96	2.57	$p \ll 0.01$	**
Male	2.898	0.016	-6.064	420	1.96	2.57	$p \ll 0.01$	**
Female	2.706	0.013	-21.680	561	1.96	2.57	$p \ll 0.01$	**

5.2.3 Von Bertalanffy Growth Function Parameters

Length-frequency distribution analysis adopted to estimate Von Bertalanffy parameters model with Pauly's function showed for male a value of $L_{\infty} = 38$ mm CL and $K = 0.83 \text{ yr}^{-1}$ ($T_0 = -0.01$ yr; Fig. 71), while showed for females a value of $L_{\infty} = 55$ mm CL and $K = 0.63 \text{ yr}^{-1}$ ($T_0 = -0.42$ yr; Fig. 72).

For juveniles from the study area the non-seasonal model estimated were $L_{\infty} = 9.39$ mm CL and $K = 0.90 \text{ yr}^{-1}$ ($T_0 = -0.80$ yr; Fig. 73).

Growth Curve = PaulySeasonal. Linf = 38,00. K = 0,83. Tzero = -0,01. Ts = 0,07. NGT = -0,14.



Growth Curve = PaulySeasonal. Linf = 38,00. K = 0,83. Tzero = -0,01. Ts = 0,07. NGT = -0,14.

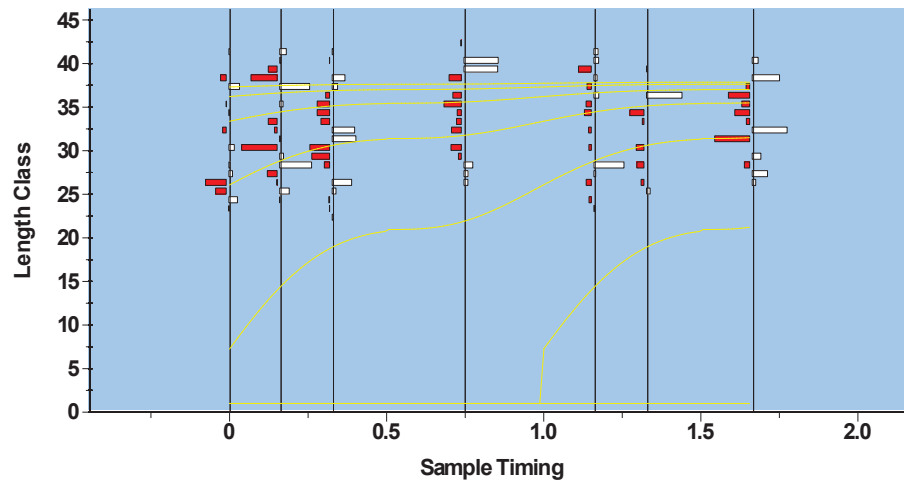
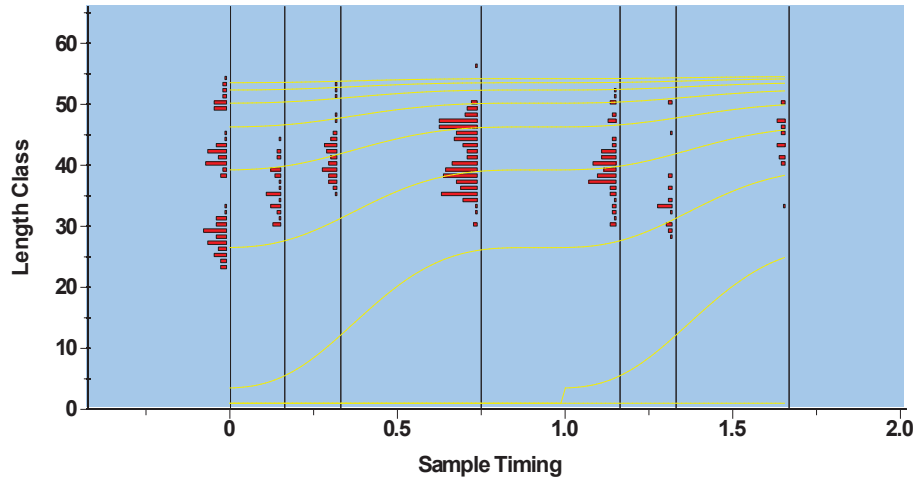


Fig. 71. Growth curve estimated from non-restructured (above) and restructured (below) length-frequency distributions for male of *P. kerathurus* (sample timing: 0= 1st January; 1= 31th December).

Growth Curve = PaulySeasonal. Linf = 55,00. K = 0,63. Tzero = -0,42. Ts = 0,42. NGT = 0,06.



Growth Curve = PaulySeasonal. Linf = 55,00. K = 0,63. Tzero = -0,42. Ts = 0,42. NGT = 0,06.

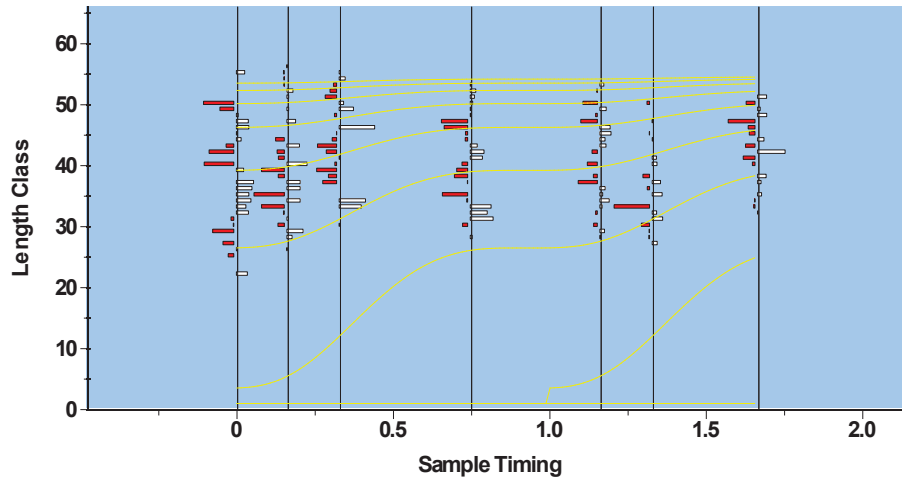
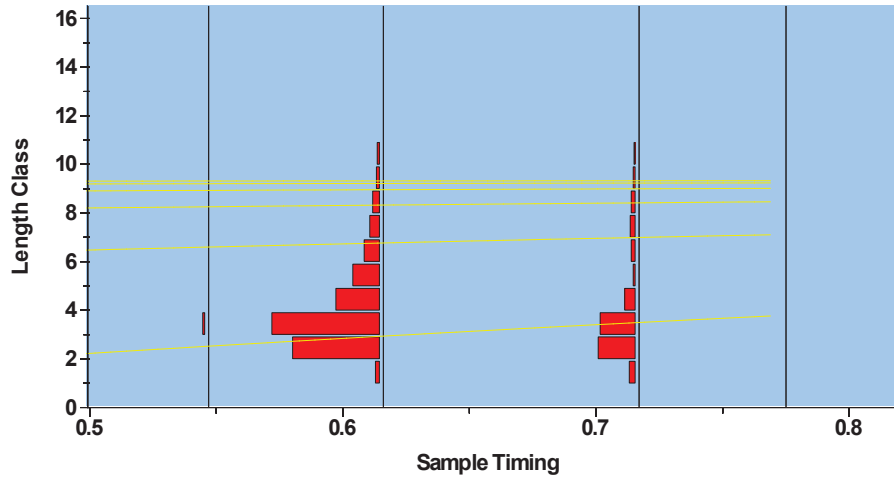


Fig. 72. Growth curve estimated from non-restructured (above) and restructured (below) length-frequency distributions for female of *P. kerathurus* (sample timing: 0= 1st January; 1= 31th December).

Growth Curve = Non Seasonal. Linf = 9,39. K = 0,90. Tzero = -0,80.



Growth Curve = Non Seasonal. Linf = 9,39. K = 0,90. Tzero = -0,80.

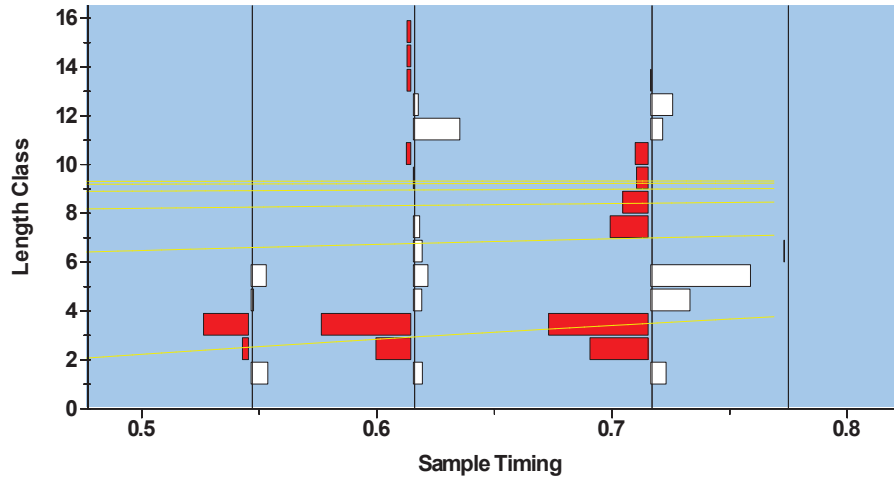


Fig. 73. Growth curve estimated from non-restructured (above) and restructured (below) length-frequency distributions for juveniles from study area of *P. keralurus* (sample timing: 0= 1st January; 1= 31th December).

5.2.4 Total Mortality Rate Z

Based on estimated Von Bertalanffy growth parameters the total mortality rate Z was estimated following the Beverton-Holt model. Z-value calculated each distributions ranged from 0.228 to 1.167 yr⁻¹ for male, with mean value (\pm SE) Z= 0.455 \pm 0.1239 yr⁻¹ (Tab. XIX).

For female Z-values ranged from 0.298 to 0.746 yr⁻¹, with mean value (\pm SE) for the entire observation period of Z= 0.515 \pm 0.0647 yr⁻¹ (Tab. XIX).

Tab. XIX. Total mortality rate Z (yr⁻¹) evaluated for each length-frequency distribution, their mean (\pm standard error), for adul male and female of *P. kerathurus*.

	Total Mortality Rate Z (yr ⁻¹)							Mean	\pm SE
Male	1.167	0.389	0.436	0.228	0.241	0.463	0.263	0.455	0.1239
Female	0.690	0.619	0.374	0.403	0.474	0.746	0.298	0.515	0.0647

5.3 Discussion

Generally, in sampling programs, it is usually easier to measure only size while weight cannot be easily recorded. Length is more relevant than age, mainly because several ecological and physiological factors are more size dependent than age-dependent (Santos et al. 2002). The morphometric relations are a useful tool in fish biology, physiology, ecology, and stock assessment, especially weight-length relationship (LWR). The LWR of a species allows the interconversion of these two parameters. In biological studies LWRs allow the assessment of seasonal variations in fish growth (Richter et al. 2000), which is frequently used in the analysis of ontogenic changes (Safran 1992), for between-region life-history comparisons (Weatherley & Gill 1987; Petrakis & Stergiou 1995) as well as to identify the spawning season (Olim & Borges 2006). In fisheries studies WLRs have many different uses, including the estimation of weight from length (Beyer 1991; Froese 2006; Froese, R. et al. 2011) and of weight-at age (Petrakis & Stergiou 1995), as well as the conversion of growth-in-length equations to growth in-weight (Pauly 1993). Furthermore, WLRs allow life history and morphological comparisons between different fish species, or between fish populations from different habitats and/or regions (Gonçalves et al. 1997).

The present study reports, not only solid morphometric relations of a very wider size range, as between carapace length (CL) – tail length, carapace

length (CL) – total length (TL), width of 2^o abdominal segment - carapace length (CL), but also the WR of *P. kerathurus* species caught in the western portion of northern and central Adriatic Sea.

This LWR comprises, for the first time, a very complete sample of ranged size, from 5 to 60 mm of carapace length, and from 0.07 to 84 g. All regressions were highly significant with high R² values. The estimation of b parameter of LWR highlighted an isometric growth of juveniles from study area (b=3), conversely showed negative allometric growth (b<3) for all the other considered groups (juveniles from Marano Lagoon, adult male and female), as confirmed by statistical analysis. This can be attributed to the very high growth, particularly pronounced during early juvenile phases. The typical growth pattern of penaeid shrimp is characterized by a rapid weight gain during the early life stage and by the achievement of asymptotic length in adults (Franco et al. 2006).

The estimated growth parameters of *P. kerathurus* derived from this study are similar to the value estimated for male and female in the Gulf of Gabès, Tunisia (Ben Mariem 2004). For the first time the growth parameters of juveniles were reported, indicating high growth coefficient K (0.90 yr⁻¹), imputable to the very low time necessary to achieve the estimated asymptotic length (L_∞= 9.39 mm carapace length CL) in this juvenile phase.

The total mortality rate Z shows a great fluctuations both for male and female, with high values (Z= 0.455 yr⁻¹ for male; Z=0.515 yr⁻¹ for female), typically

demonstrated for shorter-lived and faster-growing species such as the caramote prawn.

Sexual growth dimorphism occurs in many species; this phenomenon acquires considerable importance in the case of farmed species, in fact, some studies have been directed to assess the potential of monosex culture, as reported for *Litopenaeus vannamei* by Moss & Hennig (2002).

The mechanism by which females achieve a larger size than males is unknown. Differences of behavior and/or physiology between males and females may reflect in a sexual growth dimorphism (Moss & Hennig 2002), however, as suggested by the same authors, the growth superiority of female over male penaeid shrimp is not result of a competitive advantage in feed acquisition. More research is needed to investigate possible physiological causes of the mechanism of sexual growth dimorphism in penaeid shrimp, which still remains unclear (Moss & Hennig 2002).

The postulation provided by Myers (1978) on dimorphic niche hypothesis upon selection pressures acting only on females argues that larger female size evolved in vespertilionids bats because females (and not males) carry embryos that total 20-30% of their own body weight. There are some studies that reported a positive correlation between female size and egg number or lifetime fecundity (Hedrick & Temeles 1989; Wiklund & Karlsson 1988).

Based on what has been reported, it is quite reasonable to assume that the sexual dimorphism observed in the present study and by other work conducted on *P. kerathurus* that occurs with larger sizes females, is attributable to an evolution process that has led to an increase in reproductive output of the species.

6. CONCLUSION

The presence of *P. kerathurus* in the Adriatic Sea is recent, and the increased catches in the last years have made this species an object of interest.

To understand how the biotic, abiotic and anthropogenic factors can affect the stock of *P. kerathurus* in Northern and Central Adriatic Sea represented the ultimate goal of the study. Studying the biology and ecology of the caramote prawn was the aim of this thesis in order to recognize possible causes of the stock growth in the Adriatic Sea, in contrast with the decreasing trends observed for other stocks already exploited by the commercial fishery. In addition, in the last 50 years a variety of coastal protection structures have been developed, including breakwaters, in order to reduce coastal erosion. Another task was to understand if anthropogenic construction of breakwaters can influence the biological cycle of the selected species, and if they could affect the water temperature. Thus, filling some gaps on its reproductive biology and on the main parameters of the population in the selected area are the objectives of the present thesis.

Within the present study valid information on reproductive biology of *P. kerathurus* were obtained: a size at first sexual maturity was estimated in 40.3 mm of CL for females and 19.7 mm CL for males. While the female values were comparable with those reported in literature, the male values were well below

to whatever so far reported. In addition, presence of spermatophore adhering to *telycum* in female have been strongly connected with seasonality and size of the specimen, indicating summer season as the best period for insemination, and the percentage of inseminated female that exceed 50% were recorded only in summer months. The GSI ranged from 0.8 (December) to 11.2 (July), indicating July as the peak of the total reproductive season (summer, from May to September), as confirmed by histological analysis of ovarian maturation.

For the first time in the Mediterranean Sea, this study reported the lipid content variation in ovaries of *P. kerathurus* during the annual cycle. A statistically significant correlation of some fatty acids with GSI have been identified.

This work highlighted the important physiological changes occurring during reproductive season aimed to sustain the reproductive success, such as the ovarian development related to oocyte changes, not only in color and size, but also due to the substances reserve storage needed for the future offspring.

In addition, during gametogenesis the total lipid content increased, reaching the maximum level observed (8.8% in June), one month before to the gonado somatic peak (July), equivalent to stage IV - mature oocyte.

At this stage there was an increase of saturated fatty acids (SFAs) and mono unsaturated fatty acids (MUFAs) and a decrease of poly unsaturated fatty acids (PUFAs). Two saturated fatty acids (SFAs) such as *margaric acid* (C17) and *arachidic acid* (C20, also called *eicosanoic acid*) have shown statistically

significant trends correlated with GSI, highlighting negative linear relation the first and positive linear relation the second.

In addition, this study showed the particular thermal regime in area protected by breakwater. A broad temperature ranges were shown in the study area than the open sea (respectively -8.4 °C and +6.9 °C the wider recorded temperature differences).

The presence of juveniles of *P. kerathurus* has been demonstrated in the study area (35.9 ± 6.44 ind. 100 m^{-2}) and in a control site 80 km southward the study area during the summer season. It suggest that this “lagoon effect” exerted by breakwater constructions is not limited to the study area but it is reasonable to assume the effect as generalized in a wider area and correlated between water temperature. Comparison of length-frequency distribution of juveniles of caramote prawn collected in study area, control site and natural lagoon in summer 2015, showed very similar length-frequency distribution for study area and control site, instead they showed a certain complementarity with those recorded for natural lagoon, as confirmed by statistical analysis. This can be ascribed to one month shift in the sampling and fact that juveniles, after reaching a certain size, move from the lagoon to offshore water.

Finally, the present study reports, not only solid morphometric relations of a very wider size range, as between carapace length (CL) – tail length, carapace length (CL) – total length (TL), width of 2° abdominal segment - carapace

length (CL), but also the WR of *P. kerathurus* species caught in the western portion of northern and central Adriatic Sea.

This LWR comprises, sample of ranged size from 5 to 60 mm CL and from 0.07 to 84 g. All regressions were highly significant with high R^2 values. The estimation of b parameter of LWR highlighted an isometric growth of juveniles from study area ($b=3$), conversely showed negative allometric growth ($b<3$) for all the other considered groups (juveniles from Marano Lagoon, adult male and female), as confirmed by statistical analysis. High growth coefficient K (0.90 yr^{-1}) of the juveniles growth parameters were reported, imputable to the very low time necessary to achieve the estimated asymptotic length ($L_{\infty}= 9.39$ mm carapace length CL).

The total mortality rate Z shows a high values ($Z= 0.455 \text{ yr}^{-1}$ for male; $Z=0.515 \text{ yr}^{-1}$ for female), typically demonstrated for shorter-lived and faster-growing species such as the caramote prawn.

The mechanism by which females achieve a larger size than males is unknown. Myers (1978) postulated on dimorphic niche hypothesis upon selection pressures acting only on females, and argues that larger female size evolved in females (and not males) because for some species females carry embryos that reached 20-30% of their own body weight.

Based on what has been reported, it is quite reasonable to assume that the sexual dimorphism observed in the present study and by literature on *P. kerathurus* that

occurs with larger sizes females, is attributable to an evolution process that has led to an increase in reproductive output of the species, as well as there are some studies that reported a positive correlation between female size and egg number or lifetime fecundity (Hedrick & Temeles 1989; Wiklund & Karlsson 1988).

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