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Research Article

Evaluation of Cereals and Pseudocereals Suitability for the Development of New Probiotic Foods

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The aim of the present work was to evaluate suitability of cereals and pseudocereals for the development of new probiotic foods and to evaluate the stability of cereal and pseudocereal porridges fermented by probiotic strain *Lactobacillus rhamnosus* GG. Ten samples of cereals and pseudocereals obtained from Slovak mill house and markets were used in this work. A mixture of each cereal and pseudocereal samples with water (10% w/v) was inoculated after sterilization with coequal number of *Lactobacillus rhamnosus* GG, to obtain approximately 5-6 log colony form units per gram of suspensions. Fermentation was led at 37°C during 10 hours. Fermented suspensions were stored for 21 days at 5°C. Monitoring of cell counts, pH value, and concentration of organic acids during fermentation and storage was done.

1. Introduction

Cereals and pseudocereals belong to the most important food for the majority of mankind. They are a good source of saccharides (especially starch and fibres), proteins (with good quality of amino acids, except lysine in case of cereal grains), lipids (essential fatty acids, almost no presence of saturated fatty acids), vitamins (B group), and minerals (calcium, potassium, magnesium, ferrum, zinc, cuprum, and phosphorus) [1, 2]. On the other hand, antinutritive factors, such as phytic acid, tannins, polyphenols, and enzyme inhibitors, that decrease nutritional quality of grains by binding proteins, saccharides and mineral to complexes are naturally present in cereal and pseudocereal grains [3].

Cereals and pseudocereals are usually consumed in the form of bread, breakfast cereals, or cereal bars in developed countries. On the other hand, in developing countries the consumption of fermented cereals in the form of beverages, cakes, or porridges is common. These kinds of food would increase consumption of cereals and mainly pseudocereals in developed countries and moreover contribute to increase probiotic intake, in case of using probiotic strain for fermentation process [4].

Fermentation conduces to the enhancement of nutritional quality of end products via production of nutritive factors (e.g., biogenic amines and γ-aminobutyric acid) and reduction of antinutritive factors [3, 5]. Fermentation also makes for improvement of sensory quality of end products in comparison to raw materials by producing aroma compounds by lactic acid bacteria [3, 6] and to the extension of shelf life, through the production of antimicrobial compounds [7, 8].

Probiotics are live microorganisms which have a positive effect on consumer [9]. *Lactobacillus rhamnosus* GG is one of the most monitored probiotic strains. It belongs to Gram-positive, non-spore-forming, nonmotile, catalase-negative, facultatively anaerobic or microaerophilic, and mesophilic bacteria [10]. Metabolism of *Lb. rhamnosus* GG is facultatively heterofermentative; it can produce acetic acid, formic acid and ethanol, in addition to lactic acid, in lack of glucose in fermentation environment [11]. *Lb. rhamnosus* GG is highly tolerant to acidic environment of the stomach; it is able to survive in intestinal passage, adhere to intestinal mucosa, and colonize gastrointestinal tract after three days of consumption [12, 13]. It enhances human natural resistance and healthy digestive system and inhibits adhesion of some

pathogenic bacteria. It relieves syndromes of irritation of GI tract, atopic dermatitis, and cow milk allergy [14, 15].

In Slovak food market, probiotic foods are mainly milk based, and therefore probiotics are not available for all consumers, especially for those who suffer from milk allergies and intolerances. This was the reason why we focused our research on the selection of plant materials (cereals, pseudocereals, and legumes) on the bases of growth and metabolic characteristics of selected probiotic strain, *Lactobacillus rhamnosus* GG, which will lead to the development of new probiotic foods.

2. Materials and Methods

2.1. Starter Culture. The probiotic strain of *Lb. rhamnosus* GG was used in this work. It was provided by Dr. Salminen (University of Turku, Turku, Finland) through mediation of Dr. Lauková (State Veterinary and Food Institute, Košice, Slovakia).

2.2. Cereal and Pseudocereal Substrates. Ten samples of cereals and pseudocereals were used in this work. Rye flour (RF), rye grain (RG), barley flour (BF), and whole barley flour (WBF), amaranth flour (AF), buckwheat flour (BWF), whole buckwheat flour (WBWF), whole oat flour (WOF) were obtained from mill house (Mlyn Zrno, Šíšov, Slovak Republic); amaranth grain (AG, Primeal, Peaugres, France) and millet grain (MG, Marianna wholesale, Ivánka pri Dunaji, Slovak Republic) were obtained from market. Sixty grams of flours or milled and sieved grains were mixed with 540 mL of deionised water for preparation of fermentation substrates.

2.3. Fermentation and Storage. Substrates were inoculated after autoclaving for 15 min at 121°C and cooling down with overnight culture (MRS broth, 18 hours, 37°C) of *Lb. rhamnosus* GG to give approximately 5 to 6 log colony form units per gram of suspensions. Static fermentation was performed for 10 hours (according to the previous results) at 37°C. Samples for analyses were taken every 2 hours. Storage observations were carried out at 5°C for 21 days. Samples for analyses were taken every 2-3 days.

2.4. Viable Cell Enumeration. Enumeration of viable cells of *Lb. rhamnosus* GG was performed by estimation of colony forming unit number on MRS-agar plates according to the STN ISO 15214.

2.5. Determination of pH and Organic Acids. The pH of samples was measured by pH-meter CG 843 (Schott, Mainz, Germany). The quality and quantity of the produced organic acids were measured by isotachophoretic analysis by using the Isotachophoretic Analyser ZKI 01 (Villa Labeco, Spišská Nová Ves, Slovak Republic). Electrolytic system according to Kocková et al. [16] was used. Quantitative analysis was performed by calibration of standard solution of lactic, acetic, citric, formic, and succinic acids (Lachema, Brno, Czech Republic).

2.6. Estimation of Growth and Metabolic Parameters of Lb. rhamnosus GG. Growth curves of *Lb. rhamnosus* GG in each substrate were modelled with a mechanistic model of Baranyi and Roberts [17]. Growth and metabolic parameters were calculated from each curve.

2.7. Statistical Analyses. Each experiment was performed in three separate trials. Results represented means with standard deviations. Statistical analyses were carried out using Microsoft Excel 2007 (Microsoft, Redmond, WA, USA). Data were treated by ANOVA test with a least significant difference of 95%.

3. Results and Discussion

3.1. Fermentation. Fermentation of cereal and pseudocereal substrates for evaluation of their suitability for lactic acid fermentation was carried out. Cell counts, pH value, and organic acid concentrations were monitored in fixed period. Growth and metabolic curves are shown in Figure 1 and growth and metabolic parameters in Tables 1–5.

As it is shown in Figure 1, probiotic strain *Lb. rhamnosus* GG was able to grow and metabolize in each cereal and pseudocereal substrate during a ten-hour fermentation process. *Lb. rhamnosus* GG was able to grow from initial cell counts 5.04–6.46 log cfu g^{-1} to final 7.40–8.80 log cfu g^{-1} (Table 1), which is similar to density of *Lb. rhamnosus* GG reached in milk [10], in cereal water-based puddings [18], and maize porridges with barley addition [19]. The highest growth rate was calculated in case of amaranth flour (in which the longest lag phase was observed), 0.589 log cfu g^{-1} h^{-1}. The lowest growth rate was calculated for whole oat flour, 0.248 log cfu g^{-1} h^{-1} (Table 1). Growth rate of *Lb. rhamnosus* GG during fermentation of milk at 35°C was 0.653 log cfu mL^{-1} h^{-1} [10]. Higher growth rate was found during fermentation of amaranth milk- and water-based puddings and buckwheat milk-based pudding by *Lb. rhamnosus* GG [20]. Time to double ranged from 0.51 h (amaranth flour) to 1.21 h (whole oat flour). Lag phase, time for adapting microorganisms to new environment, ranged from 0.73 h (rye grain) to 3.88 h (amaranth flour). In substrate from millet grain, no lag phase was observed (Table 1).

During fermentation, organic acids production, as a result of metabolic activity of *Lb. rhamnosus* GG, was observed. This caused decreasing of pH values in each substrate. pH values dropped from initial 4.93–6.08 to final 4.31–5.99 (Figure 1, Table 2). Final pH value caused by metabolic activity of *Lb. rhamnosus* GG in MRS broth was 4.00 [21], in milk 6.50 [10] and in milk-based cereal puddings and maize porridges with barley under 4.00 [18, 19]. In case of substrates from amaranth grain, buckwheat flour, and millet grain, a long lag phase of reducing pH values (7.90, 8.70, and 6.50 h, resp.) was observed. In these substrates, higher rate of reducing pH (−0.355, −1.063, and −0.244 h^{-1}, resp.) was calculated. Rates of decreasing pH values in other substrates ranged from −0.003 h^{-1} (whole buckwheat flour) to −0.047 h^{-1} (barley flour) (Table 2). In amaranth

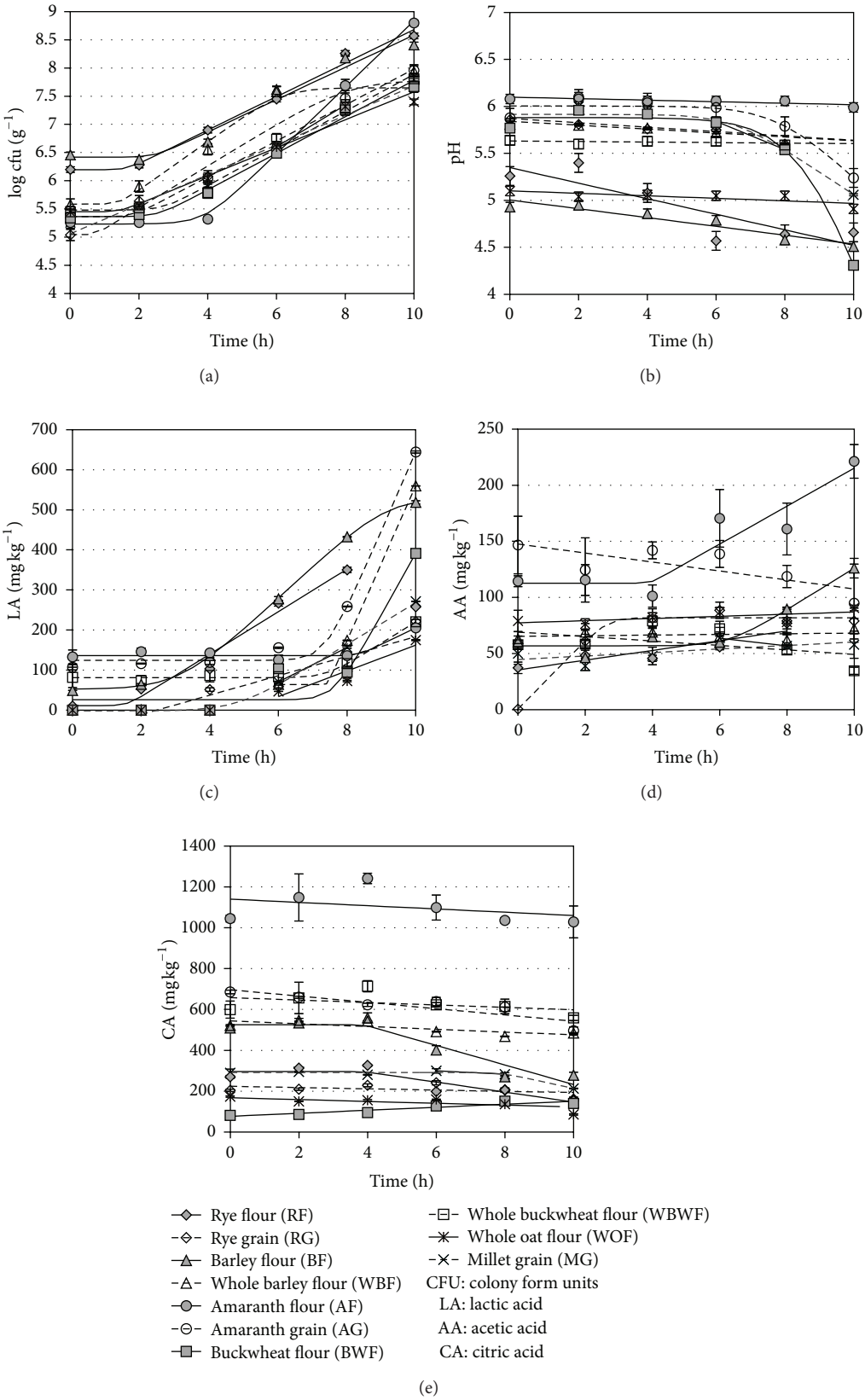

Figure 1: Evaluation of cell counts of *Lb. rhamnosus* GG (a) and changes in pH values (b) and concentration of lactic acid (c), acetic acid (d), and citric acid (e) during fermentation.

Table 1: Growth parameters of *Lb. rhamnosus* GG during fermentation of cereal and pseudocereal substrates.

Substrate	Gr [log cfu g^{-1} h^{-1}]	t_d [h]	λ [h]	N_0 [log cfu g^{-1}]	N_{max} [log cfu g^{-1}]
RF	0.297^{b}	1.01^{d}	1.64^{c}	6.20^{f}	8.57^{f}
RG	0.372^{c}	0.81^{c}	0.73^{a}	5.05^{a}	7.78^{c}
BF	0.287^{b}	1.05^{d}	2.44^{d}	6.42^{g}	8.41^{e}
WBF	0.430^{d}	0.70^{b}	1.42^{b}	5.59^{e}	7.65^{b}
AF	0.589^{e}	0.51^{a}	3.88^{e}	5.23^{b}	8.80^{g}
AG	0.319^{b}	0.94^{c}	1.79^{c}	5.36^{c}	7.96^{d}
BWF	0.324^{b}	0.93^{c}	2.53^{d}	5.36^{c}	7.67^{b}
WBWF	0.328^{b}	0.92^{c}	2.68^{d}	5.48^{d}	7.81^{c}
WOF	0.248^{a}	1.21^{d}	1.42^{b}	5.45^{d}	7.40^{a}
MG	0.263^{a}	1.15^{d}	—	5.06^{a}	7.67^{b}

RF: rye flour, RG: rye grain, BF: barley flour, WBF: whole barley flour, AF: amaranth flour, AG: amaranth grain, BWF: buckwheat flour, WBWF: whole buckwheat flour, WOF: whole oat flour, MG: millet grain.
Gr: growth rate, t_d: time to double, λ: lag phase, N_0: initial density of *Lb. rhamnosus* GG, N_{max}: final density of *Lb. rhamnosus* GG.
[a–g]Means within a column with different superscript letters are significantly different ($P < 0.05$).

Table 2: Parameters of pH value changes during fermentation of cereal and pseudocereal substrates with *Lb. rhamnosus* GG.

Substrate	k_{pH} [h^{-1}]	λ_{pH} [h]	pH_0	pH_{end}
RF	−0.083^{f}	—	5.35^{b}	4.62^{b}
RG	−0.029^{d}	—	5.87^{d}	5.60^{e}
BF	−0.047^{e}	—	5.00^{a}	4.78^{b}
WBF	−0.026^{d}	—	5.84^{d}	5.59^{e}
AF	−0.008^{b}	—	6.10^{e}	5.99^{f}
AG	−0.355^{g}	7.90^{b}	6.00^{e}	5.24^{d}
BWF	−1.063^{h}	8.70^{c}	5.88^{d}	4.31^{a}
WBWF	−0.003^{a}	—	5.63^{c}	5.59^{e}
WOF	−0.013^{c}	—	5.10^{a}	4.91^{c}
MG	−0.244^{g}	6.50^{a}	5.91^{d}	5.06^{c}

RF: rye flour, RG: rye grain, BF: barley flour, WBF: whole barley flour, AF: amaranth flour, AG: amaranth grain, BWF: buckwheat flour, WBWF: whole buckwheat flour, WOF: whole oat flour, MG: millet grain.
k_{pH}: rate of changes in pH values, λ_{pH}: lag phase of pH changes, pH_0: initial pH, pH_{end}: final pH.
[a–h]Means within a column with different superscript letters are significantly different ($P < 0.05$).

Table 3: Parameters of lactic acid concentration changes during fermentation of cereal and pseudocereal substrates with *Lb. rhamnosus* GG.

Substrate	k_{acid} [mg kg^{-1} h^{-1}]	λ_{acid} [h]	c_0 [mg kg^{-1}]	c_{end} [mg kg^{-1}]
RF	53.07^{d}	1.58^{a}	11.27^{b}	349.98^{d}
RG	24.31^{a}	2.50^{b}	0.00^{a}	221.16^{b}
BF	78.81^{e}	3.12^{c}	52.51^{c}	529.31^{f}
WBF	192.85^{g}	7.43^{e}	64.14^{d}	559.60^{g}
AF	40.42^{c}	8.30^{g}	136.40^{g}	205.15^{b}
AG	195.91^{g}	7.35^{e}	124.09^{f}	644.21^{h}
BWF	152.95^{f}	7.61^{f}	0.00^{a}	391.14^{e}
WBWF	53.22^{d}	7.38^{e}	81.05^{e}	220.40^{b}
WOF	32.02^{b}	5.00^{d}	0.00^{a}	174.66^{a}
MG	51.83^{d}	4.80^{d}	0.00^{a}	271.47^{c}

RF: rye flour, RG: rye grain, BF: barley flour, WBF: whole barley flour, AF: amaranth flour, AG: amaranth grain, BWF: buckwheat flour, WBWF: whole buckwheat flour, WOF: whole oat flour, MG: millet grain.
k_{acid}: rate of lactic acid concentration changes, λ_{acid}: lag phase of lactic acid concentration changes, c_0: initial concentration of lactic acid, c_{end}: final concentration of lactic acid.
[a–h]Means within a column with different superscript letters are significantly different ($P < 0.05$).

and buckwheat water- and milk-based pudding rates of pH value decreasing ranged from 0.144 to 0.317 h^{-1} [20].

The concentration of lactic acid at the beginning of fermentation process was established from 11.27 mg kg^{-1} in case of rye flour to 136.40 mg kg^{-1} in case of amaranth flour. In substrates from rye grain, buckwheat flour, whole oat flour, and millet grain concentration of lactic acid was under detection limit. The increase in lactic acid concentration was observed in all substrates (Figure 1, Table 3). The highest rate of production of lactic acid was calculated for whole barley flour and amaranth grain, 192.85 and 195.91 mg kg^{-1} h^{-1}, respectively. In these substrates, long lag phase of lactic acid production was observed, 7.43 and 7.35 h, respectively. Long lag phase was observed also in case of amaranth flour, buckwheat flour and whole buckwheat flour, ranged from 7.38 to 8.30 h. The slowest lactic acid production was calculated in substrate from rye grain, 24.31 mg kg^{-1} h^{-1}. Lactic acid level at the end of fermentation process ranged from 174.66 mg kg^{-1} (whole oat flour) to 644.21 mg kg^{-1} (amaranth grain) (Table 3). Lactic acid concentration after 18 hours fermentation by LAB in MRS broth was 36 g L^{-1} and in milk lower than 900 mg L^{-1} [21]. In maize porridges fermented by *Lb. rhamnosus* GG, lactic acid concentrations were 3500 mg kg^{-1} and 4000 mg kg^{-1}, respectively [19]. Lactic acid level in cereal water-based pudding fermented by *Lb. rhamnosus* GG was 2600 mg kg^{-1} and in milk-based

Table 4: Parameters of acetic acid concentration changes during fermentation of cereal and pseudocereal substrates with *Lb. rhamnosus* GG.

Substrate	k_{acid} [mg kg^{-1} h^{-1}]	λ_{acid} [h]	c_0 [mg kg^{-1}]	c_{end} [mg kg^{-1}]
RF	4.34^{f}	—	35.44^{b}	76.98^{c}
RG	30.59^{h}	—	0.50^{a}	79.01^{c}
BF	18.64^{g}	6.28^{b}	56.93^{d}	126.06^{e}
WBF	0.29^{c}	—	65.29^{e}	72.14^{c}
AF	16.92^{g}	3.95^{a}	112.77^{g}	221.21^{f}
AG	−3.99^{a}	—	147.53^{h}	94.87^{d}
BWF	—	—	—	—
WBWF	−1.99^{b}	—	69.12^{e}	34.61^{a}
WOF	0.97^{d}	—	77.35^{f}	90.64^{d}
MG	1.56^{e}	—	44.68^{c}	57.87^{b}

RF: rye flour, RG: rye grain, BF: barley flour, WBF: whole barley flour, AF: amaranth flour, AG: amaranth grain, BWF: buckwheat flour, WBWF: whole buckwheat flour, WOF: whole oat flour, MG: millet grain.
k_{acid}: rate of acetic acid concentration changes, λ_{acid}: lag phase of acetic acid concentration changes, c_0: initial concentration of acetic acid, c_{end}: final concentration of acetic acid.
[a–h]Means within a column with different superscript letters are significantly different ($P < 0.05$).

Table 5: Parameters of citric acid concentration changes during fermentation of cereal and pseudocereal substrates with *Lb. rhamnosus* GG.

Substrate	k_{acid} [mg kg^{-1} h^{-1}]	λ_{acid} [h]	c_0 [mg kg^{-1}]	c_{end} [mg kg^{-1}]
RF	−24.36^{g}	3.90^{a}	295.49^{d}	153.70^{c}
RG	−3.06^{b}	—	223.86^{c}	161.56^{c}
BF	−47.79^{i}	3.88^{a}	525.03^{e}	278.35^{e}
WBF	−6.80^{e}	—	543.68^{f}	484.96^{f}
AF	−8.02^{e}	—	1140.13^{h}	1028.97^{h}
AG	−15.40^{f}	—	696.34^{g}	496.19^{f}
BWF	7.43^{a}	—	76.71^{a}	140.23^{b}
WBWF	−5.97^{d}	—	657.78^{g}	559.08^{g}
WOF	−4.49^{c}	—	167.64^{b}	68.36^{a}
MG	−37.40^{h}	7.91^{b}	291.26^{d}	213.22^{d}

RF: rye flour, RG: rye grain, BF: barley flour, WBF: whole barley flour, AF: amaranth flour, AG: amaranth grain, BWF: buckwheat flour, WBWF: whole buckwheat flour, WOF: whole oat flour, MG: millet grain.
k_{acid}: rate of citric acid concentration changes, λ_{acid}: lag phase of citric acid concentration changes, c_0: initial concentration of citric acid, c_{end}: final concentration of citric acid.
[a–i]Means within a column with different superscript letters are significantly different ($P < 0.05$).

9800 mg kg^{-1} [18]. Rates of production of lactic acid in buckwheat and amaranth water-based puddings during 8 hours fermentation were 162 mg L^{-1} h^{-1} and 280 mg L^{-1} h^{-1}, respectively [20].

The rate of acetic acid production was slower compared to rate of lactic acid production, ranged from 0.29 mg kg^{-1} h^{-1} (whole barley flour) to 30.59 mg kg^{-1} h^{-1} (rye grain), which was good for final sensory quality of substrates. In case of amaranth grain and whole buckwheat flour, reduction in acetic acid level was observed and in substrate from buckwheat flour concentration of acetic acid was under detection limit during process (Figure 1, Table 4). Generally, production of acetic acid in our substrates was lower in compare to production of acetic acid in MRS broth or milk by the same probiotic strain [21].

Reducing in citric acid level was observed in all substrates, except those prepared from buckwheat flour. In case of rye flour, barley flour and millet grain, lag phase of reducing citric acid was observed and calculated, what caused higher rate of reducing citric acid, ranged from −24.36 to −47.79 mg kg^{-1} h^{-1}. In other substrates, rate of reducing citric acid ranged from −3.06 (rye grain) to −15.40 mg kg^{-1} h^{-1} (amaranth grain) (Figure 1, Table 5). Partial utilization of citric acid by *Lb. rhamnosus* GG was observed in maize porridges [19] and also in cereal water- and milk-based puddings [18].

Only small insignificant ($P > 0.05$) changes in formic and succinic acids content during fermentation were observed in all substrates (results are no shown), which concurs with the results of Helland et al. [19].

3.2. Storage. During storage at refrigerated temperature, changes in cell counts and metabolic activity were evaluated. For probiotic food, it is necessary to contain probiotic strain at certain level (6 log cfu per gram or millilitre of products, at least), and any metabolic activity of present microorganism is important.

As shown in Figure 2, density of *Lb. rhamnosus* GG after 21 days of storage was over the limit required for probiotic food, except substrate prepared from whole buckwheat flour, in which cell counts of *Lb. rhamnosus* GG decreased with growth rate −0.00517 log cfu g^{-1} h^{-1}. Visible reduction in density of *Lb. rhamnosus* GG was observed also in case of rye, barley, and amaranth flours (Figure 2, Table 6).

All fermented porridges were metabolically stabile except those prepared from amaranth flour, in which production of lactic acid (3.752 mg kg^{-1} h^{-1}) and reduction of citric acid (−0.732 mg kg^{-1} h^{-1}) and pH values (−0.00435 h^{-1}) continued, and from whole buckwheat flour, in which metabolic activity continued during first five days of the storage period (Figure 2, Tables 7, 8, 9, and 10). According to these results, we evaluated used substrates as metabolically stabile, except substrate prepared from amaranth flour, in which higher content of saccharides, proteins, and lipids were found [16]. Evaluation of stability and definition of shelf-life of fermented cereal products was done only in scarce research. Angelov et al. [22] estimated shelf-life of probiotic oat drink to 21 days as a result of 24-day observation during storage period.

4. Conclusion

According to our results, we can say that probiotic strain *Lb. rhamnosus* GG is able to grow and metabolize during the fermentation of cereal and pseudocereal substrates dealing to

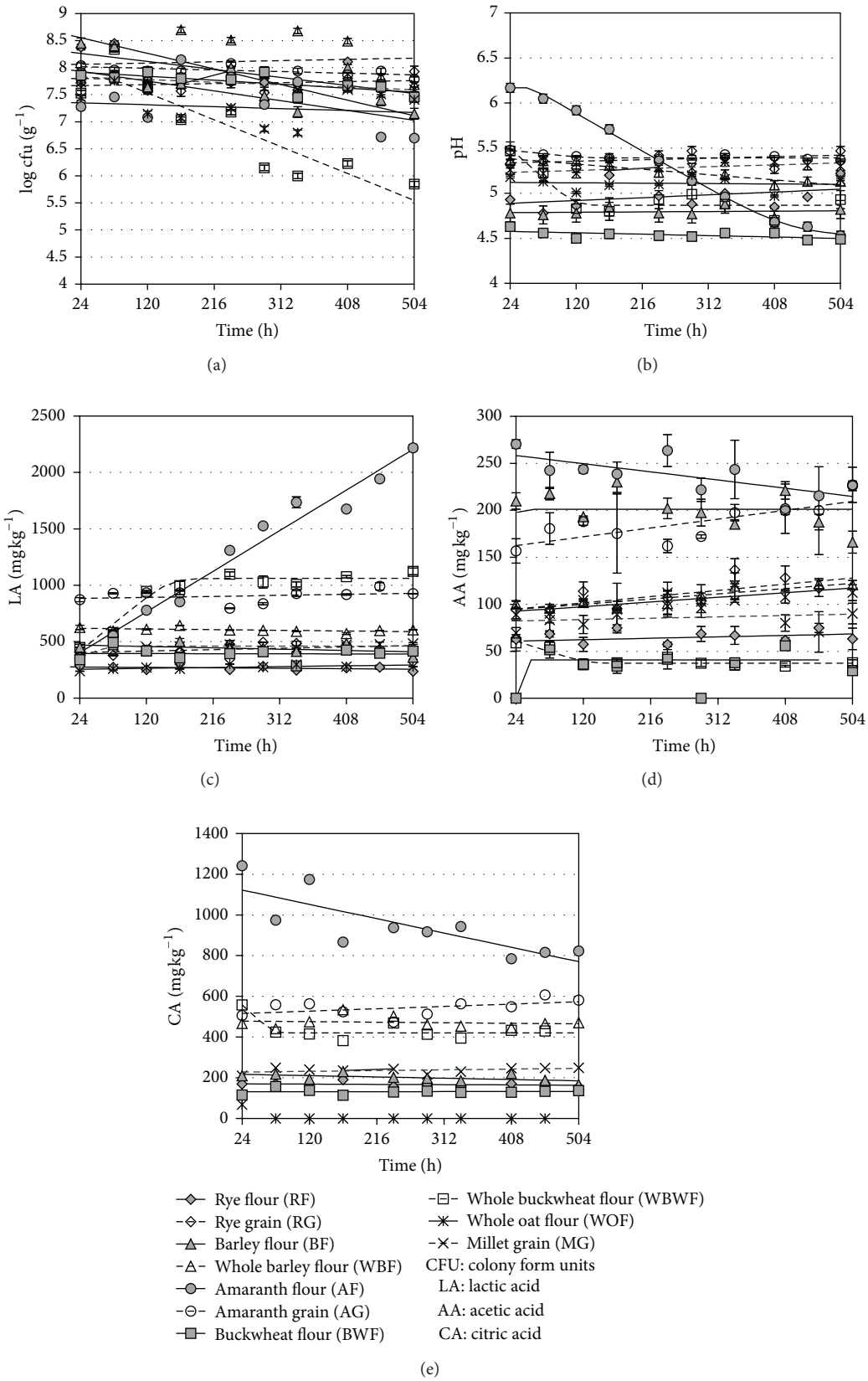

Figure 2: Evaluation of cell counts of *Lb. rhamnosus* GG (a) and changes in pH values (b), and concentration of lactic acid (c), acetic acid (d) and citric acid (e) during storage of fermented cereal and pseudocereal substrates.

Table 6: Growth parameters of *Lb. rhamnosus* GG during storage of fermented cereal and pseudocereal substrates.

Substrate	Gr [log cfu g^{-1} h^{-1}]	N_0 [log cfu g^{-1}]	N_{max} [log cfu g^{-1}]
RF	−0,00154^{d}	8,29^{e}	7,49^{d}
RG	0,00019^{i}	7,66^{b}	7,93^{g}
BF	−0,00300^{b}	8,59^{f}	7,15^{c}
WBF	0,00024^{j}	8,06^{d}	7,69^{e}
AF	−0,00188^{c}	7,96^{d}	6,70^{b}
AG	−0,00031^{h}	8,02^{d}	7,79^{f}
BWF	−0,00076^{e}	7,91^{d}	7,46^{d}
WBWF	−0,00517^{a}	7,80^{c}	5,85^{a}
WOF	−0,00037^{g}	7,35^{a}	7,41^{d}
MG	−0,00048^{f}	7,83^{c}	7,60^{e}

RF: rye flour, RG: rye grain, BF: barley flour, WBF: whole barley flour, AF: amaranth flour, AG: amaranth grain, BWF: buckwheat flour, WBWF: whole buckwheat flour, WOF: whole oat flour, MG: millet grain.
Gr: growth rate, N_0: initial density of *Lb. rhamnosus* GG, N_{max}: final density of *Lb. rhamnosus* GG.
$^{a–j}$Means within a column with different superscript letters are significantly different ($P < 0.05$).

Table 7: Parameters of pH changes during storage of fermented cereal and pseudocereal substrates.

Substrate	k_{pH} [h^{-1}]	pH_0	pH_{end}
RF	0,00033^{g}	4,89^{b}	5,22^{c}
RG	0,00018^{f}	5,34^{d}	5,47^{d}
BF	0,00005^{e}	4,78^{b}	4,82^{b}
WBF	−0,00061^{b}	5,38^{d}	5,13^{c}
AF	−0,00435^{a}	6,17^{e}	4,52^{a}
AG	−0,00077^{b}	5,48^{d}	5,39^{c}
BWF	−0,00016^{c}	4,58^{a}	4,49^{a}
WBWF	−0,00677^{a}	5,50^{d}	4,87^{b}
WOF	−0,00004^{d}	5,12^{c}	5,15^{c}
MG	0,00022^{f}	5,23^{c}	5,30^{c}

RF: rye flour, RG: rye grain, BF: barley flour, WBF: whole barley flour, AF: amaranth flour, AG: amaranth grain, BWF: buckwheat flour, WBWF: whole buckwheat flour, WOF: whole oat flour, MG: millet grain.
k_{pH}: rate of changes in pH values, pH_0: initial pH, pH_{end}: final pH.
$^{a–g}$Means within a column with different superscript letters are significantly different ($P < 0.05$).

Table 8: Parameters of lactic acid concentration changes during storage of fermented cereal and pseudocereal substrates.

Substrate	k_{acid} [mg kg^{-1} h^{-1}]	c_0 [mg kg^{-1}]	c_{end} [mg kg^{-1}]
RF	−0,041^{c}	276,52^{a}	236,41^{a}
RG	0,126^{g}	404,90^{c}	401,05^{d}
BF	−0,122^{a}	469,20^{e}	361,41^{c}
WBF	−0,060^{b}	617,95^{f}	603,81^{f}
AF	3,752^{i}	402,90^{c}	2217,50^{i}
AG	0,092^{f}	883,81^{g}	925,48^{g}
BWF	−0,011^{d}	395,75^{c}	415,53^{d}
WBWF	4,967^{j}	416,12^{d}	1122,91^{h}
WOF	0,079^{e}	255,74^{a}	279,09^{b}
MG	2,455^{h}	359,89^{b}	483,38^{e}

RF: rye flour, RG: rye grain, BF: barley flour, WBF: whole barley flour, AF: amaranth flour, AG: amaranth grain, BWF: buckwheat flour, WBWF: whole buckwheat flour, WOF: whole oat flour, MG: millet grain.
k_{acid}: rate of lactic acid concentration changes, c_0: initial concentration of lactic acid, c_{end}: final concentration of lactic acid.
$^{a–i}$Means within a column with different superscript letters are significantly different ($P < 0.05$).

Table 9: Parameters of acetic acid concentration changes during storage of fermented cereal and pseudocereal substrates.

Substrate	k_{acid} [mg kg^{-1} h^{-1}]	c_0 [mg kg^{-1}]	c_{end} [mg kg^{-1}]
RF	0,016^{d}	60,60^{b}	63,15^{b}
RG	0,069^{f}	94,91^{d}	117,07^{e}
BF	0,007^{c}	88,67^{d}	78,48^{c}
WBF	0,058^{e}	94,44^{d}	120,77^{e}
AF	−0,091^{b}	258,30^{f}	227,02^{f}
AG	0,098^{g}	162,46^{e}	225,97^{f}
BWF	6,793^{h}	0,05^{a}	40,87^{a}
WBWF	−0,236^{a}	60,94^{b}	37,78^{a}
WOF	0,052^{e}	92,55^{d}	117,28^{e}
MG	0,015^{d}	82,33^{c}	90,11^{d}

RF: rye flour, RG: rye grain, BF: barley flour, WBF: whole barley flour, AF: amaranth flour, AG: amaranth grain, BWF: buckwheat flour, WBWF: whole buckwheat flour, WOF: whole oat flour, MG: millet grain.
k_{acid}: rate of acetic acid concentration changes, c_0: initial concentration of acetic acid, c_{end}: final concentration of acetic acid.
$^{a–h}$Means within a column with different superscript letters are significantly different ($P < 0.05$).

improve sensory quality of end products. We also found out that fermented cereal and pseudocereal substrates were stable during the 21-day storage period, except those prepared from amaranth flour, due to its rich nutritional composition. We are able to summarize that selected cereals and pseudocereals are suitable for developing new probiotic foods, which is in our interest.

Conflict of Interests

The authors declare that they have no conflict of interests.

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Table 10: Parameters of citric acid concentration changes during storage of fermented cereal and pseudocereal substrates.

Substrate	k_{acid} [mg kg^{-1} h^{-1}]	c_0 [mg kg^{-1}]	c_{end} [mg kg^{-1}]
RF	−0,014^{d}	170,02^{b}	155,95^{a}
RG	—	—	—
BF	−0,066^{f}	216,76^{c}	165,06^{a}
WBF	−0,028^{e}	478,37^{d}	470,37^{d}
AF	−0,732^{g}	1121,94^{f}	822,96^{f}
AG	0,120^{b}	515,66^{d}	581,54^{e}
BWF	0,003^{a}	130,93^{a}	136,86^{a}
WBWF	−3,161^{h}	557,95^{e}	429,94^{c}
WOF	—	—	—
MG	0,037^{c}	228,06^{c}	249,15^{b}

RF: rye flour, RG: rye grain, BF: barley flour, WBF: whole barley flour, AF: amaranth flour, AG: amaranth grain, BWF: buckwheat flour, WBWF: whole buckwheat flour, WOF: whole oat flour, MG: millet grain.

k_{acid}: rate of citric acid concentration changes, c_0: initial concentration of citric acid, c_{end}: final concentration of citric acid.

$^{a–h}$Means within a column with different superscript letters are significantly different ($P < 0.05$).

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