Acta Sci. Pol. Technol. Aliment. 24(2) 2025, 217–227
www.food.actapol.net pISSN 1644-0730 eISSN 1898-9594 http://doi.org/10.17306/J.AFS.001313

ORIGINAL PAPER

Received: 18.11.2024
Accepted: 05.03.2025

# VALORISATION OF COCONUT OIL DREGS (BLONDO) AS AN INGREDIENT FOR FERMENTED PLANT-BASED ICE CREAM: A FUNCTIONAL AND SENSORY EVALUATION

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## ABSTRACT

**Background**. Blondo, a by-product of coconut oil production, is primarily used as livestock feed despite its high protein and fat content. The valorisation of blondo into ice cream products with high functional value has strong development potential. This research aims to create a LAB-fermented, blondo-based ice cream with varying fat content, and to characterise its properties.

**Material and methods**. A comprehensive investigation was conducted to determine the chemical properties of proximate composition, pH, and total acidity. The physical properties examined were overrun and melting time. The total lactic acid bacteria (LAB) was also analysed. Sensory evaluation was conducted using descriptive and hedonic methods to assess overall acceptance. Furthermore, an *in vitro* simulation of LAB resistance in the gastrointestinal tract was performed, specifically under low pH (pH:2) in the presence of pepsin in gastric simulation, as well as bile salt and pancreatin in a small intestine simulation, ensuring the validity of the findings.

**Results.** The final ice cream product consisted of three variations: light ice cream with a fat content of 4.77 ±0.36%, reduced-fat ice cream with a fat content of 7.22 ±0.86%, and regular-fat ice cream with a fat content of 9.16 ±0.25%. Higher fat content led to increased overrun and melting time. It also impeded the growth of LAB and the generation of acid. However, it enhanced the ability of LAB cells to withstand stress in the simulated gastrointestinal tract.

**Conclusion.** This research underscores the potential of blondo, a by-product of coconut oil production, to contribute to advances in food technology. It can serve as an alternative ingredient in functional foods in the form of ice cream, offering a novel method to protect LAB against extreme conditions in the gastrointestinal tract. This promising development offers hope for the future of food technology.

**Keywords:** blondo, coconut oil dregs, blondo-based fermented ice cream, in vitro gastrointestinal simulation, valorisation, functional food

## INTRODUCTION

The coconut tree is one of Indonesia's most significant agricultural commodities, and coconut oil is one of its most well-known products. There are two techniques for producing coconut oil: the dry and wet methods, which differ in the raw material used (Kurniawati et al., 2023). The dry method uses dried coconut flesh (copra), whereas the wet method uses fresh coconut flesh (Kurniawati et al., 2023).

Virgin Coconut Oil (VCO) is a coconut oil derivative. It is produced from fresh coconut flesh without

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heating, resulting in a product that is low in free fatty acids, translucent in colour and characterised by a distinctive coconut aroma (Husmaini et al., 2013). Although the processes used to produce coconut oil differ, both yield the same by-product: coconut oil dregs (blondo in Javanese). The wet method produces more blondo than the dry method. The production of VCO using a cold-pressed centrifugal method yields 68.84% coconut oil and up to 30.97% blondo as a by-product (Wijana et al., 2020).

Fifty grams of blondo contains 30.32% water, 26.59% fat, and 16.38% protein (Ansharullah et al., 2021; Harni et al., 2022). In the form of concentrate, the protein content of blondo can reach up to 80.30% (Ansharullah et al., 2021). Despite the current suboptimal use of blondo, i.e., as livestock feed, its high nutrient content indicates that it could be reutilized as an alternative protein source.

Blondo's high globulin and albumin content makes it a potential emulsifier (Kurniawati et al., 2023; Permatasari et al., 2015). Blondo concentrate dried for 3 hours at 100°C has a high emulsification capacity of 7.81% (Ansharullah et al., 2021). This suggests that coconut oil dregs (blondo) could be utilised in a wider range of food applications that rely on emulsification, while also contributing nutritional value. One of the emulsion-based products that blondo can be used to develop is ice cream. With a nutritional profile comparable to that of milk, blondo has the potential to serve as a medium for the growth of LAB (Setiaji et al., 2002), which may exhibit probiotic properties. This would enhance the functional value of blondo-based ice cream products.

Fermented ice cream is a modified form of ice cream made by adding an LAB starter culture, with fermentation occurring prior to freezing. Similar to regular ice cream, most LAB-fermented products, such as yogurt, cheese, and kefir, are milk-based (O'Brien et al., 2016). However, as innovation has progressed, many ice creams and LAB-fermented products have begun using plant-based ingredients to cater to vegetarians and consumers allergic to dairy products. The shift to plant-based food products is a growing trend despite various challenges, including the limited availability of processed plant-based products (Alcorta et al., 2021; Venter de Villiers et al., 2024). Therefore, research into innovative plant-based food products with enhanced functional properties has become increasingly urgent. The use of blondo supports this diversification by serving as a milk substitute in ice cream production. In addition, valorising blondo is a sustainable agricultural practice, transforming a low-value by-product into an innovative product with a higher market value.

Probiotics are live microorganisms that confer a health benefit on the host when consumed in adequate amounts. Generally, microorganisms with probiotic properties are derived from LAB, such as *Bifidobacterium*, *Lactobacillus*, and *Streptococcus*. However, the term probiotic can only be used to refer to a specifically identified strain; otherwise, the bacteria in question should be referred to as potentially probiotic LAB (Food and Agriculture Organization/World Health Organization, 2002). The health benefits of consuming probiotics include maintaining gastrointestinal health, modulating the immune system, and lowering blood glucose and serum cholesterol (Gul and Durante-Mangoni, 2024).

Different food matrix compositions affect the fate of LAB in the gastrointestinal tract (Rasika et al., 2021). Since LAB are expected to survive passage through the gastrointestinal tract and adhere to the surface of the colon, where they exhibit probiotic properties, the viability of LAB-containing food products is crucial. The minimum viable cell count of LAB in food believed to provide health benefits is in the range of 6–7 log CFU/g or mL (Kamil et al., 2020; 2021).

Product characteristics significantly influence consumer acceptance of new product innovations. Thanks to its protein and its fat content, blondo also contributes flavour and aroma, and may help improve texture when incorporated into food formulations. In ice cream, fat content affects not only mouthfeel and flavour but also texture and structural stability (Marimon-Valverde et al., 2024; Mostafavi, 2019). The lower the fat content, the icier and crumblier the ice cream (Rolon et al., 2017).

The main aim of this research was to reutilise blondo as an ingredient in LAB-fermented, plant-based ice cream. Specifically, the study evaluated the impact of different formulations on the quality and sensory acceptance of blondo-based, LAB-fermented ice cream, as well as its potential to serve as a carrier for LAB during gastrointestinal simulation.

## MATERIAL AND METHODS

### Research material

The materials used in this study were: blondo (Bibit Rejeki, Indonesia); coconut milk (Kara, Indonesia); sugar (Gulaku, Indonesia); Carboxy Methyl Cellulose (CMC) (Koepoe-Koepoe, Indonesia); yellow sweet potato purchased from a local market in Banyumanik, Semarang, Indonesia; LAB starter containing *Bifidobacterium longum*, *Lactobacillus rhamnosus*, *Lactobacillus casei*, *Lactobacillus helveticus*, *Lactobacillus bulgaricus*, *Lactobacillus acidophilus*, and *Streptococcus thermophilus* (Yogurtmert, Canada); de Man Rogosa Sharpe Agar (MRSA) (Merck, Germany); pepsin enzyme (Sigma-Aldrich; P7000, USA); bile salt (Oxoid No. 3, UK); pancreatin enzyme (Sigma-Aldrich; P7545, USA); HCl (Merck, Germany); NaOH (Merck, Germany); and phenolphthalein indicator.

### Production of LAB-fermented blondo-based ice cream

The LAB-fermented blondo-based ice cream was made using three formulations with different total fat contents: 5%, 8%, and 10%. The blondo was first dried at 60°C for 16 hours, then ground and mixed. The finely ground, dried blondo was combined with coconut milk in a ratio of 1:10.5, along with all the other ingredients. Soybean oil was used to adjust the fat content in each formulation: F1 (0% w/w), F2 (2% w/w), and F3 (6% w/w). The mixture was then pasteurized at 80°C for 15 minutes and allowed to cool until its temperature reached 39–41°C. It was subsequently mixed for 10 minutes while incorporating 10% LAB culture with an initial count of log 6 CFU/mL. Finally, the mixture was fermented at 40°C for 8 hours and frozen using an ice cream maker for 30 minutes.

### Proximate analysis

Proximate composition – including moisture content (gravimetrically), ash content (gravimetrically), crude protein (Kjeldhal method), and fat content (Gerber method) – was determined in accordance with the Indonesian Standard for ice cream (SNI 01-3713-1995) (Badan Standarisasi Nasional, 1995).

### Determination of total LAB

Total LAB was enumerated using the pour plate method with MRSA medium. In brief, one gram of ice cream was serially diluted, and 1 mL from an appropriate dilution was inoculated into a Petri dish, followed by the addition of MRSA medium. The plates were incubated at 37°C for 48 h. Total LAB counts are expressed as log CFU/g.

### Determination of total acid and pH value

Total acidity was analysed using the titration method with standardized 0.1 N NaOH, with results expressed as a percentage of lactic acid. The pH value was measured using a calibrated pH meter (Pen Type PH-009, Indonesia)

### Determination of overrun

The volume of ice cream mixture was measured before and after processing in an ice cream maker using a measuring cup (Marimon-Valverde et al., 2024). The percentage of overrun was calculated using Equation 1:

$$\% \text{ overrun} = \frac{\text{weight before freezing} - \text{weight after freezing}}{\text{weight after freezing}} \times 100 \qquad (1)$$

### Determination of melting time

Melting time was determined by allowing 10 grams of LAB-fermented, blondo-based ice cream – which had been frozen for 24 hours – to melt at room temperature (25–28°C) (Nugroho et al., 2020). The time taken for each sample to melt was recorded, and the results were analysed statistically.

### Determination of sensory characteristics

Sensory testing was conducted with 25 untrained male and female panellists aged 19–25 years in order to provide an overview of the product and its acceptability from the a general consumer perspective. A descriptive test was carried out to evaluate colour, taste, and texture. Meanwhile, a hedonic test was used to assess overall product acceptance. A four-point numerical scale was employed for the sensory analysis.

### Determination of LAB resistance in gastrointestinal simulation

The resistance of LAB cells in the gastrointestinal simulation was evaluated using methods adapted from previous research, with some modifications (Kamil et al., 2021). Simulated gastric juice was prepared by suspending 3 mg/mL pepsin in sterile saline solution (NaCl 0.85% w/v), which had an initial pH of 2. Simulated intestinal juice was prepared by suspending 1 mg/mL pancreatin and 0.3% bile salts in sterile saline solution (NaCl 0.85% w/v), which had an initial pH of 8. Up to 9 mL of simulated gastric and intestinal juice was added, in succession, to 1 g of ice cream and incubated at 36–37°C, which approximates normal human body temperature, accompanied by shaking using a water bath (Memmert, Germany). The gastric simulation was carried out for two hours, while the intestinal simulation was carried out for four hours. Cell viability and injured cells were analysed hourly throughout the simulations.

### Determination of injured cells, dead cells, and survival rate

Injured cells, dead cells, and survival rate were quantified to evaluate LAB cell resistance during the intestinal simulation, following Rahayu et al. (2024), with modifications. Injured cells were determined by inoculating serially diluted samples into MRSA medium supplemented with 3% NaCl (w/v), followed by incubation at 37°C for 48 hours. The following formulas (Eq. 2–4) were used to calculate the number of injured cells, dead cells, and the survival rate:

$$\%\ \text{dead cells} = \frac{(X0 - Xt)}{X0} \times 100 \qquad (2)$$

$$\%\ \text{survival} = \frac{Xt}{X0} \times 100 \qquad (3)$$

$$\%\ \text{injured cells} = \frac{(\text{viable cell in MRSA} - (\text{viable cell in MRSA} + 3\%\ \text{NaCl}))}{\text{viable cell in MRSA}} \times 100 \qquad (4)$$

where

X0 – initial viable cells (log CFU/g)

Xt – viable cells after incubation (log CFU/g).

### Statistical analysis

The parametric data were analysed using analysis of variance (ANOVA) in SPSS 26.0 for Windows, with a 5% significance level. Where significant effects were observed, Duncan's Multiple Range Test (DMRT) was used to identify differences between treatments. Sensory data were analysed using the non-parametric Kruskal-Wallis test. If significant effects were found, the Mann-Whitney test was applied to determine pairwise differences.

## RESULTS AND DISCUSSION

### Proximate characteristics

Table 1 displays the results of proximate analysis for each formulation of LAB-fermented blondo-based ice cream. Significant differences in fat, protein, water, and ash content can be observed among the formulations. Higher fat content was associated with lower levels of protein, moisture, and ash. As shown in Table 1, the measured fat content was lower than the fat content expected based on the formulation. Pasteurisation of the ice cream mixture at 80°C for 15 minutes may alter the fat composition through oxidation and the evaporation of volatile fatty acids (Pestana et al., 2015). Furthermore, the fermentation process involves catabolic reactions in which microorganisms decompose fat to support their growth, resulting in a decrease in fat content (Knez et al., 2023). According to SNI 01-3713-1995, the minimum required fat and protein contents in ice cream are 5% and 2.7%, respectively.

**Table 1.** Proximate composition of LAB-fermented blondo-based ice cream

| Component | F1 | F2 | F3 |
|---|---|---|---|
| Fat, % | 4.77 ±0.36$^{a}$ | 7.22 ±0.86$^{b}$ | 9.16 ±0.25$^{c}$ |
| Protein, % | 5.39 ±0.24$^{c}$ | 4.64 ±0.02$^{b}$ | 4.10 ±0.08$^{a}$ |
| Moisture, % | 49.44 ±0.27$^{a}$ | 49.33 ±0.32$^{a}$ | 45.15 ±0.61$^{b}$ |
| Ash, % | 2.44 ±0.04$^{a}$ | 2.33 ±0.03$^{a}$ | 1.97 ±0.08$^{b}$ |

Data are represented as mean ±standard deviation and were collected from four sets of experiments, each consisting of at least three repetitions of the analysis. Different lowercase superscripts indicate significant differences ($p < 0.05$).

Based on the classification by Chansathirapanich et al. (2016), ice cream containing ~5% fat is considered light ice cream, while ice cream with ~7.5% fat is referred to as reduced-fat ice cream. Ice cream with a fat content of ~10% is categorised as regular-fat ice cream.

## Overrun and melting time

Table 2 shows that increasing the fat content of LAB--fermented blondo-based ice cream significantly affects both overrun and melting time. Overrun is the increase in ice cream volume resulting from the incorporation of air during mixing (Marimon-Valverde et al., 2024). Because it affects ice cream quality and profitability, the amount of air is considered a critical factor in production. A similar increase in overrun was reported in the low-fat (2.5%), reduced-fat (5%), and regular-fat (10%) ice cream formulations studied by Chansathirapanich et al. (2016). Higher fat content in ice cream promotes the formation of more fat globules, which can form a three-dimensional (barrier) structure to trap air, thereby increasing overrun (Yan et al., 2021). Although a similar trend was observed in the present study, the overrun values for all LAB-fermented blondo-based ice cream were lower than those reported by Chansathirapanich et al. (2016).

**Table 2.** Overrun and melting time of LAB-fermented blondo-based Ice cream

| Formula | Overrun, % | Melting time, min. |
|---|---|---|
| F1 | 12.62 ±1.29$^a$ | 12.34 ±2.32$^a$ |
| F2 | 14.14 ±1.24$^a$ | 16.26 ±2.61$^b$ |
| F3 | 24.20 ±2.36$^b$ | 17.48 ±2.31$^b$ |

Data are represented as mean ±standard deviation and were collected from four sets of experiments, each consisting of at least three repetitions of the analysis. Different lowercase superscripts indicate significant differences ($p < 0.05$).

Various elements of the ice cream mixture and manufacturing process – such as extrusion and homogenization – can influence overrun. Overrun is affected by the presence of emulsifying agents and their emulsification capability (Kurultay et al., 2010; Rolon et al., 2017). Emulsifiers help stabilize fat and air bubbles in the ice cream after it has been mixed. However, the more fat in the ice cream mixture, the more emulsifiers are required to stabilize the fat globules, making the air bubbles in the mixture more unstable (Chansathirapanich et al., 2016).

In this study, yellow sweet potato was used to increase total solids. This likely inhibited the incorporation of air due to the denser, heavier mixture. This finding aligns with the results of Kurultay et al. (2010), who reported that higher total solid content increases viscosity, thus reducing overrun. In addition, the type of homogenization technique used can affect overrun. Conventional homogenization tends to increase overrun, while high-pressure homogenization generally reduces it (Wang et al., 2022). In the present study, the ice cream mixture was homogenized using a conventional mixer, which may explain the observed overrun values.

Gradual melting, keeping its shape, and gradual disintegration are significant qualitative factors that characterise an excellent ice cream. The higher the fat content, the longer the melting time. This is in line with the results of previous research by Abbas Syed (2018) and Marimon-Valverde et al. (2024). An increase in total fat globules can also affect the melting time of ice cream. The ice crystals that form during the freezing process will lie between the fat globules, so as the number of fat globules increases, so does the time required for the heat penetration process to melt the ice crystals, increasing the melting time (Yang et al., 2022). Hence, dairy fat or vegetable oil acts as an insulating barrier against heat by reducing the thermal diffusivity of the mixture (Marimon-Valverde et al., 2024).

## Total LAB, pH value, and total acidity

Table 3 presents the results for total viable LAB, total acidity, and pH. It can be seen that fat content significantly affects the total viable LAB and total acidity, which both tend to decrease as fat content increases, as well as pH, which tends to increase. Adding oil to the ice cream mixture, which increases the fat content, inhibits the growth of lactic acid bacteria in the ice cream mixture. This is primarily because it reduces the amount of water available for microbial growth and limits the space in which microbes can grow (Verheyen et al., 2020). Using soybean oil as a fat additive

**Table 3.** Total LAB, total acidity, and pH value of LAB-fermented blondo-based ice cream

| Formula | Total LAB log CFU/g | Total Acidity % | pH |
|---|---|---|---|
| F1 | 8.78 ±0.19$^b$ | 0.58 ±0.04$^b$ | 4.54 ±0.12$^a$ |
| F2 | 8.54 ±0.30$^b$ | 0.47 ±0.04$^a$ | 4.75 ±0.07$^b$ |
| F3 | 7.70 ±0.22$^a$ | 0.44 ±0.03$^a$ | 4.79 ±0.10$^b$ |

Data are represented as mean ±standard deviation and were collected from four sets of experiments, each consisting of at least three repetitions of the analysis. Different lowercase superscripts indicate significant differences ($p < 0.05$).

in LAB-fermented blondo-based ice cream can also reduce the metabolism of LAB. Increasing the amount of oil with high linoleic acid content in fermentation media can inhibit the growth of *Lactobacillus acidophilus* (Puniya et al., 2009). Phenolic substances in soybean oil can also inhibit the growth of *Streptococcus thermophilus* (Nurliyani et al., 2020). As a result, soybean oil can decrease the activity of LAB in the starter. Although the presence of fat inhibits the growth of LAB, the viable cell count still meets the recommended minimum for probiotic food products, which is 6–7 log CFU/g (Kamil et al., 2020; 2021).

Total acidity and pH values are related to total LAB, with which total acidity has a linear relationship. As lactic acid bacteria metabolise nutrients, they produce lactic acid; thus, a lower LAB count results in reduced total acidity (Wang et al., 2021). In contrast, pH has an inverse relationship with total acidity. During fermentation, LAB produce organic acids – such as lactic, citric, and acetic acids – which dissociate to release $H^+$ ions, thereby lowering the pH of the mixture.

## Sensory characteristics

Table 4 shows that the fat level of LAB-fermented blondo-based ice cream has a significant impact on its taste and texture. All formulations in the current study were rated between "sour" and "slightly sour". As fat content increased, the taste was perceived as less sour. Across the formulations, the texture of LAB-fermented blondo-based ice cream varied from crumbly to sandy. Crumbly ice cream is characterized by its brittleness and tendency to break apart readily when bitten, which is caused by the uneven crystallization

**Table 4.** The sensory attributes of LAB-fermented blondo-based ice cream

| Parameters | F1 | F2 | F3 | Scale |
|---|---|---|---|---|
| Taste | 3.00 ±0.63$^a$ | 2.36 ±0.93$^b$ | 2.16 ±0.83$^b$ | 1. not sour<br>2. slightly sour<br>3. sour<br>4. very sour |
| Colour | 2.12 ±0.43 | 2.12 ±0.43 | 2.20 ±0.40 | 1. broken white<br>2. light brown<br>3. brown<br>4. dark brown |
| Texture | 2.32 ±0.79$^a$ | 2.64 ±0.74$^a$ | 3.12 ±0.71$^b$ | 1. icy<br>2. crumbly<br>3. sandy<br>4. smooth |
| Overall acceptance (hedonic) | 2.44 ±0.75 | 2.64 ±0.89 | 2.52 ±0.98 | 1. dislike<br>2. slightly like<br>3. like<br>4. like a lot |

Data are represented as mean ±standard deviation from 25 panelists. Different lowercase superscripts in the same column indicate significant differences ($p < 0.05$).

of ice (Abbas Syed, 2018). Sandy ice cream is defined by a granular mouthfeel (Abbas Syed, 2018). The findings suggest that a higher fat content in ice cream leads to a smoother texture, while retaining a slightly grainy feel.

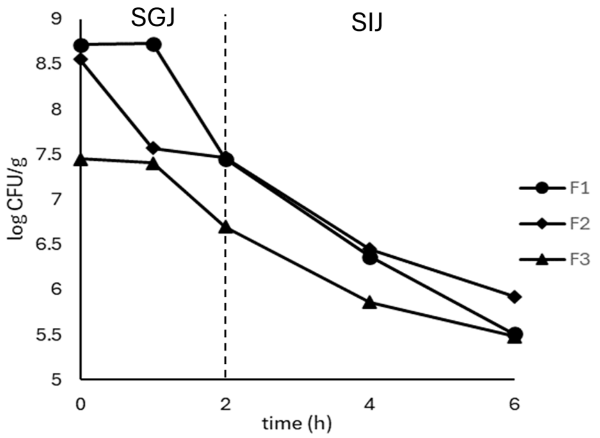
The acidity of the LAB-fermented blondo-based ice cream decreased as fat content increased, as the total acidity decreased, and as the pH value increased. As fat content increased, the sourness of the ice cream was also reduced. The sour taste is influenced by the presence of lactic acid produced by LAB. Lower LAB counts lead to reduced lactic acid formation, resulting in a less sour product (Wang et al., 2021). Increased fat content also contributes to a smoother texture. Fat plays a key role in the formation of ice cream structure: the higher the fat content, the smoother the texture (Zhao et al., 2023). Texture, in turn, affects melting time. Coarse-textured ice cream has low melting resistance, so it melts faster, while ice cream with a smooth texture melts more slowly.

Ice crystals play a crucial role in achieving a smooth and creamy texture in ice cream (Kurultay et al., 2010; Marimon-Valverde et al., 2024; Mostafavi, 2019; Rolon et al., 2017). Smaller ice crystals are necessary to achieve the desired smoothness. In contrast, when ice crystals are larger, ice cream texture becomes coarse and icy. The formation of these crystals is affected by the proportion of water in the ice cream mixture. According to the results of the proximate analysis, ice cream with higher fat levels tends to have lower water content. In addition, fat content can act as a tactile lubricant, helping reduce the perception of coarse texture caused by large ice crystals (Choi and Shin, 2014; Marimon-Valverde et al., 2024; Mostafavi, 2019). This aligns with research by Mostafavi (2019), which indicates that increased levels of fat enhance the creaminess, mouthcoating, and smoothness of ice cream while reducing its iciness and coarseness.

## Gastrointestinal tract simulation

Examining the resistance of LAB in the intestinal system is crucial, as the capacity of probiotics to grow and establish themselves in the host's intestine is directly linked to their functional utility, including their probiotic properties. Food can serve as both a carrier and a protector of probiotics under harsh gastrointestinal conditions, including low pH levels (2.5–3.5) in the stomach, the presence of bile salts in the small intestine, and the presence of digestive enzymes like pepsin and pancreatin (Matouskova et al., 2021; Ranadheera et al., 2012).

Figure 1 illustrates the viability of LAB cells during a simulation of the gastrointestinal tract for each formulation. The results unambiguously show a decline in the viability of LAB cells following a 2-hour incubation in the gastric simulation (SGJ), with reductions of 1.27, 1.08, and 0.75 log CFU/g in the F1, F2, and F3 treatments, respectively. A further drop in cell viability was observed in the small intestine simulation (SIJ) following exposure to the gastric phase, with reductions of 1.94, 1.55, and 1.22 log CFU/g in the F1, F2, and F3 treatments, respectively. These results indicate that the F3 formulation was more effective in protecting LAB cells during the simulation of the gastrointestinal tract. According to the linear equations in Table 5, the F3 treatment has the gentlest slope, where the slope represents the rate at which LAB die per hour in the simulation (Alizadeh-Sani et al., 2019). A gentler slope indicates a slower rate of bacterial death during the experiment.



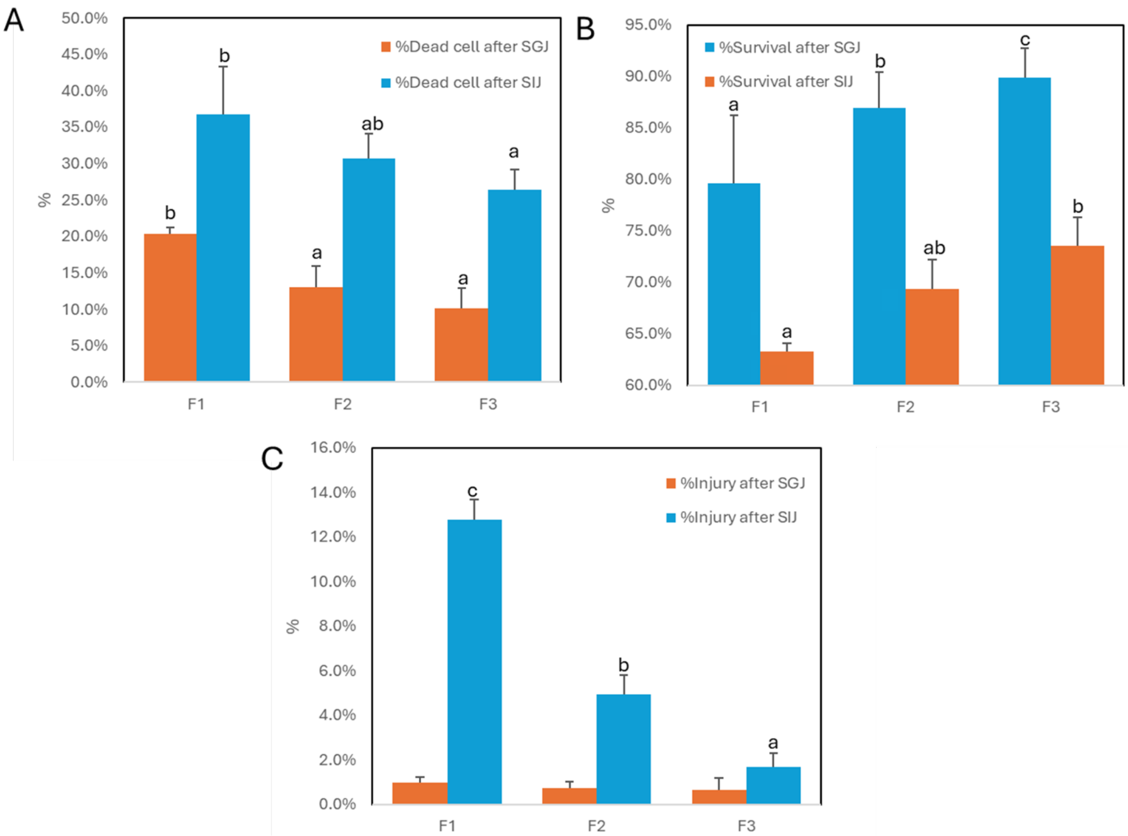

SGJ – Simulated Gastric Juice; SIJ – Simulated Intestinal Juice. Data are represented as mean ±standard deviation and were collected from three sets of experiments, each consisting of at least two repetitions of the analysis.

**Fig. 1.** Viability of LAB cells during gastrointestinal simulation

**Table 5.** Linearity of LAB cell viability during gastrointestinal tract simulation

| Formula | Equation | $R^2$ |
|---|---|---|
| F1 | y= –0.5798x + 8.859 | 0.9591 |
| F2 | y= –0.4165x + 8.2727 | 0.9510 |
| F3 | y= –0.3619x + 7.5186 | 0.9567 |

During gastrointestinal tract simulations, probiotics encounter highly challenging conditions that impact their ability to survive. Stress environments, such as those found in the gastrointestinal system, can impact the integrity of the cell membrane, intracellular pH, and the functionality of enzymes in probiotic cells, as stated by Cruz Rodrigues et al. (2019). These conditions affect the physiological state of the probiotic cells, which can be categorized as alive (active and




SGJ – Simulated Gastric Juice; SIJ – Simulated Intestinal Juice. Data are represented as mean ±standard deviation and were collected from three sets of experiments, each consisting of at least three repetitions of the analysis. Different lowercase superscripts indicate significant differences ($p < 0.05$).

**Fig. 2.** The fate of LAB cells during gastrointestinal tract simulation. A – % dead cells of LAB, B – % injured cells of LAB, C – % survival rate of LAB cells

culturable), dormant/injured (inactive but culturable), or dead (inactive and not culturable). Accordingly, this study also examined LAB cells that experienced injury or death. The results shown in Figure 2 A–C align with those in Figure 1A and Table 5, indicating that when the fat content increases, there is a corresponding decrease in the % dead cells and % injured cells, while the % survival rate increases.

A previous study revealed that ice cream produced from goat milk containing approximately 10% fat exhibited superior preservation of probiotic viability compared to both plain yogurt and fruit yogurt, which had fat concentrations of around 5% (Ranadheera et al., 2012). Elevated fat levels in the food matrix can enhance the ability of probiotic bacteria to withstand excessive acid (buffering capacity) (Matouskova et al., 2021; Rasika et al., 2021). Additionally, higher fat content can provide a physical defence against the effects of digestive enzymes (Rodrigues et al., 2019). Ranadheera et al. (2012) conducted a study that showed an increase in the pH of a gastric simulation solution after the addition of food matrices such as yogurt and ice cream, with ice cream causing the highest increase. The absence of fermentation in ice cream results in a higher final pH than is observed with yogurt. In addition, Salaün et al. (2005) define the buffering capacity of a food product as a physicochemical property that provides resistance to acidification or alkalisation. Table 1 shows a direct correlation between the fat content of LAB-fermented blondo-based ice cream and its pH, with higher fat content resulting in a higher pH. Additionally, higher fat content leads to lower total acidity. This demonstrates the buffering capacity of LAB-fermented blondo-based ice cream, with formulation F3 exhibiting the highest buffering capacity.

The % dead cells and % injured cells in the small intestine simulation are more pronounced than those observed in the gastric simulation. While differences in incubation time may have contributed to this result, gastric stimulation induces greater stress levels as a result of elevated acidity (pH 2.5–3.5), which can be mitigated by the buffering capacity of the food matrix (Matouskova et al., 2021; Ranadheera et al., 2012; Rasika et al., 2021). In the simulation of the small intestine, bile salt acts as a natural detergent, aiding in the emulsification of fat and exhibiting antibacterial activity by damaging the membranes of probiotic cells (Matouskova et al., 2021; Ranadheera et al., 2012; Rasika et al., 2021). The presence of fat in the food matrix helps protect probiotics by shielding bacterial cells from direct exposure to bile salts (Ranadheera et al., 2012; Rodrigues et al., 2019). In a previous experiment, probiotic cells exposed to simulated digestive stress showed surface damage such as shrinkage and thinning, as observed with SEM (Scanning Electron Microscopy). This damage was not observed in probiotic cells protected by a food matrix, including fat components (Cruz Rodrigues et al., 2019). Damage to the cell wall can further affect the probiotic's ability to adhere to the intestine and successfully colonize the host (Ranadheera et al., 2012).

## CONCLUSION

Blondo (also known as coconut dregs), a by-product of VCO production, holds significant promise as a substitute food ingredient, rather than being limited to its current use as low-value animal feed. When used as the primary ingredient in ice cream and combined with LAB cultures, blondo can produce a plant-based ice cream with desirable physical and chemical properties that is well-received in terms of taste and sensory experience.

Moreover, blondo-derived plant-based ice cream can serve as an effective vehicle for LAB to traverse the gastric and small intestinal environments, as its lipid content provides buffering capacity and shields cells from the harsh conditions of the digestive tract. This study presents data indicating that by-products generated during VCO production possess significant potential for utilisation in functional foods. Their use could enhance the market value of coconuts and contribute to a zero-waste manufacturing system.

## DECLARATIONS

### Data statement

All data supporting this study has been included in this manuscript.

### Ethical Approval

Not applicable.

### Competing Interests

The authors declare that they have no conflicts of interest.

## OPEN ACCESS



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