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Analytical Methods

A rapid liquid chromatography method for determination of glufosinate residue in maize after derivatisation

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ABSTRACT

A rapid liquid chromatographic method for glufosinate analysis in maize samples after derivatisation has been developed. The labelled glufosinate was separated on a Kromasil C_{18} column (250 mm × 4.6 mm, 5 μm) and UV detection was applied at 360 nm. The optimisation of derivatisation conditions and the influence of different ion-pair reagents on the separation were discussed. The method linearity correlation coefficient was 0.9998 in concentrations ranging from 0.1 to 20 mg L^{-1}. The level of quantification was set to 0.02 mg kg^{-1}, and reached pesticide EU-MRLs for glufosinate in the maize samples. The proposed method was applied to the quantitative determination of glufosinate in samples with recoveries of 98.0–100.5% and RSDs of 2.13–4.13%.

1. Introduction

Food safety has become one of the most important issues that concerns people (König et al., 2004). As the pesticides were intensively and extensively used in agriculture, pesticide residues in food have posed a great threat to the consumers (Amvrazi & Albanis, 2009; Ozbey & Uygun, 2007).

Glufosinate [DL-homoalanine-4-yl(methyl) phosphonic acid] is a broad-spectrum, non-selective and post-emergent herbicide used for the control of a wide range of weeds (Royer, Beguin, Sochor, & Communal, 2000; Zimdahl, 1993). It has been used to control weeds in the farmland such as maize, wheat, soybean and so on (MAFF, 1990). Phosphinothricin is the active ingredient of glufosinate, and leads to plant death by inhibiting the action of the enzyme glutamine synthetase. The use of glufosinate has increased dramatically since the marketing of genetically engineered crops that are resistant to this herbicide (Chompoo & Pornprom, 2008; Kumar, Bellinder, Brainard, Malik, & Gupta, 2008). As a systemic herbicide, it can be absorbed by roots and leaves and transmitted to the plant tissues, and thus indicate a possible food contamination (Tomlin, 1999), which can create a potential threat to the health of people. So far, the number of publication about residue determination of glufosinate in maize samples is limited. A simple and reliable method is therefore a must for the determination of glufosinate in maize samples.

Analytical methods for the analysis of glufosinate include capillary electrophoresis (CE) (Chang, Tseng, Mallipattu, & Chang, 2005; Goodwin, Startin, Keely, & Goodall, 2003; Orejuela & Silva, 2005; Zhou et al., 2007), enzyme-linked immunosorbent assay (ELISA) (González-Martínez et al., 2005; Ibáñez, Pozo, Sancho, López, & Hernández, 2005), gas chromatography (GC) (Kataoka, Ryu, Sakiyama, & Makita, 1996; Motojyuku et al., 2008; Royer et al., 2000; Tseng, Lo, Chang, Chou, & Chang, 2004) and liquid chromatography (LC) (Hanke, Singer, & Hollender, 2008; Hori, Fujisawa, Shimada, Sato, Kikuchi, et al., 2002; Hori, Fujisawa, Shimada, Sato, Honda, et al., 2002; Kuster, Alda, & Barceló, 2009; Sadi, Vonderheide, & Caruso, 2004; Sancho, Hernandez, López, Hogendoorn, Dijkman, & van Zoonen, 1996; Wang, Chen, Hsu, Cheng, & Lee, 2008). LC is preferred over GC because of the ionic character of glufosinate. However, for LC with conventional detection systems, such as UV–vis or fluorescence detectors, glufosinate needs to be derivatised because of the lack of chromophore or fluorophore. For these reasons, chemical derivatisation or labelling becomes a necessary procedure to transform the analytes into derivatives that can be more easily isolated, separated, and detected. Thus for sensitive determination of glufosinate, derivatisation agents reported mainly include 9-fluorenylmethyl chloroformate (FMOC-Cl) (Sancho et al., 1996; Hori, Fujisawa, Shimada, Sato, Honda, et al., 2002) and *p*-nitrobenzoyl (Hori, Fujisawa, Shimada, Sato, Kikuchi, et al., 2002).

4-Chloro-3,5-dinitrobenzotrifluoride (CNBF) is an important fine chemical, which has been known to react with primary or secondary amines in the presence of base to produce stable *N*-substituted-2,6-dinitro-4-(trifluoromethyl)-benzamine deriva-

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tives, which display satisfactory ultraviolet absorption (Pitzer, Werbovetz, Brendle, & Scovill, 1998; Qian et al., 2009). For the determination of water-soluble herbicides, ion-pair liquid chromatography–tandem mass spectrometry and ion-pair liquid chromatography coupled to inductively coupled plasma mass spectrometry were ever used (Chen, He, Beer, Megharaj, & Naidu, 2009; Wang et al., 2008). Glufosinate is a water-soluble herbicide with strong polarity, which increases the difficulty in the determination. In our present work, ion-pair reversed-phase liquid chromatographic method was used for the determination of glufosinate after derivatisation with CNBF in maize samples. In order to meet the determination standards, a C_{18} solid phase extraction (SPE) column was used to purify the sample in a single step using solid sorbents without resorting to complicated or expensive instrumentation. At the same time, the optimisation of derivatisation conditions and the effect of different ion-exchange conditions on separation efficiency were also studied.

2. Materials and methods

2.1. Instrumentation and conditions

A high performance liquid chromatography system, which consisted of two LC-10ATvp pumps and a SPD-10Avp, ultraviolet detector (Shimadzu, Japan) was used for the analysis and separation. A reversed-phase Kromasil ODS C_{18} column (250 mm × 4.6 mm, 5 μm) was used for separation at room temperature and a Chromato Solution Light Chemstation for LC system was employed to acquire and process chromatographic data. AccuBOND ODS-C_{18} (200 mg/3 mL, 40 μm) was purchased from Chubo Apparatus & Equipment Shanghai Co. Ltd. (Shanghai, China).

2.2. Chemicals and reagents

Glufosinate (99%) was friendly supplied by Bayer CropScience China Co. Ltd. (Beijing, China). A standard solution of 0.01 mol L^{-1} glufosinate was prepared in H_3BO_3–$Na_2B_4O_7$ buffer and further diluted to the required concentration when used. Working standard was prepared by mixing aliquot of the stock solution and water. The stock and working standard were stored in dark at 4 °C. Acetonitrile and methanol were HPLC grade and purchased from J.T. Baker (USA). Ultrapure water was obtained in the laboratory using a Milli-Q water purification system (Millipore, Billerica, MA). CNBF was obtained from Alfa Aesar (Ward Hill, MA, USA), and its solution was prepared in methanol and filtered through a 0.45 μm nylon membrane filter and refrigerated when not in use. Cetyltrimethylammonium bromide (CTAB), dodecyl trimethyl ammonium bromide (DTAB), methyl trioctyl ammonium bromide (MTAB) and tetrabutyl ammonium bromide (TBAB) were purchased from Sinopharm Chemical Reagent Beijing Co. Ltd. (Beijing, China). All other chemicals and solvents were of analytical grade and from commercial sources. H_3BO_3–$Na_2B_4O_7$ buffer was prepared by mixing 0.2 M H_3BO_3 solution with 0.05 M $Na_2B_4O_7$ solution to the required pH value.

2.3. Sample preparation

First of all, 2 mL of standard glufosinate solution of different concentrations was added into 2 g of maize sample, respectively. Then, the sample was set in a polypropylene centrifuge tube, milled, capped, and stood overnight at room temperature. The sample was extracted by shaking for 1 h with adding borate buffer to 20 mL and then centrifuged at 4000 rpm for 10 min. All supernatant passed through the SPE cartridge which had been rinsed with acetonitrile (3 mL) and water (3 mL). Finally, the filtrate was evaporated to dryness, and sample was fixed volume to 1 mL with borate buffer.

Two hundred microlitres of sample or glufosinate standard solution was transferred into a 1 mL vial, and then 300 μL of borate buffer (pH 9.0) and 100 μL of CNBF methanol solution were added. After the whole solution was diluted to 1.0 mL with borate buffer, and then mixed on a vortex-mixer. The mixture was incubated at 60 °C for 30 min in water-bath, shaking at 10 and 20 min and then 10 μL HCl (2 M) was added to quench the reaction. The resulting solution was filtered through 0.45 μm nylon filter and injected in the chromatographic system. Each sample was assayed in triplicate and all the assays were carried out at room temperature.

2.4. Chromatographic method

Before the analysis, the C_{18} column equipped with a guard column (4 mm × 3 mm) was pre-equilibrated with the mobile phase for 30 min. The HPLC separation of glufosinate derivative was carried out on Kromasil ODS C_{18} column. Acetonitrile (eluent A), 10 mM DTAB solution (pH 4.0 with 20 mM phosphoric acid) (eluent B) were used as mobile phase. All the solvents were filtered with a 0.45 μm membrane filter. The programme was set for a linear gradient starting from 45% to 100% of the solvent A at 10 min. The injection volume was 20 μL, and detection wavelength was 360 nm. The flow rate was constant at 0.8 mL min^{-1} and the column was at room temperature.

3. Results and discussion

3.1. Optimisation of derivatisation conditions

The reaction of CNBF with amino group on glufosinate molecule is represented in Fig. 1. CNBF is known to have good activity and selectivity for amino compounds and can be employed as an excellent active group. It can react with amines in low concentration to form stable derivatives under base conditions, and the excess reagent is hydrolysed to the corresponding phenol without any other by-products and interferences. The hydrolysis compound can be written as (CNBF)OH. Because CNBF has relatively poor solubility in water, organic solvent should be added to the derivatisation medium to avoid the precipitation of the reagent. Therefore, at least 100 μL of methanol should be added to the derivatisation medium. There is a competition between the labelling and the hydrolysis, so excess labelling reagents should be used.

The effect of glufosinate/CNBF ratio, derivatisation pH value, derivatisation reaction temperature, and derivatisation reaction time on the peak areas of CNBF-glufosinate derivative was investigated. An aliquot of glufosinate (0.5×10^{-3} M) was reacted with various concentrations of CNBF (1.0×10^{-3}, 1.5×10^{-3}, 2.0×10^{-3} and 3.0×10^{-3} M). The results showed that the peak area of derivative was highest and unchangeable when the concentration of reagent reached 2.0×10^{-3} M, and there was not a statistically significant difference between 2.0×10^{-3} and 3.0×10^{-3} M.

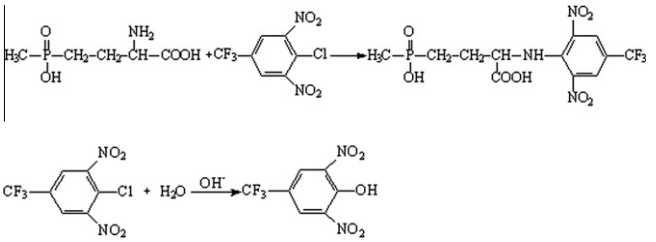

Fig. 1. The reaction scheme of CNBF with amino group on glufosinate molecule.

Therefore, 2.0×10^{-3} M was selected as the optimal concentration. The reaction of CNBF with glufosinate was also found to be pH dependent. The influence of various pH values on the peak areas was also studied. The optimum reaction pH was determined by derivatising glufosinate at pH values ranging from 7.5 to 11.0. The results showed that the peak areas of the derivative were almost stable at pH 8.5–10.0. This was probably due to deprotonation of glufosinate at the base condition, which could promote the nucleophilic addition, as observed in the case of aliphatic diamines (Zhang, Liu, Wang, & Cheng, 2004). Hence, an optimum derivatisation pH of 9.0 was selected for all subsequent experiments. Temperature was a very important factor in optimising the derivatisation rate. The values ranging from 40 to 70 °C were performed to find the best derivative temperature. It was found that peak areas of the derivative reached a plateau at 60 °C. The reaction time was a critical factor for the derivatisation reaction. The effect of reaction time on derivatisation was studied over the period from 10 to 40 min, while keeping all the other parameters constant. It was clear that peak areas reached an optimum value over a period of 30–40 min. To keep the total analysis time short, a reaction time of 30 min was chosen.

3.2. Optimisation of ion-exchange conditions

The mobile phase composition was optimised in order to achieve fast and optimum separation of glufosinate derivative, CNBF, (CNBF)OH. Chromatographic separation was carried out under gradient reversed-phase condition on Kromasil C_{18} column. Due to strong polarity of glufosinate derivative, it could not be separated completely with some polar impurities such as amino acids and small amino sugars. After ion-pair reagent was ionised in the mobile phase, counterion with hydrophobicity was absorbed on the stationary phase, and retention time of derivative increased by ion-exchange. The dynamic model view of ion exchange is shown in Fig. 2.

The effect of ion pair regent on retention time, recovery, resolution, and peak shape of derivative is shown in Table 1. As the carbochain grew, the retention time increased due to the affinity enhancement of ion-pair reagent (TBAB, MTAB, DTAB, CTAB) with stationary phase. Besides, it was found that ion-pair reagent from 5 to 10 mM could improve separation efficiency and higher concentration of ion-pair reagent caused the delay of retention time with peak broadening. The pH value of buffer in the mobile phase was also studied. Glufosinate is a kind of strong acid and can absolutely ionise to process ion-exchange. So, the retention time of derivative had no obvious change, as the pH value of mobile phase varied from 2.0 to 6.0. In this method, 10 mM DTAB and pH 4.0 was used in subsequent experiments for better and quicker separation. The chromatograms of glufosinate derivative obtained in gradient elution mode are shown in Fig. 3.

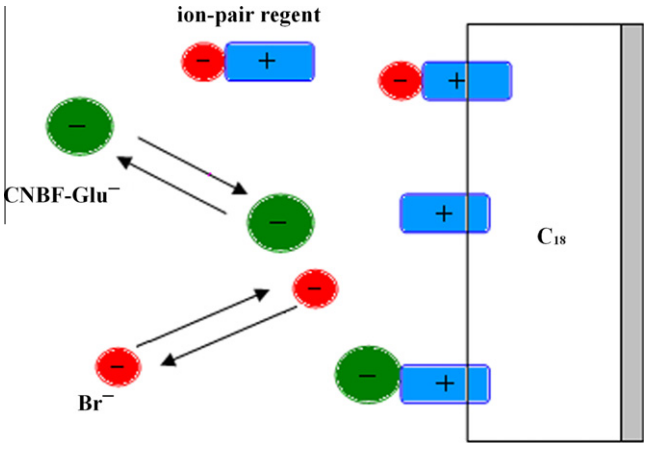

Fig. 2. The dynamic model view of ion exchange of glufosinate derivative with ion pair reagent.

Table 1
The effect of ion pair regent on retention time, recovery, resolution, and peak shape of derivative.

Ion-pair regents	Concentration (mM)	Retention time (min)	Recovery (%)[a]	Resolution of peaks	Peak shape
TBAB	2	3.7	–	0.86	Sharp
	5	4.2	73.72	1.45	Sharp
	10	4.5	87.35	1.78	Sharp
	20	4.6	88.92	1.82	Sharp
MTAB	2	4.0	–	1.03	Sharp
	5	4.5	82.23	1.57	Sharp
	10	4.8	90.45	1.72	Sharp
	20	5.0	90.36	1.83	Broaden
DTAB	2	5.8	–	1.12	Sharp
	5	8.3	90.77	1.75	Sharp
	10	8.7	99.65	>2	Sharp
	20	9.3	98.54	>2	Broaden
CTAB	2	6.6	–	1.23	Sharp
	5	9.4	93.23	1.80	Sharp
	10	10.1	99.56	>2	Sharp
	20	10.9	98.63	>2	Broaden

[a] Mean values of three determinations.

3.3. Validation of the method

The maize samples with different concentrations of glufosinate were prepared and analysed by using the optimised derivatisation procedure and separation conditions for the determination of glufosinate. The peak areas of the standards were recorded. The

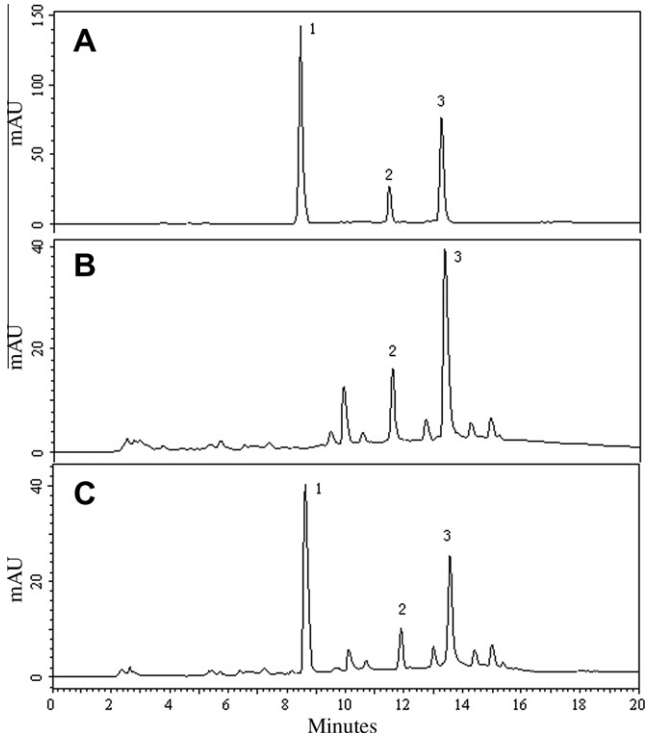

Fig. 3. Chromatograms obtained from samples (A) glufosinate standard solution; (B) blank maize sample; (C) maize spiked with 1.0 mg kg^{-1} of standard glufosinate. Chromatographic conditions: column, Kromasil C_{18} (250 mm × 4.6 mm, 5 μm); UV–DAD detection, λ = 360 nm; mobile phase, acetonitrile (eluent A), 10 mM DTAB solution (pH 4.0 with 20 mM phosphoric acid) (eluent B), a linear gradient starting from 45% to 100% of the solvent A at 10 min; flow rate, 0.8 mL/min; room temperature. Peaks: 1 glufosinate-CNBF, 2 CNBF, 3 (CNBF) OH.

Table 2
Linear calibration range, regression equation and detection limit of glufosinate.

Parameters	Glufosinate
Calibration range (mg L^{-1})	0.1–20.0
Regression equation, Y[a]	24736X[b] + 826.4
Coefficient regression, R^2	0.9998
RSD (%), n = 6, within-day	2.25
RSD (%), n = 6, between-day	3.95
LOQ (mg kg^{-1})[c]	0.02

[a] Y: peak area of glufosinate derivative.
[b] X: concentration of glufosinate (mg L^{-1}).
[c] Per 20 μL injection volume.

Table 3
Average recovery of glufosinate from maize samples by using proposed method.

Glufosinate added (mg kg^{-1})	Spring maize[a]		Summer maize[b]	
	Found (mg kg^{-1})	Recovery (%) (RSD)	Found (mg kg^{-1})	Recovery (%) (RSD)
0.5	0.49	98.0 ± 2.83	0.49	98.0 ± 2.89
1.0	0.98	98.0 ± 2.95	0.99	99.0 ± 2.16
2.0	1.99	99.5 ± 2.92	1.98	99.0 ± 2.63
5.0	4.9	98.0 ± 3.15	5.05	100.5 ± 2.13
10.0	9.9	99.0 ± 4.13	9.9	99.0 ± 3.73

[a] Nongda 108, collected from campus of China Agricultural University, Beijing, China.
[b] Zhengdan 958, collected from Zhengzhou in Henan province, China.

coefficient regression (R^2) is 0.9998. The linear calibration range, regression equation, and level of quantification are listed in Table 2. The applicability of the proposed method was evaluated in maize samples. The results obtained from the analysis of samples are shown in Table 3. The recoveries of glufosinate were from 98.0% to 100.5% and RSDs from 2.13% to 4.13%, depending on the sample investigated. The level of quantification was set to 0.02 mg kg^{-1}, and reached European Union glufosinate residue standard for maize samples (http://ec.europa.eu/sanco_pesticides/public/index.cfm).

4. Conclusions

A novel method for detection of glufosinate in maize samples was developed by ion-pair reversed-phase liquid chromatography. The optimisation of derivatisation conditions, mechanism of ion exchange and the effect of different ion-exchange conditions on separation efficiency were discussed. In this study, the optimisated conditions were used for quicker and better determination of glufosinate in the maize samples. The proposed method showed good repeatability, low quantification level (0.02 mg kg^{-1}) and excellent linearity for quantitative assay of glufosinate in maize samples.

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References

Amvrazi, E. G., & Albanis, T. A. (2009). Pesticide residue assessment in different types of olive oil and preliminary exposure assessment of Greek consumers to the pesticide residues detected. *Food Chemistry, 113*, 253–261.

Chang, S. Y., Tseng, W. L., Mallipattu, S., & Chang, H. T. (2005). Determination of small phosphorus-containing compounds by capillary electrophoresis. *Talanta, 66*, 411–421.

Chen, Z. l., He, W. X., Beer, M., Megharaj, M., & Naidu, R. (2009). Speciation of glyphosate, phosphate and aminomethylphosphonic acid in soil extracts by ion chromatography with inductively coupled plasma mass spectrometry with an octopole reaction system. *Talanta, 78*, 852–856.

Chompoo, J., & Pornprom, T. (2008). RT-PCR based detection of resistance conferred by an insensitive GS in glufosinate-resistant maize cell lines. *Pesticide Biochemistry and Physiology, 90*, 189–195.

González-Martínez, M. Á., Brun, E. M., Puchades, R., Maquieira, Á., Ramsey, K., & Rubio, F. (2005). Glyphosate immunosensor. Application for water and soil analysis. *Analytical Chemistry, 77*, 4219–4227.

Goodwin, L., Startin, J. R., Keely, B. J., & Goodall, D. M. (2003). Analysis of glyphosate and glufosinate by capillary electrophoresis–mass spectrometry utilising a sheathless microelectrospray interface. *Journal of Chromatography A, 1004*, 107–119.

Hanke, I., Singer, H., & Hollender, J. (2008). Ultratrace-level determination of glyphosate, aminomethylphosphonic acid and glufosinate in natural waters by solid-phase extraction followed by liquid chromatography–tandem mass spectrometry: Performance tuning of derivatization, enrichment and detection. *Analytical and Bioanalytical Chemistry, 391*, 2265–2276.

Hori, Y., Fujisawa, M., Shimada, K., Sato, M., Kikuchi, M., Honda, M., et al. (2002). Quantitative determination of glufosinate in biological samples by liquid chromatography with ultraviolet detection after *p*-nitrobenzoyl derivatization. *Journal of Chromatography B, 767*, 255–262.

Hori, Y., Fujisawa, M., Shimada, K., Sato, M., Honda, M., & Hirose, Y. (2002). Enantioselective analysis of glufosinate using precolumn derivatization with (+)-1-(9-fluorenyl)ethyl chloroformate and reversed-phase liquid chromatography. *Journal of Chromatography B, 776*, 191–198.

Ibáñez, M., Pozo, Ó. J., Sancho, J. V., López, F. J., & Hernández, F. (2005). Residue determination of glyphosate, glufosinate and aminomethylphosphonic acid in water and soil samples by liquid chromatography coupled to electrospray tandem mass spectrometry. *Journal of Chromatography A, 1081*, 145–155.

Kataoka, H., Ryu, S., Sakiyama, N., & Makita, M. (1996). Simple and rapid determination of the herbicides glyphosate and glufosinate in river water, soil and carrot samples by gas chromatography with flame photometric detection. *Journal of Chromatography A, 726*, 253–258.

König, A., Cockburn, A., Crevel, R. W. R., Debruyne, E., Grafstroem, R., Hammerling, U., et al. (2004). Assessment of the safety of foods derived from genetically modified (GM) crops. *Food and Chemical Toxicology, 42*(7), 1047–1088.

Kumar, V., Bellinder, R. R., Brainard, D. C., Malik, R. K., & Gupta, R. K. (2008). Risks of herbicide-resistant rice in India: A review. *Crop Protection, 27*, 320–329.

Kuster, M., Alda, M. L., & Barceló, D. (2009). Liquid chromatography–tandem mass spectrometric analysis and regulatory issues of polar pesticides in natural and treated waters. *Journal of Chromatography A, 1216*, 520–529.

MAFF, 1990. Ministry of Agriculture Fisheries and Food, London.

Motojyuku, M., Saito, T., Akieda, K., Otsuka, H., Yamamoto, I., & Inokuchi, S. (2008). Determination of glyphosate, glyphosate metabolites, and glufosinate in human serum by gas chromatography–mass spectrometry. *Journal of Chromatography B, 875*, 509–514.

Orejuela, E., & Silva, M. (2005). Rapid and sensitive determination of phosphorus-containing amino acid herbicides in soil samples by capillary zone electrophoresis with diode laser-induced fluorescence detection. *Electrophoresis, 26*, 4478–4485.

Ozbey, A., & Uygun, U. (2007). Behaviour of some organophosphorus pesticide residues in peppermint tea during the infusion process. *Food Chemistry, 104*, 237–241.

Pitzer, K. K., Werbovetz, K. A., Brendle, J. J., & Scovill, J. P. (1998). Synthesis and biological evaluation of 4-chloro-3,5-dinitrobenzotrifluoride analogues as antileishmanial agents. *Journal of Medicinal Chemistry, 41*, 4885–4889.

Qian, K., Tang, T., Shi, T. Y., Wang, F., Li, J. Q., & Cao, Y. S. (2009). Residue determination of glyphosate in environmental water samples with high-performance liquid chromatography and UV detection after derivatization with 4-chloro-3,5-dinitrobenzotrifluoride. *Analytica Chimica Acta, 635*, 222–226.

Royer, A., Beguin, S., Sochor, H., & Communal, P. Y. (2000). Determination of glufosinate ammonium and its metabolite (AEF064619 and AEF061517) residues in water by gas chromatography with tandem mass spectrometry after ion exchange cleanup and derivatization. *Journal of Agricultural Food Chemistry, 48*, 5184–5189.

Sadi, B. B. M., Vonderheide, A. P., & Caruso, J. A. (2004). Analysis of phosphorus herbicides by ion-pairing reversed-phase liquid chromatography coupled to inductively coupled plasma mass spectrometry with octapole reaction cell. *Journal of Chromatography A, 1050*, 95–101.

Sancho, J. V., Hernandez, F., López, F. J., Hogendoorn, E. A., Dijkman, E., & van Zoonen, P. (1996). Rapid determination of glufosinate, glyphosate and aminomethylphosphonic acid in environmental water samples using precolumn fluorogenic labeling and coupled-column liquid chromatography. *Journal of Chromatography A, 737*, 75–83.

Tomlin, C. D. S. (1999). The electronic pesticide manual (11th ed.), Version 1.0. British Crop Protection Council, Alton: UK.

Tseng, S. H., Lo, Y. W., Chang, P. C., Chou, S. S., & Chang, H. M. (2004). Simultaneous quantification of glyphosate, glufosinate, and their major metabolites in rice

and soybean sprouts by gas chromatography with pulsed flame photometric detector. *Journal of Agricultural Food Chemistry, 52*, 4057–4063.

Wang, K. C., Chen, S. M., Hsu, J. F., Cheng, S. G., & Lee, C. K. (2008). Simultaneous detection and quantitation of highly water-soluble herbicides in serum using ion-pair liquid chromatography–tandem mass spectrometry. *Journal of Chromatography B, 876*, 211–218.

Zhang, L. Y., Liu, Y. M., Wang, Z. L., & Cheng, J. K. (2004). Capillary zone electrophoresis with pre-column NDA derivatization and amperometric detection for the analysis of four aliphatic diamines. *Analytica Chimica Acta, 508*, 141–145.

Zhou, L., Luo, Z., Wang, S. M., Hui, Y., Hu, Z. D., & Chen, X. G. (2007). In-capillary derivatization and laser-induced fluorescence detection for the analysis of organophosphorus pesticides by micellar electrokinetic chromatography. *Journal of Chromatography A, 1149*, 377–384.

Zimdahl, R. L. (1993). *Fundamentals of weed science*. London: Academic Press.