

A study on the possible use of *Anisakis* as biological indicators of heavy metals contamination in marine environments

A COSTA¹, G CAMMILLERI¹, ML RIZZUTO¹, L PANTANO¹, FG GALLUZZO¹, A BRUNONE², E GIANGROSSO¹, MD BUSCEMI¹, V FERRANTELLI¹

¹ Istituto Zooprofilattico Sperimentale della Sicilia; Palermo, Italy ² Biologist Freelance, Palermo, Italy

INTRODUCTION

A bioindicator is an organism that provides information on the environmental conditions of its habitat based on its presence or absence or for its behavior, which vary according to the exposure or toxic effect of environmental chemicals. (Peakall, 1994). Marine fish are subject to infestation with various species of parasites (Sures et al., 2001). This aspect, together with their ability to accumulate heavy metals, suggests that parasites could be used as biological indicators of heavy metals contamination in marine environment (Sures et al., 2001). Among the species of parasites used as biological indicators, particular attention can be given to the *Anisakis* nematodes, as they respond to most of the typical characteristics of a bioindicator; they are easily identifiable and can be collected during inspection and have a high species-host adaptation (Lester & Mackenzie 2009). The aim of this work was to determine the ecological parameters useful for the recognition of *Anisakis* spp. as environmental indicators of As, Cd Hg and Pb pollution in the Mediterranean.

MATERIALS AND METHODS

A total of 87 fish samples from Mediterranean (FAO 37.1.3) and 6 specimens of stranded dolphins were examined for the detection of anisakid parasites and the detection of Cd, As, Pb and Hg in the host muscle and in the larvae themselves. The larvae collected were identified morphologically and molecularly through protocols found in literature (D'Amelio et al. 2000). About 0.1 g of sample were put into the automatic mercury analyzer (DMA 80 - Milestone) for the determination of Hg. A calibration curve was constructed by associating an absorbance value to 5 concentration points to assign a concentration value to the samples. As, Cd and Pb were quantified using a 7700x Series ICP-MS (Agilent Technologies, Santa Monica CA, USA). The extraction of the samples was performed by a digestion procedure according to the UNI EN 13805: 2002.

A maximum quantity of 10 *Anisakis* larvae/adults were considered for the extraction to eliminate any bias related to the starting quantity for extraction. The solutions have been brought up to 50 ml with ultrapure water and filtered before ICP-MS analysis.

RESULTS AND CONCLUSIONS

The As arsenic levels in the fish samples showed average maximum values in mackerel (6.856 ± 3.94 mg / kg). The mackerel samples examined also revealed the average maximum levels of cadmium (0.031 ± 0.069 mg / kg). The maximum Pb average value was reached in squid samples (0.070 ± 0.095 mg/kg).

The highest average levels of mercury were found in monkfish (0.613 ± 0.245 mg / kg). The As concentrations in the infesting anisakids showed average values up to 0.066 ± 0.120 mg/kg. The highest values of Cd were found in samples of *Anisakis* infesting squid (0.073 ± 0.09 mg / kg). This study found very high Pb concentrations (0.480 ± 0.45 mg / kg) with a maximum of 1.06 mg/kg. No Hg was detected in all the parasite samples, except for a silver scabbardfish-infesting sample which found levels of 0.076 mg/kg. The Pb levels of *Anisakis* in silver scabbard samples were significantly higher than the silver scabbard muscle samples ($p = 0.00012$), with a C[parasite]/C[host] bioconcentration ratio of 6.82 while no significant differences were found for As and Cd. The horse mackerel-*Anisakis* comparison also revealed significant differences for Pb ($p = 0.0000041$) and As ($p = 0.00077$), with C[parasite]/C[host] bioconcentration ratios of 9.275 and 0.017, respectively. The comparison of the dolphin samples with the adults of *Anisakis* did not reveal any significant differences ($p > 0.05$) in the values of Cd, Pb, As and Hg. The bioconcentration factor confirmed the clear divergence in the accumulation of lead between *Anisakis* and its hosts. *Anisakis* could be therefore confirmed as sensitive indicators of Pb in aquatic ecosystems, showing an evident signal of the load of environmental pollution at sea.