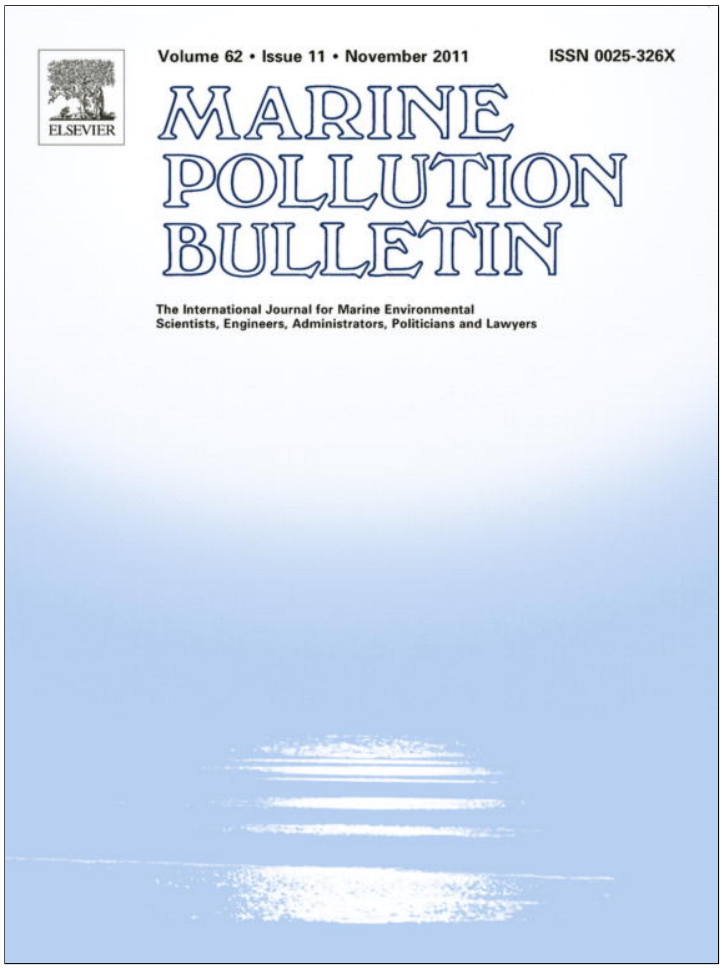
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Baseline

Temporal and spatial biomonitoring of heavy metals in eastern Aegean coastal waters using *Amphibalanus amphitrite*

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ABSTRACT

This biomonitoring study presents the spatial and temporal distributions of heavy metals in the soft tissues of a major fouling species *Amphibalanus amphitrite* living on hard substrate at different sites along the eastern Aegean coast. *A. amphitrite* has been chosen as a strong candidate for monitoring heavy metals. Sediment and seawater samples were also collected to detect their metal contents in order to gain more information on the environmental conditions and possible bioaccumulation patterns. The physico-chemical characteristics of sampling stations have been measured in order to characterize the sampling area. The order of metal concentrations in barnacles, sediment and seawater decreased in the following order Cu > Fe > Zn > Mn > Cd > Cr > Pb > Hg, Fe > Mn > Zn > Cu > Pb > Cr > Hg > Cd and Fe > Zn > Mn > Cu > Pb > Cr > Cd > Hg, respectively. These results showed that barnacles accumulate Cu in a higher degree than both sediment and seawater. Moreover, metal concentrations in barnacle have the potential for use in any future regulatory framework monitoring and eventually controlling ambient metal pollution levels.

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Chemical contamination of marine environment is a world wide problem, but it is particularly serious along the coasts of industrialized countries, where wastes from a number of human activities reach the sea. Marine environments are impacted more and more by anthropogenic activities resulting in pollution of marine sediments by persistent pollutants especially the heavy metals (Barbieri et al., 1999).

Heavy metals, pesticides and other toxic substances can be absorbed from the water column onto surfaces of fine particles and they move with the sediments. They participate in various biogeochemical mechanisms and can affect the ecosystems through bioaccumulation and biomagnification processes (GESAMP/UNESCO, 1994; Salomons and Förstner, 1984). Some of these heavy metals can represent a threat to marine life and possibly to man as a consumer of seafood.

Heavy metals are not fixed permanently in the sediment. They can be transported between sediment and water column. Once at the sediment–water interface or in the water column, metals are more likely to be transported and to enter the food web. Metals of anthropogenic origin are more loosely bound in sediments and thus are more readily available to organisms (Schropp and Windrom, 1988). The study of metal concentrations in organisms can give more reliable information on the bioavailability and consequently of the potential damage to the organism's metabolism and to the environment in general (Bryan et al., 1980). The levels of heavy metals accumulated by marine organisms are function not only of water quality, but also of seasonal factors, temperature, salinity, diet, spawning and individual variation, among other factors (Chan, 1989).

The relative contamination of coastal environments can be measured by the use of suitable biomonitors. The use of selected organisms as biomonitors of trace metal bioavailability in coastal waters facilitates comparison over space and time (Rainbow and Blackmore, 2001). There are several well-established biomonitors of contaminant trace metals in coastal environments, for which the accumulation patterns and kinetics are well known (Rainbow and Blackmore, 2001). Especially mussels and barnacles, key components of the local sublittoral benthos, are used to address the question of metal bioavailabilities. Barnacles are considered excellent metal biomonitors. Laboratory research has shown that barnacles are net accumulators of metals (Rainbow and White, 1989). It is possible to assume that higher metal bioavailability in seawater leads to greater accumulation in barnacles. Therefore, in the locations where barnacles represented with high metal levels, the waters should contain relatively elevated bioavailability (Páez-Osuna et al., 1999).

A number of studies have been carried out on the concentrations of heavy metals in sediment and biota from different sites of Aegean Sea (Kucuksezgin et al., 2001, 2002, 2006; Dassenakis et al., 2003; Zeri and Voutsinou-Taliadouri, 2003; Gonul and Kucuksezgin, 2007). To the best of our knowledge following a literature survey, no previous monitoring of metal contaminants has been carried out in barnacle species from the Aegean coast. The

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main aim of this study was to monitor the spatial and temporal variations in the bioavailability of heavy metals in four different sites in eastern Aegean, using the barnacle *Amphibalanus amphitrite* as biomonitor.

The Aegean Sea situated between Turkey and Greece and constitutes one of the main parts of the Eastern Mediterranean Sea. The study area includes four different sites; Candarli, Izmir-Levent Marina, Izmir-Pasaport, Kusadasi-Setur Marina (Fig. 1) and extends from Candarli in the north to Kusadasi in the southeastern Aegean Sea.

Candarli Gulf is located between 38°55′55″N latitude and 26°56′55″E longitude in the eastern Aegean coast. It has been strongly affected by growing population and industrialization. Great industry settlements, located in the coastal area of Candarli, have been discharging their solid and liquid wastes into Bakircay or Candarli Gulf after limited treatment. Izmir Gulf is one of the great natural inlets of the Mediterranean. The gulf is divided conventionally into three zones: inner, middle and outer by virtue of topographical, hydrological and ecological features. Industrial activities cover a large range of industries including food processing, leather tanning, paint, chemicals, textile manufacture and petroleum refining. Two stations, Levent Marina and Pasaport located in the Izmir Gulf (Fig. 1). Izmir Levent Marina 38°24′24″N, 27°04′09″E and Kusadasi-Setur Marina is located between longitudes 27°15′30″E, latitude of 37°52′00″N, respectively. The maximum depths are 5 m for two marinas. At Kusadasi-Setur Marina, all kinds of painting and maintenance, varnishing, epoxy-polyester works, boat covering, spray hood manufacture and repair, all kinds of main machinery, generator and outboard repair, plane, stainless metal and galvanizing works, electric and electronic equipment repair, bilge control, mounting and maintenance are available.

A. amphitrite were collected seasonally from the sampling sites along the eastern Aegean coast, during 1 June 2008 to 08 June 2009. Sixty organisms of the different size (rostro-carinal axis) were selected and collected from the intertidal zone at each station; they were removed from the substratum with a stainless steel scraper and placed in individual clean polyethylene bags then transported in closed, refrigerated containers and frozen at −20 °C.

Water samples were collected seasonally from sampling sites, using 1 l acid-leached polyethylene bottles. Sediment samples were taken by using Van-Veen Grab from surface sediments and they were dried in oven at 40 °C, homogenized and reduced to a fine powder by using a sieve (63 μm). Physicochemical parameters of water quality (conductivity, pH, salinity and temperature) were measured *in situ* with WTW Portable instrument and dissolved oxygen was determined using (DO) Winkler method (Methods of Seawater Analysis, 1983).

In the laboratory, identification of individuals was performed by direct examination of morphological characteristics considering Southward (2008). Their rostro-carinal axes were measured and classified different size groups. Then barnacles were defrosted, their bodies were dissected out with a stainless steel scraper and the soft parts were separated. The samples were dried to constant weight at 40 °C and weighed (Rainbow and Blackmore, 2001).

The barnacle samples were digested in a microwave digestion system because of the advantages of this technique, which include speed of digestion and less possibility of contamination during the process. A portion of the dry, finely powdered solid barnacle tissues (0.5–1 g) were accurately weighed and then digested with 5:1 $HNO_3/HClO_4$ in a microwave oven. Sediment samples were digested in microwave digestion system with a HNO_3–HF–$HClO_4$–HCl acid mixture solutions. Ion exchange technique, using Chelex-100 resin was applied for determining heavy metals in acidified water samples. All analyses were performed by flame, graphite furnace and cold vapour technique in AAS (Varian Spectraa-300 plus) by using the manufacturer's conditions and with background correction

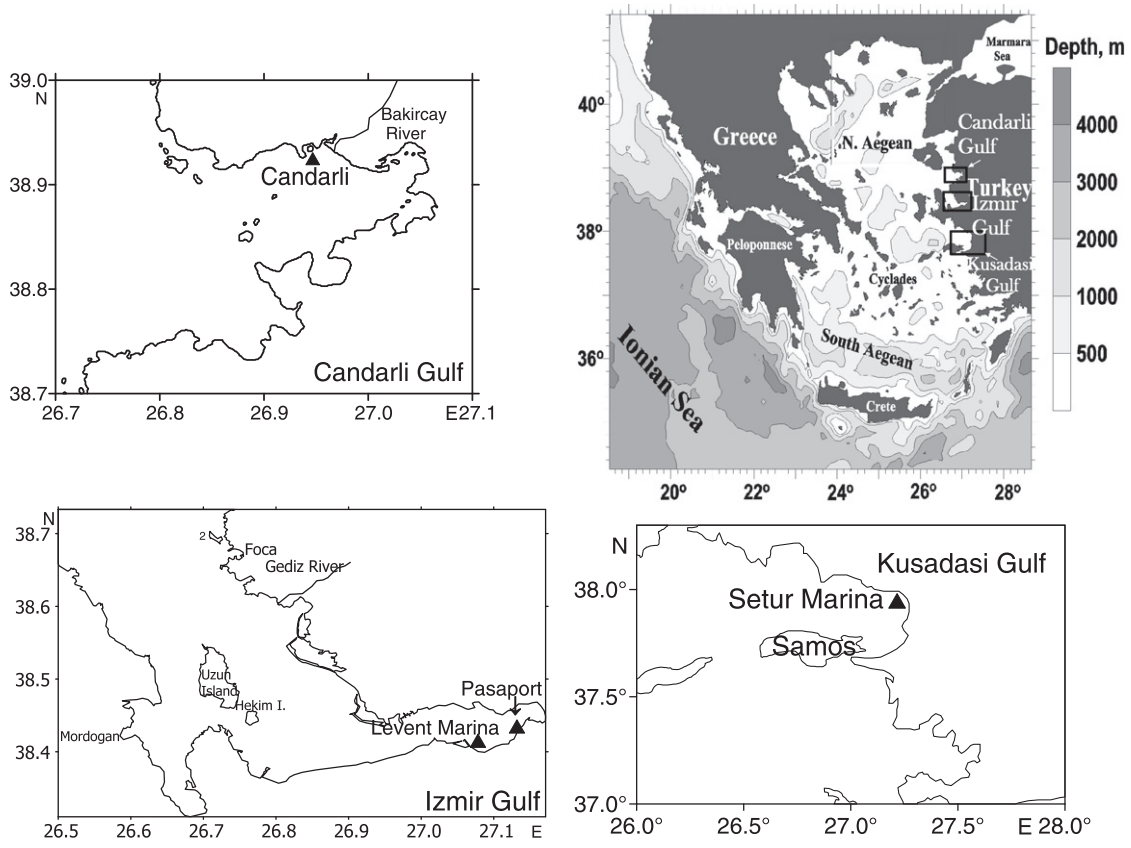


Fig. 1. The location of sampling points in the Aegean Sea.

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(UNEP, 1982, 1985). The detection limits for trace metals were: Hg: 0.05 μg kg^{-1}, Cd: 0.10 μg kg^{-1}, Pb: 0.10 μg kg^{-1}, Cr: 0.06 μg g^{-1}, Cu: 0.03 μg g^{-1}, Zn: 0.01 μg g^{-1}, Mn: 0.10 μg g^{-1}.

Mercury concentration was measured by anodic stripping voltammeter at the rotating gold electrode in seawater. With a deposition time of 90 s the calibration curve is linear from 0.4 to 15 μg l^{-1}; the limit of quantification was 0.4 μg l^{-1}. After appropriate digestion the determination of mercury is possible even in samples with a high load of organic substances. The concentration in the sample is determined by standard addition (METROHM Application Bulletin 96/5e, Determination of mercury at the rotating gold electrode by anodic stripping voltammetry).

Accuracy and validity of the analytical processes were tested with a reference material (homogenate muscle sample, IAEA-407, from the International Laboratory of Marine Radioactivity, IAEA, Monaco). The values obtained from the analysis of three replicates of this sample (certified: observed values in μg g^{-1} dry wt. ± standard deviation) were as follows: Hg, 0.22 ± 0.024:0.21 ± 0.0015; Cd, 0.19 ± 0.019:0.18 ± 0.004; Pb, 0.12 ± 0.06:0.10 ± 0.012; Cr, 0.73 ± 0.22:0.74 ± 0.062; Cu, 3.28 ± 0.40:3.39 ± 0.36; and Zn, 67.1 ± 3.8:69.4 ± 1.3.

Intercalibration sediment (IAEA-433) sample (from the IAEA) were used as a control for the analytical methods. The values obtained (in μg g^{-1} dry wt) for the analysis of six replicates of this sample were as follows: Hg (certified 0.168, s.d: 0.017; found 0.167, s.d: 0.012), Cd (certified 0.153, s.d: 0.033; found 0.140, s.d: 0.025), Cr (certified 136, s.d: 10; found 136.7, s.d: 2.0), Cu (certified 30.8, s.d: 2.6; found 30.8, s.d: 2.3), Pb (certified 26.0, s.d: 2.7; found 27.0, s.d: 3.1), Zn (certified 101, s.d: 8.0; found 103, s.d: 1.9), Mn (certified 316, s.d: 16; found 317, s.d: 1.7), Fe (certified 40800, s.d: 1900; found 40412, s.d: 293).

Statistical analysis was performed using the STATISTICA 5.1 (Stat. Soft. Inc. 1998, Tulsa). Main effect ANOVA was applied on log transformed metal data in sediment and seawater samples and was used to compare the heavy metal concentrations among seasons and sampling stations. Analysis of covariance (ANCOVA) was used to detect differences among the sites for each of the sampling seasons and the temporal differences at the sampling sites in terms of accumulated body metal burdens. In the data analysis, metal concentrations and season were considered as the dependent and independent variable, respectively, whereas mean weight was selected as the covariate. The data set was normalized using double log transformation [log body weight (x) against log metal concentration (y)] before ANCOVA test. Homogeneity of variances was examined using the Cochran-C test of heavy metal contents in samples. Spearman Rank Order Correlation test used to check for significant relationships of heavy metals in barnacle, sediment and seawater. Pearson product moment correlation test was applied to determine the relationship between physicochemical parameters and metal contents of seawater, sediment and barnacle.

Metal concentrations in the bodies of *A. amphitrite* showed significant size effect (Rainbow et al., 1993). Thus accumulated concentrations in barnacle are modelled by the power function:

$$y = ax^b$$

where y is the metal concentration (μg g^{-1}), x is the mean body dry weight (g) and a and b are constants. All data were transformed logarithmically (to the based 10) to reduce deviations of the data sets from normal distributions, creating an additive data set with skews removed.

The physico-chemical characteristics of water column have been analyzed to find possible correlations between metal concentrations and some of these parameters such as pH, salinity, conductivity, temperature and dissolved oxygen (DO) and the results were given in Table 1. The pH of the aquatic system is an important indicator of water quality and the extent of the pollution in the watershed areas. A pH range of 6.5–8.5 is normally acceptable as per guideline suggested by WHO (1993). The measured pH values (7.98–8.52) indicated moderately alkaline nature of the seawater for the sampling area. The lowest temperature value was measured as 11.9 °C at Levent Marina during winter period while the highest temperature (30.0 °C) was found at the Candarli during summer period.

Conductivity reflects the status of major ions derived from inorganic pollution and is a measure of total dissolved solids and ionized species in the water column. There were no big differences in conductivity values ranged between 57.10 and 60.70 mSi cm^{-1} at sampling stations. The lowest salinity value was measured 37.3 psu at Izmir-Levent Marina in spring while the highest value was recorded at Izmir-Pasaport (40.8 psu) in summer period.

DO concentrations at the stations varied from 3.20 to 9.40 mg l^{-1}. During the cold period DO represented with higher values considering the other periods. In this period the highest DO concentration (9.4 mg l^{-1}) was observed at the Izmir-Pasaport where the significant decrease was determined at the same station for the summer period.

Table 2 summarizes the results for the metal concentrations of water samples during sampling periods. Mercury concentrations presented with the higher values at all stations in the summer period and the highest and lowest mean values were recorded in Izmir-Pasaport (0.017 μg l^{-1}) and Candarli (0.012 μg l^{-1}), respectively. Significant temporal changes of mercury were observed at all sampling stations (Table 3).

The mean cadmium concentration in seawater showed slight fluctuations from a minimum mean value of 0.17 μg l^{-1} at Izmir-Levent Marina and maximum mean value 0.24 μg l^{-1} at Izmir-Pasaport. Moreover, seasonal and spatial patterns in Cd level were not evidenced clearly (Table 3).

Concerning the highest mean level, Pb was found in Kusadasi-Setur Marina (1.8 μg l^{-1}) and the lowest mean value was measured (1.05 μg l^{-1}) in Izmir-Levent Marina. Higher Pb and Cr values at all sampling stations were recorded in summer period. Moreover, the highest mean level for chromium was obtained from Candarli (1.2 μg l^{-1}), while the lowest mean concentration was measured at Levent Marina (0.77 μg l^{-1}). Seasonal differences in the concentrations of two metals were significant (Table 3).

The highest Cu concentration (8.14 μg l^{-1}) was determined in Kusadasi-Setur Marina in the summer period. However, no consistent seasonal fluctuations were observed. The lowest mean concentration of copper was recorded at Izmir-Pasaport in spring period (0.83 μg l^{-1}) while the highest mean was found in Kusadasi-Setur Marina (4.85 μg l^{-1}). Temporal pattern was not statistically significant for Cu (Table 3).

No significant temporal and spatial differences were found for Zn, Fe and Mn in seawater along the eastern Aegean coast. However the highest (8.5 μg l^{-1}) and the lowest (3.7 μg l^{-1}) mean value for zinc was determined at Kusadasi-Setur Marina. The minimum Zn value (1.6 μg l^{-1}) was detected in Izmir-Pasaport and maximum level (12 μg l^{-1}) was measured in Kusadasi-Setur Marina, both of them were recorded during spring period. Concerning zinc and iron, the concentrations showed an obvious increase in the selected stations from northern to southern part of the Aegean coast. Manganese concentration (9.7 μg l^{-1}) peaked only at Candarli in winter period. Concentrations of Mn reached the highest mean level in Candarli (4.5 μg l^{-1}) and the lowest mean (1.4 μg l^{-1}) level in Kusadasi-Setur Marina.

Generally, the elevated metal concentrations in different seasons were determined in Kusadasi-Setur Marina, Candarli and Izmir-Pasaport where the source of metals could be derived from not only marinas but also industrial or domestic pollution. The

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Table 1
Physicochemical parameters.

Site	Period	DO (mg l^{-1})	Salinity (psu)	Conductivity mSi cm^{-1}	pH	Temperature (°C)
Candarli	Summer	6.5	39.8	59.00	8.30	25.1
	Autumn	6.9	39.7	59.60	8.38	20.2
	Winter	7.9	38.7	58.60	8.47	16.7
	Spring	7.1	39.5	59.40	8.19	22.7
Izmir-Pasaport	Summer	3.2	40.8	60.70	7.98	26.0
	Autumn	5.6	39.5	59.90	8.18	16.6
	Winter	9.4	38.2	57.20	8.52	13.6
	Spring	6.9	38.1	57.10	8.47	25.1
Izmir-Levent Marina	Summer	4.7	39.2	58.70	8.24	24.0
	Autumn	7.3	39.9	60.00	8.32	19.8
	Winter	8.1	38.8	59.30	8.38	11.9
	Spring	6.8	37.3	58.10	8.37	24.6
Kusadasi-Setur Marina	Summer	6.7	39.6	59.10	8.27	25.2
	Autumn	7.0	38.2	57.70	8.19	20.7
	Winter	8.6	38.1	57.90	8.39	16.8
	Spring	6.8	38.2	57.80	8.09	22.8

Table 2
The mean ± SE and minimum–maximum concentrations of the heavy metals in seawater during sampling periods (μg l^{-1}).

	Candarli	Izmir-Pasaport	Izmir-Levent M.	Kusadasi-Setur M.
Hg	0.012 ± 0.001	0.017 ± 0.005	0.016 ± 0.005	0.013 ± 0.002
	0.010–0.013	0.011–0.033	0.010–0.032	0.012–0.017
Cd	0.24 ± 0.004	0.24 ± 0.020	0.17 ± 0.040	0.20 ± 0.040
	0.23–0.25	0.19–0.30	0.13–0.22	0.83–0.25
Pb	1.5 ± 0.36	1.4 ± 0.29	1.1 ± 0.29	1.8 ± 0.85
	1.0–2.5	0.91–2.2	0.66–1.9	0.53–4.3
Cr	1.2 ± 0.71	0.98 ± 0.43	0.77 ± 0.48	1.0 ± 0.70
	0.37–3.3	0.44–2.4	0.25–2.1	0.19–3.1
Cu	3.0 ± 0.85	0.83 ± 0.36	1.4 ± 0.15	4.9 ± 1.45
	1.0–4.9	0.50–1.2	0.84–2.4	2.2–8.1
Mn	4.5 ± 1.80	2.4 ± 0.29	2.2 ± 0.47	1.4 ± 0.38
	2.0–9.7	1.4–3.7	1.8–3.0	0.56–2.2
Zn	3.7 ± 0.88	4.8 ± 2.24	5.4 ± 1.61	8.5 ± 1.25
	2.2–6.0	1.6–8.2	1.8–11	5.9–12
Fe	6.9 ± 1.61	8.9 ± 2.60	17.3 ± 6.31	21.5 ± 8.90
	3.3–9.8	3.9–16	4.6–35	7.1–45

Table 3
Values of main effects ANOVA in water column.

Water Column	Season			Station		
	df	*F*	*p* level	*df*	*F*	*p* level
Hg	3	6.920	*	3	0.911	ns
Cd	3	0.238	ns	3	1.019	ns
Pb	3	9.845	**	3	1.090	ns
Cr	3	40.920	***	3	1.457	ns
Cu	3	0.248	ns	3	5.505	*
Mn	3	0.829	ns	3	2.774	ns
Zn	3	1.046	ns	3	1.473	ns
Fe	3	0.518	ns	3	1.449	ns

* $p < 0.05$.
** $p < 0.01$.
*** $p < 0.001$.

mean metal concentrations in seawater generally decreased in the following order: Fe > Zn > Mn > Cu > Pb > Cr > Cd > Hg in the Aegean coast (Table 2).

Spearman's rank correlation analysis was performed to evaluate the relationship between metal concentrations in seawater. This analysis demonstrated that there was a positive correlation ($p < 0.05$) between metal pairs; Pb with Hg ($R = 0.694$), Cd ($R = 0.507$), Cr ($R = 0.790$) and Cr with Hg ($R = 0.647$), Cd ($R = 0.569$) and Zn with Cu ($R = 0.565$), Fe ($R = 0.693$) and Hg with Fe ($R = 0.582$).

The distributions of Hg and Cu levels in seawater were similar with the data obtained from Kalloni Bay in eastern Aegean Sea (Gavriil and Angilidis, 2005) and Algericas Bay, Spain (Morillo and Usero, 2008). However, Hg, Cd, Pb values in seawater samples were lower than the results obtained by Akcali and Kucuksezgin (2011). Cu values were lower than the results obtained from Huvelva Estuary (Morillo and Usero, 2008) and from Naples Harbour-Italy (Adamo et al., 2005).

The mean ± SE and minimum–maximum concentrations of heavy metals as dry weight (dry wt.) in sediment samples were given in Table 4. Hg did not show significant temporal variation (Table 5). However, spatial variation was important and the highest mercury level which was observed at Levent Marina was approximately 20 times higher than the lowest value recorded at Kusadasi-Setur Marina.

The highest mean Cd value (1.9 μg g^{-1}) was recorded with maximum values in all seasons at the Izmir-Pasaport whereas the lowest Cd concentrations were found (0.11 μg g^{-1}) at Kusadasi-Setur Marina. Apart from Izmir-Pasaport, all stations showed a similar distribution trend with low concentration of Cd. The concentrations of cadmium showed significant variation among stations (Table 5).

The lead concentration of sediment samples from Izmir-Pasaport represented with higher values in all seasons with regarding to other stations. In that case, Izmir-Pasaport showed apparent spatial

Table 4
The mean ± SE and minimum–maximum concentrations of the heavy metals in sediments during sampling periods (μg g^{-1} dry wt.).

	Candarli	Izmir-Pasaport	Izmir-Levent M.	Kusadasi-Setur M.
Hg	1.5 ± 0.25	2.4 ± 0.20	2.0 ± 0.80	0.37 ± 0.08
	0.87–2.1	2.0–2.8	0.71–4.3	0.22–0.55
Cd	0.17 ± 0.025	1.9 ± 0.26	0.27 ± 0.059	0.11 ± 0.010
	0.11–0.22	1.2–2.3	0.19–0.44	0.084–0.13
Pb	85 ± 14	335 ± 78	89 ± 7.0	29 ± 1.4
	44–104	252–570	68–100	26–32
Cr	75 ± 3.3	164 ± 9.2	116 ± 1.6	38 ± 5.8
	69–84	146–190	114–120	23–52
Cu	256 ± 25	279 ± 22	272 ± 20	59 ± 11.6
	189–308	239–343	219–316	28–85
Mn	284 ± 18	392 ± 6.2	362 ± 4.7	354 ± 21
	256–335	374–400	352–375	295–390
Zn	199 ± 8.8	620 ± 28	336 ± 27	84 ± 19
	181–215	557–681	289–387	48–117
Fe	26,149 ± 2859	27,875 ± 1635	31,246 ± 631	9953 ± 1461
	18,074–31,253	25,038–31,861	29,645–32,550	6537–12,864

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Table 5
Values of main effects of ANOVA in sediment.

Sediment	Season			Station		
	df	*F*	*p level*	*df*	*F*	*p level*
Hg	3	2.26	ns	3	16.45	***
Cd	3	1.06	ns	3	65.01	***
Pb	3	0.81	ns	3	40.56	***
Cr	3	1.11	ns	3	53.47	***
Cu	3	0.60	ns	3	29.71	***
Mn	3	1.36	ns	3	10.64	**
Zn	3	0.40	ns	3	38.77	***
Fe	3	0.57	ns	3	24.67	***

ns: non significant.
* $p < 0.05$.
** $p < 0.01$.
*** $p < 0.001$.

patterns for the sediment samples. The mean lead value was extremely higher in Izmir-Pasaport (335 $\mu g\ g^{-1}$) where the value approximately 10 times greater than the lowest mean value found at Kusadasi-Setur Marina (29 $\mu g\ g^{-1}$).

The spatial variation of Cr levels in sediment was significant while no significant temporal variation was observed for this metal. The higher mean values were measured at two stations from Izmir-Pasaport (164 $\mu g\ g^{-1}$) and Izmir-Levent Marina (116 $\mu g\ g^{-1}$) located in Izmir Inner Bay, while the lowest mean chromium value was measured at Kusadasi-Setur Marina (38 $\mu g\ g^{-1}$).

Mean level of copper in sediment samples were nearly similar at all stations except Kusadasi-Setur Marina where maxima and minima concentrations were recorded in autumn (85 $\mu g\ g^{-1}$) and in winter (28.27 $\mu g\ g^{-1}$), respectively. There were no clear temporal patterns for Cu concentration, furthermore, a significant spatial difference was observed in sediment samples. Moreover, higher mean values were obtained from Izmir-Pasaport (279 $\mu g\ g^{-1}$), Izmir-Levent Marina (272 $\mu g\ g^{-1}$) and Candarli (256 $\mu g\ g^{-1}$), whereas the lowest level was detected in Kusadasi-Setur Marina (59 $\mu g\ g^{-1}$).

The Zn values (681 $\mu g\ g^{-1}$) in summer were found higher at Izmir-Pasaport corresponding to the areas closest to the main urban localities. Zn showed significant variation among stations and the main source of the variation derived from the values measured in Izmir-Pasaport. As a result, higher mean values were obtained from Izmir-Pasaport (620 $\mu g\ g^{-1}$) and the lowest at Kusadasi-Setur Marina (117 $\mu g\ g^{-1}$).

The highest mean Mn value was determined at Izmir-Pasaport (392 $\mu g\ g^{-1}$) and the lowest in Candarli 284 $\mu g\ g^{-1}$. However, Mn values measured in Candarli were relatively lower than the other stations. The highest mean Fe value was observed at Izmir-Levent Marina (31,246 $\mu g\ g^{-1}$) and the lowest in Kusadasi-Setur Marina 9953 $\mu g\ g^{-1}$. There was significant spatial difference among stations; however, no significant temporal difference was determined in sediment (Table 5).

The order of mean metal concentrations in sediments in this study decreased in the following order Fe > Mn > Zn > Cu > Pb > Cr > Hg > Cd for Aegean Sea. In generally the order of metals in sediments were not similar to the sequences obtained from seawater with the exception of Fe, Pb, Cu and Cr.

Spearman rank-order correlation analysis ($p < 0.05$) demonstrated that positive correlation existed between some metal concentrations in sediment. The most strong correlation was determined between lead and cadmium ($R = 0.947$). Zinc showed significant correlations with chromium ($R = 0.944$), cadmium ($R = 0.900$) and lead ($R = 0.874$). Chromium also showed marked correlations with cadmium ($R = 0.877$), mercury ($R = 0.853$) and lead ($R = 0.847$). In the study area the relationship between iron and copper was found considerably high ($R = 0.821$) in the sediment samples. Significant correlations were observed between organic matter and heavy metals except Mn and Fe.

Heavy metal concentrations in sediments from the study area were generally lower than those in Euvoikos Gulf (Greece), Ligurian Sea (Italy), the port of Naples (Italy), three docks of Naples Harbour (Italy), in three NW Mediterranean Harbours (France) except Hg (Adamo et al., 2005; Ferraro et al., 2006; Lafabrie et al., 2007). Similar Cd and Zn levels obtained in this study were found in the sediments of Huelva Estuary-Spain (Morillo and Gracia, 2004), Ligurian Sea-Italy (Bertolotto et al., 2005) and Izmir Bay (Kucuksezgin et al., 2006), southwest coast of Spain (Morillo and Gracia, 2004). Similar Mn, Fe values were found with the study performed in the sediments taken from Euvoikos Gulf, Greece (Dassenakis et al., 2003).

Mean and minimum–maximum concentrations (as $\mu g\ g^{-1}$ dry weight, dry wt.) for eight metals in soft tissue of *A. amphitrite* were given in Table 6. *A. amphitrite* showed a narrow range of accumulated body mercury concentrations with the highest mean soft tissue value in Candarli (0.76 $\mu g\ g^{-1}$) and the lowest one in Izmir-Levent Marina (0.02 $\mu g\ g^{-1}$). However Hg showed both temporal and spatial variations during sampling periods (Table 7).

The concentrations of cadmium in *A. amphitrite* varied among the sampling sites ($p < 0.001$) with the lowest to the highest measured values being 5.0–20.0 $\mu g\ g^{-1}$ in Izmir-Pasaport to Candarli in winter and spring periods, respectively. Seasonal variation was only important in Izmir-Pasaport where the highest mean value was recorded in autumn period (16.54 $\mu g\ g^{-1}$).

A. amphitrite showed a wide range of Pb concentrations with significant differences between seasons ($p < 0.001$). Pb levels in organisms ranged between 19.6 $\mu g\ g^{-1}$ at Candarli in summer and 0.97 $\mu g\ g^{-1}$ at Kusadasi-Setur Marina in spring. At all stations, lead concentration was recorded higher in summer period except Kusadasi-Setur Marina where mean Pb concentration reached its highest value in winter.

Chromium concentrations in barnacle reached almost higher in all seasons at Kusadasi-Setur Marina with the maximum value in spring (14.1 $\mu g\ g^{-1}$). In winter period the lowest Cr concentration was observed at Candarli (1.66 $\mu g\ g^{-1}$) in spring where a seasonal decreasing trend was detected. There was a significant differences among stations in which Candarli represented the mean lowest concentration (3.06 $\mu g\ g^{-1}$) in the study area (Table 7).

The barnacle showed a wide range of accumulated body Cu concentration in soft tissue. The maximum Cu level was recorded at Candarli (2372 $\mu g\ g^{-1}$) in autumn while minimum concentration was recorded at Izmir-Pasaport (27.6 $\mu g\ g^{-1}$) in winter. According to ANCOVA results, there were apparent consistent spatial ($p < 0.001$) and temporal ($p < 0.01$) patterns (Table 7).

The maximum Zn concentration was found at Izmir-Pasaport (846 $\mu g\ g^{-1}$) in autumn period and this value was four times greater than those of measured at Candarli station (179 $\mu g\ g^{-1}$) during the same period. According to ANCOVA, there were not significant differences among stations for the Zn which was showed similar trends within stations.

Minimum (9.9 $\mu g\ g^{-1}$) and maximum (72.6 $\mu g\ g^{-1}$) concentrations of Mn were found at Izmir-Pasaport in winter and Candarli in autumn, respectively. The manganese concentrations did not differ significantly among seasons and stations. However inter-seasonal variation can be only observed at Izmir-Pasaport (Table 7).

The mean concentrations of Fe in *A. amphitrite* ranged from 438 in Candarli to 698 $\mu g\ g^{-1}$ in Kusadasi-Setur Marina during the sampling periods. *A. amphitrite* showed a wide range of accumulated soft tissue iron concentrations with some significant differences between sites and seasons. Slightly higher mean levels of Fe in *A. amphitrite* were observed at Izmir-Pasaport and Kusadasi-Setur Marina in autumn.

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Table 6
The mean ± SE and minimum–maximum concentrations of the heavy metals in *Amphibalanus amphitrite* (μg g^{-1} dry wt.).

Region/Site	Length (mm)	Hg	Cd	Pb	Cr	Cu	Mn	Zn	Fe
Candarli	4–6	0.20 ± 0.05	10.5 ± 0.97	4.3 ± 1.67	2.9 ± 0.56	496 ± 47.1	19.0 ± 3.74	203 ± 8.46	407 ± 114
		0.12–0.35	8.9–13.3	1.3–7.4	2.0–4.2	429–633	10.0–25.9	179–217	179–717
	6–8	0.36 ± 0.14	11.2 ± 2.02	4.5 ± 2.19	2.7 ± 0.51	1069 ± 435	34.9 ± 12.6	286 ± 80.7	586 ± 177
		0.18–0.76	7.3–15.9	1.3–10.9	1.7–4.1	549–2371	20.8–72.6	200–528	241–1064
	8–10	0.45 ± 0.04	15.8 ± 0.92	8.0 ± 2.15	3.6 ± 0.45	1060 ± 89.7	27.8 ± 0.67	282 ± 13.4	322 ± 23.0
		0.27–0.63	11.4–20.0	1.3–19.6	2.1–6.0	752–1541	25.3–31.3	235–354	216–428
Izmir-Pasaport	4–6	0.09 ± 0.02	8.6 ± 1.22	4.7 ± 0.85	4.1 ± 0.65	599 ± 110	20.5 ± 3.46	353 ± 53.7	533 ± 58.1
		0.03–0.20	5.0–15.2	2.4–9.3	2.7–6.9	27.6–1033	9.9–39.2	197–644	224–733
	6–8	0.13 ± 0.03	9.8 ± 1.49	3.3 ± 0.23	5.5 ± 1.35	612 ± 185	24.5 ± 4.56	432 ± 76.5	528 ± 63.2
		0.03–0.31	5.1–17.8	2.3–4.5	1.7–12.9	98.1–1615	9.9–49.1	198–846	233–821
	8–10	0.06 ± 0.0	5.4 ± 0.0	3.0 ± 0.0	6.2 ± 0.0	109 ± 0.0	48.6 ± 0.0	221 ± 0.0	471 ± 0.0
		0.06	5.4	3.0	6.2	109	48.6	221	471
Izmir-Levent Marina	4–6	0.05 ± 0.01	6.5 ± 0.14	3.3 ± 1.80	7.2 ± 4.09	345 ± 44.3	23.1 ± 0.58	273 ± 64.6	476 ± 20.1
		0.04–0.05	5.6–7.4	1.5–5.08	3.1–11.2	301–389	22.5–23.6	209–337	456–496
	6–8	0.03 ± 0.00	6.6 ± 0.41	5.1 ± 2.49	7.0 ± 2.11	353 ± 56.4	23.5 ± 2.35	217 ± 11.4	481 ± 131
		0.02–0.04	5.8–7.7	1.4–12.4	2.3–10.6	196–444	19.9–30.3	197–249	314–871
	8–10	0.03 ± 0.01	7.0 ± 0.60	2.7 ± 0.49	5.4 ± 2.02	306 ± 86.8	20.3 ± 2.13	228 ± 23.0	320 ± 30.1
		0.02–0.05	5.9–8.7	1.9–4.14	1.7–10.9	125–513	16.3–26.3	198–297	251–397
Kusadasi-Setur Marina	4–6	0.21 ± 0.03	8.2 ± 0.98	7.0 ± 1.49	9.5 ± 0.82	807 ± 42.8	16.6 ± 1.14	222 ± 1.95	701 ± 43.0
		0.15–0.28	5.5–10.1	2.7–9.5	7.1–10.8	701–910	14.7–19.5	219–227	584–777
	6–8	0.28 ± 0.01	8.9 ± 1.43	5.5 ± 2.80	11.8 ± 1.17	1062 ± 65.8	18.0 ± 1.98	252 ± 14.1	695 ± 85.0
		0.26–0.31	7.2–13.2	0.97–12.7	8.6–14.1	905–1194	14.0–21.0	227–279	501–865

Table 7
Effect of inter-season and inter-station variations covariate size on the variability of soft tissues in *Amphibalanus amphitrite*.

Variable	Season			Station		
	df	*F*	*p* level	*df*	*F*	*p* level
Hg	3	3.367	*	3	59.617	***
Cd	3	1.008	ns	3	14.680	***
Pb	3	9.515	***	3	0.377	ns
Cr	3	2.938	ns	3	19.090	***
Cu	3	7.126	**	3	16.317	***
Mn	3	1.354	ns	3	1.055	ns
Zn	3	7.184	**	3	1.914	ns
Fe	3	1.004	ns	3	2.282	ns

ns: not significant.
* $p < 0.05$.
** $p < 0.01$.
*** $p < 0.001$.

The comparison of soft tissue concentrations of metals demonstrates that, there were significant ($p < 0.05$) seasonal variations for Hg, Pb, Cu and Zn. Significant inter-site differences were observed for Hg, Cd, Cr and Cu concentrations in the soft tissues of *A. amphitrite*. The test did not show statistically significant spatial differences for Pb, Mn, Zn and Fe (Table 7).

There was a significant correlation ($p < 0.05$) for Hg with Cd ($R = 0.638$), Cu ($R = 0.804$) and Zn ($R = 0.561$). The positive correlation was obtained for Hg and Zn. Some metal pairs such as Fe with Cu ($R = 0.548$) and Zn ($R = 0.572$); Cu with Zn ($R = 0.620$) and Cd ($R = 0.552$) showed significant correlations. The concentrations of all metals in seawater were lower than that measured in barnacle. Comparison of rank orders of sites from seawater and barnacle metal data using Spearman's rank correlation tests revealed that there was no significant correlation ($p < 0.05$) except Cu/W-Hg/B ($R = 0.518$), Fe/W-Mn/B ($R = -0.506$), Zn/W-Mn/B ($R = -0.610$) and Mn/W-Cr/B ($R = -0.643$).

The mean metal concentration in barnacles recorded in this study decreased in the following order; Cu > Fe > Zn > Mn > Cd > Cr > Pb > Hg for the Aegean Sea coast. This order showed differences among the stations.

The metal concentration in organisms depends on abiotic (e.g. temperature, salinity, pH, metal concentration) and biotic (e.g. growth rate, food availability, population age structure) factors that may result in a specific pattern of metal concentration in the organism (Blackmore et al., 1998). Metal levels in *A. amphitrite* were higher not only in urban centres such as Kusadasi-Setur Marina, Izmir-Pasaport and Izmir-Levent Marina but also in less urbanised areas such as Candarli suggesting that it may be due to non-sewage related inputs.

Seasonal variation of dissolved oxygen is mainly affected by freshwater flow, temperature and salinity changes. The surface water temperature showed an increasing trend from winter (11.9 °C) to summer (26 °C) and was influenced by the intensity of solar radiation, evaporation, freshwater influx and cooling and mixing water. The higher salinity values could be attributed to the low amount of rainfall, higher rate of evaporation and also due to neritic water dominance (Asha and Diwakar, 2007). pH in surface waters remained alkaline throughout the study period and generally, seasonal variation of pH could be due to CO_2 removal by photosynthesis through bicarbonate degradation, low primary productivity and decomposition of organic matter (Bragadeeswaran et al., 2007).

Metal concentrations in the bodies of marine organisms are dependent on the rates of accumulation, excretion and diluting body growth. Therefore, there is potential for an effect of size on the body metal concentration (Blackmore et al., 1998). In an effort to investigate the relationship between the tissue metal content of *A. amphitrite* and its soft tissue dry weight, linear regression analyses were conducted, taking into account all metal pairs at each site. Log-transformed data for Hg, Cd, Pb, Cr, Cu, Zn, Mn and Fe at each site were checked for significant regressions between metal concentration (y) in μg g^{-1} and body dry weight (x) in g. In many cases significant regressions ($p < 0.05$) were found for Hg, Cd, Cu, Zn, Mn and Fe at various stations. No significant relations were found between Pb and Cr concentration and body weight. The strongest correlations among metal levels and soft tissue dry weight measurements were found for Cd ($R^2 = 0.9918$), Zn ($R^2 = 0.9898$) and for Hg ($R^2 = 0.9132$) at Izmir-Pasaport, for Cu ($R^2 = 0.8880$), Zn ($R^2 = 0.9943$) at Candarli and for Zn ($R^2 = 0.9687$) at Levent Marina. *A. amphitrite* also showed negative relationships ($p < 0.01$) between weight and zinc levels at all stations except Kusadasi-Setur Marina (Fig. 2a–f).

It is well known that one of the most important factors that play a significant role in heavy metal accumulation in marine animals is the metabolic activity (Roesijadi and Robinson, 1994). It is also

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known that the metabolic activity of a young individual is normally higher than that of an older one. Thus, metal accumulation was shown to be higher in younger individuals than the older ones (Rainbow et al., 1993). The negative relationships between metal concentrations and size found in this study may be attributed to the differences in metabolic activity between younger and older barnacles. The net accumulation of heavy metals in an organism is a result of the difference between uptake and depuration and this is the most important factor in metal accumulation. According to our results, it could be said that heavy metal concentrations may be controlled by barnacle's growth. Increasing in size, metal burdens are diluted, i.e. larger barnacles have lower metal concentrations. Barnacles collected from eastern Aegean coast may have been larger and, therefore, may have comparatively lower body metal concentrations. Similar results have been found at Xiamen China coastal waters by Rainbow et al. (1993).

Whilst microphagous feeders, such as barnacles, may ingest many potentially metal-rich particles, they also pass large volumes of water across the permeable surfaces of their cirri and this further facilitates uptake (Rainbow and White, 1989). According to our results, it is possible to assume that higher metal bioavailability in seawater leads to greater accumulation of metals in barnacles.

When we compare the metal levels and weight of the soft tissue, it can be seen that the slope for Hg, Cd, Fe and Mn were greater in Izmir-Pasaport and Candarli. It could be explained either by different bioavailability.

In the current study, the mean metal concentrations generally decreased in the following order: Fe > Zn > Mn > Cu > Pb > Cr > Cd > Hg in seawater, Fe > Mn > Zn > Cu > Pb > Cr > Hg > Cd in sediment and Cu > Fe > Zn > Mn > Cd > Cr > Pb > Hg in barnacle. The highest Cu levels in seawater (2.2–8.1 $\mu g\, l^{-1}$) were measured in

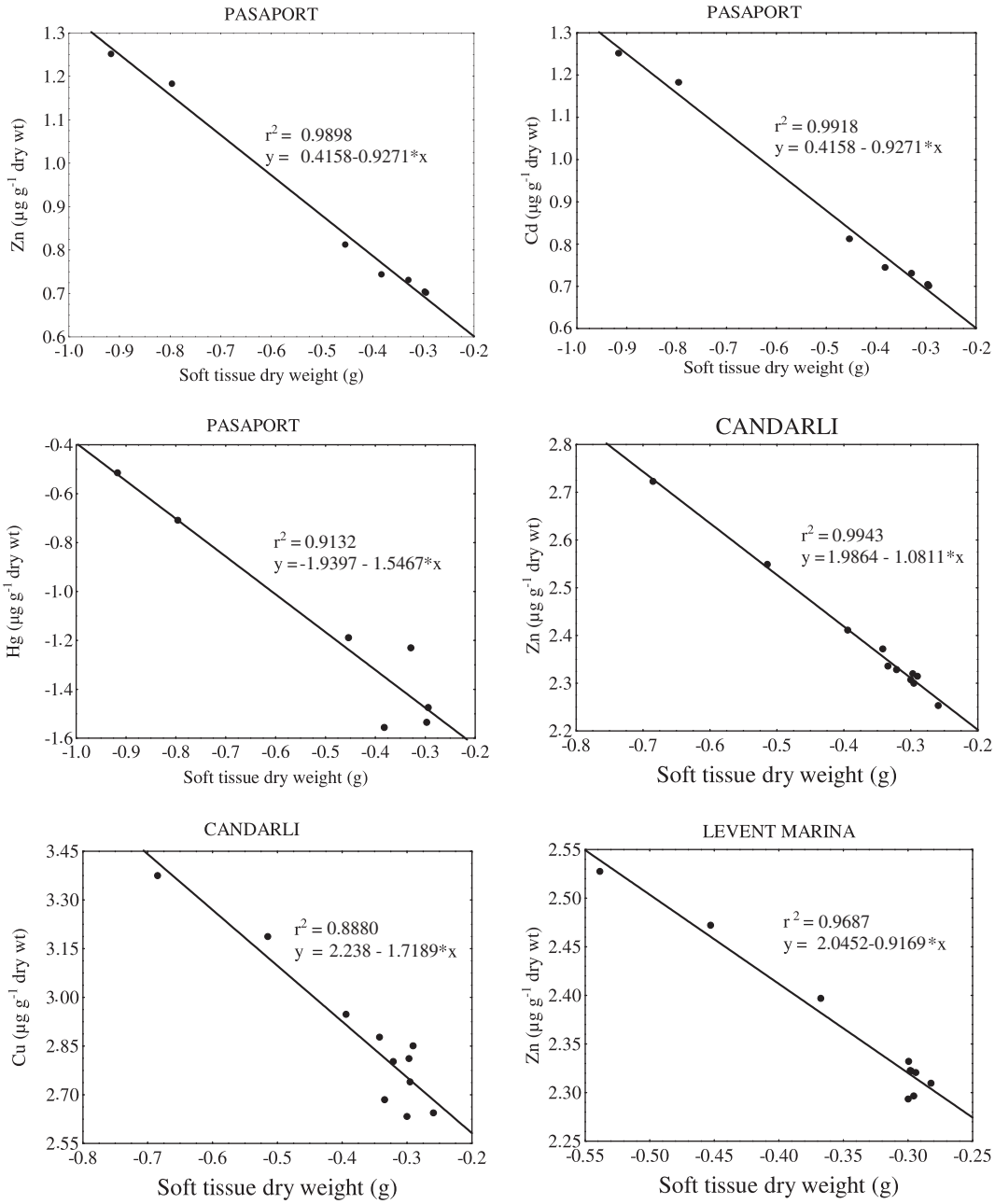


Fig. 2. Linear regression between log metal concentrations ($\mu g\, g^{-1}$, dry weight) and log soft tissue dry weight (g) of *A. amphitrite* in Izmir-Pasaport for Zn, Cd, Hg; Candarli for Zn, Cu; Izmir-Levent Marina for Zn.

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Kusadasi-Setur Marina. This may be due to boat docking, boat repairing activities and corrosion of ships' hulls which generally have the antifouling coating (Dolgen et al., 2003). Kusadasi-Setur Marina used as landing place has a capacity rising from 410 yachts in winter to 525 yachts in summer periods. Cu is the main component of antifouling paints, which is considered as an important anthropogenic source of Cu for the aquatic environment (Jones and Bolam, 2007). Other substances leached from the paints that may have potentially adverse effects, (e.g. Zn) have received little attention (Karlsson et al., 2010). Even though Cu is an essential micronutrient used in enzymes involved in several metabolic processes (White and Rainbow, 1985), the metal may negatively affect organisms at concentrations higher than physiologically necessary. Cu presented the highest values in barnacle, probably reflecting the important role played by this metal in the respiratory metabolism of these organisms in this study.

Measured copper and zinc levels in sediment samples were nearly similar along the Aegean coast. Compared to other stations this can be attributed to organic matter content of sediment. The lowest organic matter value was recorded at Kusadasi-Setur Marina. It is worth noting that there were a significant positive relationships between organic matter (%) and Cu ($p < 0.001$), Zn ($p < 0.001$). Decomposition of the organic matter are found to release heavy metals back to sediments and accumulated; and this process might be responsible for the strong association of Zn and Cu with organic carbon (Bardarudeen et al., 1996). Besides, the release of organically bound heavy metals through influx from land runoff might have also contributed elevated level of Zn and Cu, despite they are meager in amount. Zn and Cu are generally good indicators of anthropogenic inputs (Forstner and Wittmann, 1979).

In this study, Zn values in sea water which exceeded the clean offshore levels were recorded in marinas. In clean offshore areas the values range from 1 to 5 $\mu g\, l^{-1}$, and can even go down to 0.002 $\mu g\, l^{-1}$. However, the levels of Zn in seawater can be as high as 450 $\mu g\, l^{-1}$ especially in polluted harbours (UNEP, 1996). Zinc was the element that presented with the higher concentrations than Mn, Cd, Cr, Pb and Hg in the barnacles. This fact may be related with the importance of Zn in the composition of approximately 90 different enzymes of the animal metabolism. Barnacles are particularly strong net accumulators of particular trace metals, to body concentrations higher than in most invertebrates. Copper is accumulated by barnacles in copper- and sulphur-rich deposits (in extreme copper rich environment), probably representing end products of the lysosomal breakdown of copper-containing metallothioneins. Zinc for example accumulates in form of zinc pyrophosphate granules in the body tissue (Rainbow, 1998).

The comparison of metal concentrations in barnacle with results reported from other geographical areas revealed that in the Aegean coast. Hg, Pb, Cr, Cu, Mn, Zn, Fe concentrations were lower than those other areas in the world (Blackmore et al., 1998; Fialkowsky and Newman, 1998; Rainbow and Blackmore, 2001; Morillo and Usero, 2008). Cd levels were higher than the concentrations reported from other countries (Blackmore et al., 1998; Blackmore, 2001; Rainbow and Blackmore, 2001), while the levels of Cd were generally lower than the reported values at Huelva Estuary (168 $\mu g\, g^{-1}$) (Morillo and Usero, 2008).

In order to assess the concentration factors (CFs) for *A. amphitrite*, water analysis were performed at the same sites and sampling periods. The CF may be used to evaluate the state of conservation of an ecosystem or to monitor its state (Conti and Cecchetti, 2001). CF is the ratio of metal concentration in the barnacle ($\mu g\, g^{-1}$ dry wt.) to the concentration of metal in seawater ($mg\, l^{-1}$) over the sampling periods. CFs could be influenced by the passage of a contaminant through the trophic chain.

Concentration factor (CF) for Cd, Zn and Fe observed as 10^4 while Hg, Cr, Pb and Mn varied from 10^3 to 10^4 (Table 8). Concentration factor for Cu changed between 10^4 and 10^5 in *A. amphitrite*. It has the ability to accumulate metals several times more than sea water because heavy metals, pesticides and other toxic substances can be absorbed from the water column onto surfaces of fine particles (suspended material and benthic fauna). They participate in various biogeochemical mechanisms and can affect the ecosystems through bioaccumulation and biomagnification processes (GESAMP/UNESCO, 1994; Salomons and Förstner, 1984).

Barnacles are excellent biomonitors for investigating available concentrations of trace contaminants because they are sessile organisms, easy to identify, abundant, long lived, available for sampling throughout the year, tolerant of exposure to environmental variations in physico-chemical parameters such as salinity and they are a net accumulator of the relevant metal.

In seawater the highest concentrations of Hg and Cd were measured in Izmir-Pasaport due to the industrial activities. The maximum levels of Pb, Cu, Zn, Fe and Cr, Mn were measured in Kusadasi-Setur Marina and Candarli, respectively. For sediment samples, the concentrations of Hg, Cd, Pb, Cr, Cu, Zn, Mn have the highest values in Izmir-Pasaport. However, the highest mean Fe levels were found at Izmir-Levent Marina. In this study, heavy metal levels in sediment samples were generally lower in the sampling sites than the polluted areas of the Mediterranean Sea except mercury. Metal levels in the sediment showed no significant temporal variation, while all metals showed significant spatial distributions.

The highest soft tissue mean concentrations of Hg, Pb and Cd were recorded in barnacles collected from Candarli whereas the lowest mean concentrations were measured in Izmir-Levent Marina and Izmir-Pasaport, respectively. The highest mean values of Cr, Fe and Mn, Zn were obtained from Kusadasi-Setur Marina and Izmir-Pasaport, respectively. The maximum mean level of Cu in barnacles was measured in Candarli. While mercury and copper showed significant spatial and temporal differences in barnacle soft tissue, zinc and lead showed only seasonal; cadmium and chromium indicated spatial variations in this study. The manganese and iron values did not show significant differences both the stations and sampling seasons. As barnacles increase in size, metal burdens are diluted, i.e. larger barnacles have lower metal concentrations. Barnacles collected from eastern Aegean coast may have been larger and, therefore, may have comparatively lower body metal concentrations.

Generally, in the Mediterranean Sea, organic and trace metal pollution has increased over the last 20 years and this has had a perceptible effect on marine environments. The land based activities and the intensive maritime traffic and untreated domestic discharges, urban activities are the main source of heavy metals in the study area. The study area can be divided into different type of regions, with different degree of environmental pollution. In addition to metal levels investigated in this region, our results indicated that the inner of Izmir Bay especially harbour area is mostly affected by human activities.

In marine areas throughout the world where recreational boats are densely located, concentrations of copper in the water are being found to be in excess of government standards, due to the hull coatings used on these boats. The results suggest that the amounts of Zn and Cu leached from anti-fouling paints may attain toxic concentrations in areas with high recreational activity and account for potential ecological risk. Cu and Zn concentration were high in all the barnacle species from the sampling sites. It is concluded that the barnacle is suitable candidates to be used in biomonitoring surveys of the eastern Aegean coast. Therefore, for monitoring programs, the interpretation of variations in metal concentration in biomonitors would be improved by looking at the potential sources of metal and dietary routes of contaminant transfer in organisms. This would represent an important advance in the understanding of metal concentrations in biomonitor organisms.

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Table 8
Concentration factors for Hg, Cd, Pb, Cr, Cu, Mn, Zn and Fe in *Amphibalanus amphitrite* along the Aegean coast.

Site	Hg	Cd	Pb	Cr	Cu	Mn	Zn	Fe
Candarli	2.8×10^4	5.1×10^4	3.6×10^3	2.6×10^3	2.9×10^5	6.0×10^3	6.9×10^4	7.5×10^4
Izmir-Pasaport	5.9×10^3	3.7×10^4	2.8×10^3	5.1×10^3	6.5×10^4	1.1×10^4	7.7×10^4	5.8×10^4
Izmir-Levent M.	2.1×10^3	2.2×10^4	4.8×10^3	8.3×10^3	2.3×10^5	9.9×10^3	4.3×10^4	2.4×10^4
Kusadasi-Setur M.	2.4×10^4	4.2×10^4	3.5×10^3	1.0×10^4	9.5×10^4	1.2×10^4	2.8×10^4	3.2×10^4

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References

Adamo, P., Arienzo, M., Imperato, M., Naimo, D., Nardi, G., Stanzione, D., 2005. Distribution and partition of heavy metals in surface and sub-surface sediments of Naples city port. Chemosphere 61, 800–809.

Akcali, I., Kucuksezgin, F., 2011. A biomonitoring study: heavy metals in macroalgae from eastern Aegean coastal areas. Mar. Pollut. Bull. 62, 637–645.

Asha, P.S., Diwakar, R., 2007. Hydrobiology of the inshore waters off Tuticorin in the Gulf. J. Mar. Biol. Assn. India 49, 7–11.

Barbieri, P., Adami, G., Predonzani, S., Reisenhofer, E., 1999. Heavy metals in surface sediments near urban and industrial sewage discharges in the Gulf of Trieste. Toxicol. Environ. Chem. 71, 105–114.

Bardarudeen, T., Damodaran, K.T., Sajan, K., Padmalal, D., 1996. Texture and geochemistry of the sediments of a tropical mangrove ecosystem, southwest coast of India. Env. Geol. 27, 164–169.

Bertolotto, R.M., Tortarolo, B., Frignani, M., Bellucci, L.G., Albanese, S., Cuneo, C., Alvarado-Aguilar, D., Picca, M.R., Gollo, E., 2005. Heavy metals in surficial coastal sediments of the Ligurian Sea. Mar. Pollut. Bull. 50 (3), 348–356.

Blackmore, G., Morton, B., Huang, Z., 1998. Heavy metals in *Balanus amphitrite* and *Tetraclita squamosa* (Crustacea Cirripedia) collected from the coastal waters of Xiamen, China. Mar. Pollut. Bull. 36 (1), 32–40.

Blackmore, G., 2001. Interspecific variation in heavy metal bdy concentrations in Hong Kong marine invertebrates. Environ. Pollut. 114, 303–311.

Bragadeeswaran, S., Rajasegar, M., Srinivasanand, M., Kanagarajan, U., 2007. Sediment texture and nutrients of Arasalar estuary, Karaikkal, southeast coast of India. J. Environ. Biol. 28, 237–240.

Bryan, G.W., Langston, W.J., Hummerstone, L.G., 1980. The use of biological indicators of heavy-metal contamination in estuaries with special reference to an assessment of the biological availability of metals in estuarine sediments from south-west Britain. J. Mar. Biol. Assn. UK Occasional Publication No. 1. pp. 1–73.

Chan, H.M., 1989. Temporal and spatial fluctuations in trace metalconcentrations in transplanted mussels in Hong Kong. Mar. Pollut. Bull. 20, 82–86.

Conti, M.E., Cecchetti, G., 2001. Biological monitoring: lichens as bioindicators of air pollution assessment – a review. Environ. Pollut. 114, 471–492.

Dassenakis, M., Andrianos, H., Depiazi, G., Konstantas, A., Karabela, M., Sakellari, A., Scoullos, M., 2003. The use of various methods for the study of metal pollution in marine sediments in the case of Euvoikos Gulf. Greece. Appl. Geochem. 18, 781–794.

Dolgen, D., Alpaslan, M.N., Serifoglu, A.G., 2003. Best waste management programs (BWMPs) for marinas: a case study. J. Coastal Conserv. 9, 57–63.

Ferraro, L., Sprovieri, M., Alberico, I., Lirer, F., Prevedello, L., Marsella, E., 2006. Benthic foraminifera and heavy metals distribution: a case study from the Naples Harbour (Tyrrhenian Sea, Southern Italy). Environ. Pollut. 142, 274–287.

Fialkowsky, W., Newman, W.A., 1998. A pilot study of heavy metal accumulation in a barnacle from the Salton Sea, Southern California. Mar. Pollut. Bull. 36 (2), 138–143.

Forstner, U., Wittmann, G.T.W., 1979. Metal Pollution in Aquatic Environment. Springer-Verlag, Heidelberg, Berlin.

Gavriil, A.M., Angilidis, M.O., 2005. Metal and organic carbon distribution in water column of a shallow enclosed Bay at the Aegean Sea Archipelago, Kalloni Bay, island of Lesvos, Greece. Estuarine Coastal Shelf Sci. 64, 200–210.

GESAMP/UNESCO, 1994. Anthropogenic influences on sediment discharge to the coastal zone and environmental consequences. Rep. Studies 52, 4–30.

Gonul, L.T., Kucuksezgin, F., 2007. Mercury accumulation and speciation in the muscle of red mullet (*Mullus barbatus*) and annular sea bream (*Diplodus annularis*) from Izmir Bay (Eastern Aegean). Mar. Pollut. Bull. 54, 1962–1989.

Jones, B., Bolam, T., 2007. Copper speciation survey from UK marinas, harbours and estuaries. Mar. Pollut. Bull. 54, 1127–1138.

Karlsson, J., Ytreberg, E., Eklund, B., 2010. Toxicity of anti-fouling paints for use on ships and leisure boats to non-target organisms representing three trophic levels. Environ. Pollut. 158, 681–687.

Kucuksezgin, F., Altay, O., Uluturhan, E., Kontas, A., 2001. Trace metal and organochlorine residue levels in red mullet (*Mullus barbatus*) from the eastern Aegean, Turkey. Water Res. 35, 2327–2332.

Kucuksezgin, F., Uluturhan, E., Kontas, A., Altay, O., 2002. Trace metal concentrations in edible fishes from Izmir Bay, Eastern Aegean. Mar. Pollut. Bull. 449, 827–832.

Kucuksezgin, F., Kontas, A., Altay, O., Uluturhan, E., Darılmaz, E., 2006. Assessment of marine pollution in Izmir Bay: nutrient, heavy metal and total hydrocarbon concentrations. Environ. Int. 32, 41–51.

Lafabrie, C., Pergent, G., Kantin, R., Pergent-Martini, C., 2007. Trace metals assessment in water, sediment, mussel and seagrass species – validation of the use of *Posidonia oceanica* as a metal biomonitor. Chemosphere 68 (11), 2033–2039.

Methods of Seawater Analysis, 1983. Second Revised and Extended Edition. In: Grasshoff, K., Ehrhardt, M., Kremling, K. (Eds.). Verlag Chemie.

Morillo, J., Gracia, I., 2004. Heavy metal distribution in marine sediments from the southwest coast of Spain. Chemosphere 55, 431–442.

Morillo, J., Usero, J., 2008. Trace metal bioavailability in the waters of two different habitats in Spain: Huelva estuary and Algeciras Bay. Ecotoxicol. Environ. Saf. 71 (3), 851–859.

Páez-Osuna, F., Bójorquez-Leyva, H., Ruelas-Inzunza, J., 1999. Regional variations of heavy metal concentrations in tissues of barnacles from the subtropical pacific coast of México. Environ. Int. 25, 647–654.

Rainbow, P.S., 1998. Phylogeny of trace metal accumulation in crustaceans. In: Langston, W.J., Bebianno, M. (Eds.), Metal Metabolism in Aquatic Environments. Chapman and Hall, London, pp. 285–319.

Rainbow, P.S., White, S.L., 1989. Comparative strategies of heavy metal acumulation by crustaceans, zinc, copper and cadmium in a decapod, an amphipod and a barnacle. Hydrol. 174, 245–262.

Rainbow, P.S., Huang, Z.G., Songkai, Y., Smith, B.D., 1993. Barnacles as biomonitors of trace metals in the coastal waters near Xiamen, China. Asian Mar. Biol. 10, 109–121.

Rainbow, P.S., Blackmore, G., 2001. Barnacles as biomonitors of trace metal bioavailabilities in Hong Kong coastal waters: changes in space and time. Mar. Environ. Res. 51, 441–463.

Roesijadi, G., Robinson, W.E., 1994. Metal regulation in aquatic animals: mechanism of uptake, accumulation and release. In: Malins, D.C., Ostrander, G.K. (Eds.), AquaticToxicology (Molecular, Biochemical and Cellular Perspectives). London, Lewis Publishers.

Salomons, W., Förstner, U., 1984. Metals in the Hydrocycle. Springer-Verlag, Heidelberg.

Schropp, S.J., Windrom, H.L., 1988. A guide to the interpretation of metal concentrations in estuarine sediments. In: Schropp, S.J., Windom, H.L. (Eds.), Savannah, Georgia, p. 53.

Southward, A.J., 2008. Barnacles. In: Kermack, D.M., Barnes, R.S.K., Crothers, C.H., Hayward, P.J. (Eds.), Synopses of the British Fauna (New Series). The Linnean Society of London and the Estuarine and Coastal Sciences Association, Shrewsbury, pp. 98–100.

UNEP, 1982. Reference Methods for Marine Pollution Studies. No: 14.

UNEP, 1985. Reference Methods for Marine Pollution Studies. No: 26.

UNEP, 1996. Assessment of the state of pollution of the Mediterranean Sea by zinc, copper and their compounds. MAP Technical Report Series, 105.

White, S.L., Rainbow, P.S., 1985. On the metabolic requirements for copper and zinc in molluscs and crustaceans. Mar. Environ. Res. 16, 215–229.

WHO, 1993. Guidelines for Drinking Water Quality, second ed., vol. 1. Recommendations, Geneva.

Zeri, C., Voutsinou-Taliadouri, F., 2003. Processes affecting the distribution of dissolved trace metals in the North Aegean Sea (Eastern Mediterranean). Cont. Shelf Res 23 (9), 919–934.