

Probiotic and prebiotics: impact on bioavailability of aflatoxin B₁ in milk

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Introduction

Milk represents one of the most important dietary sources of nutrients for humans due to its rich nutritional composition. However, milk consumption is also potentially related to the ingestion of chemical contaminants such as mycotoxins. Aflatoxin B₁ (AFB₁) is a highly toxic mycotoxin, for both animals and humans. This toxin is frequently detected in milk, and it results from the ingestion of contaminated feed by producing animals. Given its high toxicity, methods of AFB₁ decontamination, including the use of probiotic microorganisms, have been tested in milk. But the total amount of ingested contaminant does not always reflect the amount available for absorption (bioavailability), so *in vitro* digestion models are used to determine human oral bioavailability of food contaminants. Regarding prebiotics, few studies have evaluated the decontamination of action of these compounds and it is suggested further that the probiotics to be synergistic in the removal of mycotoxins.

Materials and methods

Milk samples spiked with AFB₁ (10 µg.L⁻¹) were inoculated with *L. plantarum* and with the prebiotic compounds inulin, beta-glucan, polydextrose and oligofructose, isolated and combined, in a total of 12 different assays plus two controls (Table 1). The samples were subjected to *in vitro* digestibility (Figure 1). AFB₁ was quantified in samples by UHPLC-FLD before and after digestion.

Table 1 - Matrix planning Plackett Burman of 12 trials with 02 central points.

Test	x ₁ ^a	x ₂ ^b	x ₃ ^c	x ₄ ^d	x ₅ ^e	x ₆ ^f
1	+1	-1	+1	-1	-1	-1
2	+1	+1	-1	+1	-1	-1
3	-1	+1	+1	-1	+1	-1
4	+1	-1	+1	+1	-1	+1
5	+1	+1	-1	+1	+1	-1
6	+1	+1	+1	-1	+1	+1
7	-1	+1	+1	+1	-1	+1
8	-1	-1	+1	+1	+1	-1
9	-1	-1	-1	+1	+1	+1
10	+1	-1	-1	-1	+1	+1
11	-1	+1	-1	-1	-1	+1
12	-1	-1	-1	-1	-1	-1
13	0	0	0	0	0	0
14	0	0	0	0	0	0
CP*	0	-	-	-	-	-
CN**	-	-	-	-	-	-

^a AFB₁ concentration (µg.L⁻¹);
^b Time (h);
^c Inulin;
^d Oligofructose;
^e B-Glucan;
^f Polydextrose
 • Positive control;
 • ** Negative control

Levels:
 [AFB₁]: +1 / 0 / -1 = 10 / 7.5 / 5
 Time: +1/ 0 / -1 = 6h, 3h, 0h;
 % Prebiotics: +1/0/-1 = 0.75/0.38/0

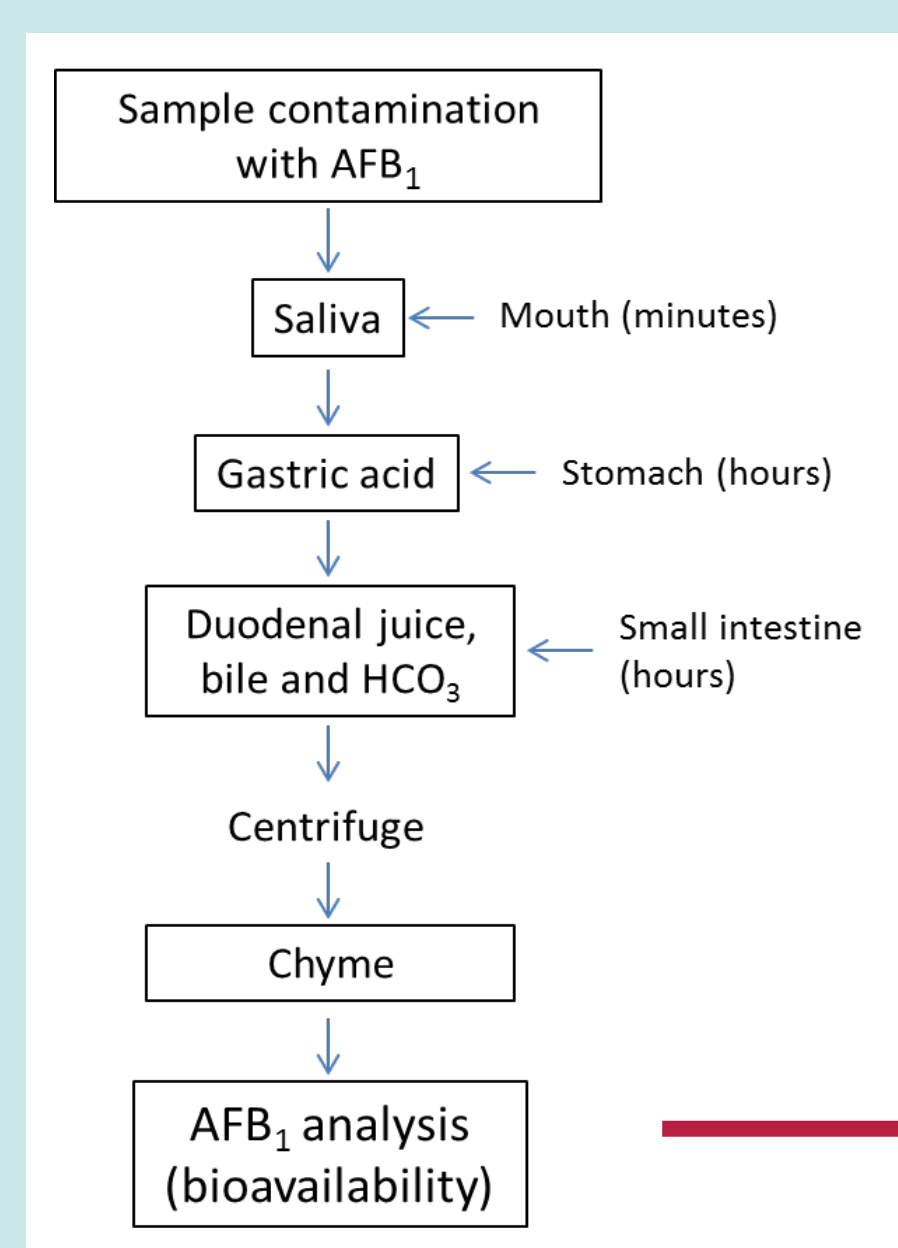
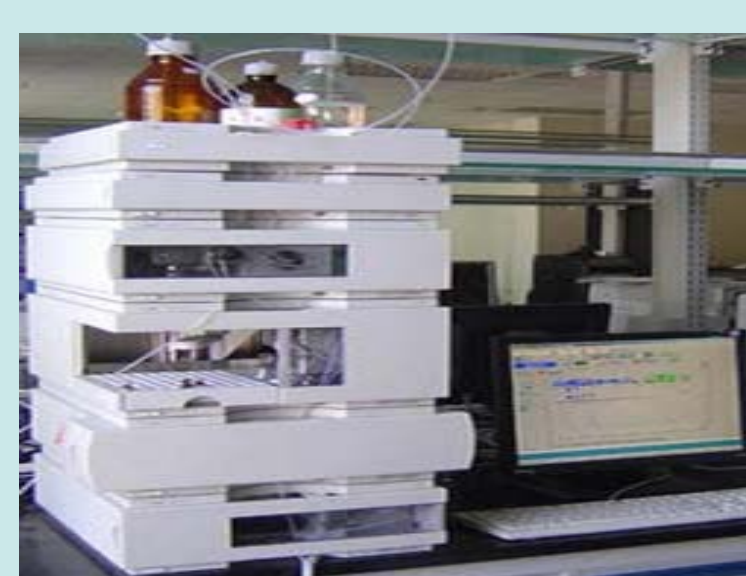


Figure 1 – Scheme of *in vitro* digestion and detection and quantification of available AFB₁ after the various treatments



Objective

The study aimed to evaluate the effect of the probiotic microorganism *Lactobacillus plantarum* BG112 and of several prebiotics (inulin, beta-glucan, polydextrose and oligofructose), alone or combined, in the bioavailability of AFB₁ in milk artificially contaminated with the mycotoxin, after digestibility in an *in vitro* digestion model.

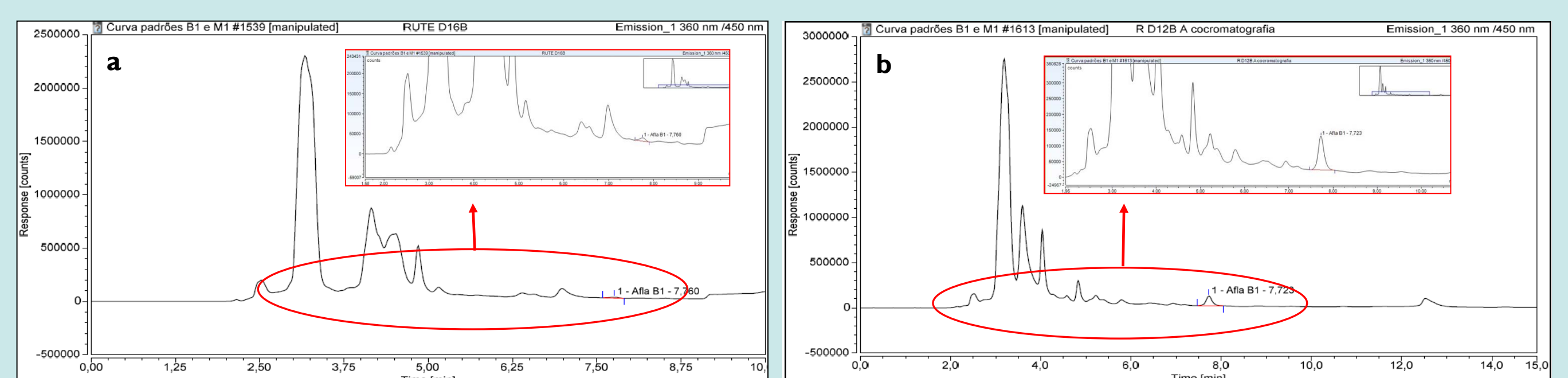
Results and Discussion

- The bioavailability of AFB₁ on milk obtained for the various probiotic and prebiotics combinations after *in vitro* digestion ranged from 15.6 to 35.6% of the spiked amount.
- The addition of *L. plantarum* BG112 alone on AFB₁ contaminated milk resulted on AFB₁ bioavailability of 27.1 ± 0.93%, a level similar to that reported by other authors, emphasizing the binding capacity of the acid lactic bacteria with AFB₁.
- Concerning prebiotics effect, bioavailability of AFB₁ varied between 15 and 51%. The best result was observed for prebiotics inulin and oligofructose when tested individually.

Table 2 – Bioavailability of AFB₁ after *in vitro* digestion with the various probiotic and prebiotics combinations.

Test	% Bioavailability
1	15,92±0,79
2	15,62±0,55
3	26,53±2,13
4	21,25±1,60
5	20,00±2,00
6	30,76±1,74
7	35,63±0,28
8	51,09±0,99
9	39,34±0,77
10	26,78±0,81
11	30,55±0,74
12	27,12±0,93
13	28,01±0,27
14	21,00±0,30
CP*	101±6,3
CN**	ND

Figure 2 - Chromatograms of milk samples fortified with AFB₁, after *in vitro* digestion: a) without detectable AFB₁; b) with AFB₁.



Conclusions

- Bioavailability of AFB₁ was reduced by the addition of *L. plantarum* and prebiotics under all tested conditions.
- *L. plantarum* can be incorporated into a food matrix in order to reduce the bioavailability of AFB₁.
- The results help explain how some natural components present in food can help modulate the intake of certain toxic compounds.