Biointerface Research in Applied Chemistry
Open-Access Journal (ISSN: 2069-5837)

Article
Volume 13, Issue 4, 2023, 354
https://doi.org/10.33263/BRIAC134.354

The Efficiency of *Saccharomyces Cerevisiae* as an Antifungal and Antimycotoxigenic Agent

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Scopus Author ID 36910157000
Received: 3.04.2022; Accepted: 3.06.2022; Published: 17.09.2022

Abstract: Aflatoxins (AFs) and zearalenone (ZEN) are the most predominant mycotoxins in various food and feed. Therefore, this study aimed to estimate the antifungal activity of *Saccharomyces cerevisiae* strains and to study their ability to remove aflatoxin B_1 (AFB_1) and zearalenone (ZEN) *in vitro*. Data revealed that *S. cerevisiae* NRLL Y-12633 showed higher antifungal activity compared to *S. cerevisiae* NRRL Y-1089. On the other hand, *S. cerevisiae* suspension exhibited higher antifungal activity than *S. cerevisiae* supernatant. Concerning the removal of AFB_1, results indicated that after 30 min. *S. cerevisiae* NRRL Y-1089 displayed a higher ability to remove AFB_1 at a concentration of 5.0 µg/mL with a percentage of reduction reaching 87.20% than *S. cerevisiae* NRRL Y-12633, which removed 21.00%. As for the removal of ZEN, results showed that after 30 min. *S. cerevisiae* NRRL Y-12633 successfully removed ZEN at a concentration of 5.0 µg/mL by 94.80%, whereas *S. cerevisiae* NRRL Y-1089 removed ZEN by 91.80%. Results also indicated that the removal of AFB_1 increased by increasing the incubation time, whereas the removal of ZEN decreased by increasing the incubation time. Therefore, it could be concluded that *S. cerevisiae* strains could be applied as an additive to decrease the concentration of mycotoxins in food and feed.

Keywords: *Saccharomyces cerevisiae*; AFB_1; ZEN; antifungal; adsorption.

1. Introduction

Mycotoxins are toxic secondary compounds synthesized by certain fungi that can grow on various foods under certain conditions [1]. Due to the use of contaminated raw materials, mycotoxins are usually found in grains, fruits, spices, and manufactured products [2]. Mycotoxins can also be detected in animal-derived products (e.g., milk, meat) [3]. Several types of mycotoxins have been detected, but only a few cause food safety concerns, such as aflatoxins (AFs), deoxynivalenol (DON), fumonisins (FBs), HT-2 toxin (HT2), ochratoxin A (OTA), patulin (PAT), T-2 toxin (T2), and zearalenone (ZEN) [4].

The most important mycotoxins are AFs and ZEN, which have attracted particular attention. The World Health Organization has classified AFs as carcinogenic and genotoxic [5]. When AFs are ingested, inhaled, or absorbed through the skin, even at very small concentrations, they cause acute or chronic toxic effects of carcinogenic, mutagenic, teratogenic, immunotoxic, or hepatotoxic [6]. At the same time, ZEN is known to have carcinogenic, genotoxic, teratogenic, immunotoxic, hepatotoxic, and hepatotoxic properties [7]. Aflatoxins are fairly stable compounds that can survive at relatively high temperatures, and

heating or cooking are not reliable for destroying AFs [8], whereas ZEN shows stability during storage and exposure to high temperatures, and pasteurization does not destroy it [9].

Since mycotoxins are relatively stable during cooking and processing [10, 11], new methods for preventing mycotoxin development in food and feed, as well as assessing the impact of current mycotoxin contamination, are greatly needed. To prevent the growth of mycotoxin-producing fungi and to decontaminate and/or detoxify foods and feeds, strategies have been developed [12]. These include 1) mycotoxin contamination prevention, 2) detoxification of mycotoxins in foods and feeds, and 3) prevention of mycotoxin absorption from the gastrointestinal system [13].

The use of chemical and physical detoxification strategies is not relevant in practice, owing to the possibility of producing hazardous residues or compromising the purified goods' nutritional value and organoleptic qualities [14]. Given physical and chemical decontamination limitations, biological methods using microorganisms or their enzymes are gaining attention [15]. Bio adsorption is a low-cost, environmentally beneficial approach for the verification of the safety of mycotoxin-contaminated foods [16]. This technology has been used to study the ability of some bacteria and molds to bind several mycotoxins from food and feed [17]. Therefore, the aim of this study was to estimate the antifungal activity of *Saccharomyces cerevisiae* strains and to study their ability to remove aflatoxin B_1 and zearalenone *in vitro*.

2. Materials and Methods

2.1. Mycotoxin standards.

Aflatoxin B_1 (≥98%) was obtained from Sigma-Aldrich (St Louis, MO 68178, USA), and zearalenone (≥98%) was obtained from Santa Cruz Biotechnology Inc. (Dallas, Texas 75220, USA).

2.2. Fungal cultures.

Aspergillus parasiticus, *Aspergillus niger*, *Alternaria tennis*, *Penicillium chrysogenum*, *Penicillium expansum*, and *Fusarium graminearum* were preserved in the Department of Food Toxicology and Contaminants, National Research Centre.

2.3. Saccharomyces cerevisiae cultures.

Saccharomyces cerevisiae NRRL Y-12633, and NRRL Y-1089, were kindly provided by Agriculture Research Service Culture Collection (National Center for Agriculture Utilization Research, Illinois 61604, USA)

2.4. Saccharomyces cerevisiae preparation.

The yeast tubes were wiped with alcohol (70%), opened, and lightly flamed at the open end. The yeast pellets were then poured into a tube with 0.5 mL yeast extract, peptone, malt extract, glucose (YPMG 3g yeast extract, 5g peptone, 3g malt extract, and 10g glucose), and let stand for a few minutes to allow the pellets to dissolve. Then a lapful of suspension was streaked on YPMG agar and incubated at 25°C for 2 to 4 days.

2.5. Antifungal activity.

Saccharomyces cerevisiae isolates were grown on YPMG broth at 25°C for 2-4 days. After that, the broth was divided into two parts, one part was used as suspension, and the other was centrifuged at 5000 ×g for 5 min. (Thermo Fisher Scientific, USA), and the upper layer was taken and used as supernatant. Fungal isolates were grown on Potato Dextrose Agar slants (PDA, Conda, Spain) and incubated for 7 days at 28°C. In an aqueous solution containing 0.1 % Tween 80, fungal spore suspensions (10^6 spores/mL) were prepared. One mL of each yeast culture (suspension or supernatant) for each strain was placed in a 250 mL conical flask containing 100 mL of Potato Dextrose broth (PDP, Conda, Spain) and infected with 1 mL of fungal spore suspension. Conical flasks were incubated for 7 days at 28°C. The mycelium mats were collected and filtered using Whatman filter paper No. 4, washed twice with water, and dried in an oven at 95°C until the consistent weight was achieved and weighed.

2.6. Detoxification of aflatoxin B_1 and zearalenone.

Erlenmeyer flasks 250 mL containing 100 mL of YPMG broth were inoculated with the yeast cultures at a concentration of 10^4 cells/mL and incubated at 25°C. After incubation, one mL of cell suspension was taken into Eppendorf tubes and centrifuged at 5000 ×g for 5 min. (Labnet International, Inc. USA), the supernatant was completely removed, and to remove any residual culture medium, and the yeast biomass was washed three times with Phosphate buffer saline (PBS).

Aflatoxin B_1 and zearalenone binding assay were performed by modifying the method of El-Nezami *et al*. [18] and Chlebicz and Śliżewska [19]. 980 µl of PBS (pH 7.0) and 20 µl of the mycotoxin standard containing 5.0 and 8.0 µg/mL were added to the Eppendorf and mixed thoroughly. The Eppendorfs were incubated at 25 °C with agitation at 200 rpm for 30, 60, and 120 minutes. Following the incubation period, the Eppendorfs were centrifuged for 10 min. (Labnet International, Inc. USA) at 10,000 ×g, the supernatants were filtered with PTFE syringe filters with 0.45-µm-diameter pores (Millex-GS, Millipore, USA). A solution of analyzed mycotoxin in PBS was used as a positive control, whereas yeast suspension served as a negative control. High-Performance Liquid Chromatography was used to evaluate the concentrations of mycotoxins in the samples.

2.7. The HPLC analysis.

The HPLC analysis was performed using an Agilent 1260 series (Agilent Technologies, USA). The fluorescence detector was monitored at 365 nm excitation at 435 nm emission. Using a C_{18} column (4.6mm x 250mm i.d., 5 µm), the separation was executed.

2.8. Method validations.

The calculated validation parameters were as follows: linearity, the limit of quantification (LOQ), the limit of detection (LOD), relative standard deviation (%RSD), and recovery. The linearity was assessed by the determination of the calibration curve equations and coefficients of determination (R^2). The LOQ and LOD were determined using the MS Excel "SLOPE" and STEYX functions. The repeatability of the method was calculated by the coefficient of variation (%RSD). Known concentrations of analytes (25, 50, and 100 ng/kg for AFB_1 and 5, 10, and 20ng/kg for ZEN) were used for recovery experiments.

2.9. Statistical analysis.

The SPSS statistical tool for Windows (Version 21) was used for the statistical analysis (SPSS Inc., Chicago, IL, USA). All data were statistically examined with analysis of variance (ANOVA), and the results were deemed significant at $P \leq 0.05$. Fisher's Protected Least Significant Difference was also used to compare the differences between means.

3. Results

3.1. Antifungal activity.

Figures 1 and 2 show the antifungal activity of S. cerevisiae NRRL Y-1089 and *S. cerevisiae* NRRL Y-12633 supernatant and suspension. Results revealed that *S. cerevisiae* NRRL Y-1089 suspension exhibited significant inhibition of fungal growth compared to the supernatant (Figure 1). *Saccharomyces cerevisiae* NRRL Y-1089 suspension reduced *Penicillium chrysogenum*, *Alternaria tennis*, *Penicillium expansum*, and *Aspergillus parasiticus* by 90.43%, 85.12%, 77.02%, and 69.59%, respectively. On the other hand, *S. cerevisiae* NRRL Y-1089 supernatant exhibited low antifungal activity, whereas it reduced *Fusarium graminearum*, *Alternaria tennis*, *Aspergillus niger*, and *Penicillium chrysogenum* by 41.75%, 13.56%, 8.82%, and 6.70% respectively (Figure 1). Meanwhile, *S. cerevisiae* NRRL Y-1089 supernatant did not show any antifungal activity against *Penicillium expansum* and *Aspergillus parasiticus*.

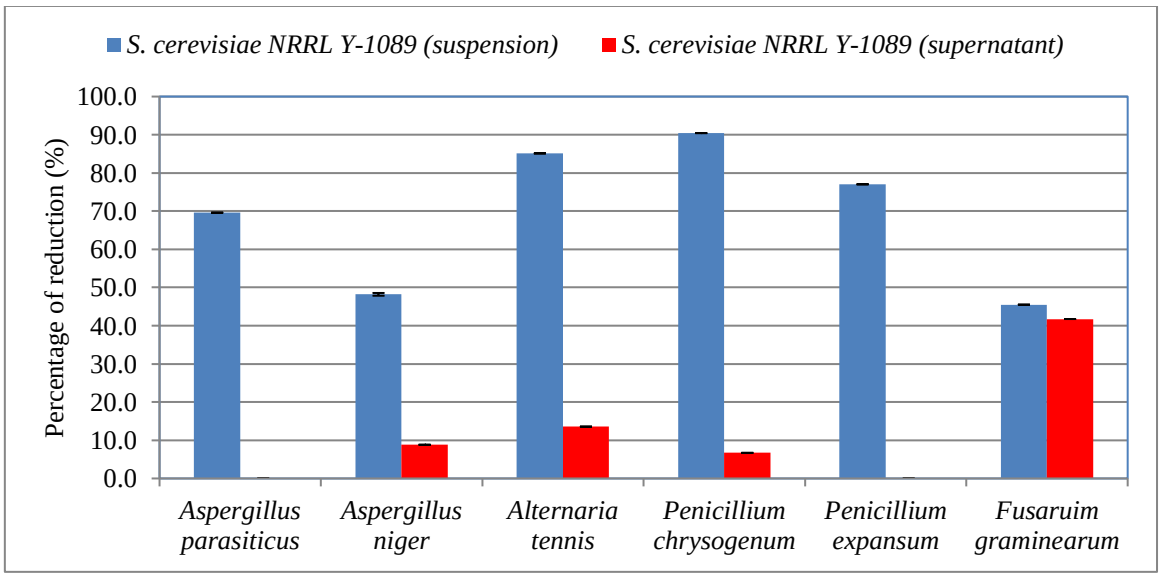

Figure 1. Antifungal activity of *S. cerevisiae* NRRL Y-1089. Results are expressed as mean ± SD. Bars represent SD. Results revealed significant differences $P \leq 0.05$ between yeast suspension and supernatant.

Data in Figure 2 showed the antifungal activity of *S. cerevisiae* NRRL Y-12633, whereas *S. cerevisiae* NRRL Y-12633 suspension caused a percentage of reduction of over 80.00% for all fungal strains. On the other hand, *S. cerevisiae* NRRL Y-12633 supernatant caused a percentage reduction of over 70.00% for all fungal species, except for *Alternaria tennis* which was inhibited by 47.23%. It could be observed that *S. cerevisiae* NRRL Y-12633 showed higher antifungal activity than *S. cerevisiae* NRRL Y-1089, and *S. cerevisiae* suspensions showed higher antifungal activity compared to *S. cerevisiae* supernatant.

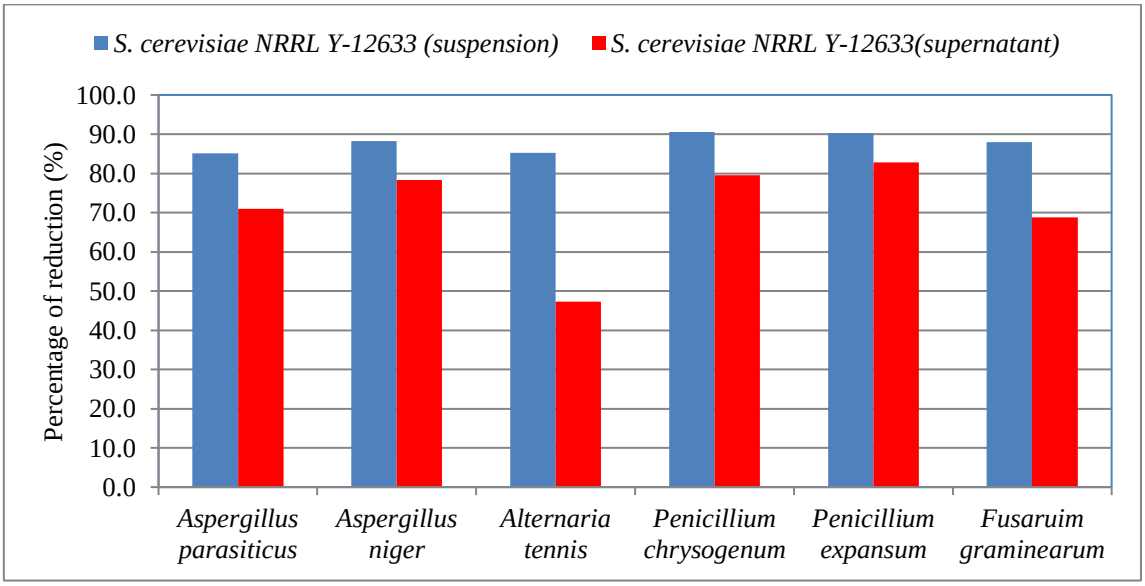

Figure 2. Antifungal activity of *S. cerevisiae* NRRL Y-12633. Results are expressed as mean ± SD. Bars represent SD**.** Results revealed significant differences $P \leq 0.05$ between yeast suspension and supernatant.

3.2. Method validations.

Good linearity was obtained for AFB_1 and ZEN with an R^2 equal to 0.999 and 0.992, respectively (Table 1). The RSD was below 1 for both mycotoxins. The mean recoveries evaluated were 104.93 and 104.11% for AFB_1 and ZEN, respectively.

Table 1. Method validation.

Mycotoxins	Calibration curve equation	R^2	LOD (ng/mL)	LOQ (ng/mL)	RSD	Recovery (%)
AFB_1	y = 3.9792x	0.999	2.71	8.22	0.43	104.93
ZEN	y = 2.9089x	0.992	2.94	8.92	0.65	104.11

LOD: Limit of detection; LOQ: Limit of quantification; RSD: Relative standard deviation

3.3. Detoxification of AFB_1.

Data in Table 2 showed the ability of both *S. cerevisiae* NRRL Y-1089 and *S. cerevisiae* NRRL Y-12633 to reduce AFB_1 in PBS during the different incubation times. Results revealed that *S. cerevisiae* NRRL Y-1089 showed a higher ability to remove AFB_1, whereas the percentage of reduction of AFB_1 at a concentration of 5.0 µg/mL reached 99.40% after 120 min. of incubation. Meanwhile, the percentage of reduction of AFB_1 at a concentration of 8.0µg/mL reached 83.05% after 120 min.

Table 2. Reduction of aflatoxin B_1 (µg/mL) by *Saccharomyces cerevisiae* at different incubation times in phosphate buffer saline.

Yeast isolates	AFB_1 concentration (µg/mL)	Incubation time (minutes)		
		30	60	120
		Concentration µg/mL (Reduction %)		
Saccharomyces cerevisiae NRRL Y-1089	5	0.64±0.25 (87.20)	0.59±0.07 (88.20)	0.03±0.00 (99.40)
	8	1.56±0.30 (80.50)	1.44±0.08 (82.00)	1.32±0.08 (83.50)
Average		1.10	1.02	0.83
Saccharomyces cerevisiae NRRL Y-12633	5	3.95±1.26 (21.00)	3.38±2.13 (32.40)	0.64±0.23 (87.20)

Yeast isolates	AFB_1 concentration (µg/mL)	Incubation time (minutes)		
		30	60	120
		Concentration µg/mL (Reduction %)		
	8	6.36±0.57 (20.50)	6.13±0.23 (23.38)	4.94±3.17 (38.25)
Average		5.16	4.76	2.79

Results are mean ± SD
Within each raw results showed no significant differences $P>0.05$
Within each column, results showed significant differences $P\leq0.05$

On the other hand, *S. cerevisiae* NRRL Y-12633 showed lower AFB_1 removal ability, whereas, after 30 min. of incubation, the percentage of reduction of AFB_1 at a concentration of 5.0 µg/mL reached 21.00%. Meanwhile, the percentage of reduction of AFB_1 at a concentration of 8.0 µg/mL reached 20.50% after 30 min. of incubation. Results showed that by increasing the incubation time, the reduction of AFB_1 increased.

3.4. Detoxification of ZEN.

Data in Table (3) showed the ability of both *S. cerevisiae* NRRL Y-1089 and *S. cerevisiae* NRRL Y-12633 to remove ZEN from PBS during the different incubation times. Results showed that *S. cerevisiae* NRRL Y-1089 reduced the concentration of ZEN by 91.80% and 94.25% after 30 min. at a concentration of 5.0 and 8.0 µg/mL, respectively. Meanwhile, *S. cerevisiae* NRRL Y-12633 reduced the concentration of ZEN by 94.80% and 95.75% after 30 min. at a concentration of 5.0 and 8.0 µg/mL, respectively. Results also revealed that by increasing the incubation time, the percentage of ZEN reduction was decreased, indicating that the binding of ZEN was fast after 30 min.

Table 3. Reduction of zearalenone (µg/mL) by *Saccharomyces cerevisiae* at different incubation times in phosphate buffer saline.

Yeast isolates	ZEN concentration (µg/mL)	Incubation time (minutes)		
		30	60	120
		Concentration µg/mL (Reduction %)		
Saccharomyces cerevisiae NRRL Y-1089	5	0.41±0.30 (91.80)	1.38±0.55 (72.40)	1.98±0.32 (60.40)
	8	0.46±0.61 (94.25)	3.19±0.42 (60.13)	3.23±1.12 (59.63)
Average		0.44	2.29	2.61
Saccharomyces cerevisiae NRRL Y-12633	5	0.26±0.007 (94.80)	0.73±0.33 (85.40)	2.83±0.21 (43.40)
	8	0.34±0.47 (95.75)	2.62±0.25 (67.25)	4.48±0.24 (44.00)
Average		0.30	1.68	3.66

Results are mean ± SD
Within each raw results showed significant differences $P\leq0.05$
Within each column, results showed no significant differences $P>0.05$

4. Discussion

Among some of the mycotoxin detoxification methods is the use of microorganisms, such as *S. cerevisiae*, that could be used for mycotoxin detoxification of food and feed [20]. European countries have stated that *S. cerevisiae* is a safe feed additive for animal feeds. Likewise, the United States For Food And Drug Administration confirmed that yeast strains are generally considered safe (GRAS)[21]. Therefore, the ability of two *S. cerevisiae* strains to lower the concentration of mycotoxins AFB_1, and ZEN in PBS solution was demonstrated *in vitro*.

Data showed that *S. cerevisiae* strains exhibited variable antifungal activity against several pathogenic fungi. Similarly, Zhu *et al*. [22] reported that among the 73 yeast strains investigated, five displayed substantial inhibitory effects on both *Aspergillus carbonaris* and *Aspergillus ochraceus,* including *Metschnikowia aff. fructicola* M179, *Pichia kluyveri* M117, and *Candida zemplinina* M3, as well as *S. cerevisiae* M114 and C297. Meanwhile, Pretscher *et al*. [23] revealed that at least one yeast species inhibited pathogenic fungi. Several studies extensively studied the applications of yeast against postharvest spoilage molds [24-27]. Recently, *S. cerevisiae* Y33 inhibited both *in vitro* and *in situ* the growth of most fungal strains [28]. In another study, the cell walls of six yeast strains were found to inhibit *A. flavus* growth [29]. It was hypothesized that a combination of numerous enzymes, such as β-1, 3- and β-1, 6-glucanase, chitinase, mannanase, or protease, performed interactively to lyse the fungal cell walls [30]. On the other hand, Bleve *et al*. [31] assumed that the competition effect might explain the antagonistic behavior of yeast against fungi for a particular growth-limiting factor, such as vitamins or another metabolite.

In vitro results demonstrated the ability of *S. cerevisiae* strains to reduce the concentration of AFB_1 in PBS solution. Data revealed that *S. cerevisiae* strains varied in their reduction of AFB_1 concentration. Although, *S. cerevisiae* NRRL Y-1089 caused an average reduction of AFB_1 by 87.20%, 88.20%, and 99.40% after 30, 60, and 120 min., respectively, *S. cerevisiae* NRRL Y-12633 caused a lower average reduction of AFB_1 by 21.00%, 32.40%, and 87.20% after 30, 60, and 120 min., respectively. The results of *S. cerevisiae* NRRL Y-1089 were considered similar to those reported by Gonçalves *et al*. [32], who stated that the mean percentages of AFB_1 bound to dried yeast ranged from 96.5 to 99.3 %. On the other hand, the results of *S. cerevisiae* NRRL Y-12633 were consistent with those obtained by Chlebicz and Śliżewska [19], who revealed that AFB_1 reduction ranged from 20 to 65% reduction. These results also indicated strain-dependent AFB_1 detoxification by *S. cerevisiae* [33, 34]. Recently, the cell walls of six yeast strains prevented aflatoxin synthesis and removed mycotoxins from contaminated buffers (at pH 3, 5, and 7) and milk [29].

Data showed that the percentage of reduction increased by increasing the incubation time. Similar observations were reported by Shetty *et al*. [35]. The ability of yeast to adsorb mycotoxins may be related to the glucomannan on the surface of yeast cell walls [36-38]. Recently, Xu *et al*. [39] reported that the adsorption of mycotoxins could be achieved via ionic bonds, hydrogen bonds, and/or hydrophobic forces with polysaccharides, proteins, lipids, and other substances on the yeast cell wall. Thus, yeast or yeast cell wall components can be used to adsorb mycotoxins in food and feed.

Data revealed that *S. cerevisiae* strains reduced ZEN concentration in PBS solution. Results showed that *S. cerevisiae* NRRL Y-1089 caused an average reduction of 91.80%, 72.40%, and 60.40% after 30, 60, and 120 min., respectively. Meanwhile, *S. cerevisiae* NRRL Y-12633 caused a higher average reduction of 94.80% and 85.40% after 30 and 60 min., respectively. Similar observations were reported by Keller *et al*. [40], who observed that a large amount of ZEN is bound to the yeast cell wall due to the higher concentration of mycotoxin. The detoxification activity of yeast strains was found to be strain-dependent, as shown by Armando *et al*. [41] and Faucet-Marquis *et al*. [42]. Results revealed that the reduction percentage was reduced by increasing the incubation time. This was similar to those reported by Chlebicz and Śliżewska [19]. *In vitro* testing of the encapsulated red yeast cells for ZEN adsorption capacity revealed that the red yeast cells had the maximum binding capacity for

ZEN by 86.67% at a concentration of 2.5 g/mL [43]. The yeast autolysates YA1 and YA2 demonstrated considerable adsorption of ZEA in the range of 49.7% to 64.7% [44].

Shang *et al*. [45] reported that yeast is a relatively stable adsorbent of ZEN, mostly by functional carbohydrates (glucomannan polymer) in the cell wall. Due to the nature of the complexes formed between yeast cell walls and certain mycotoxins, there has been some assumption that hydrophobic interactions could occur between ZEN and yeast cell wall components [46]. Yiannikouris *et al*. [47, 48] reported that the binding of *S. cerevisiae* to ZEN has been attributed to glucan components. Moreover, Yiannikouris *et al*. [49, 50] described the mechanism of binding yeast-modified glucan with ZEN, and it was suggested that non-covalent bonds are involved in the interactions between -D-glucans and ZEN, making them an "adsorption type" rather than a "binding type". In recent years, *S. cerevisiae* has been settled as a nutritional additive when added to food or feed and inhibits mycotoxin toxicity during production [51-53]. Also, because they are known to create complex sets of aroma/flavor and texture components, the genus *Saccharomyces* has recently garnered significant scientific impetus in food processing, preservation, and fortifications [54]. Despite the fact that the mechanisms responsible for yeast antagonism against mycotoxin-producing fungi and bio-detoxification are not completely understood and require further research, yeasts can be an appealing alternative detoxification option [55].

5. Conclusions

The results showed that the two strains of *S. cerevisiae* bound considerable amounts of AFB_1 and ZEN. These strains could have great possibilities to be added as an additive to food and feed to prevent mycotoxin contamination in humans and animals. Nevertheless, further research is necessary to recognize the binding mechanism and investigate the stability of the complex formed in physicochemical settings.

Funding

This research was funded by National Research Centre, grant number 12020212.

Acknowledgments

The financial support by the National Research Centre, Cairo, Egypt (Grant No. 12020212). was greatly appreciated.

Conflicts of Interest

The authors declare no conflict of interest.

References

1. Freire, L.; Sant'Ana, A.S. Modified mycotoxins: An updated review on their formation, detection, occurrence, and toxic effects. *Food Chem Toxicol* **2018**, *111*, 189-205, https://doi.org/10.1016/j.fct.2017.11.021.
2. Kępińska-Pacelik, J.; Biel, W. Alimentary risk of mycotoxins for humans and animals. *Toxins (Basel)* **2021**;*13*, 822, https://doi.org/10.3390/toxins13110822.
3. Muaz, K.; Muhammad, R.; Oliveira, C.A.F.D.; Akhtar, S.; Habibullah, S.W.A.; Park, N.S.; Balasubramaniane, B. Aflatoxin M_1 in milk and dairy products: global occurrence and potential decontamination strategies. *Toxin Rev* **2022**, *41*, 588-605, https://doi.org/10.1080/15569543.2021.1873387.
4. Yang, Y.; Li, G.; Wu, D.; *et al*. Recent advances on toxicity and determination methods of mycotoxins in foodstuffs. *Trends Food Sci Technol* **2020**, *96*, 233-252, https://doi.org/10.1016/j.tifs.2019.12.021.

5. Food and Agriculture Organization of the United Nations and World Health organization. *FAO/WHO Guide for Application of Risk Analysis Principles and Procedures during Food Safety Emergencies*. Rome; **2011**, https://apps.who.int/iris/handle/10665/44739.
6. Kowalska, A.; Walkiewicz, K.; Kozieł, P.; Muc-Wierzgoń, M. Aflatoxins: characteristics and impact on human health. *Postep Hig Med Dosw* **2017**, *71*, 315-327, https://doi.org/10.5604/01.3001.0010.3816.
7. Bai, X.; Sun, C.; Xu, J.; *et al*. Detoxification of zearalenone from corn oil by adsorption of functionalized GO systems. *Appl Surf Sci* **2018**, *430*, 198-207, https://doi.org/10.1016/j.apsusc.2017.06.055.
8. Alamu, E.O.; Gondwe, T.; Akello, J.; *et al*. Nutrient and aflatoxin contents of traditional complementary foods consumed by children of 6–24 months. *Food Sci Nutr* **2018**, *6*, 834-842, https://doi.org/10.1002/fsn3.621.
9. Rogowska, A.; Pomastowski, P.; Sagandykova, G.; Buszewski, B. Zearalenone and its metabolites: Effect on human health, metabolism and neutralisation methods. *Toxicon* **2019**, *162*, 46-56, https://doi.org/10.1016/j.toxicon.2019.03.004.
10. Karlovsky, P.; Suman, M.; Berthiller, F.; *et al*. Impact of food processing and detoxification treatments on mycotoxin contamination. *Mycotoxin Res* **2016**, *32*, 179-205, https://doi.org/10.1007/s12550-016-0257-7.
11. Alshannaq, A.; Yu, J.H. Occurrence, toxicity, and analysis of major mycotoxins in food. *Int J Environ Res Public Health* **2017**,*14*, 632, https://doi.org/10.3390/ijerph14060632.
12. Kabak, B.; Dobson, A.D.W.; Iş[idot], Var. Strategies to Prevent Mycotoxin Contamination of Food and Animal Feed: A Review. *Crit Rev Food Sci Nutr* **2006**, *46*, 593-619, https://doi.org/10.1080/10408390500436185.
13. Hathout, A.S.; Aly, S.E. Biological detoxification of mycotoxins: A review. *Ann Microbiol* **2014**, *64*, 905-919, https://doi.org/10.1007/s13213-014-0899-7.
14. Marshall, H.; Meneely, J.P.; Quinn, B.; *et al*. Novel decontamination approaches and their potential application for postharvest aflatoxin control. *Trends Food Sci Technol* **2020**, *106*, 489-496, https://doi.org/10.1016/j.tifs.2020.11.001.
15. Piotrowska, M. Microbiological decontamination of mycotoxins: Opportunities and limitations. *Toxins (Basel)* **2021**, *13*, 819, https://doi.org/10.3390/toxins13110819.
16. Pfliegler, W.P.; Pusztahelyi, T.; Pócsi, I. Mycotoxins - prevention and decontamination by yeasts. *J Basic Microbiol* **2015**, *55*, 805-818, https://doi.org/10.1002/jobm.201400833.
17. Shetty, P.H.; Jespersen, L. *Saccharomyces cerevisiae* and lactic acid bacteria as potential mycotoxin decontaminating agents. *Trends Food Sci Technol* **2006**, *17*, 48-55, https://doi.org/10.1016/j.tifs.2005.10.004.
18. El-Nezami, H.; Kankaanpaa, P.; Salminen, S.; Ahokas, J. Ability of dairy strains of lactic acid bacteria to bind a common food carcinogen, aflatoxin B_1. *Food Chem Toxicol* **1998**, *36*, 321-326, https://doi.org/10.1016/S0278-6915(97)00160-9.
19. Chlebicz, A.; Śliżewska, K. *In vitro* Detoxification of Aflatoxin B_1, Deoxynivalenol, Fumonisins, T-2 Toxin and Zearalenone by Probiotic Bacteria from Genus *Lactobacillus* and *Saccharomyces cerevisiae* Yeast. *Probiotics Antimicrob Proteins* **2020**, *12*, 289-301, https://doi.org/10.1007/s12602-018-9512-x.
20. Nahle, S.; El Khoury, A.; Savvaidis, I.; Chokr, A.; Louka, N.; Atoui, A. Detoxification approaches of mycotoxins : by microorganisms , biofilms and enzymes. *Int J Food Contam* **2022**, *2*, 1-14, https://doi.org/10.1186/s40550-022-00089-2.
21. Bilal, R.M.; Hassan, F.U.; Saeed, M.; *et al*. Role of Yeast and Yeast-Derived Products as Feed Additives in Broiler Nutrition Role of Yeast and Yeast-Derived Products as Feed Additives. *Anim Biotechnol* **2021**, *0*, 1-10, https://doi.org/10.1080/10495398.2021.1942028.
22. Zhu, C.; Shi, J.; Jiang, C.; Liu, Y. Inhibition of the growth and ochratoxin A production by *Aspergillus carbonarius* and *Aspergillus ochraceus in vitro* and *in vivo* through antagonistic yeasts. *Food Control* **2015**, *50*, 125-132, https://doi.org/10.1016/j.foodcont.2014.08.042.
23. Pretscher, J.; Fischkal, T.; Branscheidt, S.; *et al*. Yeasts from different habitats and their potential as biocontrol agents. *Fermentation* **2018**, *4*, 31, https://doi.org/10.3390/fermentation4020031.
24. Druvefors, U.A.; Schnürer J. Mold-inhibitory activity of different yeast species during airtight storage of wheat grain. *FEMS Yeast Res* **2005**, *5*, 373-378, https://doi.org/10.1016/j.femsyr.2004.10.006.
25. Oro, L.; Feliziani, E.; Ciani, M.; Romanazzi, G.; Comitini, F. Biocontrol of postharvest brown rot of sweet cherries by *Saccharomyces cerevisiae* Disva 599, *Metschnikowia pulcherrima* Disva 267 and *Wickerhamomyces anomalus* Disva 2 strains. *Postharvest Biol Technol* **2014**, *96*, 64-68, https://doi.org/10.1016/j.postharvbio.2014.05.011.
26. Cordero-Bueso, G.; Mangieri, N.; Maghradze, D.; *et al*. Wild grape-associated yeasts as promising biocontrol agents against *Vitis vinifera* fungal pathogens. *Front Microbiol* **2017**, *8*, 2025 https://doi.org/10.3389/fmicb.2017.02025.
27. Grzegorczyk, M.; Żarowska, B.; Restuccia, C.; Cirvilleri, G. Postharvest biocontrol ability of killer yeasts against *Monilinia fructigena* and *Monilinia fructicola* on stone fruit. *Food Microbiol* **2017**, *61*, 93-101, https://doi.org/10.1016/j.fm.2016.09.005.
28. Tryfinopoulou, P.; Skarlatos, L.; Kaplani, P.; Panagou, E.Z. Antifungal activity of *Saccharomyces cerevisiae* and assessment of ochratoxigenic load on currants by means of Real-Time PCR. *Int J Food Microbiol* **2021**, *344*, 109111, https://doi.org/10.1016/j.ijfoodmicro.2021.109111.

29. Ul Hassan, Z.; Al Thani, R.; Atia, F.A.; Alsafran, M.; Migheli, Q.; Jaoua, S. Application of yeasts and yeast derivatives for the biological control of toxigenic fungi and their toxic metabolites. *Environ Technol Innov* **2021**, *22*, 101447, https://doi.org/10.1016/j.eti.2021.101447.
30. Salazar, O.; Asenjo, J.A. Enzymatic lysis of microbial cells. *Biotechnol Lett* **2007**, *29*, 985-994, https://doi.org/10.1007/s10529-007-9345-2.
31. Bleve, G.; Grieco, F.; Cozzi, G.; Logrieco, A.; Visconti, A. Isolation of epiphytic yeasts with potential for biocontrol of *Aspergillus carbonarius* and *A. niger* on grape. *Int J Food Microbiol* **2006**, *108*, 204-209, https://doi.org/10.1016/j.ijfoodmicro.2005.12.004.
32. Gonçalves, B.L.; Rosim, R.E.; de Oliveira, C.A.F.; Corassin, C.H. The *in vitro* ability of different *Saccharomyces cerevisiae* - based products to bind aflatoxin B_1. *Food Control* **2015**, *47*, 298-300, https://doi.org/10.1016/j.foodcont.2014.07.024.
33. Pizzolitto, R.P.; Bueno, D.J.; Armando, M.R.; Cavaglieri, L.; Dalcero, A.M.; Salvano, M.A. Binding of Aflatoxin B_1 to Lactic Acid Bacteria and *Saccharomyces cerevisiae in vitro*: A Useful Model to Determine the Most Efficient Microorganism. In: *Aflatoxins - Biochemistry and Molecular Biology* **2011**, 323-346, https://doi.org/10.5772/23717.
34. Poloni, V.; Salvato, L.; Pereyra, C.; *et al*. Bakery by-products based feeds borne-*Saccharomyces cerevisiae* strains with probiotic and antimycotoxin effects plus antibiotic resistance properties for use in animal production. *Food Chem Toxicol* **2017**, *107*, 630-636, https://doi.org/10.1016/j.fct.2017.02.040.
35. Shetty, P.H.; Hald, B.; Jespersen, L. Surface binding of aflatoxin B_1 by *Saccharomyces cerevisiae* strains with potential decontaminating abilities in indigenous fermented foods. *Int J Food Microbiol* **2007**, *113*, 41-46, https://doi.org/10.1016/j.ijfoodmicro.2006.07.013.
36. Raju, M.V.L.N.; Devegowda, G. Influence of esterified-glucomannan on performance and organ morphology, serum biochemistry and haematology in broilers exposed to individual and combined mycotoxicosis (aflatoxin, ochratoxin and T-2 toxin). *Br Poult Sci* **2000**, *41*, 640-650, https://doi.org/10.1080/713654986.
37. Parlat, S.S.; özcan, M.; Oguz. H. Biological suppression of aflatoxicosis in Japanese quail *(Coturnix coturnix* japonica) by dietary addition of yeast (*Saccharomyces cerevisiae*). *Res Vet Sci* **2001**, *71*, 207-211, https://doi.org/10.1053/rvsc.2001.0512.
38. Bueno, D.J.; Casale, C.H.; Pizzolitto, R.P.; Salvano, M.A.; Oliver, G. Physical adsorption of aflatoxin B_1 by lactic acid bacteria and *Saccharomyces cerevisiae*: A theoretical model. *J Food Prot* **2007**, *70*, 2148-2154, https://doi.org/10.4315/0362-028X-70.9.2148.
39. Xu, H.; Wang, L.; Sun, J.; *et al*. Microbial detoxification of mycotoxins in food and feed. *Crit Rev Food Sci Nutr* **2021**, *0*, 1-19, https://doi.org/10.1080/10408398.2021.1879730.
40. Keller, L.; Abrunhosa, L.; Keller, K.; Rosa, C.A.; Cavaglieri. L.; Venâncio, A. Zearalenone and its derivatives α-zearalenol and β-zearalenol decontamination by Saccharomyces cerevisiae strains isolated from bovine forage. *Toxins (Basel)* **2015**, *7*, 3297-3308, https://doi.org/10.3390/toxins7083297.
41. Armando, M.R.; Pizzolitto, R.P.; Dogi, C.A.; *et al*. Adsorption of ochratoxin A and zearalenone by potential probiotic *Saccharomyces cerevisiae* strains and its relation with cell wall thickness. *J Appl Microbiol* **2012**, *113*, 256-264, https://doi.org/10.1111/j.1365-2672.2012.05331.x.
42. Faucet-Marquis, V.; Joannis-Cassan, C.; Hadjeba-Medjdoub, K.; Ballet, N.; Pfohl-Leszkowicz, A. Development of an *in vitro* method for the prediction of mycotoxin binding on yeast-based products: Case of aflatoxin B_1, zearalenone and ochratoxin A. *Appl Microbiol Biotechnol* **2014**, *98*, 7583-7596, https://doi.org/10.1007/s00253-014-5917-y.
43. Tapingkae, W.; Srinual, O.; Lumsangkul, C.; *et al*. Industrial-Scale Production of Mycotoxin Binder from the Red Yeast *Sporidiobolus pararoseus* KM281507. *J Fungi* **2022**, *8*, 353, https://doi.org/10.3390/jof8040353.
44. Bruinenberg, P.G.; Castex, M. Evaluation of a Yeast Hydrolysate from a Novel Strain of *Saccharomyces cerevisiae* for Mycotoxin Mitigation using *In vitro* and *In vivo* Models. *Toxins (Basel)* **2022**,*14*, 1-12, https://doi.org/10.3390/toxins14010007.
45. Shang, Q.H.; Yang, Z.B.; Yang, W.R.; Li, Z.; Zhang, G.G.; Jiang, S.Z. Toxicity of mycotoxins from contaminated corn with or without yeast cell wall adsorbent on broiler chickens. *Asian-Australasian J Anim Sci* **2016**, *29*, 674-680, https://doi.org/10.5713/ajas.15.0165.
46. Freimund, S.; Sauter, M.; Rys, P. Efficient Adsorption of the Mycotoxins Zearalenone and T - 2 Toxin on a Modified Yeast Glucan Efficient Adsorption of the Mycotoxins Zearalenone and T-2 Toxin on a Modified Yeast Glucan. **2011**, 1234, https://doi.org/10.1081/PFC-120019892.
47. Yiannikouris, A.; Gwénaëlle, A.; Buléon, A.; *et al*. Comprehensive conformational study of key interactions involved in zearalenone complexation with beta-D-glucans. *Biomacromolecules* **2004**, *5*, 2176-2185, https://doi.org/10.1021/bm049775g.
48. François, A.Y.J.; Poughon, L.; Dussap, C-G.; Bertin, G.; Jeminet, G.; Jouany, J-P. Alkali extraction of beta-D-glucans from *Saccharomyces cerevisiae* cell wall and study of their adsorptive properties toward zearalenone. *J Agric Food Chem* **2004**, *52*, 3666-3673, https://doi.org/10.1021/jf035127x.
49. Yiannikouris, A.; François, J.; Poughon, L.; *et al*. Adsorption of zearalenone by β-D-glucans in the *Saccharomyces cerevisiae* cell wall. *J Food Prot* **2004**, *67*, 1195-1200, https://doi.org/10.4315/0362-028X-

67.6.1195.
50. Yiannikouris, A.; François, J.; Poughon, L.; *et al*. Influence of pH on complexing of model β-D-glucans with zearalenone. *J Food Prot* **2004**, *67*, 2741-2746, https://doi.org/10.4315/0362-028X-67.12.2741.
51. dos Santos, V.M.; da Silva, O.G.; de Lima, C.A.R.; Curvello, F.A. Broiler chick performance using *Saccharomyces cerevisiae* yeast cell wall as an anti-mycotoxin additive. *Czech J Anim Sci* **2021**, *66*, 65-72, https://doi.org/10.17221/237/2020-CJAS.
52. Mgbeahuruike, A.C.; Ejiofor, T.E.; Ashang, M.U.; *et al*. Reduction of the adverse impacts of fungal mycotoxin on proximate composition of feed and growth performance in broilers by combined adsorbents. *Toxins (Basel)* **2021**, *13*, 1-14, https://doi.org/10.3390/TOXINS13060430.
53. Nešić, K.; Habschied, K.; Mastanjević, K. Possibilities for the Biological Control of Mycotoxins in Food and Feed. *Toxins (Basel)* **2021**, *13*, 198, https://doi.org/10.3390/toxins13030198.
54. Leo, V.V.; Viswanath, V.; Purbajyoti, *et al*. *Saccharomyces* and Their Potential Applications in Food and Food Processing Industries. In: Abdel-Azeem, A.M.; Yadav, A.N.; Yadav, N.; Usmani, Z. ed. *Industrially Important Fungi for Sustainable Development.* Springer, **2021**, 393–427. https://doi.org/10.1007/978-3-030-67561-5_12.
55. Papp, L.A.; Horváth, E.; Peles, F.; Pócsi, I.; Miklós, I. Insight into Yeast – Mycotoxin Relations. *Agricult* **2021**, *11*, 1290, https://doi.org/10.3390/agriculture11121291.