INTRODUCTION

As a consequence of agricultural, urban and industrial development, the chemical profiles of air, soils and water bodies are being altered. The alteration of the habitat may have deleterious effects on native flora and fauna. As we modify environment for our own needs, the destruction of the habitat of various species occurs that directly leads to the disappearance of many of them. The current global loss of biodiversity is a process generated by such anthropogenic interventions.

Pesticides have become an indispensible part of modern agricultural practices and one of the vital factors in increasing food production. No doubt, the use of pesticides has helped in an increase of the agricultural production, but their indiscriminate use has also led to destructions of many plants and animals. Every year millions of tons of agrochemicals are applied to soils in and around aquatic ecosystems that eventually pollute rivers, streams, lakes, wetlands and other water bodies. Despite the hazards that pesticides cause to the environment and human health, farmers apply ever increasing amounts of these toxic chemicals to their fields. The extensive use of pesticides to control pests and to increase agricultural output has resulted in their adverse effect on non-target species. The poisoning of agricultural fields by pesticides is a serious pollution problem and its environmental long term effect may result in the incidence of poisoning of fish and other aquatic forms.

The impact of environmental contaminants on amphibians is a subject that has received limited attention. It is possible that amphibians are responding adversely to the environmental changes. They are abundant and functionally important elements in many terrestrial and most freshwater ecosystems and constitute a significant component of world’s biota. They are the only class of vertebrates that do not include any pests or species harmful to man. Amphibians play key roles in many aquatic and
terrestrial ecosystems. As tadpoles they devour vast quantities of algae and other plants, thus regulating aquatic oxygen levels. On land adult amphibians act as excellent biological control agents as they consume large quantities of insects and other invertebrates. The pest control benefits of amphibians have long been recognized. Amphibians also support the many different types of animals that prey upon them: they are important in the diet of many reptiles, birds, mammals and freshwater fishes. Thus in both aquatic and terrestrial food webs, amphibians often play an important role in energy transfer and nutrient cycles. They have survived more than 300 million years through drastic environmental changes that led to the disappearance of dinosaurs and many other species. Yet scientists are alarmed by the recent rapid decline of amphibians in many parts of the world. In many instances their decline is attributed to adverse human influences acting locally such as deforestation, draining of wetlands and pollution.

Among amphibians, anurans, particularly frogs are likely to be exposed to dangerous levels of chemicals because of their higher concentrations in irrigation ditches, ponds and marshes. They are greatly valued as indicators of environmental stress as they are in close contact with water as larvae and have some contact with land as adults. Therefore, they experience both aquatic and terrestrial stressors. In light of their apparent sensitivity, and extremely permeable skin the frogs may serve as early warning systems for environmental degradation. There has been dramatic increase in sightings of deformed frogs all over the world, and as we choke our streams, ponds and wetlands with lead, mercury, fertilizers and pesticides, frogs are being poisoned and deformed on a massive scale.

During the last two decades amphibian decline has emerged as a key example of the global biodiversity crisis. Concern is so great for the phenomenon of amphibian declines, that the International Union for
Conservation of Nature and Natural Resources (IUCN) has set up the Declining Amphibian Population Task Force (DAPTF) to investigate the matter.

**Experimental animal: Euphlyctis hexadactylus (Lesson)**

*Euphlyctis hexadactylus* is one of the common amphibian species found in Kerala. They are distributed in countries like Sri Lanka, Nepal, India and Bangladesh. They live in and around water bodies, and water logged wetlands and paddy fields form their favorite dwelling places. They are known to be the natural enemy of agricultural pests, especially paddy pests. They are commonly used as a laboratory specimen and are large-sized frogs, females reaching 113 mm in snout-vent length and weighing up to 160 gms. Males are comparatively smaller than females, reaching 77 mm in snout-vent length and weighing up to 45 gms. Skin is smooth and slimy, bright grass green or olive green above and pale yellow below. The colour varies slightly with the habitat in which they live. A pale yellow vertebral stripe from snout to vent may be present in some specimens. They have a flat snout and a distinct tympanum. Toes are fully webbed.

**Pesticides used in experiments**

Amphibians are not likely to be killed by normal exposures to the pesticides in nature, due to low concentrations that are likely to occur in aquatic systems. Therefore it is important to assess the sublethal impacts of these chemicals on different aspects of these organisms. In the present study sublethal and median lethal effects of two commonly used pesticides – nuvacron and carbaryl – on the histopathological and biochemical aspects of the green frog *Euphlyctis hexadactylus* were investigated. Nuvacron is an organophosphate pesticide and carbaryl is a carbamate.
Nuvacron. Nuvacron is a soluble concentrate containing 360 gm monocrotophos as its active ingredient per kg of product. Its chemical name is dimethyl (E)-1-methyl-2-(methyl carbamoyl) vinyl phosphate. It is an organophosphorus insecticide and acaricide, which is a systemic as well as contact poison. It is used to control a variety of sucking chewing and boring insects, spiders and mites on a variety of cultivation including paddy in Kerala. It is a direct acting cholinesterase inhibitor capable of penetration through the skin. Many workers have studied the mode of action of organophosphorus pesticides. Primarily they appear to inhibit the working of the enzyme cholinesterase and so act on the nervous system and they are also responsible for a number of physiological and biochemical disturbances in vertebrates (Natarajan, 1981).

Carbaryl. Carbaryl (1-Naphthyl N-methyl carbamate) is one of the world’s most commonly used broad spectrum pesticide. Because of their low persistence carbamate pesticides have attained a wide spread acceptance as a replacement for the more traditionally used organochlorines (Mann and Bidwell, 1999). Carbaryl is a widely used carbamate insecticide used to control over 100 species of insects and is applied on a variety of crops, animals, ornamental plants and indoor areas. It is commonly sold under the brand name sevin and is formulated as a solid, which varies from colorless to grey depending upon the purity of compound. It is neurotoxic in action inhibiting the action of cholinesterase leading to the blocking of smooth transmission of nerve impulses. Muscle weakness and anorexia are caused by prolonged low level exposure to carbaryl. Its use has also been associated.
with a large number of health problems such as suppression of immune system responses, reproductive abnormalities and even genetic problems.

![Chemical structure](image)

**Significance of the study**

The fact that modern agricultural practices have their own contribution to the diminishing of our fauna and flora is not a matter of dispute. But its impact on amphibian population has not received much attention in India. As stated earlier, amphibians are of considerable significance to man and his environment. They are not only important to man but also to other animals within the community. They make major contribution to community biomass and play significant roles in the trophic levels as a predator of many invertebrates and prey to many vertebrates. The introduction of chemicals or other anthropogenic modifications to an aquatic habitat can produce profoundly different responses in consecutive life stages of a single amphibian species. The eggs of amphibians are naked without any protective hard shell and are susceptible to all sorts of aquatic pollution. Their reproductive seasons overlap with agrochemical applications that affect their reproduction since there is greater probability of exposure, particularly to pesticides in agricultural landscapes.

Frogs can be considered as good bioindicators because most of them spend time in freshwaters as aquatic eggs and larvae and out of water as terrestrial immatures and adults. Review of literature reveals that they show moderate to high sensitivity in acute and chronic sensitivity tests compared to other aquatic organisms. At present there are no regulatory criteria on toxicants for frogs. Instead, data from fish studies are often
assumed to provide knowledge about toxicants on frogs also. This implies that fish and frogs are equally sensitive to the toxicants for which criteria have been established. But Birge et al. (2000) compared the toxicity of a variety of toxicants to amphibians and many fish species. In all, 50 metals and compounds were tested. Their results showed that amphibians have lower LC50 values than fishes. The researcher’s overall conclusion was that there exists great variation among amphibian species in their sensitivity to metal and other organic contaminants; they generally are more sensitive than fishes and the water quality criteria established for fish may not be protective of amphibians.

Hence, a scientific understanding of ecotoxicological aspects of amphibians in general and frogs in particular is relevant and justifiable. Although toxicants impair the metabolic and physiological activities of the organisms, physiological studies alone do not satisfy the complete understanding of pathological conditions of tissues under toxic stress. Therefore, a histological analysis of the vital tissues along with a biochemical study is inevitable.
REFERENCES


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