The use of lactic acid fermentation and propionic acid fermentation to obtain fermented milk and bean drinks

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Introduction

- Fermented drinks are highly valued for their organoleptic and health-promoting properties.
- The most popular and appreciated are those based on milk in the form of fermented milks, but in recent years drinks based on plant matrices have been also gaining popularity.
- The most popular microorganisms used in such production are lactic acid bacteria (LAB) and Bifidobacterium bacteria.
- There are more and more suggestions indicating the potential of propionic acid fermentation bacteria in this application

Aim of study

The aim of this study was to obtain fermented drinks based on a milk and plant matrix using:

- mesophilic lactic acid bacteria (*Lactococcus lactis* ATCC 11454),
- propionic acid bacteria (*Propionibacterium freundenreichii* subsp. *shermanii* PS4),
- bifidobacteria (*Bifidobacterium animalis* subsp. *lactis* Bb-12)

and their selected analysis.

Materials and methods

- Commercial UHT cow milk with a fat content of 3.2%, as well as a bean drink obtained in laboratory conditions from germinated Adzuki beans (*Vigna angularis* var. *angularis*) were used to obtain the drinks.

- The bean seeds were germinated in a professional germinator at 25°C for 72 hours. Then drinking water was added to the germinated bean seeds and the whole was blended with a food processor. The obtained homogenate was poured through a fine sieve and brought to the boil in order to gelatinize the starch. The obtained vegetable drink was sterilized at 121°C for 20 minutes.

Materials and methods

- The starter bacteria (as monocultures and their mixture) were added to the sterilized portions of the plant drink and the portions of cow's milk in order to carry out the fermentation process.

Milk drinks	Bean drinks
L. lactis ATCC 11454 (0.04%)	*L. lactis* ATCC 11454 (0.04%)
P. freundenreichii subsp. *shermanii* PS4 (0.04%)	*P. freundenreichii* subsp. *shermanii* PS4 (0.04%)
B. animalis subsp. *lactis* Bb-12 (0.04%)	*B. animalis* subsp. *lactis* Bb-12 (0.04%)
their mixture (1:1:1)	their mixture (1:1:1)

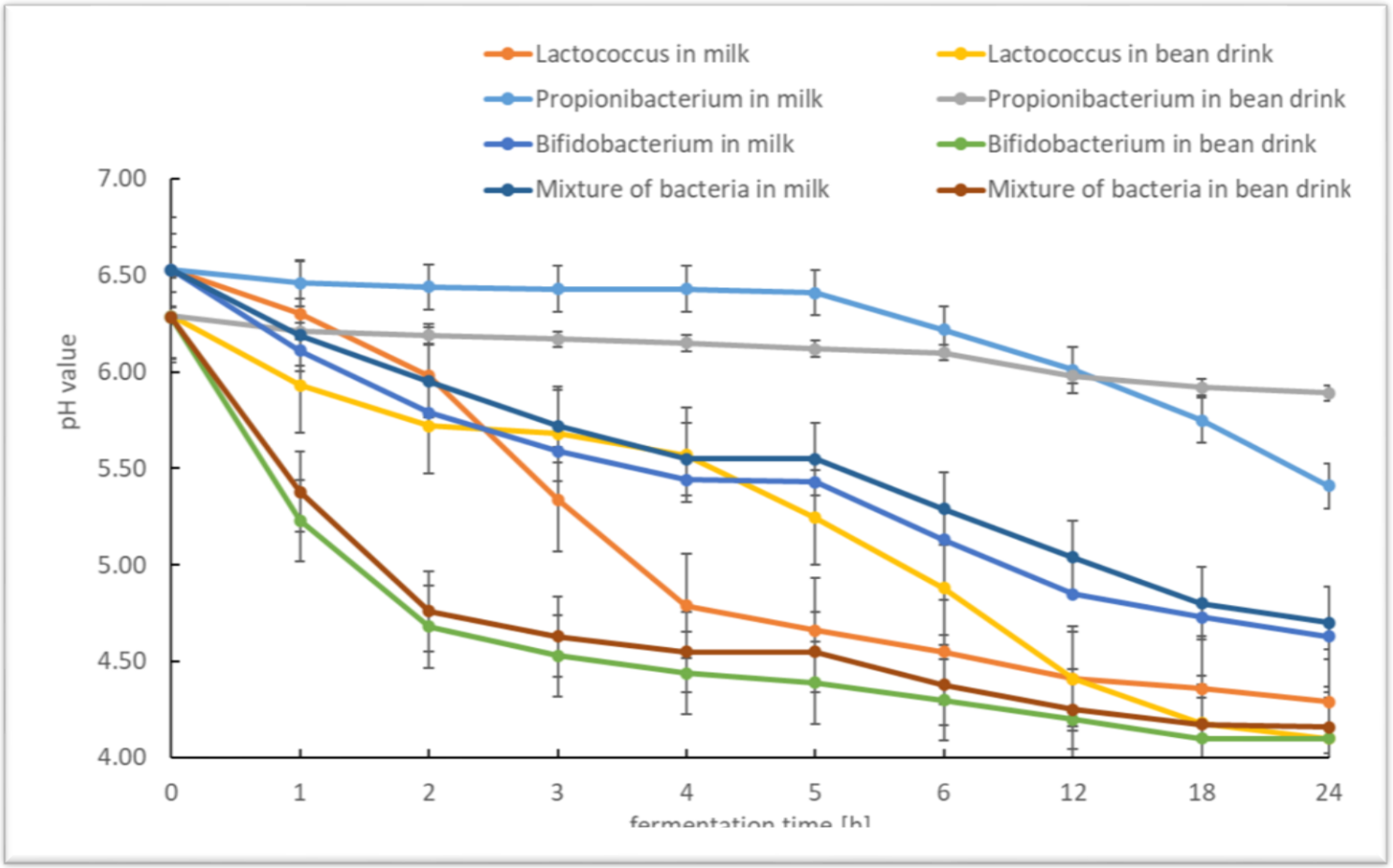
Materials and methods

- Fermentation was carried out at 30°C (for *P. freundenreichii* subsp. *shermanii* PS4 and *L. lactis* ATCC 11454) and at 37°C for 24 h (for *B. animalis* subsp. *lactis* Bb-12) for 24 h.

- The value of pH was measured during the fermentation, and the number of bacterial cells (plate method) and the content of selected B vitamins (HPLC-UV) were determined before and after the fermentation process.

- Two independent experiments were performed and each analysis was performed in duplicate.

Results - pH



- The highest increase in acidity was observed for beverages fermented by *L. lactis* ATCC 11454 (4.10-4.29 after 24 h) or *B. animalis* subsp. *lactis* BB-12 (4.10-4.63 after 24 h), regardless of the type of the fermented food matrix.

Results - pH

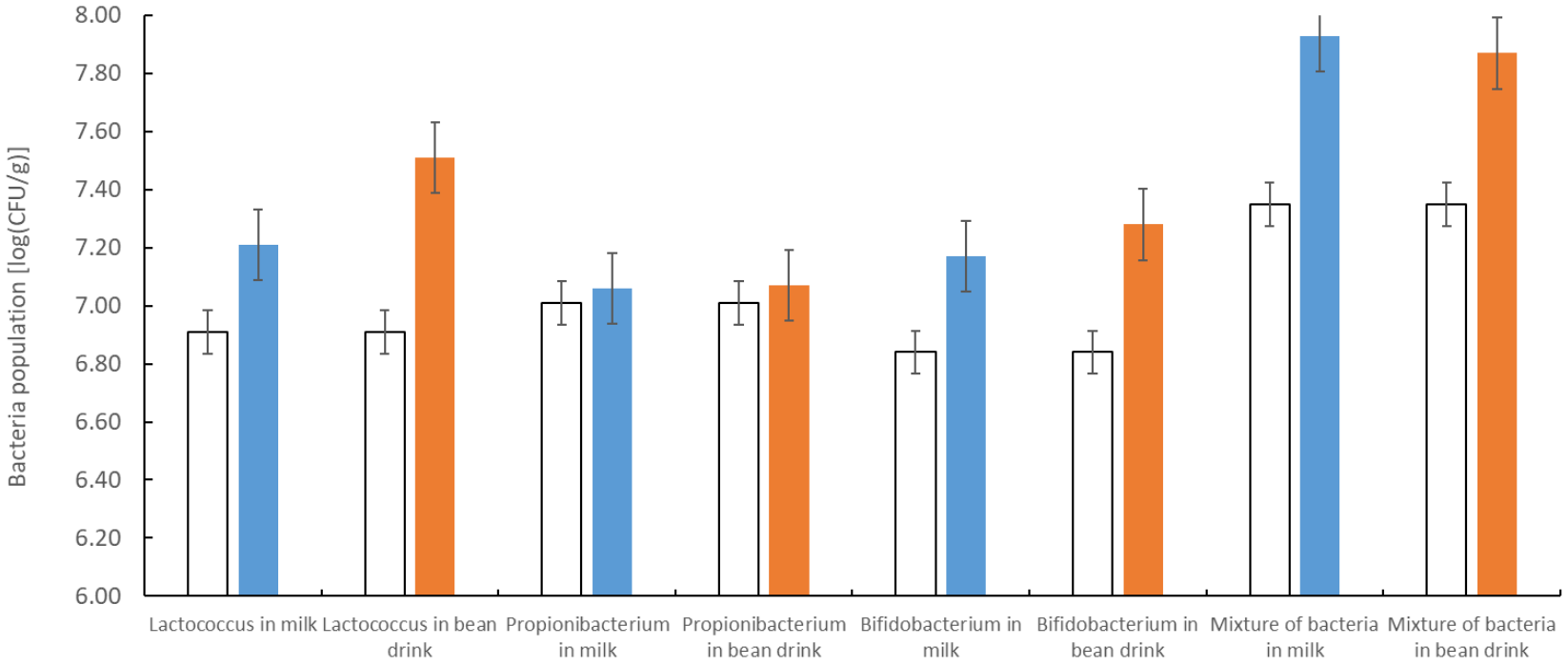
- In the case of beverages fermented by *P. freundenreichii* subsp. *shermanii* PS4 the final pH value was significantly higher than for the remaining samples of milk drinks (5.41) and bean drinks (5.89).

- On the other hand, the mixture of the studied strains turned out to be useful for the effective fermentation of both a milk drink (4.70) and a bean drink (4.16).

- The determined acidification curves indicate that the bean drink is a good matrix to ferment with *L. lactis* ATCC 11454, *B. animalis* subsp. *lactis* BB-12 or a mixture of these bacterial strains.

Results – bacterial population

- Changes in the population of live cells of the starter bacteria were observed in all the drink samples, confirming their metabolic activity.



Results – bacterial population

- In the case of *L. lactis* ATCC 11454 the initial was 6.91 log(cfu/ml), while the final population was 7.21 log(cfu/ml) and 7.51 log(cfu/ml) in a milk drink and the bean drink, respectively.
- For *B. animalis* subsp. *lactis* BB-12 initial cell population averaged 6.84 log(cfu/ml) and after fermentation the cell population averaged 7.17 log(cfu/ml) and 7.28 log(cfu/ml) in the milk drink and bean drink, respectively.
- The smallest changes in the cell population of the starter strains were observed in the drinks fermented with *P. freundenreichii* subsp. *shermanii* PS4 (7.01 log(cfu/ml) and 7.06 log(cfu/ml) before and after fermentation, respectively.
- The greatest changes in the population were recorded in drinks fermented by a mixture of strains - 7.35 log(cfu/ml) before fermentation, as well as 7.93 log(cfu/ml) and 7.87 log(cfu/ml) after fermentation of milk and bean drinks, respectively.

Results – vitamins

- Fresh cow's milk contained 0.400 µg of cyanocobalamin, 0.170 mg of riboflavin, 0.036 mg of thiamine, 0.100 mg of niacin and 0.050 mg of pyridoxine per 100 g.

- The detectable amounts of riboflavin (0.023 mg/100 g), thiamine (0.067 mg/100 g), niacin (0.220 mg/100 g) and pyridoxine (0.049 mg/100 g) were recorded before fermentation of the bean drink.

Results – vitamins

- After fermentation with strains of *P. freundenreichii* subsp. *shermanii* PS4 and *L. lactis* ATCC 11454 drinks based on Adzuki beans showed increased cyanocobalamin content.
- Meanwhile, *B. animalis* subsp. *lactis* BB-12 showed a relatively low synthesis capacity of this substance.

		L. lactis ATCC 11454		*P. freundenreichii* subsp. *shermanii* PS4		*B. animalis* subsp. *lactis* BB-12		Mixture of bacteria	
		milk drink	bean drink	milk drink	bean drink	milk drink	bean drink	milk drink	bean drink
Cyanocobalamin (μg/100 g)	initial	0,400±0,030	0,000±0,000	0,400±0,020	0,000±0,000	0,400±0,020	0,000±0,000	0,400±0,020	0,000±0,000
	final	0,150±0,005	0,015±0,001	0,360±0,020	0,070±0,003	0,240±0,020	0,050±0,002	0,320±0,020	0,080±0,004

Results – vitamins

- As for riboflavin, the bean drink was a less favorable environment for its biosynthesis than milk, and *L. lactis* ATCC 11454 turned out to be the best producer of this substance in milk.

		L. lactis ATCC 11454		*P. freundenreichii* subsp. *shermanii* PS4		*B. animalis* subsp. *lactis* BB-12		Mixture of bacteria	
		milk drink	bean drink	milk drink	bean drink	milk drink	bean drink	milk drink	bean drink
Riboflavin (mg/100 g)	initial	0,170±0,006	0,023±0,002	0,170±0,005	0,023±0,002	0,170±0,008	0,023±0,002	0,170±0,009	0,023±0,002
	final	0,190±0,008	0,013±0,001	0,150±0,008	0,013±0,001	0,130±0,006	0,015±0,001	0,170±0,009	0,016±0,001

Results – vitamins

- None of the fermented drinks showed an increase in thiamine, niacin, and pyridoxine content after the fermentation process (a reduction in the content of these substances has been found many times).

		L. lactis ATCC 11454		*P. freundenreichii* subsp. *shermanii* PS4		*B. animalis* subsp. *lactis* BB-12		Mixture of bacteria	
		milk drink	bean drink	milk drink	bean drink	milk drink	bean drink	milk drink	bean drink
Thiamine (mg/100 g)	initial	0,036±0,002	0,067±0,001	0,036±0,002	0,067±0,004	0,036±0,002	0,067±0,004	0,036±0,002	0,067±0,003
	final	0,023±0,001	0,024±0,003	0,026±0,002	0,024±0,002	0,024±0,002	0,030±0,002	0,024±0,002	0,013±0,001
Niacin (mg/100 g)	initial	0,100±0,005	0,220±0,020	0,100±0,005	0,220±0,020	0,100±0,005	0,220±0,020	0,100±0,005	0,220±0,020
	final	0,030±0,002	0,090±0,005	0,106±0,008	0,090±0,005	0,030±0,002	0,070±0,006	0,108±0,009	0,100±0,005
Pyridoxine (mg/100 g)	initial	0,050±0,002	0,053±0,003	0,050±0,003	0,053±0,003	0,050±0,003	0,053±0,003	0,050±0,003	0,053±0,002
	final	0,037±0,002	0,049±0,002	0,036±0,002	0,051±0,003	0,045±0,002	0,051±0,003	0,041±0,003	0,051±0,002

Cunclusion

- The research proved the possibility of using lactic acid fermentation and propionic acid fermentation in the production of fermented drinks, not only fermented milk, but also fermented drink obtained from germinated Adzuki beans.

- The tested starter cultures, both in the form of monocultures and a starter mixture, were able to ferment the milk and plant matrix well, maintaining the viability of bacterial cells, but without significantly increasing the content of substances showing the activity of B vitamins other than cyanocobalamin

Thank you for your attention