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Analysis of 12 mycotoxins in calves' milk replacer by means of UHPLC-MS/MS

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In Belgium, veal calves are predominantly fed liquid milk replacers, based on powder milk and vegetable protein sources such as soy, corn and wheat. These ingredients may imply a risk of mycotoxin contamination. The aim of this study was to develop a multi-mycotoxin ultra-high pressure liquid chromatographic-tandem mass spectrometric (UHPLC-MS/MS) method for the detection, quantification and identification of 12 mycotoxins in milk replacer. The mycotoxins included in this study were aflatoxins (AFB_1 and B_2), alternariol (AOH), alternariol monomethyl ether (AME), deoxynivalenol (DON), 3-acetyl-DON (3-Ac-DON), 15-acetyl-DON (15-Ac-DON), fumonisins (FB_1 and B_2), ochratoxin A (OTA), T-2 toxin (T-2) and zearalenone (ZEN). $^{13}C_{15}$-DON, $^{13}C_{17}$-3-AcDON, $^{13}C_{24}$-T-2 and $^{13}C_{18}$-ZEN were used as internal standards.

Gradient chromatographic separation was performed on an Acquity™ UHPLC (Waters) system equipped with an Acquity CSH Fluoro Phenyl column (1.7 µm, 2.1 x 150 mm, Waters). The mobile phase consisted of a mixture of H_2O + 0.3% acetic acid (A) and methanol (MeOH) + 0.3% acetic acid (B). Detection was performed with a Xevo TQ-S MS triple quadrupole system (Waters). The extraction procedure consisted of a first extraction step with MeOH followed by a second extraction step with acetonitrile/H_2O/acetic acid (79/20/1). Specificity, linearity (R^2), apparent recovery (R_A), repeatability (RSD_r), reproducibility (RSD_R) , limit of detection (LOD) and quantification (LOQ) were determined for the method developed. For the calculations of R_A, RSD_r and RSD_R two concentration levels were considered.

For all mycotoxins considered, the criteria of linearity ($R^2 \geq 0.99$) and specificity were fulfilled. The average R_A (over the two concentration levels) varied between 85% and 107%, which is in agreement with the ranges stipulated in Commission Decision 2002/657/EC. Average RSD_r and RSD_R ranged between 0.9 and 10.5% and between 1.5 and 12.8%, respectively. The LOD values were within the range of 1-89 µg/kg and 2-171 µg/kg , respectively.
The developed method was used to determine the degree of mycotoxin contamination in different samples of calves' milk replacer (Antonissen *et al.*, 2014).

References

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