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SOME GENOTOXICOLOGICAL AND ECOTOXICOLOGICAL STUDIES ON AMPHIBIANS

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ABSTRACT

Amphibians contain some of the most endangered species and their population is declining worldwide. Amphibian population decline is related with habitat degradation, climate change, diseases, pesticide use, introduced species and pollution. Pollutants can affect amphibian habitats and population gene pool. The goal of this paper is to summarize some of the studies that have been done on the genotoxicology and ecotoxicology of amphibians.

INTRODUCTION

The pollutants and heavy metals in the water affect amphibians directly or indirectly as they spent some parts of their life in water. Anthropogenic toxicants in the environment can affect natural population's gene pool. Chemical substances cause mutation; affecting genetic diversity, population reduction and migration changes (1,2).
Toxicology studies on pollution in amphibians have increased in the last two decades due to the decrease in populations in the world (3). Amphibians, mammals and birds show a higher extinct in nature (4). Chemical contamination to their habitats is one of the factors that cause extinction in nature (5). Most of the amphibian ecotoxicology literature focuses on the effects of water pollution in early stage of their life. Studies about all stages of amphibian life is limited.
The metals in the study area of ecotoxicology, which studies the effects of environmental pollutants on living organisms, are biologically accumulating in the body. Some heavy metals give damage to organism by affecting the enzyme structure. Many studies have been done on amphibia on heavy

metals, metal toxicity and bioaccumulation in the contaminated areas (6,7). There is an increasing evidence that chemical contaminants are somewhat responsible for the reduction of amphibians (8). Heavy metal term refers to any metallic chemical element that has a relatively high density and is toxic or harmful even at low concentrations. Metals and metallic compounds are the natural components of all ecosystems and cycled within and between the atmosphere, hydrosphere, lithosphere and biosphere (9). Havy metals that causes contamination are copper (Cu), zinc (Zn), nickel (Ni), mercury (Hg), arsenic (As), chromium (Cr), cadmium (Cd), lead (Pb). High concentrations of metals such as Hg, Pb, Cd, Cu, Cr, Zn, Ni in the aquatic environment can be toxic to organisms, although some metals are necessary for the organisms (10). And also, if these contaminating agents interact with DNA, they can cause alteration of the genes and passed down to the next generations.

There is two methods of heavy metal deduction. One of them is to analyze the habitat (water, soil or sediment), other one is detecting heavy metal concentrations of organisms as bioindicators. Studies about the effects of pollutants on the amphibian population and the effects of pollutants on DNA is limited (3,11,12). The most commonly used genotoxicity tests to detect disorders that occur in amphibian DNA are; Chromosome aberration test, sister chromatid exchange test and micronucleus test (13).

In this review, studies on the effect of pollutants (heavy metals) on amphibian habitats and amphibian genetics have been examined. This review will shed light on the conservation action plan of endangered amphibian populations.

MATERIALS AND METHODS

The effects of contaminat is different on each organism. Cadmium is hazardous to animals whereas freshwater organisms are more sensitive to this metal. A possible accumulation of cadmium may be considered when the concentration in vertebrate organisms exceeds 2 ppm (14). Chromium has both lethal and non-lethal effects on wildlife organisms. From the previous studies it is conducted that amounts exceeding 4 ppm in tissues are indicative of chromium accumulation (15). Copper is a necessary element for living things, high concentrations can cause toxic effects. Data on copper concentrations in filed collections of amphibians are scarce (16). Iron has a toxic effect above 5 ppm concentrations. Lead is neither essential nor beneficial to organisms. Accumulation of lead in tissues may have a toxic effect (17). Zinc is an essential element for enzymes and proteins in living organisms. Zinc plays an important role in nucleic acid and protein metabolism and in cell division. The most sensitive aquatic species were adversely affected at nominal water concentrations between 10 and 25 μg/L, including amphibians (18).

Lefcort et al. (1998) conducted three different investigations on the effects of heavy metals on Rana luteiventris in the Ihoado valley and determined pollution. They investigated the effects of heavy metals on Rana luteiventris, their survival and duration. Over time, heavy metal deposits were observed in the Rana luteiventris habitats due to the silver mining and they found that the metamorphosis of Rana luteiventris was delayed (19). Freda argues that the most important role in the effects of acidification-induced heavy metals on frogs is aluminum (20). Sparling et al. (2010) examined the effects of heavy metal concentrations on three different species of amphibians. And they found that Al, Fe, Mg and Mn concentrations were high. Be and Sr concentrations which were high can be reduced by acidification process (3). Adlassing et al. (2013) observed eight locations that were found to be heavily contaminated with heavy metals and discussed the negative effect of heavy metals on amphibians (21).

Shin et al. (2008) examined the impact of sodium nitrate on early larval stage of *Epidalea calamita*. *Pelophylax perezi* ve *Hyla meridionalis* that were exposed during 10 to 16 days. They found that mountain populations were more sensitive to polluted enviranments that coastal populations. Their results showed that geographic and genetic variation and evolutionary adaptation of tolerance may also be the keys to variation amongst populations of the same species (11).
Xenopus laevis and *Pleurodeles waltl* larvae were investigated on the toxic and genotoxic potential of Cadmium using micronucleus test and comet assay. The results of micronucleus test showed that 2 mg/L concentration of Cd had a genotoxic effect on *Xenopus laevis* and there was no genotoxic effect on *Pleurodeles waltl* at all concentrations tested. According to the Comet assay test, the Cd genotoxic effect has been observed to be released from the first day of exposure. It has been determined that the comet assay test is more appropriate and sensitive in amphibians detecting genotoxicology (22). The toxic and genotoxic effects of $CdCl_2$ concentrations in *Rana limnocharis* tadpoles were investigated and it was determined that the rates of 0.1 and 0.4 mg/L had lethal effect. According to the micronucleus test and the comet assay test, $CdCl_2$ was found to be genotoxic (23).
According to International Nature Conservation Union (IUCN), 787 amphibian species are endangered (24). Amphibian population declines were widespread in occurrence around the world, due to habitat degradation, climate change, diseases, pollution, pesticide use, introduced species and pollution (4,25–27). Around 20 years ago researches point out the global crisis of amphibians, due time it is still topical (4,28). There are several studies to understand what is the major reason for the amphibian declines and appears to be no single cause for amphibian population declines.

RESULTS AND DISCUSSION

The pollution of the environment affects the living organisms in some way. Environmental pollutants are asscociated with physical, chemical and biological structure of the environment, the amount of pollutants, and depends on the species and size of the affected organism. When the pollutants are present in a small amount, the effect can be partly reduced by the ecosystem. When the pollution reachs the critical level, it affects the fauna and flora and can carry this effect up to the upper levels of food chain. Environmental condition changes may affect the sensibility of organisms to toxicants such as pesticides and heavy metals.
Amphibians have an important role in food chain. The toxicity studies on amphibians will give us a clear view about the amphiban population dynamis and potential accumulation in human beigns.
When we consider the protection status of amphibians, it is important to establish a wider database to understand how ecotoxicological agents affect populations. Genomic scans, including the identification of candidate genes responding to the novel selection pressure of ecotoxicological contaminants, are a promising area of future research to identify candidate populations for reintroductions and assisted migrations (29).

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