ISSN 1392-3196 / e-ISSN 2335-8947
Zemdirbyste-Agriculture, vol. 106, No. 1 (2019), p. 59–64
DOI 10.13080/z-a.2019.106.008

The use of rice polish medium for the evaluation of antifungal activity of lactic acid bacteria

Rūta VAITKEVIČIENĖ[1], Daiva ŽADEIKĖ[1], Elena BARTKIENĖ[2], Vita KRUNGLEVIČIŪTĖ[2], Violeta BALIUKONIENE[2], Skaidrė SUPRONIENĖ[3], Gražina JUODEIKIENĖ[1]

[1]Kaunas University of Technology
Radvilėnų 19, Kaunas, Lithuania
E-mail: ruta.vaitkeviciene@ktu.lt

[2]Lithuanian University of Health Science
Tilžės 18, Kaunas, Lithuania

[3]Institute of Agriculture, Lithuanian Research Centre for Agriculture and Forestry
Instituto 1, Akademija, Kėdainiai distr., Lithuania

Abstract

The aim of this study was to evaluate rice polish as a substrate for the production of metabolites characterised by antifungal activity by using lactic acid bacteria. The *Lactobacillus plantarum, L. brevis*, *L. paracasei*, *L. uvarum*, *L. farraginis*, *Pediococcus pentosaceus* and *P. acidilactici* strains were tested in the experiment. The antifungal activity of the strains was evaluated against filamentous fungi, such as *Aspergillus versicolor*, *A. terreus*, *A. niger*, *A. fumigatus* and *Penicillium spinulosum*, *P. viridicatum*, *P. palitans*. The rice polish fermentation was carried out in a single step without growth supplementation at submerged fermentation conditions. The obtained results confirmed that rice polish is a suitable substrate for the production of antifungal metabolites depending on lactic acid bacteria (LAB) strain. The tested *P. pentosaceus* strains showed strong and moderate inhibitory activity against *P. viridicatum*, *A. versicolor* fungi, while *P. acidilactici* strains showed moderate and low activity against *Aspergillus* sp. fungi. None of the tested LAB exhibited antifungal activity against *A. terreus* and *A. niger.*

Key words: antifungal activity, *Aspergillus*, lactobacilli, pediococci, *Penicillium*, rice bran.

Introduction

The widespread fungi are important contaminants of food crops, food raw materials and processed foods. Fungal contamination causes not only substantial economic losses, but also food safety issues (Hawksworth, 2001). Prevention and control of fungal contamination is an important problem in the field of agriculture and industry. In food industry, chemical preservatives are generally used to control the growth and reproduction of fungi. However, due to abuse of chemical preservatives, their residues and food safety issues become more and more important. Also, consumer demand for safe food with a long shelf life and preference for minimally processed products that do not contain chemical preservatives make it more challenging for food processing (Cotter et al., 2005). The use of microorganisms to prevent fungal contamination has been gaining interest during the recent years as they can mitigate the adverse effects of chemical preservatives (Prema et al., 2010). Antifungal peptides are reported to be one of the most important natural preservatives which are effective against most of the fungal pathogens. According to the literature, most strains of *Bacillus* sp. produced only one or two antimicrobial substances (Mora et al., 2011). Lactic acid bacteria can provide different kinds of bioactive molecules, such as organic acids, fatty acids, hydrogen peroxide and bacteriocins (Asmahan, 2010; Schnurer, Magnusson, 2005). Recently, several antimicrobials have been discovered; however, it has become imperative to search for more stable antimicrobials with a broad range of activities, especially against various spoilage moulds.

The ability to produce several antibacterial and antifungal substances confers biopreservation potential to lactic acid bacteria (Cabo et al., 2002; Cheong et al., 2014). The lactic acid bacteria have been used as bio-preservative organisms in food and feed systems. They are Gram-positive bacteria and include species of the genera *Enterococcus*, *Lactobacillus*, *Leuconostoc*, *Lactococcus*

Please use the following format when citing the article:
Vaitkevičienė R., Žadeikė D., Bartkienė E., Krunglevičiūtė V., Baliukonienė V., Supronienė S., Juodeikienė G. 2019. The use of rice polish medium for the evaluation of antifungal activity of lactic acid bacteria. Zemdirbyste-Agriculture, 106 (1): 59–64. DOI 10.13080/z-a.2019.106.008

and *Pediococcus*. For centuries, fermentation with lactic acid microorganisms has been a simple and inexpensive method to improve the functional value and safety of foods. The production of inhibitory substances and organic acids by the LAB is an indicator that these bacteria could be widely used in the food industry as bio-preservatives due to their broad inhibition spectrum.

Antifungal metabolite production by lactic acid bacteria using natural by-products from food production is a very promising area. Rice polish contains various nutrients, such as protein, fat, carbohydrates, minerals, and vitamins, making it a beneficial food source. The ability to produce several antibacterial and antifungal substances confers biopreservation potential to lactic acid bacteria. Using alternative biotechnology tools for the evaluation of antimicrobial activity against undesirable microorganisms in the food industry specific metabolites would be suitable candidates for use as antimicrobial agents.

The aim of this study was to analyse the potential of brown rice polishing waste as a substrate for antifungal metabolite production by lactic acid bacteria (LAB) newly isolated from cereal based sourdoughs.

Materials and methods

Materials and microorganisms. Rice polish (moisture 11.38 g 100 g^{-1}, protein 11.05 g 100 g^{-1}, fat 1.27 g 100 g^{-1}, sugars 205 mg 100 g^{-1}, dietary fibre 7.46 g 100 g^{-1}) was obtained from SC "Ustukiu malunas" (Pasvalys, Lithuania) after processing of brown rice. Before analysis, rice polish was ground in a laboratory mill (Buhler Bros Inc., Switzerland) and sterilized at 121°C for 15 min. Eleven lactic acid bacteria (LAB): *Lactobacillus paracasei* LUHS244 (NBRC 15889), *Lactobacillus uvarum* LUHS245 (strain 8), *Lactobacillus farraginis* LUHS206 (NRIC 0676), *Lactobacillus brevis* LUHS140, LUHS173 (ATCC 367; n = 2), *Lactobacillus plantarum* LUHS18 (WCFS1), *Lactobacillus plantarum* LUHS135 (JCM 1149), *Pediococcus acidilactici* LUHS29, LUHS236 (DSM 20284; n = 2), *Pediococcus pentosaceus* LUHS22, LUHS100 (ATCC 25745; n = 2), previously isolated from a spontaneously fermented cereal based media and identified by phenotypic and molecular techniques (Bartkiene et al., 2017), were used in this study during 2016–2018. For the experiment, each strain was cultured in an MRS (de Man, Rogosa and Sharpe) broth CM0359 (Oxoid Ltd., UK) for 48 h at 30°C in the dark.

Rice polish fermentation. For fermentation assay, the rice polish sample (40 ± 0.01 g) was mixed with the distilled water (60 mL) wherein 2% (w/v) of the freshly prepared culture of each LAB strain was added. The samples were incubated for 72 h at 30°C in the dark. The acidity characteristics (pH values) were evaluated after 24, 48 and 72 h of fermentation; the antifungal activity was evaluated after 48 h of fermentation.

Determination of acidity (pH). The pH values of nutritional (MRS) and rice polish (RPL) media after fermentation by different lactic acid bacteria were measured and recorded by a pH electrode (Sartorius, Germany). The pH value was determined for a 10 g sample of sourdough homogenized with 90 mL of distilled water, filtrated and measured. All analyses were performed in duplicate.

Preparation of concentrated culture filtrate. LAB strains were propagated for 24 h in MRS (Oxoid Ltd.) nutritional medium at optimal temperature (30–45°C). The cells were harvested by centrifugation (4500 g, 20 min, 5°C), whereas the supernatant was filtered using a 0.22 μm sterile membrane filter (Merck, Germany) and separated into two subsamples. The pH value of one subsample was adjusted to 6.5 with 5 mol L^{-1} NaOH to eliminate the effect of organic acids. Fermented rice polish also was harvested by centrifugation and separated into two subsamples, one of them had pH 6.5.

Antifungal assay. The plant pathogenic fungal strains of *Penicillium* spp., *Aspergillus* spp. (Table 1) were cultured on sabouraud dextrose agar (SDA) (Oxoid Ltd.) (per 1 L: 10 g peptone, 40 g glucose and 15 g agar) plates using aseptic procedures to avoid contamination. Conidia from fungal mycelium were obtained from 7-day-old SDA cultures of the fungus incubated at 25–30°C in the dark and then stored at 5°C. A spore suspension was collected after 7 days by vigorously shaking the slants with sterile peptone water. Spore concentrations were then adjusted to 1×10^6 spores mL^{-1} in half-strength using a Neubauer Chamber cell counting method (Paudel, Tyagi, 2014).

Table 1. Fungal strains* and their optimal temperature

Fungal strain	Optimal temperature
Penicillium viridicatum	25°C
Penicillium spinulosum	25°C
Penicillium palitans	25°C
Aspergillus terreus	35–40°C
Aspergillus niger	25°C
Aspergilus versicolor	22–26°C
Aspergillus fumigatus	25°C

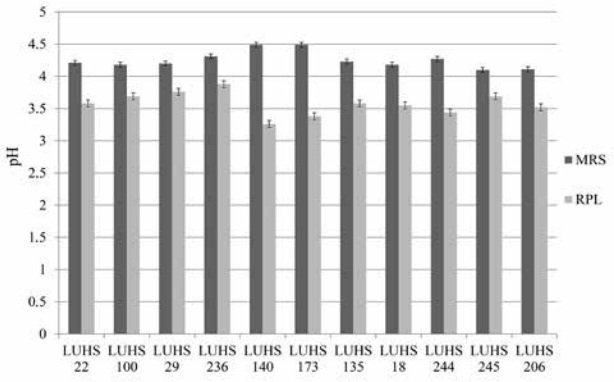
* – growth on sabouraud dextrose agar (Samson, 2000)

Determination of antifungal activity. The antifungal activity of the strains was measured using an agar well diffusion method (Magaldi et al., 2004) and disk-diffusion method (Balouiri et al., 2015). For the assay, the SDA (25 ml) cooled to 45°C was poured into Petri dish and overlaid with 100 μL of fungal suspension. For agar well diffusion method, the antifungal activity against fungi was determined by measuring the inhibition zones (mm). The LAB culture suspension (100 μL) after centrifugation (4500× rpm, 20 min) was added to each well (6 mm in diameter) punched in the cooled agar plates following the incubation for 48 h at the 25°C. After the inhibition zones (mm) had been measured, the antifungal activity was recorded according to the following scale (Manini et al., 2016): (−) no inhibition, (+) delay of spore formation (1–3 mm), (++) delay of spore formation with a small clear zone of inhibition (3–6 mm), (+++) a very good inhibition of mycelium growth and sporulation with large clear zones (>6 mm) around the punched well. For agar disk-diffusion method, the antifungal activity against fungi was determined by measuring the inhibition zones (mm) formed around each disk. Sterile filter paper disks 6 mm diameter (Oxoid Ltd.) impregnated with 100 μl of LAB culture suspension were placed on the inoculated agar surfaces. All tests were performed in duplicate, and the

diameters of the growth inhibition zones around each disk were measured (in mm) after incubation at 25°C for 48 h.

Results and discussion

Changes in acidity (ph) during rice polish fermentation by the tested lactic acid bacteria (LAB). The higher acidification rate of rice polish medium was achieved by fermentation with lactobacilli compared to pediococci. The highest pH values were of the medium fermented with *L. brevis* strains (pH = 3.26 for LUHS140, pH = 3.38 for LUHS173) after 48 h of fermentation. The lowest pH values were of the samples fermented with *P. acidilactici* strains LUHS29 (pH = 3.76) and LUHS236 (pH = 3.88) (Fig. 1). According to the obtained results, the more intensive production of organic acids by the tested LAB was noticed in rice polish medium compared to nutritional medium (MRS), the pH values of rice polish samples fermented with lactobacilli and pediococci were by pH 3.58 and by 15.76% lower on average than those of MRS samples.

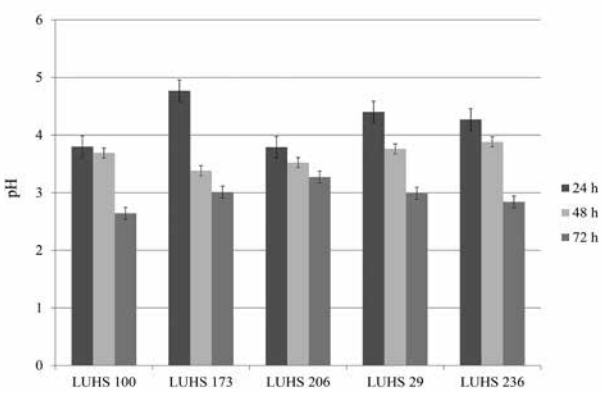


LUHS22, LUHS100 – *Pediococcus pentosaceus*, LUHS29, LUHS236 – *P. acidilactici*; LUHS140, LUHS173 – *Lactobacillus brevis*, LUHS135, LUHS18 – *L. plantarum*, LUHS244 – *L. paracasei*, LUHS245 – *L. uvarum*, LUHS206 – *L. farraginis*

Figure 1. The acidity (pH) values of nutritional (MRS) and rice polish (RPL) media after 48 h fermentation by different lactic acid bacteria

The highest acidity in rice polish medium was shown by five strains of all the LAB tested. As is seen from Figure 2, the highest acidity and the lowest pH values were produced by the LUHS100 (pH 3.80) after 24 h fermentation, following the LUHS206 (pH 3.79). The highest acidity was determined in rice medium fermented by LUHS173 (pH 3.38) and LUHS206 (pH 3.52) after 48 h fermentation, and in samples fermented by LUHS100 (pH 2.64) and LUHS236 (pH 2.84) after 72 h of fermentation. The drastic decrease in pH during 72 h fermentation was shown by the LUHS173 (36.90%) and LUHS29 (34.09%), LUHS236 (33.49%) (Fig. 2).

Lactic acid bacteria are able to produce different kind of bioactive molecules, such as organic acids, fatty acids, hydrogen peroxide and bacteriocins (Gerez et al., 2009), equally to produce other small compounds with a low molecular weight during fermentation processes (Cizeikiene et al., 2013), also provide metabolites that can inhibit the growth of fungi under the native conditions



LUHS100 – *Pediococcus pentosaceus*; LUHS173 – *Lactobacillus brevis*, LUHS206 – *L. farraginis*, LUHS29, LUHS236 – *P. acidilactici*

Figure 2. Changes in the acidity (pH) of medium during fermentation of rice polish by selected lactic acid bacteria

(Hassan, Bullerman, 2008). According to the literature, the acetic acid and the lactic acid produced was likely the primary factor responsible for the *Lactobacillus* antimicrobial properties (Cabo et al., 2002). Also, it was reported that *P. pentosaceus* produced three antifungal substances: cyclo(L-Phe-L-Pro), cyclo(L-Phe-trans-4-OH-L-Pro) and phenyllactic acid (Magnusson et al., 2003).

Antifungal activity of tested LAB in rice polish medium determined by agar well diffusion assay. According to the results, rice polish substrate showed the potential for production of antifungal metabolites by the tested LAB. The tested LAB strains showed different inhibition capability on the growth of *Penicillium* and *Aspergillus* sp. strains (Table 2). *Lactobacillus* strains showed strong and moderate activity against *Penicillium* sp., while *Pediococcus* strains showed moderate and low activity against *Aspergillus* sp.

From the tested LAB, *P. acidilactici* LUHS236 and *L. faraginis* LUHS206, strains were shown to moderate inhibition and delay of spore formation against *P. spinulosum*. The *P. pentosaceus* LUHS100 and *L. brevis* LUHS173 produced the high (++) antifungal activity against *A. versicolor* strain. Among other tested LAB, only *L. plantarum* LUHS18 initiated the delay of fungal sporulation (+) against *P. viridicatum*, as well as *P. acidilactici* LUHS29 and lactobacilli LUHS173, LUHS244 and LUHS245 delayed fungal sporulation (+) against *P. spinulosum*. The *P. acidilactici* LUHS29, LUHS236, *P. pentosaceus* LUHS22, *L. brevis* LUHS173 and *L. uvarum* LUHS245 were slightly active (+) against *P. palitans*. From the tested strains, *P. pentosaceus* LUHS100 and *L. brevis* LUHS173 were able to inhibit *A. fumigatus* and *L. brevis* LUHS173 was able to inhibit *A. versicolor*. The others LAB (LUHS140, LUHS135 and LUHS206) did not form the inhibition zones (Table 2).

In previous studies, *P. acidilactici* was found to inhibit the growth of fungi, including species of *Aspergillus* and *Penicillium spp.* (Mandal et al., 2013). *P. pentosaceus* and *P. acidilactici* strains have been shown to inhibit the growth and sporulation of the *A. niger* (Cizeikiene et al., 2013). Also, the supernatant of *P. pentosaceus* and *P. acidilactici* strains showed the high antifungal activity against *A. versicolor* strain (Juodeikiene et al., 2016).

Table 2. The antifungal activity of lactic acid bacteria (LAB) supernatants against the *Penicillium* and *Aspergillius* sp. (an agar well diffusion method)

Micro-organisms	Medium	*P. pentosaceus*		*P. acidilactici*		*L. brevis*		*L. plantarum*		*L. paracasei*	*L. uvarum*	*L. farraginis*
		LUHS 22	LUHS 100	LUHS 29	LUHS 236	LUHS 140	LUHS 173	LUHS 135	LUHS 18	LUHS 244	LUHS 245	LUHS 206
P. viridicatum	MRS	++	++	+	++	+	+	+	+	+	++	+
	RPL	−	−	−	−	−	−	−	+	−	−	−
P. spinulosum	MRS	−	−	+	++	−	+	−	−	+	+	++
	RPL	−	−	+	++	−	+	−	−	+	+	++
P. palitans	MRS	+	+	−	+	−	+	+	+	−	+	−
	RPL	+	−	−	+	−	+	−	+	−	+	−
A. terreus	MRS	−	−	−	−	−	−	−	−	−	−	−
	RPL	−	−	−	−	−	−	−	−	−	−	−
A. versicolor	MRS	+	++	+	+	+	++	+	+	+	+	+
	RPL	−	++	−	−	−	++	−	+	−	−	−
A. niger	MRS	−	−	−	−	−	−	−	−	−	−	−
	RPL	−	−	−	−	−	−	−	−	−	−	−
A. fumigatus	MRS	+	+	+	−	−	+	−	+	+	−	+
	RPL	−	+	−	−	−	+	−	−	−	−	−

Note. MRS – nutritional medium, RPL – rice polish medium; interpretation of zone diameter of inhibition: (−) = no inhibition, (+) = delay of spore formation (1–3 mm), (++) = delay of spore formation with a small clear zone of inhibition around the punched well (3–6 mm), (+++) = a very good inhibition of mycelium growth and sporulation with large clear zones around the punched well (>6 mm).

As the low acidity (pH) has always been associated with antifungal activity (Cabo et al., 2002), the antifungal activity assay was repeated by analysing neutralized LAB supernatants.

Neutralised LAB rice polish supernatants (Table 3) showed lower antifungal activity compared to that of not neutralised (Table 3). The delay of fungal sporulation (+) was shown against the *P. spinulosum* by four LAB strains, against the *P. viridicatum* by three LAB strains, against the *P. palitans* by only one strain. Regarding *Aspergillus* sp., the delay of fungal sporulation (+) was shown against *A. versicolor* and *A. fumigatum* by five LAB strains. The pediococci LUHS100, LUHS29 and *L. brevis* LUHS173 were slightly active (+) against 2–3 pathogens, the other LAB did not form the inhibition zones.

Table 3. The antifungal activity of neutralised (pH 6.5) lactic acid bacteria (LAB) supernatants against the *Penicillium* and *Aspergillus* sp. (an agar well diffusion method)

Micro-organisms	Medium	*P. pentosaceus*		*P. acidilactici*		*L. brevis*		*L. plantarum*		*L. paracasei*	*L. uvarum*	*L. farraginis*
		LUHS 22	LUHS 100	LUHS 29	LUHS 236	LUHS 140	LUHS 173	LUHS 135	LUHS 18	LUHS 244	LUHS 245	LUHS 206
P. viridicatum	MRS	+	−	−	−	−	+	+	+	+	+	−
	RPL	−	−	−	−	−	+	+	+	−	−	−
P. spinulosum	MRS	−	+	+	+	−	−	−	−	+	−	−
	RPL	−	+	+	+	−	−	−	−	+	−	−
P. palitans	MRS	+	−	−	−	−	−	−	−	−	−	−
	RPL	+	−	−	−	−	−	−	−	−	−	−
A. terreus	MRS	−	−	−	−	−	−	−	−	−	−	−
	RPL	−	−	−	−	−	−	−	−	−	−	−
A. versicolor	MRS	−	+	−	−	−	+	−	−	−	+	−
	RPL	−	+	+	+	+	+	−	+	−	−	−
A. niger	MRS	−	−	−	−	−	−	−	−	−	−	−
	RPL	−	−	−	−	−	−	−	−	−	−	−
A. fumigatus	MRS	+	+	+	−	−	+	−	+	+	−	+
	RPL	−	+	+	−	+	−	+	−	+	−	−

Note. MRS – nutritional medium, RPL – rice polish medium; interpretation of zone diameter of inhibition: (−) = no inhibition, (+) = delay of spore formation (1–3 mm), (++) = delay of spore formation with a small clear zone of inhibition around the punched well (3–6 mm), (+++) = a very good inhibition of mycelium growth and sporulation with large clear zones around the punched well (>6 mm).

No inhibition zones were fixed around the punched well against *P. palitans* (except LUHS22), *A. terreus* and *A. niger*. According to the literature, *P. pentosaceus* and *P. acidilactici* strains have no inhibition zones against *A. terreus* (Cizeikiene et al., 2013).

Antifungal activity of selected LAB in rice polish medium determined by an agar disk-diffusion assay. For the optimization of antifungal activity production, five LAB strains (three *Pediococcus* strains and two *Lactobacilli* strains) showing the highest antifungal

activity were tested by an agar disk-diffusion assay for antifungal activity against *Penicillium spinulosum* and *Aspergillus versicolor*. Metabolites produced by LAB in rice polish medium showed various degrees of antifungal activity after 24, 48 and 72 h of fermentation (Table 4). According to the results, rice substrate extracts fermented by tested LAB showed inhibition zones varying from 1.0 to 5.0 mm. The *L. brevis* LUHS173 showed the highest antifungal activity against *A. versicolor* (the inhibition zones 3 and 5 mm, respectively) and *P. spinulosum* (inhibition zones 1.5 and 3 mm, respectively) after 48 and 72 h of fermentation. The *L. farraginis* inhibited the growth of *P. spinulosum* (zone 4 and 5 mm) after 48 and 72 h of fermentation.

Table 4. Antifungal activity against indicatory microorganisms produced by lactic acid bacteria (LAB) in rice polish medium (an agar disk-diffusion method)

Microorganisms		Inhibition zones on an agar plate, mm					
		Penicillium spinulosum			*Aspergillus versicolor*		
		24 h	48 h	72 h	24 h	48 h	72 h
Pediococcus pentosaceus	LUHS100	< 1.0	< 1.0	< 1.0	< 1.0	2.0 ± 0.2	2.0 ± 0.2
Lactobacillus brevis	LUHS173	1.0 ± 0.2	1.5 ± 0.2	3.0 ± 0.2	1.0 ± 0.2	3.0 ± 0.2	5.0 ± 0.2
L. farraginis	LUHS206	3.00 ± 0.2	4.00 ± 0.2	5.00 ± 0.2	< 1.0	< 1.0	1.0 ± 0.2
P. acidilactici	LUHS29	0.10 ± 0.2	0.10 ± 0.2	0.10 ± 0.2	< 1.0	< 1.0	< 1.0
P. acidilactici	LUHS236	0.20 ± 0.2	0.30 ± 0.2	0.30 ± 0.2	< 1.0	< 1.0	< 1.0

According to the literature, different *Lactobacillus* strains producing acids were able to inhibit some species of fungi (Lavermicocca et al., 2000) thus LAB may be considered as an alternative for bio-conservation (Gerez et al., 2009). Also, it was reported that the *P. acidilactici* strain produced broad-spectrum antifungal compounds with fungicidal activity (Mandal et al., 2013). The observed effect of the acidity (pH) suggested that organic acids present in the supernatants could be involved in the detected antifungal activity (Cabo et al., 2002). The results of our study indicated that the fermentation with LAB could enhance production of certain bioactive compounds in rice polish. These LAB naturally dominate in sourdough, and they prooduce metabolites that can inhibit the growth of fungi under the native conditions (Hassan et al., 2015). The selection of different LAB (or their combinations) with a wide spectrum of antifungal activities is an important issue for improving the safety of rice products in various food production.

Conclusions

1. *Lactobacillus* strains showed a higher acidification rate in rice polish medium compared to *Pediococcus* strains.

2. Rice polish medium was tested as a substrate for antifungal activity production against *Penicillium* and *Aspergillus* fungi without additional supplements. All *Lactobacillus* strains produced the antifungal activity against *Penicillium* sp., while *Pediococcus* strains showed low inhibitory activity against *Aspergillus* spp.

3. None of the tested strains exhibited activity against the *A. terreus* and *A. niger*.

4. Given the fact that neutralized supernatants showed inhibitory activity, it can be concluded that the observed antifungal effects cannot be attributed to supernatant acidity, as long as the pH-value was adjusted to 6.5.

5. The further study will be focused on the optimization of culture medium based on rice polish for the production of high antimicrobial activity by tested microorganisms.

Received 16 04 2018
Accepted 15 10 2018

References

1. Asmahan A. A. 2010. Beneficial role of lactic acid bacteria in food preservation and human health: a review. Research Journal of Microbiology, 5: 1213–1220. https://doi.org/10.3923/jm.2010.1213.1221
2. Balouiri M., Sadiki M., Koraichi Ibnsouda S. 2016. Methods for *in vitro* evaluating antimicrobial activity: a review. Journal of Pharmaceutical Analysis, 6: 71–79. https://doi.org/10.1016/j.jpha.2015.11.005
3. Bartkiene E., Bartkevics V., Pugajeva I., Krungleviciute V., Mayrhofer S., Domig K. 2017. The contribution of *P. acidilactici*, *L. plantarum*, and *L. curvatus* starters and L-(+)-lactic acid to the acrylamide content and quality parameters of mixed rye-wheat bread. LWT – Food Science and Technology, 80: 43–50. https://doi.org/10.1016/j.lwt.2017.02.005
4. Cabo M. L., Braber A. F., Koenraad P. M. 2002. Apparent antifungal activity of several lactic acid bacteria against *Penicillium discolour* due to acetic acid in the medium. Journal of Food Protection, 65: 1309–1316. https://doi.org/10.4315/0362-028X-65.8.1309
5. Cheong Y. L. E., Sandhu A., Jayabalan J., Le T. T. K., Nhiep N. T., Ho H. T. M., Tumer M. S. 2014. Isolation of lactic acid bacteria with antifungal activity against the common cheese spoilage mould *Penicillium commune* and their potential as biopreservatives in cheese. Food Control, 46: 91–97. https://doi.org/10.1016/j.foodcont.2014.05.011
6. Cizeikiene D., Juodeikiene G., Paskevicius A., Bartkiene E. 2013. Antimicrobial activity of lactic acid bacteria against pathogenic and spoilage microorganism isolated from food and their control in wheat bread. Food Control, 31 (2): 539–545. https://doi.org/10.1016/j.foodcont.2012.12.004
7. Cotter P. D., Hill C., Ross R. P. 2005. Bacteriocins: developing innate immunity for food. Nature Reviews Microbiology, 3: 777–778. https://doi.org/10.1038/nrmicro1273
8. Gerez C. L., Torino M. I., Rollán G., Valdez G. F. 2009. Prevention of bread mould spoilage by using lactic acid bacteria with antifungal properties. Food Control, 20 (2): 144–148. https://doi.org/10.1016/j.foodcont.2008.03.005
9. Hassan Y. I., Bullerman L. B. 2008. Antifungal activity of *Lactobacillus paracasei* ssp. tolerans isolated from a sourdough bread culture. International Journal of Food Microbiology, 121: 112–115. https://doi.org/10.1016/j.ijfoodmicro.2007.11.038

10. Hassan Y. I., Zhou T., Bullerman L. B. 2015. Sourdough lactic acid bacteria as antifungal and mycotoxin-controlling agents. Food Science and Technology International, 22 (1): 79–90. https://doi.org/10.1177/1082013214565722
11. Hawksworth D. L. 2001. The magnitude of fungal diversity: the 1.5 million species estimate revisited. Mycological Research, 105: 1422–1432. https://doi.org/10.1017/S0953756201004725
12. Juodeikiene G., Bartkiene E., Cernauskas D., Cizeikiene D., Zadeike D., Lele V., Bartkevics V. 2016. Antifungal activity of lactic acid bacteria and their application for *Fusarium* mycotoxin reduction in malting wheat grains. LWT – Food Science and Technology, 89: 307–314. https://doi.org/10.1016/j.lwt.2017.10.061
13. Lavermicocca P., Valerio F., Evidente A., Lazzaroni S., Corsetti A., Gobbeti M. 2000. Purification and characterization of novel antifungal compounds from the sourdough *Lactobacillus plantarum* Strain 21B. Applied and Environmental Microbiology, 66: 4084–4090. https://doi.org/10.1128/AEM.66.9.4084-4090.2000
14. Magaldi S., Mata-Essayag S., Hartung de Capriles C., Perez C., Colella M. T., Olaizola C., Ontiveros Y. 2004. Well diffusion for antifungal susceptibility testing. International Journal of Infectious Diseases, 8: 39–45. https://doi.org/10.1016/j.ijid.2003.03.002
15. Magnusson J., Strom K., Roos S., Sjogren J., Schnurer J. 2003. Broad and complex antifungal activity among environmental isolates of lactic acid bacteria. FEMS Microbiology Letters, 219: 129–135. https://doi.org/10.1016/S0378-1097(02)01207-7
16. Mandal V., Sen S. K., Mandal N. C. 2013. Production and partial characterisation of an inducer-dependent novel antifungal compound(s) by *Pediococcus acidilactici* LAB 5. Journal of the Science of Food and Agriculture, 93 (10): 2445–2453. https://doi.org/10.1002/jsfa.6055
17. Manini F., Casiraghi M. C., Poutanen K., Brasca M., Erba D., Plumed-Ferrer C. 2016. Characterization of lactic acid bacteria isolated from wheat bran. LWT – Food Science and Technology, 66: 275–283. https://doi.org/10.1016/j.lwt.2015.10.045
18. Mora I., Cabrefiga J., Montesinos E. 2011. Antimicrobial peptide genes in *Bacillus* strains from plant environments. International Microbiology, 14: 213–223.
19. Paudel R., Tyagi S. 2014. Effect of different pH on the growth and sporulation of *Fusarium oxysporum*: the causal organism of wilt disease of tomato. International Journal of Basic and Applied Biology, 2: 103–106.
20. Prema P., Smila D., Palavesam A. Immanuel G. 2010. Production and characterization of an antifungal compound (3-phenyllactic acid) produced by *Lactobacillus plantarum* strain. Food and Bioprocess Technology, 3: 379–386. https://doi.org/10.1007/s11947-008-0127-1
21. Samson R. A. (ed.). 2004. Introduction to food- and airborne fungi. Amer Society for Microbiology.
22. Schnurer J., Magnusson J. 2005. Antifungal lactic acid bacteria as biopreservatives. Trends in Food Science and Technology, 16: 70–78. https://doi.org/10.1016/j.tifs.2004.02.014

ISSN 1392-3196 / e-ISSN 2335-8947
Zemdirbyste-Agriculture, vol. 106, No. 1 (2019), p. 59–64
DOI 10.13080/z-a.2019.106.008

Ryžių poliravimo atliekų panaudojimas įvertinti pieno rūgšties bakterijų priešgrybiniam aktyvumui

R. Vaitkevičienė[1], D. Žadeikė[1], E. Bartkienė[2], V. Krunglevičiūtė[2], V. Baliukonienė[2], S. Supronienė[3], G. Juodeikienė[1]

[1]Kauno technologijos universitetas
[2]Lietuvos sveikatos mokslų universitetas
[3]Lietuvos agrarinių ir miškų mokslų centro Žemdirbystės institutas

Santrauka

Tyrimo tikslas – įvertinti ryžių poliravimo atliekas kaip substratą priešgrybiniu aktyvumu pasižyminčių metabolitų gamybai naudojant pieno rūgšties bakterijas. Eksperimento metu buvo naudotos *Lactobacillus plantarum*, *L. brevis*, *L. paracasei*, *L. uvarum*, *L. farraginis*, *Pediococcus pentosaceus* ir *P. acidilactici* bakterijų padermės. Tirtas jų aktyvumas prieš *Aspergillus versicolor*, *A. terreus*, *A. niger*, *A. fumigatus*, *Penicillium spinulosum*, *P. viridicatum* ir *P. palitans* grybus. Ryžių poliravimo atliekos fermentuotos tradiciniu būdu, nenaudojant papildomų maisto medžiagų.

Tyrimo rezultatai patvirtino, kad ryžių poliravimo atliekos yra tinkamas substratas siekiant sukelti bakterijų priešgrybinį aktyvumą, priklausomai nuo naudotos pieno rūgšties bakterijų padermės. Analizuotos *P. pentosaceus* bakterijų padermės pasižymėjo stipriu ir vidutiniu inhibitoriniu aktyvumu prieš *P. viridicatum* ir *A. versicolor* grybus, o *P. acidilactici* padermės buvo vidutiniškai ir silpnai aktyvios prieš *Aspergillus* spp. grybus. Nė viena iš tirtų pieno rūgšties bakterijų nepasižymėjo priešgrybiniu aktyvumu prieš *A. terreus* ir *A. niger* grybus.

Reikšminiai žodžiai: *Aspergillus*, lactobacilli, pediococci, *Penicillium*, priešgrybinis aktyvumas, ryžių sėlenos.