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Aflatoxin M1 retention by exopolysaccharides from kefir grains: Impact of extraction method on binding efficiency

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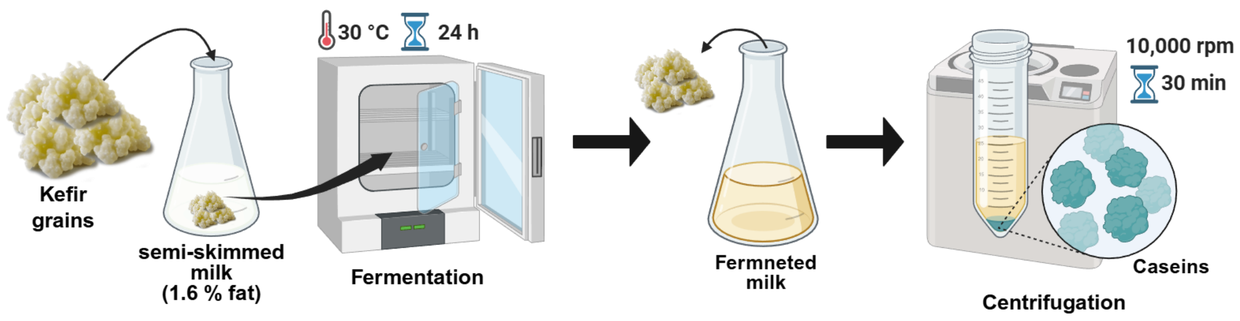
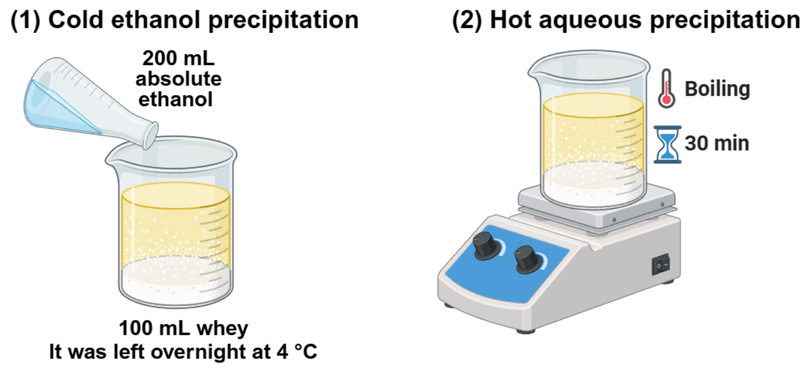
INTRODUCTION & AIM

Aflatoxin M1 (AFM1) is a carcinogenic and thermally stable mycotoxin frequently found in dairy products, where conventional heat treatments fail to eliminate it (1). This challenge has led to increasing interest in natural, food-grade alternatives for toxin mitigation. Exopolysaccharides (EPS) produced by kefir grains have gained attention due to their structural complexity and their potential to interact with contaminants (2). Because the chemical composition of kefir EPS depends strongly on the extraction method, differences in protein and carbohydrate content may influence their ability to retain AFM1 (2, 3). To explore this relationship, EPS obtained through cold ethanol precipitation and hot aqueous extraction were compared to determine how extraction-driven compositional changes affect their AFM1-binding performance. This approach provides insight into the potential application of kefir EPS as a natural strategy for reducing AFM1 in dairy systems.

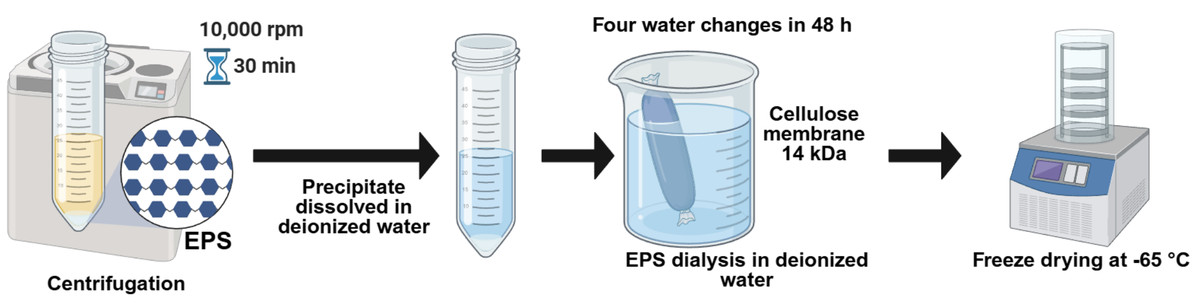
METHOD

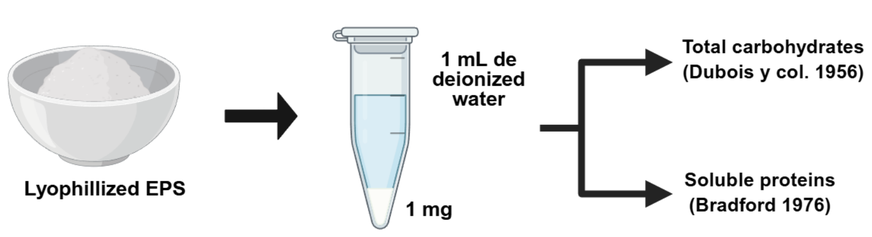
EPS production and precipitation



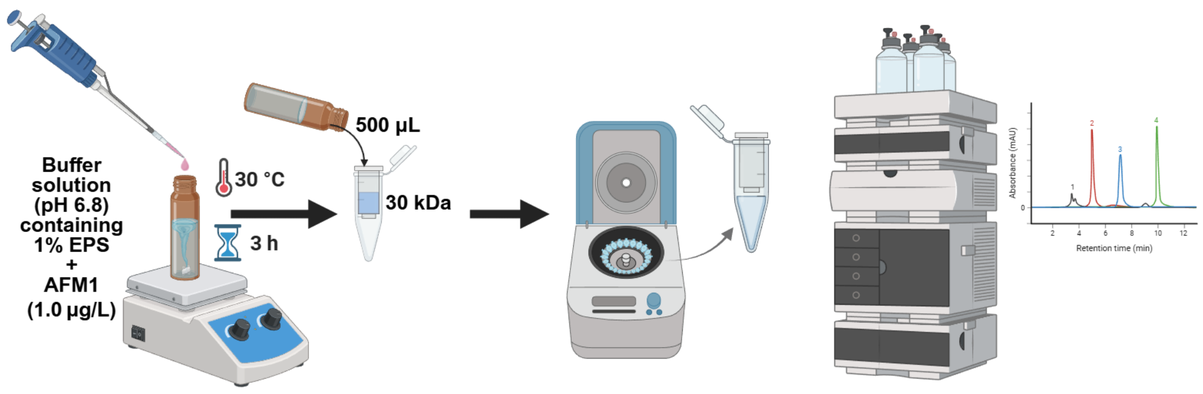


Purification and drying of extracted EPS





AFM1-EPS interaction assay



RESULTS & DISCUSSION

Cold and hot extraction methods produced EPS with markedly different physicochemical characteristics, which directly influenced their AFM1-binding behavior. Cold extraction yielded the highest amount of solids (9,400 mg) and preserved a balanced sugar-to-protein ratio, indicating minimal structural disruption during processing. In contrast, hot extraction drastically altered EPS composition, reducing soluble protein content by 69% while increasing sugar concentration nearly eight-fold. As illustrated in Figure 1, these shifts in matrix composition underscore the strong dependence of EPS structure on extraction conditions and provide insight into their functional divergence.

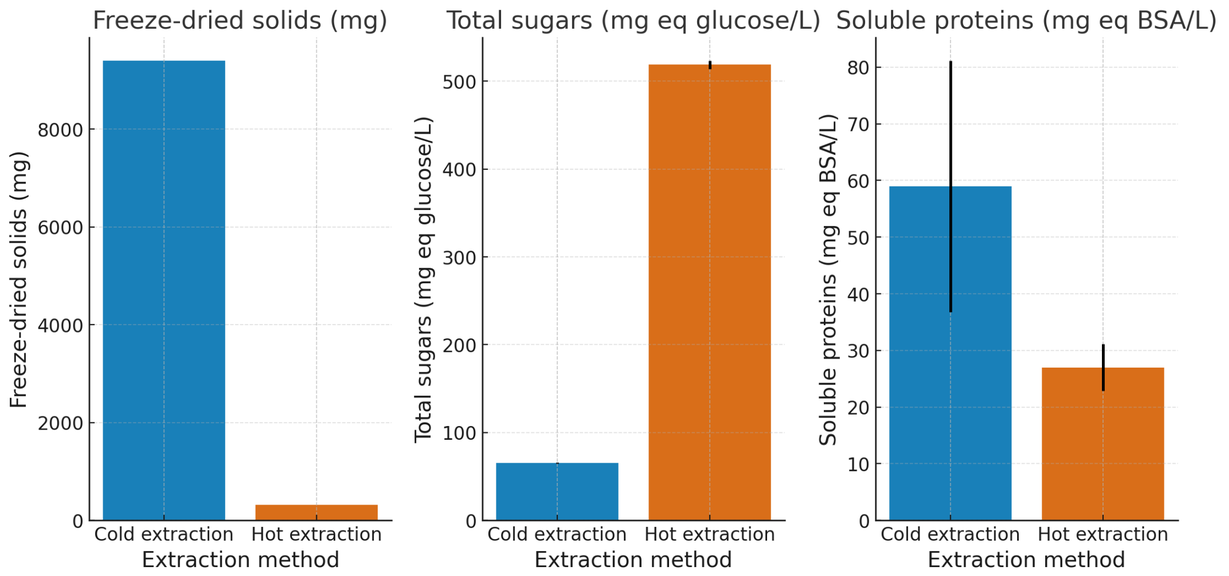


Figure 1. Comparative effects of cold and hot extraction on EPS yield and biochemical composition.

These compositional differences translated into significant variation in AFM1 sequestration efficiency. Cold-extracted EPS exhibited consistently high retention (>80%) throughout the assay, suggesting that the presence of proteinaceous components facilitates stable toxin binding, likely through complementary interaction sites or synergistic sugar–protein domains. Conversely, hot-extracted EPS displayed lower and gradually declining retention, consistent with a more purified polysaccharide matrix lacking protein-based binding motifs. The statistically significant differences between extraction methods ($p < 0.05$) emphasize the functional relevance of protein–mycotoxin interactions and highlight the potential of minimally processed EPS for toxin-binding applications.

CONCLUSION

Extraction methodology significantly influenced both EPS composition and AFM1-binding performance. Cold-extracted EPS, which retained protein components, demonstrated superior and stable AFM1 sequestration, supporting the role of protein–mycotoxin interactions in binding efficiency. In contrast, hot extraction produced a purer but functionally less active EPS fraction. Overall, these findings highlight the potential of kefir-derived EPS as a natural strategy for mycotoxin mitigation in dairy products and support further investigation into their industrial application.

ACKNOWLEDGMENT

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