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Pollution by Mercury, Arsenic, Lead, Chromium, Cadmium, and Polycyclic Aromatic Hydrocarbons of Fish and Mussels from the Gulf of Naples, Italy

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Along the Italian Western central coasts the Gulf of Naples is considered very polluted by civil and industrial wastes (EEA, 2000). In the last ten years many important industries have been closed and environmental protection actions have been developed in the area.
The purpose of this study was to assess the Hg, As, Pb, Cr, Cd and PAH pollution level and its trend in fish and mussels occurring in the zone.

MATERIALS AND METHODS

Site of sampling in the Gulf of Naples was the Campi Flegrei Bay (Fig.1) where until 1991 were located one of the most important steel plants (ILVA) in the Mediterranean and many industrial connected developments now closed.
To assess the pollution level and its trend in marine organisms, samplings of fish (n. 60) and mussels (n. 60) were carried out, about every 2.5 months, between January 1998 and December 1999, and repeated between January 2000 and December 2001. Fish species - pandora *(Pagellus erythrinus)*, bogue *(Boops boops)*, rainbow wrass *(Coris julis)*, rock goby *(Gobius paganellus)*, scorpion fish *(Scorpaena scrofa)*, spotted weever *(Trachinus araneus)* - were caught with bow-nets, gill nets and trawls within 1500 m from the coast. Mussels *(Mytilus galloprovincialis)* were hand collected. Sampling, handling and storage of the specimens were carried out according to the FAO (1976) recommendation. Samples were stored below –20°C until analyses.

Composite samples of each fish species, when lower than 100 g, were prepared blending together muscle tissue of three specimens of almost similar size; fish more than 100 g were analyzed individually. For analyses of mussels, individuals were partially thawed, the shell removed and a composite sample of at least 10 individuals was made (mean wet weight 4 g/individual).

For toxic element determinations, fish and mussel homogenates were dried at 110°C to constant weight for about 24 h; aliquots of 0.5 g of dried samples were closed-vessel digested in concentrated nitric acid, allowed to settle for about 8 h and finally heated at 130-140°C for 1 h. The cooled solutions were transferred to Pyrex tubes with Teflon caps, diluted with distilled water to 15 ml and analyzed. Hg and As were detected by a MHS-l0 mercury hydride system; Pb, Cr and Cd by a Perkin Elmer Atomic Absorption Spectrophotometer 1100 equipped with a HGA – 300 heated graphite furnace system (Perkin Elmer Co., Norwalk, CT-

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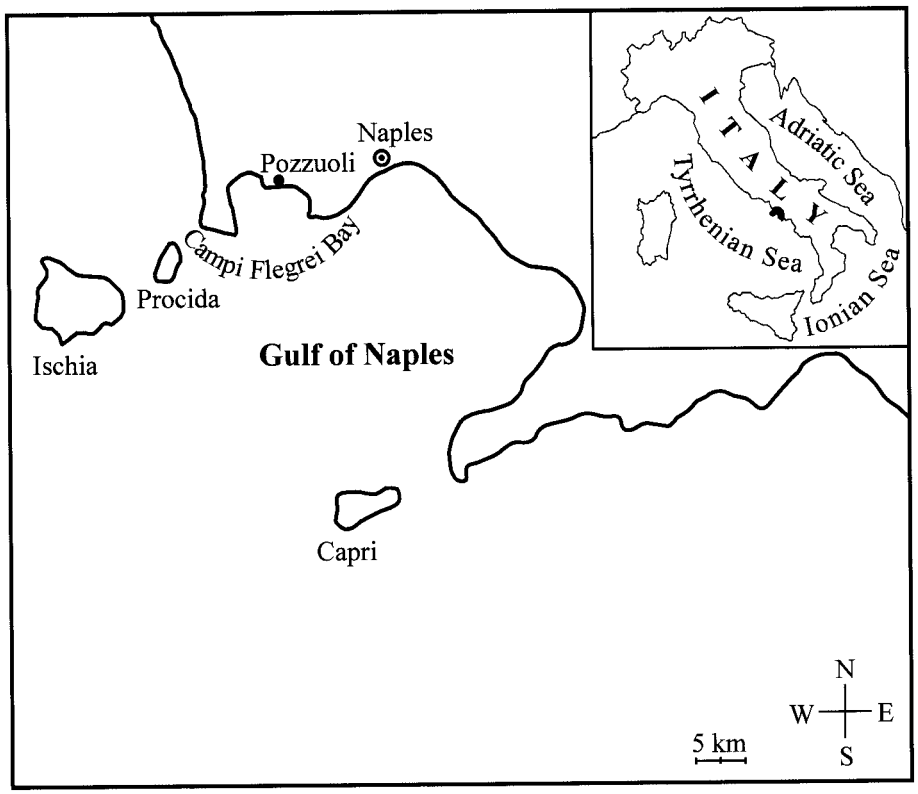

Figure 1. Sampling site in the Gulf of Naples (Italy).

USA) (Medina et al. 1986). Several blanks were included with the reagents to check for possible contamination. Hg and As concentrations were calculated by direct comparison with standard (Sigma-Aldrich Fine Chemicals, St. Louis-Missouri- USA) calibration curves. For quantitation of Pb, Cr and Cd, the standard addition method was applied. The minimum detectable levels, on the basis of wet weight (w wt), were 3 $\mu g\ kg^{-1}$, for Hg and As; 5 $\mu g\ kg^{-1}$, for Pb; 1 $\mu g\ kg^{-1}$, for Cr and Cd. Recovery rates obtained by standard fortified samples were: 76 ± 7% for Hg; 79 ± 9% for As; 80 ± 5% for Pb and Cd and 78 ± 7% for Cr.

To detect PAHs, 30-50 g of the fish/mussel homogenates were digested, extracted, purified and analyzed as described by Takatsuki et al. (1985). HPLC analyses were carried out by a Beckman Liquid Chromatograph equipped with a Model 165 variable wavelength UV detector at 254 nm, a Shimadzu C-R3A integrator and a Supelcosil LC-PAH 25 cm 4.6 column. Elution was carried out at a flow-rate of 2 ml min^{-1}, using a gradient of mobile phase acetonitrile/water starting from 35:65 (v/v) to 100% acetonitrile over 14 min. Sixteen PAHs (naphthalene, acenaphthylene, acenaphthene, fluorene, phenanthrene, anthracene, fluoranthene, pyrene, benz(a)anthracene, chrysene, benzo(b)fluoranthene, benzo(k)fluoranthene, benzo(a)pyrene, dibenzo(a,h)anthracene, benzo(g,h,i)perylene, indeno (1,2,3-cd) pyrene) were investigated. PAH's concentrations were determined comparing the

peak areas with those obtained from the reference standards (PAH mixture 610-M Supelco Inc.Bellefonte, PA, USA).
Detection limits varied from 1 μg kg^{-1} w wt (anthracene) to 20 μg kg^{-1} w wt (acenaphthylene). Experiments undertaken with PAH's standard fortified samples showed a recovery rate ranging from 64 ±10% (fluorene) to 88 ± 4% (anthracene).

Results are summarized as median value and range of pollutant concentrations found in fish and mussels collected over the two sampling periods, corrected on the basis of their recovery rates, and expressed as μg kg^{-1} w wt of each element and total PAHs (TPAHs).

Statistic analysis was performed by SYSTAT 5.2 for MacIntosh.

RESULTS AND DISCUSSION

Concentrations of toxic elements and TPAHs obtained on the biota sampled during 1998 - 1999 and 2000 - 2001 are shown in Tables 1-3, compared with the available literature data.
All the analytical samples showed measurable concentrations of toxic elements and PAHs.

In fish, Hg concentrations varied from 5 to 120 μg kg^{-1} w wt, median value 30 μg kg^{-1} w wt, during 1998 - 1999, and between 5 and 340 μg kg^{-1} w wt, median value 33 μg kg^{-1} w wt, during 2000 - 2001. These medians are lower than mean values obtained by Giordano et al. (1991) in the same zone and by Amodio-Cocchieri et al. (1993) in the Ionian sea.

In fish sampled during 1998 - 1999 As contents were in the range 5 - 110 μg kg^{-1} w wt (median value 18 μg kg^{-1} w wt) and in the range 5 -150 μg kg^{-1} w wt (median value 10 μg kg^{-1} w wt) in those captured in the 2000 - 2001 years. Recent data are not available about fish As contamination in the zone nor in other Italian zones. The As levels found in the course of the present study are much lower than those obtained in Mediterranean sea by Grimanis et al. (1978) and by Alberti et al. (1995) and in North East Irish sea by Leah et al. (1992).

Pb levels ranged from 5 to 180 μg kg^{-1} w wt (median 39 μg kg^{-1} w wt) in fish captured during 1998 - 1999 years; over 2000 - 2001 the range was the same reported above, but the median value was 42 μg kg^{-1} w wt. In fish captured in the same zone in 1986 over the summer Giordano et al. (1991) found detectable amounts of Pb only in *Serranus cabrilla* at a mean value of 50 ± 5 μg kg^{-1} w wt. Our values are much lower than those referred by Amodio-Cocchieri et al. (1993) about fish species residing in the Ionian sea.

During 1998 - 1999 Cr concentrations varied from 3 to 355 μg kg^{-1} w wt (median value 102 μg kg^{-1} w wt); in fish sampled over the 2000 - 2001 years Cr ranged between 3 and 180 μg kg^{-1} w wt, with a median value of 48 μg kg^{-1} w wt. As observed for As, no data on the Cr contamination of fish living in the zone investigated in the present study are available. The values found in the present study are much lower than those obtained in the Ionian sea (Amodio-Cocchieri et al. 1993).

In fish sampled in the course of 1998 - 1999 Cd concentrations varied from 4 to 110 μg kg^{-1} w wt (median 9 μg kg^{-1} w wt); in the following years the range was 5 - 75 μg kg^{-1} w wt with a median value of 10 μg kg^{-1} w wt. About fifteen years before in the same sampling zone (Ischia, Capri, coast of Naples) Giordano et al. (1991) did never obtained detectable amounts of Cd in analyzed fish. In the Ionian sea Amodio-Cocchieri et al. (1993) found in fish mean levels of Cd varying from 10 to 70 μg kg^{-1} w wt.

The TPAH content varied between 5 and 812 μg kg^{-1} w wt (median 120 μg kg^{-1} w wt) and between 5 and 414 μg kg^{-1} w wt (median 22 μg kg^{-1} w wt) during 1998 - 1999 and 2000 - 2001 years, respectively. These values are much lower than those obtained by Amodio-Cocchieri et al. (1990) in the same zone and in the Ionian (Amodio-Cocchieri et al. 1993).

Among the detected pollutants, only chromium and TPAHs showed in fish collected over the two sampling periods significant variations *($p < 0.001$)* of the contamination levels.
The contents of toxic elements in fish occurring in the zone are relatively low; mercury, arsenic, lead and cadmium concentrations are very similar over the two sampling periods considered, but chromium and TPAHs are lowering over the time. The contamination levels of fish are lower than those obtained when many industrial developments were located in the zone. The pollutant levels are also lower than those exhibited by fish living along other Italian coasts or in other Mediterranean or North European sites. In particular, the arsenic contamination in fish captured within the Campi Flegrei Bay appears much more lower than that found in fish from Spain, Greece and Ireland.

The concentrations of toxic elements and TPAHs obtained on the mussels sampled during 1998 - 1999 and 2000 - 2001 are listed in Tables 2-3.

The concentration ranges of Hg have been 5-620 μg kg^{-1} w wt (median 243 μg kg^{-1} w wt) and 5-366 μg kg^{-1} w wt (median 204 μg kg^{-1} w wt) in 1998 - 1999 and 2000 - 2001 years, respectively. These Hg pollution levels are in a good agreement with those observed in the Pozzuoli area during a surveillance plan on marine pollution carried out along the Italian coasts by the Italian Department of Environment (Ministero dell'Ambiente 2001), but are much higher than those found by Giordano et al. (1991) in the same sampling zone about fifteen years before. They are much higher also than those detected along the other Italian coasts (Giordano et al. 1990; Amodio-Cocchieri et al. 1993; Regoli and Orlando 1994; Renzoni 1998). The results obtained in the first sampling period agree with Hg contents found by De Martin et al. (2001) in molluscs other than mussels. The results obtained in the second sampling period agree with Hg contents found in the Ionian sea (Martella et al. 1997).
Anyway over 1998 - 1999 the Hg concentrations in mussels were in most cases below the law limit for edible molluscs (500 μg kg^{-1} in muscle tissue). Over 2000 - 2001 Hg contents did never exceed the law limit (D.M. 14 luglio 1971).

Over the two sampling periods, As concentrations varied in mussels from 6 to 219 μg kg^{-1} w wt (median 60 μg kg^{-1} w wt) and from 5 to 149 μg kg^{-1} w wt (median 42 μg kg^{-1} w wt), respectively. As noted about fish, data on As pollution in mussel in the zone are not available, while in the Ionian sea along the coasts of Salento, Brindisi and Taranto, Martella et al. (1997) observed in *Mytilus galloprovincialis Lmk* mean concentrations of As varying from 1216 and 15,667 μg kg^{-1} w wt. In

Spain, Alberti et al. (1995) found on *Mytilus edulis* collected in the Barcelona zone an average As amount of 1410 μg kg^{-1} w wt.

Pb concentrations varied from 5 to 310 μg kg^{-1} w wt (median 231 μg kg^{-1} w wt) over 1998 - 1999 and from 5 to 272 μg kg^{-1} w wt (median 210 μg kg^{-1} w wt) over the next two years. These findings, in a good agreement with those obtained by the Italian Department of Environment (Ministero dell'Ambiente 2001) in the Pozzuoli area, are much lower than those meanly detected by Giordano et al. (1991) in the same site some years before. Available data about Pb contamination in *Mytilus galloprovincialis* collected along the Italian coasts other than the Gulf of Naples in the last years (Giordano et al. 1990; 1991; Amodio-Cocchieri et al. 1993; Regoli and Orlando 1994; Martella et al.1997; Renzoni 1998; Ministero dell'Ambiente 2001) show levels much higher than those found in the course of the present study in the zone considered.
Pb concentrations in mussel muscle tissue were always below the law limit - 2000 μg kg^{-1} w wt - for edible molluscs (Ministero della Sanità 1999).

Cr concentrations varied between 3 and 237 μg kg^{-1} w wt over 1998 - 1999 and the median value was 165 μg kg^{-1} w wt; in the next period they ranged from 3 to 210 μg kg^{-1} w wt, with a median value of 150 μg kg^{-1} w wt. These values are lower than those obtained in various sites along the Italian coasts (Amodio-Cocchieri et al. 1993; Regoli and Orlando 1994; Martella et al. 1997; Renzoni 1998; Ministero dell'Ambiente 2001).

During 1998 - 1999 Cd concentrations ranged between 1 and 126 μg kg^{-1} w wt with a median value of 60 μg kg^{-1} w wt; over the next two years, the range was 1 - 100 μg kg^{-1} w wt and the median 33 μg kg^{-1} w wt, according to those meanly obtained by the Italian Department of Environment (Ministero dell'Ambiente, 2001) along the coasts of Campania Region. More than ten years before Giordano et al. (1991) found in *Mytilus galloprovincialis* collected in the Gulf of Naples mean levels of Cd of 130-150 μg kg^{-1} w wt, while along the other Italian coasts, over the 1986 -1998 years, Cd contents varied from 12 to 352 μg kg^{-1} w wt (Ministero dell'Ambiente, 2001).
The law limit for Cd concentration in muscle tissue of edible molluscs - 2000 μg kg^{-1} w wt - was never exceeded in the course of the present study (Ministero della Sanità 1999).

The total concentration of PAHs ranged from 105 to 831 μg kg^{-1} w wt (median 334 μg kg^{-1} w wt) and from 16 to 627 μg kg^{-1} w wt (median 241 μg kg^{-1} w wt) over the two sampling periods, respectively. The contamination levels lowered over the time but the difference obtained was not significant. These results agree with mean level of PAH's pollution found more than ten years before in the same zone by Amodio-Cocchieri et al. (1990).

Over the sampling years in *Mytilus galloprovincialis* significant variations of the contamination levels were only observed for Hg *($p = 0.005$)* and Cd *($p < 0.001$)*, while As, Pb, Cr and PAH pollution haven't exhibited a significant reduction.
In *Mytilus galloprovincialis,* a significant reduction of mercury and cadmium contents over 1999 - 2001 was found, while the concentrations of the other pollutants appear stationary. Even if these pollution levels can be considered very low in comparison with those found in other sites and don't exceed the law limits for edible molluscs, the levels of mercury, as noted also by the Italian Department of Environment (Ministero dell'Ambiente 2001), are higher than those obtained in

Table 1. Concentrations of toxic elements and total PAHs (μg Kg^{-1} wet weight) in fish from the Campi Flegrei Bay in the Gulf of Naples (Italy) over 1998 - 1999 / 2000 - 2001 years and comparison with the literature data.

ELEMENT	PRESENT RESULTS				LITERATURE DATA		
	1998-1999		2000-2001				
	MEDIAN	RANGE	MEDIAN	RANGE	MEAN	SITE	AUTHORS
Mercury	30	5-120	33	5-340	102-192	Italy-Gulf of Naples	Giordano et al.1991
					40-180	Italy-Ionian Sea	Amodio Cocchieri et al.1993
Arsenic	18	5-110	10	5-150	420-23,700	Greece- Aegean Sea	Grimanis et al.1978
					2400-21,300	Eire- N-E Irish Sea	Leah et al.1992
					4140-6150	Spain- Barcelona Coast	Alberti et al.1995
Lead	39	5-180	42	5-180	50	Italy- Gulf of Naples	Giordano et al.1991
					70-1480	Italy- Ionian Sea	Amodio Cocchieri et al.1993
Chromium	102	3-355	48	3-180	80-2200	Italy- Ionian Sea	Amodio Cocchieri et al.1993
Cadmium	9	4-110	10	5-75	NO DETECT.	Italy- Gulf of Naples	Giordano et al.1991
					10-70	Italy- Ionian Sea	Amodio Cocchieri et al.1993
Total PAHs	120	5-812	22	5-414	94-1930	Italy- Gulf of Naples	Amodio Cocchieri et al.1990
					22-340	Italy- Ionian Sea	Amodio Cocchieri et al.1993

Table 2. Concentrations of Mercury and Arsenic (μg Kg^{-1} wet weight) in *Mytilus galloprovincialis* from the Campi Flegrei Bay in the Gulf of Naples (Italy) over 1998 - 1999 / 2000 - 2001 years and comparison with the literature data.

ELEMENT	PRESENT RESULTS				LITERATURE DATA		
	1998-1999		2000-2001				
	MEDIAN	RANGE	MEDIAN	RANGE	MEAN	SITE	AUTHORS
Mercury	243	5-620	204	5-366	5-54	Italy-Tyrrhenian Sea	Giordano et al. 1990
					15	Italy-Adriatic Sea	" "
					10-32	Italy-Gulf of Naples	Giordano et al. 1991
					10	Italy-Ionian Sea	Amodio Cocchieri et al.1993
					5-18	Italy-Adriatic Sea	Regoli and Orlando 1994
					8-327	Italy-Ionian Sea	Martella et al. 1997
					50	Italy-Tyrrhenian Sea	Renzoni 1998
					27-61	Italy-Adriatic Sea	" "
					10-640	" "	De Martin et al. 2001
					251	Italy-Gulf of Naples	Ministero dell'Ambiente 2001
Arsenic	60	6-219	42	5-149	1410	Spain-Barcelona Coast	Alberti et al. 1995
					1216-15,667	Italy-Ionian Sea	Martella et al. 1997

Table 3. Concentrations of Lead, Chromium, Cadmium and total PAHs (μg Kg^{-1} wet weight) in *Mytilus galloprovincialis* from the Campi Flegrei Bay in the Gulf of Naples (Italy) over 1998 - 1999 / 2000 - 2001 years and comparison with the literature data.

ELEMENT	PRESENT RESULTS				LITERATURE DATA		
	1998-1999		2000-2001				
	MEDIAN	RANGE	MEDIAN	RANGE	MEAN	SITE	AUTHORS
Lead	231	5-310	210	5-272	40-1350	Italy- Tyrrhenian Sea	Giordano et al.1990
					88-7600	Italy- Italian Coasts	Giordano et al.1990, 1991; Amodio Cocchieri et al.1993; Regoli and Orlando 1994; Martella et al. 1997; Renzoni 1998; Ministero dell'Ambiente 2001
					276	Italy- Gulf of Naples	Ministero dell'Ambiente 2001
Chromium	165	3-237	150	3-210	440	Italy- Ionian Sea	Amodio Cocchieri et al.1993
					102-1710	Italy- Adriatic Sea	Regoli and Orlando 1994
					133-1200	" "	Martella et al.1997
					266	Italy- Tyrrhenian Sea	Renzoni 1998
					230-755	Italy- Adriatic Sea	" "
					86-276	Italy- Gulf of Naples	Ministero dell'Ambiente 2001
Cadmium	60	1-126	33	1-100	130-150	Italy- Gulf of Naples	Giordano et al.1991
					12-352	Italy- Italian Coasts	Ministero dell'Ambiente 2001
					28-96	Italy- Campania Coasts	" "
					42	Italy- Gulf of Naples	" "
Total PAHs	334	105-831	241	16-627	295	Italy- Gulf of Naples	Amodio Cocchieri et al.1990

the same zone about fifteen years before and than those recently obtained from other Italian areas.

In conclusion, the investigated zone can be considered at low level of pollution by Hg, As, Pb, Cr and Cd. The decreasing of levels of PAHs in fish is probably attributable to industrial dismantling and to the environmental protection actions developed in the Gulf of Naples last years. Nevertheless, the levels of PAHs in mussel, which is considered one of the best biological markers in defining and monitoring water pollution, are similar to those of molluscs living in zones considered moderately polluted (Pancirov & Brown 1977; Iosifidiou et al. 1982; Rainio et al. 1986). They haven't shown significant variations over the studied periods nor from 1988 (Amodio-Cocchieri et al. 1990). As the waters of the Campi Flegrei Bay are extensively utilized for tourist and mariculture purpose, a special attention must be put in achieving some knowledge on the mercury and PAH local pollution sources.

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