ejpmr, 2024, 11(7), 119-125

SJIF Impact Factor 7.065

EUROPEAN JOURNAL OF PHARMACEUTICAL AND MEDICAL RESEARCH

www.ejpmr.com

Research Article

ISSN 2394-3211

EJPMR

DETECTION OF THE ABILITY OF MANNOPROTEIN EXTRACTED FROM*SACCHAROMYCES CEREVISIAE*TO INHIBIT THE ABILITY OF SOME SPECIES OF BACTERIA ISOLATED FROM DIFFERENT INFECTION SOURCES TO PRODUCE VIRULENCE FACTORS

Ashwaq Muthanna Wahib[1*], Mohammed Nadhir Maaroof[2]

[1]Ph.D.Student College of Education for Pure Science, University of Tikrit.
[2]College of Education for Pure Science, University of Tikrit.

***Corresponding Author: Ashwaq Muthanna Wahib**

Ph.D.Student College of Education for Pure Science, University of Tikrit.

Article Received on 14/05/2024 **Article Revised on 04/06/2024** **Article Accepted on 24/06/2024**

ABSTRACT

The study included (60) bacterial isolates from the total number of samples (100) samples, collected from different sources of infections and at different ages including both sexes, collected from patients with wound, burn and urinary tract infections. The isolates were diagnosed based on culture, microscopic and biochemical tests. Two isolates of *Saccharomyces cerevisiae* were selected for the production of mannoprotein .Mannoprotein was used in conjunction with urease, hemolysin and biofilm virulence media to demonstrate the effect of mannoprotein in preventing the ability of some pathogenic bacteria such as (*Staphylococcus aureus*, *Escherichia coli*, *Proteus mirabilis*, *Klebsiella pneumonia*) to product virulence factors. The results of the current study showed the ability of mannoprotein extracted from baker's yeast of the second and third types to prevent *Staph. aureus* bacteria from product the enzyme hemolysin. As for the bacterial isolates (*K.Pneumoniae*) under study, their ability to produce the virulence factor (urease) and (biofilm) was tested. The isolates under study were tested for the production of the enzyme urease. The results of the current study showed that the bacterial isolates did not produce the virulence factor (urease) for both types of mannoprotein due to its effect on preventing the bacteria from product the virulence factor at all concentrations. The ability of bacteria to product the virulence factor (biofilm) is considered a qualitative test to detect the ability to form biofilms based on the thickness and density of the biofilms associated with the inner wall of the test tube. Through the results obtained, the results of the study showed that the first isolate of bacteria (*K.Pneumoniae*) did not produce the virulence factor for the two types of mannoprotein used. As for the second isolate of bacteria, it was not productive of the virulence factor for the type of mannoprotein extracted from the first isolate used in neutralization and produced it when using the mannoprotein extracted from the second isolate and for all concentrations. Bacterial isolates (*E.coli*) Two isolates of the bacteria under study were used to detect the ability of mannoprotein to prevent bacteria from product (biofilm). The bacteria did not product the virulence factor due to their inability to produce the biofilm formation factor due to the effect of mannoprotein on preventing bacteria from producing the virulence factor. This is for the first bacterial isolate and for the two types of mannoprotein and at all concentrations. As for the second bacterial isolate, it produced the cell membrane factor when using mannoprotein from the first yeast isolate. However, when using mannoprotein from the second yeast isolate, it did not produce the virulence factor. As for Proteus bacteria, the first isolate of (*P.mirabilis*) bacteria was unable to product the virulence factor urease for the mannoprotein extracted from both yeast isolates for the first concentration only (100%) of mannoprotein, while the concentration (75%) and (50%) were ineffective in preventing the bacteria from product the virulence factor urease, and thus the bacteria were able to produce the virulence factor. As for the second isolate of (*P.mirabilis*) bacteria, it was unable to product the virulence factor urease for the mannoprotein of both yeast isolates at the concentration (100%) and (75%), while the concentration (50%) and as for the virulence factor biofilm, the first isolate of Proteus bacteria was unable to product biofilm in the presence of the mannoprotein extracted from both yeast isolates for the first concentration only (100%) of mannoprotein, while the concentration (75%) and (50%) were ineffective in preventing the bacteria from product biofilm. As for the second isolate of Proteus bacteria, it was unable to Biofilm product for mannoprotein of both yeast isolates at concentrations of (100%) and (75%), while concentration of (50%) was ineffective in neutralizing bacteria.

KEYWORDS: Mannoprotein, Saccharomyces cerevisiae, Biofilm, urease, Hemolysin.

INTRODUCTION

Yeasts are fungi. Eukaryotic., unicellular with properties completely different from those of bacteria. They have the ability to reproduce asexually by simple fission. Or budding, either sexually by spores. One of the most economically, industrially and medically important yeasts is baker's yeast *Saccharomyces cerevisiae*.[1] *S. cerevisiae* is a unicellular, eukaryotic yeast with vegetative forms that are oval, round, conical and oblong. It is solitary or arranged in small groups, thus appearing as small, white or cream-coloured colonies with regular margins and a convex apex, and with a sticky, mucous consistency.[2] *S. cerevisiae* is heterotrophic, due to the lack of chlorophyll, and is a saprophytic. *S. cerevisiae* is mesophilic, and the optimum temperature for its growth is 30°C. As for the yeast's need for air, it is facultative, meaning it can grow in both aerobic and anaerobic conditions, but yeast growth is slow in anaerobic conditions, unlike its growth in aerobic conditions.[3] The cell wall is a functional interface between the interior and exterior of the cell. It plays an important role in determining the shape and size of the cell, as well as maintaining the osmotic balance of the cell. The cell wall has a multifunctional role in the living processes of yeast. The cell wall in *S.cerevisiae* is composed primarily ofmannoproteins and beta-glucans.[4] Electron microscopy shows that the cell wall of S. cerevisiae is organized into two layers, inner and outer. The inner layer is composed of beta-glucan and the outer layer is composed of mannoprotein. Mannoprotein plays a role in protecting the inner layer (glucan) from cell wall-degrading enzymes and also plays a role in regulating cell wall permeability, as well as in sexual differentiation between cells and their formation of biofilms.[5]Mannoprotein is a bioemulsifier, composed of protein and sugar, known as amphiphilic with lipophilic and hydrophilic properties. This bioemulsifier is able to stabilize oil-in-water emulsions.[6] The emulsifying properties of the mannoprotein of the yeast S. cerevisiae were first described in 1988 by Cameron et al.[7] Mannoproteins are part of the outer layer of the yeast cell wall, which not only provides the cells with the rigidity that protects them from osmotic stress but also helps maintain their shapes, allowing them to change shape in accordance with the phase of the cell cycle. The structural mannoproteins are interwoven within a network of glucans to form the outer layer of the *S. cerevisiae* cell wall.[8] Bakery yeast is an inexpensive, non-toxic source used to produce this bio-emulsifier Mannoprotein, and has emulsifying properties that can be of commercial importance. These good properties include non-toxicity, high foaming ability, synthesis from natural substrates under mild environmental conditions, high biodegradability, high activity at extreme temperatures, and tolerance to different levels of salinity and pH. Thus, it is increasingly attracting the attention of the scientific and international community as a promising candidate alternative to a number of synthetic surfactants (emulsifiers).[7] Since *S. cerevisiae* yeast is edible (non-toxic), the bio-emulsifier of mannoprotein is expected to be non-toxic, and is generally recognized as safe.[8] Therefore, mannoprotein can be used in the food and beverage industry. Mannoprotein and beta-glucan are mainly used as animal feed ingredients for their nutritional value. Bioemulsifiers can be used to produce mayonnaise along with Carboxymethyl cellulose (CMC), instead of using expensive ingredients such as ginseng to form mayonnaise. Mannoprotein extracted from the cell wall of baker's yeast has a long history as non-specific biomodifiers. Mannoprotein recognition is an important mechanism of host interaction with various pathogens, which may provide a therapeutic or preventive approach to oral bacterial infections by elevating host antibacterial mechanisms.[6]

MATERIALS AND WORKING METHODS

Table (1-1): Laboratory devices and equipment used in the study.

Manufacturer and Origin	Materials and equipment
Hirayama (Japan)	Autoclave
Memmert (Germany)	Incubator
KERN (Germany)	Sensitive electric balance
Nuva (Turkey)	Laminer flow Hood
Stuart (UK)	Hot Plate & Magnetic Stirrer
Memmert (Germany)	Oven
Gallenkamp (England)	Water Distiller
Hi-Media (India)	Standard Wire Loop
Al-Hani (USA)	Disposable Petri dishes
Biozek (Metherlands)	P-line Tubes
Sterelline (England)	Sterilized cotton swabs
GEMA (Spain)	Millipore Filter
BBL (USA)	Conical flasks
Canfort Lab (China)	Cork Boere
CHMLAB (Spain)	Filter Paper
TEKA / Spain	Refrigerator
Fisher Scientific (USA)	Centrifuge
Volac (England	Pasteur pipettes
Sun (China)	Tibs
Brand (Germany)	Glass cylinder
Amal (Turkic)	Burner
Bioneer(Korea)	Rack Tube
Kardelen (Turkey	Medical Cotton

Table (1-2): The culture media used in the study.

Manufacturer and Origin	Culture Media
Hi-Media (India)	Blood base agar
Hi-Media(India)	MacConkey Agar
Hi- Media (India)	Urea agar base
Oxoid (England)	Muller-Hinton agar
Oxoid (England)	Nutrient broth
Oxoid (England)	Manitol Salt agar
Oxoid (England)	Saboroid dextrose agar
Oxoid (England)	Saboroid dextrose broth

Growth of *S. cerevisiae* yeast isolates

Dry yeast isolates were activated and grown according to[9], by taking 1 g of yeast powder and inoculating it in

10 ml of liquid Sabouraud dextrose broth medium and incubating it at 30°C for half an hour. After the incubation period was over, it was planted on solid Sabouraud dextrose medium and then incubated for 48 hours at 30°C.

Production Mannoprotein

Yeast isolates were activated on Sabouraud dextrose liquid medium, mixed well and incubated at 30°C in a shaking incubator for 4 days.[10]

Mannoprotein extraction

The obtained cells were separated by centrifugation at 10,000 rpm for 20 min, then the cells were suspended in phosphate buffer and the suspended cells were boiled for 5 min and then centrifuged at 3000 rpm for 10 min. Then the supernatant representing the crude mannoprotein was taken and placed in dialysis bags to obtain the mannoprotein after getting rid of the excess water by filtering it through the dialysis bags after placing the dialysis bags containing the mannoprotein in the sucrose and covering them completely to complete the filtration.[10]

RESULTS

(60) isolates were obtained from the total number of samples of (100) samples, collected from different infection sources and at different ages including both sexes, collected from patients with wound, burn and urinary tract infections, for the period from 8/20/2022 to 1/20/2023 from Baqubah Teaching Hospital and Muqdadiyah General Hospital in Diyala Governorate. The samples were collected under the supervision of a specialist doctor and from both sexes, females and males, whose ages ranged between (9 months - 50 years). MacConkey agar, blood agar and mannitol saline agar were used to culture the samples, then the plates were incubated aerobically inverted at a temperature of 37 C for 24 hours and their diagnosis was confirmed by biochemical tests. The results showed that 46 samples (46%) showed bacterial growth on the culture media used. The reason for the appearance of this high percentage is that burns, wounds, and the urinary tract represent a fertile environment suitable for the growth of bacteria, while 54 samples (54%) were negative for bacterial growth. The reason for the absence of growth in this percentage is due to good sterilization conditions, and it is also due to the fact that the burn has not yet reached the degree of bacterial infection, and thus negatively affects the absence of bacterial growth.[11]

The effect of mannoprotein on neutralizing the ability of bacteria to product virulence factors

The study included 8 bacterial isolates from the total isolates obtained from different isolation sources. Two isolates were taken from each type of bacteria (*Staph. aureus*), (*K.Pneumoniae*), (*E.coli*), and (*P. mirabilis*). These isolates were among the isolates capable of product virulence factors (hemolysin, urease, biofilm). A test was conducted to show the effect of the ability of mannoprotein to prevent bacteria from product virulence factors by using media specific to virulence factors, as shown in the following tables:

1- The effect of mannoprotein on neutralizing the ability of *Staph.aurase* bacteria to product the virulence factor hemolysin.

The bacterial isolates (*Staph. aurase*) under study were subjected to detecting the ability of mannoprotein to prevent bacteria from product the enzyme hemolysin by growing them on blood agar medium (containing 5% human blood) with the addition of (mannoprotein) of the second and third types and at three different concentrations (100%), (75%) and (50%). After incubation for 24 hours at 37°C, the isolates did not show transparent decomposition areas around the growing bacterial colonies for all concentrations due to their inability to produce the enzyme hemolysin. Through the results obtained, it was shown that (2) bacterial isolates did not produce this enzyme, as shown in Table (1-3).

Table (1-3): Shows the effect of mannoprotein on neutralizing the ability of *Staph.aurase* bacteria to product the virulence factor hemolysin.

Virulence factor	%50	%75	%100	Type of isolate Yeast (Mannoprotein)	Bacteria
Hemolysin	+	+	+	Type I	*Staph.aurase* isolate 1
Hemolysin	+	+	+	Type II	*Staph.aurase* isolate 1
Hemolysin	+	+	+	Type I	*Staph.aurase* isolate 2
Hemolysin	+	+	+	Type II	*Staph.aurase* isolate 2

The results of our study show that *Staph. aurase* isolates are unable to product the virulence factor hemolysin at a rate of (100%) for both yeast isolates due to the effect of mannoprotein on preventing the bacteria and neutralizing them from secreting the virulence factor.

2- The effect of mannoprotein on neutralizing the ability of *K.Pneumoniae* bacteria to product the virulence factor urease.

Urea agar medium was used and poured into tubes. After reaching a certain temperature and before it solidified, (1 ml) of mannoprotein extracted from two isolates of baker's yeast was added to each tube at concentrations of (100%, 75%, 50%) and placed in an inclined manner until it solidified. After the medium solidified, bacterial isolates capable of producing urease were cultivated. After incubating the bacteria for (24) hours, the results were read as shown in Table (1-4).

Table (1-4): Shows the effect of mannoprotein on neutralizing the ability of *K.Pneumoniae* bacteria to product the virulence factor urease.

Virulence factor	%50	%75	%100	Type of isolate Yeast (Mannoprotein)	Bacteria
urease	+	+	+	Type I	*K. Pneumoniae* Isolation 1
urease	+	+	+	Type II	*K. Pneumoniae* Isolation 1
urease	+	+	+	Type I	*K. Pneumoniae* Isolation 2
urease	+	+	+	Type II	*K. Pneumoniae* Isolation 2

The results of our study show that *K. Pneumoniae* isolates are unable to product the virulence factor urease at a rate of (100%) for both yeast isolates due to the effect of mannoprotein on preventing the bacteria and neutralizing them from secreting the virulence factor urease.

3- The effect of mannoprotein on neutralizing the ability of *K. Pneumoniae* bacteria to product the virulence factor biofilm.

The liquid neutron medium was used and poured into tubes. After reaching a certain temperature, (1 ml) of the mannoprotein extracted from two isolates of baker's yeast was added to each tube at concentrations of (100%, 75%, 50%). After that, the bacterial isolates capable of producing biofilm were cultured. After incubating the bacteria for (24) hours, the results were read as shown in Table (1-5).

Table (1-5): Shows the effect of mannoprotein on neutralizing the ability of *K. Pneumoniae* bacteria to product the virulence factor biofilm.

Virulence factor	%50	%75	%100	Type of isolate Yeast (Mannoprotein)	Bacteria
Biofilm	+	+	+	Type I	*K. Pneumoniae* Isolation 1
Biofilm	+	+	+	Type II	*K. Pneumoniae* Isolation 1
Biofilm	+	+	+	Type I	*K. Pneumoniae* Isolation 2
Biofilm Produced the virulence factor	-	-	-	Type II	*K. Pneumoniae* Isolation 2

The results of our study showed that the first isolate of *K. Pneumoniae* bacteria was unable to product the biofilm virulence factor at a rate of (100%) for the mannoprotein extracted from both yeast isolates and for all concentrations. As for the second isolate of *K.Pneumoniae* bacteria, it was unable to product the biofilm virulence factor at a rate of (50%) for the mannoprotein of the second isolate and was able to product biofilm for the mannoprotein of the third isolate, due to the effect of mannoprotein on preventing the bacteria and neutralizing them from product the biofilm virulence factor.

4- The effect of mannoprotein on neutralizing the ability of *E.coli* bacteria to product the virulence factor biofilm.

The liquid neutron medium was used and poured into tubes. After reaching a certain temperature, (1 ml) of the mannoprotein extracted from two isolates of baker's yeast was added to each tube at concentrations of (100%, 75%, 50%). The bacterial isolates capable of producing biofilm were cultured. After incubating the bacteria for (24) hours, the results were read as shown in Table (1-6).

Table (1-6): Shows the effect of mannoprotein on neutralizing the ability of *E.coli* bacteria to product the virulence factor biofilm.

Virulence factor	%50	%75	%100	Type of isolate Yeast (Mannoprotein)	Bacteria
Biofilm	+	+	+	Type I	*E.coli* Isolation 1
Biofilm	+	+	+	Type II	*E.coli* Isolation 1
Biofilm	-	-	-	Type I	*E.coli* Isolation 2
Biofilm	+	+	+	Type II	*E.coli* Isolation 2

The results of our study showed that the first isolate of *E. coli* bacteria was unable to product the virulence factor biofilm at a rate of (100%) for the mannoprotein extracted from both yeast isolates and for all concentrations. As for the second isolate of E. coli bacteria, it was able to product the virulence factor biofilm at a rate of (50%) for the mannoprotein of the second isolate and was unable to product biofilm for the mannoprotein of the third isolate, due to the effect of mannoprotein on preventing bacteria and neutralizing them from product the virulence factor biofilm.

5- The effect of mannoprotein on neutralizing the ability of *P. mirabilis* bacteria to product the virulence factor biofilm.

The liquid neutron medium was used and poured into tubes. After reaching a certain temperature, (1 ml) of the

mannoprotein extracted from two isolates of baker's yeast was added to each tube at concentrations of (100%, 75%, 50%). After that, the bacterial isolates capable of producing biofilm were cultivated. After incubating the bacteria for (24) hours, the results were read as shown in Table (1-7).

Table (1-7): Shows the effect of mannoprotein on neutralizing the ability of *P. mirabilis* bacteria to product the virulence factor biofilm.

Virulence factor	%50	%75	%100	Type of isolate Yeast (Mannoprotein)	Bacteria
Biofilm	-	-	+	Type I	*P.mirabilis isolate 1*
Biofilm	-	-	+	Type II	*P.mirabilis isolate 1*
Biofilm	-	+	+	Type I	*P.mirabilis isolate 2*
Biofilm	-	+	+	Type II	*P.mirabilis isolate 2*

The results of our study showed that the first isolate of *P. mirabilis* bacteria was unable to product the biofilm virulence factor for the mannoprotein extracted from both yeast isolates for the first concentration only (100%) of mannoprotein, while the concentrations (75%) and (50%) were ineffective in preventing the bacteria from secreting the biofilm virulence factor, and thus the bacteria were able to produce the virulence factor. As for the second isolate of Proteus bacteria, it was unable to product the biofilm virulence factor for the mannoprotein of both yeast isolates at concentrations (100%) and (75%), while the concentration (50%) was ineffective in neutralizing the bacteria, and thus the second isolate was able to product the biofilm for the concentration (50%) only.

6- The effect of mannoprotein on neutralizing the ability of *P. mirabilis* bacteria to product the virulence factor urease.

Urea agar medium was used and poured into tubes. After reaching a certain temperature and before it solidified, (1 ml) of mannoprotein extracted from two isolates of baker's yeast was added to each tube at concentrations of (100%, 75%, 50%) and placed in an inclined manner until it solidified. After the medium solidified, bacterial isolates capable of producing urease were cultivated. After incubating the bacteria for (24) hours, the results were read as shown in Table (1-8).

Table (1-8): Shows the effect of mannoprotein on neutralizing the ability of *P. mirabilis* bacteria to product the virulence factor urease.

Virulence factor	%50	%75	%100	Type of isolate Yeast (Mannoprotein)	Bacteria
urease	-	-	+	Type I	*P.mirabilis isolate 1*
urease	-	-	+	Type II	*P.mirabilis isolate 1*
urease	-	+	+	Type I	*P.mirabilis isolate 2*
urease	-	+	+	Type II	*P.mirabilis isolate 2*

The results of our study showed that the first isolate of Proteus bacteria was unable to product the virulence factor urease for the mannoprotein extracted from both yeast isolates for the first concentration only (100%) of mannoprotein, while the concentrations (75%) and (50%) were ineffective in preventing the bacteria from product the virulence factor urease, and thus the bacteria were able to produce the virulence factor. As for the second isolate of Proteus bacteria, it was unable to secrete the virulence factor urease for the mannoprotein of both yeast isolates at concentrations (100%) and (75%), while the concentration (50%) was ineffective in neutralizing the bacteria, and thus the second isolate was able to product urase for the concentration (50%) only.

DISCUSSION

The results of our current study matched the results of the researcher (Anna) et al.[12] in (2022), where he explained that adding mannoprotein to bacteria has a positive effect on the growth of bacteria and that the Mannoproteins isolated from *S. cerevisiae* have a clear effect on the growth of (*E. coli*), Pseudomonas aureoginosa and *P. mirabilis*. It also matched the study of Saleh et al.[13], where his study showed that the partially and completely purified mannoprotein isolated from *S. cerevisiae* has an effect on the selected bacterial isolates. After purifying the yeast mannoprotein by gel filtration, the effectiveness of its inhibitory activity increased against eleven bacterial isolates that were studied, including bacterial isolates: *E. coli*, *P. aeruginosa*, *K. pneumoniae*, and *Staph.aureus.*

Our study differed from the study of researcher Walencka et al., where no direct antibiotic activity was observed for mannoprotein (*S. cerevisiae*) against *Staph. aureus* and *Staph.epidermidis* cells, but mannoprotein had an effective effect in reducing the initial deposition of staphylococci and in reducing the formation of the formed biofilm, which accelerates the detachment of mature staphylococcal biofilms.[14] The results were

consistent with the study of researchers (Rashee and Haydar[15], where the activity of mannoprotein was evaluated as an antibacterial, antiadhesive and antibiofilm against some pathogenic bacteria. The results showed an inhibitory effect against *P. aerugenosa* and *S. aureus*, and the results of his study also showed a high antiadhesion activity of mannoprotein against *P. aeruginosa* bacteria. This indicates that mannoprotein has an inhibitory effect against pathogenic bacteria.

It also agreed with the results of researcher Ghada et al.[16] who mentioned that *Staph.aureus* and *E. coli* were inhibited by using mannoprotein.

Our current study is also consistent with the study of the researcher who indicated that mannoprotein extracted from *S.cerevisiae*, which has activity against pathogenic bacteria, can be used as an inhibitor of *Staph.aureus*. Mannoprotein also accelerates the detachment of mature staphylococcal biofilms, which were previously formed under ideal conditions. It was found that the anti-biofilm effect of mannoprotein reflects its effect on the hydrophobic cell surface.[17]

From the above, the results obtained indicate that the mannoprotein naturally present in the cell wall has an inhibitory effect on pathogenic bacteria and works to limit the growth of unwanted bacteria. The research should be expanded to include the analysis of the effect of the tested preparation on the growth of selected bacteria in the food environment and in the digestive tract. In the future, it would be useful to evaluate how the purification process of mannoprotein affects its antimicrobial properties.

WHO encourages research into the prevention and treatment of diseases using alternative treatments other than antibiotics, such as treatments derived from natural materials, plant extracts, etc.[18] This results from the development and spread of resistance of pathogenic microorganisms to antibiotics and the presence of drug residues in the environment and food chain, which endangers the health and safety of humans and animals.[16] Natural medicines and some other natural products, including derivatives of yeast cell wall components, can be used to treat infections as alternatives to common antibiotics that have become useless for bacteria due to their resistance.[19]

REFERENCES

1. Phale, S. Yeast: characteristics and economic significance. J. Bioprocess. Biotech., 2018; 8(5).
2. Stefanini, I.; Dapporto, L.; Legras, J. L.; Calabretta, A.; Paola, M. D.; Filippo, C. D.; Viola, R.; Capretti, P.; Polsinelli, M.; Turillazzi, S. and Cavalieri, D. Role of social wasps in Saccharomycescerevisia ecology and evolution. PNAS., 2012; 109(33): 13398–13403.
3. Salari, R.and Salari, R. Investigation of the Best Saccharomycescerevisiae growth condition. Electronic physician, 2017; 9(1): 3592-3597.
4. Ueda, M. (Ed.). (2019). Yeast cell surface engineering: biological mechanisms and practical applications. Springer.
5. Molon, M.; Woznicka, O. and Zebrowski, J. Cell wall biosynthesis impairment affects the budding lifespan of the Saccharomyces cerevisiae yeast, Biogerontology, 2018; 19: 67–79.
6. Alizadeh-Sani M, Hamishehkar H, Khezerlou A, Azizi-Lalabadi M, Azadi Y, Nattagh-Eshtivani E, Fasihi M, Ghavami A, Aynehchi A, and Ehsani A. Bioemulsifiers derived from microorganisms: applications in the drug and food industry. Pharm. Bull., 2018; 8(2): 191-199.
7. De Iseppi A, Curioni A, Marangon M, Vincenzi S, Kantureeva G, and Lomolino G. Characterization and emulsifying properties of extracts obtained by physical and enzymatic methods from an oenological yeast strain. J Sci Food Agric., 2019; 99(13): 5702-5710.
8. Liu H, Liu L, Hui H and Wang Q . Structural characterization and antineoplastic activity of Saccharomyces cerevisiae mannoprotein. Int. J. Food Prop., 2014; 18:359–371.
9. Al-Goshaah FAS.Studying the effect of inhibitory substances produced by Saccharomyces boulardii on virulence factors of some enteric bacteria. P.H.D thesis, Al- Mustansiriya University,College of Science, Baghdad, Iraq. (2005).
10. Alcantara V A, Pajares IG, Simbahan JF, Villarante NR. and Rubio M LD. Characterization of biosurfactant From Saccharomyces cerevisiae 2031 and evaluation of emulsification activity for potential application in bioremediation . Philippine. Agric. Sci., 2010; 93(1): 22-30.
11. Araujo, Katy L. B. , Juthani-Mehta. M , Pisani M. A. , Van Ness P-H-..(2016) . outocomes of older Adults with Sepsis At Admission to an Intensiv care Unite. National Inatitutes of Health , open forum infectionsnDiseaseJan (1) of W DIO.
12. Anna BW, Pavol F, Paulina C, Dominika P, Alicja S, Katarzyna P, and Monika JAntimicrobial and prebiotic activity of mannoproteins isolated from conventional and nonconventional yeast species—the study on selected microorganisms.World J MicrobiolBiotechnol., 2022; 38(12): 256. Published online 2022 Nov 2. doi: 10.1007/s11274-022-03448-5.
13. Saleh AYA, Salman JAS, Aziz RA. Study of the effect of mannoprotein extracted from Saccharomyces cerevisiae on some pathogenic bacteria. Eurasia J Bio. sci. 2020; 14: 7297–7300. [Google Scholar].
14. Walencka E, Wieckowska-Szakiel M, Rozalska S, Sadowska B, Rozalska B. A surface-active agent from Saccharomyces cerevisiae influences staphylococcal adhesion and biofilm development. ZeitschriftFürNaturforschung C.,

2007; 62(5–6): 433–438. doi: 10.1515/znc-2007-5-618. [PubMed] [CrossRef] [Google Scholar].

15. Rasheed H. GH..andHaydar N. H. Purification, characterization and evaluation of the biological activity ofmannoproteinproduced from Saccharomycescerevisiaeby. Iraqi Journal of Agricultural Sciences, 2023; 54(2): 347- 359.
16. Ghada SM, Hayder NH and Mahmood M S. Synergistic effect of biosurfactant and prodigiosin produced by Serratiamarcescens as antimicrobial agent, 2018; 6(2): 1601-1615.
17. Elzbieta W, Marzena WS, SylwiaR,Beata S. and Barbara R. A Surface-Active Agent from Saccharomyces cerevisiae Influences Staphylococcal Adhesion and Biofilm Development. Z. Naturforsch.. journal ZeitschriftfürNaturforschung C2007; 62c: 433 - 438.
18. Perez-Sotelo LS, Talavera-Rojas M, Monroy-Salazar HG, Lagunas-Bernabé S, Cuarón-Ibargüengoytia JA, de Montes Oca Jiménez R, Vázquez-Chagoyán JC. In vitro evaluation of the binding capacity of Saccharomycescerevisiae Sc47 to adhere to the wall of Salmonella spp. Rev LatinoamMicrobiol., 2005; 47(3-4): 70–75. [PubMed] [Google Scholar].
19. Valarikova J, Cízova A, Rackova L, Bystricky S. Anti-staphylococcal activity of quaternizedmannan from the yeast *Candida albicans*. CarbohydrPolym., 2020; 240(15): 116288. doi: 10.1016/j.carbpol.2020.116288. [PubMed] [CrossRef] [Google Scholar].