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**An Investigation into the Presence of Organochlorinated
Pesticides in Fruits and Vegetables Grown in Lancaster County**

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Abstract

Chlorpyrifos is a heterocyclic organophosphate neurotoxin used as an effective broad-spectrum pesticide. Literature implicates that it may also be a teratogenic agent. Laboratory studies were conducted to detect the presence of chlorpyrifos in agricultural products grown in Lancaster County. Acetone extraction was used to prepare sample for analysis. Enzyme-linked ImmoSorbant assay was performed on all samples to confirm the presence of the pesticide and its metabolic derivative. Studies reflected seasonal usage of the pesticide with detectable concentrations as high as 374 ppb in some produce. Levels of the pesticide in produce exceed the LD₅₀ of 4 ppb.

Introduction

Chlorpyrifos [o,o-diethyl,O-(3,5,6-trichloro-2-pyridinyl)phosphorothioate] is a broad-spectrum organophosphate insecticide. It was introduced in 1965. Chlorpyrifos is effective in controlling a variety of insects, including cutworms, corn rootworms, cockroaches, grubs, flea beetles, flies, termites, fire ants, and lice. It is used as an insecticide on grain and cotton fields, fruit, nut and vegetable crops, as well as on lawns and ornamental plants. It is also registered for direct use on sheep, turkey, for horse site treatment, for treatment of dog kennels and for domestic dwellings. Chlorpyrifos is available in emulsifiable concentrate, dust, pellet, pray, and granular and wettable powder formulations. Chlorpyrifos is sold as Dursban[®] for domestic use and as Lorsban[®] for agricultural use.

Chlorpyrifos acts on insects primarily as a contact poison. Animals exposed to the insecticide exhibit stomach poison symptoms. In plants the poison effects are confined in the tissue where it comes into direct contact with the plant and is not transported to other plant parts.

Chlorpyrifos is one of a class of insecticides referred to as organophosphates. These chemicals act by interfering with the activities of cholinesterase, an enzyme that is essential for the proper working of the nervous system of both humans and insects degrading acetylcholine to choline and acetate.

In general, organophosphate pesticides are rapidly metabolized in the body by multifunctional oxidases (MFOs) on the P₄₅₀ cytochrome system located in the liver to allow solubility in the blood. This pathway leading to solubility does one of two things: it either detoxifies the harmful compounds or it increases their toxicity. Chlorpyrifos does the latter since it is metabolized to chlorpyrifos-oxon, a more toxic metabolite, and finally to 3,5,6-trichloro-2-pyridinol (TCP), the excreted form of chlorpyrifos.

In this study, we wanted to determine if chlorpyrifos is used in the fruits and vegetables of Lancaster County and, if it is, what kind of seasonal variations, if any, are evident. The method chosen to detect chlorpyrifos is by a modified enzyme linked immunoabsorbent assay (ELISA) technique. This technique is based on combining selective antibodies with sensitive enzyme reactions to produce analytical systems capable of detecting minute levels of chemicals.

The immunochemical reaction was selected because it contributed high selectivity since antibodies have a discriminatory property and high sensitivity since enzymes are capable of powerful catalytic activity.

Procedural Information

Enzyme-Linked Immunosorbent Assay

A. Assay and Intended Use

The Chlorpyrifos Rapid Assay? from **Strategic Diagnostics (SDIX)** was selected for detection of chlorpyrifos, which is capable of detecting the pesticide in water, soil, crop and foods, including eggs and tissue.

B. Procedure

The Chlorpyrifos Rapid Assay? applies the principles of enzyme-linked immunosorbent assay (ELISA) to the determination of chlorpyrifos. The acetone extracted sample to be tested (produce or tissue) was added, along with an enzyme conjugate, to a disposable test tube followed by paramagnetic particles with antibodies specific to chlorpyrifos attached. Both chlorpyrifos (which may be in the sample) and the enzyme labeled chlorpyrifos (the enzyme conjugate) compete for the antibody binding sites on the magnetic particles. At the end of the incubation period, a magnetic field was applied to hold the paramagnetic particles (with chlorpyrifos and labeled chlorpyrifos analog bound to the antibodies on the particles, in proportion to their original concentration) in the tube and allow the unbound reagents to be decanted. After decanting, the particles were washed with Washing Solution.

The presence of chlorpyrifos was detected by adding the enzyme substrate and the chromogen. The enzyme labeled chlorpyrifos analog bound to the chlorpyrifos antibody catalyzed the conversion of the substrate/chromogen mixture to a colored product. After an incubation period, the reaction was stopped and stabilized by the addition of acid. Since the labeled chlorpyrifos (conjugate) was in competition with the unlabeled chlorpyrifos (sample) for

the antibody site, the color developed is inversely proportional to the concentration of chlorpyrifos in the sample.

Results and Conclusion

A standard curve for both chlorpyrifos and TCP assays allowed concentrations in produce to be determined (Figure 1). As shown by Table 1, California grapes contained the highest concentration of chlorpyrifos (374 ppb) followed by apple skin (373 ppb), apple pulp (362 ppb), corn (307 ppb), and with peach having the least (11-30 ppb). All of these levels exceed the LD₅₀ of 4 ppb. No detectable amounts of pesticide were evident in summer fruits and vegetables.

In conclusion, levels of chlorpyrifos exceed the LD₅₀ of 4 ppb in Lancaster County's produce.

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