Effects of Probiotics, Paraprobiotics and Postbiotics against Food Borne Pathogens: A Review

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Baskar *et al.*: Probiotics and Other Forms of Probiotics against Food Borne Pathogens

Food borne pathogens (bacteria, viruses, parasites, fungi or their mycotoxins) are the major cause of substantial number of diseases with an important impact on human well-being and economy. Contamination or spoilage of food by pathogens is linked with a diverse range of outbreaks of food-borne diseases. World Health Organization defined food-borne disease as an infectious disease which is caused by contaminated food or water. However, antibiotics are vital tools used in the healthcare sector to antagonize food-borne pathogens. The frequent usage of antibiotics has resulted in antimicrobial resistance which is causing major threats throughout the world. Over the past years, there has been an increasing interest in the use of probiotic bacteria as alternatives to antibiotics. Generally, the use of probiotics improves gastrointestinal well-being has been suggested for several years however many critical issues arise in the use of probiotics. Therefore, the notion of paraprobiotics and postbiotics are comparatively novel concepts which do not fit in terms of probiotics. Paraprobiotics and postbiotics are produced from probiotic organisms which provide beneficial impacts on food commodities and human health. This review will provide insight into probiotics, paraprobiotics and postbiotics against food-borne pathogens; and their probiotic challenges.

Key words: Postbiotics, paraprobiotics, beneficial microbes, health benefits, pathogens

The link between the food consumption and human diseases was primarily reported in 460 BC. Food-borne illness or diseases are major public health issue affecting human health and food safety around the world[1]. The main cause for food-borne illness is consumption foodstuff or animal product contaminated with pathogens (bacteria, viruses, parasites, fungi or their mycotoxins)[2,3]. According to the 2018 World Bank report Food Borne Diseases (FBD) affect nation economy in low and mild incoming countries. Globalization, climate change, inadequate food safety regulation and poor sanitation and food handling practices are the common cause for outbreak of FBD. More than 200 types of food borne illness have been recognized and enormous numbers of people are affected by FBD. FBD are a rising worldwide concern owing to their growing mortality and morbidity. In 2010, FBD Burden Epidemiology Reference (FERG) was established by World Health Organization (WHO) to measure the food borne illness globally[4]. In accordance with the various reports, FBD is a major problem across worldwide which causes death and illness in million people annually[5]. To diminish the food borne pathogens, various control methods should be adopted in order to decrease the occurrence of FBD and food spoilage through pathogens[6,7]. The uses of biological, chemical and physical techniques are generally employed alone or together with other methods to preserve the food so as to reduce food spoilage by microorganisms[8,9]. Among these methods, chemical preservation is the most

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common approach for food preservation to regulate pH, to serve as antioxidant and antimicrobial factors[10,11], though, nowadays an increasing heath concerns of consumers considering chemical preservatives as unhealthy[12]. For this reason, recently many researchers have been focused to create food products with less additives or utilizing natural compounds as additives to assure the quality and safety of food products[13-15]. Also, natural compounds as antimicrobial agent has less probability to develop Antimicrobial Resistance (AMR) on food borne pathogens. In this regard, probiotics, paraprobiotics and postbiotics as natural antimicrobial agents have gained more interest among the researchers[16,17]. Probiotics or Lactic Acid Bacteria (LAB) is one of the most investigated natural antimicrobial agents owing to their health well-being properties[18]. Despite the fact, the viable probiotic has some disadvantages; whereby to address this issue, paraprobiotics and postbiotics that were also from probiotics is an emerging concept to control the FBD and enhance human health. This review aimed to emphasize the advantageous effect of probiotics, paraprobiotics and postbiotics against food borne pathogens. The possible applications of paraprobiotics and postbiotics over probiotics were also discussed.

FOOD BORNE PATHOGENS

FBD are often caused by contaminated food products. The causative agent for contaminated food products is illustrated in fig. 1. Bacteria and its toxins, viruses, fungi and parasites compounds are the major common reason for food-borne illness. The most common food-borne pathogens are listed in the Table 1. To treat the food-borne illness caused by food borne pathogens, antibiotics have been widely used. According to 2018 Food and Agricultural Organizational report 25 % of food borne pathogens exhibits resistance to one or more group of antibiotics. Therefore, most of the food-borne pathogens has resistance to antibiotics. Antibiotic resistance studies displays that most of the food-borne pathogens were resistant to minimum one antibiotic[19,20]. Therefore, overuse of antibiotics cause resistance in pathogenic genes in addition to their pathogenicity[21,22]. As a result, the food-borne pathogens act as a reservoir of resistance genes in addition of their pathogenicity[23]. In addition, impulsive usage of antibiotics is a major threat worldwide to the human population[22]. To overcome this drawback biological methods such as probiotic, postbiotic and parabiotic are a novel alternative approach.

PROBIOTICS, POSTBIOTICS AND PARAPROBIOTICS AGAINST FOOD-BORNE PATHOGENS

Probiotic bacteria:

World Health Organization defined probiotics are live microorganism that confer a health benefit on the host and prevent diseases like inflammatory diseases, obesity, diabetes, food allergic reaction, lactose intolerance and food borne illness when administered in adequate amounts[24]. Probiotic bacteria eliminate the pathogenic organisms by the following mechanisms. Synthesis of antimicrobial compounds; immunomodulation; improvement of intestinal barrier performance; exclusion of pathogenic organisms by competitive mechanism. The existence of probiotics in gastro-intestinal tract interferes the pathogenic organism adhesion and undermining the pathogenicity as exhibited in fig. 2. Numerous investigations stated that probiotic bacteria have ability to reduce the propagation of pathogenic bacteria. Kariyawasam *et al.*[25] reported that *Lactobacillus brevis* act as antagonist against *Escherichia coli* O157:H4, *Listeria monocytogenes*, *Salmonella enteritis* and *Staphylococcus aureus*[25]. Another investigation revealed that five isolates of LAB from kimchi were inhibited the biofilm formation of *Listeria monocytogenes*[26]. As similar, *Lactobacillus brevis* identified from Italian cheese has investigated the Lactobacillus auto-aggregation and co-aggregation properties against *Escherichia coli*, *Salmonella typhimurium*, *Staphylococcus aureus* and *Pseudomonas aeruginosa*. The outcome of the study revealed that *Lactobacillus* strain has limited the proliferation of food borne pathogens by co-aggregation and auto-aggregation mechanisms[27]. A study conducted by Mahjoory *et al.*[28] stated that probiotic strain *Limosilactobacillus fermentum* fight against the aflatoxins producing organisms such as *Aspergillus niger* and *Aspergillus flavus* and it eliminate these organisms through auto-aggregation and co-aggregation mechanisms[28]. Also, probiotics was directly incorporated into the food products as bio-preservative in order to reduce or eliminate the foodborne pathogens in food products. Several studies were investigated the incorporation of probiotics into food products to hinder the proliferation of foodborne pathogens. In fresh cut pear researchers investigated the inter linkage between *L. rhamnosus* and pathogenic bacteria in fresh-cut pear and they found that *L. rhamnosus* reduces level of invasiveness of pathogenic bacteria[29]. Similarly in another investigations, *Pediococcus acidilactici* and *Lactobacillus plantarum* TN8 introduced into the food matrices of beef sausage substantially reduced the count of Enterobacteriaceae hence also extend the shelf life of sausage[30,31]. Likewise, the defensive effects of integration of probiotic organisms into food matrix were investigated in numerous studies. An extensive *in vitro* and *in vivo* research was conducted on probiotic strains such as *Bifidobacterium brevis*, *L. paracasei*, *L. plantarum*, *L. delbrueckii*, *Clostridium butyricum*, *L. rhamnosus* GG, *L. gasseri*, *L. helveticus*, *L. acidophilus*, *L. reuteri* ATCC 55730, *Bacillus thermophilum* RBL67, *Enterococcus faecium*, *L. crispatus*, *L. gallinarum*, *L. rhamnosus* J10-L and *L. casei* Q8-L and the outcome of the studies showed that probiotics have reduced the effect of major food borne pathogens namely *E. coli* O157:H7, *Salmonella* species, *Campylobacter jejuni*, *Listeria monocytogenes* and *Shigella* species[32-34]. In addition, animal studies displays that certain probiotics has antiviral effect by blocking the viral adhesion, replication and antiviral compound synthesis[35,36].

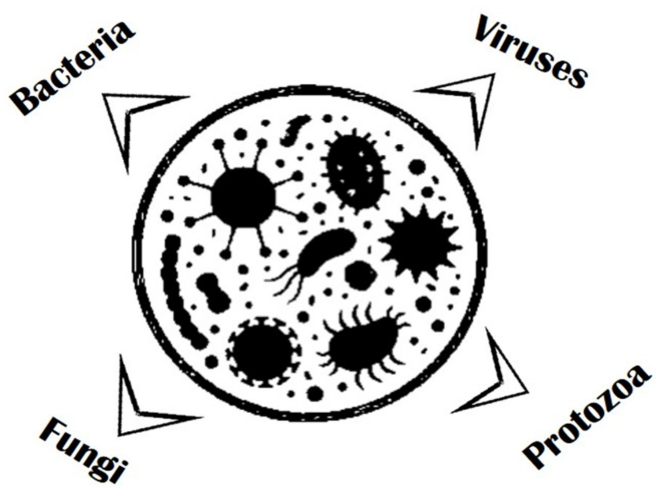

Fig. 1: Food borne pathogens

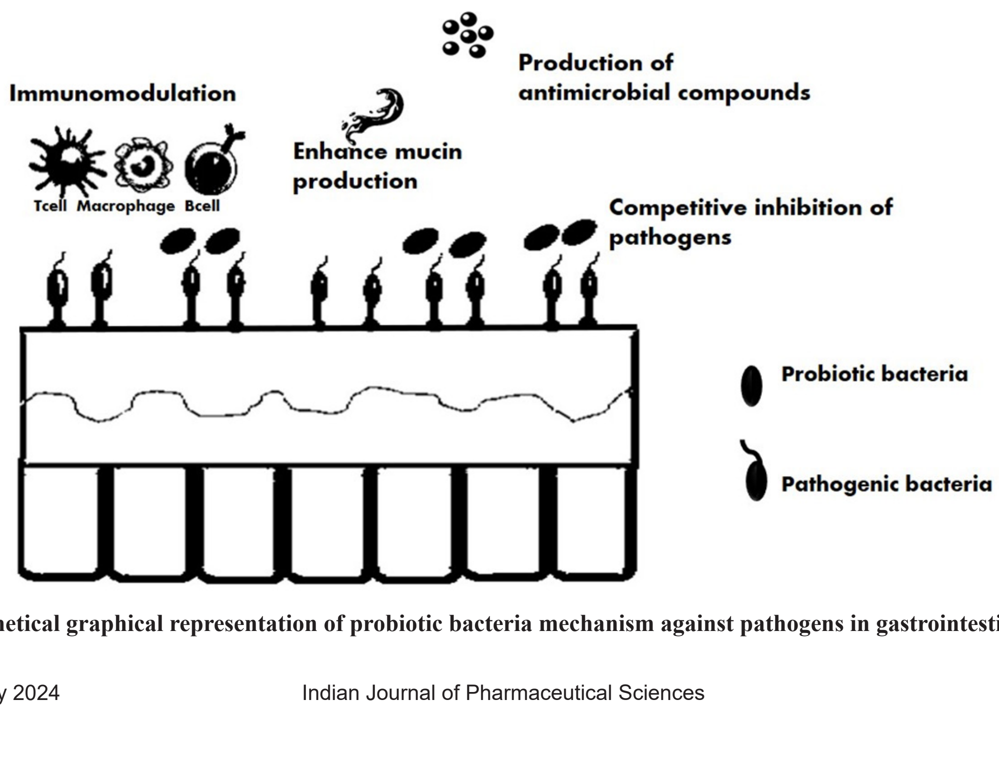

Fig. 2: A hypothetical graphical representation of probiotic bacteria mechanism against pathogens in gastrointestinal tract

Numerous clinical evidences have confirmed that probiotic strains reduce the risk associated with food borne pathogens[37,38]. Even if the utilization of probiotics is a promising approach, there are some concerns for the usage of probiotics such as transfer of virulence gene, emergence of bacteremia in immuno compromised patients, development of antibiotic resistance, also short durability and stability have hinder their prospective application in the pharmaceuticals and food sectors[39-41]. To overcome these issues, paraprobiotics and postbiotics the other form of probiotics are being investigated against food-borne pathogens owing to the enhanced health and well-being properties of host[42].

Postbiotics and paraprobiotics:

Paraprobiotics are also called dead cells or non-viable probiotics or ghost-probiotics which provides health benefits to animals and humans when administered in sufficient quantity. The probiotics inactivation can be achieved by different methods as represented in fig. 3. Paraprobiotics entirely lost their viability after being exposed to elements that change microbial cell structures, such as mechanical damage to the envelope of cell, key enzyme's inactivation and DNA filament breaks[43]. Hence, the paraprobiotics are the inactivated probiotic bacterial cells contain cell constituents include teichoic acid, surface proteins, or crude extracts of microbial cells holds complex composition of chemicals[44]. However, several studies have stated that paraprobiotics display a tremendous health benefit characteristics more than the probiotic bacteria[45,46]. A very modest investigation was conducted on paraprobiotics against food-borne pathogens. Tareb *et al.*[47] examined the activity of heat inactivated or paraprobiotics *L. farciminis* CNCM-I-3699 and *L. rhamnosus* CNCM-I-3698 activity against the poultry based food borne pathogen *Camphylobacter jeujeni* and found that paraprobiotics strongly inhibited the adhesion of *Camphylobacter jeujeni* in the intestinal mucin[47]. In a mice model of *Helicobacter pylori* infection, the paraprobiotic *Lactobacillus johnsonii* has decreased the cell count of *H. pylori* after the recurrent consumption of heat killed *L. johnsonii* strain[48]. *In vitro* investigation of five viable and non-viable strains Levi*lactobacillus brevis* O22, Levi*lactobacillus brevis* O24, *Lacticaseibacillus casei* O12, *Lacticaseibacillus casei* O16, and *Lactiplantibacillus plantarum* O20 were tested against pathogens *L. monocytogenes* 15313 and *S. aureus* 25923. The outcome of the investigation showed that the five viable and non-viable LAB strains were preventing the colonization of pathogenic strains by coaggregation. Also, non-viable LAB strains have high activity against the pathogen than viable LAB strains[49]. Similarly, heat inactivated LAB strains such as *L. fermentum*, *Enterococcus faecium*, *L. acidophilus*, and *L. plantarum* reduced the *Salmonella* infection in a mice model. The authors reported that antagonist effect of heat inactivated LAB strains might attributed to the presence exopolysaccharides and lipoteichoic acid[50].

Postbiotic refers to inactive microbe's cell components, or metabolites that are released through fermentation by probiotic microorganisms. In most of the studies, postbiotics are referred as filtered Cell Free Supernatant (CFS) which is produced from the fermented probiotic culture media, is where postbiotics are derived in the majority of research[51]. Postbiotics is a mixture of bioactive substances instead of single purified compounds; the postbiotics are the intricate composition of metabolic end products produced by probiotics in CFS included secreted proteins, short chain fatty acids, organic acids, peptides, vitamins, enzymes, secreted biosurfactants and amino acids, etc.,[51,52]. The mechanisms of postbiotics are not clearly understood. However, postbiotics might modulate and provoke the immunological response of host.

Postbiotics antimicrobial mechanisms majorly depend on the compounds secreted by probiotic bacteria. Therefore, aforesaid postbiotic compounds might interpret the enzyme's structure and activity, electron transport chain, membrane degradation of pathogenic organisms, prohibition of macromolecule synthesis and create unfavorable condition for survival of pathogenic organisms through reduce the pH of the environment[54]. Due to the advantages of postbiotics, researches on postbiotics against food borne pathogens are increasing. Moreover, many *in vitro* and *in vivo* studies conducted by researchers showed that antimicrobial activity of postbiotics against pathogenic organism. The studies which used postbiotics against foodborne pathogens are listed in Table 1 and Table 2.

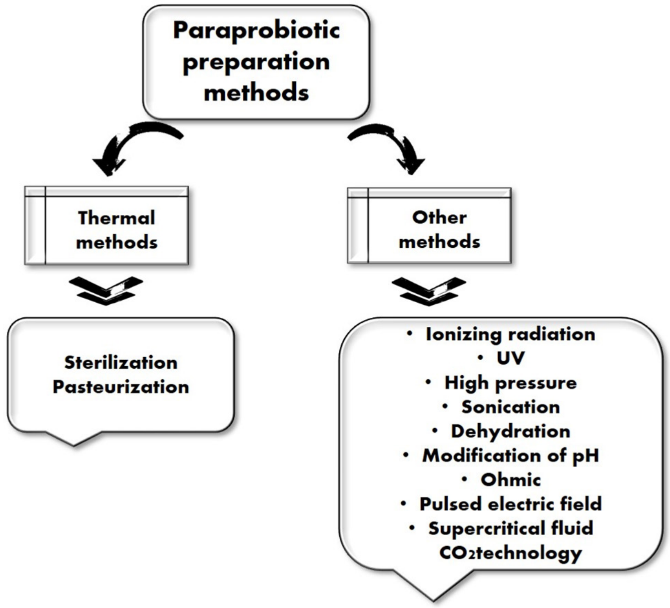

Fig. 3: Inactivation methods of probiotics organisms in paraprobiotic preparation

TABLE 1: THE COMMON FOOD BORNE PATHOGENS AND ITS EFFECT ON HUMAN HEALTH

S. No	Food-borne bacteria	Sources	Incubation period	Symptoms	References
1	*Salmonella* species and its enterotoxins Ex: *Salmonella typhi*	Eggs, fruits, beef, sprouts, vegetables, pork, chicken, and nuts	7-28 d	Vomiting, abdominal ache, constipation, rose spots, fever, nausea, headache, cough, chills, bloody stools, and fever	[55-60]
2	*Staphylococcus* and its enterotoxin Ex: *Staphylococcus aureus*	Meat, puddings, pastries, and sandwiches,	1 d	Diarrhea, nausea, vomiting, and stomach cramps	
3	*Clostridium* and its neuro toxins	Meat (beef, poultry), food left for prolonged periods	2 h-6 d, more often than not 12-36 h	Respiratory, loss of energy, paralysis vertigo, blurry vision, light reflex loss, complexity in swallowing, and dry mouth	
5	*Bacillus cereus*	Various foods especially rice and left overs, soups, sauces, and other cooked meals kept for extended period at ambient temperature.	24 h	Vomiting, watery diarrhea, nausea and abdominal cramps	
6	*Escherichia coli* 0157:H7	Contaminated water and food, uncooked beef, unpasteurized milk and juice, soft cheeses, infected people feces, cows, goats and sheep animal farms	5-10 d	Hemolytic Uremic Syndrome (HUS) comprises less production of urine. Chronic diarrhea and stomach ache, no or low-grade fever, vomiting, urine is dark colored, and color change in lower eyelids and cheeks	
7	*Listeria monocytogenes*	Dairy products, soft cheese, raw fruits and vegetables, ready-to-eat foods; hot dogs and deli meats refrigerated foods such as pastries, meats, and smoked sea foods.	Days to several weeks	Fever, muscle ache, stiff neck, unsteadiness, and convulsions	

8	*Shigella*	Intake of contaminated food and water or via infected person	5-7 d	Nausea, sudden stomach cramping, fever, diarrhea that might be bloody or mucus, and rectal tenesmus
9	Norovirus	Ready-to-eat foods touched by infected food, intake of	12-48 h	Vomiting, nausea, diarrhea and abdominal pain or cramp
	(NoV, SRSV, NLVs)	contaminated raw oysters or some other contaminated foods		
10	Hepatitis A and E	Intake of uncooked shellfish, drinking contaminated water.	15-50 d	Dark urine or light-colored stools, jaundice and joint pain
11	*Toxoplasma gondii*	Contaminated food or water consumption of uncooked meat or from infected animals and oocysts.	10-23 d	Fever, inflamed lymph nodes, head and muscle ache
12	*Aspergillus* species	Aflatoxins, ochratoxin, patulin	Certain stored fruits and vegetables, mushroom, wheat, maize, dairy products cereals and nuts	Vomiting abdominal pain, liver damage and cancer

TABLE 2: ANTIMICROBIAL ACTIVITY OF POSTBIOTICS AGAINST FOOD BORNE PATHOGENS

Postbiotic solution obtained probiotic bacteria	Target pathogenic bacteria	References
Lactobacillus plantarum	*Klebsiella pneumonia, Cronobacter sakazakii, Vibrio parahaemolyticus, Pseudomonas aeruginosa, Salmonella enterica, Staphylococcus mutans, S. aureus, Listeria monocytogenes*	
Pediococous pentosaceus 4I1	*Bacillus subtilis, S. enterica, S. aureus, E. coli* O157:H7, *L. Monocytogenes,*	
L. acidophilus, L. salivarius	*E. coli*	
Lactobacillus delbrueckii subsp.	*Clostidium perfringens*	
L. rhamnosus		
P. acidilactici		
L. fermentum		[61-66]
L. casei 431	*S. aureus*	
L. acidophilus LA5		
Leuconostoc mesenteroides	*E. coli, Vibrio parahaemolyticus* and *Pseudomonas aeruginosa*	
Pediococcus acidilactici	*L. monocytogenes, Salmonella typhirium,*	
	Escherichia coli O157:H7,	
Lactic acid bacteria (*Pediococcus acidilactici, Streptococcus thermophilus, Leuconostoc mesenteroides*)	*Escherichia coli* and *Salmonella paratyphi* A	

Paraprobiotics and postbiotics have various benefits over probiotics i.e. pure form of their availability, easy storage and synthesis, precise mechanism of action and no possibility to antibiotic resistance genes transfer. The process flowchart for the synthesis of paraprobiotics and postbiotics is represented in fig. 4[55]. The use paraprobiotics and postbiotics allow overtaking the several disadvantages generated by probiotics. Therefore, the unique characteristics of postbiotic and paraprobiotics have drawn attention in other research areas such as obesity, hypertension, cancer, longer storage stability, and ability to trigger several systems governing inflammation, cardiovascular disease and oxidative stress. Similar to how human health has drawn attention, postbiotic and paraprobiotics played an important role in animal health as well[52,53]. Also, postbiotics and paraprobiotics uncover new prospects in the pharmaceutical and food industry.

FUTURE REMARKS

Many food-borne pathogens are resistant to antibiotics, which are used to treat the FBD. Meanwhile, another therapeutic strategy is being examined to minimize the up-surging antibiotic resistant pathogens. Probiotics, paraprobiotics and postbiotics are the promising alternative natural antibiotic therapy against food-borne pathogens. On the other hand, probiotic provides numerous health benefits to the host, but the above-mentioned drawbacks are the major limit for the probiotic organisms. Therefore, paraprobiotics and postbiotics would be a great substitute for antibiotic therapy, but efficiency of paraprobiotics and postbiotics relies on the efficiency of probiotic strains. On the contrary, lack of information about the paraprobiotics and postbiotics in *in vivo*, *in vitro* and clinical studies restrict the therapeutic and industrial application. Hence, it is significant to perform investigation to understand effects of paraprobiotics and postbiotics on human health and their gut microbiome interaction, also their incorporation into food products prevent the food spoilage by pathogens which are the significant cause of FBD. The deeper examination of paraprobiotics and postbiotics by the researcher will aids to develop novel prospect for the production of healthier, sustainable, natural and safer products.

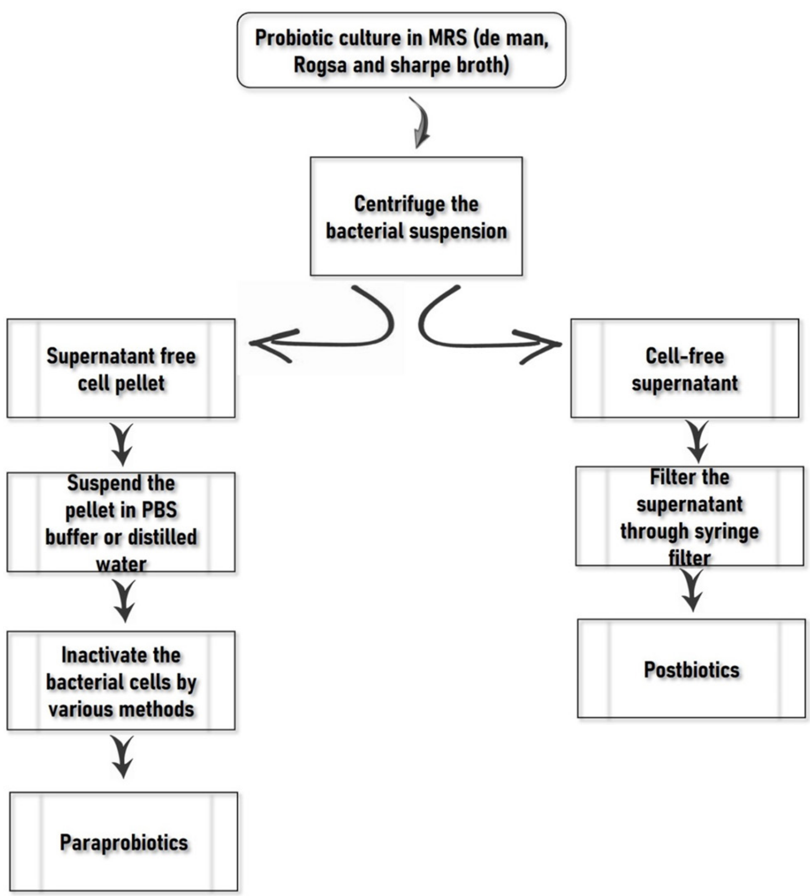

Fig. 4: Inactivation methods of probiotics organisms in paraprobiotic preparation

Author's contribution:

Ragavi Baskar and Vishnupriya Subramaniyan contributed equally to this work.

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Conflict of interest:

The author declares no conflict of interest.

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