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Ecotoxicology: Why is it a discipline of growing importance?

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Ecotoxicology is the study of the toxic effects of chemicals on biological organisms, at the level of the organism, population, community or ecosystem. It is a multidisciplinary field, which integrates broad areas of toxicology, ecology and environmental chemistry. Its ultimate goal is to predict adverse effects of environmental contaminants and devise efficient and effective means of action to prevent or rectify any identified detrimental effects. In instances where ecosystems have already become severely degraded due to environmental pollution, ecotoxicology will allow us to take the best course of action to restore the system to a healthy state.

The beginnings of ecotoxicology

It is not wrong to say that *Silent Spring* by Rachel Carson published in June 1962 had the greatest impact on the attitude of humans towards environmental contamination. It was the single most influential book that caused an awakening to the reality that substances synthesized and used for our benefit may cause considerable harm to non-target organisms and the environment. Six years after its publication, a sinister effect of pesticide use was documented: the accumulation of DDE, a compound produced when DDT is degraded, were causing reproductive failure in several predatory birds, including Peregrine falcons, Brown pelicans, Osprey and Bald eagles. Not only was this compound toxic to developing embryos, it also caused eggs to have abnormally thin shells that easily cracked under the weight of the adult bird during incubation. This resulted in the ban of DDT in 1972. It was only much later that the true impact of Rachel Carson's book was realized, and in the 1990s it was named 'one of the 25 greatest science books of all time' by the editors of the Discover Magazine. According to Rodrick (1992) this book created fears in humans about synthetic chemicals in the environment and, among other things, fostered interest in the science of toxicology.

Ecotoxicology could be considered a science of the twentieth century, which had its early development in the late sixties. The term ecotoxicology was first used by Truhaut in 1969 (Truhaut 1977). He defined ecotoxicology as "a branch of toxicology concerned with the study of toxic effects caused by natural or synthetic pollutants to the constituents of the ecosystem, animals, vegetables and microbes, in an integral context". This differed significantly from the 'classical toxicology' which was mainly concerned with the effects of contaminants at subcellular to organism levels, with a heavy emphasis on humans. Prior to the 1960s, little attention was paid to the impacts on populations and ecosystems. The radical shift from toxicology to ecotoxicology resulted from the recognition that the passage of chemical constituents in organisms through food chains may have consequential impacts at levels beyond the organisms that are directly affected.

This field has now progressed far beyond these simple beginnings, with the past few decades becoming exceptionally exciting for the ecotoxicologist. Increasing quantities of deleterious substances are being relentlessly discharged into natural ecosystems, while there has been a transformation of fundamental sciences such as ecology and toxicology into more complex and applied disciplines. We are now in a better position to understand the mechanisms and pathways of contaminants and therefore to predict their impacts on the environment. Consequently, recent years have seen dramatic advances in our recognition of the impacts on species and ecosystems.

Evidence of adverse impacts of pollutants on species and ecosystems

A contaminant is described as a biological, chemical, physical, or radiological substance which is not usually present in the environment and which, in sufficient concentrations, can adversely affect living organisms. Such contaminants may include deleterious gases, agrochemicals, heavy metals, radioactive compounds and pharmaceuticals which are being discharged into the environment in increasing quantities principally through anthropogenic activities. These environmental contaminants enter organisms primarily via inhalation, ingestion or absorption through the nose, mouth or skin. Once entered, the substance can elicit both direct and indirect toxic effects in the exposed organism. Some contaminants are toxic in any quantity, while others exert toxic effects only when undesirable levels are reached. The manifested effects can also be described in terms of their magnitude and duration. A direct lethal effect is known as an acute effect while a prolonged effect is termed chronic. From the late 1960s scientists have been investigating the modes of action, levels of accumulation and the manifested toxic effects of various contaminants in different test organisms and, consequently, a wealth of information has been generated. Let us look at some of the documented findings of other ecotoxicologists and of my work with other co-workers in the Department of Zoology, University of Colombo.

At the level of the organism, enhanced mortality is one of the most commonly observed direct effects, which has been demonstrated for many species exposed to ecologically relevant levels of aquatic toxicants. Some examples are the high levels of mortality in fish and amphibians exposed to pesticides and heavy metals (Lefcort *et al.* 1998; Elskus 2007; Wijesinghe and Ratnasooriya 2010; Ranatunga *et al.* 2012). Such impacts have been also shown, to a lesser extent, in higher taxonomic groups. In 1989, the Environmental Protection Agency of the USA estimated that carbofuran alone kills 1-2 million birds in that country every year (Schauber *et al*. 1997). In 1995, it was reported that the pesticide monocrotophos, sprayed to kill grasshoppers, was responsible for the deaths of at least 20,000 Swainson's Hawks in Argentina (Hooper *et al.* 2002). They state that birds swallowed pesticide granules directly mistaking it for seed, and indirectly, by consuming contaminated voles and deer mice.

Apart from direct mortality, scores of studies have established that contaminants bring about sublethal effects. Among them are growth retardation, delayed development and metamorphosis, and behavioural changes with notable declines in activity. Deformities have been also induced by contaminants. An example would be the loss of limbs and spinal cord deformities in frogs induced by pesticides (Jayawardena *et al.* 2011). Additionally histopathological alterations have been reported to occur in critical organs such as the liver, gills and kidney. For instance, Bandara *et al.* (2008) have shown degenerative changes in the liver and muscles of the Asian common toad exposed to field levels of pesticides. The hematological profile of a species is a good indicator of the levels of environmental stress because blood parameters are altered with exposure to pollutants as revealed in many a study (e.g. Parma *et al.* 2007). Immuno-suppression has also been noted in certain species (Halloran *et al.* 1997). Others have demonstrated genotoxic effects (Reicher *et al.* 1999) which are triggered by exposure to environmental contaminants and include structural changes to the DNA, which if not repaired, can lead to the appearance mutations.

What is of concern is that, while sublethal damage may not always cause direct mortality, there is a strong possibility that reduced survival may occur in the long term. Some of the undesirable sublethal effects may result in decreased life span and reduced breeding success. Lethargy coupled with smaller sizes of exposed individuals reduces breeding success, makes them more susceptible to predation or reduces their competitive ability at securing food resources (Crossland 1998). Decreased nutrition causes growth retardation that in turn leads to reproductive failure. Inhibition of the innate immune defense system can also lead to greater susceptibility to lethal pathogens. Davidson *et al.* (2007) has shown that carbaryl exposure increases susceptibility of the foothill yellow-legged frogs to the chytrid fungus which is widely associated with amphibian declines.

The magnitude of the manifested effects in organisms exposed to a contaminant will depend on several factors relating to the scale of the dose and the duration and pattern of exposure. It will also be very much dependent on the species, age, stage of development and the health status of the exposed animal.

The effects of contaminants on organisms will have connotations at the population level. The enhanced mortality and the diminution in fitness will lower rates of recruitment in the long run causing populations to decline in some cases to the point of local extinction. This in turn will lead to narrowing of species distributional ranges. Here in Sri Lanka, we are constantly made aware of the increasing numbers of species that have become nationally threatened; pollution has been identified as one of the major contributors. Groups particularly at risk are the bees, butterflies, fish and herpetofauna. It is clearly apparent that many species that were once common in agro-ecosystems are now far less common or extremely rare.

What impacts could one expect at the community or ecosystem level? Any serious changes in species populations would inevitably cause imbalances and lead to malfunctioning of entire ecosystems. Soil contaminants have been shown to inhibit growth and seed germination of plants and because they are at the bottom of the pyramid, every other organism in the ecosystem would consequently be affected. Animals have gone to the brink of extinction because of the feeding associations that exist within communities. In some cases the predator is affected by a toxin resulting in a decline of predator populations, which then results in an unlimited expansion of the prey population, which in turn brings about destruction to the entire ecosystem. Sometimes prey are affected resulting in a decline in the predator population. Because of the differences in tolerance levels some adaptable species may also stand to benefit and expand at the expense of the less tolerant specialist species. Local population extinctions will evidently bring about changes in species composition and shifts in food habits of species.

Some substances (e.g. organochlorines) possess two dangerous traits – persistence i.e. the ability to remain chemically active for a long time and the solubility in fat resulting in accumulation in fatty tissues within organisms over time. This is commonly referred to as bioaccumulation. Because of these two traits, contaminants can be transported along a food chain with the harmful substances generally becoming more concentrated at the higher trophic levels, a process known as biomagnification. Some exceptions have, however, been documented. A combined effect of bioaccumulation and biomagnification would probably explain why the invertebrate community in a pond was found to be significantly different in species diversity and abundance from the original community two years after the application of the insecticide fenvalerate (Relyea 2005). The effects of environmental toxicants therefore have dangerous ramifications that flow from organisms through to populations and communities and ultimately to ecosystems.

Increasing deterioration of the natural environment

All over the world, the ongoing discharge of pollutants in the form of industrial effluents and domestic sewage and effluents has caused considerable and irreversible degradation of our natural habitats which makes ecotoxicology a discipline of growing importance. This interest is particularly strong in aquatic systems where many non-target organisms are more frequently exposed to deleterious substances. Therefore let me, as example, consider the pollution of the freshwater ecosystems stemming from the intensified use of agrochemicals in Sri Lanka.

Pesticides were first introduced to Sri Lanka in the 1940s to control malaria, but since then, there has been a significant rise in the import and application of agrochemicals in the country. According to the Registrar of Pesticides the total quantity of pesticides imported to the country has doubled over the last decade. The insecticide formulations imported increased from 849 tonnes in 2001 to 1713 tonnes in 2011. The respective values for herbicide and fungicide formulations are 2056 and 553 in 2001 and 5031 and 949 in 2011. According to a World Bank report, farmers in Sri Lanka use 284.3 kg of fertilizer per hectare of arable land which is far greater than that in other countries of the Asian region (India – 153.5 kg.; Bangladesh – 164.5 kg; Indonesia – 189.1 kg). The development of the agricultural sector over the years, particularly in the dry zone with the Accelerated Mahaweli Project, has no doubt intensified the use of agrochemicals. Agrochemicals and their residues enter waterways directly, or as surface runoff contributing heavily to the problem of water pollution in these areas. Despite the seriousness of the situation, only a handful of studies have attempted to record levels of pesticide residue in water. A study in Polonnaruwa, revealed the presence of Propanil (1.02 - 1.18 mgl^{-1}) and Chlorpyriphos (1.45 - 6.77 mgl^{-1}) in water samples collected from the Meegalawa Wewa, where irrigated agriculture was practiced. More information exists, nevertheless, with regard to nutrient enrichment in both surface and ground water sources in agro-ecosytems. Ground water, in intensively cultivated areas, typically have nitrate concentrations in the range of 10-15 mg 1^{-1} compared to 0.2 mg 1^{-1} within the non-cultivated lands. A study of 58 irrigation reservoirs across the island revealed that all of them were in a state of eutrophication in both paddy growing seasons, having total phosphorus levels of more than 30 μgl^{-1} (Yatigammana *et al.* 2007). In fact, 79% of the wells in the dry zone had nitrate concentrations which exceed levels recommended by the World Health Organisation (WHO) for safe drinking water. Leaching of agrochemicals from intensively cultivated soil is also responsible for elevated concentrations of chloride, nitrate and potassium observed in many irrigation wells in the Kalpitiya peninsula (Lawrence and Kuruppuarachchi 1986). Consequences of excessive nutrient pollution i.e. eutrophication has been reported from many areas across the country. Eutrophication is also a matter of concern in the Kotmale reservoir (Piyasiri 2000). Phosphate enrichment has been reported from Nuwara wewa and to a lesser extent in Tissa Wewa in the Anuradhapura district. Nutrient enrichment is occurring in the Kandy Lake and Lake Gregory in the hill country. In all these cases increased nutrient loads in water have been largely attributed to the excessive use of fertilizers in the catchment areas.

The future challenges for ecotoxicologists

The merging of the two disciplines ecology with toxicology has resulted in the relatively young field of ecotoxicology. Despite the wealth of evidence generated so far, we have only yet begun to address the diversity of problems and questions concerning the effects of pollutants on individuals, species and ecosystems. Some of the areas that pose challenges in the ecotoxicological arena are given below.

One of the most common methods of assessing effects of various pollutants at the basic level of the individual is through empirical trials. Standard toxicity tests are carried out exposing selected study species to different concentrations of the pollutant to observe its effects. There are, however, several limitations in the selection of taxa/species for conducting such laboratory-based toxicity tests. Some taxa (e.g. amphibians and fish) have been found to be ideal indicators of the levels of pollutants in water bodies because of their high sensitivity to environmental stressors. Additionally, other characters that make species suitable for such tests would be short generation time, ease of rearing and handling. Apart from practical constraints, there are ethical issues that prevent the use of animals from higher taxa (birds and mammals). We must, however, devise practical and ethically acceptable means of investigating or making realistic predictions of the adverse impacts on these species as well. It should also be borne in mind, before generalizing, that the toxicity of a given substance may be highly variable between taxonomic groups and even between related species.

There are also major problems in extrapolating the findings generated from empirical trials to natural scenarios. One of the basic problems, particularly in countries such as Sri Lanka, is the lack of quantitative data on the levels of pesticides found in the water bodies within the country. Exposure trials will become more realistic only when field levels of the tested pollutants are used. In the case of agrochemicals in particular, it has been the view of some toxicologists that research should always focus on formulated products entering the natural environment, instead of assessing effects of the individual active ingredients. This stems from findings that commercial products elicit effects that are different to those induced by the active ingredients. A case in point is the northern leopard frog which suffered reduced larval growth and deformities when exposed to the formulation round up but not when exposed to the active ingredient glyphosate (Howe *et al.* 2004). We need also to take into consideration that, in the field, more often than not, pollutants occur as mixtures, making it difficult to identify abnormal conditions and relate them to likely causes. Effects are, at times, accelerated because of synergism among environmental pollutants where the combined effect of two or more pollutants is greater than that caused by the individual pollutants acting alone. Also, some pollutants may react with natural elements in the environment forming substances that have a greater potency than the primary compound. Another confounding problem is the alteration of toxic effects by environmental factors such as solar radiation, sediment loads and pH. These factors may facilitate the conversion of the active ingredients to more potent or less potent substances which in turn would alter the magnitude of the manifested effects.

It is because of these complications that systematic field studies are limited both here and elsewhere in the world. Under the prevailing conditions, therefore, standard toxicity trials using realistic exposure levels would, without doubt, provide a strong basis for predicting the toxicity of various contaminants on non-target organisms.

One of the areas where information is most lacking is how the effects of pollutants on individuals affect population dynamics. For example, we must know, or at least be able to predict, what concentrations of a particular pollutant cause levels of mortality that would in turn affect population recruitment and growth. To make extrapolations from individuals to species, we need to also know more about the factors that bring about natural mortality.

In conclusion, it is my belief that scientists involved in ecotoxicological research fervently hope that their findings would lead to a better understanding of the impacts of contaminants on organisms, species and ecosystems. Hopefully this should in turn indicate possible ways of efficiently managing and conserving natural ecosystems and providing a safe and healthy environment for our present and future generations.

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