



Phomopsin A in lupin flour and lupin containing food in the Netherlands

Monique de Nijs*, Diana Pereboom-de Fauw, Theo de Rijk, Hans Mol

Introduction

Phomopsin A (PHOA) (figure 1) produced by *Diaporthe toxica*² causes liver damage in sheep. Lupinosis is reported in sheep in Australia and in the past in Europe^{4, 5}. Australia and New Zealand have a regulatory limit of 5 µg PHOA/kg product¹. To meet sustainability goals and to ensure food security, the industry aims to replace soy with locally produced lupin. Numerous food products containing lupin are available on the European market^{1, 3}.



Objective

Survey on the occurrence of phomopsin A in lupin flour and lupin containing foods using an LC-MS/MS method.

Experimental

Extraction:

An 2.5 g test portion was extracted with ACN/H₂O (80%/20%) and 1% AA. The suspension was shaken (30 min.) centrifuged (3500 RPM; 10 min; RT). The supernatant was diluted 1:1 in water, cooled (30 min.), filtered (0.45 µm PTFE filter) and cooled until analysis. PHA contamination of the samples was determined by using the external standard method based on average matrix matched standard measurements before and after samples.

LC-MS/MS analysis: 20 µL was injected on a RP HPLC column. Mobile phases A (5 mM ammonium formate buffer pH 5) and B (5 mM ammonium formate buffer in 95% methanol), changing from 90% to 0% and 10% to 100% resp. over 8 min., flow rate of 400 µL/min. The Shimadzu HPLC system was coupled with a mass spectrometer (Applied Biosystems, MDS AB Sciex API Q-trap 5500) for detection. Two transitions of PHOA were measured (m/z 789.2>226.1; m/z 789.2>323.1) with the first one mentioned as the quantifier.

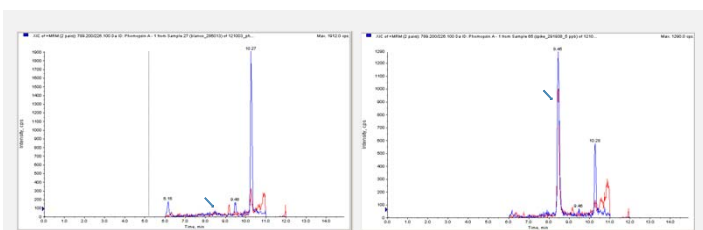
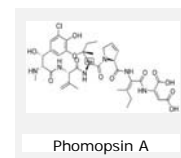


Figure 1: Combined chromatograms of a blank sample 286013 (left) and a blank sample 286013 spiked at a level of 5 µg PHOA/kg (right) for the quantifier 789.2>226.1 (blue) and qualifier 789.2>323.1 (red)

Samples

Sample	#
Lupin flour	5
Flour for bread	1
Bread	5
Biscuit/cakes	4
Frozen lupin flakes	3
Lupin snacks	3
Dry pasta products	2
Lupin flour for bakery	5
Netherlands lupin	11
Lupin for animal feed	3



Results

The limit of detection (LOD) of PHOA in lupin flour was 1.2 µg/kg, with a limit of quantification of 5 µg/kg. Results of analytical quality controls were RSD of 9% and an average recovery of 79%.

No phomopsin A was detected above LOD in any of the samples.

Conclusions

Lupin is used in a broad range of food products available in retail shops in the Netherlands. The LC-MS/MS method performs well for the determination of phomopsin A in these products.

No phomopsin A was detected above LOD in the samples analysed in this survey.

Acknowledgements

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References

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