

Characterization of 70.5% cocoa content dark chocolate incorporated with *Bifidobacterium animalis* subspecies *lactis* BB-12®



CATOLICA

CBQF - CENTRE FOR BIOTECHNOLOGY
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PORTO

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Introduction

Currently, **probiotics** are defined as "live microorganisms that, when administered in adequate amounts, confer a health benefit on the host" [1]. *Bifidobacterium animalis* subsp. *lactis* **BB-12®** is one of the most common probiotics used as a **food supplement** [2].

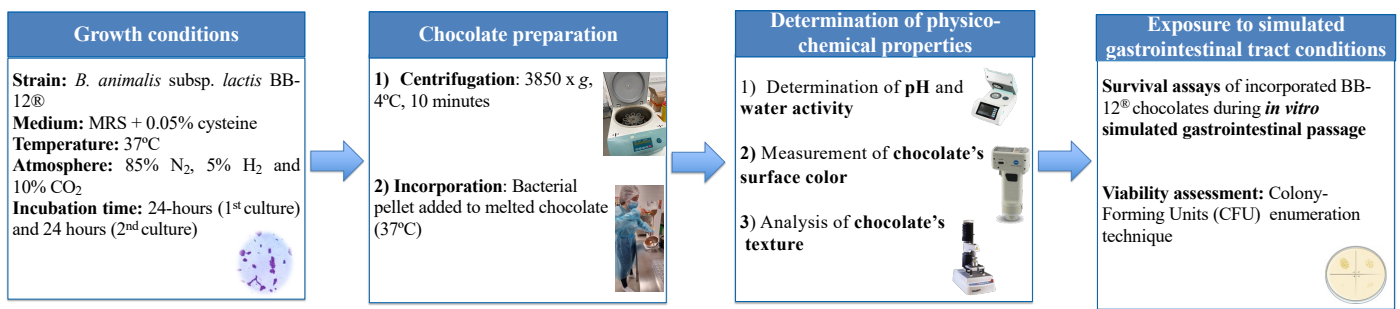
Chocolate is one of the most popular and appealing culinary products, with various **cocoa percentages** available to consumers [3;4]. Several studies have suggested that chocolate, particularly **dark chocolate**, may offer **health benefits** due to the presence of a diverse array of **bioactive compounds** [5].

In this sense, a growing interest in using chocolate as a **carrier for probiotic delivery** has emerged.

Objectives

This study aimed to characterize a chocolate matrix with 70.5% cocoa content incorporated with *B. animalis* subspecies *lactis* BB-12® in the following parameters: i) **probiotic viability** in the chocolate matrix **throughout manufacture**; ii) **physicochemical properties**: pH, water activity, surface color and texture and iii) **probiotic survival** throughout *in vitro* **gastrointestinal passage**.

Methods



Main findings

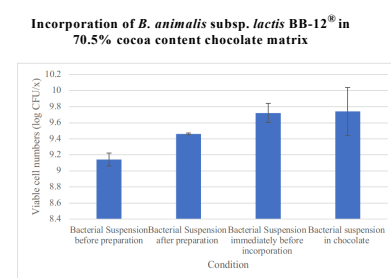


Fig. 1 Viable cell number of *B. animalis* subsp. *lactis* BB-12® in bacterial suspension before and after bacterial pellet preparation (log CFU/ml), before incorporation (log CFU/ml) and when incorporated in the 70.5% cocoa content chocolate (log CFU/g)

Table 1 Physico-chemical properties of the incorporated *B. animalis* subsp. *lactis* BB-12® chocolate with 70.5% cocoa content

Overall quality aspect	Chocolate with 70.5% (w/w) cocoa content	
	Control	With <i>B. animalis</i> BB-12®
Weight	2.28 ± 0.05 g	2.29 ± 0.06 g
Water activity	0.36 ± 0.01	0.62 ± 0.01
pH	6.20 ± 0.16	6.40 ± 0.04
Whiteness Index	22.0 ± 0.51%	22.1 ± 0.53%
Texture		
Firmness (Work of Penetration)	9824.68 ± 86.27 N	14513.92 ± 207.22 N
Hardness (Maximum Force)	1.38 ± 0.04 N	2.31 ± 0.19 N

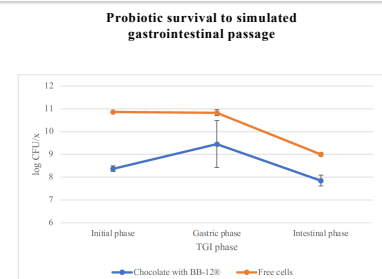


Fig. 2 Evolution of viable cell numbers of *B. animalis* subsp. *lactis* BB-12® in free form (log CFU/ml) or when incorporated in chocolate with 70.5% cocoa content (log CFU/g) during *in vitro* passage through gastrointestinal tract.

Conclusions

A chocolate matrix containing 70.5% cocoa content allowed the survival of *B. animalis* BB-12® at viable cell number levels of at least 10⁸ CFU/g throughout manufacture. The incorporation of probiotic in the chocolate matrix did not alter the physico-chemical properties of the food matrix. After an *in vitro* simulated GIT passage, this matrix ensured viable cell numbers of *B. animalis* BB-12® in level higher than the minimum required threshold of 10⁶ – 10⁷ CFU/g for a probiotic product.

Acknowledgements

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