

Lecture: free communications

***In vitro* model for evaluation of zineb induced oxidative stress, immunosuppression and apoptosis**

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Pesticides usage has become an ever increasing phenomenon despite their adverse effects on environment. Exhaustive information is available on the pesticides and their deleterious effects on animal and human health, but it is painful to note that very little emphasis has been directed towards studying pesticide induced immunosuppression and apoptosis employing *in vitro* cell culture system of avian splenocytes.

Zineb is a dithiocarbamate fungicide with some insect repellent properties. It is a metabolic poison of low acute toxicity to mammals: Zineb is toxic to zinc sensitive plants. It has also been used extensively in the rubber industry as a promoter of vulcanization. Present communication evaluates oxidative stress, immunosuppression and apoptosis in chicken lymphocytes due to *in vitro* exposure of low level dose of zineb employing nitric oxide estimation, lymphocyte proliferation assay, cytokine assay, annexin V assay and DNA fragmentation assay, respectively.

Splenocytes were isolated and treated with thousand times dilution of No Observable Effect Level (NOEL/103) dose of zineb. Chicken lymphocytes displayed elevated level of Nitric Oxide when exposed to zineb. Lymphocyte proliferation assay revealed significant decrease in B and T lymphocytes levels. In cytokine assay there was down regulation in interleukin I and II levels in zineb treated cells as compared to control cells. Thus zineb was found to be immunotoxic even at low dose level. Annexin V assay revealed increased number of lymphocytes undergoing apoptosis. It was further confirmed by agarose gel electrophoresis of genomic DNA of zineb exposed cells which displayed typical apoptotic DNA ladder pattern. It is also worthwhile to mention that such *in vitro* tests in immunotoxicology can be useful in the quick preliminary evaluation and toxicity assessment of pesticides and will be helpful in reducing the use of laboratory animals.

Keywords: zineb, immunotoxicity, DNA fragmentation, oxidative stress, cytokine assay, apoptosis, avian lymphocytes, annexin V assay