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Oration

Pesticide Effects on Earthworms: An Integrated Approach to Tropical Risk Assessment

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Intensification and Expansion of agriculture in tropical regions is heavily relying on the use of pesticides. Pesticides however, are generally toxic to non-target soil organisms and as a consequence may hamper proper functioning of the soil. Therefore, to comprehend about the sustainability of tropical agriculture, information on the effects of pesticides on beneficial soil organisms like earthworms, termites, springtails which play an important role in the soil ecosystem are required. Nevertheless, pesticide toxicity under tropical conditions is only rarely assessed and understanding the effects in such conditions remains a priority to date. Tropical risk assessment often is relying on data generated under temperate conditions (De Silva, 2009., Owojori *et al,* 2013). Such an approach is rather questionable, mainly due to deviating climatic conditions. Consequently, it is essential to understand pesticide toxicity and the consequent ecological effects under tropical conditions.

Temperature and soil type may act as critical factors that modify pesticide toxicity. The effects of three commonly used pesticides, chlorpyrifos, carbofuran and carbendazim, on the survival, growth and reproduction of the standard test species *Eisenia andrei* were investigated in different soil types at representative temperate and tropical conditions. In case of survival, toxicity of chlorpyrifos and carbofuran in artificial soil was higher at 26 °C than at 20 °C. But in natural soils, carbofuran was most toxic at 20 °C while chlorpyrifos remained more toxic at 26 °C. The higher activity of earthworms at higher temperatures may trigger a higher uptake of pesticides causing larger effects on survival under representative tropical conditions. However, sub-lethal endpoints, such as growth and reproduction, did not show clear differences between the tested temperatures and varied with different soil types indicating that the nature of the pesticide and soil type were more important than temperature. Effects of carbendazim on survival, growth and reproduction of *Eisenia andrei* were lower at 26 °C than at 20 °C suggesting temperature may also influence the stability of the pesticide and its degradation kinetics, resulting in a lower toxicity at higher temperatures. It is suggested that pesticide toxicity may differ with temperature and soil type; therefore temperate data should only be used with caution in tropical risk assessment (De Silva *et al,* 2009., De Silva, 2011., De Silva, 2013).

Tropical risk assessment currently uses data generated with the temperate com-

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post worms *Eisena fetida* and *Eisena andrei*. These species are less ecologically relevant as they are rarely found in natural soils. One good alternative is to use indigenous species specific to tropical regions. Therefore the toxicity of chlorpyrifos, carbofuran and mancozeb to the tropical earthworm species *Perionyx excavatus* was investigated. In addition effects of both pure compounds and formulated products were compared to estimate the possible added toxicity of formulated products that is often neglected in toxicity studies. Carbofuran was most toxic to *Perionyx excavatus* under tropical conditions followed by chlorpyrifos and mancozeb. In case of survival, comparison with toxicity data generated for chlorpyrifos and carbofuran revealed that *Perionyx excavatus* was more sensitive than the standard species *Eisenia andrei*. However, for sub-lethal effects differences in sensitivity were relatively small. Generally, the toxicity of formulated products was higher than that of the pure compounds (De Silva *et al*, 2010b., De Silva & Amarasinghe, 2008). Nevertheless, results suggest that the toxicity of formulated products may be masked by interactions with the soil resulting in more or less similar toxicities of formulations and the pure compounds. In addition studies show that *Perionyx excavatus* may be used as a good alternative test species under tropical conditions in future studies, applying the available standard test guidelines for *Eisenia* sp.

Lack of cost-effective tests with short duration often hampers the risk assessment of pesticides under tropical conditions. Recently the earthworm avoidance test has been considered a good alternative in toxicity studies. Earthworm avoidance tests were performed with the standard temperate test species *Eisenia andrei* and the tropical earthworm species *Perionyx excavatus*, using a in two-chamber test system with artificial soil and a selected natural soil at a representative tropical temperature (26°C). The EC_{50} values for the effects on avoidance behaviour for both chlorpyrifos and carbofuran indicated that *Eisenia andrei* was a factor of 2-3 more sensitive than *Perionyx excavatus* under tropical conditions. An important finding was the attraction of *Perionyx excavatus* at lower concentrations of both pesticides tested and in both soils; hence it was suggested to use avoidance tests with local species with extra caution as it might lead to wrong estimations of the effects. Comparison with literature data showed that endpoints generated through earthworm avoidance tests are generally less sensitive than reproduction but more sensitive than survival and therefore could be used in the initial risk assessment to facilitate pesticide regulation in tropical regions (De Silva & van Gestel, 2009a., De Silva & Seniviratne, 2008., De Silva, 2009)

The standard test guidelines for earthworm toxicity testing often use the OECD artificial soil, which is composed of sand, kaolin clay and sphagnum peat. Development of a tropical artificial soil is timely needed due to increasing costs, non-availability and environment concerns associated with sphagnum peat. The suitability of locally available, environmentally friendly substrates such as coco peat (composted and non-composted), saw dust and paddy husk was investigated. Modified artificial soils were prepared by sub-

stituting sphagnum peat in the standard artificial soil. Survival, growth and reproduction of *Eisenia andrei* in the modified artificial soils were tested in the first phase with comparison to standard artificial soil. The results indicated the suitability of modified artificial soils prepared with composted coco peat and paddy husk (De Silva & van Gestel, 2009b., Hansani *et al,* 2013). During the second phase toxicity of chlorpyrifos, carbofuran and carbendazim for *Eisenia andrei* was determined to validate the two selected substrates. For all three pesticides LC_{50} values found in standard artificial soil and soils modified with composted coco peat and paddy husk did not differ significantly. EC_{50} values for the effects on reproduction were similar in standard artificial soil and soil modified with composted coco peat but lower in soil modified with paddy husk. These results suggest that composted coco peat might be a good alternative for sphagnum peat and can be used in tropical ecotoxicology. Standardization of this new substrate however, warrants further studies.

Effects of pesticides on structural properties (species diversity, abundance, biomass) and soil functions such as organic matter breakdown & nutrient cycling in soil ecosystems have rarely been studied in the field under tropical conditions. And predicting field effects based on data from laboratory studies is a challenge. Hence effects of chlorpyrifos on the diversity, abundance and biomass of earthworms and termites in relation to organic matter break down were studied in tropical soil. To link laboratory results and realistic field conditions, doses relevant to normal agricultural practice, representing the laboratory EC_{50} for earthworm reproduction and double the EC_{50} were applied. Diversity, abundance and biomass of earthworms and termites were observed prior to application and 1, 2, 3, 6 and 12 months after pesticide application. In addition, using litter bags, organic matter breakdown was investigated until complete decomposition in the control plots was achieved. Chlorpyrifos caused adverse effects on decomposition even 2-3 months after application. Biomass and abundance of earthworms and termites were also reduced and the decrease could be linked with decreased litter decomposition. Nevertheless effects on termites and earthworms were diminished after 6 months and populations were completely recovered after 12 months. The faster litter decomposition rates under tropical conditions showed the importance of modifying the current standard guidelines on effects of pesticides in field conditions for use under tropical conditions (De Silva *et al,* 2010a., De Silva, 2013)

The extrapolation of laboratory studies to real time filed situations still remains a challenge. As an example, effects of pesticides on structural properties (species diversity, abundance, biomass) and functions such as organic matter breakdown, nutrient cycling and soil formation of soil ecosystems have rarely been studied in the field under tropical conditions hence future experiments based on litter bag test and earthworm field test that have been introduced and standardized by the OECD should be encouraged under tropical conditions. In addition it should be noted that incorporation of generated scientific information into policy decisions still remains very poor and effective mechanism is urgently needed.

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