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The Influence of Protein Loading in Emptied Yeast on its Bactericidal and Anticancer Effectiveness

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ABSTRACT

While yeasts were employed for delivery of drugs, active RNA or DNA, they were in minor cases utilized in delivery of certain antigenic proteins. The loading of bioeffective proteins inside emptied cells can preserve their structure and bioefficiency. In the present work, loaded lactoferrin inside emptied yeast was assayed for bactericidal and anticancer effectiveness after loading and compared with free protein. Its bactericidal efficiency was assayed against *S. aureus, S. typhi, K. pneumonia, S. sonnei, P. vulgaris, S. marcescens*, and *E. coli*. Its cytotoxicity was checked on human skin fibroblast (HSF) and potential anticancer impact on epidermoid skin carcinoma of human (A-431). *S. aureus, S. sonnei*, and *E. coli* were the most susceptible to loaded protein (concentration of minimum inhibition (MIC) of 0.65 mg/ml), but K. pneumonia and P. vulgaris were the least susceptible ones having MIC equal to 2.6 mg/ml. The MIC for loaded protein remained the same against K. pneumonia compared to free protein. Conversely, MIC for loaded protein amplified twice against *S. aureus, S. typhi, S. sonnei, S. marcescens*, and *E. coli*, and amplified four times in case of *P. vulgaris*. IC50 of loaded protein on A-431 was >1.25 mg/ml, and on HSF was 2.77 mg/ml. These outcomes pointed out the drop in bactericidal potency for loaded protein inside emptied yeast against six of seven tested pathogens related to free protein. Moreover, the toxicity was comparable on both normal (HSF) and anticancer cells (A-431).

Keywords: Loaded Protein; Emptied Yeast; Bactericidal; Anticancer; Bioeffective Proteins

Abbreviations: A-431: Human Epidermoid Skin Carcinoma; FBS: Fetal Bovine Serum; HSF: Human Skin Fibroblast; MIC: Concentration of Minimum Inhibition

Introduction

Since the eighties of 19 century, proteins have gained extensive acceptance as drugs, with insulin as a unique model [1,2]. Protein therapies or therapeutic candidates include either purified proteins from natural sources or recombinant ones for instance hormones, enzymes, antibodies, cytokines, vaccines from protein subunits, etc. [3-5]. However, their delivery remains a hot topic for research [6]. Lactoferrin as therapeutic candidate was earlier verified [7]. Its bactericidal potency was showed to be based on binding to various sites on cell surfaces of bacteria [8-12]. Its anticancer effectiveness was also verified [7,13,14]. Lactoferrin does its anticancer potential via provoking caspase-1 as well as IL-18, triggering CD8+ and CD4+, activating natural killer and IFN-γ T cells, hindering angiogenesis, and inducing apoptosis [15]. Lactoferrin had the capacity for constraining or motivating division of cells, reliant on whether its impact intended for healthy or cancer cells [16,17]. It was also found to affect cells that produce melanin resulting in approximately twenty reduction percent in pigmentation. It was immersed transdermally conquering production of melanin [18]. Its recombinant form could trigger propagation and migration of fibroblasts, and keep their survival [19]. Yeasts (*S. cerevisiae*) are profitable for carrying drugs owing to their safety and cost effectiveness. Furthermore, they are cultivable lacking whichever extra costs. Also, phospholipids in their membranes behave in a similar way to liposomes and thus could encapsulate various molecules [20-23].

Their thick wall containing glucan, mannoprotein layer, and chitin (only minor quantity) made them a type of continuous discharge system for delivery of drugs [24]. Yeast was formerly chemically emptied

from all of its contents [25]. Drugs like berberine and gossypol acetic acid were introduced into yeast cells for their delivery [26,27]. While, delivery of certain antigenic proteins was also reported for yeast [28]. In an earlier study (under publication), lactoferrin derived from milk of camel was introduced into emptied yeast. The present work was conducted to examine the influence of protein loading inside emptied yeast on its bactericidal and anticancer effectiveness.

Material and Methods

Introduction of Chloramphenicol into Emptied Yeast

Chloramphenicol (Bioshop, Canada) was dissolved at concentration 50 µg/ml in absolute ethanol, and filter sterilized. Emptied yeast was added to 2 ml of chloramphenicol, let at room temperature for half hour, followed by evaporating ethanol. Chloramphenicol in emptied yeast was used as bactericidal standard. Protein (lactoferrin derived from milk of camel) previously loaded inside emptied yeast (under publication) was involved in the coming assays.

Assessment of Chloramphenicol into Emptied Yeast

To conclude the quantity of taken chloramphenicol dissolved in ethanol by emptied yeast, a microscopic glass slide was weighted and emptied yeast was prepared as a slide smear without heating during fixation. As a substitute, cells were left to dry at 37 °C. After drying, slide was weighted again to calculate the smear weight. About 500 µl of chloramphenicol (50 µg/ml dissolved in ethanol) was added on top of the smear. The slide was left to enable the cells to take the drug for 20 min, and then the slide was dried again. After washing the slide with distilled water to get rid of any excess drug (outside emptied yeast), the slide was dried again at 37 °C and weighed. The amount of chloramphenicol contained within the cells was calculated from the difference in smear weight before and after drug addition.

Bactericidal Assessment for Loaded Lactoferrin Inside Emptied Yeast

The utilized bacterial pathogens in broth microdilution check to value bactericidal efficiency of loaded lactoferrin/chloramphenicol inside emptied yeast include *Staphylococcus aureus* ATCC 25923, *Salmonella typhi* ATCC 19430, *Klebsiella pneumonia, Shigella sonnei* ATCC 25931, Proteus vulgaris, Serratia marcescens, and Escherichia coli ATCC 25922. All of which incubated overnight at 37 °C in LB broth. To measure bactericidal efficiency of loaded lactoferrin/chloramphenicol inside emptied yeast and assess their MIC, broth microdilution was done. Dilutions (serial) were done at two-fold from loaded protein (starting with 5.2, and reaching 0.325 mg/ml) and added to plates of bacteria. Additionally, the same was done for loaded chloramphenicol (starting 20, and reaching 1.25 µg/ml). After 12 h incubation at 37 °C, growth was assessed. Test was carried out in triplicates [29].

Cultures of Skin Cells

HSF and A-431 have been gotten from Nawah Scientific Inc. (Cairo, Egypt). Cells maintenance was carried out at 37°C in DMEM supplemented media (10% of heat inactivated fetal bovine serum; FBS) in humidified, 5% (v/v) CO_2 atmosphere.

Cytotoxicity and Anti-Carcinogenicity Assays

Standard MTT test has been conducted for cell viability estimation for HSF and anti-carcinogenicity for A-431 [30,31]. Aliquots of cells suspension (100 µL at 5x 10^3 cells) were loaded in plates and incubated in complete DMEM media for 24 h. Cells were treated with different concentrations of Cisplatin (standard drug) and loaded protein inside emptied yeast. Following 48 h of exposure, medium was removed and MTT introduced. The released formazan was detected with DMSO. The absorbance was valued at ʎmax 570 nm.

Results and Discussion

Bioeffective proteins are interesting candidates that can be applied in different applications [3-5]. Some of them even enclose short peptide(s) that add to their functionality in a similar pathway or different one. Lactoferrin is one of those proteins that is extensively consumed for different purposes. This protein is a scavenger for so many and variable activities beneficial for us [7-15]. The fascinating thing is that lactoferrin is already naturally produced within our secretions. In the present work, lactoferrin derived from milk of camel previously introduced into emptied yeast (under publication) was analyzed to examine the influence of protein loading inside emptied yeast on its bactericidal and anticancer effectiveness. Bactericidal efficiency of loaded protein was checked against seven bacterial pathogens. Its cytotoxicity on HSF and anticancer efficacy on A-431 were also assessed.

Assessment of Chloramphenicol into Emptied Yeast

The amount of chloramphenicol contained within emptied yeast was about 20 µg.

Bactericidal Assessment for Loaded Lactoferrin Inside Emptied Yeast

In view of Table 1 and presented MIC, *S. aureus, S. sonnei*, and *E. coli* were the most susceptible to loaded protein (MIC of 0.65 mg/ml), but *K. pneumonia* and *P. vulgaris* were the least susceptible ones having MIC equal to 2.6 mg/ml. The MIC for loaded protein remained the same against *K. pneumonia* compared to free protein. Conversely, MIC for loaded protein amplified twice against *S. aureus, S. typhi, S. sonnei, S. marcescens*, and *E. coli,* and amplified four times in case of *P. vulgaris*. This drop in bactericidal potency may be caused by slow freeing of the loaded protein or may be its binding to yeast surface.

Table 1: Bactericidal efficacy for loaded lactoferrin inside emptied yeast.

Strain	MIC			
	Free Chloramphenicol (µg/ml)	Free lactoferrin derived from milk of camel (mg/ml)	Loaded chloramphenicol inside emptied yeast (µg/ml)	Loaded lactoferrin inside emptied yeast (mg/ml)
S. aureus	1.25	0.32	5	0.65
S. typhi	2.5	0.65	10	1.3
K. pneumonia	5	2.6	20	2.6
S. sonnei	2.5	0.32	10	0.65
P. vulgaris	5	0.65	20	2.6
S. marcescens	5	0.65	10	1.3
E. coli	2.5	0.32	5	0.65

Cytotoxicity and Anti-Carcinogenicity Assays

A normal cell line (HSF) was used to investigate the toxicity of loaded protein and A-431 cancer cells were for anticancer evaluation. Both cell types were treated using Cisplatin/standard anticancer drug (0.03-300 µg/ml) and loaded protein inside emptied yeast (0.004-1.25 mg/ml). The aim was to investigate the impact of protein loading on its anticancer efficacy on skin cancer cells. Figure 1, plates showed clearly that Cisplatin behave differently on normal than cancer cells, yet loaded protein behaved nearly the same way especially at concentration of 1.25 mg/ml it even caused more toxicity to healthy cells. The MTT data for HSF are in Tables 2 & 3. While, those for A-431 are showed in Tables 4 & 5. The LC_{50} details on both cell lines were calculated as in Figures 2 & 3. The findings displayed that the cytotoxicity against the normal cell line under the experimental condition for Cisplatein was 4 µg/ml, while for loaded protein it was 2.77 mg/ml. The anti-carcinogenicity for Cisplatin on cancer cells was 5.24 µg / ml, while for the loaded protein it was >1.25 mg/ml. The above-mentioned result indicated that the loaded protein under the experimental conditions is highly toxic for cancer as well as healthy cells. In contrast, Cisplatin exhibited the standard criteria of the antitumor compound and proved to be selective. These outcomes differ significantly from a former study that applied free lactoferrin (non-loaded).

Table 2: The viability % of HSF after treatment with various Cisplatin concentrations.

Cisplatin (µg/ml)	Raw data			Blank Corrected Data			Viability %			Mean	STD
	1	2	3	1	2	3	1	2	3		
Control	1.0345	1.0351	1.0281	0.99907	0.99967	0.99267	100	100	100	100	0
0.03	0.9371	0.95412	0.9474	0.90167	0.91869	0.91197	90.4259	92.1328	91.4588	91.3392	0.701956
0.1	0.9151	0.9225	0.9215	0.87967	0.88707	0.88607	88.2196	88.9617	88.8614	88.6809	0.328764
0.3	0.9232	0.9181	0.9102	0.88777	0.88267	0.87477	89.0319	88.5204	87.7282	88.4268	0.536348
1	0.8193	0.8157	0.8867	0.78387	0.78027	0.85127	78.612	78.251	85.3714	80.7448	3.274816
3	0.7731	0.7251	0.7569	0.73767	0.68967	0.72147	73.9787	69.1649	72.3541	71.8326	1.999522
10	0.1084	0.1101	0.1236	0.07207	0.07377	0.08727	7.23391	7.40456	8.75966	7.79938	0.682587
30	0.0783	0.0801	0.0836	0.04197	0.04377	0.04727	4.21253	4.39321	4.74454	4.4501	0.220883
100	0.0725	0.0679	0.0725	0.03617	0.03157	0.03617	3.63034	3.1686	3.63034	3.47643	0.217666
300	0.061	0.0618	0.0585	0.02467	0.02547	0.02217	2.47599	2.5563	2.22505	2.41911	0.141086
Blank	0.0344	0.038	0.0366	Blank Average		0.03633	Control average		0.99623		

Note: Control: untreated cells, Blank: DMSO only

Table 3: The viability % of HSF after treatment with various loaded protein concentrations.

Loaded protein (mg/ml)	Raw data			Blank Corrected Data			Viability %			Mean	STD
	1	2	3	1	2	3	1	2	3		
Control	1.0049	1.0331	1.0214	0.9702	0.9984	0.9867	100	100	100	100	0
0.004	1.0222	1.0321	1.0114	0.9875	0.9974	0.9767	100.244	101.249	99.1473	100.213	0.858126
0.009	1.0104	0.9515	0.9321	0.9757	0.9168	0.8974	99.0458	93.0667	91.0974	94.4033	3.379766
0.019	0.9983	0.9155	0.9501	0.9636	0.8808	0.9154	97.8175	89.4122	92.9246	93.3848	3.446819
0.039	0.9273	0.9232	0.8915	0.8926	0.8885	0.8568	90.6101	90.1939	86.9759	89.26	1.623968
0.078	0.9043	0.9131	0.8879	0.8696	0.8784	0.8532	88.2753	89.1686	86.6105	88.0181	1.06006
0.15	0.8998	0.8747	0.8778	0.86493	0.83983	0.84293	87.8164	85.268	85.5828	86.2224	1.13444
0.3125	0.8554	0.8523	0.8657	0.82053	0.81743	0.83083	83.3085	82.9938	84.3543	83.5522	0.581533
0.625	0.7828	0.8579	0.8583	0.74793	0.82303	0.82343	75.9375	83.5623	83.603	81.0342	3.604014
1.25	0.5754	0.6062	0.5321	0.54053	0.57133	0.49723	54.8802	58.0073	50.484	54.4572	3.085928
Blank	0.0342	0.0352	0.0352	Blank Average		0.03487	Control average		0.98493		

Note: Control: untreated cells, Blank: DMSO only

Table 4: The viability % of A-431 after treatment with various Cisplatin concentrations.

Cisplatin (µg/ml)	Raw data			Blank Corrected Data			Viability %			Mean	STD
	1	2	3	1	2	3	1	2	3		
Control	1.137	1.1679	1.2224	1.10237	1.13327	1.18777	100	100	100	100	0
0.03	1.2211	1.1985	1.207	1.18647	1.16387	1.17237	103.973	101.992	102.737	102.901	0.816762
0.1	1.2032	1.1621	1.2401	1.16857	1.12747	1.20547	102.404	98.8024	105.638	102.281	2.791852
0.3	1.1114	1.2012	1.1665	1.07677	1.16657	1.13187	94.3594	102.229	99.1879	98.592	3.240171
1	1.1551	1.1635	1.1547	1.12047	1.12887	1.12007	98.1889	98.925	98.1539	98.4226	0.355556
3	1.0239	1.0841	1.0998	0.98927	1.04947	1.06517	86.6916	91.9671	93.3429	90.6672	2.866723
10	0.1169	0.1254	0.1663	0.08227	0.09077	0.13167	7.20921	7.95408	11.5382	8.90051	1.889783
30	0.0633	0.066	0.0707	0.02867	0.03137	0.03607	2.51212	2.74873	3.1606	2.80715	0.267944
100	0.0543	0.0592	0.0597	0.01967	0.02457	0.02507	1.72343	2.15283	2.19665	2.0243	0.213498
300	0.0516	0.0556	0.0568	0.01697	0.02097	0.02217	1.48683	1.83735	1.94251	1.75556	0.194816
Blank	0.0342	0.035	0.0347	Blank average		0.03463	Control average		1.14113		

Note: Control: untreated cells, Blank: DMSO only

Table 5: The viability % of A-431 after treatment with various loaded protein concentrations.

Loaded protein (mg/ml)	Raw data			Blank Corrected Data			Viability %			Mean	STD
	1	2	3	1	2	3	1	2	3		
Control	1.3444	1.3731	1.2332	1.30977	1.33847	1.19857	100	100	100	100	0
0.004	1.3321	1.3102	1.3215	1.29747	1.27557	1.28687	101.185	99.4775	100.359	100.341	0.697371
0.009	1.2898	1.281	1.3121	1.25517	1.24637	1.27747	97.8866	97.2003	99.6257	98.2375	1.020785
0.019	1.2362	1.2614	1.2854	1.20157	1.22677	1.25077	93.7065	95.6717	97.5434	95.6405	1.566586
0.039	1.2484	1.2321	1.2211	1.21377	1.19747	1.18647	94.6579	93.3867	92.5289	93.5245	0.874621
0.078	1.2064	1.2023	1.2015	1.17177	1.16767	1.16687	91.3824	91.0627	91.0003	91.1485	0.167384
0.15	1.2032	1.1995	1.1021	1.16857	1.16487	1.06747	91.1329	90.8443	83.2484	88.4085	3.650666
0.3125	1.0951	1.0537	1.1177	1.06047	1.01907	1.08307	82.7025	79.4738	84.465	82.2138	2.06673
0.625	0.9998	1.0858	1.0141	0.96517	1.05117	0.97947	75.2704	81.9772	76.3856	77.8777	2.934328
1.25	1.0115	1.0321	1.0412	0.97687	0.99747	1.00657	76.1828	77.7893	78.499	77.4904	0.96893
Blank	0.0342	0.035	0.0347	Blank average		0.03463	Control average		1.28227		

Note: Control: untreated cells, Blank: DMSO only

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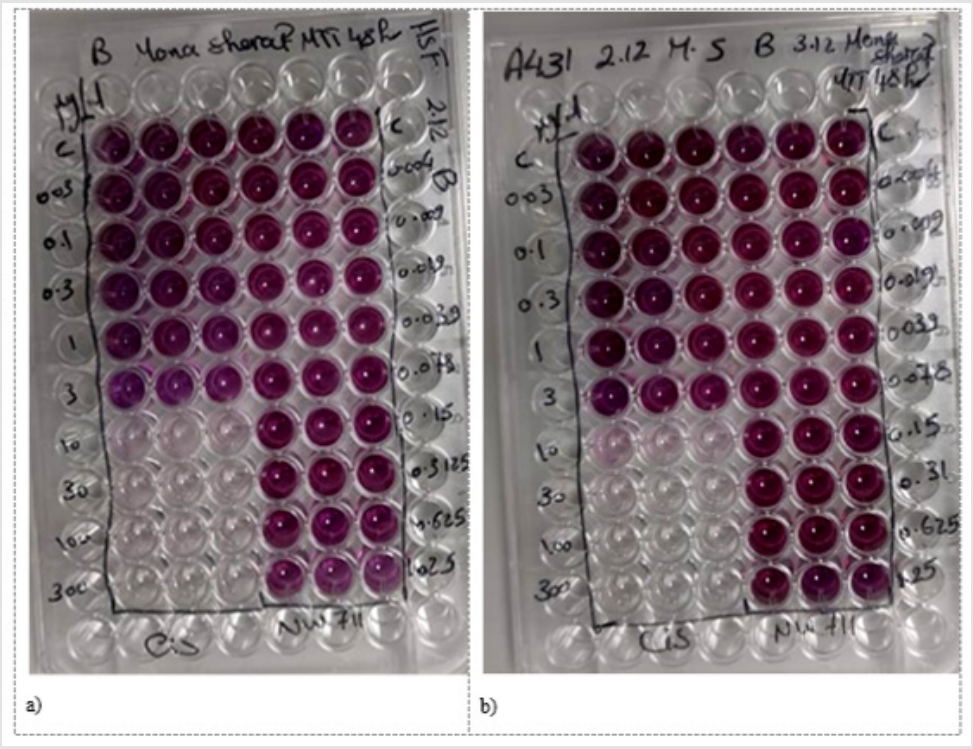
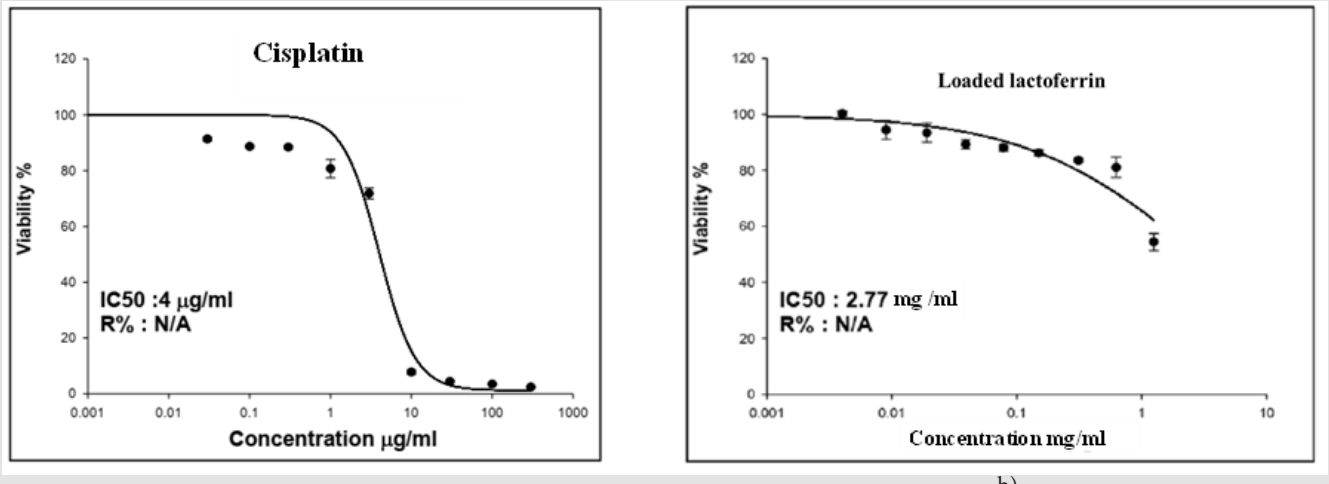


Figure 1:
a) Multi-well plate of normal HSF cells treated by Cisplatin (0.03-300 µg/ml) and loaded protein inside emptied yeast (0.004-1.25 mg/ml),
b) Multi-well plate of cancer A-431 cells treated by Cisplatin (0.03-300 µg/ml) and loaded protein inside emptied yeast (0.004-1.25 mg/ml).



a) b)

Figure 2:
a) LC_{50} of Cisplatin on HSF,
b) LC_{50} of loaded protein on HSF.

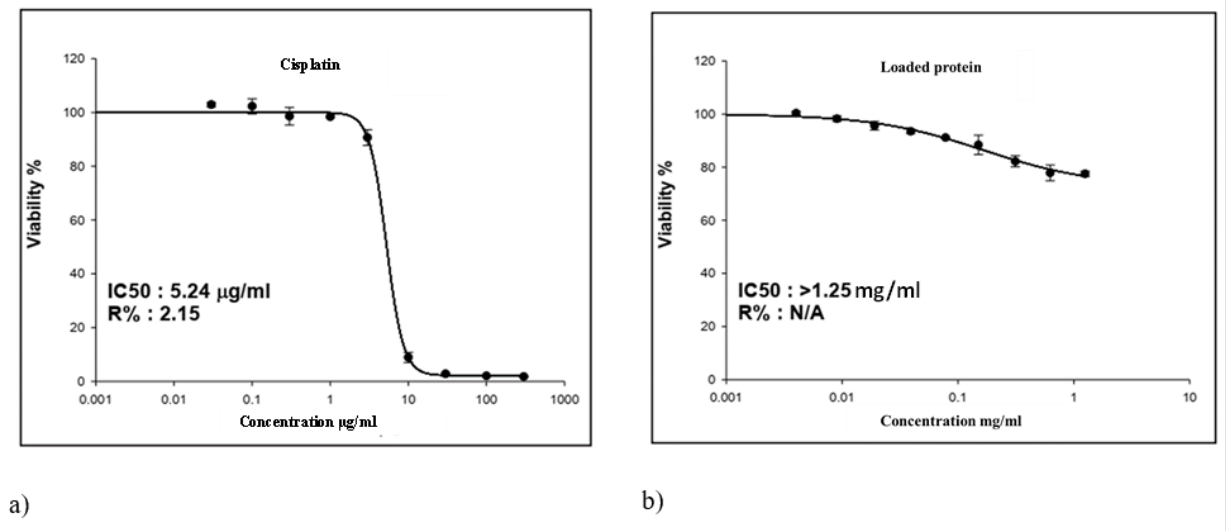


Figure 3:
a) LC_{50} of Cisplatin on A-431,
b) LC_{50} of loaded protein on A-431.

The obtained data of the free protein demonstrated that the concentration of free protein that spared the life of all WI-38 cells (Human normal cells) was 425.2 µg/ml while the concentration that could kill 50% of cancer cells; HepG-2, Caco-2, MCF-7 and Hela were 1011, 2127, 1229, 1352 µg/ml respectively, which means that free protein could selectively kill cancer cells [14]. One could also conclude that the loaded protein in emptied yeast apparently enables a higher concentration around the yeast cells before full dissociation in the surrounding medium. That might explain its high toxicity on both of normal and cancer cells. More investigations are needed to adjust the release of the loaded protein by yeast and the best conditions that can readjust its toxicity based on its concentration in certain volume during the start point of the release till the full re-evacuation of the loaded protein from emptied yeast.

Conclusion

The influence of protein loading in emptied yeast was unfortunately negative on its bactericidal and anticancer effectiveness. That might indicate a controlled freeing of the protein from emptied yeast in case of bacterial control. The high toxicity in case of cell lines treatment can be linked to a higher concentration around the yeast cells before full dissociation in the surrounding medium. More investigations are needed.

Conflicts of Interest

The authors declare no conflict of interest.

References

1. Johnson IS (1983) Human insulin from recombinant DNA technology. Science 219(4585): 632-637.
2. Walsh J (2004) Second generation biopharmaceuticals. Eur J Pharma Biopharma 58(2): 185-196.
3. Gerngross TU (2004) Advances in the production of human therapeutic proteins in yeasts and filamentous fungi. Nat Biotechnol 22(11): 1409-1414.
4. Dimitrov DS (2012) Therapeutic proteins. Methods Mol Biol 899: 1-26.
5. Ebrahimi SB, Samanta D (2023) Engineering protein-based therapeutics through structural and chemical design. Nat Commun 14: 2411.
6. Bruno BJ, Miller GD, Lim CS (2013) Basics and recent advances in peptide and protein drug delivery. Ther Deliv 4(11): 1443-1167.
7. Abuelgasim KA, Alsharhan Y, Alenzi T, Alhazzani A, Ali YZ, et al. (2018) The use of complementary and alternative medicine by patients with cancer: a cross sectional survey in Saudi Arabia. BMC Complementary Medicine and Therapies 18: 88.
8. Arnold RR, Brewer M, Gauthier JJ (1980) Bactericidal activity of human lactoferrin: sensitivity of a variety of microorganisms. Infection and Immunity 28(3): 893-898.
9. Ellison III RT, LaForce FM, Giehl TJ, Boose DS, Dunn BE, et al. (1990) Lactoferrin and transferrin damage of the gram-negative outer membrane is modulated by Ca2+ and Mg2+. The Journal of General Microbiology 136(7): 1437-1446.
10. Appelmelk BJ, An YQ, Geerts M, Thijs BG, de Boer HA, et al. (1994) Lactoferrin is a lipid A-binding protein. Infection and Immunity 62(6): 2628-2632.

11. Drago-Serrano ME, dela Garza-Amaya M, Luna JS, Campos-Rodríguez R (2012) Lactoferrin-lipopolysaccharide (LPS) binding as key to antibacterial and antiendotoxic effects. International Immunopharmacology 12(1): 1-9.
12. Drago-Serrano ME, Campos-Rodríguez R, CØsar Carrero J, Garza Mdela (2017) Lactoferrin: balancing ups and downs of inflammation due to microbial infections. International Journal of Molecular Sciences 18(3): 501.
13. Habib HM, Ibrahim WH, Schneider-Stock R, Hassan HM (2013) Camel milk lactoferrin reduces the proliferation of colorectal cancer cells and exerts antioxidant and DNA damage inhibitory activities. Food Chemistry 141(1): 148-152.
14. EL Baky NA, Abu Serie MM, Redwan EM (2021) De novo designed lactoferrin–oleic acid-loaded chitosan nanoparticles with improved activity and selectivity toward four human cancer cells as compared to conventional complexes. Journal of Applied Pharmaceutical Science 11(3): 60-70.
15. Ramírez Rico G, Drago Serrano ME, León Sicairos N, de la Garza M (2022) Lactoferrin: A Nutraceutical with Activity against Colorectal Cancer. Front Pharmacol 13: 855852.
16. Eliassen LT, Berge G, Sveinbjornsson B, Svendsen JS, Vorland LH, et al. (2002) Evidence for a direct antitumor mechanism of action of bovine lactoferricin. Anticancer Res 22(5): 2703-2710.
17. Spadaro M, Caorsi C, Ceruti P, Varadhachary A, Forni G, et al. (2008) Lactoferrin, a major defense protein of innate immunity, is a novel maturation factor for human dendritic cells. FASEB J 22(8): 2747-2757.
18. Ishii N, Ryu M, Suzuki YA (2017) Lactoferrin inhibits melanogenesis by down-regulating MITF in melanoma cells and normal melanocytes. Biochem Cell Biol 95(1): 119-125.
19. Hassoun LA, Sivamani RK (2017) A systematic review of lactoferrin use in dermatology. Crit Rev Food Sci Nutr 57(17): 3632-3639.
20. Bishop JRP, Nelson G, Lamb J (1998) Microencapsulation in yeast cells. J Microencapsul 15(6): 761-773.
21. Chow C, Palecet P (2004) Enzyme encapsulation in permeabilized Saccharomyces cerevisiae cells. Biotechnol Progr 20(2): 449-456.
22. Shi G, Rao L, Yu H, Xiang H, Pen G, et al. (2007) Yeast-cell based microencapsulation of chlorogenic acid as a water-soluble antioxidant. J Food Eng 80(4): 1060-1067.
23. Shi G, Rao L, Yu H, Xiang H, Yang H, et al. (2008) Stabilization of photosensitive resveratrol within yeast cell. Int J Pharmaceut 349: 83-93.
24. De Nobel JG, Klis FM, Munnik T, Priem J, Van Den Ende H, et al. (1990) An assay of the relative cell wall porosity of Saccharomyces cerevisiae, Kluveromyces lactis and Schizosaccharomyces pombe. Yeast 6(6): 483-490.
25. Amara AA (2015) Saccharomyces cerevisiae ghosts using the Sponge-Like Re-Reduced protocol. SOJ Biochem 1(1): 4.
26. Salari R, Bazzaz BSF, Rajabi O, Khashyarmanesh Z (2013) New aspects of Saccharomyces cerevisiae as a novel carrier for berberine. DARU J Pharm Sci 21(1): 73.
27. Amara AA (2015) Bacterial and yeast ghosts: E. coli and Saccharomyces cerevisiae preparation as drug delivery model. Int Sci Invest J 4(7): 11-22.
28. Tan Y, Chen L, Li K, Lou B, Liu Y, et al. (2022) Yeast as carrier for drug delivery and vaccine construction. J Control Release 346: 358-379.
29. Almehdar HA, El-Baky NA, Alhaider AA, Almuhaideb SA, Alhaider AA, et al. (2020) Bacteriostatic and bactericidal activities of camel lactoferrins against Salmonella enterica Serovar Typhi. Probiotics & Antimicro 12: 18-31.
30. Mosmann T (1983) Rapid colourimetric assay for cellular growth and survival: application to proliferation and cytotoxicity assays. Journal of immunological methods 65(1-2): 55-63.
31. Ahmed AHH, Mohamed MFA, Allam RM, Nafady A, Mohamed SK, et al. (2022) Design, synthesis, and molecular docking of novel pyrazole-chalcone analogs of lonazolac as 5-LOX, iNOS and tubulin polymerization inhibitors with potential anticancer and anti-inflammatory activities. Bioorg Chem 129: 106171.

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