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Effects of the Glucose Addition during Lactic Fermentation of Rice, Oat and Wheat Flours

Marianna Gallo[1,2*], Francesca Passannanti[1], Rosa Colucci Cante[1], Federica Nigro[2], Dana Salameh[1], Paola Schiattarella[3], Concetta Schioppa[3], Andrea Budelli[4], Roberto Nigro[1]

1- University of Naples Federico II, DICMAPI, Piazzale Tecchio 80, 80125, Naples, Italy.

2- University of Rome Niccolò Cusano, Engineering Department, Via Don Carlo Gnocchi, 3, 00166Rome, Italy.

3- ITP s.r.l., via Bisignano a Chiaia 68, 80121 Napoli.

4- KraftHeinz Innovation Center, NieuweDukenburgseweg 19, 6534 Nijmegen, Netherlands.

Abstract

Background and objective: Consumer interests in probiotic foods have increased in recent decades. Food industries respond to these growing interests by developing innovative products and guaranteeing high production efficiency. Cereals, due to their prebiotic nature, are good fermentable substrates; from which, potentially functional foods could be achieved. The aim of this study was to verify effects of D-glucose addition on fermentation of rice, oat and wheat flours.

Material and methods: Suspensions of 15% of cereals flours (rice, oat and wheat) in distilled water added with increasing glucose concentrations (2, 5, 7 and 10% w v^{-1}) were fermented by *Lactobacillus paracasei* CBA L74 for 24 h. Then, pH, microbial growth and lactic acid production were assessed.

Results and conclusion: Rice fermentation was not affected by glucose addition. For oat and wheat, addition of D-glucose increased bacterial concentration, as well as lactic acid production. In particular, the best growth was achieved by the addition of 2 and 5% of glucose. Furthermore, lactic acid concentration increased with increased glucose concentration. In conclusion, D-glucose addition seems to be unnecessary for the improvement of rice fermentation. On the contrary, oat and wheat fermentations need further available carbon sources for a better Lactobacillus growth and a higher lactic acid production.

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***Corresponding author:**
Marianna Gallo,
University of Naples Federico II, and University of Rome Niccolò Cusano, Italy.

Tel: +390817685943
Fax: +390815936936
E-mail: marianna.gallo@unina.it

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1. Introduction

Functional foods or foods with additional functions (e.g. health-promotion and disease prevention) have become a new challenge for the food industries. A particular class of functional foods is represented by the probiotics. Probiotics are defined as "live microorganisms which when administered in adequate amounts confer a health benefit to the host" [1]. Based on the literature, the number of lactic fermenting bacteria necessary to temporary colonize the intestine is at least 10^7 CFU g^{-1} [2]. Despite the fact that the most well-known functional foods are dairy products [3], nondairy products such as cereals may contain probiotics [4-6], demostrating their functional activities in several In vitro and In vivo experiment. Cereal flours (rice, oat and wheat) fermented with *Lactobacillus* (*L.*) *paracasei* CBA L-74 have shown abilities to decrease gliadin peptide entrance in Caco-2 cells and promote innate immunity peptides [7-9]. Similarly, a clinical trial has demonstrated the ability of rice flour fermented with a similar bacterial strain in prevention of common infectious diseases in children [10].

The aqueous suspensions of cereal flours are good examples of fermentable matrices since they are rich sources of interesting nutrients such as resistant starch, water-soluble and insoluble β-glucans and arabinoxylans.

These nutrients can help microbial growth as prebiotics, which can selectively stimulate growth and/or activity of a limited number of resident bacterial species in colon and hence improve the host health [11]. A previous study [12] has demonstrated that fermentation of three various water/cereal (rice, oat and wheat) suspensions with a solid content of 15% is possible. In fact, a value considered as sufficient for potentially functional foods was achieved at the end of the fermentation. However, rice flour seemed the best substrate for the microbial growth and lactic acid production. Based on these results, a laboratory-scale experimental study was carried out to understand if glucose addition could improve performance of the fermentation in microbial growth and lactic acid production. Increase of glucose concentration from 0 to 10% w v^{-1} was used in this experiment. The aim of this study was to assess effects of lactic fermentation on cereal flour and potential differences between different flours as well as the potential role of D-glucose on the process.

2. Materials and methods

2.1 Bacterial strain

The bacterial strain used as starter culture was *L. paracasei* CBA L74, (Heinz Italia SpA) with International Depository Accession Number of LMG P-24778. This strain was a Gram-positive homo-fermentative, facultative anaerobic bacteria. The bacteria was stored in freeze-dried form at-20°C and recovered at 37°C, 10 min before the fermentation process, using 0.9% sodium chloride solution. The inoculum bacterial density was 10^8 CFU ml^{-1}.

2.2 Rice, oat and wheat flours

Rice, oat and wheat flours were provided by Heinz Italia SpA. Before fermentation processes, each flour was subjected to heat treatment (120°C for 90 min) to decrease possible microbial loads.

2.3 Laboratory-scale bioreactor for fermentation

The experimental laboratory-scale bioreactor included four parts of a vessel, a mixing system, a system of thermal conditioning and a system for pH and temperature measurements. The vessel was a cylindrical Pyrex glass with an external jacket, allowing circulation of the service fluid. This was necessary to preserve constant temperature of the system. The inner height and diameter respectively included 18 and 10 cm and the external dimensions included 21 and 12 cm, respectively. The vessel maximum capacity was 1.5 L. The mixing system consisted of a stainless steel impeller with a pitched blade turbine and a Rushton turbine blade connected to a motor (a three-phase asynchronous electric motor with 0.25 hp/0.18 kW; 1310 rpm with a speed reducer; 170-880 rpm) that allowed the adjustment of stirring speed. Turbines allowed the axial and radial flows. Therefore, efficient mixing and good homogeneity of the substrates were achieved using a mixing speed of 180 rpm. This was demonstrated by preliminary mixing tests with a food dye (data not shown). Briefly, the best mixing speed (currently 180 rpm) was considered a speed, which allowed perfect distribution of the food dye in suspensions within 10 min. The vessel, the mixing system and the entire mechanism are respectively shown in Figures 1A, B and C. The service fluid used for controlling the fermentation temperature included distilled water (D.W.), thermally regulated using thermo-stated bath, (HAAKE G, USA) at 37°C. A Mettler Toledo probe was used (In Pro 3100, Mettler Toledo, USA) for the pH and temperature measurements.

Figure 1. The vessel and the mixing system. A) The vessel was made of Pyrex glass. It included a jacket vessel; in which, the thermo-stated water was circulated; B) The stainless steel impeller with two various types of turbines: a blade turbine to achieve axial flow and a Rushton turbine to achieve radial flow; and C) The complete equipment used for the laboratory-scale experiments.

2.4 Laboratory-scale fermentation protocol

The fermentation protocol was originally described by Gallo et al. [12] with the exception of added glucose. Briefly, a submerged fermentation under aerobic conditions was carried out in a 1.5-L fermenter with 1-L working volume. First, fermenter and impeller were sterilized at 121°C for 20 min using autoclave. Then, 150 g of each flour (rice, oat or wheat flour) were treated using

dry heat at 120°C for 90 min. Treated flour was mixed with 0.850 L of sterilized D.W. with addition of increasing glucose concentration (0, 2, 5, 7 and 10% w v^{-1} of D-glucose) (Oxoid, UK). This was subjected to a tyndallisation process with two consecutive cycles of heating (90°C) and cooling (37°C) to guarantee the decrease of contaminants [12,13]. During the following fermentation process, temperature of the substrate was preserved at 37°C and the fermentation process was stopped after 24 h. A similar protocol was used for the three flours.

2.5 Analytical methods

Samples were aseptically collected from the bioreactor at various fermentation times (t_0, t_4, t_{14}, t_{20}, t_{24} after the inoculum phase). These samples were used for the pH measurement and microbiological and chemical analyses. Samples were cultured on MRS agar (Oxoid, UK) Petri dishes after serial dilutions and incubated at 37°C for 48 h under anaerobic conditions using special anaerobic kits (Anaerogen Compact, Oxoid, UK). To exclude the contaminants, samples were cultured on Petri dishes prepared with PCA (bacteriological agar, yeast extract, peptose peptone and D-glucose) (Oxoid, USA), MacConkey agar (Oxoid, USA) and gelatine peptone agar (Biolife, Italy). The pH analysis was carried out using In Pro 3100 Probe (Mettler Toledo, USA). Lactic acid concentration was assessed using high performance liquid chromatography (HPLC) (Agilent Technologies 1100, USA) equipped with a C18 Column (Agilent Zorbax C18 4.6 × 150 mm, pore size of 80 A) and a visible/UV detector. The eluent was 0.1 M $NH_4H_2PO_4$ at a flow rate of 0.8 ml min^{-1} and the mobile phase was ammonium phosphate with pH of 2.7 with detection at 218 nm. The analysis temperature was 30°C. Secondary acids such as butyric, acetic and propionic acids were assessed using gas chromatography (Agilent Technologies 6890, USA) equipped with a capillary Poraplot Q Column (25 m × 0.32 mm). The flow rate was 200 ml min^{-1} and the mobile phase was helium gas.

2.6 Statistical analysis

Each experiment was carried out in triplicate. Statistical analysis was carried out using GraphPad Prism Software v.7.0a (San Diego, CA, USA). Mean and standard deviation of the results were calculated and their significance was assessed by Student's t-test. Results were reported as significant when $P \leq 0.05$.

3. Results and discussion

Although dairy foods are the most common substrates used for probiotic production, cereal based foods offer valuable alternatives. Therefore, three various flours (rice, oat and wheat flours) were fermented. The *L. paracasei* CBA L74 was used a fermenting culture. This strain was a homo-fermentative bacterium, meaning that the product is only lactic acid. To verify the success of flour fermentation, lactic acid concentration, pH value and *L. paracasei* growth were assessed.

3.1 Rice flour with increasing glucose concentration

The pH value, bacterial growth and lactic acid production from rice flour are shown in Figure 2 and Table 1. Comparison of results between the rice fermentations with no glucose and with 2% of glucose showed no significant differences in pH value, microbial growth and lactic acid production, as shown in Figures 2A, 2B and 2C, respectively. In both conditions (with no glucose and with 2% of glucose), the statistically significant differences were recorded at t_4 ($P<0.05$), t_{14} ($P<0.01$) and t_{20} ($P<0.05$) regarding bacterial growth and at t_{14} ($P<0.01$) and t_{20} ($P<0.05$) regarding lactic acid production. However, no differences were recorded at the end of the process (t_{24} pH value of 3.42 ±0.044 with no added glucose and 3.42 ±0.190 with 2% of added glucose; t_{24} bacterial growth of 9.5×10^8 CFU ml^{-1} $\pm 1.33 \times 10^8$ with no added glucose and 1×10^9 CFU ml^{-1} $\pm 2.57 \times 10^8$ with 2% of added glucose, starting with an initial bacterial load of nearly 5×10^6 CFU ml^{-1}; lactic acid production of 3100 mg l^{-1} ±134.55 with no added glucose and 3200 mg l^{-1} ±94.79 with 2% of added glucose). Moreover, presence of the secondary acids (acetic, propionic and butyric acids) was assessed. No significant quantities of these acids were detected (data not shown), demonstrating absence of the contaminants. Considering no differences between the fermentations with no added glucose and that with 2% of added glucose, no other experiments with increasing glucose concentrations were reported as useful.

Table 1. Summary of the results from the rice fermentation without or with 2% of added glucose

		Rice	
	Time (h)	0%	2%
pH	0	6.32 ± 0.14	6.04 ± 0.26
	24	3.42 ± 0.04	3.42 ± 0.19
Growth (CFU ml^{-1})	0	$5.0\times10^6 \pm 6.0\times10^5$	$6.0\times10^6 \pm 5.5\times10^5$
	24	$9.5\times10^8 \pm 1.33\times10^8$	$1.0\times10^9 \pm 2.57\times10^8$
Lactic Acid (mg l^{-1})	0	0	0
	24	3100 ±134.55	3200 ± 94.80

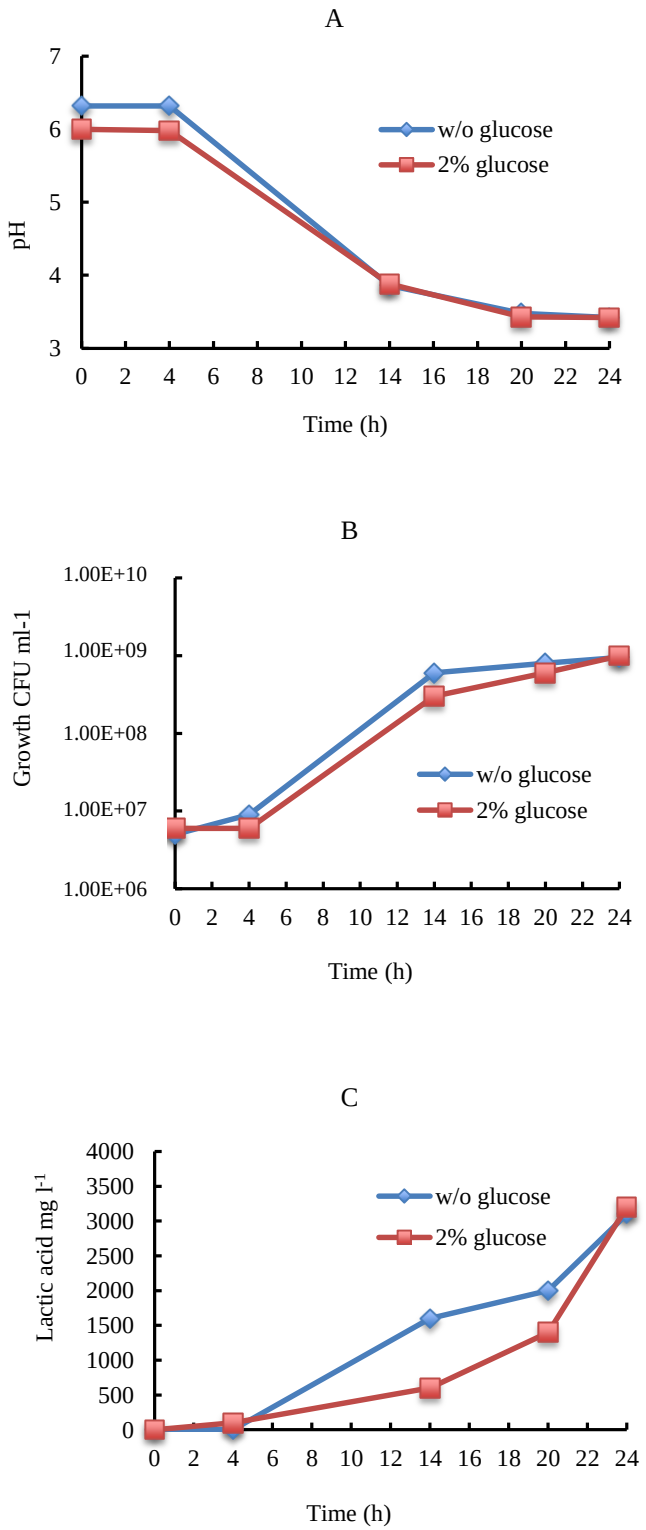

Figure 2. Analytical results from fermentation of the rice. A) The pH values of the rice fermentation with no glucose addition and with 2% of added glucose; B) The bacterial load (CFU ml^{-1}) of the rice fermentation with no glucose addition and with 2% of added glucose; and C) The lactic acid production (mg l^{-1}) during rice fermentation with no glucose addition and with 2% of added glucose. Each result was the mean value of a triplicate analysis, Student t-test; *P<0.05; **P<0.01

3.2 Oat flour with increasing glucose concentration

The pH value, bacterial growth and lactic acid production from oat flour are shown in Figure 3 and Table 2. Comparison of results from oats with no added glucose and with 2% of added glucose showed multiple differences, which led to test glucose concentrations greater than 2%. To assess effects of glucose addition (5-10%), bacterial growth and lactic acid concentration were assessed at t_0 and t_{24} and the values were compared with the values of 0 and 2% of glucose. The pH results are demonstrated in Figure 3a. All fermentations included an initial pH of nearly 6.2.

At t_{24}, oat fermentations with 0, 2 and 5% of added glucose reached similar pH values (0%, 4.51 ±0.11; 2%, 4.51 ±0.097 and 5%, 4.56 ±0.015), while fermentations with 7 and 10% of added glucose reached a lower value (7%, 3.84 ±0.026 and 10%, 3.84 ±0.067). Differences between the fermentations with lower glucose concentrations (0, 2 and 5%) and those with higher glucose concentrations (7 and 10%) were statistically significant (P<0.001) (Figure 3B).

Results of the microbial growth are shown in Figure 3C. Significant differences were seen at t_{24} between the no added glucose condition and all other conditions (0%, 2.00 × 10^8 CFU ml^{-1} ±3.79 × 10^7; 2%, 5.47 × 10^9 CFU ml^{-1} ±5.86 × 10^8; 5%, 8.1 × 10^9 CFU ml^{-1} ±4.36 × 10^8; 7%, 1.29 × 10^9 CFU ml^{-1} ±4.52 × 10^8 and 10%, 1.53 × 10^9 CFU ml^{-1} ±6.2 × 10^8). Statistically significant differences were reported for all the results, except for the difference between the fermentations with 7 and 10% of glucose (Figure 3D). Interestingly, a higher bacterial load was reached at t_{24} by adding 5% of glucose. For the concentrations of glucose greater than 5%, the final bacterial load decreased significantly. However, this load was still higher than that observed when no glucose was added. Therefore, oat fermentation possibly needs addition of glucose. However, glucose concentrations greater than 5% result in substrate inhibition, at least when bacterial growth is concerned. Results of the lactic acid production are shown in Figure 3E. As the glucose concentration increased; lactic acid production increased with statistically significant differences (Figure 3F). Starting all conditions with the absence at t_0 of lactic acid, they reached t_{24} 2000 mg l^{-1} ±94.98 when no glucose was added and 3300 mg l^{-1} ±132.64, 3850 mg l^{-1} ±147.42, 4500 mg l^{-1} ±196.22 and 5800 mg l^{-1} ±314.22 when 2, 5, 7 and 10% of glucose were respectively added. This could explain the lowest pH levels observed at t_{24}, when 7 and 10% of glucose were added to the oat. In this study, presence of the secondary acids (acetic, propionic and butyric acids) was assessed. No significant quantities of these acids were seen (data not shown), proving absence of the contaminants.

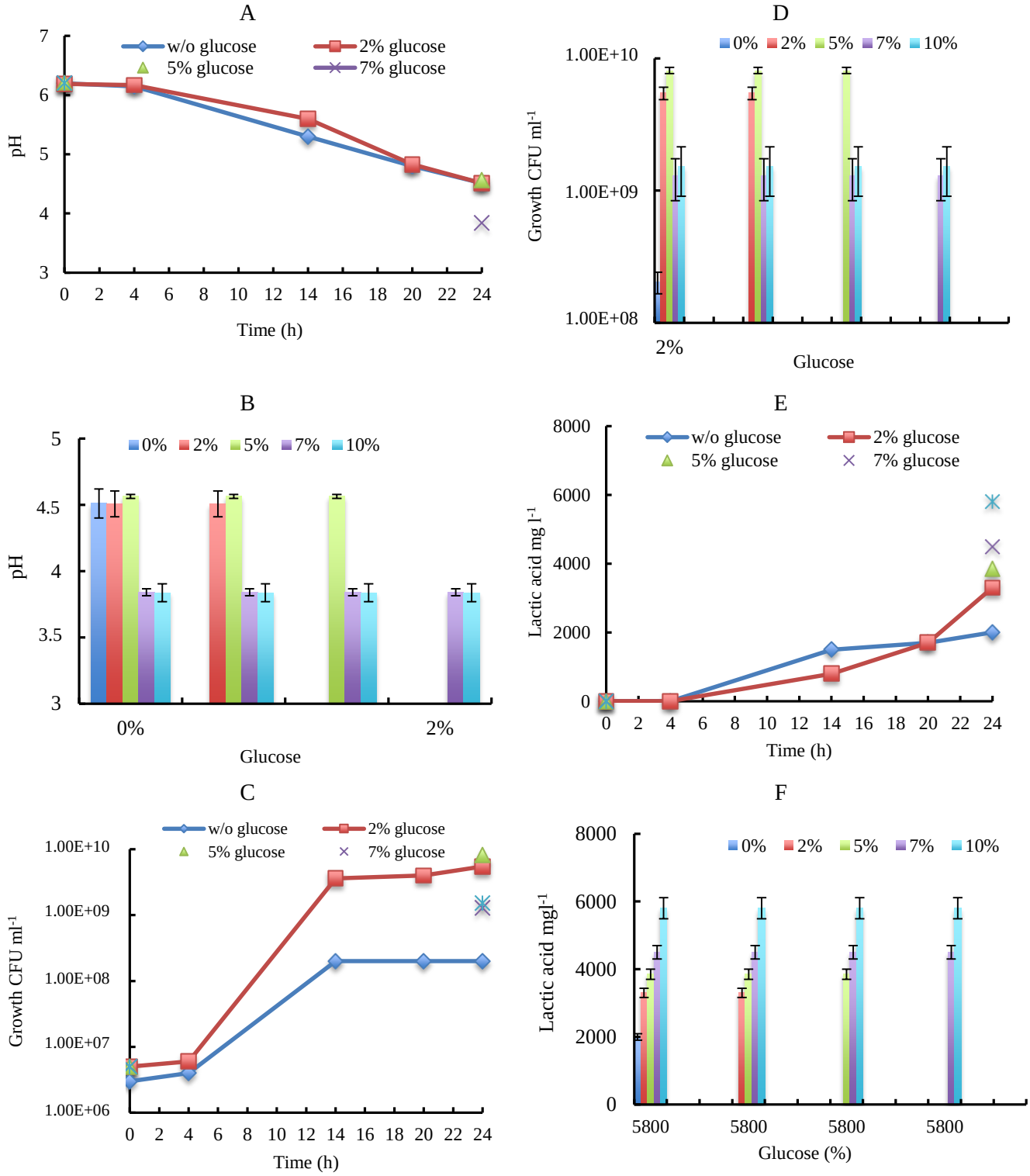

Figure 3. Analytical results of the fermentation of oat flour with increasing added glucose concentration. A) The pH values of the oat fermentation with 0, 2 (t_0, t_4, t_{14}, t_{20}, t_{24}), 5, 7 and 10% (t_0, t_{24}); B) Statistical results by the comparison between t_{24} pH values of the glucose concentrations; C) Bacterial load (t_0) and growth of the oat flour fermented with 0, 2 (t_0, t_4, t_{14}, t_{20}, t_{24}), 5, 7 and 10% (t_0, t_{24}) of glucose; D) Statistical results by the comparison of t_{24} bacterial load achieved using each glucose concentration with t_{24} bacterial load achieved using other glucose concentrations; E) Lactic acid production (mg l^{-1}) in the oat flour fermented with 0, 2 (t_0, t_4, t_{14}, t_{20}, t_{24}), 5, 7 and 10% (t_0, t_{24}) of glucose; and F) Statistical results by the comparison of t_{24} lactic acid produced using each glucose concentration with t_{24} lactic acid produced using other glucose concentrations. To make a statistically significant experimental campaign, each experiment was carried out in triplicate, Student t-test; *p< 0.05; **P<0.01; ***P<0.001

Table 2. Summary of the results from the oat fermentation with increasing glucose addition

		Oat				
	Time (h)	0%	2%	5%	7%	10%
pH	0	6.20 ± 0.04	6.19 ± 0.025	6.20 ± 0.021	6.20 ±0.036	6.20 ± 0.032
	24	4.51 ± 0.11	4.51 ± 0.097	4.56 ± 0.015	3.84 ± 0.026	3.84 ± 0.067
Growth (CFU ml^{-1})	0	$3.0\times10^6 \pm 7.4\times10^5$	$5.0\times10^6 \pm 3.8\times10^5$	$5.0\times10^6 \pm 3.6\times10^5$	$5.0\times10^6 \pm 3.1\times10^5$	$5.0\times10^6 \pm 3.2\times10^5$
	24	$2.0\times10^8 \pm 3.8\times10^7$	$5.5\times10^9 \pm 5.86\times10^8$	$8.1\times10^9 \pm 4.4\times10^8$	$1.3\times10^9 \pm 4.5\times10^8$	$1.5\times10^9 \pm 6.2\times10^8$
Lactic Acid (mg l^{-1})	0	0	0	0	0	0
	24	2000 ± 94.98	3300 ± 132.64	3850 ± 147.42	4500 ± 196.22	5800 ± 314.22

Differences with no statistical significance were recorded when glucose was added (Figure 4D), verifying the use of glucose for a better microbial growth in wheat fermentation. No statistically significant differences were seen between 7 and 10% of added glucose. Similar to oat, a higher t_{24} bacterial load was reached by adding 5% of glucose. For glucose concentrations greater than 5%, the final bacterial load significantly decreased. The lactic acid production results are shown in Figure 4E. The initial lactic acid concentration was 0 mg l^{-1}. At t_{24}, production of 2300 mg l^{-1} ±174.51 was achieved when no glucose added and 3500 mg l^{-1} ±116.66, 5850 mg l^{-1} ±94.18, 5860 mg L^{-1} ±32.88 and 8000 mg l^{-1} ±133.69 when 2, 5, 7 and 10% of glucose were used, respectively. As glucose concentration increased, lactic acid production increased with statistically significant differences between the increasing concentrations, except between fermentations with 5 and 7% of glucose (Figure 4F). Presence of the secondary acids (acetic, propionic and butyric acids) was assessed. No significant quantities of these acids were seen (data not shown), demonstrating absence of the contaminants.

3.3 Wheat flour with increasing glucose concentration

The pH values, bacterial growth and lactic acid production with wheat flour are shown in Figure 4 and Table 3. Comparison of results from wheat with no added glucose and with 2% of added glucose showed several differences, which led to test concentrations of glucose greater than 2%. To assess effects of glucose addition (5-10%), bacterial growth and lactic acid concentration were assessed at t_0 and t_{24}. These values were compared to those with 0 and 2% of glucose. The pH results are shown in Figure 4A. For all conditions, pH value at t_0 included nearly 6.2. Furthermore, significant differences were seen at t_{24} between the fermentation with no glucose addition and all other conditions (pH values at t_{24} included 4.1 ±0.1 for 0%; 3.26 ±0.27 for 2%; 3.3 ±0.27 for 5%; 3.20 ±0.21 for 7% and 3.5 ±0.2 for 10% of the added glucose) (Figure 4B).

Results of the microbial growth are demonstrated in Figure 4C. In general, the bacterial load for all conditions 5×10^6 CFU ml^{-1}. The bacterial load at t_{24} included 3×10^8 CFU ml^{-1} $\pm 4.58 \times 10^7$ when no glucose was added and 4.5×10^9 CFU ml^{-1} $\pm 4.36 \times 10^8$ for 2%, 8×10^9 CFU ml^{-1} $\pm 4.51 \times 10^8$ for 5%, 2×10^9 $\pm 3.22 \times 10^8$ for 7% and 1.53×10^9 CFU ml^{-1} $\pm 5.13 \times 10^8$ for 10% of added glucose, respectively.

Table 3. Summary of the results from the wheat fermentation with increasing glucose addition

		Wheat				
	Time (h)	0%	2%	5%	7%	10%
pH	0	6.0 ± 0.10	6.0 ± 0.10	6.2 ± 0.07	6.2 ± 0.08	6.2 ± 0.05
	24	4.10 ± 0.10	3.26 ± 0.27	3.3 ± 0.27	3.20 ± 0.21	3.5 ± 0.2
Growth	0	$6.0\times10^6 \pm 9.3\times10^5$	$4.4\times10^6 \pm 4.1\times10^5$	$5.1\times10^6 \pm 6.6\times10^5$	$5.1\times10^6 \pm 1.0\times10^6$	$5.1\times10^6 \pm 6.7\times10^5$
(CFU ml^{-1})	24	$3.0\times10^8 \pm 4.6\times10^7$	$4.5\times10^9 \pm 4.4\times10^8$	$8.0\times10^9 \pm 4.5\times10^8$	$2.0\times10^9 \pm 3.2\times10^8$	$1.5\times10^9 \pm 5.1\times10^8$
Lactic Acid	0	0	0	0	0	0
(mg l^{-1})	24	2300 ± 174.51	3500 ± 116.66	5850 ± 94.18	5860 ± 32.88	8000 ± 133.69

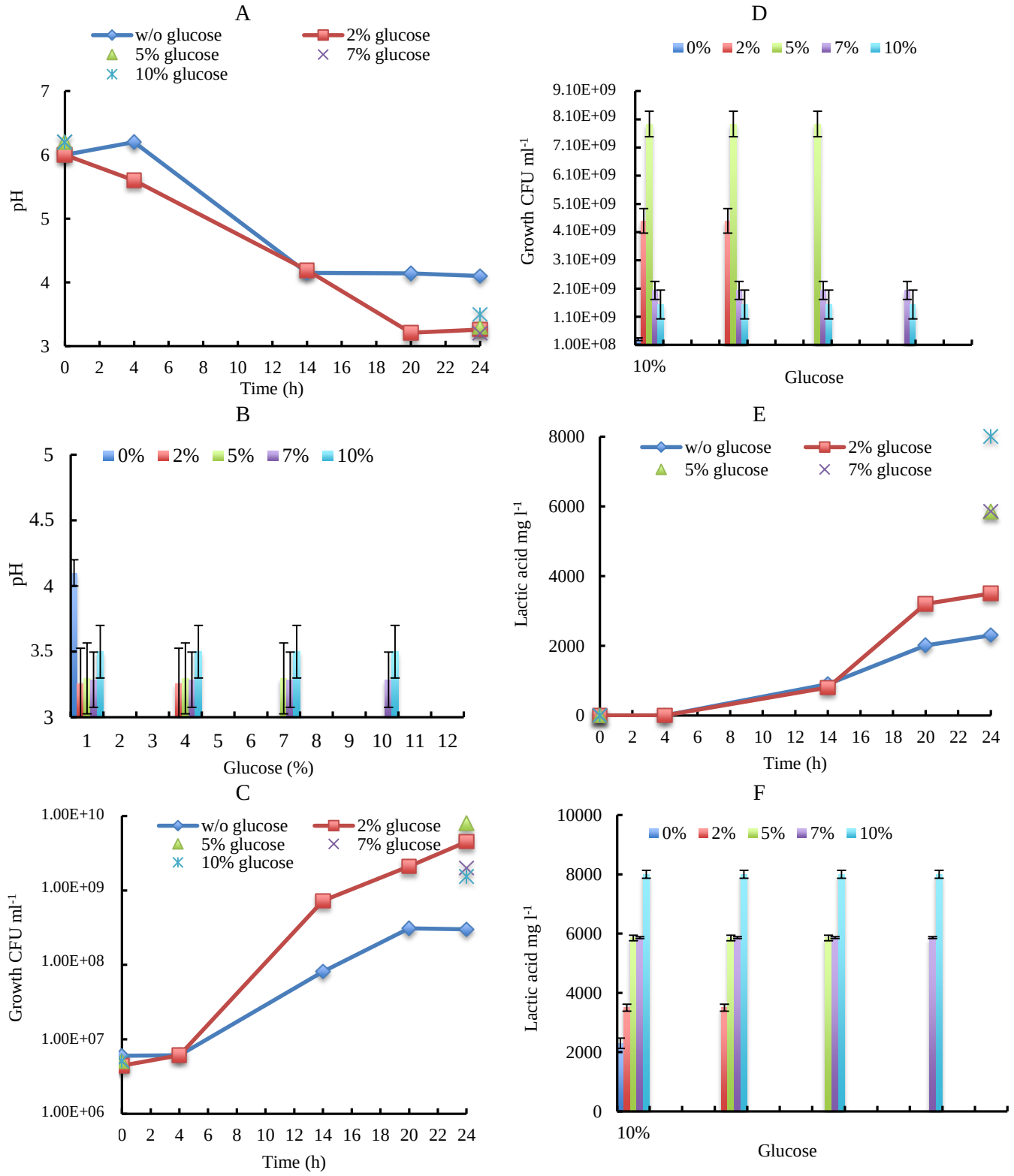

Figure 4. Analytical results of the fermentations of wheat flour with increasing added glucose concentration. A) The pH values of the wheat fermentation with 0, 2 (t_0, t_4, t_{14}, t_{20}, t_{24}), 5, 7 and 10% (t_0, t_{24}) of glucose; B) Statistical results by the comparison of t_{24} pH value using each glucose concentration with t_{24} pH value using other glucose concentrations; C) Bacterial load (t_0) and growth in wheat fermented with 0, 2 (t_0, t_4, t_{14}, t_{20}, t_{24}), 5, 7 and 10% (t_0, t_{24}) of glucose; D) Statistical results by the comparison of t_{24} bacterial load achieved using each glucose concentration with t_{24} bacterial load achieved using other glucose concentrations; E) Lactic acid production (mg l^{-1}) of the wheat flour fermented with 0, 2 (t_0, t_4, t_{14}, t_{20}, t_{24}), 5, 7 and 10% (t_0, t_{24}) of glucose; and F) Statistical results by the comparison of t_{24} lactic acid produced using each glucose concentration with t_{24} lactic acid produced using other glucose concentrations. Each experiment was carried out in triplicate, Student t-test; *P<0.05; **P<0.01; ***P<0.001

3.4 Comparison between rice, oat and wheat fermentations with increasing glucose concentration

Comparison of the results from experiments with no glucose addition to rice, oat and wheat flours with the results from the addition of 2% w v^{-1} of glucose showed an improved process for the oat and wheat flours but not for the rice. Therefore, experiments continued on oat and wheat flours, investigating increased concentration of D-glucose (5, 7 and 10%). In this preliminary study, only two sampling times of t_0 and t_{24}were used. By the addition of 2% of D-glucose, Lactobacillus growth on the oat and wheat flours was a log higher, compared to that with no glucose addition. This was not observed at the end of the rice flour fermentation. The lack of bacterial growth promotion could be due to several factors. First, rice starch granules include the smallest size [14,15] and, as suggested by Bhatty and Vasanthan [16], small granules are hydrolyzed more rapidly than large granules by α-amylase, which could make rice starch more accessible than oat and wheat starch to hydrolysis. Second, high amylose starches are particularly resistant to hydrolysis [17] and rice seems to include a smaller amylose content (nearly 21-25% w w^{-1} on a total starch dry basis) [18] than that oat (nearly 27.5-29.8% w w^{-1} on a total starch dry basis) [19] and wheat (nearly 25.6% w w^{-1} on total starch dry basis) [18] do. These data of granule size and amylose content could explain easier hydrolysis of the rice starch. Another factor that facilitates greater accessibility of the rice starch could be linked to its crystalline type. The rice starch is composed of an A structure, while oat and wheat starches are composed of two A and B types of the crystal structures. It has been reported [16,20] that type B starches resist enzymatic hydrolysis, compared to that type A starches do. This could explain why no differences were seen with or without glucose addition at the end of the rice fermentation in the current study. In contrast, the higher difficulty in hydrolyzing oat and wheat granules was possibly linked to D-glucose. For the lactic acid production at the end of the fermentations with no glucose addition, the highest concentration was reported in rice flour (3100 mg l^{-1}), while significantly lower concentrations were reported in oat (2000 mg l^{-1}) and wheat (2300 mg l^{-1}) flours. By the addition of 2% w v^{-1} of D-glucose, results changed drastically since lactic acid production increased to 3300 mg l^{-1} for oat and to 3500 mg l^{-1} for wheat flours. However, no similar results were seen in the rice flour (3200 mg l^{-1}). This was possibly due to the growth of Lactobacillus. Assessing effects of increased glucose concentration of D-glucose (0-10%) on oat and wheat flours demonstrated improved bacterial growth. within a certain threshold for oat and wheat flours, the greatest growth was achieved with the addition of 2 and 5% of glucose. However, addition of higher quantities resulted in substrate inhibition. For lactic acid production in oat and wheat flours, increases in the added glucose increased the lactic acid production. It is well-known that glucose is the major substrate to improve lactic fermentation. The unusual finding based on the present study is the improved process in terms of pH value, bacterial growth and lactic acid production in wheat and oat flours with various glucose concentrations. Calderon [21,22] carried out a similar study, using potato starch alone and potato starch with glucose. He reported increased bacterial growth of *L. fermentum* Ogi E1 and doubled lactic acid production when glucose was added to the starch, as reported from the present study on oat and wheat flours.

4. Conclusion

In the current study, lactic fermentation of cereal flours was possible but results were different in terms of strain growth and lactic acid production. To achieve the best results for oat and wheat fermentations, it is preferable to add D-glucose to the flour. Furthermore, the sugar concentration depends on the target; therefore, using a maximum concentration of 5% D-glucose is favorable for a better bacterial growth while a glucose concentration higher than 5% can be used for a higher lactic acid production. However, rice fermentation does not need this sugar additive.

Contributions: Roberto Nigro and Marianna Gallo designed the research; Rosa Colucci Cante, Federica Nigro, Francesca Passannanti, Dana Salameh, Paola Schiattarella and Concetta Schioppa carried out the research; Andrea Budelli provided the raw materials and *Lactobacillus paracasei* CBA L74; Marianna Gallo and Francesca Passannanti wrote the paper and have the primary responsibility for the final content. All authors have read and approved the final manuscript.

5. Acknowledgements

Dr. Andrea Budelli is currently employed by Heinz BV, Netherlands. He provided the raw materials (rice, oat and wheat flours) and *Lactobacillus paracasei* CBA L74 and participated in design of the study. He had no additional role in data collection and analysis, decision to publish or preparation of the manuscript. Heinz BV did not provide any financial supports to the authors for experimental activities and did not have additional roles in study design, data collection and analysis, decision to publish or preparation of the manuscript.

6. Conflict of interest

The authors declare no conflict of interest.

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اثرات افزودن گلوکز هنگام تخمیر لاکتیکی آردهای برنج، جو دوسر و گندم

ماریانا گالو[۱و۲*]، فرانسیسکا پاساننانتی[۱]، رزا کلاسی کانته[۱]، فدریکا نیگرو[۲]، دانا سالامه[۱]، پائولا سیتارلا[۳]، کنستا سیوپا[۳]، آندرآ بادلی[۴]، روبرتو نیگرو[۱]

۱- دانشگاه ناپل فدریکو II، DICMAPI، پیازال تکنیکو ۸۰، ۸۰۱۲۵، ناپل، ایتالیا.

۲- دانشگاه رم نیکولو کاسانو، گروه مهندسی، از طریق دون کارلو گنوچی، ۳، ۰۱۶۶ رم، ایتالیا.

۳- آی تی پی اس. آر.۱، از طریق بیسیگنانو در چیائیا ۶۸، ۸۰۱۲۱ ناپولی.

۴- مرکز نوآوری کرفت هینس، NieuweDukenburgseweg ۱۹، ۶۵۳۴ نیجمگن، هلند.

چکیده

سابقه و هدف: علاقه مصرف کنندگان مواد غذایی زیست‌یار[1] در دهه های اخیر افزایش یافته است. صنایع غذایی با تولید فرآورده های نوآورانه و تضمین بهره‌وری بالای تولید، به این توجه فزاینده پاسخ می دهند. غلات، به علت ماهیت کمک زیست‌یاری[2]، رشدمایه هایی[3]قابل تخمیر مناسبی می باشند؛ که امکان تولید مواد غذایی فراسودمند[4] از آن وجود دارد. هدف این مطالعه تایید اثرات افزودن د-گلوکز بر تخمیر آردهای برنج، جو دوسر و گندم بود.

مواد و روش ها: سوسپانسیون ۱۵ درصدی آرد غلات (برنج، جو دوسر و گندم) در آب مقطر با افزایش غلظت گلوکز (۲، ۵، ۷ و ۱۰ وزنی حجمی) به مدت ۲۴ ساعت توسط با *لاکتوباسیلوس پاراکازئی* CBA L74 تخمیر شدند. سپس، pH، رشد میکروبی و میزان تولید لاکتیک اسید بررسی شد.

یافته‌ها و نتیجه‌گیری: تخمیر برنج تحت تاثیر افزودن گلوکز نمی باشد. درمورد جو دوسر و گندم، افزودن د-گلوکز غلظت باکتریایی و نیز تولید لاکتیک را افزایش داد. به خصوص، بیشترین رشد با افزودن ۲ و ۰/۵ درصد گلوکز به دست آمد. علاوه بر این، غلظت لاکتیک اسید با افزایش غلظت گلوکز افزایش یافت. در نتیجه، به نظر می رشد افزودن د-گلوکز برای بهبود تخمیر برنج ضرورتی نداشته باشد. در مقابل، تخمیر جو دوسر و گندم برای رشد بهتر لاکتوباسیلوس و تولید بیشتر لاکتیک اسید، نیازمند دسترسی به منابع کربن بیشتر می باشد.

تعارض منافع: نویسندگان اعلام می‌کنند که هیچ نوع تعارض منافعی مرتبط با انتشار این مقاله ندارند.

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واژگان کلیدی

- آرد غلات
- گلوکز
- تخمیر لاکتیکی
- *لاکتوباسیلوس پاراکازئی* CBA L74
- زیست یار

***نویسنده مسئول**

ماریانا گالو

دانشگاه ناپل فدریکو II و دانشگاه رم نیکولو کاسانو، ایتالیا.

تلفن: ۳۹۰۸۱۷۶۸۵۹۴۳+
نمابر: ۳۹۰۸۱۵۹۳۶۹۳۶+

پست الکترونیک:
marianna.gallo@unina.it

[1] Probiotic foods
[2] Prebiotic
[3] Substrate
[4] Functional foods