DRF-390
[1-7]

RESEARCH ARTICLE Asian Journal of Dairy and Food Research

Possibility of Concentrating Milk Enriched with Lemongrass (Cymbopogon) as a Preservative using Ultrafiltration Technology

Hamdia Al-Hamdani[1], Amer Alnaemi[2], Azhar Getan[2]

10.18805/ajdfr.DRF-390

ABSTRACT

Background: This study aimed at the possibility of concentrating milk in the laboratory using ultra-filtration technology instead of the traditional evaporation method that is still used in Iraq.

Methods: Therefore, 5 litres of raw, cow milk were taken from a reliable source and two different concentrations of lemongrass were added as a preservative to observe the changes occurring during concentration, chemically and microbial. The best conditions were found to concentrate the milk 2.7 once and improve it over many days with the addition of lemongrass extract as a preservative. A material with a flat membrane polyethyle-solphen with a measurement of 10 kdalton (Snyder USA) was selected. Throughout the duration of the more than three-hour experiment, the measurements were taken. Two sets of data were examined: in the first set, the flow rate was varied between 0.15 and 0.25 L/min while the trans membrane pressure (TMP) was kept constant at 2 bar. The value of 0.15 L/min was determined to be the ideal flux. In the second group, the trans membrane pressure was varied between 0.5 and 1,2 bar while the flow rate was kept constant at 0.15 L/min. At TMP 2 bar, the ideal flux value was discovered at 35°C, the permeate flux was around 5-7 L/m2.hr.

Result: It was found the change of the total solid and protein were excellent percentages with time of filtration. The total number of bacteria at both concentrations decreased significantly ($p<0.05$) in the biological test results.

Key words: Lemongrass, Pre-concentrated milk, Ultra-filtration technology.

INTRODUCTION

Membrane technology, which has been introduced in the dairy industry over the past thirty years, is one of the more advanced technologies (Agarwal *et al*., 2015). It is preferred for many industrial processes that require the separation or concentration of products without the use of heat, particularly in the cheese industry and the fractionation of milk components (Arunkumar and Etzel, 2015).

Ultrafiltration membranes are semi-permeable membranes that prevent the passage of substances bigger than 30 nanometers, which is the basis for this method of concentrating milk. This spectrum encompasses most proteins. The separated fine components have molecular weights between 1 and 1000 kdalton and molecular sizes between 0.01 and 0.02 µm. The filtered milk is often separated into two portions: permeate, which comprises water, lactose, dissolved salts, NPN and dissolved vitamins and retentat, which contains proteins, lipids and dissolved salts. The liquid is typically forced to the surface under low pressure (Arunkumar and Etzel, 2018). The majority of industrialized nations, even those that are adjacent, now employ ultrafiltration technology more frequently in the production of different dairy products (Agarwal *et al*., 2015). This may be attributed to the several benefits this technology offers, chief among them being enhanced production and improved nutritional value (Muehlhoff *et al*., 2013). Low molecular weight (MWCP) milk protein concentrate (MPC) with a range of 10 kDa to 30 kDa is produced using ultrafiltration membrane technology. The membranes hold the proteins in place while allowing lactose and extremely minute particles to pass through Arunkumar and Etzel, (2015).

About 8.6% of total solids are found in cow milk; 3.2% are proteins; 5.4% are ash, non-protein nitrogen (NPN) and other tiny molecules; and the remaining 91.4% is water. Casein and whey proteins, which range in molecular mass from 14.4 to 150 kDa, make up the majority of milk proteins (Arunkumar *et al*., 2016).

One issue with low-pressure membrane technology is fouling, which compromises the precision and efficiency of the filtration process (Salunke *et al*., 2021). The buildup of particles that need to be separated, including dirt or proteins, on the surface or within the pores causes the treatment process to take longer. This increases energy and cleaning costs, decreases separation efficiency, or even totally clogs the pores (Muhammad *et al*., 2018).

[1]Market Research and Consumer Protection Center/Univ. of Baghdad/Baghdad/Iraq.
[2]Ministry of Science and Technology. Directorate of Environment and Water/Baghdad/Iraq.

Corresponding Author: Hamdia Al-Hamdani, Market Research and Consumer Protection Center/Univ. of Baghdad/Baghdad/Iraq. Email: cioffi16@yahoo.com

How to cite this article: Al-Hamdani, H., Alnaemi, A. and Getan, A. (2024). Possibility of Concentrating Milk Enriched with Lemongrass (Cymbopogon) as a Preservative using Ultrafiltration Technology. Asian Journal of Dairy and Food Research. DOI: 10.18805/ajdfr.DRF-390.

Submitted: 29-02-2024 **Accepted:** 21-05-2024 **Online:** 24-07-2024

On the other hand, the accumulation of particles that create a thin layer on the membrane's surface over the first few minute's leads to the steady, balanced production. Because of electrostatic interactions on the surface, the flux steadily decreases and remains constant throughout the filtration process until the membrane stops filtering owing to a phenomena concentration polarization, necessitating washing as (Judd and Carra, 2021).

In an attempt to solve this issue, producers hurried to develop membranes that are lower pollutant and more selective in their manufacturing process Prestes *et al*. (2022), as well as regular cleaning systems, transverse speed control, uniform membrane pressure, the use of ultrasonic waves, ceramic membranes and other potential future fixes (Marella *et al*., 2013).

Lemongrass (*Cymbopogon citratus*) is native to Asia and in all Asian regions but now grows all over the world Sharma *et al*. (2022), for its subtle flavor in its fresh and dried leaves common to Asian cuisine and use in curries, tea, soups and milk, it is also suitable for cooking with poultry, seafood and fish (Wifek *et al*., 2016). And it was also used in folk medicine (Singh *et al*., 2011), where it was mixed with tea as a sedative and to treat various fever-related diseases. Additionally, it served as an immune stimulant in India, Egypt and Iran Amirdivani and (Baba, 2011; Tilaye *et al*., 2018). The antimicrobial activity of lemongrass is due to its high content of phytochemicals such as citrate and essential oils (Zulfa *et al*., 2016; Al-Hamdani, 2022). Currently, in our country, there are many cow breeders and an abundance of milk. We must investigate the possibility of filtering and concentrating the milk using internationally advanced techniques, such as ultrafiltration technology and its application with the addition of lemongrass to the milk as a preservative and its effect on the concentrated milk produced chemically and microbial.

MATERIALS AND METHODS

Five kilograms of raw cow's milk was brought from the College of Agriculture/University of Baghdad/Abu-Ghraib to the laboratory and placed in the refrigerator. Experimental testing was conducted after preparing the instruments in the laboratory of the Ministry of Science and Technology/ Environment and Water Department, using flat sheet ultra-filter membrane with 10 kdalton (Snyder USA) made from polyethylene-sulphone (PES) and surface area of 266 cm^2 and two percentage of lemongrass extract (100 mg/ml, 200 mg/ml) .

Equipment

To complete the experiment, the milk concentration process was carried out using Membrane testing device manufactured by the American company as shown in (Fig 1).

Stages of operating the system

The process was carried out to achieve a concentration approximately 2.7 times calculated based on the fat concentration, with an estimated retention rate of almost 99% and it took about a 3 hour and a half. For this study, five kilograms of fresh milk were used. It underwent pasteurization at 73°C for 15 minutes, followed by cooling to a temperature of 5°C. Subsequently, the milk was heated at a temperature of 35±2°C and a press by pumping it into the separation chamber (sepa), as illustrated in (Fig 1) the pressure and flow rate are monitored by recording the readings of the pressure gages and set the stopwatch to read the gradients of cylinder in which the permeate filter is collected respectively., Samples of 50 ml of milk were taken in sterile glass containers upon receipt, from the center and the filtrate during the concentration process every 15 minutes. Subsequently, the samples taken were subjected to chemical tests to determine the changes in composition between the milk and the concentrate, comparing them with the filtrate.

Analytical methods

Analysis was performed on milk, UF concentrated samples. The Bradford method was chosen to measure the percentage of protein concentration in milk, which is a spectrophotometric method based on the principle of shifting the absorbance of the blue dye under acidic conditions from brown to blue. The spectrophotometer has a wavelength of 595 nm and the standard curve was made, then the readings were taken for the different models and the protein concentration was measured compared to the standard solution according to Bradford (1976); Schiano *et al*. (2017).

The percentage of lactose in whey was determined using the colorimetric method, based on its sugar reduction ability in the presence of substances that aid reduction (copper sulfate solution, sodium tungsten solution, sulfuric acid, phenol and lactose standard solution). The absorbance was then measured at a wavelength of 490 nm, following the procedure outlined by (Hieda *et al*., 2018).

The resultant concentrated milk were microbiologically examined for total bacterial count, mold and yeasts count and coliform group according to American Public Health Association (Vinderola and Reinheimer, 2000).

RESULTS AND DISCUSSION

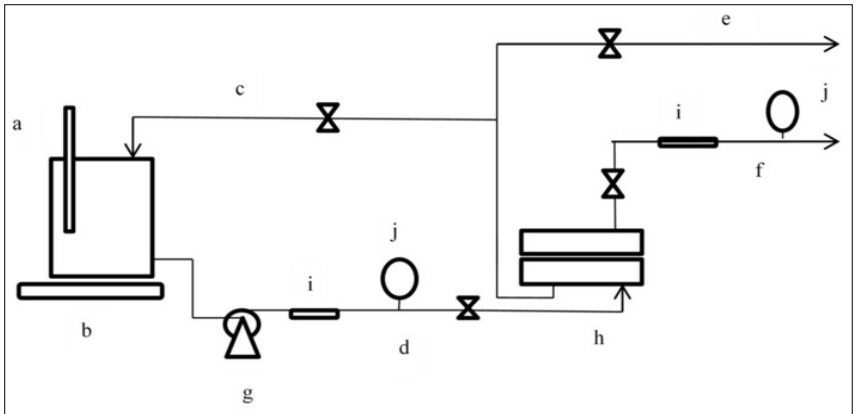
Ultra-filtration measuring

Fig 2 showed the scheme of working efficiency of the Ultra-filtration measuring on 10 kdalton to isolate milk protein in the form of concentrated milk and and exudes the lactose at pressure 2 bar.

Flow rate effect

Fig 3 represents the effect of changing the flow rate with constant transmembrane pressure at 2 bar and the values of the flow rates are (0.15,0.25 L/min). The graph shows the flow decreasing with time. This rapid decrease occurs at the beginning of filtration and slows down when

continuing, for both values. This is due to the decrease in the protein concentration at the boundary layer of the membrane, which leads to a decrease in the concentration polarization and with Continuing filtration, the protein is better preserved after approximately 25 minutes (Porter, 1972). The viscosity of the solution gradually increases due to the total solids it contains. This accumulation of dissolved materials spreads the protein molecules in the other direction, away from the membrane, because they spread from the higher concentration to the lower concentration. Fortunately, the protein concentration decreases from the surface of the membrane to the top of



1= Heat sensor, 2= Tank, 3= Recycle, 4= Flow meter, 5= Permeate, 6= Retentate, 7= Pump, 8= Sepa, 9= Flow meter, 10= Pressure gage.

Fig 1: A simplified piping and instruments flow chart to concentrate milk using the membrane testing system.

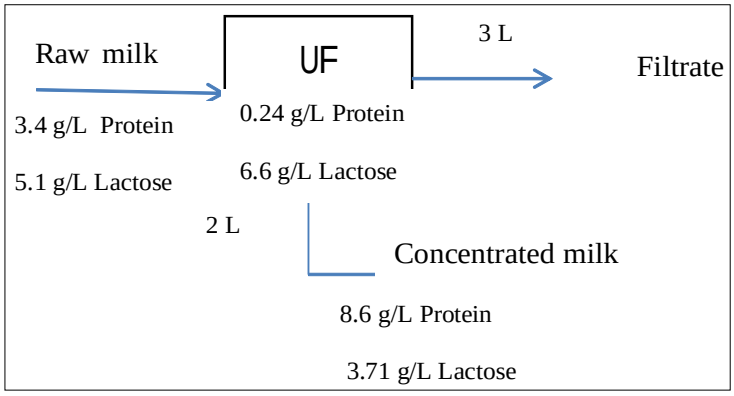


Fig 2: Scheme of Ultra-filtration measuring at 10 kdalton.

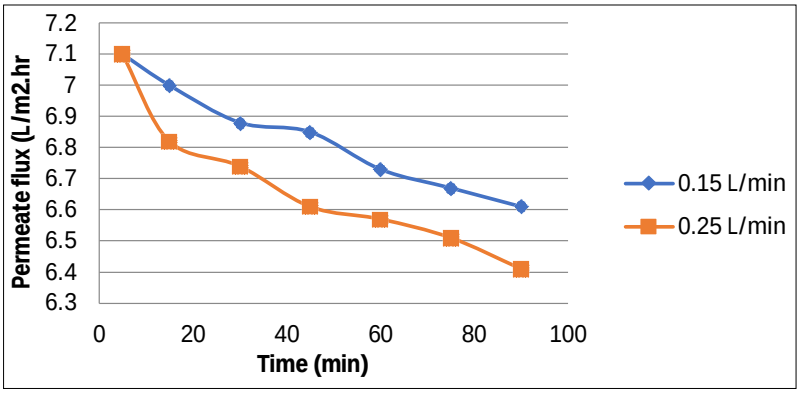


Fig 3: Change the flux with time of filtration at constant flow rate.

the boundary layer, its concentration is the same as the concentration of the bulk current through the membrane (Lipnizki, 2010).

Pressure effect

Fig 4, showed the filtration mechanism which can be followed by changing the pressure values (0.5, 1 and 2 bar) while maintaining a constant flow rate of filtration (0.15 L/min). The flux can be observed over time for the first 20 minutes and for all values due to the natural decrease in hydraulic pressure on the surface of the membrane, which leads to the precipitation of dissolved substances in the milk, forming a layer of cake, which causes an increase in Polarizes protein and total solids (Li *et al*., 2017). The small filter pores that do not allow the passage of protein molecules larger than 10 kDa such as α-lactalbumin and its molecular weight is 14 kDa β-lactoglobulin with a molecular weight of 18 kDa. This could be a result of the gradual reducing flux over the 90-minute filtration period (Ferrer *et al*., 2011).

Change in protein concentration over time

Fig 5 showed the change in protein concentration with time when the filtration conditions were fixed at the pressure and flow rate. Samples were taken every 15 minutes. From the graph, it can be seen that the percentage of protein coding in the milk increased from 3.2 mg/l to 8.1 mg/l and this increase can be monitored after 20 minutes of filtration (Meyer *et al*., 2015).

Change in total solids percentage with time

Fig 6 showed the change of percentage of total solids with time in minutes for milk concentration using ultrafiltration technique which was used in this study. Through the results, an increase in the percentage of total dry matter can be observed during the filtration process with a significant difference. The average rate at the beginning of the filtration process for raw milk reached 11%, while the average rate at the end of the filtration reached 24%. The reason is attributed to the presence of some dissolved components in the water such as mineral salts, lactose and others, which are included in the dry ingredients, which the membrane cannot seize. These results are similar to what was founded by (Moreno-Montoro *et al*., 2015).

Microbial assay

The contamination of membranes with microbes is a significant concern in the dairy industry, posing limitations on the use of membrane filtration. This includes issues such as pore blockage and the formation of a layer of various sediments and bacteria present in milk, commonly referred to as a 'cake.' These challenges can reduce efficiency and

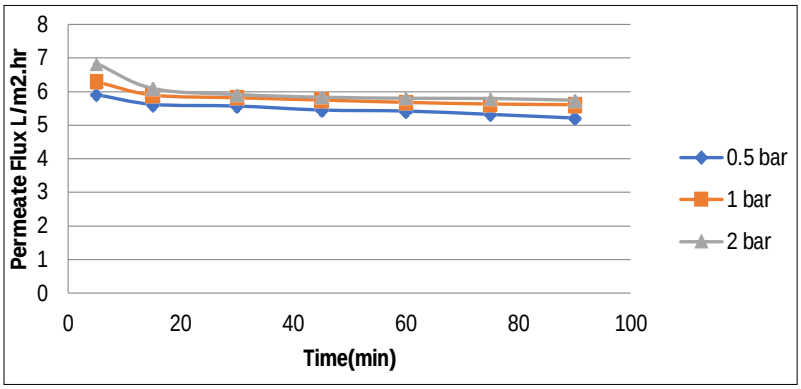


Fig 4: Change the flux with time at constant transmembrane pressure (TMB).

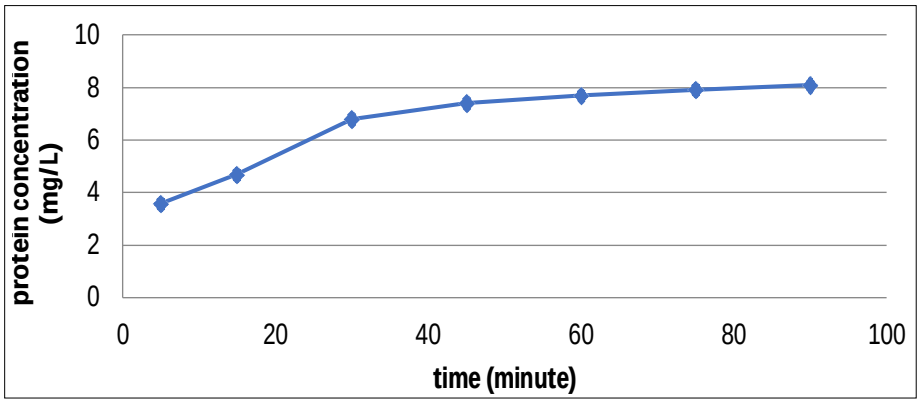


Fig 5: Change of protein in milk with time of filtration.

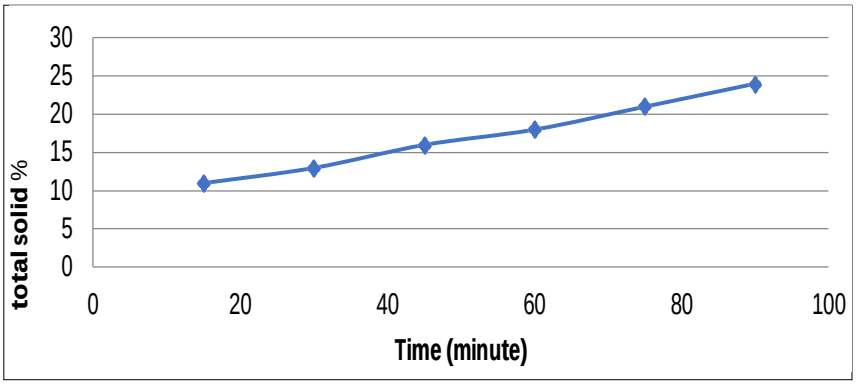


Fig 6: Change of total solids% with time of filtration.

Table 1: Total counts of bacteria in treated concentrated milk with lemongrass extraction.

		Time		P-value
Conc. of milk with lemon grass extraction		24 hr	48 hr	
	100 mg/L	90×10^1	62×10^3	67.127*
	200 mg/L	45×10^1	50×10^2	54.948*
P-value		35.073*	52.665*	---
	*(P≤0.05)			

hinder the preservation of milk after concentration for an extended period. To address these concerns, an extract of lemongrass was prepared with two concentrations: the first at 100 mg/L and the second at 200 mg/L and added to the milk concentrate. Both concentrations were cultured on standard media for the purpose of conducting a biological examination, The results of the current study on the total bacterial counts (T.C.) of concentrated and fortified milk at a concentration of 100,200 mg/L milk from lemongrass extract are represented in (Table 1).

The results of the study showed a highly significant decrease ($p \leq 0.05$) in the total bacterial counts for both concentrations, especially after 24 hours of storage in the refrigerator, while increased significantly and by a small percentage, the total number of bacteria for both concentrations of 100 and 200 mg/L became (62×10^3) and (50×10^2) respectively after 48 hours of storage (Knight-Jones *et al*., 2016), but this increase is much less than what was stipulated in the Iraqi standard (1/693, 1988). to the acceptable microbial limits in concentrated milk. The reason for the high decrease in bacterial numbers is attributed to several factors, including passing the concentrated milk through many heat treatments and the filtration used ultrafiltration for this study, which leads to the absence of microorganisms. In addition, the use of the lemongrass, which contains many active substances such as poly phenols, flavonoid, polysaccharides, also contributed to preserving the microbial load of the laboratory-produced concentrated milk (Tilaye *et al*., 2018). The presence of a high percentage of solids in the concentrated milk, represented by sugar (lactose) and the concentration of the milk generated another pressure on the contaminated microorganisms, which increases the effect of thermal treatments on them, as most anaerobic microbes, if they exist, can survive and have the ability to produce high sugar concentrations if they are packaged and stored well. These results were identical to what was found by (Knight-Jones *et al*., 2016; Muhammad *et al*., 2018)

CONCLUSION

Milk filtration methods rely on the membrane's selectivity for proteins in particular, as well as on the membrane's general selectivity for proteins and flow rate, which are governed by the membrane's pressure and flow rate laws. Based on the findings, the proper operating parameters were established, which included a flow rate of 0.15 L/min and 2 bar, as well as the percentage of particulates and lactose isolation, in order to concentrate the milk to 2.7 times. Taking into account the flow rate entering the system, the size of the pores, the molecular weight of the protein molecules and the total number of dissolved components in the raw milk, we can deduce that the flow rate is all connected. The results of the study also showed that adding 200 mg/L of lemongrass extract to milk significantly reduced the total bacterial count, which confirms to us the effectiveness of lemongrass as a preservative in terms of microbial growth. It was found the change of the total solid and protein were excellent percentages with time of filtration and also more acceptable to the consumer in terms of flavor, color and overall appearance, making it more attractive.

ACKNOWLEDGEMENT

I would like to extend my sincere thanks to the Market Research and Consumer Protection Center at the University

of Baghdad in Iraq and the Ministry of Science and Technology, specifically the Directorate of Environment and Water in Baghdad. Their significant assistance and support were instrumental in completing this research.

Conflict of interest

We, authors of this research project collectively declare that we do not have any conflicts of interest related to this research. We have no financial interests, personal relationships, or any other factors that could potentially influence the impartiality and objectivity of this research. We affirm our commitment to conducting this research in accordance with the highest ethical standards and in compliance with all relevant institutional or organizational policies. We acknowledge that the information provided in this statement is true and complete to the best of our knowledge.

REFERENCES

Agarwal, S., Beausire, R.L.W., Patel, S. and Patel, H. (2015). Innovative uses of milk protein concentrates in product development. Journal of Food Science. 80(S1): A23-A29.

Al-Hamdani, H.M.S. (2022). study of basic chemical components and antimicrobial activity of lemongrass leaves (*Cymbopogon citratus*): Study of basic chemical components and antimicrobial activity of lemongrass leaves (*Cymbopogon citratus*). Iraqi Journal of Market Research and Consumer Protection. 14(1): 94-100.

Amirdivani, S. and Baba, A.S. (2011). Changes in yogurt fermentation characteristics and antioxidant potential and *in vitro* inhibition of angiotensin-1 converting enzyme upon the inclusion of peppermint, dill and basil. LWT-Food Science and Technology. 44(6): 1458-1464.

Arunkumar, A. and Etzel, M.R. (2015). Negatively charged tangential flow ultrafiltration membranes for whey protein concentration. Journal of Membrane Science. 475: 340-348.

Arunkumar, A. and Etzel, M.R. (2018). Milk protein concentration using negatively charged ultrafiltration membranes. Foods. 7(9): 134. https://doi.org/10.3390/foods7090134.

Arunkumar, A., Molitor, M.S. and Etzel, M.R. (2016). Comparison of flat-sheet and spiral-wound negatively-charged wide-pore ultrafiltration membranes for whey protein concentration. International Dairy Journal. 56: 129-133.

Bradford, M.M. (1976). A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Analytical Biochemistry. 72(1-2): 248-254.

Ferrer, M.A., Alexander, M. and Corredig, M. (2011). Does ultrafiltration have a lasting effect on the physico-chemical properties of the casein micelles? Dairy Science and Technology. 91: 151-170.

Hieda, N., Nagatoshi, M., Ikeuchi, Y., Iga, Y. and Goto, T. (2018). Improvement of versatility and analytical range of AOAC official method 2015.06 for selenium. Journal of AOAC International. 101(4): 1215-1218.

Judd, S.J. and Carra, I. (2021). Low-pressure membrane technology for potable water filtration: True costs. Water Research. 191: 116826. https://doi.org/10.1016/j.watres.2021.116826.

Knight-Jones, T.J.D., Hang'ombe, M.B., Songe, M.M., Sinkala, Y. and Grace, D. (2016). Microbial contamination and hygiene of fresh cow's milk produced by smallholders in Western Zambia. International Journal of Environmental Research and Public Health, 13(7): 737. https://doi.org/10.3390/ijerph13070737.

Li, H., Hsu, Y.C., Zhang, Z., Dharsana, N., Ye, Y. and Chen, V. (2017). The influence of milk components on the performance of ultrafiltration/diafiltration of concentrated skim milk. Separation Science and Technology. 52(2): 381-391.

Lipnizki, F. (2010). Cross flow membrane applications in the food industry. Membrane Technology: Volume 3: Membranes for Food Applications. 3: 1-24.

Muhammad, A., Muhammad, W., Faiza, T. and Irum, A. (2018). Lemongrass (*cymbopogon citratus*): A valuable ingredient for functional foods. International Journal of Food and Allied Sciences. 4(2): 11-19.

Marella, C., Muthukumarappan, K. and Metzger, L.E. (2013). Application of membrane separation technology for developing novel dairy food ingredients. J. Food Process. Technol. 4: 269. doi: 10.4172/2157-7110.1000269.

Meyer, P., Mayer, A. and Kulozik, U. (2015). High concentration of skim milk proteins by ultrafiltration: Characterisation of a dynamic membrane system with a rotating membrane in comparison with a spiral wound membrane. International Dairy Journal. 51: 75-83.

Moreno-Montoro, M., Olalla, M., Giménez-Martínez, R., Bergillos-Meca, T., Ruiz-López, M.D., Cabrera-Vique, C., Artacho, R. and Navarro-Alarcón, M. (2015). Ultrafiltration of skimmed goat milk increases its nutritional value by concentrating nonfat solids such as proteins, Ca, P, Mg and Zn. Journal of Dairy Science. 98(11): 7628-7634.

Muehlhoff, E., Bennett, A. and McMahon, D. (2013). Milk and dairy products in human nutrition. Food and Agriculture Organisation of the United Nations (FAO), Rome.

Porter, M.C. (1972). Concentration polarization with membrane ultrafiltration. Industrial and Engineering Chemistry Product Research and Development. 11(3): 234-248.

Prestes, A.A., Helm, C.V., Esmerino, E.A., Silva, R. and Prudencio, E.S. (2022). Conventional and alternative concentration processes in milk manufacturing: A comparative study on dairy properties. Food Science and Technology. 42: e08822. doi: https://doi.org/10.1590/fst.08822.

Salunke, P., Marella, C. and Metzger, L.E. (2021). Microfiltration and ultrafiltration process to produce micellar casein and milk protein concentrates with 80% crude protein content: Partitioning of various protein fractions and constituents. Dairy. 2(3): 367-384.

Schiano, A.N., Harwood, W.S. and Drake, M.A. (2017). A 100-year review: Sensory analysis of milk. Journal of Dairy Science. 100(12): 9966-9986.

Sharma, N., Roy, P.S.D., Majeed, A., Salaria, K.H., Chalotra, R., Padekar, S.K. and Guleria, S. (2022). Nanoencapsulation-A novel strategy for enhancing the bioactivity of essential oils: A review. Indian Journal of Agricultural Research. 56(3): 241-248. doi: 10.18805/IJARe.A-5806.

Singh, B.R., Singh, V., Singh, R.K. and Ebibeni, N. (2011). Antimicrobial activity of lemongrass (*Cymbopogon citratus*) oil against microbes of environmental, clinical and food origin. Int Res J. Pharm Pharmacol. 1(9): 228-236.

Tilaye, M., Girma, T. and Philipose, M. (2018). Cost benefits analysis of lemongrass [*Cymbopogon citratus* (DC) Stapf.] variety: WG-Lomisar-Java for herbal production at wondogenet. Journal of Agricultural Economics and Rural Development. 4(2): 494-500.

Vinderola, C.G. and Reinheimer, J.A. (2000). Enumeration of *Lactobacillus casei* in the presence of *L. acidophilus*, bifidobacteria and lactic starter bacteria in fermented dairy products. International Dairy Journal. 10(4): 271-275.

Wifek, M., Saeed, A., Rehman, R. and Nisar, S. (2016). Lemongrass: A review on its botany, properties, applications and active components. International Journal of Chemical and Biochemical Sciences. 9(1): 79-84.

Zulfa, Z., Chia, C. T., and Rukayadi, Y. (2016). *In vitro* antimicrobial activity of *Cymbopogon citratus* (lemongrass) extracts against selected foodborne pathogens. International Food Research Journal. 23(3): 1262-1267.