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Effect of potentially probiotic lactic acid bacteria on the physicochemical composition and acceptance of fermented cereal beverages

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ARTICLE INFO

Article history:
Received 17 November 2014
Received in revised form 4 March 2015
Accepted 6 March 2015
Available online 27 March 2015

Keywords:
Fermented cereal beverages
Lactobacilli
Probiotic
Acceptance
PCA

ABSTRACT

The physicochemical characteristics and acceptance of Lactobacilli-fermented cereal beverages individually inoculated with human derived *Lactobacillus acidophilus* NCIMB 8821, *Lactobacillus plantarum* NCIMB 8826, and *Lactobacillus reuteri* NCIMB 11951 were evaluated after 10 h of incubation at 37 °C. Values of cell viability, pH, titratable acidity (TA), soluble solids content (SSC), free amino nitrogen content (FAN) and colour were correlated with the acceptance using the Person correlation and a principal component analysis (PCA) was used to assess the relationship and trends of these data. Additionally, volatile and non-volatile compounds determinations were performed. The cell viability was around 10^8 cfu/mL with pH values ranging from 3.3 to 3.7. The beverage formulated with *L. plantarum* and malt substrate exhibited greater acceptance and it encompassed the highest concentration of acetaldehyde. The Pearson correlation and PCA depicted that the pH of the beverages was related to their acceptance.

1. Introduction

Consumers worldwide are becoming increasingly aware of the relationship between diet and health and have a better nutrition education. A large number of individual food constituents are known or suspected to have a direct positive or negative effect on human health, and increasingly new foods have associations to different aspects of human health. Rapid advances in science and technology, increasing healthcare costs, changes in food laws affecting label and product claims, an ageing population, and rising interest in attaining wellness through diet are some of the factors driving the interest on functional foods worldwide (Siró, Kápolna, Kápolna, & Lugasi, 2008). Within these foods dairy based products targeting gut health (probiotics and/or prebiotics) constitute one of the largest and fastest growing sectors (Molin, 2001). It has been estimated that probiotic foods comprise between 60 and 70% of the total functional food market (Tripathi & Giri, 2014). Probiotics (live microbial food supplements that stimulate the growth of preferred microflora when consumed on a regular basis providing a health benefit on the host) are regularly formulated with food grade lactic acid bacteria (Reid, 2008; Rodgers, 2008). These microorganisms have been linked with a range of health benefits and are currently going under significant research in order to support such claims. The diseases that have been treated or that have

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http://dx.doi.org/10.1016/j.jff.2015.03.012
1756-4646/

the potential to be healed with probiotic formulations include diarrhoea, gastroenteritis, irritable bowel syndrome, and inflammatory bowel disease (IBD; Crohn's disease and ulcerative colitis), cancer, depressed immune function, inadequate lactase digestion, infant allergies, failure-to-thrive, hyperlipidaemia, hepatic diseases and *Helicobacter pylori* infections (Parvez, Malik, Ah Kang, & Kim, 2006).

The increasing demand for these health-enhancing products has created the need for new non-dairy substrates for probiotic production. Previous works have reported that beverages such as fruit and vegetable juices may be the next category of food matrices to serve as carriers of probiotic bacteria. Furthermore, cereals are also potential viable substrates as they contain nutrients easily assimilated by probiotics (Martins et al., 2013). It has been demonstrated that oat, barley and malt substrates can improve lactobacilli tolerance to the harsh conditions of the gastrointestinal tract, and they can support the growth of single and mix-culture fermentations of probiotic microorganisms (Charalampopoulos & Pandiella, 2010; Charalampopoulos, Pandiella, & Webb, 2003; Herrera-Ponce, Nevárez-Morillón, Ortega-Rívas, Pérez-Vega, & Salmerón, 2014). Additionally, to the proven potential of cereal substrates to support the growth of probiotic microorganisms these have been associated with the risk reduction of chronic diseases such as obesity, cardiovascular disease, type 2 diabetes, and some cancers (Wang, He, & Chen, 2014). Thus, the use of cereal substrates has a great potential to develop novel functional beverages that promote the gastrointestinal health and can prevent chronic diseases.

Cereals could be used to design cereal-based fermented beverages with probiotic characteristics if these formulations fulfil probiotic requirements and have acceptable physicochemical characteristics and organoleptic properties. Food technology encounters several challenges during the development of novel products. Consumer sensory evaluation is used to asses whether a consumer likes a product, prefers it over another, or finds it acceptable based on its sensory characteristics (Earle, Earle, & Anderson, 2001). Thus, fruit juices, vegetables and cereals constitute an emerging segment of nonconventional dairy substrates for the design of probiotic beverages that will require sensory and physicochemical characterization for quality control and further product development. Most of the current work in the area of development of non-dairy probiotics has been performed in fruit and soy-based fermented products where sensory, colour and rheological evaluations have been carried out (Chattopadhyay, Raychaudhuri, & Chakraborty, 2013; Fonteles et al., 2013; Pereira, Almeida, de Jesus, da Costa, & Rodrigues, 2013; Perricone, Corbo, Sinigaglia, Speranza, & Bevilacqua, 2014). It has also been observed that the addition of sugar and an artificial flavour can significantly improve the acceptance of fermented coconut water (Camargo Prado et al., 2015). Nevertheless, the analysis of volatile flavour compounds in novel non-dairy probiotic products has not been studied in depth. The volatile compounds content in fermented food significantly affects its aroma and substantially influences all of its organoleptic characteristics. The flavour and aroma of fermented food is composed of many volatile compounds; however, only a few are designated as aroma-impact compounds. In a previous work we studied the fermentation kinetics of nine similarly formulated and fermented cereal beverages and evaluated the effect of cereal substrates and lactobacilli strains on the formation of volatile and non-volatile compounds (Salmerón, Thomas, & Pandiella, 2014), though, the sensory characteristics of such beverages were not studied. The purpose of this work was to assess physicochemical, volatile and non-volatile flavour compounds of nine lactic acid fermented cereal beverages and to correlate these with acceptability tests. The understanding of this relationship will improve our knowledge on how these parameters can make this type of fermented cereal beverages more acceptable and in what way the relation of lactobacilli cultures and cereal media affect the sensorial attributes of novel non-dairy functional beverages.

2. Materials and methods

The cereal lactic acid fermented beverages were produced for the first time only for this study in order to evaluate the relationship of their chemical and physicochemical composition and their sensory acceptance. Such cereal based fermented beverages were formulated with cereal media individually inoculated with human derived lactobacilli strains at 37 °C for 10 h.

2.1. *Microorganisms and inocula preparation*

The strains *Lactobacillus acidophilus* NCIMB 8821 (isolated from human saliva), *Lactobacillus plantarum* NCIMB 8826 (isolated from human saliva), and *Lactobacillus reuteri* NCIMB 11951 (isolated from human intestine) were obtained from the National Collection of Industrial and Marine Bacteria, Aberdeen, Scotland, UK. The cultures were stored at –30 °C in de Man Rogosa Sharpe (MRS) broth (Sigma-Aldrich Co. Ltd., Dorset, UK) containing 10% (v/v) of glycerol. For the inocula preparation these frozen ampoules were added directly to sterilized MRS broth and incubated at 37 °C for 12 h. Stocks were prepared routinely in MRS medium. Before use, the lactobacilli cultures were propagated in sterile cereal media (starch-free extracts of flour–water suspensions prepared from oats, and malt) at 37 °C for 20 h. Individual cultures were used as inocula at 1% (v/v) in each cereal media during the preparation of the fermented beverages as in previous studies this ratio of inoculum–medium has produced fermented cereal substrates with lactobacilli cell viability above the minimum recommended for a probiotic product (6 Log_{10} cfu/mL based on a daily dose of 100 mL) (Rathore, Salmerón, & Pandiella, 2012; Rozada-Sánchez, Sattur, Thomas, & Pandiella, 2008).

2.2. *Cereal substrates processing*

The cereal media used were mainly starch-free extracts of flour–water suspensions prepared from oats (from a local producer), barley (Brewing Research International, Surrey, UK), and malt. The malt was produced from the same barley used in this study under the following conditions: steeping at 20 °C for 36 h, germination at 10 °C for 24 h, 18 °C for 72 h, and kilning at 45 °C for 20 h and 95 °C for 3 h. The grains were ground using a laboratory Falling number hammer mill (Perten Instruments,