FATE OF PERSISTENT CHEMICAL WARFARE AGENTS IN A BENTHIC ECOSYSTEM OF THE SOUTHERN ADRIATIC SEA

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Abstract

In this study we analyze the status of health of a benthic ecosystem affected by chemical warfare agents (CWAs), loaded in weapons that were dumped in southern Adriatic Sea during and after WWII. Following a multidisciplinary approach, samples of sediment, water and fish were collected both in the area of study and in a reference site. Analysis have been carried out to determine the contamination and biotoxicity of water and sediments by CWAs. Stress index were developed in order to evaluate the toxicological effect of CWAs in fish and macroscopic analysis of liver, spleen and skin were utilized to assess the health status of fish. The results provide a initial worrying picture of the benthic ecosystem healthiness even though more work is needed.

Keywords: Adriatic Sea; bathyal, ecotoxicology, monitoring, pollution

Introduction

Among war material of any sort, chemical weapons were dumped in the southern Adriatic Sea during and after WW II. An evidence is the fact that from 1946 until 1996 more than two hundred Apulian fishermen have been hospitalised because of being severely by chemical injured by chemical warfare agents (CWAs) (1, 2). Bis-(2-chloroethyl) sulpide (Cl-CH_2CH_2-S-CH_2CH_2-Cl), a CWA commonly known as "mustard gas" or yperite ("H", Levinstein process) and dichloro-(2-chlorovinyl)arsine (Cl-CH=CH-$As(Cl)_2$) ("lewisite") they are among the twenty-four different CWAs that were contained in bombs, grenades and drums dumped in different sites of the southern Adriatic Sea at depths in the range of 150 ÷ 1000 m (Fig. 1). Eighteen of these compounds pose major risks to the concerned benthic ecosystem because their physical-chemical properties make their noxiousness persistent in sea water. Hydrolysis products, such as 1,4-thioxane (S-CH_2CH_2-O-CH_2CH_2) and bis(2-hydroxyethyl)sulfide (HO-CH_2CH_2-S-CH_2CH_2-OH) from yperite and 2-chlorovinylarsenious acid (Cl-CH=CH-$As(OH)_2$) and 2-chlorovinylarsenious oxide (Cl-CH=CH-As=O) from lewisite, have shown to be even more toxic than their parent products (3).

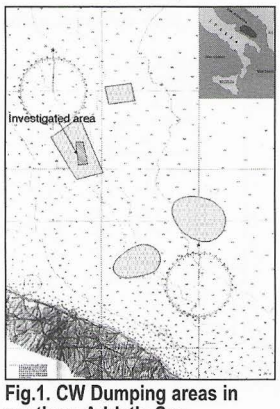

Fig.1. CW Dumping areas in southern Adriatic Sea

Methods and Materials

In June 1999 a survey was carried out on a ten square nautical miles rectangle of the sea-floor. This site was chosen from one of the four different dumping zones that the authors discovered in the southern Adriatic Sea (Fig. 1). A digital DATASONIC CHIRP SIS 1000 side scan sonar and sub bottom profiler coupled with a magnetometer (mod. G880 GEOMETRICS) were employed for the localisation of "targets" on the sea-bottom at depths in the range of 200 - 300 m. The analysis of the recorded courses allowed the ranking of 102 targets and the choice of the ones considered as representative. Nine aerial and two artillery bombs were observed by means of a remotely operated vehicle (mod. SEASURVEYOR SEA EYE 215) that was specially equipped to collect samples of sediment and water. In three cases, the CWAs contained in the observed rusted shells were clearly visible both from holes and fractures of the bomb body as well as on the surrounding sea-floor (hard substrata made by coarse sediments enriched with fine particles).

By means of the R.O.V, macrozoobenthos (*Thenea muricata, Abra sp., Cidaris cidaris, Echinus acutus*) and demersal fishes (*Helicolenus dactylopterus* and *Conger conger*) were observed to behave apparently normally while they were very close to what was considered as an yperite-based partially hydrolised product.

Sampling surveys were carried out to collect demersal fishes both in the area of risk and in two areas located in the southern Tyrrenian Sea supposed to be unaffected by war material dumping.

A multidisciplinary approach was used to investigate the noxiousness of the CWAs on the benthic environment. Sediments and fish tissues (muscle and liver) were analysed by means of GC-MS full scanning in order to detect yperite and lewisite products traces. Water and sediment samples were tested by means of MICROTOX® and tissues (liver, brain, muscle and gills) of *Conger conger, Helicolenus dactylopterus, Raja asterias, Raja clavata, Trigla lyra* and *Trigla lucerna* were analysed to detect traces of arsenic by means of AAS and to measure some "stress indices" (CYP-450 1A, Ech). Furthermore, a health assessment index (HAI) was assigned to each individual, according to the number of macroscopic alterations observed. Histopatological analysis were performed on liver and spleen of the same fishes.

Results and Discussion

Sediment samples revealed the presence of some decomposition products of yperite and of bis-(2-chloroethyl) disulpide (impurity product of Levistein's process): 1,4-tioxane; 1,4-dithiane; 1-oxa-4,5-dithiapane; 1,2,5-Trithiapane. Sediment samples analysed with the MICROTOX, test showed biotoxicity both in the solid phase and in the interstitial water.

Arsenic levels in *Helicolenus dactylopterus* (8 individuals) collected from the area of study were significantly higher (Mann Withney *U* test, $p<0,05$) compared to those found in the 6 individuals of the same species collected from the control site (Fig. 2).

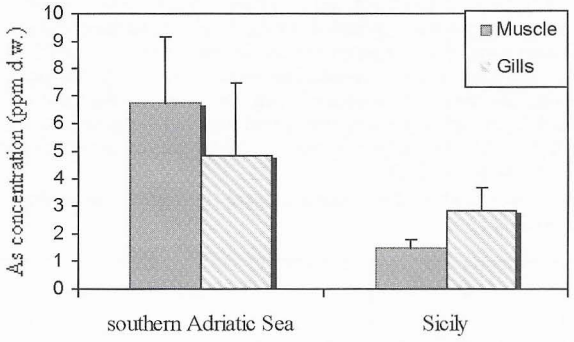

Fig. 2. As concentration (ppm d.w.) in specimens of *H. dactylopterus* collected

Moreover the enzyme activity of *Helicolenus dactylopterus* involved in detoxifying processes in liver tissues (CYP-450 1A) and the physiological activity of enzymes in brain and muscle tissues (AchE) showed significant differences compared to the controls values.

The "Health Assessment Index (HAI)" (4), counted 15 out of 18 individuals of *Helicolenus dactylopterus* as damaged whilst in the control site only 11 out of 21 specimen of *H. dactylopterus* showed significant macroscopic alterations. Histological analysis revealed evident damages (steatosis, fibrosis, granuloma, and atrophy of linphatic centres) in liver and spleen tissues of 16 individuals among the 18 *H. dactylopterus* analyzed.

The analytical results indicate that the leakage of CWAs from the rusted bomb shells is likely to produce negative effects on the concerned benthic ecosystem. In order to assess the environmental risk, more data and a wider biological sample size are needed. At the moment, the Italian Ministry of the environment is taking into consideration this request.

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