

Analysis of glyphosate residues in manure fertilizers

Hakala K¹, Lindroth T¹, Torniainen M², Nieminen J¹

Finnish Food Safety Authority Evira,

¹ Research and Laboratory Department, Chemistry and Toxicology Research Unit

² Control Department, Feed and Fertilizer Control Unit

Introduction

Glyphosate is the most commonly used herbicide in the agriculture and gardens worldwide.¹ Farmed animals diet typically include cereal and soya based feed; both being components whose cultivation glyphosate products are generally used against weeds. Obviously this could lead to some small allowed residues in feed. However, glyphosate may be excreted unchanged to the manure and lead to unwanted residue in it. This could be problematic as manure is valuable and widely used fertilizer in organic farming.

In this work, method based on derivatization with FMOC-Cl and UPLC-MS/MS analysis was optimized and validated for the determination of glyphosate residues in manure fertilizers.

Method

Fertilizer samples are extracted with water. Extracts are eluted through SPE cartridges to remove matrix components and derivatized with FMOC-Cl (fluorenylmethoxycarbonyl chloride). Samples are analyzed by liquid chromatography mass spectrometry with positive electrospray ionization (Shimadzu LCMS-8050, Shimadzu Nexera X2).



Figure 1. Sample pre-treatment and derivatization procedure.

Table 1. LC-MS/MS parameters

Column	Acquity UPLC@BEH C18 1.7µm 2.1x100 mm
Flow rate	0.250 µl/min
Injection volume	5 µl
Eluents	10 mM NH ₄ OAc (A) and ACN (B)
Gradient profile	10%→95% B 5 min, 95% B 5-8 min, 95%→10%B 1 min
Monitored reactions and corresponding collision energies (eV)	Glyphosate: m/z 392.0→88.0 (CE -25 eV), 392.0→214.0 (-10 eV), 392.0→170.0 (CE -13 eV), 392.0→179.0 (CE -20 eV) Glyphosate 2-C13 (IS) 394.0→90.0 (CE -25 eV), 394.0→172.0 (-13 eV), 394.0→216.0 (CE -13 eV)
Interface voltage	3.5 kV
Temperatures (mass spectrometry)	Interface Temp 350 °C, DL Temp: 250 °C
Gas flows (mass spectrometry)	Neb gas flow 1.5 L/min, Heating gas flow:10.00L/min, Drying gas flow 5.00L/min.

Results

Method optimization

The analytical method that is routinely used in our lab for cereals, was not applicable to manure samples because of remarkably higher detection level of glyphosate in manure. Therefore, following improvements to the method were made in order to minimize matrix effects and decrease detection level:

- exchange of column: C18 3 µm 2.1x150 mm → C18 1.7µm 2.1x100 mm
- lower sample size: 5g→3g
- add of purification step with Oasis PRIME HLB cartridge
- remove of borate buffer (pH=9) from the derivatization step

Repeatability

Table 2. Within-day and between-day repeatability (Relative standard deviation, RSD%).

Spiking level (mg/kg)	Repeatability within-day (n=5)		Repeatability between three day (n=6)	
	Average concentration (mg/kg)	RSD%	Average concentration (mg/kg)	RSD%
0.050	0.050	3.8	0.048	6.2
0.500	0.518	2.1	0.511	7.0

Linearity

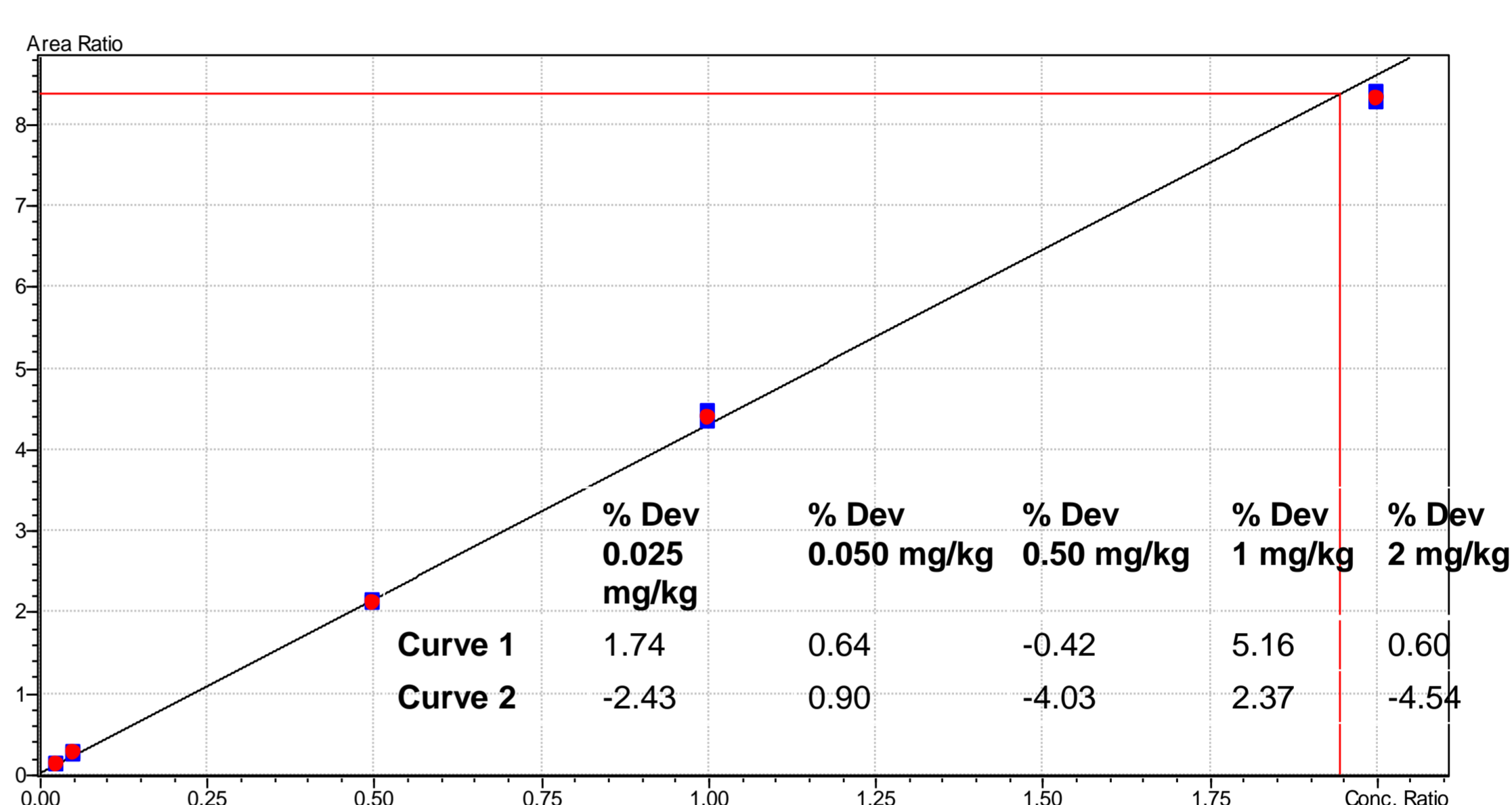


Figure 2. Linearity of procedural calibration curve (spiked to blank manure fertilizer). Quantification MRM reaction m/z 392.00 → 88.00.

Limit of quantification

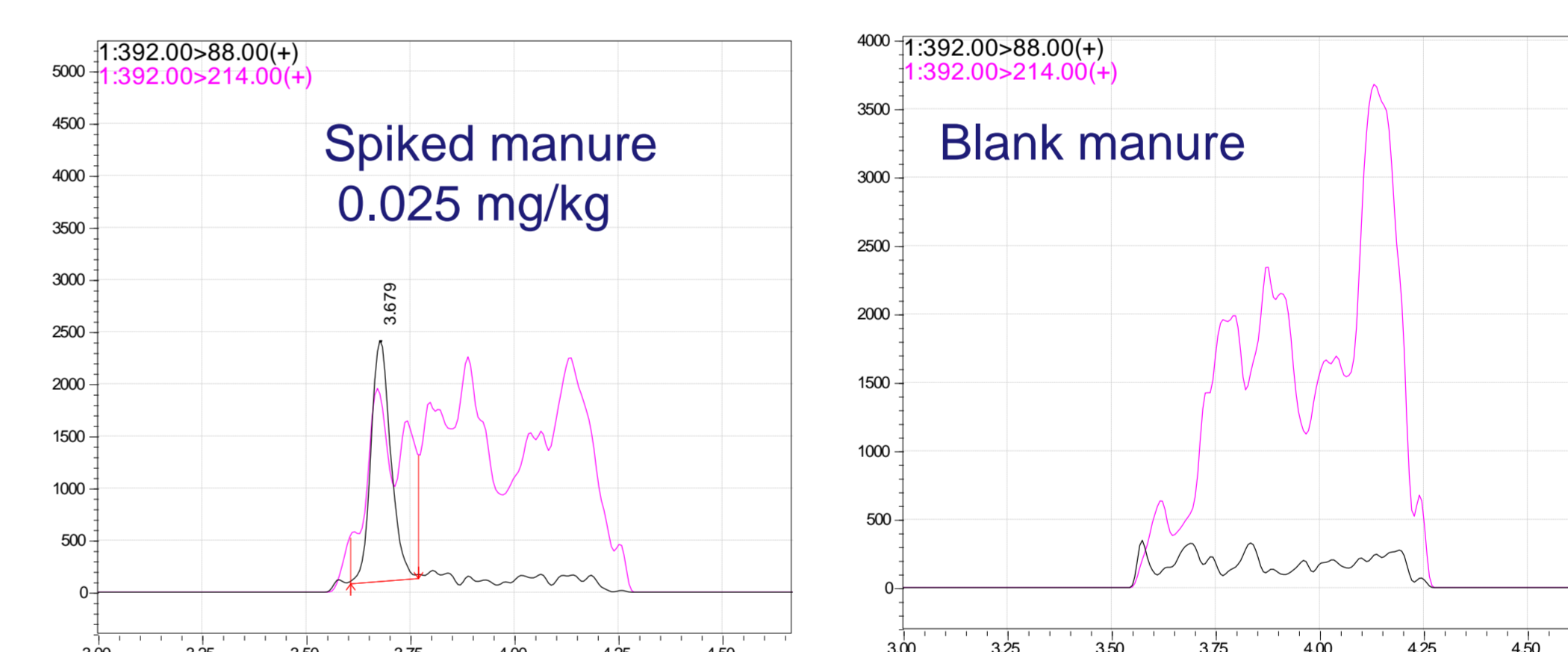


Figure 3. Chromatogram for the spiked sample at lowest calibration level 0.025 mg/kg and for the corresponding blank sample.

Application to fertilizer samples

Seven different fertilizer products containing manure as a main component were analyzed. Results for five samples were: 0.729 mg/kg, 0.352 mg/kg, 0.526 mg/kg, 0.836 mg/kg and 0.412 mg/kg. Two of samples contained glyphosate < LOQ.

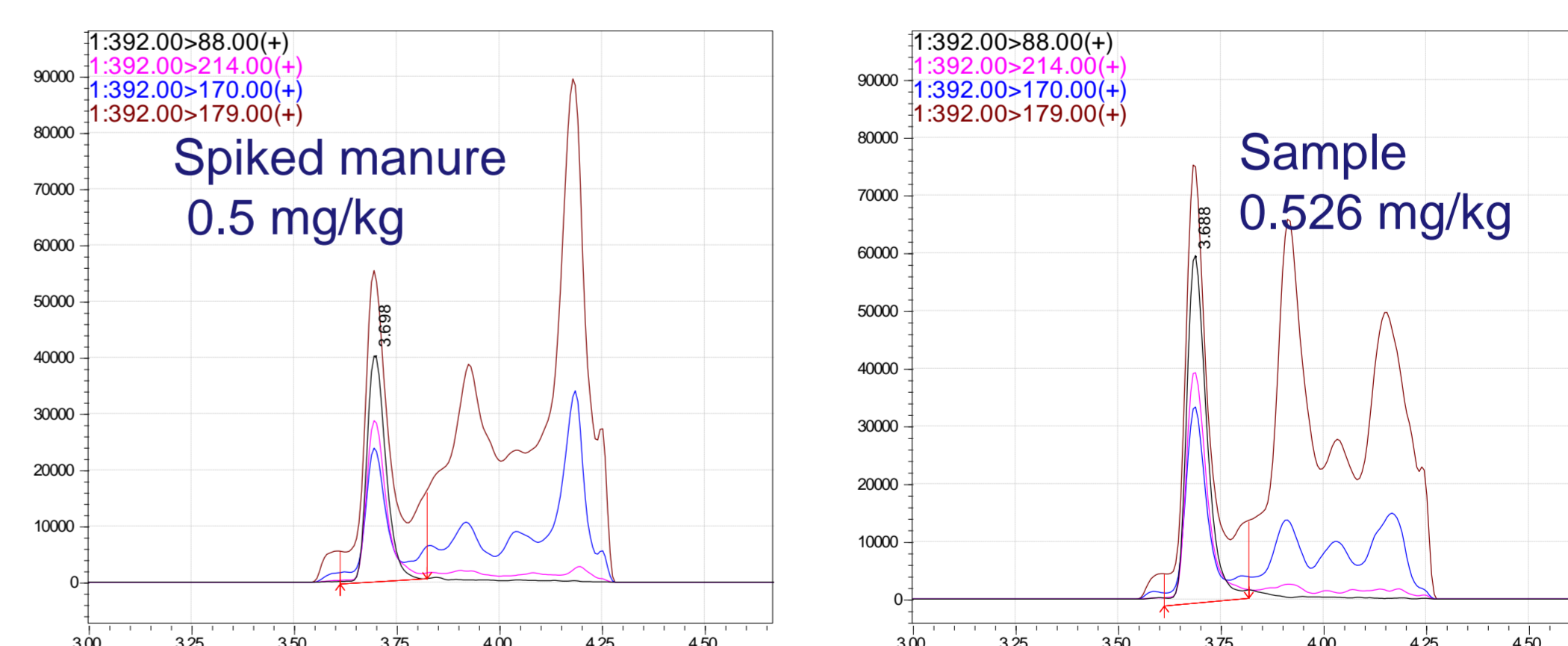


Figure 4. Chromatogram for the spiked sample at level 0.05 mg/kg and for the corresponding blank sample.

Conclusions

- The enhanced analytical method enables analysis of glyphosate residues at 0.025 mg/kg in manure fertilizer samples.
- Manure fertilizers may contain residues of glyphosate. In this work glyphosate concentrations between 0.352- 0.836 mg/kg were analyzed in fertilizer samples.

Chemistry and Toxicology Research Unit is an accredited laboratory according to ISO 17025.

Reference

¹ Benbrook, C.M, Environmental Sciences Europe (2016) 28:3