



# Evaluation of *Lactobacillus rhamnosus* as a Protective Agent against *Bacillus cereus* Contamination in Legume-based Infant Foods

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## ABSTRACT

**Background:** *Bacillus cereus* poses a critical food safety challenge in developing countries, emphasising the need to find ways to combat its toxicity. Commercially produced baby food is authorized as a main source of nourishment for infants; thus, it should include a high concentration of nutrients and be free from any potential hazards.

**Methods:** 300 random food samples were obtained from supermarkets and pharmacies in Taif and Makkah cities, Saudi Arabia. Microbiological examinations, molecular techniques and anatomical examinations were applied to confirm *B. cereus* pathogenicity. Probiotic bacteria were tested for their antimicrobial activity.

**Result:** The bacteria were observed in almost all samples with mean total viable counts of 1.586 to 3.543 log<sub>10</sub> cfu/g. Furthermore, 196 *Bacillus* colonies were identified and 27% of them were *B. cereus*. Finally, *L. rhamnosus* is an anti-*B. cereus* agent was used for detoxification and to reduce the pathogenicity of this pathogen. After mixing *L. rhamnosus* with *B. cereus* and/or its emetic toxins and feeding it to mice, the toxicity of *B. cereus* and its enterotoxins was reduced by 37 to 62%, paving the way for using this strain to protect infant meals from hazardous microbial elements. Anatomical examination showed that mice fed with *B. cereus* exhibited severe gastrointestinal abnormalities, whereas those treated with combinations of *B. cereus* and *Lactobacillus rhamnosus* displayed normal anatomical features. This study concluded that *Lactobacillus rhamnosus* may have significantly mitigated the toxicity of *Bacillus cereus* in infant meals, highlighting its potential as a protective agent in food safety.

**Key words:** *Bacillus cereus*, Detoxification, Enterotoxins, Infant foods, *L. rhamnosus*.

## INTRODUCTION

*Bacillus cereus* plays a significant role as a foodborne pathogen in developing countries, where the combination of environmental conditions and limited public health infrastructure heightens the risk of outbreaks (Woh and Ng, 2024). In these regions, food safety is often compromised by inadequate refrigeration, poor sanitation and lack of awareness about proper food handling practices. The toxins produced by *B. cereus* can cause severe gastrointestinal illnesses characterised by vomiting and diarrhoea (Duan *et al.*, 2023).

The challenge is exacerbated by the bacteria's ability to form durable spores, allowing it to survive. Strategies targeting hygiene, storage and cooking practices remain relevant in reducing the impact of *B. cereus*.

Because infants rely heavily on limited food sources during early development, any microbial contamination poses a direct health risk due to their immature immune systems. For this reason, the hygienic quality of commercially prepared infant foods is critical, particularly during the complementary feeding period. Foodborne diseases remain a major global health concern, especially where bacterial spores can persist through processing and storage (Wang *et al.*, 2022). Among these pathogens, *Bacillus cereus* has been repeatedly identified as one of the most common contaminants in pasteurized and ready-to-eat infant foods,

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emphasizing the need for strict safety guidelines and targeted hygienic control measures to prevent exposure in this vulnerable age group (Radmehr *et al.*, 2020).

*B. cereus* is a psychrotrophic, facultative anaerobic, spore-forming pathogen associated with food-borne outbreaks (Zhao *et al.*, 2020). It can grow at low temperatures (6-10°C) and even under refrigeration, producing emetic and diarrheal toxins (Granum and Lund, 1997). Diarrheal illness results from vegetative cells producing enterotoxins when ingested at levels exceeding 10<sup>5</sup> cfu/g (Warda, 2016). In contrast, several *Lactobacillus* species and lactic acid bacteria produce antimicrobial substances that can inhibit pathogens and help prevent food spoilage (Jørgensen *et al.*, 2017; Mori *et al.*, 2026; Timothy *et al.*, 2021).

Despite previous studies addressing *Bacillus cereus* contamination in food products, limited research has specifically evaluated the detoxifying role of probiotic strains such as *Lactobacillus rhamnosus* against *B. cereus* and its toxins in legume-based infant foods (Abdel-Rauf *et al.*, 2025; Granum and Lund, 1997; Zhao *et al.*, 2020). Therefore, this study aimed to (i) evaluate the microbiological quality of infant foods, (ii) determine the prevalence and toxigenic potential of *Bacillus cereus* and (iii) investigate the antimicrobial and detoxifying effects of *Lactobacillus rhamnosus* against *B. cereus* and its enterotoxins.

## MATERIALS AND METHODS

### Infant food sample collection and preparation

A total of 300 samples of ready-to-use baby milk-based powders were obtained randomly from diverse supermarkets and pharmacies in Taif and Makkah cities, Saudi Arabia during the period from 2024 to 2025, as shown in Table 1.

### Probiotic strain growth and preservation

*Lactobacillus rhamnosus* B-445 and *Lactobacillus bulgaricus* subsp. *bulgaricus* Lb-12 DRI-VAC strains were delivered from the Northern Regional Research Laboratory (NRRL), Illinois, USA: Hansen's Lab, Denmark. *Lactobacillus cremoris* was obtained from the Department of Dairy, National Research Centre, Egypt. They were proven to be probiotics (Adebayo-Tayo and Fashogbon, 2020) by the possession of their characteristics.

### Bacteriological analysis of infant foods

For all collected samples, 25 g of each sample in 225 mL of sterile peptone water was mixed well by homogenizer for 2 min. Total bacterial counts were tested using tryptic soya agar and plate count agar media (Darwesh *et al.*, 2019). In the case of psychrotrophic spore-forming bacterial counts, the procedure followed (Sadek *et al.*, 2018).

### Isolation and identification of psychrotrophic *Bacillus* spp.

Three to five isolates from each plate of typical colonies of the total psychrotrophic spore-forming plates were picked randomly. Each colony was purified on PEMBA medium, characterized as psychrotrophic *Bacillus* spp. and then inoculated into 10 mL of tryptic soya broth at 37°C for 24 h and then subjected to several biochemical examinations and API 50 CHB (Biomerieux, France) for identification.

### Molecular detection of toxicogenic-related genes and identification of their producing pathogens

Extraction of bacterial DNA of the 22 *B. cereus* isolates was obtained by the lysozyme (20 mg/mL) and proteinase K (1 mg/mL) method, which was detailed previously (18). Three virulence genes, hbla, nhc and cytk, were amplified as enterotoxin genes using extracted DNA and specific primers as shown in Table 2 (19-23). PCR amplification was carried out starting with denaturation at 95°C for 5 min, followed by 30 cycles of 95°C for 1 min, 58°C for 1 min, 72°C for 1 min and a final elongation step at 72°C for 10 min, with a final hold at 4°C. PCR products were analysed in 1% (w/v) TAE agarose gel. All PCR experiments for each strain were carried out twice (Barakat *et al.*, 2015). One isolate of *B. cereus* had the three tested virulence genes. Thus, it was identified using molecular biology techniques according to Adesetan *et al.* (2022).

### Antibacterial and antagonistic activities of the probiotic strains

The antimicrobial activity of the four probiotic strains against *B. cereus* was evaluated using cell-free supernatants obtained after centrifugation of overnight MRS cultures. The inhibitory effect was assessed by the agar well diffusion test and the inhibition zones were measured (Khalid *et al.*, 2021).

**Table 1:** Type, numbers and locations of the collected infant foods.

Type of sample	No. of samples	Location
Honey and milk based	15	Taif city
	15	Makkah city
Wheat and milk based	15	Taif city
	15	Makkah city
Rice and milk based	15	Taif city
	15	Makkah city
Ready to use with vegetables	15	Taif city
	15	Makkah city
Ready to use with fruit	15	Taif city
	15	Makkah city
Fruit and milk based	25	Taif city
	25	Makkah city
Milk powder	35	Taif city
	35	Makkah city
Vegetables and milk based	15	Taif city
	15	Makkah city

**Table 2:** Primer sequences and amplicon sizes used in PCR analysis.

Gene	Primer type	Primer sequences	Amplicon size (bp)
		F: Forward; R: Reverse	
HblA	F	5' AAGCAATGGAATACAATGGG 3'	1154
	R	5' AGAATCTAAATCATGCCACTGC 3'	
CytK	F	5' CGACGTCAAGTTGTAACA 3'	565
	R	5' CGTGTGTAATACCCAGTT 3'	
NheC	F	5' CGGTAGTGATTGCTGGG 3'	581

The antagonistic activity of *Lact. rhamnosus* B-445 against *B. cereus* was further examined in broth co-culture. Standardized dilutions of both strains were mixed in tryptic soya broth and bacterial counts were determined at selected time intervals. *B. cereus* was enumerated on PEMBA medium (Finlay *et al.*, 1999), while *Lact. rhamnosus* was counted on MRS agar under anaerobic conditions.

#### Detoxification and reduced pathogenicity of *B. cereus* after *Lact. rhamnosus* treatment

Evaluation of the pathogenicity and production of toxins by the *B. cereus* isolate was carried out using the mouse toxicity technique. Additionally, the effect of *Lact. rhamnosus* on this pathogenicity was evaluated. *B. cereus* was cultured in 10% skim milk medium to produce enterotoxin as a supernatant. The pellet was re-suspended in a suitable quantity of phosphate buffer to attain a bacterial suspension equivalent to 0.5 McFarland standard (Darwesh *et al.*, 2020). In another case, *Lact. rhamnosus* strain was cultured in 1% (w/v) of MRS. After bacterial cultures and *B. cereus* crude toxins were prepared, an equal volume of *Lact. rhamnosus* culture was mixed with *B. cereus* culture or its emetic toxin and incubated at 37°C for 6 h.

To evaluate the pathogenicity of *B. cereus* and study the role of *Lact. rhamnosus* in attenuating the toxicity of *B. cereus*, 30 adult male mice (*Mus musculus domesticus*) with an average weight of 100 g were obtained from the animal house at Taif University. The mice were divided into six groups containing five mice each. The animals were fed a basal diet containing 50% corn starch, 20% casein, 10% sugar cane, 10% corn oil, 5% cellulose, 4% salt mixture and 1% vitamin mixture (Darwesh *et al.*, 2023). A total of 0.5 mL of each treatment was given orally to the mice by intragastric gavage for three days. The trials (6 groups) were as follows: The control group was fed phosphate-buffered saline and their regular food, the 2<sup>nd</sup> group was fed *B. cereus* and the emetic toxin, the 3<sup>rd</sup> group was fed *B. cereus* culture, the 4<sup>th</sup> group was fed a mixture of *B. cereus*, the emetic toxin and *Lact. rhamnosus* culture, the 5<sup>th</sup> group was fed a mixture of *B. cereus* and *Lact. rhamnosus* cultures and the 6<sup>th</sup> group

was fed *Lact. rhamnosus* culture. The results were recorded for the mice after three days and the animals were thoroughly observed for any toxic signs through this period. Convulsions, motor activity, tremors, sedation, aggressiveness, relaxation of muscle, analgesia, hypnosis, ptosis, paralysis, lacrimation, skin colour and diarrhoea were noted during the experiment. The observed death was also recorded in each group.

#### Statistical analyses

All results were expressed as the mean±standard deviation (SD). Statistical significance was evaluated using analysis of variance (ANOVA, SAS software) followed by determining the least significant difference at 0.05.

#### Ethical approval

All experiments followed Taif University's Ethics Committee's guidelines (Approval No. TU 20-050, Taif, Saudi Arabia).

## RESULTS AND DISCUSSION

#### Spreading of microbial communities in infant foods

Three hundred infant food samples were collected randomly from pharmacies and supermarkets in Makkah and Taif cities. The collected samples were subjected to microbiological analyses. The results showed that the total bacterial counts (TBCs) were detected in almost all examined samples, with mean total viable counts ranging from 1.586 to 3.543 log<sub>10</sub> cfu/g. The results revealed that the total psychrotrophic spore bacteria were noticed in 40 and 60% of the whole samples of milk-based infant food with vegetables and fruits, respectively.

#### Distribution and identification of *Bacillus* spp

The samples were examined to determine their content of *Bacillus* spp. by isolation of different morphological aerobic spore-forming colonies and then characterized (morphologically, biochemically and API CHB50 tests) using acceptable protocols of *Bacillus* spp. identification. The distribution of diverse isolated *Bacillus* species is illustrated in Fig 1. The results revealed that *B. subtilis* was

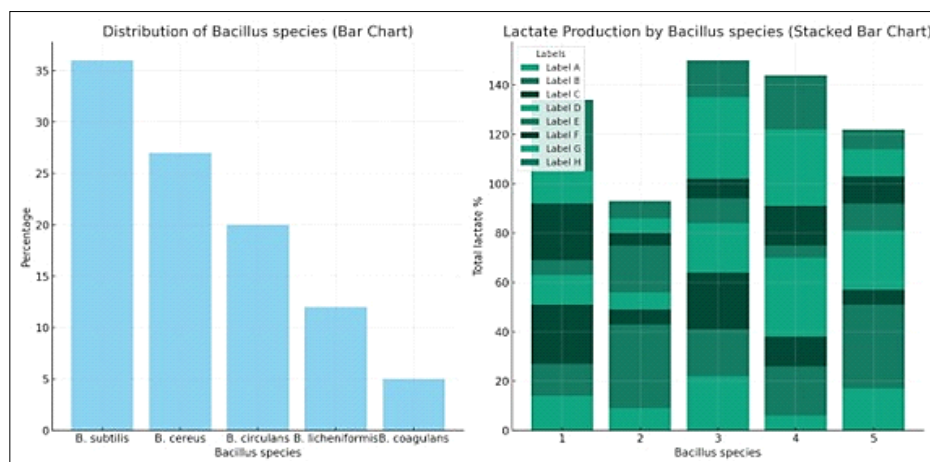


Fig 1: Distribution of different total psychrotrophic *Bacillus* species in the obtained samples.

the most common species compared with other *Bacillus* spp. with an incidence of 36%, followed by *B. cereus* (27%), *B. circulans* (20%), *B. licheniformis* (12%) and *B. coagulans* (5%). Vegetables and fruit milk-based infant foods had the highest numbers of *Bacillus* spp. From the obtained *Bacillus* isolates, *B. subtilis* and *B. cereus*, due to their hydrolytic activities, are essential for food hygiene.

#### Distribution and identification of *Bacillus cereus*

*B. cereus* bacteria calculated on PEMBA medium were detected in almost all tested samples with the highest percentages in milk-based infant food with fruits and vegetables, which reached 62.2 and 26.6%, respectively (Fig 2).

In contrast, the *B. cereus* count was not high in the ready-to-use infant food with fruits and vegetables. Generally, 40 out of 300 samples (approximately 26.67%) of the examined infant foods were positive for *B. cereus* counts.

Due to *Bacillus cereus* isolate No. 8 pathogenicity and production of the 3 virulent harmful toxin-related genes, *B. cereus* was identified using molecular biology techniques to confirm the biochemical and API CHB50 tests. The obtained sequence was compared with available sequences in the GenBank database and the similarity percentage of this isolate accounted for 98% with *Bacillus cereus* strains. Additionally, a phylogenetic tree was constructed and it showed that this strain was very close to the type strains of *B. cereus* deposited in the culture collection centre of the National Center for Biotechnology Information (Fig 3).

#### Molecular detection of toxicogenic-related genes

Three virulence genes (*hbla*, *nhc*, *cytK*) were screened in 25 *B. cereus* isolates using PCR. Only two isolates (8 and 9) carried all three genes. The *hbla* gene (1200 bp) appeared in five isolates and was mainly detected in samples from milk-based infant foods with fruit, rice, wheat and honey but with variable frequencies. The *nhc* gene showed a wider distribution across isolates from infant foods

containing fruit, vegetables, honey and wheat, while it was absent in rice-based samples. In contrast, the *cytK* gene exhibited the highest prevalence, being detected in 92% of isolates collected from all infant food types examined. Overall, isolate 8-obtained from milk-based infant food with fruit-showed the strongest *hbla* band intensity among the positive samples (Fig 4).

#### Detoxification and reduced pathogenicity of *B. cereus* using *Lact. rhamnosus*

The cell-free supernatant containing bioactive metabolites of probiotic strains was obtained and employed against the *B. cereus* strain using an agar well diffusion assay. The data from the pictures illustrated in Fig 5 show that the strain *Lact. rhamnosus* B-445 produced high antibacterial activity against the *B. cereus* strain, with a 21 mm inhibition zone diameter, followed by *Lb. dulbrueckii* and *Lb. cremoris*, but *Leuconostoc mesenteroides* appeared to have no activity. The antagonistic activity of *Lact. rhamnosus* against *B. cereus* was assessed in the broth culture media system and the results are illustrated in Fig 5. From that, we can clearly show the antagonism between the two tested strains. The growth of the *B. cereus* strain was inhibited under mixed culture with *Lact. rhamnosus* starting from 6 h of incubation. *Lact. rhamnosus* exposed a high inhibitory activity against the *B. cereus* strain, resulting in a decrease in the total counts from 5.3 to 3.3 Log<sub>10</sub> cfu/mL after 24 h with a reduction of 37.7% and reached 2.0 Log<sub>10</sub> cfu/mL with a reduction of 62.2% after 48 h of incubation.

#### Toxicology studies of *B. cereus* cells and crude toxins

The probiotic strain *Lact. rhamnosus* was mixed with *B. cereus* and/or its emetic toxins and added to the diets of the experimental mice. After three days of the experiment, the groups treated with *B. cereus* cells and its emetic toxin, groups two and three, showed some clinical signs, such as increased mortality and decreased weight compared with the other groups. The control group did not show any abnormal weight changes (ranging between 100 and 103 g)

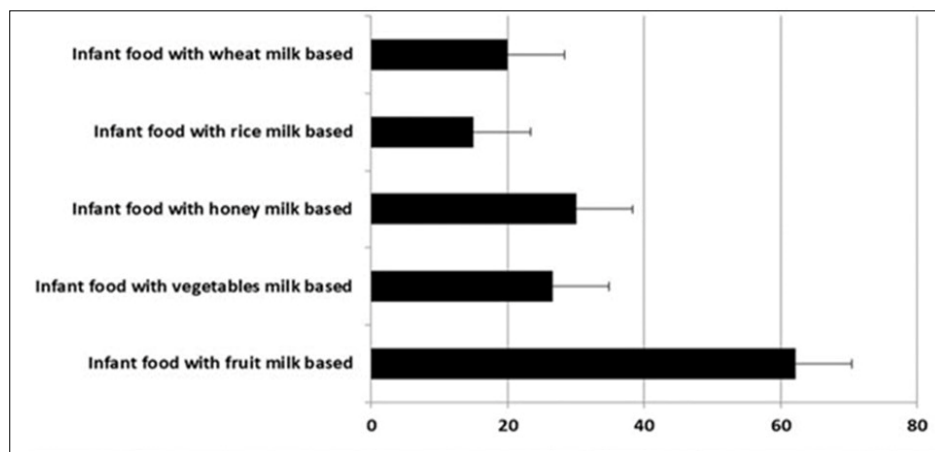
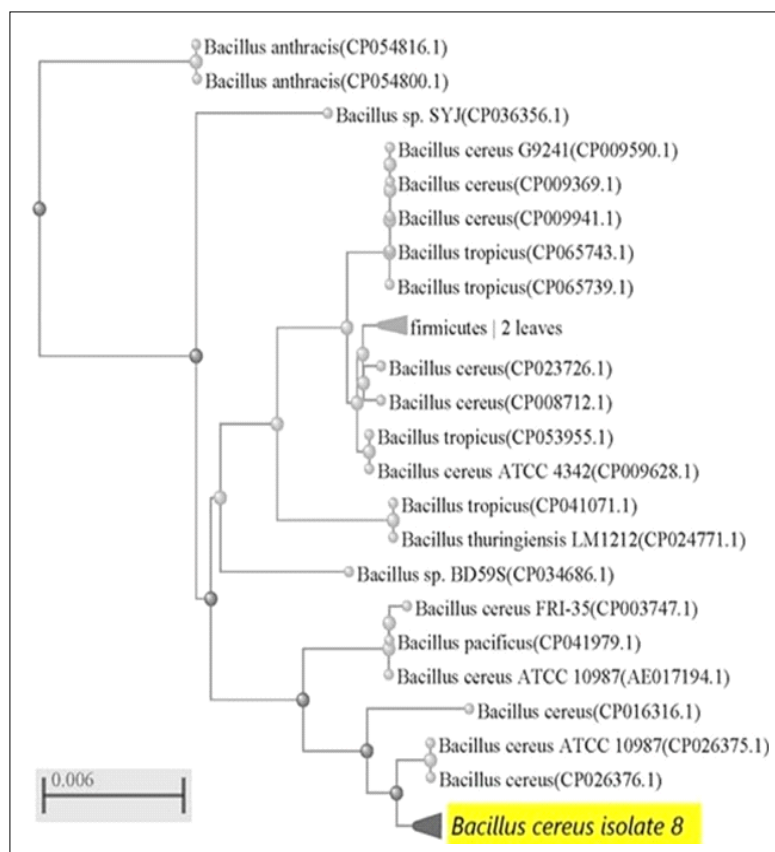


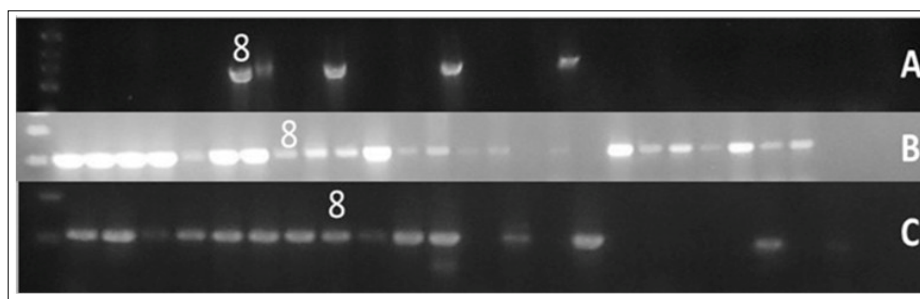
Fig 2: Distribution of *B. cereus* in the obtained samples.

after three days. While mice fed with bacterial cells of *B. cereus* showed loss of their body weight and slower movement; their weights ranged from 72 to 75 g and two of five mice died. Additionally, four out of five mice in the group fed emetic toxins died and the remaining mice had a clear decrease in their weight to 60 g and slower movement. However, the group fed a mixture of *B. cereus* cells and *Lact. rhamnosus* exhibited normal appearance and movement with weights reaching 93-94 g. Moreover, the mouse group provided a mix of crude toxins and *Lact. rhamnosus* culture displayed normal appearance, motion and weight, with weights ranging between 98 and 101 g. The group was fed with a diet containing *Lact. rhamnosus* culture appeared to exhibit

normal behaviour and their weight reached 100-105 g. Anatomical examination of the gastrointestinal tract of dissected mice revealed swelling of the intestinal tract due to fluid accumulation in the intestine's lumen and inflammation of the intestines and stomach, particularly when mice were fed with the *B. cereus*. Additionally, ulcers and dark red spots were observed in the liver of groups fed with *B. cereus*, indicating severe vascular effects of the toxin. In contrast, no significant changes were noted in the stomach and intestines of mice treated with crude emetic enterotoxin. However, the group fed a mixture of *B. cereus* and *Lactobacillus rhamnosus* showed a normal stomach, intestine and liver compared to the control group.



**Fig 3:** Phylogenetic tree of *B. cereus* and related species in the gene bank.



**Fig 4:** Agarose gel pictures for the PCR product of *hbla* (A), *cytk* (B) and *nhec* (C) genes in *B. cereus* isolates compared with the marker (1 kb).



Similarly, the group fed a mixture of *B. cereus* emetic enterotoxin and *Lactobacillus rhamnosus* B-445 exhibited normal stomach, intestine and liver conditions. Furthermore, the group fed the diet with *Lactobacillus rhamnosus* B-445 appeared normal in all examined body contents (Fig 6).

Infant foods are certified as a primary nutrition source for babies from seven months to 2 years old. Thus, they should represent a rich source of nutrients from several origins. However, germs in babies' food can cause severe infection due to their immune systems not being completely developed (Martin *et al.*, 2010). This study evaluated the hygiene of infant foods and tried to improve the quality of these foods. To achieve this objective, three hundred infant food samples were collected randomly from pharmacies and supermarkets in Makkah and Taif cities, Saudi Arabia. The collected samples were subjected to microbiological analyses. The results showed that the total bacterial counts (TBCs) were detected in almost all samples, with mean complete viable counts ranging from 1.586 to 3.543 log<sub>10</sub>

cfu/g. A similar observation was reported by other researchers (Gautam, 2015; Luby *et al.*, 2011).

The TBC falls within standard regulations; therefore, it is less critical than spore-forming and pathogenic bacterial counts. High TBC values generally indicate poor hygienic practices during handling, packaging, or the use of low-quality ingredients (Mokgaotsi, 2019). For this reason, total psychrotrophic spore-forming bacteria were also evaluated and they were detected in 40% and 60% of milk-based infant foods with vegetables and fruits, respectively.

At this stage, psychrotrophic spore bacteria in infant milk meals were reported at  $2.9 \times 10^2$  cfu/g (Ahmed *et al.*, 2008) and another study showed that 40% of vegetable-based baby foods tested positive (Sadek *et al.*, 2018). *Bacillus spp.* were isolated and identified following standard morphological, biochemical and API CHB50 protocols, with *B. subtilis* being the most common species in milk-based vegetable and fruit foods. Controlling *B. subtilis* and *B. cereus* is essential due to their hydrolytic activities, toxin

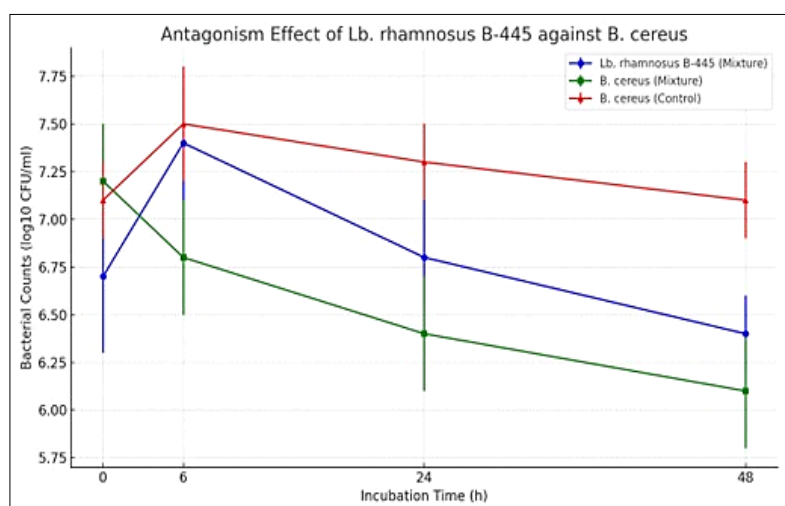
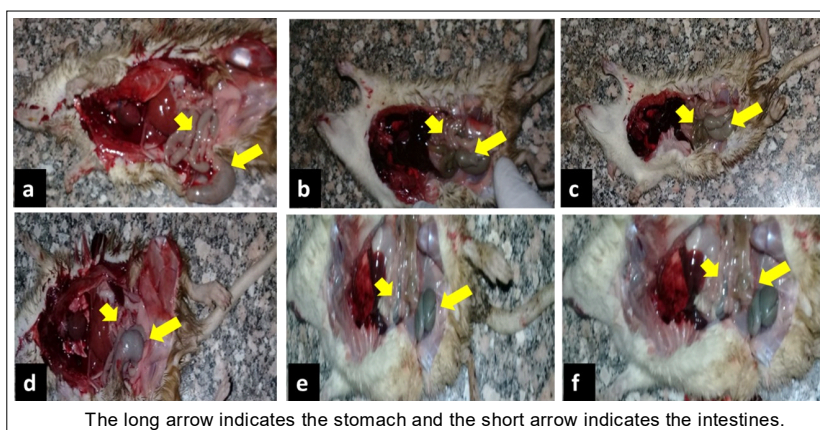


Fig 5: Anti-*Bacillus cereus* activity of some probiotic strains.



The long arrow indicates the stomach and the short arrow indicates the intestines.

Fig 6: Anatomical examination of mice under various treatments like control (a), bacterial cells of *B. cereus* (b), *B. cereus* the emetic toxins (c), mixture of *B. cereus* toxins with *L. rhamnosus* B-445 culture (d), mixture from bacterial cells of *B. cereus* and *L. rhamnosus* B-445 culture (e) and *L. rhamnosus* B-445 culture (f).

production and ability to grow at refrigeration temperatures. These findings agree with previous reports detecting *B. subtilis* (28%), *B. licheniformis* (20%) and *B. cereus* (14%) in infant formulae (Mostafa *et al.*, 2002). Although less prevalent, *B. cereus* remains the most harmful due to its pathogenicity and toxin production. FDA regulations require *B. cereus* levels to remain below 100 cfu/g. In this study, *B. cereus* was detected in all tested samples, especially milk-based infant foods with fruits and vegetables, while absent in ready-to-use products. Other studies similarly reported *B. cereus* contamination in powdered milk and a variety of infant foods (Kim *et al.*, 2011). Differences in prevalence are likely related to sample type, testing method and product formulation. Previous research has suggested applying stable, nontoxic bioactive compounds and food-grade natural materials to reduce *B. cereus* in infant foods (Sadek *et al.*, 2018).

The pathogenicity of *B. cereus* was assessed by detecting three key enterotoxin-related genes *hbla*, *nhec* and *cytK* using PCR. Two isolates (8 and 9) carried all three virulence genes. Toxigenic *B. cereus* strains have been previously reported in a wide range of starchy and dairy-based foods (Jovanovic *et al.*, 2021; Mohammadi, 2023), emphasizing the need to evaluate the safety of infant foods commonly marketed in Saudi Arabia. PCR detection methods are increasingly applied for identifying these genes (Kim *et al.*, 2000). In this study, isolate 8 originating from milk-based infant food with fruit was positive for the *hbla* gene, consistent with earlier reports detecting *hbla* in dairy and child-related foods (Rahimi *et al.*, 2013). Gene distribution patterns varied according to food type, season and collection conditions. The *nhec* gene was widespread among isolates from fruit-, vegetable-, honey- and wheat-based infant foods but absent in rice-based samples, matching previous findings where *nhec* occurred in 80% of infant-food isolates (Rahimi *et al.*, 2013). The *cytK* gene showed the highest prevalence, appearing across all food categories examined. As *cytK* is strongly associated with severe food-poisoning outbreaks, isolate 8 was selected as the pathogenic reference strain. Its genomic DNA was extracted and the 16S rRNA gene sequence showed 98% similarity to known *B. cereus* strains. Phylogenetic analysis confirmed its close relationship to established *B. cereus* type strains in the NCBI database.

The antimicrobial activity of the probiotic strains was assessed using cell-free supernatants in an agar well diffusion assay, where *Lact. rhamnosus* B-445 showed the strongest inhibition against *B. cereus* (21 mm), followed by *Lb. dulbrueckii*, *Lb. cremoris* and *Leuconostoc mesenteroides*. This variation reflects differences in the production of bioactive compounds such as organic acids, polysaccharides and bacteriocins (Liu *et al.*, 2023; Moniri *et al.*, 2017). LAB strains, including *Lact. rhamnosus* B-445 and *Lb. dulbrueckii*, are known for their antibacterial activity against *Bacillus* spp. (Ahmad *et al.*, 2022; Zeinab *et al.*, 2015). In broth co-culture, *Lact. rhamnosus* markedly

suppressed *B. cereus* growth, reducing counts from 5.3 to 3.3 Log<sub>10</sub> cfu/mL at 24 h and to 2.0 Log<sub>10</sub> cfu/mL at 48 h. This inhibition is attributed to bacteriocins and related metabolites with applications in food biopreservation (Fuochi *et al.*, 2019).

Infant foods require high hygiene standards because *B. cereus* is a pathogenic bacterium that poses significant health risks. The bacterium is particularly dangerous due to its spore-forming ability, which makes it resistant to thermal treatments and its toxins remain stable even under hygienic controls. To investigate a safe protective agent, the probiotic strain *Lact. rhamnosus* was mixed with *B. cereus* cells and/or emetic toxins and incorporated into mice diets. Mice fed only *B. cereus* cells or toxin alone experienced severe clinical signs including significant weight loss (60-75 g), reduced mobility and high mortality with 4 out of 5 mice in the toxin-treated group dying. However, mice fed *B. cereus* (cells or toxins) combined with *Lact. rhamnosus* maintained normal weight (93-105 g) and exhibited normal behavior and movement. Pathological examination revealed that mice exposed to *B. cereus* alone developed swollen intestinal tracts, ulcers and dark coloration of the liver. In contrast, groups treated with *B. cereus* mixed with *Lact. rhamnosus* showed normal stomach, intestine and liver tissues. These findings demonstrate that *Lact. rhamnosus* B-445 could serve as an effective and environmentally safe protective agent against *B. cereus* in infant foods. Probiotic supplementation has been widely applied to improve microbial balance and health performance in food-related biological systems, as reported in previous publications (Barde *et al.*, 2025).

## CONCLUSION

The findings demonstrate that *Bacillus cereus* contamination in infant foods represents a significant health risk due to its toxigenic potential and ability to persist under storage conditions. Moreover, *Lactobacillus rhamnosus* effectively reduced bacterial pathogenicity and toxin-associated effects, supporting its potential application as a natural bioprotective agent to enhance the safety of infant food products.

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## Author contributions

The principal author completed all research work items in this study.

## Data availability statement

Data is contained within the article.

## Conflict of interest

The authors declare no conflict of interest.

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