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Improving Probiotic Viability: The Effect of *Alyssum homolocarpum* gum as an Encapsulation Material

Akram Sadat Tabatabaee Bafroee[1], Negar Heydari[2], Elahe Nazari[2], and Saeedeh Shojaee-Aliabadi[3]*

ABSTRACT

As a practical approach, spray-drying microencapsulation is employed to protect probiotic cultures from intense thermal treatments and physiological stresses in the gut. Nonetheless, ensuring the survival of *Lactobacillus acidophilus* La-5 remains a significant concern when subjected to industrial-scale drying at high inlet temperatures (140–150°C). This study investigated the protective effects of *Alyssum homolocarpum* seed (AHS) gum, combined with two wall systems: low methoxyl pectin/β-glucan and sodium alginate/maltodextrin. Fabrication of the microcapsules involved a spray-dryer operating with a feed-in temperature of 140–150°C, while the discharge air was maintained at 80–85°C. The findings indicated that AHS gum significantly boosted both encapsulation efficiency (EE %) and the resilience of probiotic cells. Specifically, the survival rate of these microencapsulated strains improved from 68.4% up to 82.6% when exposed to thermal stress (55–75 °C) for periods of 1 to 10 minutes. Furthermore, SEM images revealed that the addition of AHS gum resulted in smoother microcapsule surfaces with reduced porosity (mean diameter: 18.5–22.3 µm). Under simulated gastrointestinal conditions, the LMP/β-glucan/AHS formulation retained 79.8% viability after 120 min in SIF, significantly higher than the control (28.0%). These findings confirm that AHS gum acts as an effective co-wall material, enhancing both the physical stability and probiotic resilience during processing and storage.

KEYWORDS: *Alyssum Homolocarpum* Seed (AHS), *L. acidophilus* La-5, Microencapsulation, Probiotic viability, Spray-drying.

[1] Department of Biology, ET.C, Islamic Azad University, Tehran, Islamic Republic of Iran.
[2] Department of Nutrition, Electronic Health and Statistics Surveillance Research Center, Science and Research Branch, Islamic Azad University, Tehran, Islamic Republic of Iran.
[3] Department of Food Science and Technology, Faculty of Nutrition Sciences and Food Technology, National Nutrition and Food Technology Research Institute, Shahid Beheshti University of Medical Sciences, Tehran, Islamic Republic of Iran.

*Corresponding author; e-mail: s_shojaee@sbmu.ac.ir

1-INTRODUCTION

The integration of probiotics into functional foods is a critical strategy for enhancing consumer health, offering benefits such as protection against intestinal diseases and mitigating the risk of colorectal malignancies (Villarruel-López et al., 2017; Bhagwat et al., 2020). However, a significant hurdle for food manufacturers involves the substantial loss of cell integrity during transit through the digestive system and the harsh conditions of industrial processing. In order to provide therapeutic benefits, these beneficial microbes must arrive at the intestinal site with a viable count of no less than 10^9 CFU per daily serving (Commission, 2011; Soukoulis et al., 2014). Microencapsulation via spray drying has been confirmed as a desirable and cost-effective technology to protect sensitive core materials from environmental stresses (Bhagwat et al., 2020; Dong et al., 2020; Ahmad et al., 2021). Despite its efficiency, the high thermal stress during the process remains a significant drawback. Therefore, the selection of wall materials is crucial; they must possess high glass transition temperatures and excellent film-forming properties to create a physical barrier. While sodium alginate and low methoxyl (LM) pectin are frequently used for their biocompatibility, they often result in porous structures that may not provide sufficient protection against intense heat or rapid dehydration (Fritzen-Freire et al., 2012; Rajam et al., 2012). Recently, natural seed gums have gained attention as co-encapsulants due to their unique functional properties. The gum extracted from *Alyssum homolocarpum* seeds (AHS) constitutes a sophisticated, high-molecular-weight carbohydrate polymer, predominantly featuring xylans and glucomannans in its structural composition (Koocheki et al., 2010). AHS gum exhibits high viscosity at low concentrations and excellent thermal stability, which are essential for protecting probiotic cells during heat processing. Despite its promising techno-functional attributes, the application of AHS gum as a novel wall material for the encapsulation of probiotics has not been previously explored. This study addresses this knowledge gap by investigating the synergistic effects of AHS gum in combination with conventional carriers.

In this study, we hypothesize that the interaction between the carboxylic groups of sodium alginate/pectin and the branched structure of AHS gum can lead to a more cohesive and less porous wall matrix. This architectural refinement is expected to reduce the heat transfer rate to the encapsulated cells, thereby improving their survival during both the spray-drying process and subsequent exposure to the acidic environment of the gastrointestinal tract (Homayouni-Rad et al., 2021).

This research aimed to develop microcapsules for the probiotic strain *L. acidophilus* La-5 through a spray-drying approach. Polysaccharide-based coatings, specifically combinations of low methoxyl pectin/β-glucan and sodium alginate/maltodextrin, were evaluated for their encapsulation efficiency, both with and without the incorporation of AHS gum as a stabilizing agent**.** Furthermore, the study investigated the effectiveness of these wall structures in maintaining cell viability during thermal exposure and under simulated digestive environments, including SGF and SIF. In addition, the impact of AHS gum on the microstructural characteristics and dimensional distribution of the fabricated particles was rigorously investigated. This evaluation aimed to identify the optimal formulation for achieving robust and durable probiotic carrier systems.

2- MATERIALS and METHODS

2-1- Materials and Bacterial Cell

Low Methoxyl Pectin (LMP) and sodium alginate (Sigma-Aldrich, USA) were supplied by Qlab (Canada). β-glucan and maltodextrin were sourced from CFF GmbH & Co. KG (Germany) and Behmalt (Iran), respectively. *Alyssum homolocarpum* seeds were acquired from a regional botanical market in Tehran, Iran. The seed gum was subsequently extracted and purified according to the established mucilage extraction methodology (Koocheki et al., 2010). Freeze-dried gum was ground, sieved, packed, and kept in cool and dry condition. Following lyophilization, the gum was pulverized, passed through a sieve, and stored in an airtight container under refrigerated and low-humidity conditions.

The *Lactobacillus acidophilus* La-5 probiotic culture (ID K08016), purchased from Tak Gen Company (Tehran, Iran), was activated in MRS broth (Qlab, Canada (under a 15 h incubation period at a constant temperature of 37°C.

2-2- Probiotic Inoculum Preparation

To prepare the probiotic inoculum, *Lactobacillus acidophilus* La-5 was activated through sequential cultivations in MRS broth, incubating at 37 °C for 18 h until the late exponential growth phase was reached. Harvesting of bacterial cells was performed through centrifugation (2683 × g, 20 min, 4 °C), followed by two washing cycles with sterile 0.1% (w/v) peptone water to remove residual media. The resulting pellet was subsequently dispersed within peptone water, yielding a standard stock suspension adjusted to a cell density of approximately 10^{10} CFU/mL. To confirm

the final viable count, the pour plate technique was employed on MRS agar after a 72 h anaerobic incubation at 37 °C (Arenales-Sierra et al., 2019).

2-3- Fabrication of the Microencapsulation Mixture

The microencapsulation mixtures were prepared using two different polysaccharide bases: low methoxyl pectin (LMP) / β-glucan and sodium alginate/maltodextrin. For the LMP-based matrix, 3 g of LMP and 1.5 g of β-glucan were dissolved in 225 mL of sterile deionized water followed by 1 h of magnetic stirring at 60 °C to yield a transparent, uniform blend. This mixture was then pasteurized at 90 °C for 10 min to eliminate potential microbial contamination. For the alginate-based system, a 3% (w/v) sodium alginate solution (225 mL) was formulated and allowed to hydrate at 5 °C for a 24 h period. Thereafter, 24 g of maltodextrin was incorporated over a 15 min mixing interval, and the final solution was subjected to thermal sterilization via autoclaving (121 °C for 15 min). To investigate the effect of AHS gum, each of the prepared base solutions (LMP-based and alginate-based) was divided into two equal portions of 112.5 mL each. One portion of each base was kept as a control, while the other portion was incorporated with 1.5 g of AHS gum (pre-dissolved in 75 mL of sterile water via 15 min of stirring). This resulted in four distinct wall material treatments, including; LMP/β-glucan (Control), LMP/β-glucan with AHS gum, alginate/maltodextrin (Control), and alginate/maltodextrin with AHS gum. Finally, 50 mL of the probiotic cell suspension (approximately 10^{10} CFU/mL) was homogenized into each matrix for 5 min. Consequently, the total feed volume for the spray-drying process was162.5 mL for the control treatments and 237.5 mL for the treatments containing AHS gum. Initial cell viability was assessed using the conventional pour-plate methodology on MRS agar after 72 h of anaerobic incubation at 37 °C (Uday et al., 2014; Mahmoud et al., 2020).

2-4- Spray Drying

Microencapsulation was performed using a Büchi Mini Spray Dryer B-290 (Büchi Labortechnik AG, Flawil, Switzerland). The drying parameters were fixed at an air inlet temperature of 120 ± 2 °C, an aspiration capacity of 100% (approx. 35 m^3/h), and a liquid delivery rate of 3 mL/min. Throughout the process, the exit air temperature was stabilized at 67 ± 2 °C across all formulations. The produced microencapsulated powders were harvested and preserved in sealed, moisture-resistant pouches at 4 °C prior to subsequent characterization.

2-5- Encapsulation Efficiency (EE)

The efficiency of encapsulation (EE), reflecting the survival rate of probiotic cells post-spray drying, was assessed by dissolving 1 g of the microencapsulated powder in 9 mL of sterile phosphate buffer solution (PBS, 0.1 M, pH 7.4). To achieve complete liberation of encapsulated cells, the resulting suspension was agitated for 30 min at ambient temperature. Viable cell counts were then quantified via the pour-plate method on MRS agar, following the established protocol. The encapsulation efficiency was computed using the equation below:

(1) EE (%) = (N / N0) x 100

Where N denotes the count of viable cells (log CFU/g) in the final microencapsulated powder, and N_0 represents the count of viable cells (log CFU/g) in the feed mixture prior to spray drying (Martin et al., 2013).

2-6- Microcapsule Morphology and Size

To investigate the structural morphology, a scanning electron microscope (SEM; LEO S-440i, Cambridge, UK) was employed. For this purpose, a thin layer of the microencapsulated powder was deposited onto a sample holder and gold-sputtered (SC-7160, England) to enhance electrical conductivity. Surface imaging was subsequently conducted via secondary electron detection at a 15 kV accelerating potential. Furthermore, the particle size distribution of the resulting powders was quantified using a laser diffraction-based analyzer (NanoTEC, Germany), following the manufacturer's analytical protocols.

2-7- Thermal Stability of Microcapsules

The thermal stability of the microencapsulated probiotics was investigated following the methodology described by Lasta et al. (2021), with minor adaptations. For each of the four treatments, as well as the free-cell control, 1 g samples were independently suspended in 29 mL of phosphate-buffered saline (0.1 M, pH 7.4) within sterile test tubes. These preparations were subsequently subjected to controlled heat stress at 55 °C, 65 °C, and 75 °C for durations of 1 min and 10 min. Following the thermal exposure, the survival of the bacterial cells was quantified using the pour-plate procedure detailed in the preceding sections (Lasta et al., 2021).

2-8- Survival Assessment of Microencapsulated *Lactobacillus acidophilus* La-05 in Simulated Gastrointestinal Conditions

To evaluate the gastrointestinal resilience of both free and microencapsulated *L. acidophilus* La-05, 1 g of each sample was subjected to 9 mL of simulated gastric fluid (composed of 0.08 mol HCl and 0.2% NaCl, adjusted to pH 1.55). This phase was conducted at 37 °C under strictly anaerobic conditions, with bacterial enumeration performed at 30 min intervals (0, 30, 60, 90, and 120 min) using the pour-plate technique on MRS agar. For the subsequent intestinal transit simulation, 1 mL aliquots were harvested via centrifugation and introduced into 9 mL of a specific intestinal fluid (containing 0.6 g NaCl, 0.0835 g KCl, 0.022 g $CaCl_2$, and 0.1368 g sodium bicarbonate, pH 7.5). These preparations were incubated for an additional 150 min at 37 °C. The survival rates of the encapsulated and free *L. acidophilus* LA-05 cells were then quantified by plating on MRS agar media to determine their final viability (Mirzaei et al., 2011).

2-9- Statistical Analysis

Data analysis was performed using SPSS statistical software (version 21). To assess the significance of differences between the various treatments, a one-way analysis of variance (ANOVA) was executed, complemented by Duncan's multiple range test for post-hoc comparisons. Statistical significance was established at a confidence level of 95% ($p < 0.05$).

3- RESULTS and DISCUSSION

3-1- Microstructural Morphology and Particle Size

The physical architecture and morphological characteristics of microcapsules affect their permeability, size, and particle fragility features of trapped probiotic cells. Figure 1(a-d) shows the morphology of the encapsulated *L. acidophilus* La-05 after the spray-drying technique. SEM micrographs of the bacteria encapsulated using solely LMP/β-glucan or alginate/maltodextrin matrices (in the absence of AHS gum) exhibited notable surface irregularities, including grooves, wrinkles, and roughness (Figure 1a and 1c). These features likely originated from rapid dehydration during the spray-drying operation, leading to significant particle shrinkage. Conversely, the incorporation of AHS gum into both wall systems resulted in microcapsules with a more spherical geometry and a smoother surface (Figure 1b and 1d). Such structural uniformity is crucial for enhancing the loading efficiency of probiotic cells. It appears that the synergistic effect of diverse wall materials plays a pivotal role in the surface formation of the resulting

powders. These observations align with previous research investigating the use of maltodextrin, β-glucan, Guar gum, Arabic gum, and various seed oils in spray-drying applications (Fazilah et al., 2019; Kuck and Noreña, 2016; de Barros Fernandes et al., 2014; Chew et al., 2018). Furthermore, the presence of surface fractures or cracks is known to compromise cell viability during gastrointestinal transit by allowing the penetration of detrimental agents such as bile salts, gastric acids, and gases. As noted by Fernandes et al.(de Barros Fernandes et al., 2014), excessive surface wrinkling can also impair the flow-ability of spray-dried powders. Consequently, the AHS gum-coated microcapsules in this study, characterized by their smoother surfaces, are expected to exhibit superior fluidity and protection for the encapsulated *L. acidophilus* La-05.

The particle size analysis of microcapsules containing the entrapped probiotic cells (Figures 2(a-d)) revealed that the average diameter (VMD) for LMP/β-glucan-based and sodium alginate/maltodextrin-based microcapsules without AHS gum was 19.24 µm and 24.42 µm, respectively, while the VMD for LMP/β-glucan-based microcapsules with AHS gum and for the sodium alginate/maltodextrin-based microcapsules with AHS gum were 21.12 µm and 28.54 µm, respectively. These results indicate that microcapsules coated with AHS gum were slightly larger than those without it, which is supported by related studies (Akbarbaglu et al., 2021; Damodharan et al., 2017). Moreover, irrespective of the AHS gum coating, the sodium alginate/maltodextrin matrices consistently yielded larger particle dimensions compared to the LMP/β-glucan systems. This observation underscores the significant influence of the wall material's chemical nature on the final diameter of the microcapsules (Călinoiu et al., 2019). Overall, the sizes recorded in this study were well-aligned with the 10–100 µm range proposed by Fang et al. (2012) for spray-dried probiotic powders (Fang et al., 2012). It is also noteworthy that maintaining a diameter below 40 µm is essential for achieving an optimal sensory profile (mouth-feel) in food applications. Furthermore, the spray-drying process remains a cost-effective strategy for producing microparticles within the sub-100 µm threshold (Călinoiu et al., 2019; Magazù et al., 2008).

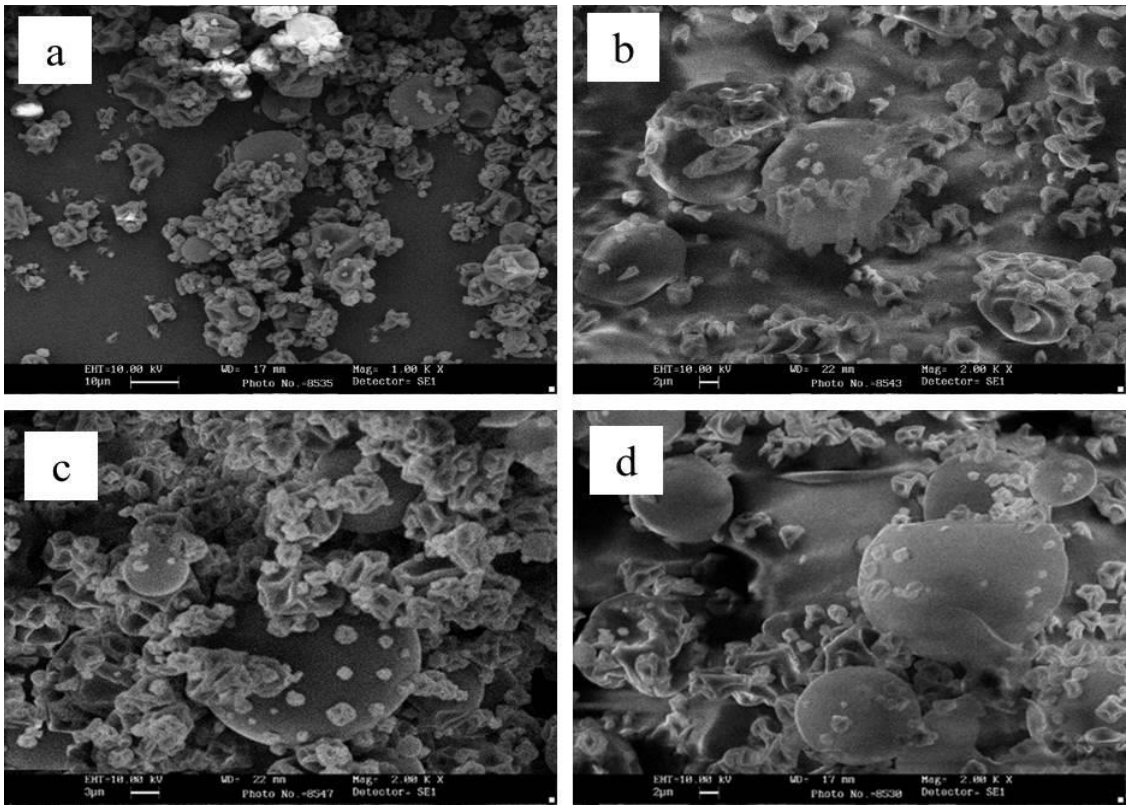

Figure 1. SEM micrographs illustrating the morphological features of encapsulated *L. acidophilus* La-05 produced via the spray-drying method. **(a-b)** Sodium alginate/maltodextrin-based microcapsules with and without *Alyssum homolocarpum* seed gum. **(c-d)** LMP/β-glucan-based microcapsules with and without *Alyssum homolocarpum* seed gum.

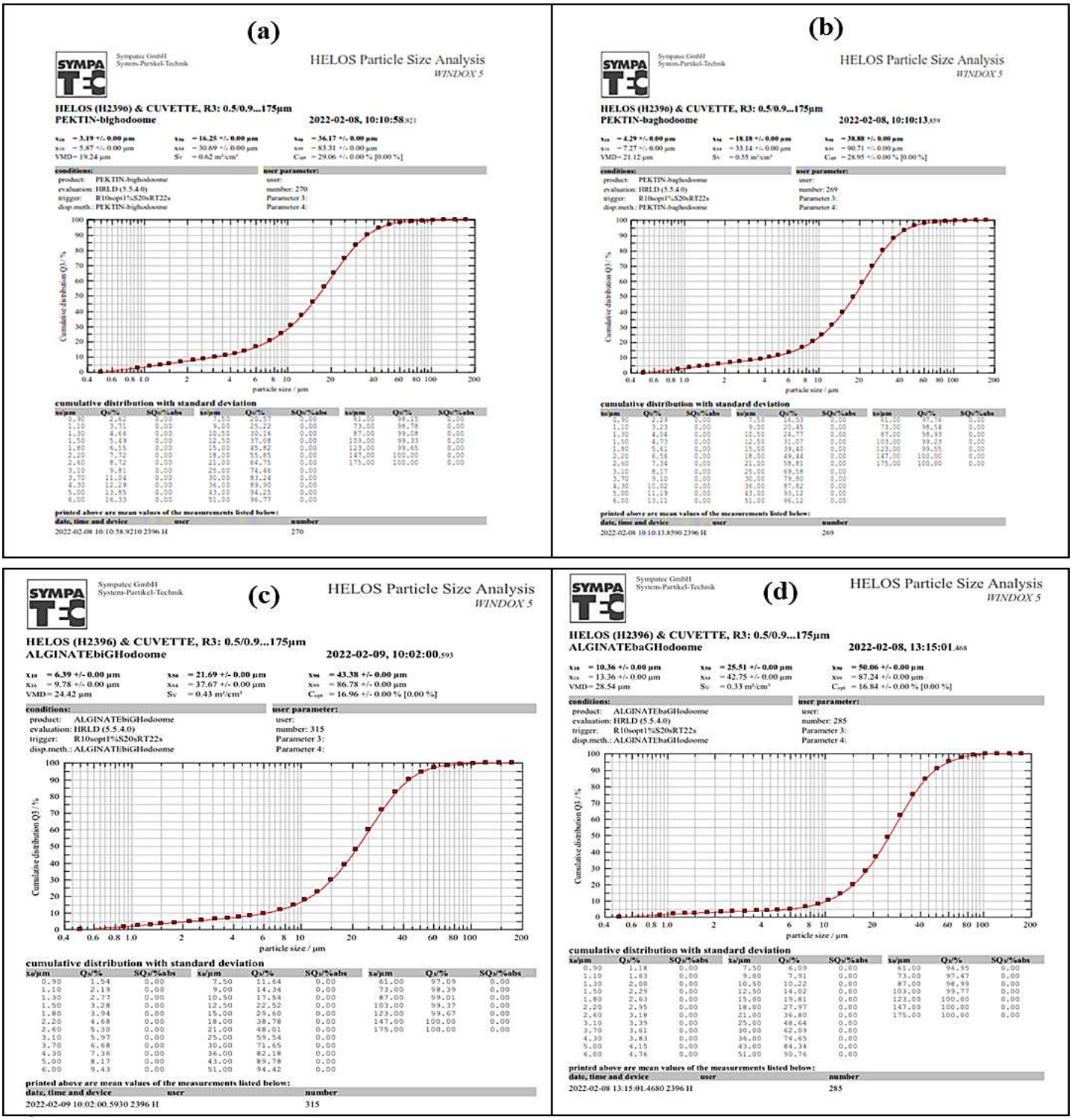

Figure 2. Particle Size Analysis of the encapsulated *L. acidophilus* La-05 obtained through the spray-drying process. **(a-b)** LMP/β-glucan-based microcapsules with and without *Alyssum homolocarpum* seed gum. **(c-d)** Sodium alginate/maltodextrin-based microcapsules with and without *Alyssum homolocarpum* seed gum.

3-2- Thermal Stability and Microencapsulation Efficiency (EE)

The results indicated that the dehydration via spray drying led to a decrease in the concentration of the bacterial cells in all tested samples. However, the loss of viability was less significant in the alginate-encapsulated groups than in those using pectin-based wall materials. Various process factors affect the thermal resistance of particles during spray drying, including atomization air pressure, feeding rate, and particle diameter, among which the particle size during the process is very effective (Huq et al., 2013). The presence of a thicker layer around the cells and larger particle size in samples with alginate may be a reason why probiotic viability was higher after spray drying. Additionally, alginate is a polysaccharide with high thermal resistance. Adding AHS gum increased bacterial viability in both alginate and pectin treatments. In general, the presence of polysaccharides in the capsule formula can increase probiotic cell viability. According to the data presented in Table 1, the sodium alginate/AHS formulation exhibited the maximal entrapment efficiency (94.2%) following the spray-drying operation (at 25 °C). This was closely followed by the LMP/β-glucan composite containing AHS, which achieved an efficiency of 91.2%. It seems that AHS gum acted as a thermoprotective compound for cells during drying and increased bacterial survival (Homayouni-Rad et al., 2021). Moreover, pectin- and alginate -based capsules are naturally porous, but this disadvantage can be overcome by modifying the capsule structure using different compounds (Dong et al., 2013). Therefore, adding AHS gum to the encapsulation formulations was successful in filling these pores and increasing the uniformity of the capsules, which is consistent with the SEM images. Dense structures create more thermal resistance by increasing particle size and densification (Shaharuddin and Muhamad, 2015).

The impact of thermal treatment on the viability of both free and microencapsulated bacterial cells, along with their respective EE values, is summarized in Table 1. Thermal treatment caused a decrease in bacterial population compared to ambient temperature at all three investigated temperatures, which significantly increased with exposure time ($p < 0.05$). Notably, thermal exposure at 55 °C for 1 versus 10 min produced statistically comparable results, indicating no marked divergence based on duration at this temperature. Exposure to elevated temperatures can trigger the thermal denaturation of critical macromolecules, including proteins and nucleic acids, ultimately leading to bacterial inactivation. After 10 min of thermal stress at 75 °C, all samples exhibited a substantial decline in viability. Nevertheless, the microencapsulated strains demonstrated significantly higher resilience than their free counterparts. Specifically, the sodium

alginate/AHS matrix preserved encapsulation efficiency (EE) of 64.4% even under these extreme conditions, whereas the non-encapsulated population suffered a much more drastic collapse. This suggests that the complex polymeric layers served as an effective thermal barrier, slowing the heat penetration and fostering superior probiotic survivability within the core.

The results indicated that alginate-derived capsules surpassed LMP/β-glucan samples in thermal endurance ,showing enhanced EE values due to their intrinsic heat-resistant characteristics (Corona-Hernandez et al., 2013). AHS addition bolstered the thermal defense mechanism of the studied matrices, with the alginate/maltodextrin/AHS blend achieving peak overall stability. This phenomenon is likely due to AHS acting as a structural modifier that increases particle size and density, effectively prolonging the heat diffusion route and reducing the rate of thermal transfer (Shaharuddin and Muhamad, 2015). Notably, AHS played a more critical role in the LMP/β-glucan systems, as it successfully compensated for the limited heat resistance typically associated with pectin-based microparticles (Homayouni-Rad et al., 2021).

Table1. Survival of free and LMP/β-glucan and sodium alginate/maltodextrin-based encapsulated *L. acidophilus* La-05 with and without *Alyssum homolocarpum* seed (AHS) gum and Encapsulation Efficiency (EE) after thermal treatment.

Time (min)	Temperature (°C)	free cells (Log CFU.g^{-1})	Sodium alginate-based encapsulated cells				LMP/β-glucan -based encapsulated cells			
			without AHS (Log CFU.g^{-1})	EE (%)	with AHS (Log CFU g$^{-1)}$	EE (%)	without AHS (Log CFU.g^{-1})	EE (%)	with AHS (Log CFU.g^{-1})	EE (%)
0	25	10.00±0.01^{a}	8.85±0.045^{c}	88.50	9.42±0.06^{b}	94.20	8.60±0.01^{d}	86	9.12±0.02^{c}	91.20
1	55	8.60±0.005^{a}	7.46±0.02^{b}	74.60	8.12±0.04^{c}	81.20	7.39±0.01^{d}	73.90	7.69±0.01^{c}	76.90
10	55	8.54±0.01^{a}	7.43±0.035^{b}	74.30	8.01±0.03^{b}	80.10	7.11±0.02^{d}	71.10	7.56±0.04^{c}	75.69
1	65	8.42±0.02^{a}	7.00±0.005^{b}	70.00	7.67±0.03^{b}	76.70	6.75±0.02^{c}	67.50	6.88±0.07^{c}	68.80
10	65	7.51±0.00^{a}	6.22±0.04^{c}	62.20	7.14±0.03^{b}	71.40	5.15±0.03^{e}	51.50	5.42±0.05^{d}	54.20
1	75	7.44±0.03^{a}	5.75±0.025^{c}	57.50	6.52±0.05^{b}	65.20	4.12±0.04^{e}	41.20	5.34±0.05^{d}	53.40
10	75	6.10±0.02^{a}	4.80±0.01^{c}	48.00	6.44±0.06^{b}	64.40	3.62±0.08^{d}	36.20	4.73±0.01^{c}	47.30

Notes: Encapsulation efficiency (EE, %) reflects the post-treatment survival rate of the probiotic cells. Within each column, means followed by distinct lowercase superscripts are significantly different according to the statistical analysis ($P < 0.05$).

3-3- Probiotic Resilience under Simulated Gastrointestinal Stress

The viability profiles of both non-encapsulated and micro-entrapped *L. acidophilus* La-05 during consecutive exposure to simulated gastric fluid (SGF) and simulated intestinal fluid (SIF) are summarized in Tables 2 and 3. The data reveal that although a general reduction in microbial counts occurred across all groups, the encapsulation process significantly bolstered the recovery rate relative to the non-incorporated cells ($p<0.05$). As evidenced in Table 2, 120-min- incubation in SGF (pH 2.0) triggered a severe population collapse in the free-cell group, underlining their vulnerability to highly acidic conditions. Conversely, matrices composed of LMP/β-glucan demonstrated superior shielding efficacy compared to alginate-based systems. This phenomenon is largely linked to the inherent acid-tolerant nature of pectin ,which maintains its structural stability at low pH, thereby effectively insulating the probiotic cargo from the gastric environment (Tarifa et al., 2021). The integration of AHS gum further fortified the barrier properties of the delivery systems. The most robust survival after the 2-hour gastric challenge was observed in the LMP/β-glucan/AHS formulation (EE = 85.6%). This suggests that AHS successfully minimized the internal porosity of the microcapsules, consequently impeding the influx of hydrogen ions toward the core (Hu et al., 2021). Following the gastric transit, the microbial persistence was monitored in SIF (Table 3). While probiotic counts continued to diminish under the influence of bile salts and physiological pH levels ,the microencapsulated cells maintained a significantly higher density than the free counterparts. Although alginate-based carriers are prone to rapid disintegration in the small intestine due to sensitivity toward neutral environments and chelating agents, the inclusion of AHS gum remarkably delayed this dissolution by enhancing the density and thickness of the capsule shell. The LMP/β-glucan/AHS composite again exhibited the highest performance, retaining 79.8% viability after 120 min. These results underscore the cooperative interaction between LMP and AHS gum in generating a durable protective framework that ensures the delivery of a therapeutic dose of probiotics to the lower intestinal tract.

Table 2. In vitro persistence and encapsulation efficiency (EE) of free and micro-entrapped *L. acidophilus* La-05 (log CFU/g) during sequential exposure to simulated gastric fluid (SGF).

Time (min)	free cells (Log CFU.g^{-1})	Simulated gastric fluid							
		Sodium alginate-based encapsulated cells				LMP/β-glucan -based encapsulated cells			
		without AHS (Log CFU.g^{-1})	EE (%)	with AHS (Log CFU.g^{-1})	EE (%)	without AHS (Log CFU.g^{-1})	EE (%)	with AHS (Log CFU.g^{-1})	EE (%)
0	10.00±0.03^{aA}	8.85±0.06^{dA}	-	9.42±0.04^{cA}	-	8.60±0.02^{cA}	-	9.12±0.06^{bA}	-
30	9.71±0.01^{aB}	8.63±0.02^{eB}	97.50	9.15±0.02^{dB}	97.10	8.39±0.04^{cA}	97.50	8.99±0.03^{bB}	98.60
60	9.32±0.01^{aC}	7.53±0.02^{dC}	85.10	8.64±0.02^{dC}	91.70	7.95±0.03^{cB}	92.50	8.70±0.06^{bC}	95.40
90	7.98±0.03^{aD}	5.04±0.02^{dD}	57.00	7.90±0.02^{eD}	83.80	6.02±0.03^{cC}	70.00	8.55±0.04^{bD}	93.70
120	7.50±0.01^{aE}	3.62±0.03^{dE}	40.90	6.80±0.09^{eF}	72.10	4.86±0.07^{cD}	56.60	7.80±0.05^{bE}	85.60

Notes: EE (%) represents the survival ratio relative to the baseline population (Time 0). Statistically significant disparities ($p < 0.05$) are denoted by distinct uppercase superscripts (A–E) for column-wise comparisons and lowercase superscripts (a–e) for row-wise comparisons.

Table 3. Comparative viability and encapsulation efficiency (EE) of *L. acidophilus* La-05 in free form versus LMP/β-glucan and alginate/maltodextrin-based microcapsules (with or without AHS gum supplementation) during simulated intestinal fluid (SIF) challenge.

Time (min)	free cells (Log CFU.g^{-1})	Simulated intestinal fluid (150min)							
		Sodium alginate-based encapsulated cells				LMP/β-glucan -based encapsulated cells			
		without AHS (Log CFU.g^{-1})	EE (%)	with AHS (Log CFU.g^{-1})	EE (%)	without AHS (Log CFU.g^{-1})	EE (%)	with AHS (Log CFU.g^{-1})	EE (%)
0	10.00±0.05^{aA}	8.85±0.03^{dA}	-	9.42±0.05^{cA}	-	8.60±0.04^{cA}	-	9.12±0.05^{bA}	-
30	9.77±0.03^{aB}	8.09±0.01^{eB}	91.40	9.32±0.09^{dB}	99.00	8.31±0.02^{cA}	96.70	8.90±0.00^{bB}	97.60
60	9.40±0.05^{aC}	6.19±0.03^{dC}	70.00	8.33±0.15^{dC}	88.50	7.48±0.03^{cB}	87.00	8.60±0.05^{bC}	94.30
90	8.01±0.07^{aD}	4.78±0.05^{dD}	54.10	7.27±0.02^{eD}	77.20	5.06±0.04^{cC}	58.80	7.92±0.05^{bD}	86.90
120	6.87±0.04^{aE}	2.48±0.01^{dE}	28.00	6.14±0.03^{eF}	65.20	3.81±0.03^{cD}	44.30	7.28±0.03^{bE}	79.80

Notes: EE (%) values reflect the survival rate relative to the initial population (Time 0). Statistically significant disparities ($p<0.05$) are denoted by distinct uppercase superscripts (A–E) for column-wise comparisons and lowercase superscripts (a–e) for row-wise comparisons.

CONCLUSIONS

In this study, the synergistic effects of wall materials (LMP/β-glucan and sodium alginate/maltodextrin) and *Alyssum homolocarpum* seed (AHS) gum on the physicochemical properties and survival of spray-dried *L. acidophilus* La-05 were investigated. The results demonstrated that the incorporation of AHS gum significantly improved the morphological integrity and optimized the particle size of the microcapsules. Importantly, the addition of AHS gum acted as a protective barrier, significantly enhancing the viability of probiotic cells during the harsh spray-drying process. Furthermore, the gastrointestinal simulation tests revealed that the LMP/β-glucan matrix combined with AHS gum provided superior protection compared to alginate-based formulations, maintaining the highest encapsulation efficiency (EE %) under both acidic gastric and bile-containing intestinal conditions. While alginate-based capsules showed acceptable initial recovery, the pectin-based system (LMP) demonstrated greater structural resilience in simulated physiological environments. Overall, this study suggests that the combination of LMP, β-glucan, and AHS gum is a highly efficient and promising delivery system for enhancing the stability and delivery of probiotics in functional food applications.

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بهبود زنده‌مانی پروبیوتیک: اثر صمغ قدومه شیرازی (*Alyssum homolocarpum*) به عنوان ماده انکپسولاسیون

اکرم السادات طباطبایی بفرویی، نگار حیدری، الهه نظری، و سعیده شجاعی علی آبادی

چکیده

به عنوان یک رویکرد کاربردی، ریزپوشانی به روش خشک‌کردن پاششی جهت محافظت از سویه‌های پروبیوتیک در برابر تیمارهای حرارتی شدید و تنش‌های فیزیولوژیک دستگاه گوارش به کار گرفته می‌شود. با این حال، تضمین بقای *لاکتوباسیلوس اسیدوفیلوس* La-5 در هنگام مواجهه با خشک‌کردن در مقیاس صنعتی و دمای ورودی بالا (۱۵۰-۱۴۰ درجه سلسیوس)، همچنان یک چالش جدی محسوب می‌شود. در مطالعه حاضر، اثرات محافظتی صمغ قدومه شیرازی (AHS) در ترکیب با دو سیستم دیواره‌ای شامل «پکتین با متوکسیل پایین/بتاگلوکان» و «آلژینات سدیم/مالتودکسترین» مورد بررسی قرار گرفت. فرآیند تولید ریزکپسول‌ها با استفاده از یک خشک‌کن پاششی با دمای ورودی ۱۵۰-۱۴۰ درجه سلسیوس و دمای هوای خروجی ۸۵-۸۰ درجه سلسیوس انجام شد. یافته‌ها نشان داد که صمغ قدومه شهری به طور معنی‌داری موجب ارتقای بازده ریزپوشانی (%EE) و تاب‌آوری سلول‌های پروبیوتیک شده است. به طور مشخص، نرخ بقای این سویه‌های ریزپوشانی شده در مواجهه با تنش حرارتی (۷۵-۵۵ درجه سلسیوس) طی بازه‌های زمانی ۱ تا ۱۰ دقیقه، از 4/68٪ به 6/82٪ بهبود یافت. علاوه بر این، تصاویر میکروسکوپ الکترونی روبشی (SEM) نشان داد که افزودن صمغ قدومه شهری منجر به ایجاد سطوح صاف‌تر در ریزکپسول‌ها و کاهش تخلخل آن‌ها (با میانگین قطر 22/18-3/5میکرومتر) شده است. در شرایط شبیه‌سازی شده گوارشی، فرمولاسیون حاوی پکتین/بتاگلوکان/قدومه شهری پس از ۱۲۰ دقیقه ماندگاری در مایع روده (SIF)، 79/8٪ از زیست‌پذیری خود را حفظ کرد که به طور معنی‌داری بالاتر از نمونه شاهد (0/28٪) بود. این نتایج تأیید می‌کند که صمغ قدومه شیرازی به عنوان یک ماده کمکی در دیواره (Co-wall material) عمل کرده و پایداری فیزیکی و مقاومت پروبیوتیک‌ها را در طول فرآوری و انبارداری بهبود می‌بخشد.