



Trace elements bioaccumulation in liver and fur of *Myotis myotis* from two caves of the eastern side of Sicily (Italy): A comparison between a control and a polluted area[☆]

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ABSTRACT

Environmental pollution is a topic of great interest because it directly affects the quality of ecosystems and of all living organisms at different trophic and systematic levels. Together with the global climate change, the long-term surviving of many species of plants and animals is threaten, distributional patterns at global and regional levels are altered and it results in local assemblages of species that are quite different from those that currently constitute coevolved communities. For this study, the species *Myotis myotis* was used as bioindicator and it was sampled from two caves in the south-east of Sicily, Pipistrelli chosen as control area and Palombara chosen as polluted area, to measure the concentrations of trace elements in fur and liver tissues. Results showed higher content of essential elements in fur in bats sampled from Pipistrelli. Conversely, higher concentrations of toxic metals in liver such as As, Cd, Pb and Hg were measured in bat samples in Palombara cave, where specimens have a hunting area extended within the boundaries of the petrochemical plant. Nevertheless, we cannot consider Palombara population as polluted by metal contamination since their tissue concentrations are overall lower than toxic thresholds values suggested for small mammals. Likewise, we cannot exclude other kind of pollutants as potential stressors of the examined population, contributing with the decreasing of bat colonies in Sicily.

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1. Introduction

Environmental pollution is a topic of great interest because it directly affects the quality of ecosystems and of all living organisms at different trophic and systematic levels (Ferrante et al., 2017b). Pollutants can be directly uptaken by organisms through ingestion of contaminated prey items, dermal exposure or inhalation. One of the most effective approaches for evaluating metals in the environment, and thus of the risk associated for organisms living in a metal contaminated environment, is the biological monitoring (Ferrante et al., 2015; Mazzei et al., 2014). From this perspective, it is

crucial to assess the concentrations of contaminants bio-accumulated in organisms to understand their potential adverse effects, especially of long-term persistent chemicals such as metals. The most toxic metals are As, Cd, Pb and Hg, which represent the ultimate form of persistent pollutants. Unlike other pollutants, metals are not degraded in the environment and can accumulate through the food chain posing potential risks to the human health and the health of ecosystems (Mazzei et al., 2014). Nevertheless, some metals are essential for organisms and are naturally available in food and water, but in the case of high levels or lack of these essential elements (e.g., manganese, nickel, cobalt, copper, iron, and zinc), adverse health effects may occur (Ferrante et al., 2018; Mansour et al., 2016).

Insectivorous bats are considered reliable bioindicator organisms as: 1) they are vulnerable to a wide range of environmental stressors; 2) they are long-lived mammals and occupy higher

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positions in the trophic chain; 3) they occur on a wide geographic range; 4) their population dynamics can present rapid declines due to their slow reproductive rates; 5) they provide key ecosystem services; 6) they are more likely to show the effects of pollutants than fructivorous bats (Alleva et al., 2006; Jones et al., 2009; Zukal et al., 2015).

Furthermore, due to their relatively long life, up to 37 years recorded for *Myotis brandtii* (Gaisler et al., 2003), compared to their small body size, and their high daily food intake (e.g. up to 0.5 g/bw/day on a wet basis measured experimentally for *Myotis lucifugus*) (Anthony and Kunz, 1977), bats can be particularly prone to chemical exposure, especially to contaminants such as metals accumulated through the food chain (Hernout et al., 2016a). The chemical contamination is one of many stressors, which is implicated in the demographic decline of many bat populations across Europe and North America, but the risks to bats remain poorly understood (Hernout et al., 2016a). To biomonitor environmental contamination in wildlife species, non-invasive proxy have been proposed and investigated such as feathers and fur. Concentrations of pollutants in fur reflect the exposure during the period in which the hair is growing, which occurs once annually between summer and fall (Fraser et al., 2013). Bat fur can be a reliable non-invasive tool to investigate metal contamination, however, the method has been evaluated on a limited number of metals (Hernout et al., 2016b). In addition, and with the focus mainly on long-term exposure, adipose tissue and other tissues like bone, liver, brain or kidney, were used as long-term biomonitors of certain elements/metals (Domingo et al., 2017). *Myotis myotis*, the greater mouse-eared bat, is an European species included in the Red List as Least Concern 3.1 (IUCN, 2017) and Vulnerable A2c in the Red List of Italian Vertebrate (IUCN-Italy, 2013). *M. myotis* is widely distributed in Europe and the Mediterranean area and is an insectivorous species with an average life expectancy ranging from 2.4 to 2.7 to 4–5 years, with a maximum longevity of 22 years (Dietz et al., 2009). In Italy, the *M. myotis* population is stable and occurs on the entire territory (Lanza and Agnelli, 1999). However, in Sicily, the insular population of *M. myotis* decreased and several colonies disappeared compared to the past decades (Agnelli et al., 2004). Whereas the causes of the population decline of *M. myotis* in Sicily have not been identified, further research is needed to investigate the potential causes of declines of bat population. Several studies have shown metal exposure and accumulation in bat tissues (Hernout et al., 2016a; Lisón et al., 2017) which could be contributing to the decline of bat populations in Sicily, and should therefore be further evaluated.

In this study, we choose to focus on two sites in Sicily. One used

as a control site and one as polluted site, based on its proximity (1 km) to a petrochemical plant (Fig. 1). In this polluted area, the industrial activities started in the late 1950s and 1960s and contain several oil refineries, chemical plants, mineral deposits, a military base and many other industrial installations. During the last few decades, the industrial activities have caused progressive contamination of the different environmental matrices through the presence of compounds that are mainly toxic, persistent and can bioaccumulate. The marine environment across the petrochemical plant has been more studied than the terrestrial environment and the overall results demonstrated a severe As, Cd, Cr, Hg, Pb, polychlorobiphenyls (PCBs) and polycyclic aromatic hydrocarbons (PAHs) contamination. The concentrations determined in sediments and seafood were exceeding the standard limit reported by national and international sediment quality guidelines (SQGs) (Ministerial Decree No. 260/2010; (De Domenico et al., 2013) and the standard limit for human consumption (D.lgs n.1881/2006), respectively. DNA damage, reduced genetic variability and oxidative stress were observed in marine organisms (Ausili et al., 2008; De Domenico et al., 2011; Di Leonardo et al., 2009; Ferrante et al., 2017b; Longo et al., 2013; Romano et al., 2016; Tomasello et al., 2012). The Italian National Institute of Health evaluated the toxicological properties of the dangerous substances in the petrochemical area and emphasized the correspondence between the types of contaminants and soil, groundwater and sediments. The recognition of an area as being at high risk entails priority allocation of resources, for cleaning up contamination and for rehabilitation in general.

The aim of this study was: 1) to assess the bioaccumulation of Arsenic (As), Cadmium (Cd), Cobalt (Co), Chromium (Cr), Copper (Cu), Mercury (Hg), Manganese (Mn), Nickel (Ni), Lead (Pb), Selenium (Se), Antimony (Sb), and Vanadium (V) in fur and liver of individuals of *M. myotis* sampled from two different caves used as nursery roosts in Sicily; 2) to compare the amount of metals accumulated in bat tissues between the two sites; 3) to compare the levels of metals with toxic thresholds for toxic metals and upper range values for essential metals in small mammals; and 4) to establish positive and negative correlations between metal concentrations determined in fur and liver. The present study is the first monitoring survey of trace elements in bats carried out in Italy.

2. Material and methods

2.1. Sampling area

The exposure of bats to metals was compared between two caves. Pipistrelli was chosen as control area and Palombara as polluted area since it is located at 1 km to a petrochemical plant of southern Italy (Fig. 1).

The control site is located in the Pantalica area, awarded in 2005 as UNESCO world heritage site for its history, archeology, speleology and landscape. The area is characterized by a nature plateau, deeply engraved by the quarries of the Anapo valley and the Calcinara river. Rocks are predominantly calcareous but we also found volcanoclastites, minor lava flows and diatremes (maars) associated with biocalcarene deposits (Grasso and Lentini, 1982; Savelli, 2001). The site is a variable of natural and semi-natural habitats along with arable land, which are essential habitats for the invertebrate and vertebrate communities. Pipistrelli cave is an important bat cave in Sicily since it hosts the largest colonies of bats and it represents the biggest nursery roost of Sicily (Spena et al., 2013). The following taxa were observed: *Rhinolophus ferrumequinum*, *R. euryale*, *R. hipposideros*, *Myotis myotis vel blythii*, *M. capaccinii* and *Miniopterus schreibersii* (Grasso et al., 2013; Spena et al., 2013). The maximum number of occurrences (taking into account all the

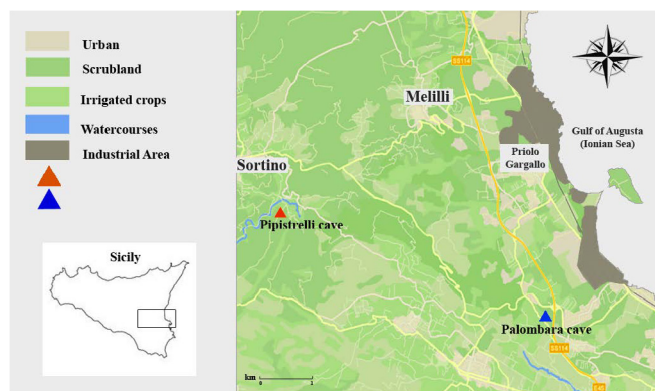


Fig. 1. Map showing the study areas (southern Italy – Sicily). In particular, the triangles indicate the geographic position of the caves and the color gradations indicate the different exploitation of the territory.

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