Concentrated autochthonous culture to be used on the production of safe meat products

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Introduction

Since food safety has become an increasingly important international concern, the application of antimicrobial peptides from LAB, that target food pathogens without toxic or other adverse effects, has received great attention.[1,2] The possibility of including live bacteriocin-producing LAB in foods has been a promising area: "protective culture concept". Bioprotective cultures may act as starter cultures in the food fermentation process, such as dry sausage manufacturing process, or they may protect foods without any detrimental organoleptic changes.[3] The addition of LAB cultures to food products requires previous encapsulation by methods, such spray drying, to guarantee their easy storage, handling and longer shelf life.[4]

The objective of this study was to test different conditions of growth and drying of a *Lactobacillus* strain in order to obtain a concentrated bioprotective culture to be used in the production of fermented meat products.

Methodology

Lactobaccilus strain grown in

Man, Rogosa & Sharpe broth

Reconstituted Skimmed Milk 11% (w/v)

Growth curve during 36h to evaluate the survival of the *Lactobacillus* strain (log N/N0); pH value of the culture and the antimicrobial activity (AU/mL) against 4 *L. monocytogenes* and 2 *L. innocua* strains in both MRS broth and RSM

After growing *Lactobacillus* strain until the time the AU/mL was maximal

18-21h

15-30h

Cells washed and ressuspended in RSM (11% v/v) as drying agent

Spray drying

Niro atomizer

Constant parameters:
-Outlet air temperature (80 ºC)
-Feed temperature (40 ºC)
-Drying air flow rate (25 ml/min)

Variable parameters:
-Inlet air temperature (140 and 150 ºC)
-Atomizer pressure (1 and 1.5 bar)

Powder analysis:
-a_w and yield (%)

Rehydrated powder analysis:
-Survival of the *Lactobacillus* strain (log N/N0)
-Antimicrobial activity (AU/mL) against 6 *Listeria* spp.

Results

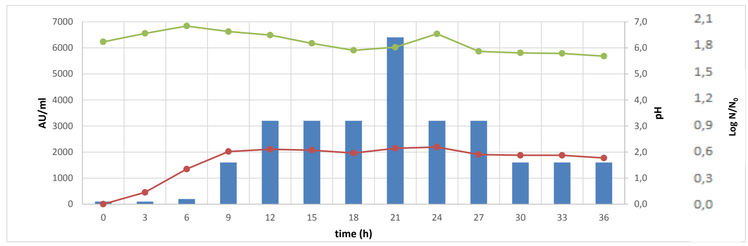

Figure 1. Growth curve in MRS broth of *Lactobacillus* strain expressed in Log N/N_0 (red), pH value (green) and bacteriocinogenic activity (AU/mL) against *Listeria monocytogenes* CLIP 80459 (blue bar) along 36 h.

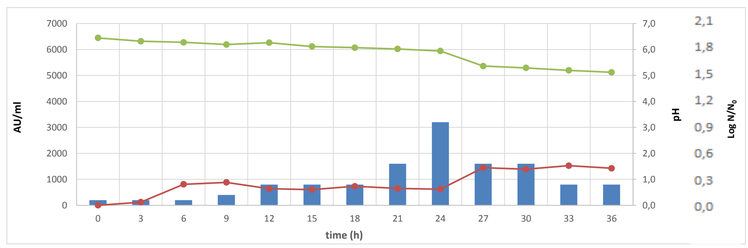

Figure 2. Growth curve in RSM of *Lactobacillus* strain expressed in Log N/N_0 (red), pH value (green) and bacteriocinogenic activity (AU/mL) against *Listeria monocytogenes* CLIP 80459 (blue bar) along 36 h.

Table 1. Data obtained after analysis of spray dried powders (aw and yield) and of rehydrated powders (survival of *Lactobacillus* strain (log N/N_0) and bacteriocinogenic activity (AU/mL) against *Listeria monocytogenes* CLIP 80459)

Growth conditions	Spray drying conditions				log CFU/mL		Bacteriocinogenic activity (AU/mL)*	
	Inlet temperature (ºC)	Pressure (bar)	a_w	Yield (%)	Before SD	After SD	Before SD	After SD
MRS broth	140 ºC	3	0.271±0.092	26.0	9.1±0.1	9.9±0.1	9600±4525	1200±566
		3.5	0.148±0.030	43.5	9.2±0.1	10.0±0.2	3200±0	800±0
	150 ºC	3	0.312±0.076	20.5	9.4±0.3	10.0±0.1	12800±0	12800±0
		3.5	0.251±0.015	12.5	9.4±0.4	10.0±0.2	12800±0	12800±0
Skim milk	140 ºC	3	0.328±0.075	19.0	8.0±0.1	9.1±0.1	800±0	500±424
		3.5	0.309±0.017	13.5	8.1±0.1	8.9±0.3	1600±0	200±0
	150 ºC	3	0.315±0.044	11.0	7.4±0.0	7.7±0.1	1200±566	1200±566
		3.5	0.244±0.025	20.5	7.5±0.3	6.7±0.4	600±283	800±0

*Results for *L. monocytogenes* CLIP 80459; SD – spray drying.

Conclusions

- Maximum antilisterial activity occurred after 18-21h (3200-6400 AU/ml) of growth in MRS and after 15-30h (800 and 3200 AU/ml) in RSM.
- The antimicrobial activity varied slightly between the different target microorganisms, being higher against *L. monocytogenes* isolates.
- Although no significant differences ($p<0.05$) were observed in the survival of the culture during spray drying, the antilisterial activity decreased for all the conditions investigated; highest decrease for those cells grown in RSM.
- When the same dried cells were ressuspended in MRS medium and incubated overnight, they were able to recover and maintain and/or increase their antilisterial activity.

These preliminary results are promising to obtain a concentrated culture to be use as Direct VAT inoculation, which could be used in food industry.

References

[1] Ahmad, V., Khan, M.S., Jamal, Q.M.S., Alzohairy, M.A., Karaawi, M.A.A., Siddiqui, M.U. (2017). Antimicrobial potential of bacteriocins: in therapy, agriculture and food preservation. International Journal of Antimicrobial Agents 49: 1-11.
[2] Balciunas, E.M., Martinez, F.A:C., Todorov, S.D., Franco, B.D.G.M., Converti, A., Oliveira, R.P.S. (2013). Novel biotechnological applications of bacteriocins: A review. Food control 32: 134-142.
[3] Woraprayote, W., Malila, Y., Sorapukdee, S., Swetwiwathana, A., Benjakul, S., Visessanguan, W. (2016). Bacteriocins from lactic acid bacteria and their applications in meat and meat products. Meat Science 120: 118-132.
[4] Barbosa, J., Borges, S., Teixeira, P. (2015). *Pediococcus acidilactici* as a potential probiotic to be used in food industry. International Journal of Food Science & Technology 50: 1151-1157.

Acknowledgements

Financial support for author J. Barbosa was provided by a post-doctoral fellowship SFRH/BPD/113303/2015 (FCT) and by the project DEM@BIOFUMADOS - *Demonstrador do Biofumados - Tradição vs Qualidade - Produção de Enchidos e Fumados Tradicionais Portugueses* (Ref. 17634 BI_1).

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