이학석사 학위논문

Liquid extraction surface analysis coupled with capillary electrophoresis to determine pesticides on apple skin

사과 표면의 농약 검출을 위한 액체 추출 표면 분석법과 연동된 모세관 전기영동법

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액체 추출 표면 분석법과 연동된 모세관 전기영동법

지도교수 정 두 수

이 논문을 이학석사 학위논문으로 제출함.

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Abstract

Liquid extraction surface analysis coupled with capillary electrophoresis to determine pesticides on apple skin

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A surface-sampling technique of liquid extraction surface analysis (LESA) was coupled with capillary electrophoresis (CE) to determine organophosphorous pesticides, including glufosinate-ammonium, aminomethylphosphonic acid, and glyphosate on the external surface of a fruit such as an apple. Without any sample pretreatment, the analytes sprayed on the surface of a half apple were directly extracted into a liquid microjunction formed by dispensing the extractant from the inlet tip of a separation capillary. The extraction efficiency was enhanced by repeating the steps of dispensing and aspirating the extractant drop. After extraction, the analytes were derivatized in-capillary with a fluorophore 4-flouro-7-nitro-2,1,3-benzoxadiazole and analyzed with CE-laser induced fluorescence (LIF) detection. The developed LESA-CE/LIF carries out quantitative analysis in a convenient and rapid manner, which is made possible since the extraction and separation of analytes can be performed in-line. The

limits of detection for glufosinate-ammonium, aminomethylphosphonic acid, and glyphosate with LESA-CE/LIF were 2.5, 1, and 10 ppb, respectively, which are at least 20 times lower than the tolerance established by the U.S. Environmental Protection Agency. Thus, we demonstrated that LESA-CE is a quite sensitive and convenient method to determine analytes on sample surfaces avoiding the dilution problem from sample pretreatment procedures including homogenization of bulk samples.

Keywords:

Liquid extraction surface analysis, Capillary electrophoresis, Organophosphorous pesticides, Pesticide residue analysis

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1 INTRODUCTION

Capillary electrophoresis (CE) is well suited for the analysis of analytes in aqueous solutions. For water insoluble analytes, nonaqueous CE (NACE) [1-5] and micellar electrokinetic chromatography (MEKC) [6-8] can be used. In addition, headspace extraction has recently been in-line coupled to CE [9] for the analysis of gaseous analytes evaporated from a liquid sample. However, it is not yet easy to analyze solid samples with CE since labor-intensive sample pretreatment processes are required. For example, a solid sample should be grounded, homogenized, centrifuged, extracted into an organic solvent, and then reconstituted into an aqueous solution suitable for CE.

Here, we suggest in-line coupling of liquid extraction surface analysis (LESA) with CE to extract analytes directly from a solid surface and subsequently analyze the extracts. LESA is a surface sampling technique that extracts chemicals on the surface of various solid samples without sample pretreatment. Since the development of a commercial LESA device using a chip-based robotic nanoelectrospray platform [10], LESA-ESI/MS using the platform has been utilized in the analysis of not only food surfaces but also biological tissues [11-15]. However, LESA-ESI/MS without separation provides limited quantitative information due to the lack of separation [11].

Glufosinate-ammonium and glyphosate are widely used non-infiltrative organophosphorous pesticides (OPPs) for the last thirty years worldwide. When non-infiltrative pesticides are applied to fruits such as apples, they remain on the fruit skin without translocation to the fruit pulp. The U.S. Environmental Protection Agency (EPA) limits the allowable amount of glufosinate-ammonium and glyphosate in an apple to 50 [16] and 200 ppb [17], respectively. Until now, analyses of these OPPs with GC [18, 19], LC [20, 21], and CE [22-24] require sample pretreatment processes including homogenization of a bulk sample causing dilution of the analytes existing only on the sample surface. In addition, these methods have other analytical challenges not only from the low liquid-liquid extraction efficiency due to the high water solubility and polarity of the OPPs, but also from a lack of chromophores.

We solved the challenges of analyzing these OPPs (glufosinate-ammonium, glyphosate, and aminomethylphosphonic acid (AMPA) which is a metabolite of glyphosate) on apple skins through LESA in-line coupled with CE using a separation capillary instead of the robotic arm of a commercial LESA device [10]. The OPPs on the apple skin were directly extracted into the separation capillary and then the extracts in the capillary were derivatized by a fluorophore in-capillary and detected by laser induced fluorescence (LIF). LESA-CE/LIF is a quite simple but sensitive method not only increasing the extraction efficiency of the OPPs by using an aqueous extraction solvent but

also avoiding the dilution problem of homogenization of a bulk sample. As a result, we could quantitatively determine the OPPs on an apple in the range of 5-200 ppb for glufosinate-ammonium and AMPA, and 20-800 ppb for glyphosate. The limits of detection (LODs) of glufosinate-ammonium, AMPA, and glyphosate with LESA-CE/LIF were 2.5, 1, and 10 ppb, respectively, which are at least 20 times lower than the tolerance limit set by the EPA. One important achievement of LESA-CE is the expansion of specimen types applicable to CE to solid surface.

2 EXPERIMENTAL

2.1 Reagents

Glufosinate-ammonium, glyphosate, AMPA, ethanol, sodium dodecyl sulfate (SDS), sodium tetraborate, and HCl were obtained from Sigma (St. Louis, MO, USA). Acetonitrile (ACN) was from Burdick & Jackson (Radnor, PA, USA). 4-flouro-7-nitro-2,1,3-benzoxadiazole (NBD-F) was from TCI (Tokyo, Japan). NaOH was from Dae Jung Chemicals (Siheung, Korea). For each pesticide, 0.04 mg mL^{-1} standard stock solution was prepared in ethanol and stored at 4℃. Pesticide mixtures were prepared by diluting the stock solutions with ethanol. Derivatization reagents of NBD-F were prepared in ACN. 100 mM sodium tetraborate and 100 mM SDS were used as stock buffer solutions. A run buffer of 10 mM sodium tetraborate containing 10 mM SDS and 10% (v/v) ACN was prepared by mixing appropriate volumes of the stock solutions and ACN, and then by titrating to pH 9.90 with 1.0 M NaOH. The run buffer solution was filtered through a 0.45-μm syringe filter (Sigma). An extraction solvent of 10 mM sodium tetraborate was prepared by diluting the 100 mM sodium tetraborate with deionized water. Before use, the extraction solvent was adjusted to pH 8.75 with 1.0 M HCl for compatibility with the in-capillary derivatization of analytes dissolved in the solvent [25]. Deionized water was prepared with a NANOpure II purification system (Barnstead, Dubuque, IA, USA).

2.2 Apparatus

Experiments were performed with a homemade CE system comprised of a high voltage power supply (CZE1000R; Spellman, Hauppage, NY, USA) and an LIF detection system. Analytes were excited at 488 nm with a 10-mW argon-ion laser (Omnichrome, Chino, CA, USA). Fluorescent light was collected at a right angle through a 10× microscope objective (Edmund Industrial Optics, Barrington, NJ, USA), passed through a 488 nm notch filter (NT67-108; Edmund Industrial Optics) and a 520 nm band pass filter (Melles Griot, Irvine, CA, USA), and detected with an integrated photomultiplier tube system (PMT; HC 120-01, Hamamatsu, Bridgewater, NJ, USA). The power supply and PMT were interfaced to a computer using an MIO-16-XE DAQ board (National Instruments, Austin, TX, USA). The LabView 7.0.1 program (National Instruments) was used for data acquisition and analysis. A fused silica capillary of an internal diameter of 70 μm and 60 cm in length (50 cm to the detector) from Polymicro Technologies (Phoenix, AZ, USA) was used. Before a run, the capillary was rinsed with 0.1 M NaOH, water, and then a run buffer each for at least 10 min with a syringe. Separation was performed at 10 kV.

2.3 Sample preparation

Apples grown with a conventional farming system and a plastic household pump sprayer were purchased from a local store. Pesticide-free apples were obtained from a local shop for organic agricultural products without pesticides. Both unwashed and washed apples were analyzed. Apple halves were weighted individually. A mixture of three pesticides in 5 mL of ethanol was sprayed against the surface of an apple half and air-dried for 15 min. A random slice of apple skin was fixed on a 50 mm Teflon plate with scotch tape, and placed on the homemade CE set up for LESA.

2.4 LESA

Fig. 1 shows the scheme for LESA-CE/LIF. The inlet end of a capillary filled with a run buffer was immersed in an extraction solvent vial and the outlet end in a destination vial containing the run buffer. The extraction solvent was injected by gravity, raising the solvent vial by 7.5 cm above the destination vial for 315 s. The volume of injection estimated with the Poiseuille equation was 350 nL. The inlet of the capillary containing the extraction solvent was transferred to a capillary holder aligned to a target spot on the sample surface. The distance between the sample surface and the capillary was about 0.4 mm. To form a liquid microjunction on the target spot, the sample assembly (sample and capillary holder) was lowered by 18.5 cm below the destination vial for 120 s dispensing 330 nL of the extraction solvent (dispensing step). The surface

area wetted with the extraction solvent was about 0.5 mm^2. Then, 340 nL of the extraction solvent was aspirated into the capillary by raising the sample assembly by 7.5 cm above the destination vial for 300 s (aspiration step). The extraction efficiency was enhanced by repeating the dispensing/aspiration cycle 3 times. Note that the extracted analytes were injected into the capillary through the last aspiration step. To minimize the peak broadening, a small portion (45 nL) of the extractant drop formed during the third dispensing step was injected into the capillary by gravity at 7.5 cm for 40 s. After injection, the capillary was removed from the capillary holder and placed into an NBD-F vial. During this process taking about 7 s, 20 nL of the injected extractant drop was re-dispensed. Thus, the actual injection volume was about 25 nL.

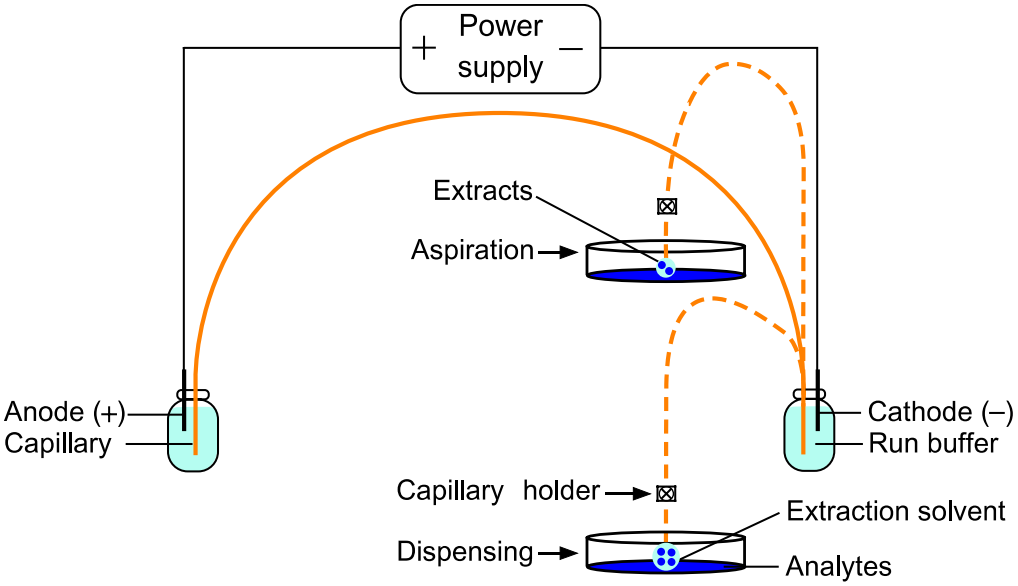


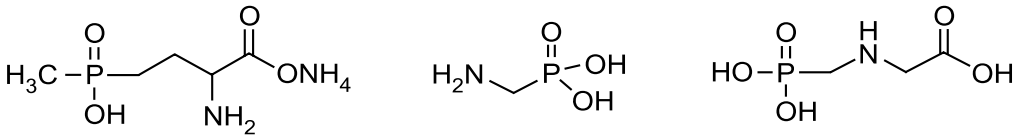
Figure 1. Scheme for LESA-CE. Dashed lines represent the extraction cycle of dispensing and aspiration steps. LESA: extraction solvent of 10 mM sodium tetraborate buffer of pH 8.75; injection of 350 nL extraction solvent; dispensing of 330 nL; aspiration of 340 nL; 3 times of dispensing/aspiration cycle.

2.5 CE-LIF

LIF detection was used after derivatizing the OPPs with NBD-F in-capillary [25]. NBD-F reacts fast with both primary and secondary amines with high efficiency (see Fig. 2) [26]. Following the injection of extracted analytes through the last aspiration step, 25 nL of the NBD-F solution was injected by gravity. Then, the anionic sample and neutral NBD-F zones were electrokinetically mixed by applying a potential of 5 kV for 21 s [25]. Afterwards, the reaction was allowed to proceed for 10 min without an electric field. During the in-capillary derivatization and separation steps, the temperature inside the homemade CE set up was maintained at 32 ± 2℃ by a heater.

The NBD-derivatized analytes were successfully separated in a micellar electrokinetic capillary chromatography (MEKC) mode with SDS as a surfactant and ACN as an organic modifier. The composition of the separation run buffer was 10 mM sodium tetraborate, 10 mM SDS, and 10% (v/v) ACN at pH 9.90 [25]. Separation was performed at a constant voltage of 10 kV.

i) Analytes



ii) Derivatization with NBD-F

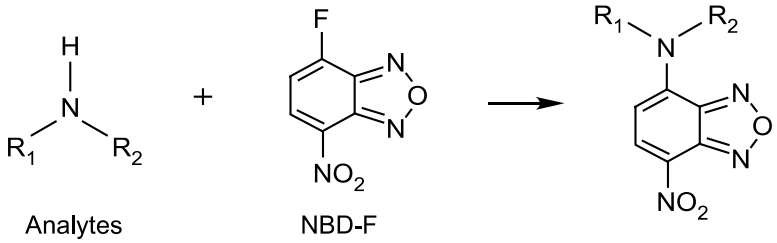


Figure 2. Analytes and the derivatization reaction with NBD-F.

2.6 Solid phase extraction-CE/LIF

For the validation of LESA-CE, solid phase extraction (SPE) [27-29] off-line coupled with CE was performed as described in Ref [30]. Apple halves sprayed with AMPA (having the highest extraction efficiency among the OPPs) at levels of 25, 50, 100, and 200 ppb were milled using a home mixer. A 1-mg aliquot of each milled apple sample was added to 20 mL of 10 mM sodium tetraborate buffer, shaken for 1 h, and then centrifuged at 4000 rpm for 20 min. 2 mL of each supernatant was passed through a SPE cartridge (SampliQ Silica C_{18}, Agilent Technologies, Seoul, Korea) pre-rinsed with 3 mL ACN and 3 mL water. An aliquot of the collected sample and 20 mM NBD-F reagent were introduced sequentially into the capillary at 0.5 psi for 5 s, using a commercial CE instrument (P/ACE MDQ, Beckman, Fullerton, CA, USA) for in-capillary derivatization. In-capillary derivatization and CE conditions were the same as in section 2.5. The temperature of the capillary cartridge was maintained at 32℃.

3 RESULTS AND DISCUSSION

3.1 Number of extraction cycles

Extraction cycles were repeated from 1 to 4 times on a target spot of the apple skin sprayed with the OPPs at the concentration of 50 ppb glufosinate-ammonium, 200 ppb glyphosate (EPA tolerance limits), and 50 ppb AMPA. The extraction efficiency increased with the extraction cycles up to 3 times, whereas repeating the extraction cycle 4 times caused decreased peak heights and poor reproducibility, probably due to the loss of an extractant drop. The relative standard deviations (RSDs) of the peak heights were less than 16% with 3 extraction cycles.

3.2 Optimization of in-capillary derivatization

A commercial CE instrument was used for the optimization of the in-capillary derivatization conditions. A previous report on the in-capillary derivatization of the OPPs served as a starting point for the re-optimization: 5 mM of NBD-F, 30 s of mixing time, and 7.5 min of stand-by time at 35℃ [25]. We increased the NBD-F concentration and adjusted the mixing and stand-by times. After the re-optimization, the fluorescence signal intensity was improved by 6-7 times.

The NBD-F concentration was varied from 5 to 40 mM while the volumes of the sample (200 μM glufosinate-ammonium, AMPA, and

glyphosate in the extraction solvent) and NBD-F solutions were maintained at 25 nL (see Fig. 3a). The standard analytes were more effectively derivatized up to 20 mM NBD-F. Any further increase in NBD-F concentration contributed little to improving the peak heights but the baseline noise was increased. Thus, 20 mM NBD-F was chosen as the optimal concentration.

The effect of the electrokinetic mixing time was investigated from 0 to 60 s at 5 kV. As seen in Fig. 3b, the peak heights increased with the mixing time up to 21 s. With a longer mixing time, the peak heights decreased. Thus, a mixing time of 21 s was chosen.

Once the two zones were overlapped, the derivatization reaction proceeded without a potential during a stand-by time. The effect of the stand-by time in the range of 0-15 min was investigated. As shown in Fig. 3c, when the stand-by time was increased from 0 to 10 min, the fluorescence signal intensity increased about 5 times for glufosinate-ammonium or 1.5-2 times for glyphosate and AMPA. However, a longer stand-by time caused a decrease; thus, 10 min was chosen as the optimal stand-by time.

The effect of temperature on the derivatization in the range of 23-40℃ was investigated. As seen in Fig. 3d, the derivatization efficiency increased as the temperature increased from 23 to 32℃, and then decreased afterwards. Thus, the ambient temperature around the homemade CE set up was maintained at about 32℃ with a heater.

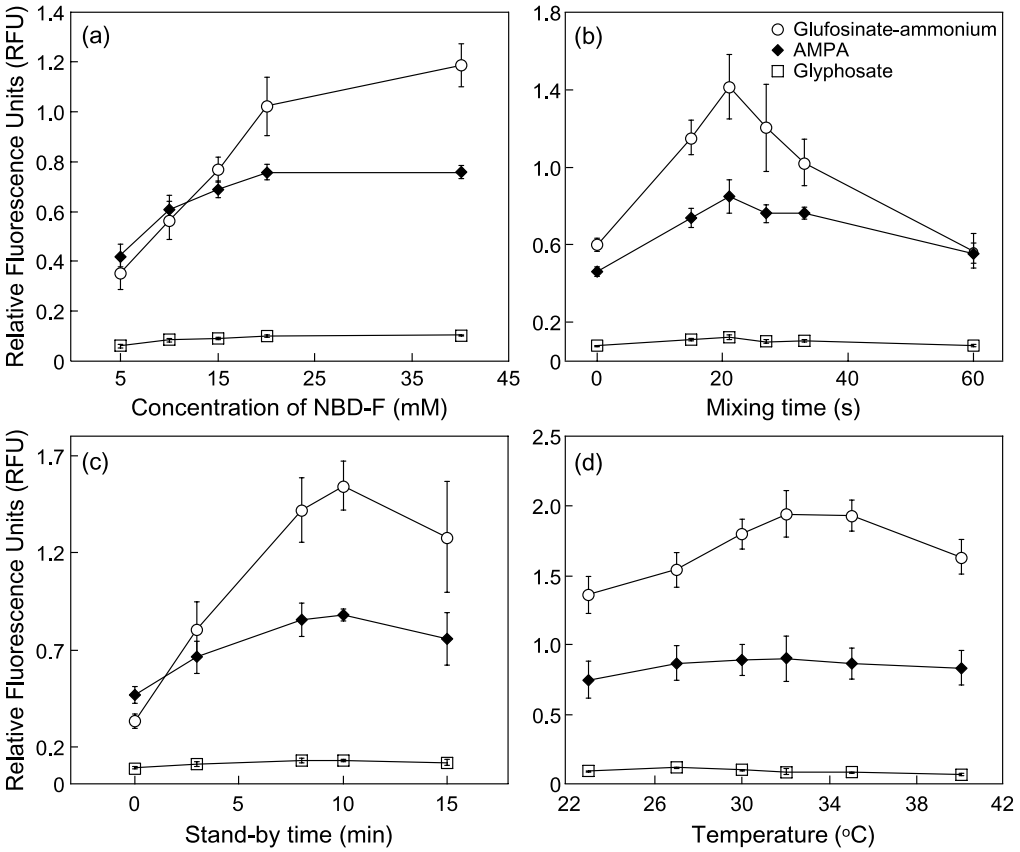


Figure 3. Optimization of the (a) concentration of NBD-F, (b) electrokinetic mixing time, (c) stand-by time, and (d) temperature. Sample solution, 200 μM glufosinate-ammonium, AMPA, and glyphosate in 10 mM sodium tetraborate of pH 8.75; sequential injection of sample and NBD-F at 0.5 psi for 5 s; fused silica capillary, 75 μm ID, 50/60 cm; run buffer, 10 mM sodium tetraborate, 10 mM SDS, 10 % (v/v) ACN (pH 9.90); 10 kV; P/ACE MDQ CE with LIF detection, λex = 488 nm, λem = 520 nm. Error bars represent the standard deviations (n = 4).

3.3 Analytical performance of LESA-CE/LIF

Under the optimal conditions, the RSDs (n = 4) of the peak heights for the OPPs sprayed on an apple skin at 50 ppb glufosinate-ammonium, 200 ppb glyphosate (EPA tolerance limit), and 50 ppb AMPA were 15%, 16%, and 11%, respectively. The LODs (S/N = 3) were 2.5, 10, and 1 ppb, respectively, which are at least 20 times lower than the EPA tolerance limit. The linear responses were excellent in the ranges of 5-200 ppb for glufosinate-ammonium and AMPA, and 20-800 ppb for glyphosate with a linear correlation coefficients (r) greater than 0.9913. The results summarized in Table 1 clearly show that our LESA-CE/LIF scheme can be used for the determination of the non-infiltrative OPP residues on an apple with great sensitivity and ease. The sensitivity of LESA-CE/LIF was comparable to that obtained with a conventional pesticide analysis method of off-line SPE-HPLC which included labor-intensive pretreatment processes [30].

To further evaluate the quantifying reliability of LESA, the results for AMPA of 25, 50, 100, and 200 ppb obtained from in-line LESA-CE/LIF and off-line SPE-CE/LIF were compared. Fig. 4 shows that the two results agreed well. The two data sets yielded a regression curve of $y = 0.9177x + 3.4589$ with $r = 0.9934$, where x and y are the AMPA concentrations in ppb obtained with SPE-CE/LIF and LESA-CE/LIF, respectively.

Table 1. Analytical performance of LESA-CE

Compound	Tolerance limit (ppb)	Linear range (ppb)	RSD (n=4)	*r*	LOD (ppb)
Glufosinate-ammonium	50	5-200	15%	0.9953	2.5
AMPA	–	5-200	11%	0.9955	1
Glyphosate	200	20-800	16%	0.9913	10

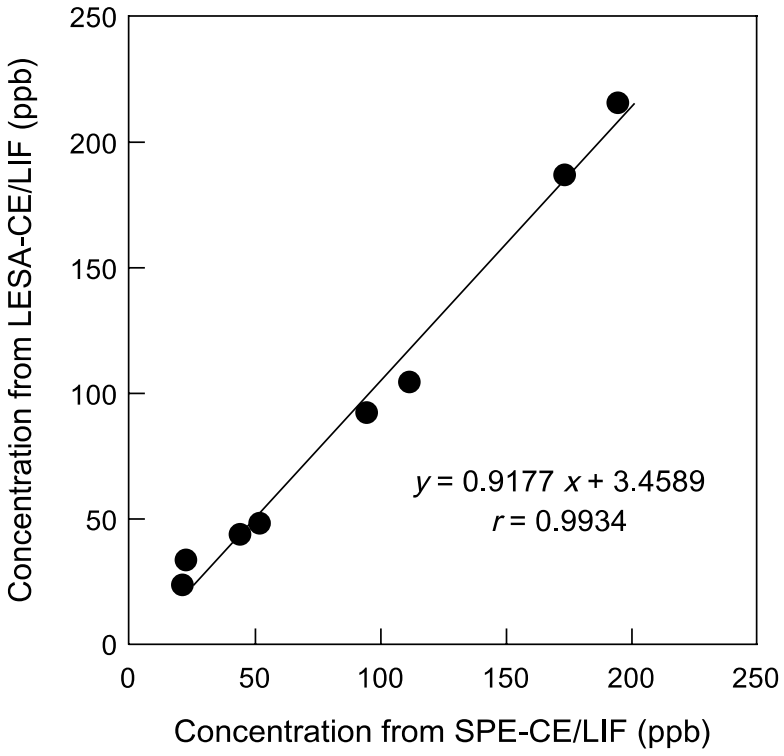


Figure 4. Comparison of LESA-CE/LIF and SPE-CE/LIF.

3.4 LESA-CE from an unspiked apple surface

Apples purchased from a local store without spraying of the OPPs were evaluated in the same manner as the artificially sprayed apples at 50 ppb glufosinate-ammonium, 200 ppb glyphosate, and 50 ppb AMPA (see Fig. 5a). Fig. 5b shows the electropherogram from a control apple peel without spraying. Interestingly, a peak at the same migration time as the AMPA peak in Fig. 5a also appeared in Fig. 5b. The peak became absent from the surface of an apple rinsed under running tap water using a dish sponge (Fig. 5c). When LESA-CE/LIF was carried out for unwashed pesticide-free apples, the AMPA peak was absent (Fig. 5d). The peak of AMPA, which is a metabolite of glyphosate, in Fig. 5b suggests the possibility of the application of glyphosate to the apple. The corresponding glyphosate residue can be estimated with the following relation:

Total ppb of glyphosate = ppb of glyphosate + ppb of AMPA × 1.52

where the conversion factor 1.52 is the molecular weight ratio of glyphosate (169.1) and AMPA (111.0). The calculated glyphosate residue by the expression was at least 20-fold below the EPA tolerance level.

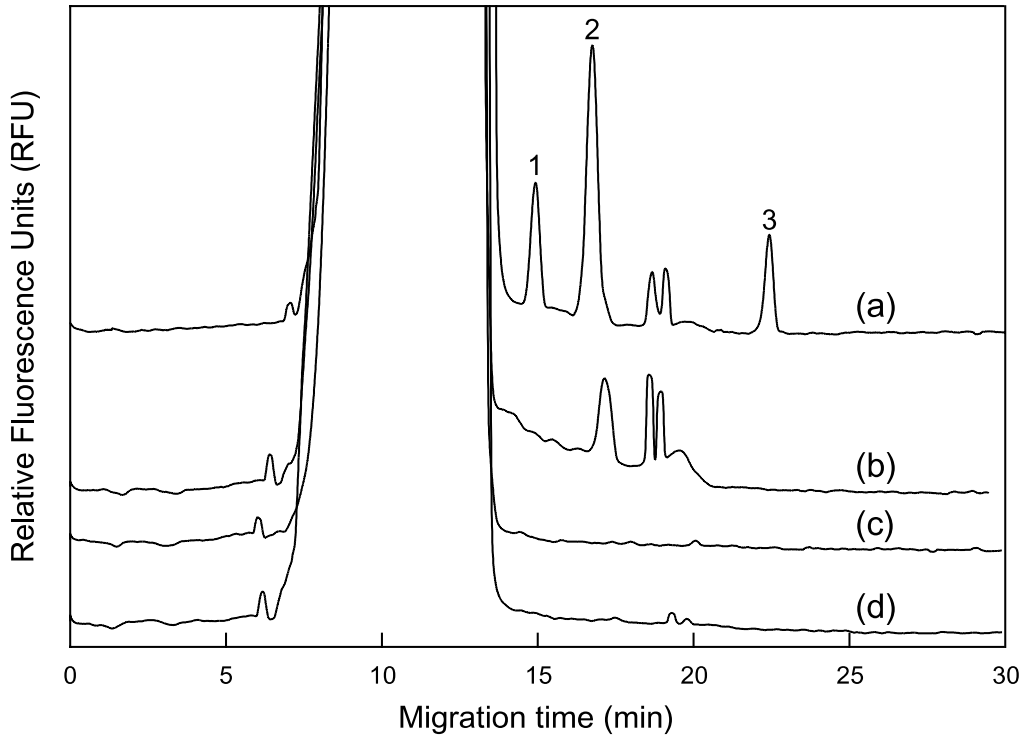


Figure 5. Electropherograms for (a) an apple peel artifitially sprayed (50 ppb glufosinate-ammonium, 50 ppb AMPA, and 200 ppb glyphosate) after washing, (b) apple peel without washing and spraying, (c) apple peel after washing, and (d) organic apple peel without washing and spraying. Other conditions as in Figs. 1 and 3. Peak identification: 1) glufosinate-ammonium, 2) AMPA, 3) glyphosate.

4. CONCLUSIONS

In-line coupling of LESA with CE was demonstrated for the analysis of non-infiltrative OPPs on an apple skin, overcoming the limitation on specimen types applicable to CE. The advantage of in-line coupled LESA-CE is that quantitative analysis can be performed in a convenient manner without dilution of the target analytes on a solid surface during bulk sample preparation. By in-capillary derivatization with the fluorophore NBD-F, the LODs were 2.5, 1, and 10 ppb for glufosinate-ammonium, AMPA, and glyphosate, respectively, which are about 20-fold lower than the EPA tolerance limits. LESA-CE/LIF using a commercial CE instrument is in progress.

REFERENCES

[1] B. Kim, M.S. Chun, S. Shin, D.S. Chung, B Kor Chem Soc 20 (1999) 1483.

[2] B. Kim, D.S. Chung, Electrophoresis 23 (2002) 49.

[3] J. Kim, M.S. Chun, K. Choi, D.S. Chung, Electrophoresis 30 (2009) 1046.

[4] K. Choi, Y.G. Jin, D.S. Chung, J Chromatogr A 1216 (2009) 6466.

[5] Y. Lu, M.C. Breadmore, J Chromatogr A 1217 (2010) 7282.

[6] M.C. Breadmore, M. Macka, P.R. Haddad, Anal Chem 71 (1999) 1826.

[7] S.H. Kang, H. Jung, N. Kim, D.H. Shin, D.S. Chung, J Chromatogr A 866 (2000) 253.

[8] I. Lee, M.C. Boyce, M.C. Breadmore, Anal Chim Acta 663 (2010) 105.

[9] S.T. Park, J. Kim, K. Choi, H.R. Lee, D.S. Chung, Electrophoresis 33 (2012) 2961.

[10] V. Kertesz, G.J. Van Berkel, J Mass Spectrom 45 (2010) 252.

[11] D. Eikel, J. Henion, Rapid Commun Mass Sp 25 (2011) 2345.

[12] D. Eikel, M. Vavrek, S. Smith, C. Bason, S. Yeh, W.A. Korfmacher, J.D. Henion, Rapid Commun Mass Sp 25 (2011) 3587.

[13] M.J. Walworth, M.S. ElNaggar, J.J. Stankovich, C. Witkowski, J.L. Norris, G.J. Van Berkel, Rapid Commun Mass Sp 25 (2011) 2389.

[14] S. Schadt, S. Kallbach, R. Almeida, J. Sandel, Drug Metab Dispos 40 (2012) 419.

[15] W.B. Parson, S.L. Koeniger, R.W. Johnson, J. Erickson, Y. Tian, C. Stedman, A. Schwartz, E. Tarcsa, R. Cole, G.J. Van Berkel, J Mass Spectrom 47 (2012) 1420.

[16] http://www.ecfr.gov/cgi-bin/text-idx?c=ecfr&SID=add74377d7733bc8c5ba517c8b34a44a&rgn=div8&view=text&node=40:25.0.1.1.27.3.19.209&idno=40, 26. 11. 2013.

[17] http://www.ecfr.gov/cgi-bin/text-idx?c=ecfr&SID=add74377d7733bc8c5ba517c8b34a44a&rgn=div8&view=text&node=40:25.0.1.1.27.3.19.124&idno=40, 26. 11. 2013.

[18] C.D. Stalikas, G.A. Pilidis, J Chromatogr A 872 (2000) 215.

[19] S.H. Tseng, Y.W. Lo, P.C. Chang, S.S. Chou, H.M. Chang, J Agr Food Chem 52 (2004) 4057.

[20] E.A. Hogendoorn, F.M. Ossendrijver, E. Dijkman, R.A. Baumann, J Chromatogr A 833 (1999) 67.

[21] I. Hanke, H. Singer, J. Hollender, Anal Bioanal Chem 391 (2008) 2265.

[22] H.Y. Chiu, Z.Y. Lin, H.L. Tu, C.W. Whang, J Chromatogr A 1177 (2008) 195.

[23] C.C. Hsu, C.W. Whang, J Chromatogr A 1216 (2009) 8575.

[24] H.H. See, P.C. Hauser, M.M. Sanagi, W.A.W. Ibrahim, J Chromatogr A 1217 (2010) 5832.

[25] L. Zhou, Z. Luo, S.M. Wang, Y. Hui, Z. Hu, X. Chen, J Chromatogr A 1149 (2007) 377.

[26] S. Hu, P.C.H. Li, J Chromatogr A 876 (2000) 183.

[27] D. Schaller, E.F. Hilder, P.R. Haddad, Anal Chim Acta 556 (2006) 104.

[28] O.G. Potter, E.F. Hilder, J Sep Sci 31 (2008) 1881.

[29] E. Candish, A. Gooley, H.J. Wirth, P.A. Dawes, R.A. Shellie, E.F. Hilder, J Sep Sci 35 (2012) 2399.

[30] K. Qian, T. Tang, T. Shi, P. Li, J. Li, Y. Cao, J Sep Sci 32 (2009) 2394.

국문 초록

표면으로부터의 시료추출법인 액체 추출 표면 분석법 (LESA)이 모세관 전기영동 (CE)과 연동되어 사과와 같은 과일 표면에 잔류하는 유기인계 농약을 분석하는데 이용되었다. 분석물질인 농약이 뿌려진 반 쪽짜리 사과 껍질 위에 모세관 주입부의 끝 단을 놓고 추출용매 방울을 만듦으로써 사과 표면과 추출용매 사이에 액체 미세 접합부분을 형성시켜 이를 통해 시료전처리 과정 없이 분석물질을 바로 추출 할 수 있다. 시료 표면 위에 추출용매 방울을 형성시키고 다시 흡입시키는 일련의 추출과정을 반복함으로써 추출 효율을 높일 수 있다. 추출 후, 모세관에 주입된 분석물질을 모세관 내에서 4-fluoro-7-nitro-2,1,3-benzoxadiazole (NBD-F)와 형광 유도체화 반응을 시켜 레이저 유도 형광 (LIF) 검출기로 검출하였다. 액체 추출 표면 분석법과 연동된 모세관 전기영동 형광 검출법 (LESA-CE/LIF)은 분석물질의 추출과 분리과정을 인라인으로 연동함으로써 정량분석을 편리하고 빠르게 수행하였다. LESA-CE/LIF를 사과 표면에 잔류하는 유기인계 농약을 분석하는데 사용했을 때 glufosinate-ammonium과 aminomethylphosphonic acid와 glyphosate의 검출한계는 2.5, 1, 10 ppb 로 미국환경보호청 (EPA)이 제시하는 허용량의 20분의 1정도의 농도에서 검출이 가능했다. 개발된 LESA-CE/LIF는 시료의 전처리 과정을 거치지 않으므로 전처리 과정 중 벌크 시료의 균질화 과정에서 유발되는 표면 잔류 분석물질의 묽힘 문제가 생기지 않게 되어, 매우 민감하고 편리한 시료 표면의 분석물질 검출법이라고 할 수 있다.

주요어: 액체 추출 표면 분석, 모세관 전기영동, 유기인계 농약, 잔류농약 분석

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