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Yeast Probiotic Isolated from Fish Fermented (Budu) with Promising AFB1 Biodetoxify

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ABSTRACT

Yeast probiotics which adsorb molecules into the cell walls of microorganisms have the benefit of degrading and preventing the negative effects of mycotoxin toxicity, especially aflatoxin B1 (AFB1). The research used five types of probiotic yeast (*Pichia kudriavzevii strain B-5P*, *Pichia kudriavzevii strain CBS 5147*, *Saccharomyces cerevisiae* (SC1), *Saccharomyces cerevisiae* (SC2), and *Saccharomyces cerevisiae* (SC3) to degrade aflatoxin B1 (AFB1). The qualitative selection used coumarin as a carbon source and using supernatants and non-viable cells and then tested their ability to degrade AFB1. The second selection was carried out quantitatively using HPLC to obtain the final result and find the best yeast probiotic for degrading AFB1 *in vitro*. The research results showed that five yeast probiotics could degrade AFB1 using coumarin as a carbon source. The next selection was to use supernatant and non-viable cells in degrading AFB1. The non-viable cells provide the highest degradation for *P. kudriavzevii strain* B-5P, *P. kudriavzevii strain CBS* 5147 and *S. cerevisiae* (SC1). Both supernatant and viable cells were almost the same in their ability to degrade AFB1 in *S. cerevisiae* (SC2), on the contrary, *S. cerevisiae* (SC3) had a higher ability in supernatant than viable cells. Gastrointestinal quantitative selection obtained that *P. kudriavzevii* CBS 5147 has the highest degradation of AFB1 was 69.35%. Meanwhile, *P. kudriavzevii B-5P* has the highest qualitative degradation of AFB1 (78.83%) and quantitative degraded AFB1 in the digestive tract including isolates treatments non-viable (81.69%), viable (89.26%), and supernatant (77.54%). Based on the research results, *P. kudriavzevii B-5P* is an attractive candidate to be selected for the biological detoxification (biodetoxification) of AFB1 *in vitro*.

Key words: In-vitro; Mycotoxin; *Pichia kudriavzevii*; Qualitative; Quantitative; *Saccharomyces cerevisiae*

INTRODUCTION

The main source of aflatoxin contamination is grain feed ingredients such as corn which is needed quite a lot as a basic ingredient in poultry rations (50-60%). The problem of contamination of the fungus which can produce AFB1 causes a decrease in the standard condition of farmer's products (Zolfaghari et al. 2020; Natarajan et al. 2022). This could be due to farmers only rely on sunlight for drying, so it takes a long time to reach a corn moisture content of 14% (dry wet). Inadequate post-harvest handling often results in feed ingredients being susceptible to fungi that cause aflatoxin. The growth of pathogenic fungi such as *Aspergillus flavus* produces aflatoxin originating from the soil and lasts from planting to harvest (Nji et al. 2023). The aflatoxin limit in feed ingredients or rations based on SNI 8926:2020 is 100pbb.

Various previous studies have reported strategies applied to remove toxigenic agents, destroy, reduce or prevent the effects of aflatoxin, or inactivate its bioavailability in contaminated corn/feed ingredients (Huang et al. 2019; Guo et al. 2021). Currently, the use of probiotic yeast is considered quite popular to eliminate mycotoxins, especially aflatoxins (Ismail et al. 2018). Yeast has two biological system mechanisms to reduce aflatoxin which are an absorption mechanism and an enzymatic mechanism. In the absorption mechanism, yeast absorbs aflatoxin onto the surface of its cells.

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Meanwhile, in the enzymatic mechanism, oxidative enzymes, carboxypeptidases, and nicotinamide adenine dinucleotide phosphate enzymes which depend on NADPH degrade mycotoxins (Zolfaghari et al. 2020).

The cell ability of yeast to reduce aflatoxin was reported by several previous studies to depend on the yeast strain (Chlebicz and Slizewska 2020; Kalita et al. 2023). Several reports indicate the effects of aflatoxin breakdown by yeast cells. Zolfaghari et al. (2020) reported that *Saccharomyces cerevisiae* isolated from dairy products in Iran had an increased aflatoxin binding capability of 30.46%. Meanwhile, Slizewska and Smulikowska (2011), in their study reported that a consortium of several microbes consisting of *Lactobacillus brevis, Lactobacillus plantarum, Lactobacillus paracasei* and *Saccharomyces cerevisiae* succeeded in reducing aflatoxin by 55%. In other studies, various types of strains such as *S. cerevisiae, Candida krusei* and *Lactobacillus sp.* which were evaluated for their potentiality to bind aflatoxin were found capable of attaching up to 60% (Shetty and Jespersen 2006). In another study by Bovo et al. (2015), *S. cerevisiae* in the citrate-phosphate buffer can remove aflatoxin by 55%. Meanwhile, the use of *Kluyveromyces lactis* and *S. cerevisiae* can decrease aflatoxin by 54 and 42% respectively, and if using a combination of the two strains it can experience a higher reduction of up to 66.6% (Hamad et al. 2017; Magnoli et al. 2024). Chlebicz and Slizewska (2020) in their experiment used six strains of *S. cerevisiae* and showed a high reduction in aflatoxin detoxification rates of 65%. There are no studies that report the use of yeast strains from Indonesia that have functioned as detoxification on aflatoxins, especially AFB1. A previous study by Marlida et al. (2021) found three local isolate yeast strains namely *S. cerevisiae* isolated from a fermented fish (Budu). Marlida et al. (2023a) found *Lactobacillus harbinensis strain 487* which has the activity of slowing the spread of pathogenic fungi and detoxifying aflatoxin B1 to 43%. Meanwhile, Marlida et al. (2024) also reported metagenomic analysis and microbial biodiversity from Budu and various potentials.

In this study, yeast *S. cerevisiae* (SC1), *S. cerevisiae* (SC2), and *S. cerevisiae* (SC3) isolated from Budu origin West Sumatra, Indonesia have the ability as a probiotic for poultry can kill pathogenic bacteria *Staphylococcus aureus, Salmonella Enteritidis* and *Escherichia coli* resistant to pH 2.5 to 5 hours and also resistant to bile at a concentration of 0.3% (Marlida et al. 2021). A previous investigation by Ardani et al. (2023) explored that two yeast from Budu had the potential as probiotics for ruminants, also called DFM (direct-fed microbial) which can increase feed digestibility and ruminal fermentation. The study reported that after identification using 16s rRNA, the isolates were *Pichia kudriavzevii strain B-5P* and *Pichia kudriavzevii strain CBS 5147*. In this research, we explored the ability of five local yeast isolated from Budu to biodetoxify mycotoxins AFB1 in feed.

MATERIALS AND METHODS

Ethical approval

This research does not require ethical approval because it was carried out *in vitro* and did not use any animals.

Study location and period

The *in vitro* study was arranged from May to October 2023 at the Laboratory of Feed Industry Technology Faculty of Animal Sciences, Andalas University, Indonesia. Observation of AFB1 degradation using HPLC was carried out at the Pharmaceutical Laboratory, Faculty of Pharmacy, Andalas University, Indonesia.

Yeast suspension preparation

The preparation of yeast suspensions followed the method of Ghofrani et al. (2018) with some modifications. This research used five probiotic yeasts, namely *Pichia kudriavzevii strain B-5P, Pichia kudriavzevii strain CBS 5147, S. cerevisiae (SC1), S. cerevisiae (SC2)* and *S. cerevisiae (SC3).* Yeast mold Broth (YMB) culture media was used to activate the five yeasts at 25°C; 24h. The cell culture was centrifuged at 3000rpm; 10min and then the supernatant was dispensed. Yeast cells were washed twice using a Phosphate Buffer Saline (PBS) solution. At last, using a PBS solution (pH 7.2), the opacity was measured with a spectrophotometer at 600nm and an absorbance of 1.170 corresponds to 2×10^8 cells/mL of yeast cells.

Aflatoxin B1 stock solution preparation

In this study, Aflatoxin B1 stock solution was prepared using a method modified by Zinedine et al. (2005). AFB1 powder (Sigma, NY, USA) containing 1 g of venom powder was suspended in benzene-acetonitrile with a ratio of 97:3 (v/v). PBS (pH 7.2) was prepared and benzene-acetonitrile was separated by rotary evaporator for 10min at 80°C and the final concentration of AFB1 (10ppm) was made by dilution with PBS. Then the stock solution was stored at 4°C in an amber glass until used.

Qualitative selection of yeast probiotics in degrading AFB1

According to Zhang et al. (2020), coumarin can be used to measure the ability of yeast probiotic strains to reduce AFB1. Each yeast probiotic was incubated for 24h at 37°C in YMB broth. Cultures then inoculated (5%; v/v) in 50mL of YMB broth and incubated for 48h at 37°C. Next, each inoculum was measured in OD600 and adjusted to 2.0 so that each strain had the same number of cells. Then the medium containing 15% coumarin was inoculated with probiotic yeast inoculum and incubated for 24h at 28ºC (150rpm). YMB media was used with coumarin as a control. After incubation, turbidity was measured using a spectrophotometer with a wavelength of 600nm.

Digestive fluids (gastric juices and intestinal fluids) preparation

Digestive fluid preparations consist of gastric juice and intestinal fluid. The simulated gastric fluid suspension contained 6.2g/L NaCl, 2.2g/L KCl, 1.2g/L NaHCO3, 0.22g/L CaCl2 and 0.3% pepsin. Meanwhile, the simulated intestinal fluid suspension contains 5g/L NaCl, 0.6g/L KCl, 0.45% bile salts, 0.3g/L CaCl2, and 0.1% pancreatin. The pH of the gastric fluid was shortened to 2.5 by adding 0.1N HCl and the pH of the intestinal fluid was increased to 7.5 by adding 0.1N NaOH. Both suspensions were filtrated with a size of 0.22µM (de Palencia et al. 2008).

Sample culture in gastrointestinal simulation

A total of 1mL of culture of each strain (10^{10} CFU/mL) was added on 9mL of gastric fluid (pH 2.5; pepsin/HCl) infected with 10ppb of AFB1 toxin to simulate gastric conditions. Then vortex for 15 seconds and incubate for 120min at 37°C. Next, 1mL of simulated gastric fluid containing the culture of all strains and AFB1 toxin was added to 9mL of simulated intestinal fluid (pH 7.5; pancreatin/bile salts). Simulated intestinal fluid was incubated for 120min at 37°C. After the incubation period, the sample was transferred to a microtube for centrifugation for 15min at 7500rpm. Then the yeast sample was precipitated. The resulting supernatant was discarded and the centrifugation step was repeated to separate all the yeast and the supernatant was discarded again. After that, the quantity of AFB1 residual in the test solution was determined using HPLC and compared with the quantity of AFB1 obtainable in the control solution. The control contained the same quantity of AFB1 but no yeast. The reduction of AFB1 quantity in the test solution compared to the control indicated the yeast's capability to absorb toxins and excrete them.

Analysis statistically

Experimental data were examined using analysis of variance (ANOVA). Data with differently significant results (P<0.05) was continued by the Duncan multiple range test (DMRT).

RESULTS

Subsection qualitative selection of yeast probiotics in degrading AFB1

The selection of yeast probiotics in degrading AFB1 can be tested using coumarin. Coumarin can be used as a carbon source by yeast which will have an impact on increasing yeast growth. The increase in yeast growth indicates that yeast can degrade AFB1 because the structure of coumarin resembles that of AFB1. Table 1 shows a qualitative selection of 5 probiotic yeast strains.

Table 1: Degradation AFB1 using coumarin with probiotic yeast

No.	Probiotic Yeast	AFB1 Degradation (%)
1.	*Pichia kudriavzevii* strain B-5P	78.83
2.	*Pichia kudriavzevii* strain CBS 5147	74.15
3.	*Saccharomyces cerevisiae* (SC1)	58.78
4.	*Saccharomyces cerevisiae* (SC2)	61.62
5.	*Saccharomyces cerevisiae* (SC3)	59.72

The highest degradation of AFB1 based on coumarin use was found in *P. kudriavzevii strain B-5P*, which was 78.83%, followed by *P. kudriavzevii strain CBS 5147*, *S. cerevisiae* (SC2), *S. cerevisiae* (SC3), and *S. cerevisiae* (SC1), are 74.15; 61.62; 59.72; and 58.78%, respectively (Table 1).

Qualitative selection of probiotic yeast in degrading AFB1 using non-viable cells and supernatant

Several secondary metabolite compounds play a role in selecting the ability of probiotic yeast to degrade AFB1, namely teichoic acid and AFB1 degrading enzymes. Two main mechanisms in the yeast biological system have been identified to deal with aflatoxins, namely absorption and enzymatic mechanisms. Aflatoxin is absorbed onto the surface of probiotic yeast. Meanwhile, in the enzymatic mechanism, the breaking up of mycotoxins by oxidative enzymes, carboxypeptidase A, and nicotinamide adenine dinucleotide phosphate (NADPH) is based on enzymes (Zolfaghari et al. 2020). Uptake by yeast cells is seen as viable cells (live yeast cells) or non-viable cells (live yeast cells).

Fig. 1 shows the ability of probiotic yeast in the form of supernatant and non-viable (dead cells) in degrading AFB1. Each yeast probiotic strain appears to have different abilities using both supernatant and non-viable cells (dead cells) (Fig. 1). Isolates *P. kudriavzevii strain B-5P, P. kudriavzevii strain CBS 5147* and *S. cerevisiae (SC1)* showed high AfB1 degradation using non-viable cells rather than supernatant, 71.07; 71.73 and 73.42%, respectively used non-viable cells and 69.42; 71.48 and 69.11%, respectively used supernatant. Meanwhile, isolates *S. cerevisiae (SC2)* and *S. cerevisiae (SC3)* degraded AFB1 in the supernatant higher than non-viable cells, 70.99 and 75.33%, respectively when used supernatant and 70.65%; 70.14%, respectively when used non-viable cells. The best results in degrading AFB1 were using supernatant from *S. cerevisiae (SC3)* isolate.

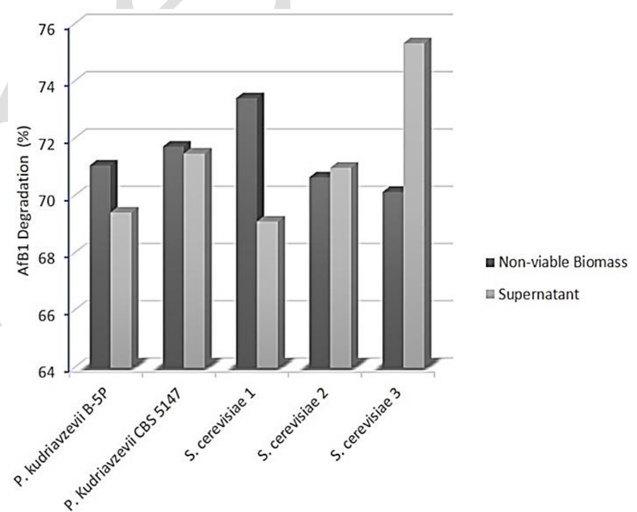

Fig. 1: Probiotic yeast in the form of non-viable cells (dead cells) and supernatant in degrading AFB1

Quantitative selection of yeast probiotics to degrade AFB1

The results of the research at this stage used the Elisa method which was modified using HPLC, where AFB1 as a control contained 200ppb of standard aflatoxin and a mixture of probiotic yeast cultures in a gastrointestinal simulation (i.e. mimicking the gastric and intestinal fluids of poultry) after which the aflatoxin content of the standard and sample was measured, which contains probiotic yeast cultures in the form of non-viable cells (yeast cells that have been killed). The research results are shown in Table 2.

Degrade AFB1 by yeast probiotics in digestive tract

The results of the research which were analyzed statistically showed that there were very significant differences (P<0.01) in the interaction between the type of treatment given and the type of isolate used, and in each single factor, there were also very significant differences (P<0.01), namely the treatment given and the type of isolate

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Table 2: Aflatoxin B1 degradation by non-viable biomassa of yeast isolates in gastrointestinal tract

No.	Probiotic Yeast	Gastrointestinal degrading AFB1 (%)
1.	*Pichia kudriavzevii* strain B-5P	67.97
2.	*Pichia kudriavzevii* strain CBS 5147	69.35
3.	*Saccharomyces cerevisiae* (SC1)	68.74
4.	*Saccharomyces cerevisiae* (SC2)	66.36
5.	*Saccharomyces cerevisiae* (SC3)	68.64

Table 3: Degrade AFB1 by Yeast Probiotics in Digestive Tract

Isolates	Isolates					Average
Treatment	*P. kudriavzevii* B-5P	*P. kudriavzevii CBS 5147*	*S. cerevisiae* (SC1)	*S. cerevisiae* (SC2)	*S. cerevisiae* (SC2)	
Non-viable	81.69^{B}	78.25^{C}	76.76^{E}	75.97^{F}	74.47^{H}	77.43^{A}
Viable	89.26^{A}	77.35^{D}	74.86^{G}	68.95^{J}	64.27^{M}	74.94^{B}
Supernatant	77.54^{D}	70.50^{I}	77.99^{C}	67.40^{K}	65.57^{L}	71.80^{C}
Average	82.83^{A}	75.36^{C}	76.54^{B}	70.77^{D}	68.10^{E}	

Different superscripts between treatments for each column and row indicated statically highly significant differences (P<0.01).

used to reduce the percentage of aflatoxin in the digestive tract (Table 3). Based on DMRT tests on the interaction of the two factors, it was found that the viable cell treatment from *P. kudriavzevii* B-5P had the highest value in reducing AFB1 in the digestive tract, the non-viable, viable, and supernatant cell of P. kudriavzevii B-5P showed highly significantly differences (P<0.01) results compared to other treatments.

DISCUSSION

Based on this research, the probiotic yeast *P. kudriavzevii* has a higher degradation ability compared to the probiotic yeast *S. cerevisiae*. This is thought to be because the aflatoxin reduction ability of yeast cells depends on the type of yeast strain. These impacts are consistent with previous research by Zolfaghari et al. (2020) who declared that applying different strains can produce different abilities. The *S. cerevisiae* culture isolated from yoghurt has an increased aflatoxin binding ability of 30.46% (Zolfaghari et al. 2020; Ismael et al. 2022; Coniglio et al. 2023), the consortium of *S. cerevisiae* LOCK 0140 and several strains of *Lactobacillus sp.* reduced aflatoxin until 55% (Slizewska and Smulikowska 2011). In another study, six strains of *S. cerevisiae* also showed upper levels of detoxification with an average reduction of 65% (Chlebicz and Slizewska 2020). The ability of *P. kudriavzevii strain B-5P*, *P. kudriavzevii strain CBS 5147* and *S. cerevisiae* (SC2), both supernatant and viable cells, is almost the same in degrading AFB1, but different things were obtained in *S. cerevisiae* (SC1) and . *S. cerevisiae* (SC3), where *S. cerevisiae* (SC1) has a high ability on non-viable cells (dead cells) while *S.cerevisiae* (SC3), on the contrary, has a higher ability on supernatant than viable cells (dead cells).

Previous studies reported that microbial adsorption of AFB1 plays an crucial role in decreasing or minimizing its bioavailability and toxic effects in mycotoxin-contaminated feedstuffs (Slizewska and Smulikowska 2011; Saleemi et al. 2019; Guo et al. 2021). Lactic acid bacteria (LAB) have been proven to be able to bind mycotoxins by using cell wall element such as peptidoglycan and polysaccharides to carry out this binding (Sadiq et al. 2019; Marlida et al. 2023a). Meanwhile, LAB strains are generally recognized as safe or GRAS, making them promising candidates for use as biopreservative agents in foodstuffs and animal feed (Zuo et al. 2013). The use of LAB isolates from Budu was previously studied by Susalam et al. (2024) as a probiotic in improving meat quality and broiler performance. Meanwhile, the use of yeast can detoxify mycotoxins in various ways, including inhibiting mycotoxin production, biodegradation or bioabsorption (Ndiaye et al. 2022).

Enzymes derived from yeast itself can be used as biodegradation agents (Huang et al. 2019). Cao et al. (2010) revealed that the fungus *Armillariella tabescens* can degrade aflatoxin B1 through oxidase activity. The ability of AFB1 degradation via the oxidase enzyme can be proven by high-performance thin-layer chromatography (HPTLC) with the bis-furan ring cleavage mechanism of the aflatoxin molecule. Other studies report that the toxicity of degraded compounds is very crucial because they can be more or less toxic than their parent compounds. However, various investigations have tested cytotoxicity. Investigation from Adebo et al. (2016) shows the toxicity resulting from AFB1 degradation by *Sporosarcina sp.* and *Staphylococcus warningeri*. The investigation was carried out by observing the death of lymphocyte cells (from human blood) that have been exposed to degraded compounds.

The process of yeast cell walls forming complexes with several mycotoxins does not result in a lack of bioavailability of definite nutrients and does not affect the environment. Other research consistently shows that *S. cerevisiae* cell walls can be added on contaminated feed and the results can selectively bind mycotoxins (Yiannikouris et al. 2004b). They declare that the complex bond between toxin-yeast can pass through the gastrointestinal tract of livestock without bad effects or resulting in residues in animal products such as meat, milk or eggs (Yiannikouris et al. 2004b). In vitro studies using *L. plantarum and S. cerevisiae* supplemented to fermented rice straw-based ratios can improve nutrient digestibility (Marlida et al. 2023b; Fathanah et al. 2024).

The biological system that uses yeast and bacteria to overcome AFB1 has two main mechanisms, namely absorption and enzymatic mechanisms (Khadivi et al. 2020; Sun et al. 2023). In its absorption mechanism, yeast absorbs aflatoxin into the outside layer of its cells (Zolfaghari et al. 2020). The AFB1 compound is trapped by the β-D-glucan component in the yeast cell wall. The AFB1 compound will be confined in a single helical chain (1→3)-β-Dglucan and a branched chain (1→6)-β-D-glucan, thus being able to keep the toxic compound in a helical structure trap (Yiannikouris et al. 2004a; 2006).

Meanwhile, in the enzymatic mechanism, the mycotoxin degradation process is carried out by two different enzymes. First, 17-hydroxy-steroid dehydrogenase converts AFB1 into aflatoxin by adding a hydroxyl group to the double bond of the dihydrofuran. The final results of this process are excreted through faeces and urine (Liu et al. 2021). The second way is oxidative enzymes such as carboxypeptidase-A. This enzyme plays a role in breaking the bis-furan ring bonds of AFB1 and β esters into final products that can be broken up such as aflatoxin, aflatoxin AFD1, AFD2, AFD1, AFO, AFB2 and B2. The microtoxin compound AFB1 is absorbed onto the surface of yeast and probiotic bacteria (Hamad et al. 2017; Zolfaghari et al. 2020).

Conclusion

Based on the research results, *Pichia kudriavzevii strain B-5P* is an interesting candidate to be selected as an *in vitro* AFB1 biodetoxifier in terms of various conditions such as qualitative highest degradation of AFB1 and quantitative degraded AFB1 in the digestive tract. However, in the future, *in vivo* research is needed to strengthen the findings of this study.

Conflict of interest: The authors declare that they do not have competing interest

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Author's Contribution: Yetti Marlida, Harnentis, and Nurul Huda arranged the experimental design and wrote the original manuscript Yetti Marlida and Lili Anggraini conducted the experiment in the laboratory. Lili Anggraini and Laily Rinda Ardani analyzed the data and finalized the draft. The final version of the manuscript was revised and approved by all authors.

REFERENCES

Adebo OA, Njobeh PB and Mavumengwana V, 2016. Degradation and detoxification of AFB1 by Staphylocococcus warneri, Sporosarcina sp. and Lysinibacillus fusiformis. Food Control 68: 92–96. https://doi.org/10.1016/j.foodcont.2016.03.021

Ardani LR, Marlida Y, Zain M, Jamsari and Fassah DM, 2023. Lactic acid bacteria and yeast strains isolated from fermented fish (Budu) identified as candidate ruminant probiotics based on in vitro rumen fermentation characteristics. Veterinary World 16: 395-402. https://doi.org/10.14202/vetworld.2023.395-402

Bovo F, Franco LT, Rosim RE, Barbalho R and de Oliveira CA, 2015. In vitro ability of beer fermentation residue and yeast-based products to bind aflatoxin B1. Brazilian Journal of Microbiology 46(2): 577. http://doi.org/10.1590/S1517-838246220130400

Cao H, Liu D, Mo X, Xie C and Yao D, 2011. A fungal enzyme with the ability of aflatoxin B_1 conversion: purification and ESI-MS/MS identification. Microbiological Research 166(6): 475-483. http://doi.org/10.1016/j.micres.2010.09.002

Chlebicz A and Slizewska K, 2020. In vitro detoxification of aflatoxin B1, deoxynivalenol, fumonisins, T-2 toxin and zearalenone by probiotic bacteria from genus Lactobacillus and Saccharomyces cerevisiae yeast. Probiotics and Antimicrobial Proteins 12: 289–301. http://doi.org/10.1007/s12602-018-9512-x

Coniglio MV, Luna MJ, Provensal P, Watson S, Ortiz ME, Ludueña HR, Cavaglieri L and Magnoli AP, 2023. Use of the probiotic Saccharomyces cerevisiae var. boulardii RC009 in the rearing stage of calves. International Journal of Agriculture and Biosciences 12(3): 188-192. https://doi.org/10.47278/journal.ijab/2023.063

de Palencia PF, Lopez P, Corbi AL, Pelaez C and Requena T, 2008. Probiotic strains: survival under simulated gastrointestinal conditions, in vitro adhesion to Caco-2 cells and effect on cytokine secretion. European Food Research and Technology 227(5): 1475-1484. http://doi.org/10.1007/s00217-008-0870-6

Fathanah SYE, Rahardja DP and Purwanti S, 2024. Impacts of probiotic and dietary Lamtoro leaf meal on the growth performance, digestibility and small intestinal morphometry of Kampung chicken. International Journal of Agriculture and Biosciences 13(3): 333-339. https://doi.org/10.47278/journal.ijab/2024.127

Ghofrani TD, Kermanshahi H, Golian A and Majidzadeh HR, 2018. In vitro binding potentials of bentonite, yeast cell wall and lactic acid bacteria for aflatoxin B1 and ochratoxin A. Iranian Journal of Toxicology 12(2): 7-13. http://doi.org/10.29252/arakmu.12.2.7

Guo HW,Chang J, Wang P, Yin QQ, Liu QC, Xu XX, Dang XW, Hu XF and Wang QL, 2021. Effects of compound probiotics and aflatoxin degradation enzyme on alleviating aflatoxin induced cytotoxicity in chicken embryo primary intestinal epithelium, liver and kidney cells. AMB Express 11: 35. http://doi.org/10.1186/s13568-021-01196-7

Hamad GM, Zahran E and Hafez EE, 2017. The efficacy of bacterial and yeasts strains and their combination to bind aflatoxin B1 and B2 in artificially contaminated infants food. Journal of Food Safety 37(4): e12365. https://doi.org/10.1111/jfs.12365

Huang WW, Chang J, Wang P, Liu CQ, Yin QQ, Song AD, Gao TZ, Dang XW and Lu FS, 2019. Effect of compound probiotics and mycotoxin degradation enzymes on alleviating cytotoxicity of swine jejunal epithelial cells induced by aflatoxin B1 and zearalenone. Toxins 11: 12. https://doi.org/10.3390/toxins11010012

Ismail A, Goncalves BL, de Neeff DV, Ponzilacquab B, Coppab CFSC, Hintzschec H, Sajide M, Cruzf AG, Corassin CH and Oliveirab CHF, 2018. Aflatoxin in foodstuffs: occurrence and recent advances in decontamination. Food Research International 113: 74–85. https://doi.org/10.1016/j.foodres.2018.06.067

Ismael E, Ismail EM, Khalefa HS, Elleithy EMM, Elmosalamy SH, Marouf S and Fahmy KNE, 2022. Evaluation of Saccharomyces cerevisiae Yeast Fermentate and Xylanase in Reduced Energy Diet Fed to Broiler Chicken. International Journal of Veterinary Science 11(2): 141-150. https://doi.org/10.47278/journal.ijvs/2021.096

Kalita R, Pegu A and Baruah C, 2023. Prospects of probiotics and fish growth promoting bacteria in aquaculture: a review. International Journal of Agriculture and Biosciences 12(4): 234-244. https://doi.org/10.47278/journal.ijab/2023.070

Khadivi R, Razavilar V, Anvar SAA and Akbari-adergani, 2020. Aflatoxin M1-binding ability of selected lactic acid bacteria strains and Saccharomyces boulardii in the experimentally

In Press

contaminated milk treated with some biophysical factors. Archives of Razi Institute. 75(1): 63-73. http://doi.org/10.22092/ari.2019.123985.1265

Liu Y, Huijia M, Kalekristos WY, Zhen W, Yating C, Thierry T, Junfang L, Yixuan J, Hongyan L and Jing W, 2021. Degradation of aflatoxin B1 by a recombinant laccase from Trametes sp. C30 expressed in Saccharomyces cerevisiae: A mechanism assessment study in vitro and in vivo. Food Research International 145: 110418. https://doi.org/10.1016/j.foodres.2021.110418

Magnoli AP, Parada J, Luna MarÃa J, Corti M, Escobar FM, FernÃ¡ndez C, Coniglio MV, Ortiz ME, Wittouck P, Watson S, Cristofolini LA and Cavaglieri L, 2024. Impact of probiotic *Saccharomyces cerevisiae* var. *boulardii* RC009 alone and in combination with a phytase in broiler chickens fed with antibiotic-free diets. Agrobiological Records 16: 1-10. https://doi.org/10.47278/journal.abr/2024.006

Marlida Y, Huda N, Harnentis H, Nur YS, Lestari MN, Adzitey F and Sulaiman MR, 2021. Potential probiotic yeast isolated from an Indonesian indigenous fermented fish (Ikan Budu). Potravinarstvo Slovak Journal of Food Science 215: 460–466. https://doi.org/10.5219/1544

Marlida Y, Nurmiati N, Husmaini H, Huda N, Anggraini L and Ardani LR, 2023a. The potential of lactic acid bacteria isolated from ikan budu (fermented fish) to inhibit the growth of pathogenic fungi and detoxify aflatoxin B1. Veterinary World 16(7): 1373–1379. www.doi.org/10.14202/vetworld.2023.1373-1379

Marlida Y, Harnentis H, Nur YS and Ardani LR, 2023b. New probiotics (*Lactobacillus plantarum* and *Saccharomyces cerevisiae*) supplemented to fermented rice straw-based rations on digestibility and rumen characteristics *in vitro*. Journal of Advanced Veterinary and Animal Research 10(1): 96-102. https://doi.org/10.5455/javar.2023.j657

Marlida Y, Susalam MK, Harnentis H, Jamsari J, Huda N, Noordin WNM, Anggraini L and Ardani LR, 2024. Metagenomic analysis and biodiversity of bacteria in traditional fermented fish or Budu from West Sumatera, Indonesia. Journal of Advanced Veterinary and Animal Research 10(4): 801-808. https://doi.org/10.5455/javar.2023.j736

Natarajan S, Balachandar D, Senthil N, Velazhahan R and Paranidharan V, 2022. Volatiles of antagonistic soil yeasts inhibit growth and aflatoxin production of Aspergillus flavus. Microbiological Research 263: 127150. http://doi.org/10.1016/j.micres.2022.127150

Ndiaye S, Zhang M, Fall M, Ayessou NM, Zhang Q and Li P, 2022. Current review of mycotoxin biodegradation and bioadsorption: microorganisms, mechanisms, and main important applications. Toxins 14(11): 729. https://doi.org/10.3390/toxins14110729

Nji QN, Babalola OO and Mwanza M, 2023. Soil *Aspergillus* Species, Pathogenicity and Control Perspectives. Journal of Fungi 29(7): 766. http://doi.org/10.3390/jof9070766 .

Sadiq FA, Yan B, Tian F, Zhao J, Zhang H and Chen W, 2019. Lactic acid bacteria as antifungal and anti-mycotoxigenic agents: a comprehensive review. Comprehensive Reviews in Food Science and Food Safety 18(5): 1403-1436. http://doi.org/10.1111/1541-4337.12481

Saleemi MK, Ashraf K, Gul ST, Naseem MN, Sajid MS, Mohsin M, He C, Zubair M and Khrar A, 2019. Toxicopathological effects of feeding aflatoxins B1 in broilers and its ameliosration with indigenous mycotoxin binder. Exotoxicology and Environmental Safety 187:109712. https://doi.org/10.1016/j.ecoenv.2019.109712

Shetty PH and Jespersen L, 2006. Saccharomyces cerevisiae and lactic acid bacteria as potential mycotoxin decontaminating agents. Trends in food Science and Technology 17: 48–55. https://doi.org/10.1016/j.tifs.2005.10.004

Slizewska K and Smulikowska S, 2011. Detoxification of afiatoxin B1 and change in microflora pattern by probiotic in vitro fermentation of broiler feed. Journal of Animal and Feed Sciences 20: 300–309. https://doi.org/10.22358/jafs/66187/2011

Sun H, Ziqi He, Dongwei Xiong and Miao Long, 2023. Mechanisms by which microbial enzymes degrade four mycotoxins and application in animal production: A review. Animal Nutrition 15: 256-274. https://doi.org/10.1016/j.aninu.2023.09.003

Susalam MK, Harnentis, Marlida Y, Jamsari and Ardani LR, 2024. The effect of probiotics consortium isolated from fermented fish (Budu) on broiler performances and meat quality. International Journal of Veterinary Science 13(1): 100-107. https://doi.org/10.47278/journal.ijvs/2023.066

Yiannikouris A, Andre G, Poughon L, Francois J, Dussap CG and Jeminet G, 2006. Chemical and conformational study of the interactions involved in mycotoxin complexation with beta-D-glucans. Biomacromolecules 7(4): 1147-1155. http://doi.org/10.1021/bm050968t

Yiannikouris A, Andre G, Buleon A, Jeminet G, Canet I, François J, Bertin G and Jouany JP, 2004a. Comprehensive conformational study of key interactions involved in zearalenone complexation with beta-D-glucans. Biomacromolecules 5(6): 2176-2185. http://doi.org/10.1021/bm049775g

Yiannikouris A, Francois J, Poughon L, Dussap CG, Bertin G, Jeminet G and Jouany JP, 2004b. Alkali extraction of beta-d-glucans from Saccharomyces cerevisiae cell wall and study of their adsorptive properties toward zearalenone. Journal of Agricultural and Food Chemistry 52(11): 3666-3673. http://doi.org/10.1021/jf035127x

Zhang X, Li B, Zhang Z, Chen Y and Tian S, 2020. Antagonistic yeasts: a promising alternative to chemical fungicides for controlling postharvest decay of fruit. Journal of Fungi 6: 158. http://doi.org/10.3390/jof6030158

Zinedine A, Faid M and Benlemlih M, 2005. In vitro reduction of aflatoxin B1 by strains of lactic acid bacteria isolated from Moroccan sourdough bread. International Journal of Agriculture and Biology 7(1): 67-70. https://doi.org/1560–8530/2005/07–1–67–70

Zolfaghari H, Khezerlou A, Ehsani A and Yari KA, 2020. Detoxification of Aflatoxin B1 by probiotic yeasts and bacteria isolated from dairy products of Iran. Advanced Pharmaceutical Bulletin 10(3): 482-487. http://doi.org/10.34172/apb.2020.060

Zuo RY, Chang J, Yin QQ, Wang P, Yang YR, Wang X, Wang GQ and Zheng QH, 2013. Effect of the combined probiotics with aflatoxin B_1-degrading enzyme on aflatoxin detoxification, broiler production performance and hepatic enzyme gene expression. Food and Chemical Toxicology 59: 470-475. http://doi.org/10.1016/j.fct.2013.06.044