ATR –FTIR Spectroscopy in Chlorpyrifos Residue Investigation

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Outline

General information about chlorpyrifos (CPS) and chlorpyrifos-oxon (CPO)
Results and discussion
Spectroscopic data and 2 D chromatography
Conclusions

Aim

-to optimize spectroscopic technique FTIR, ATR-FTIR (Attenuated Total Reflectance) with ATR sample compartment for CPS determination

-and the most toxic degradation products CPO (residues) in pesticides formulations and samples
-to analyze the chemical changes of the samples before and after pesticides treatment
-to analyze CPS degradation process
-to investigate matrix effect (after identification of CPS and CPO surrounding-components)

Introduction

The apples are treated by EC– emulsifiable concetrate and emulsion in water EW based on chlorpyrifos

The ATR- FIR for qualitative and qualitative analysis of CPO and CPS residues

The pesticides based on chlorpyrifos

-Chlorpyrifos (O,O-Diethyl-O,3,5,6-trichloro-2-pyridylphosphorothioate, CPS, (Figure 1) is an organophosphorous insecticide used in agriculture .

-It is very persistent (resistant to degradation) and non-volatile compound.

-Chlorpyrifos (CPS) - low degradability by natural processes.

The degradation process of CPS leads to the formation of chlorpyrifos-oxon (O,O-diethyl-O-3,5,6trichloro-2-pyridylphosphate, CPO) and 3,5,6-trichloro-2-pyridinol (TCP).

-CPS residues may be detectable in fruit, vegetables, water, soil and body fluids, from months to years after its application.
-This occurs especially after unprofessional application of agricultural formulations

-CPS is very toxic to humans, causing tens of thousands of deaths per year worldwide because of many reasons: type of pesticide application, short waiting period between treatments, doses over allowed)

Toxicity mechanism: The formation of the most toxic metabolite CPO (Figure 1) occurs in liver, where P=S bond of CPS is replaced by a P-O bond.

CPF degradation till CPO

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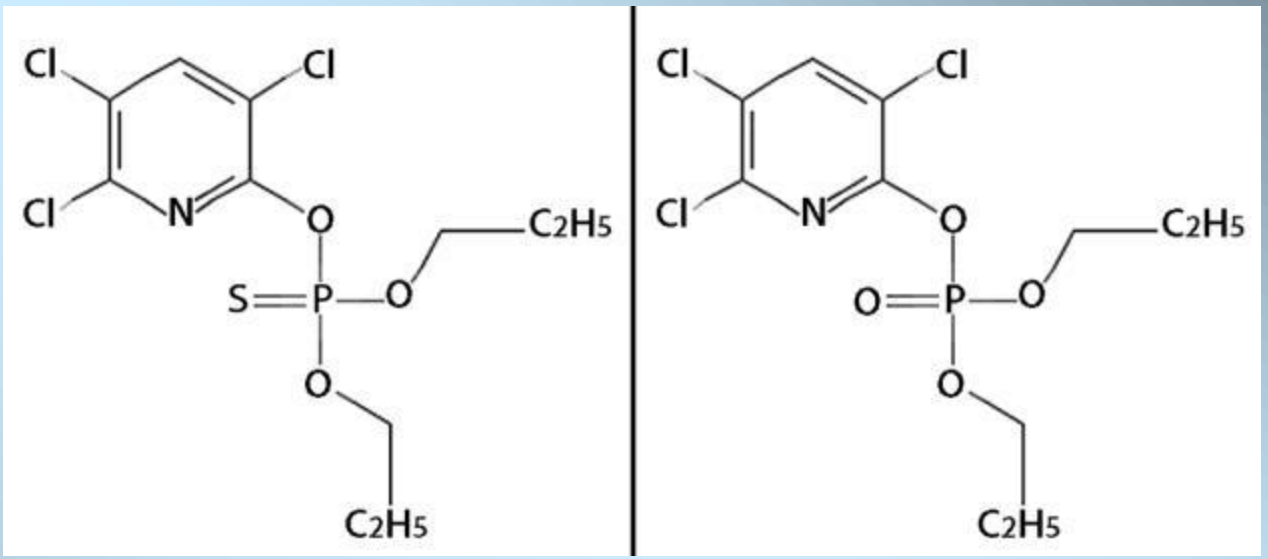

Figure 1. *O,O-Diethyl-O,3,5,6-trichloro-2-pyridylphosphorothioate (CPS, left) and O,O-diethyl-O-3,5,6-trichloro-2-pyridylphosphate (CPO, right).*

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1.Spectroscopic technique:

The Fourier transform infrared spectroscopy (FT-IR) and FT-IR with Attenuated total reflection comparment (for determination of chlorpyrifos and chlorpyrifos-oxon in apples and substances that give contribution to matrix effect)

FTIR (ATR –FTIR)

- High resolution
- Successfully used to study pesticide behaviour
- Follow up reactivies products
- Confirmation of pathway in reaction mechanisms
- for chemical changes of the samples before and after pesticides treatment.

2. Chromatographic (GC MS and 2D Chromatography)

2 D chromatography has been used for matrix effect studies and gas chromatography with mass spectrometry (GC-MS) based on component retention time and target ion

ATR –FTIR

The Fourier transform infrared spectra (FT-IR) were recorded with a resolution of 2 cm-1, 32 scans on a Thermo Nicolet 6700 FT- IR spectrometer .

Attenuated total reflection Fourier transform infrared techniques, sample „introduction" equipment (ATR-FTIR) used to analyzed the chemical changes of the samples-apples before and after treatment.

FT- IR spectra of CPS and CPO analytical standards have been recorded firstly.

The P=O stretch vibration, ν=537.12cm-1 originated from chlorpyrifos-oxon and the P=S vibration, v=634.59cm-1 originated from CPS, both in fingerprint area in IR spectrum.

The intensities of these vibration's bands are monitored in EC and EW formulations.

FTIR Spectroscopy in investigation CPS degradation

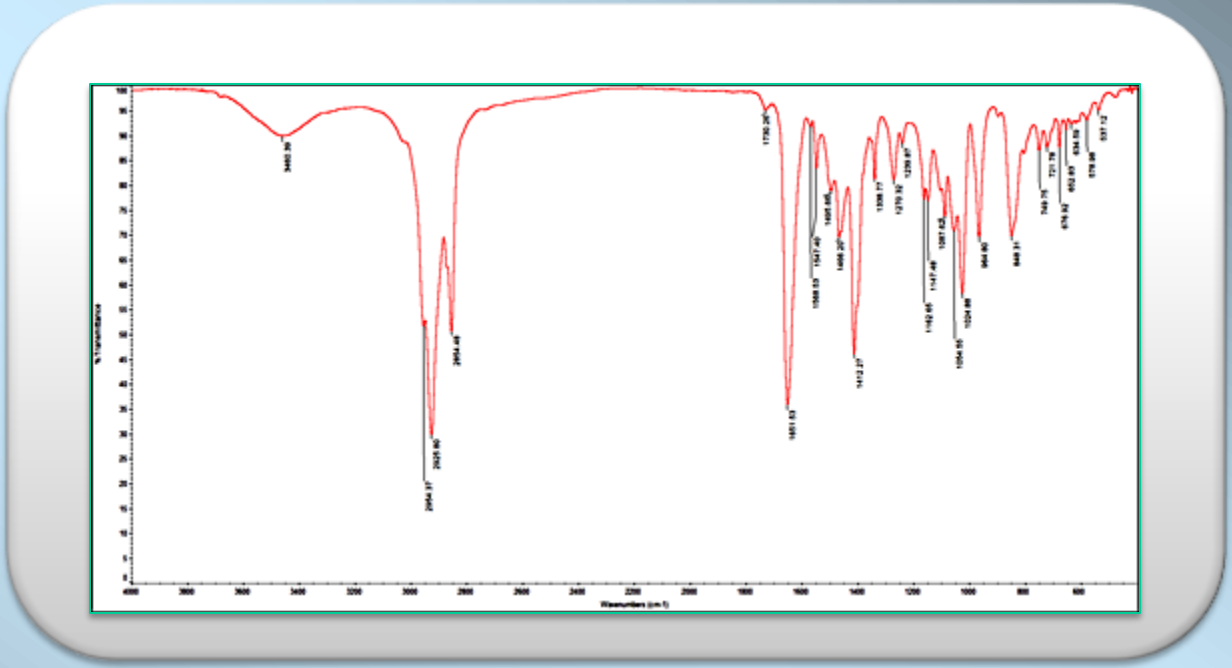

Figure 2: FTIR spectrum of CPS

The peak at 634.59 cm-1 is assigned to P=S stretching, characteristic for chlorpyrifos, the peak at 537.12 cm-1 is assigned to P-O stretch, and is ascribed to chlorpyrifos-oxon.
The Peak at 1466 cm-1 belong to –CH2 and –CH3 groups.

FTIR Spectroscopy in investigation CPS degradation

During degradation of CPS in FTIR spectrum fig 2. peaks of C-Cl with 600-800 cm-1 and CH with 2954 cm-1 were obviously dropped.

It means that cleavage of C-Cl and C-H bonds occurred, and chlorine was further converted into chloride.

The peaks at 1651.30, 1588.96 and 1495.06 cm-1 were contributed by benzene vibrations.

During stimulated degradation process aromatic ring of cpf was destroyed also.

ATR-FTIR Spectroscopy – apple skin

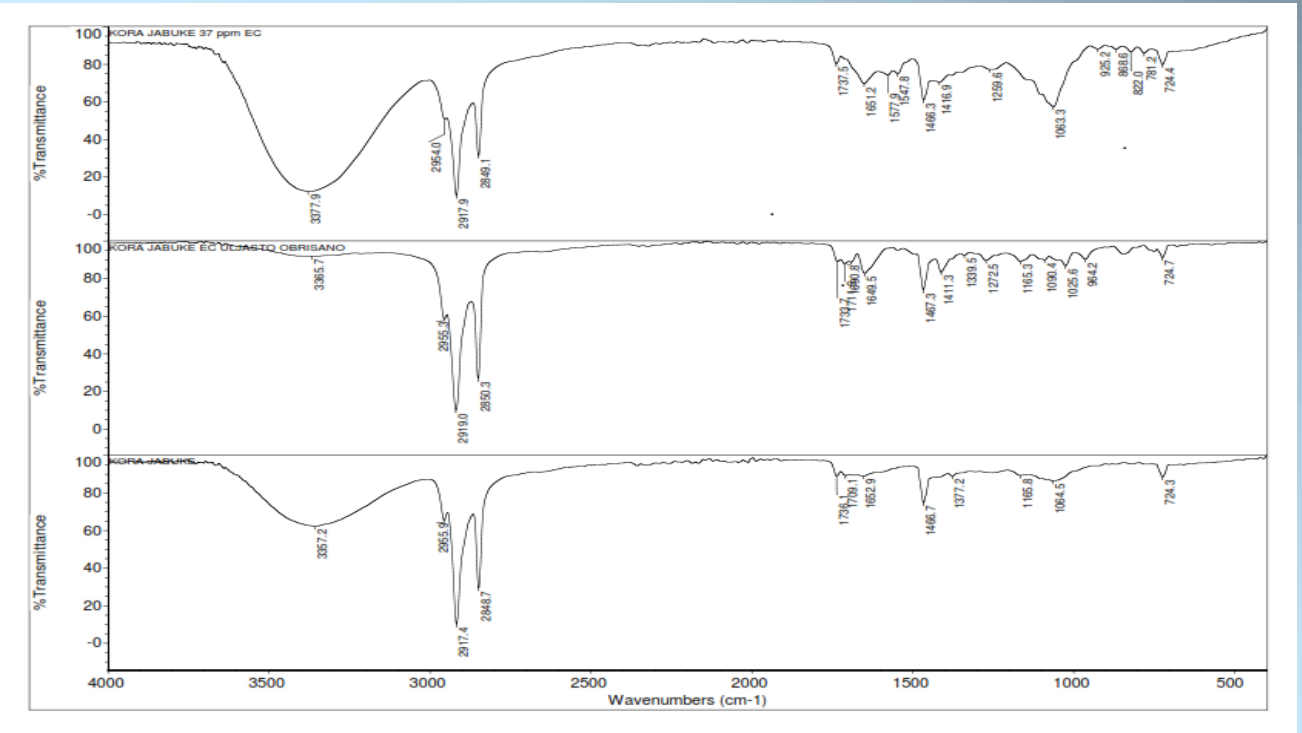

*Figure 3: **ATR –FTIR spectra of apple samples:** a) dried skin apple, b) dried skin apple treated by EC and c) dried skin apple treated by 37 ppm solution of EC formulations*

ATR -FTIR Spectroscopy

1.ATR-FTIR in real sample investigation has advantages in comparation to other techniques :

-No sample preparation
-No changes in samples during analysis
-No toxic solvents for extractions

2.Treated apples peaks differ in intensities–bases for quantitative analysis

.

interpretation - Figure 3

ATR-FTIR spectra, consists of four wavenumber interval:
-(3100-3400)cm-1 typical for water,
-(2700-3600)cm-1 typical for compound with carbonyl group,
-(2100-2700)cm-1, area without significant compound and
-(450-1700)cm-1 characteristic for esters and
In ATR-Fourier transform infra-red spectra was assigned P=O stretch vibration, ν=537.12 cm-1 originated from chlorpyrifos-oxon, in area of fingerprint in spectrum.

Matrix effect

-FTIR Spectrometry used for qualitative analysis of CPS based formulation

Result for EW formulations by ATR-FTIR :

Active supstance for both is CPS with characteristic wavenumbers : 749.75, 848.31, 1466,20, 1588,96 1651,53 cm-1

From FTIR spectra of EW formulation:
characteristic wavenumbers that indicated presence of **thickener** with characteristic wavenumers 3443.50, 1455.41, 1163.20 cm-1

And 2872. 47 , 1240.54; 1087.48, 872,00 cm-1 that belong to **antifreeze**.
The confirmation has been done by 2 D chromatography and confirmed bgc ms chromatography

2D chromatograhy in matrix investigation

The different surroundings of active component is checked by GCxGC MS spectra of these two formulations

This most abundant species chosen in matrix are: esters, methyl-benzene and oxadecans that can obstruct determination of analyte. (EC- consist of CPF in high-boiling mineral oils, solvent aliphatic hydrocarbonate HCs and emulsifiers).

The EW formulation except CPS consist of emulsifier, antifreeze , antifoam, thickener, biocide, stabilizer, buffer.

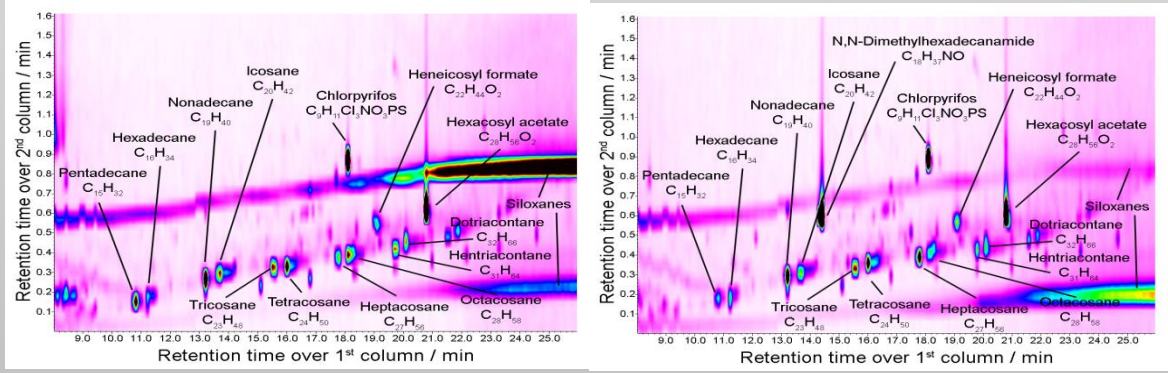

Fig 4 : GCXGC of 3 ppm EW formulation left and 3 ppm EC formulation right.

It has been used 2D termal modulation in GC x GC analysis.

Two used software is Chrom Square ver 2.1.

On the basis of GCxGC-MS, differences were observed in the existence of the components in the EC formulation that are not detected in relation to the EW formulation. In accordance to intensities the highest difference is for N,N–dimethyl hexadecanamide.

For identification it has been used GC-MS based on retention time and 3 confirmatory ions.

Conclusion :

- ATR –FTIR and 2D chromatography are good combination for degradation process study of pesticides as well as investigation of matrix effect in the determination of analytes.

- Possible mechanism of CPF degradation involves breakage of P=S bond in chlorpyrifos

- These two techniques can be used to develope simple and environmental friendly methods for the determination of CPS in different samples, with no special preparation
- In further investigation data from ATR FTIR spectra will be used for mathematical calculation by genic algorithms and artificial neural network (for active supstaces in complex organic compound such as pesticides)
- ATR -FTIR techniques provides excellent quality data in conjunction with best possible reproducibility
- To our knowledge there is no characterization of EC and EW formulation by 2 D chromatography

Thank you for your attention!