Solid phase microextraction,
GC-MS, weeds, CAR/PDMS

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APPLICATION OF SPME/GC-MS FOR DETERMINATION OF CHLOROPHENOXY HERBICIDE RESIDUES WITHIN WEED TISSUES

Application of SPME/GC-MS for determination of accumulation of the MCPA (4-chloro-2-methylphenoxy)acetic acid in aerial parts of three groups of cereal weeds: highly sensitive (scentless mayweed, shepherd's purse), sensitive species (field horsetail, garden cornflower) and resistant ones (corn cockle, wireweed) has been studied. Dichloromethane extract was performed and identification and quantification of the MCPA was done using a gas chromatography-mass spectrometry (GC-MS) system with a selective ion monitoring (SIM). The chlorophenoxy herbicide showed different level of accumulation within tissues of the studied weeds. The highest concentration of MCPA was found within tissues of the sensitive weed species and the lowest within tissues of the resistant ones.

INTRODUCTION

Chlorophenoxy acetic acid derivatives were the first herbicides to be registered in the United States and have been widely used in control of broadleaf and woody plants on rangelands, lawns, golf courses, forests, roadways, parks and agricultural land [1]. Comercial preparations containing (4-chloro-2-methylphenoxy)acetic acid (MCPA) and 2,4-dichlorophenoxyacetic acid (2,4-D) salts, amines or esters are still widely used as herbicides for cereal crops in Europe and North America [2-3]. 2,4-D's structure is similar to that of the plant hormone indole acetic acid and acts as a plant growth regulator that can interfere with hormones and affect of plant growth [4]. The MCPA (CAS No. 94-74-6) is an izosteric compounds of 2,4-D. MCPA is usually formulated as either an aqueous salt (e.g. dimethylamine (DMA) or sodium salts) or as an ester (e.g. 2-ethylhexyl, 2-EHE). Formulations of MCPA are registered world-wide for use on agricultural crops including cereals and are adsorbed through both roots and leaves of the most plants, especially broadleaf species [5]. Aminopielik (active agent – DMA-2,4-D) and Chwastox (active agent – DMA-MCPA) are the most popular herbicides used in cereal protection in Siedlce Administrative District (Fig. 1).

LLE (*liquid liquid extraction*) and SPE (*solid phase extraction*) have been traditionally used for herbicide residue determination in environmental samples. However, LLE methods require large volume of solvent and long extraction. On the other hand, the SPE requires less solvent volume than the LLE while offering a limited reduction in sample preparation time [6]. Solid phase microextraction (SPME) is a new, fast and simple analytical technique which uses coated fused-silica to extract

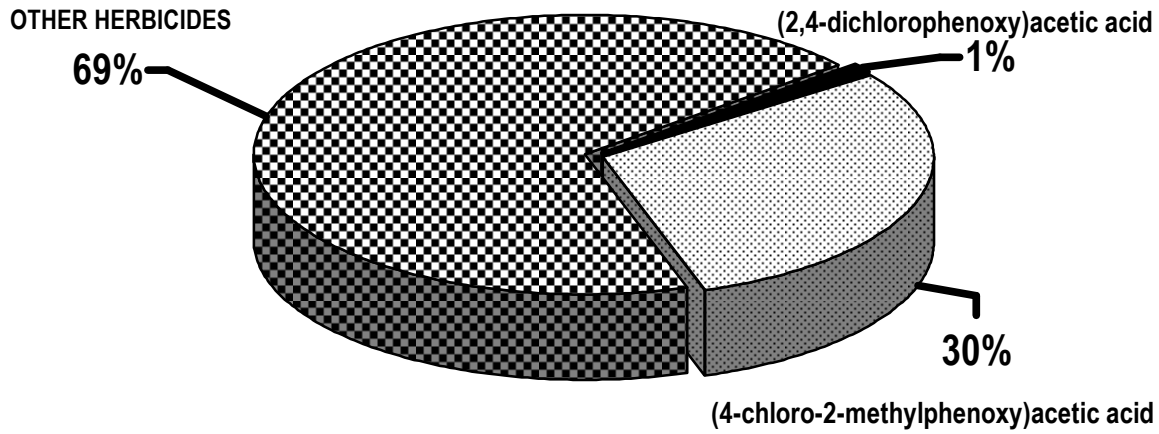

Fig.1. Percentage of the chlorophenoxy herbicides used in the Siedlce Administrative District.

herbicides from aqueous samples. The SPME requires a small volume of sample and may be followed by gas chromatography combined with mass spectrometry [7-9]. Initially polydimethylsiloxane (PDMS) and polyacrylate (PA), have been suggested for the herbicides analyses and now it is extended to other coatings such as Carboxen-PDMS [10].

This paper report on possibility of application of the solid phase microextraction (SPME) combined with GC-MS for monitoring of the (4-chloro-2-methylphenoxy) acetic acid residues within tissues of three groups of weeds growing on agricultural field of winter wheat.

MATERIALS AND METHODS

The field experiments were performed on commercial field of winter wheat (Roma cv.) located near Siedlce. The following three groups of weeds were used in the carried out experiments: high sensitive (*Matricaria inodora*, *Capsella bursa pastoris*), sensitive (*Equisetum arvense*, *Centaurea cyanus*) and resistant species (*Agrostemma githago*, *Polygonum aviculare*).

The experimental field was sprayed out with 300 g/l MCPA in May (active agent a commercial preparation Chwastox Extra 300 SL containing 30% of MCPA as sodium salt and 70% of nonspecified ingredient was purchased from „Organika Sarzyna" - Nowa Sarzyna, Poland). The spraying of the cereal field was carried out at growth stage 43 of winter wheat according to the BBCH codes [11]. Then 100 g of aerial parts of each weed were collected after 1, 4, 8, 12, 24 hrs and 2, 3, 4, 5, 12, 19, 33, 61, 90 days.

30 g of weed's material was shaken with mixture of acetone – NaCl - water (3:1:6 v/v) for 15 min, and then filtered through a Whatman No 1 filter paper.

Dichloromethane extraction of the plant filtrates was performed and the extracts were evaporated to dryness at 35 ^{o}C in a rotary evaporator. The residue was redissolved in 1 ml of methanol, and then the solid phase microextraction was performed.

75 μm Carboxen/Polydimethylsiloxane (CAR/PDMS) fiber was conditioned before initial application in the injection port of the gas chromatograph by heating at 280 ^{o}C for 1h. Then the fiber was exposed to the stirred sample with addition of 0.5% of NaCl for adsorption. When the adsorption was completed, the fiber was removed from the better sample vial and introduced into the GC injector where the thermal desorption of the analyte was carried out [12].

GC-MS separation was performed using, QP-5050 Shimadzu instrument equipped with capillary column BPX-5 (30 x 0.25 mm x 0.25 μm), under the following chromatographic conditions: injector temperature 220 ^{o}C, column temperature programmed as follows: 80 ^{o}C (5 min), 80-280 ^{o}C (20 ^{o}C/min), 280 ^{o}C (5 min). Helium was used as the carrier gas at 9.8 ml/min, 56.7 kPA. MS measurements were performed with electron impact (EI) ionization at 70 eV, and SIM three characteristic ions were selected for each compound and scanned using corresponding time windows between 45-450 mz per ion.

RESULTS

Obtained results showed the highest concentration of the studied herbicide within tissues of the sensitive weeds, *M. indora* and *C. bursa pastoris*, and it was reaching 0.37 μg/l and 0.36 μg/l, respectively. Eight hours after application, plants of the sensitive weed species died, and there was no clear differences between plants of the scentless mayweed and shepherds purse (Fig. 2).

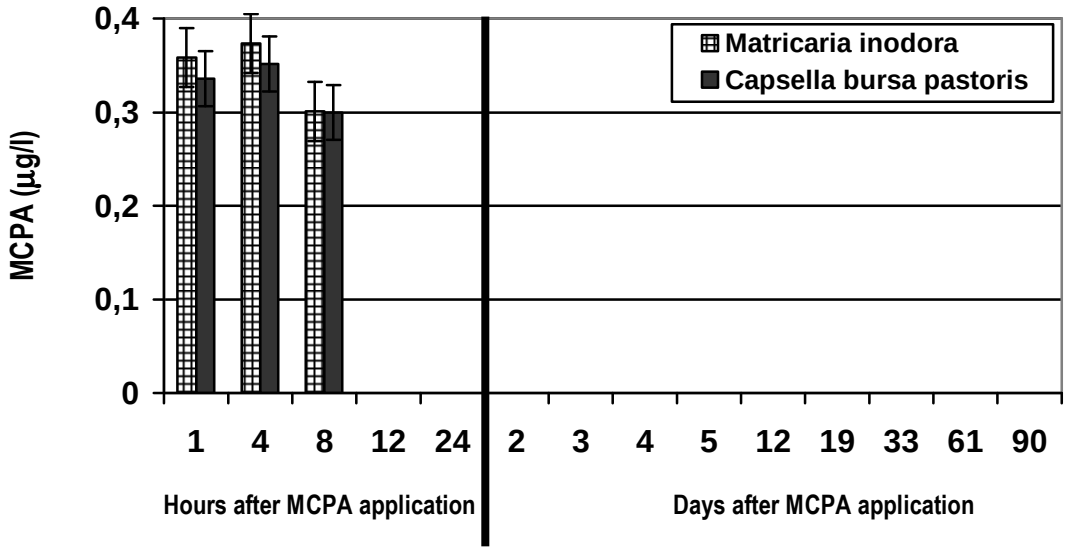

Fig. 2. Presence of the (4-chloro-2-methylphenoxy)acetic acid within sensitive weeds.

The lower concentration of the herbicide was found within the less sensitive weeds, such as field horsentail and garden cornflower. Higher level of MCPA occurred within tissues of the *E. arvense* than the *C. cyanus*. Moreover, the plants of *E. arvense* highly accumulating (4-chlor-2-methylphenoxy)acetic acid died after 19 days, instead plants of *C. cyanus* survied even 90 days after MCPA application (Fig. 3).

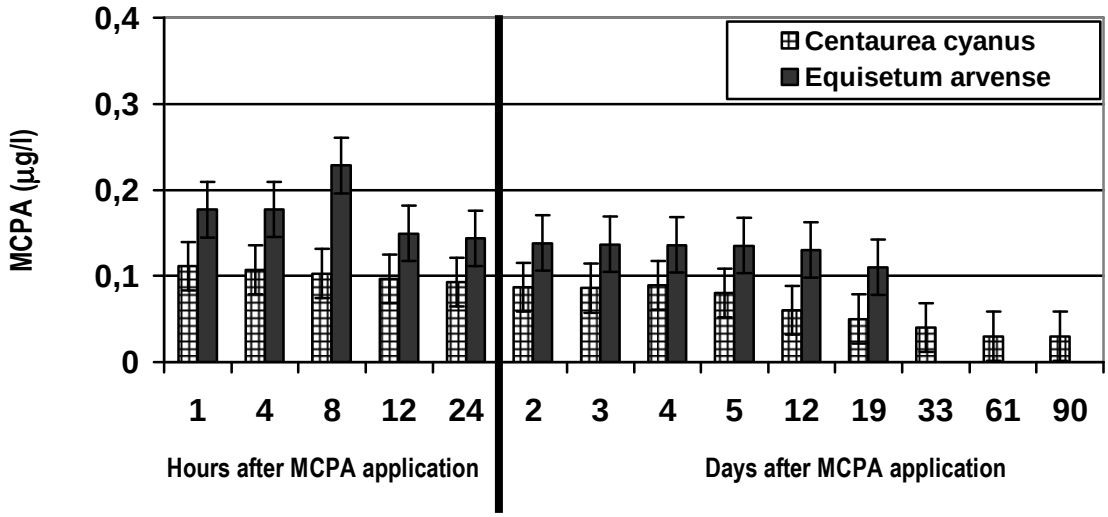

Fig. 3. Presence of the (4-chloro-2-methylphenoxy)acetic acid within less sensitive weeds.

When the resistant weeds (*A. githago* and *P. aviculare*) were studied they showed much lover accumulation of the chlorophenoxy herbicide. Higher concentration of MCPA existed within aerial parts of the *P. aviculare* than then within tissues of the *A. githago*. However, plants of the both species survived the herbicide application until 90 days of observation (Fig. 4).

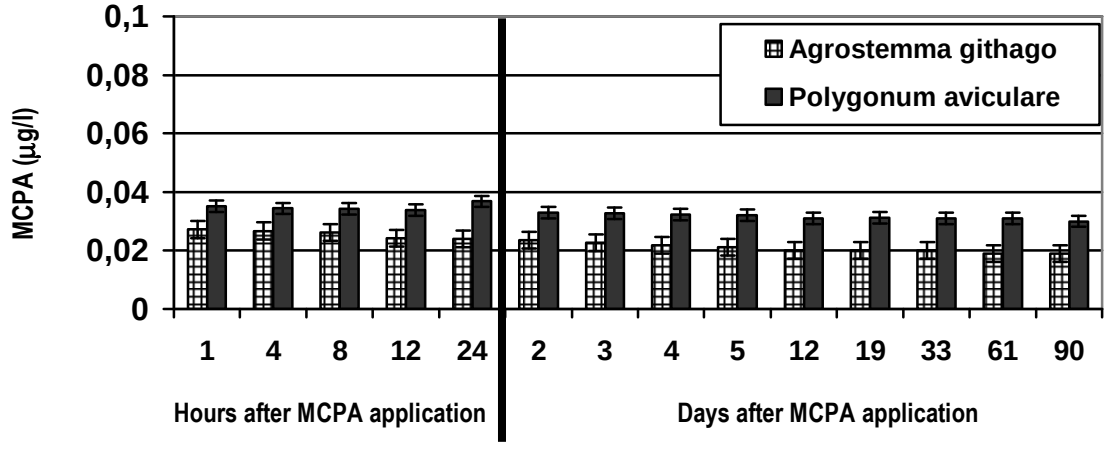

Fig. 4. Presence of the (4-chloro-2-methylphenoxy)acetic acid within resistant weeds.

CONCLUSIONS

SPME/GC-MS analysis proved to be a fast and sensitive method for routinely and precisely determination of (4-chloro-2-methylphenoxy)acetic acid residues within tissues of cereal weeds.

ACKNOWLEDGEMENTS

This work was supported by the Polish Scientific Committee in the framework of a grant no 2P06S 058 30

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