# DETERMINATION OF DDT AND DDE RESIDUES IN COTTON, *GOSSYPIUM HIRSUTUM* FOLIAGE

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**Abstract**

DDT and DDE as DDT metabolite on cotton foliage were studied. Seven rounds of DDT 50EC @1.5 liter /acre were deployed on cotton crop. First deployment was made after 40 days of spouting, thereafter, six subsequent applications were made at the intervals of 20, 20 15, 30, 15 and 20 days. Initially cotton foliage samples were drawn at 10 days post treatment and thereafter, sample were collected after an interval at 90 days age of the crop. The crop was found contaminated maximum with 0.25 mg/kg DDT and its metabolite as DDE and minimum 0.15 and 0.19 mg/kg DDT and its metabolite, respectively, while DDD was not detected in any case, employing GC-ECD.

## Introduction

Nowadays there is lot of worries about the affects of residues pesticides at the post application period, those stay in the agro produce up to the time of consumption. Preferably, human consumption articles should remain free from insecticide or alike biologically active material, but for the crops protection purpose it become essential to apply with insecticides and thus residues may leave, consequently. However, to some extent insecticides application is unavoidable. Nevertheless, there are two reasons to measure insecticides residues on plants. The first is to determine at what time the quantity of insecticide remaining become at a minimum level that makes it out of consideration as its hazard level, this give the minimum period between treatment and harvesting. The second is to find out how longer the chemical remain to provide a good protection as some insecticides in residual status work as repellant or deterrent etc so that the maximum benefit of used insecticides could be obtained, such economy in the insecticides usage would saves capital and pesticide usage attempts to reduction the latent residues risk.(Baig et al, 1972) It is considerable that if the chemicals are decomposed or metabolized it is important to be sure that the decomposed products are nontoxic. Detection of metabolites of unknown composition is difficult and perhaps the most satisfactory way of following the decomposition or metabolism of traces of insecticides is to use compound labeled with radioactive isotopes, which can be followed and used to trace the decomposition of metabolic products from the labeled chemicals, since the proteins are known to have affinity with particular insecticides therefore, specific protein based techniques are also useful in certain cases. Because persistence of pesticides is related with the environment where it is used and it fluctuates with respect to position, thus it is necessary to make such studies where an insecticide is used.

Nevertheless, the nearly all the persistent Organo-chlorine insecticides are deregistered compounds in this country. However, despite legislations against such compounds, it is anticipated that to overcome pest problem instantly, farmers may cover their crops with persistent insecticides like DDT, as Khan (1998) has indicated evidence that cotton bollworm may even develop insecticide resistance up to 2000 fold against pyrethroid therefore, without consideration of hazardous consequences, the worried small farmer jumps to promising persistent OC. In this scenario it is conceivable that somehow, though at a little scale, DDT is still in use. Therefore, Iram et al (2009), Ahad et al. (2005) and Anwar et al (2012) have reported it from Lake environment and soil respectively, Uddin (2011) and Khan *et al*. (1996) have reported it on food produce in Pakistan. However, Khan et al (1996) suggested that only small farmers are using deregistered products on locally consumable produce. Presently, a small experiment was conducted to determine extent of the DDT and it metabolites residues in cotton crop during and after spray period.

## Materials and Methods

The crop was treated with DDT seven times. First deployment was made after 40 days of spouting, followed by 6 more at intervals of 20, 20 15, 30, 15 and 20 days, depending on the situation of pest appearance. Stage of picking of cotton was reached when the crop was nearly 3 months old. Each sample was collected 10

days after spray on crop; the cotton foliage samples were preserved in a deep freezer before proceeding of extraction and clean-up. Fortification of untreated cotton samples with respective insecticides and metabolites was made as five dilutions of analytical grade of DDT, DDE and DDD were prepared in 50, 100, 200 and 500 µg/10ml in n- hexane as solvent. These dilutions were added to untreated cotton samples, and seed for fortified samples' determination of recoveries. Moreover, GC-standard curves for retention times and standard peak were also obtained from these dilutions for the comparison. A four hours Soxhlet extraction in 1:3 acetone: n-hexane solvent system was employed, for the extraction of DDT & metabolites from crude crop samples. The obtained material was first dehydrated through passing over anhydrous sodium-sulfate. For the extraction and cleanup, 50 ml of the extract was mixed with 50ml dimethyl formamide (DMF) in a separating funnel and agitated strongly there after left stand to allow the layers separate. DMF was taken into another separating funnel. The n-hexane layer was washed two times with 25 ml 92% DMF and obtained DMF layers were collected in the same separating funnel and combined DMF was mixed with 400 ml 4% sodium sulfate thereafter, agitated strongly and left on a stand for 15 minutes to allow the layers separated. The non-polar layer was got dry passing through sodium sulfate that was condensed and pass through a cleanup column. At $500C^{o}$ activated, 10 g aliumina was poured in a column of 2cm diameter, containing 2 g anhydrous $Na_2SO_4$ at the apex. It was rinsed with 25ml n-hexane than the 5ml extract was poured on to the column that was followed by n-hexane 100ml. The obtained extractive was brought to minimum volume on a rotary evaporator and brought that up to a 5 ml for GC-ECD analysis.

**Results and Discussions**

GC-ECD column material was SE 30, 10% and 5% QF-1 on DC 200 and on chromosorb w, column temperature was set on $150^0$ and ECD temperature was set $175^0$ centigrade on sensitivity of $3x\ 10^{-10}$ while 5.5ml/min nitrogen was used as a carrier gas. On these parameters around 90% recoveries of DDT and its metabolites were achieved. While the various amount of DDT and DDE detected in the treated samples and the fortified ones are presented in table 1. 90% DDT and DDE recoveries were achieved in fortified samples. In Table 1 it is indicated that the several samples, collected during and a long period after spraying of the crop with DDT, contained maximum 0.25 mg/kg DDT and its metabolite as DDE and minimum 0.15 and 0.19 mg/kg DDT and its metabolite, respectively. It is suspected that to combat with the pest problem, small farmers may use deregistered insecticides like DDT, despite the deregistered of nearly all the persistent Organochlorine insecticides and legislations against use of such compounds. As Khan (1998) has indicated evidence that cotton bollworm may even develop insecticide resistance up to 2000 fold, in this scenario it is conceivable that somehow, though at a little scale, DDT is still in use. Therefore, Iram et al (2009), Ahad et al. (2005) and Anwar et. al (2012) have reported it from Lake environment and soil respectively, Uddin (2011) and Khan et al.(1996) have reported it on food produce in Pakistan, which can be followed and used to trace the decomposition of metabolic products from the labeled chemicals, since the proteins are known to have affinity with particular insecticides therefore, specific protein based techniques are also useful in certain cases. HPLC and GC have been used successfully for the purpose (Tabssum et al 2008). However, Khan et al (1996) suggested that only small farmers are using deregistered products on locally consumable produce.

**Table 1. DDT and DDE residues found in fortified and treated cotton samples**

| #Samples | Insecticides | | | |
|---|---|---|---|---|
| | DDE | | DDT | |
| | Sprayed Sample | Untreated Fortified* | Sprayed sample | Untreated Fortified* |
| 1st | 0.2 | 0.8 | 0.15 | 0.78 |
| 2nd | 0.25 | 1.0 | 0.17 | 0.78 |
| 3rd | 0.20 | 1.0 | 0.2 | 1.0 |
| 4th | 0.19 | 1.0 | 0.18 | 0.9 |
| 5th | 0.25 | 1.1 | 0.25 | 1.0 |
| 6th | 0.19 | 1.0 | 0.18 | 0.9 |
| 7th | 0.19 | 1.0 | 0.2 | 0.9 |

*Fortified cotton samples showed 90% recoveries

**References**

Ahad, K., Mohammad, A.F., Mehboob, A.S. and Ahmad, I. (2005). Pesticide Residues in Rawal Lake, Islamabad, Pakistan. *Bul.Environ. Contam. Toxicol*. 76: 463-470.

Akhtar, S. and Hassan, N. (2002). Efficiency of mini column for detection of multiple pesticide residues in vegitables. *Philippine J. Sci*. 131: 149-153.

Anwar, T., Ahmad, I. and Tahir, S. (2012). Determination of pesticide residues in soil of Nowshehra, Distt. Sindh, Pakistan. *Pakistan J. Zool*. 44: 87-93.

Baig, M.M.H., Khan, M.S., Khan, N., Ferhat, S., Hossain, A., Osmani, M.J.A. and Lord, K.A. (1972). Gaschromatographic and radiometric study of the behavior of $C^{14}$-DDT on mustard plants under tropical conditions. *Pakistan J. Sci. Ind. Res*. 15(3): 220- 226.

Iram, S., Ahmad, I., Ahad, K., Muhammad, A. and Anjum, S. (2009). Analysis of Pesticides Residues of Rawal and Simly Lakes. *Pakistan J. Bot*. 41(4): 1981-1987.

Khan, M.F., Naqvi, S.N.H., Azmi, M.A., Khan, M.I. and Qadri, S.S. (1996). DDT residues on apple after spray for codling moth control. *Pakistan J. Entomol*. 11:21-22.

Khan, M.F. (1998). Determination of resistance in cotton bollworm *Earias fabia* stol against local neem product in comparison with monocrotophos and cypermethrin. PhD thesis submitted to the University of Karachi.

Uddin, R., Khan, M.F., Iqbal, S. and Abbas, M. (2011). Determination of multi- residues insecticides of Organochlorine,Organopohosphorous and pyrethroids in wheat. *Bull. Environ. Contam. Tixicol*. 87(3): 303-6.

Tabassum, R., Naqvi, S.N,H. and Khan, M.F. (2008). Quantitative estimation of residues of neem compounds (NfC &NC) and Dimilin (IGR)in treated *Callosobrucus analis* by HPLC. *Turkish J. Zool*. 31: 131-136.