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Characterization of 27 mycotoxin detoxifiers and the relation with *in vitro* zearalenone binding

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Introduction and Aims

Zearalenone is a *Fusarium* mycotoxin which exerts estrogenic properties leading to reduced reproductive capabilities of several animal species. To alleviate the effect of this and other mycotoxins, mycotoxin detoxifiers are often mixed in animal feed. These detoxifiers bind or modify the mycotoxin in the intestinal tract leading to a reduced bioavailability of mycotoxins. Commercially available detoxifiers can be classified in organic and anorganic detoxifiers, of which smectites are the largest subgroup within the anorganic detoxifiers. However, most of them are not well characterized and their mycotoxin binding capacity can vary significantly according to literature.

The aims of present study were to: 1/ Characterize the physico-chemical properties of 27 different mycotoxin detoxifiers
2/ Correlate these characteristics with *in vitro* zearalenone binding capacities

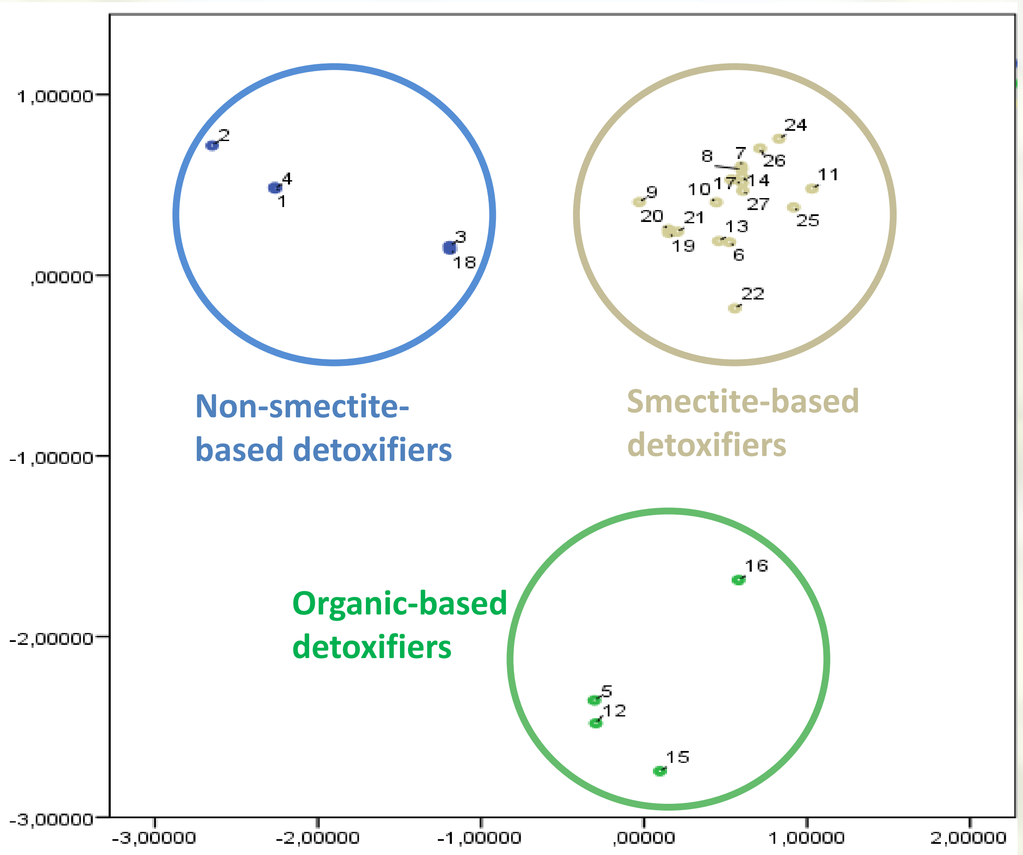

Figure 1 (↑): The XRD- and mineral fraction data were analyzed with principal component analysis and explorative cluster analysis which enabled the identification of three distinct groups: smectite based (n=18), non-smectite based (n=5) and organic based detoxifiers (n=4).

Materials and methods

1/ Characterization of the mycotoxin detoxifiers
Twenty-seven commercially available mycotoxin detoxifiers from Belgium and the Netherlands were characterized. This comprised X-ray diffraction (XRD)-profiling, cation exchange capacity (CEC) and exchangeable cation (Ca^{2+}, Mg^{2+}, Na^{+} and K^{+}) analysis, mineral fraction, moisture content, pH and swelling volume.

2/ *In vitro* binding of zearalenone
A similar protocol as Sabater-Vilar et al. (2007) was used. In brief, 20 mg of binder was shaken for 4 hours in 5 mL PBS-buffer (pH 2.5, 6.5 and 8). The buffer contained zearalenone in a concentration of 200 ng/mL. Zearalenone content in the supernatans was determined according to De Baere et al. (2012) with a modified sample clean-up procedure. The % binding was calculated as follows: (free concentration of zearalenone – total concentration)/total concentration * 100%.

Results

Figure 1 displays the segmentation of the additives based on the XRD-analysis and mineral fraction, the zearalenone binding is depicted in figure 2. The zearalenone binding of the smectite containing group was related to the physico-chemical properties using a multivariate linear regression model. The retained multivariate linear model included exchangeable K^{+} ($p \leq 0.05$), moisture content ($p \leq 0.05$) and mineral fraction ($0.05 \leq p \leq 0.1$), indicating a statistical relation of these parameters with the *in vitro* zearalenone binding (figure 3).

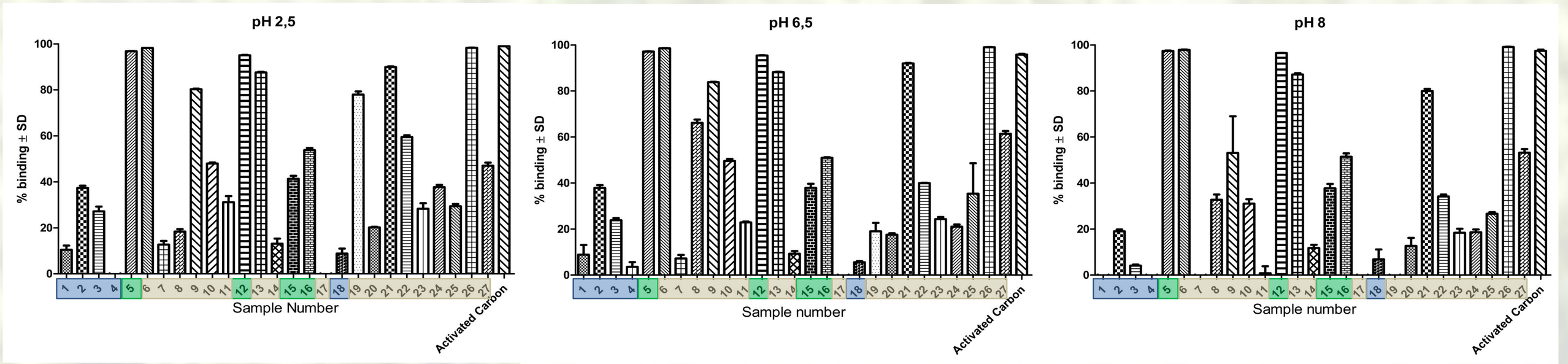

Figure 2 (↑): *In vitro* binding of zearalenone to 27 commercially available mycotoxin detoxifiers. The % binding was calculated as follows: (free concentration of zearalenone – total concentration)/total concentration * 100%.

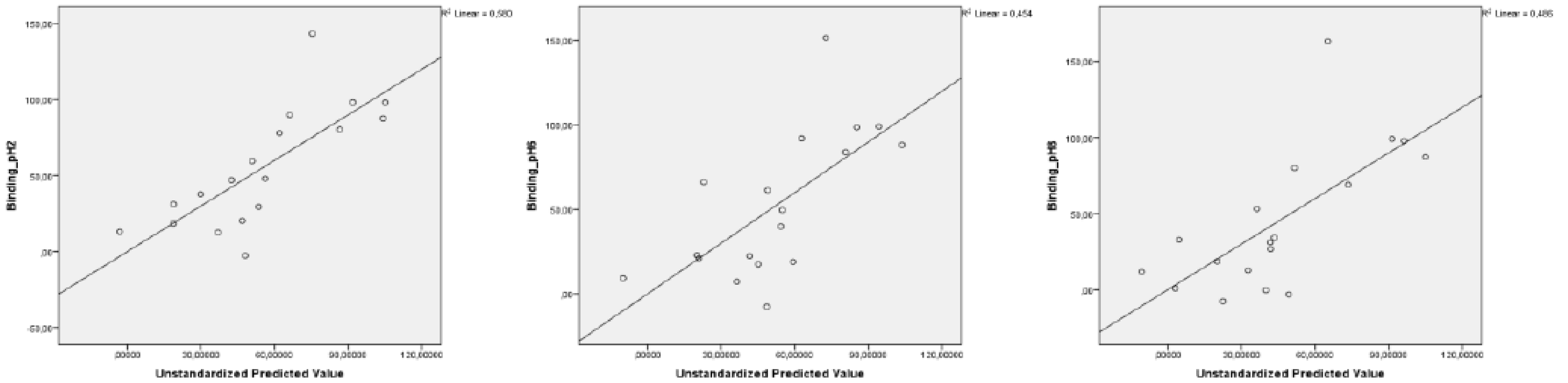

Figure 3 (↑): Observed binding vs. predicted binding of zearalenone according to the selected model, for pH 2.5, 6.5 and 8. This model includes exchangeable potassium, moisture content and mineral content. All the included parameters are fitted with a negative coefficient indicating an inverse correlation.

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Acknowledgements
The financial support of the Federal Public Service Public Health and Food chain safety is gratefully acknowledged (RF 11/6255)

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