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Probiotic effervescent tablet for gut health improvement and immune support

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ABSTRACT

This study focuses on the development and formulation of an effervescent probiotic tablet designed to enhance gut health and immune support while addressing the limitations of traditional oral dosage forms, such as poor swallowing compliance and slow absorption. By utilising an effervescent couple of citric acid and sodium bicarbonate, the tablet rapidly disintegrates in water to release carbon dioxide, which facilitates the pre-solubilization of active ingredients and masks unpleasant tastes. The tablet includes a mix of spore-forming probiotics (*Bacillus subtilis* and *Bacillus coagulans*), which are more stable and survive better in the stomach than regular probiotics, along with *fructo-oligosaccharides* (FOS) as a prebiotic fibre. To further boost immune function and metabolic health, the tablet is fortified with vitamin C and folic acid. Manufactured under strictly controlled climatic conditions (25% RH) to prevent premature reactivity, the final product provides a highly bioavailable, palatable, and stable delivery system particularly beneficial for geriatric patients and health-conscious consumers seeking an efficient synbiotic supplement.

KEYWORDS

Effervescent tablet; Probiotics (*Bacillus coagulans*, *Bacillus subtilis*); Prebiotics (FOS – *fructo-oligosaccharides*); Gut-immune axis; Vitamin C; Folic acid; Synbiotic; Bioavailability

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Introduction

Probiotics are defined as "live microorganisms that, when administered in adequate amounts, confer a health benefit on the host" [1]. This definition, established by key international organizations, underscores two critical criteria: the viability of the microorganisms and the demonstration of a specific, measurable health benefit. Historically, the use of fermented foods containing beneficial microbes dates back centuries, but the modern scientific understanding of probiotics began in the early 20th century. Today, these beneficial microbes are central to the burgeoning field of gut health, with extensive research exploring their diverse mechanisms of action and therapeutic potential [2,3]. Beyond their widely known effects on digestive health, emerging evidence points to their profound influence on systemic functions, including a promising role in improving bone health through a complex biological relationship known as the gut-bone axis [4,5,6]. Probiotics derive their health benefits from a multifaceted interaction with the host's intestinal environment and immune system, rather than a single mechanism [7]. These mechanisms can be broadly categorized into gut lumen effects, intestinal barrier modulation, and systemic immune and metabolic modulation. Within the gut lumen, probiotic microorganisms engage in competitive exclusion, occupying ecological niches and physically preventing pathogenic bacteria from colonizing the intestinal wall [8]. This is a primary method for maintaining a healthy balance of the gut microbiota. Additionally, many probiotic strains, such as those from the Lactobacillus and Bifidobacterium genera, produce antimicrobial substances like bacteriocins, hydrogen peroxide, and organic acids (e.g., lactic acid) that can directly inhibit the growth of pathogens or kill them, creating a hostile environment for unwanted microbes [9]. Probiotics can also influence the availability of nutrients in the gut by fermenting indigestible dietary fibers and carbohydrates, producing beneficial byproducts, most notably short-chain fatty acids (SCFAs) like butyrate, acetate, and propionate. These SCFAs serve as a primary energy source for colonocytes (cells lining the colon) and are vital to sustaining gut health [10].

Prebiotics are a class of food ingredients that the human body cannot digest, yet they confer a significant health benefit on the host by selectively stimulating the growth and activity of beneficial microorganisms, such as "probiotics," already present in the gut [11,12]. Prebiotics are different from dietary fibers because they only affect one or a few types of gut bacteria, which makes the host healthier overall. Research has extensively documented their positive impact on various physiological processes [13]. For example, fructo-oligosaccharides (FOS) and inulin, which are prebiotics, have been shown to help different groups of people, from young men to postmenopausal women, absorb important minerals like calcium [14,15]. This enhanced mineral bioavailability is a key mechanism through which prebiotics support bone health [16]. Prebiotics not only help the body absorb minerals, but they also help strengthen the intestinal barrier, which is an important line of defense against pathogens and toxins [15]. They also affect the immune system by affecting the gut-associated lymphoid tissue [17]. However, the true potential of these compounds shines when combined with probiotics, the live microorganisms that provide direct health benefits when consumed [19]. This symbiotic relationship, known as a synbiotic, is founded on the principle that the prebiotic component acts as a selective "food source" that enhances the survival, growth, and metabolic activity of the probiotic strain, ensuring a more potent and durable effect within the gut ecosystem [20]. This synergy ensures that the beneficial effects of both components, such as improved nutrient absorption and immune function, are amplified. As such, the targeted use of both prebiotics and probiotics represents a powerful strategy for maintaining gastrointestinal health and promoting systemic well-being [19,20].

Oral medicine is the most common and preferred way for most people. However, this type of medicine may exhibit a delayed onset of action due to the longer time taken for absorption in the body [21]. When medicines are taken in liquid form, they can work faster and sometimes require lower doses. But there's a problem with liquid medicines, too. The active ingredients (the part of the medicine that does the work) are usually less stable in liquid form, meaning they can break down or lose effectiveness over time. This limitation can be addressed by administering the drug in liquid form. Yet, many active pharmaceutical ingredients (APIs) exhibit limited stability in liquid preparations.

Effervescent tablets have the property of dissolving rapidly in water just prior to consumption, forming a stable, immediate-use

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solution or dispersion. The effervescence arises from the reaction of tartaric and citric acids with carbonates or bicarbonates, liberating carbon dioxide (CO_2). The CO_2 facilitates quick tablet disintegration, enhances API dissolution, and improves palatability through taste masking. Compared to traditional oral dosage forms, effervescent tablets provide several advantages, including improved taste, a gentler effect on the stomach, and favorable marketing opportunities [22]. The manufacturing shall be done under controlled climatic conditions to avoid effervescent reactions. The packaging is done under 25% RH at 25°C. Effervescent tablets stand out with better bioavailability, a milder effect on the stomach, and greater ease for patients, especially older adults or those with swallowing difficulties who can't handle solid pills [23]. Since the active ingredients dissolve in water prior, they spread evenly in the stomach for quicker uptake. Additionally, they promote drinking more water, supporting hydration. All these qualities make effervescent tablets a flexible, appealing choice that boosts treatment effectiveness [24,25].

Advantages of effervescent tablets [26-28]:

1. Better taste because the fizzy drink can mask the unpleasant taste of some medicines.
2. Faster absorption occurs because the medicine is already dissolved in water, allowing it to start working more quickly.
3. The fizzing sensation of the drink makes it both attractive and enjoyable, appealing to those who prefer refreshing drinks.

Characteristics

Effervescent tablets are very similar to normal tablets. The powder used to make these tablets needs to have some specific qualities such as:

- The particles should be of the right shape and size.
- They should be evenly mixed.
- It should be easy to press into a solid tablet [16].

Moisture

One of the crucial things when making effervescent tablets is moisture. These tablets fizz when dropped into water [17]. That is because they contain:

- An acid (like citric acid)
- A base (like sodium bicarbonate or baking soda)

When these two mix with water, they create a chemical reaction that makes carbon dioxide gas (CO_2) [19]. That gas helps the tablet break up quickly in the water [29]. But here is the problem: if the materials used in the tablet already have a little moisture in them, this reaction can start too early, even inside the bottle. If that happens, the tablets can spoil or break down before use, and once the reaction starts, it gets faster and worse because it creates more water, which keeps it going [20]. To avoid such reactions, we use ingredients that are either very dry or that contain water in a stable, safe form (called a hydrate). But we still need a tiny bit of water in the mixture so the powder can stick together and form a solid tablet. That's why we carefully choose ingredients that have just the right amount of water.

Solubility

Another key thing is solubility, which means how well each ingredient dissolves in water [28].

- If an ingredient does not dissolve well, the tablet will not fizz properly.
- If some parts dissolve faster than others, the tablet may not break apart evenly and can leave chunks or residue in the glass.

So, it is best if all the ingredients dissolve quickly and at about the same speed [24].

Materials & Methodology

Materials

Active ingredients

Spore-forming probiotic strains (Bacillus subtilis and Bacillus coagulans), fructo-oligosaccharides (FOS-prebiotic fibre), Vitamin C (ascorbic acid), and folic acid (Vitamin B9).

Excipients

Citric acid (anhydrous, acid source), sodium bicarbonate (alkaline carbonate source), sucralose (sweetener), lemon flavour (flavouring agent), silicon dioxide (E551, anti-caking agent), and sodium benzoate (preservative).

Methodology

Procedure for preparation of effervescent tablets:

Procedure

Weighing of ingredients: Accurately weigh the required quantities of active pharmaceutical ingredients (Vitamin C and Folic Acid), flavouring agents, probiotic strains, and excipients, including Sodium benzoate, Sodium bicarbonate, Citric acid, Silicon dioxide, Sucralose, and Fructo-oligosaccharides (FOS), as per the specified formulation [28].

Blending of materials: Transfer all the weighed ingredients into a mortar and thoroughly mix using a pestle to achieve a uniform blend.

Tablet compression: Compress the blended powder into tablets using a manual tablet compressor. The tablets are prepared by the direct compression method.

Drying: Dry the compressed tablets in a hot-air oven at a controlled temperature of 35°C for 1 hour to remove residual moisture and enhance stability (Table 1 and Table 2).

Table 1. Formulation trial numbers 1 to 11.

Ingredient (mg)	T1	T2	T3	T4	T5	T6	T7	T8	T9	T10	T11 (Final)	Role / Significance
Citric Acid	1100	1100	1100	1100	1100	1100	900	800	700	700	700	Acid source
Sodium Bicarbonate	1200	1300	1400	1300	1300	1300	1300	1300	1300	1200	1200	Carbonate source

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Table 1 (continued)

Ingredient (mg)	T1	T2	T3	T4	T5	T6	T7	T8	T9	T10	T11 (Final)	Role / Significance
Flavor (Orange/ Lemon)	600	500	400	350	350	350	350	350	350	350	350	Flavoring agent
Sucralose	150	150	150	150	150	150	150	250	300	400	450	Sweetener
FOS (Fructo-oligosaccharides)	—	—	—	150	150	150	150	150	150	150	150	Prebiotic fibre
Probiotic Strains	100	100	100	100	100	100	100	100	100	100	100	Probiotic strains (B. subtilis, B. coagulans)
Silicon Dioxide	20	20	20	20	30	40	40	40	40	40	40	Anticaking agent
Vitamin C (Ascorbic Acid)	40	40	40	40	40	40	40	40	40	40	40	Vitamin source
Sodium Benzoate	20	20	20	20	20	20	20	20	20	20	20	Preservative
Folic Acid (Vitamin B)	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	Vitamin source
Total Weight (mg)	3230. 2	3230. 2	3230. 2	3230. 2	3240. 2	3250. 2	3050. 2	3050. 2	3000. 2	3000. 2	3050.2	—
Trial Outcome	Caught moisturefrom flavor	Still Caught moisture	Caught moisture; lower flavor needed	Caught moisure from FOS	Moisture retained; SiO_2 increased	Citric taste dominates flavor	Citric taste dominates; low sweetness	Citric tast high; sweetness low	Sweetnes still insufficient	Sweetness slightly low	Final formulation accepted	

Table 2. Final Formulation of Prebiotic-Probiotic Effervescent tablet.

Ingredients	Quantity (mg)	Significance
Sodium Bicarbonate	1200	Carbonate source
Citric Acid	700	Acid source
Sucralose	450	Sweetener
Flavor (Lemon)	350	Flavoring agent
Fructo-oligosaccharides	150	Prebiotic fibre
Strains (*B. subtilis* & *B. coagulans*)	100	Probiotic strains
Ascorbic acid (Vitamin C)	40	Vitamin source
Silicon dioxide	40	Anticaking agent
Sodium benzoate	20	Preservative
Folic acid (Vitamin B)	0.2	Vitamin source

Determination of pH of effervescent tablets

One effervescent tablet was placed in a 250 mL beaker, where it was allowed to dissolve completely. Once the reading on the pH meter stabilized, the pH value of the solution was recorded accurately. To ensure accuracy and reliability of results, the measurement was repeated using three separate effervescent tablets, each dissolved under the same experimental conditions. The mean pH value was calculated from the three replicate readings obtained, and the standard deviation (SD) was determined to evaluate the variability

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among the measurements. The final results were reported in the format: pH = (mean ± SD) [11,23].

Uniformity of thickness

Thirty tablets were randomly selected from the batch. 10 of these were assayed individually, and standard deviation (s) and relative standard deviation (RSD) were calculated. The batch passed the test if 9 of the 10 tablets were within the acceptable limit of deviation [13].

Disintegration/dissolution time

To determine the disintegration time of the tablet, a single tablet was placed in a beaker containing 200 mL of purified water at 20°C ± 1°C. The effervescence time was recorded as complete whenever a clear solution without particles was obtained. The mean of three measurements for each formulation was reported [14].

Viable count of probiotics after preparation

To determine the viable count of probiotics after preparation, a serial dilution and plating procedure is followed to ensure the resulting colonies are countable and representative. Initially, one probiotic tablet is thoroughly dissolved in 200 ml of distilled water to create the primary suspension. This mixture is then diluted serially. From each of the last three dilutions, a 1 ml aliquot is extracted and transferred into sterile Petri dishes, and molten plate-count agar is then poured on the plates and incubated at 37°C. After incubation, a count is taken, and an average viability is calculated [5].

Microbial limit test for prebiotic and probiotic effervescence tablets

To perform the microbiological analysis, a single probiotic tablet was dissolved in 200 ml of distilled water, and pentagonal streaking was performed on various selective and differential media to screen for specific pathogens: MacConkey's agar for *Escherichia coli*, mannitol salt agar for *Staphylococcus aureus*, cetrimide agar for *Pseudomonas aeruginosa*, and xylose lysine deoxycholate (XLD) agar for Salmonella and Shigella species. All inoculated plates were then incubated at 37°C for 24 to 48 hours to observe for characteristic colony morphology, such as pink lactose-fermenting colonies on MacConkey or yellow colonies on Mannitol Salt Agar, which would indicate the presence of these opportunistic pathogens [8].

Results and Discussion

Using the direct compression method the prebiotic and probiotic effervescent tablets were prepared successfully, the tablet appeared uniformly circular and pale white/off white in colour (consistent with the addition of vitamin C and flavorings). The manual compression at low pressure successfully produced tablets with sufficient structural integrity. The probiotic effervescent tablet is an off-white, uniform disk with clean edges and high structural integrity, showing no physical defects. Its granular surface reflects a well-blended mixture, ensuring the probiotic spores are evenly distributed throughout the effervescent base (Figure 1).

Figure 1. Visual appearance of the effervescent tablet showing physical integrity and uniform circular shape.

Determination of pH of effervescence tablets

Figure 2. Measurement and determination of the dissolved effervescent tablet solution pH using a digital pH meter (pH = 6.00).

Tablet was dissolved in water, the pH of the dissolved tablet solution was found to be 6.0, which is slightly acidic. The final pH of 6.00 suggests that the acid and base components were well-balanced and the solution in a range that is gentle on the gastrointestinal tract. From a formulation perspective, a pH of 6.00 is generally associated with a pleasant, mild taste that is neither too sour (low pH) nor soapy/bitter (high pH), which is suitable for oral administration. For stability and viability probiotic strains often prefer slightly acidic environments. A pH of 6.00 is near-neutral but leans acidic, which helps maintain the integrity of the microorganisms while they are in the liquid state before consumption (Figure 2).

Uniformity of thickness

Table 3. Observation: Thickness of effervescent tablet.

Tablet	Thickness in mm
1	4.5
2	4.6
3	4.5
4	4.4
5	4.5
6	4.4
7	4.5
8	4.5
9	4.5

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Table 3 (continued)

Tablet	Thickness in mm
10	4.6

Average Thickness = (4.5 + 4.6 + 4.5 + 4.4 + 4.5 + 4.4 + 4.5 + 4.5 + 4.5 + 4.6)/10 = 4.50 mm

Standard deviation = 0.067 mm Relative standard deviation = 1.48%

The average thickness of the tablets was 4.50 mm, with a standard deviation (s) of 0.067 mm and a relative standard deviation (RSD) of 1.48%. which indicates that the batch was well within the acceptable limit of 5% deviation set by the USP <905> and the British Pharmacopeia Appendix XII G (Table 3 & Figure 3).

Figure 3. Measurement of tablet thickness using a vernier caliper to check dimensional uniformity.

Disintegration Time: When immersed in water, the tablet exhibited rapid effervescence, achieving complete disintegration in 40 ± 5 seconds the resulting mixture appeared as a white, translucent suspension. The observed disintegration time of 40 seconds indicates a highly efficient effervescent mechanism. Standard pharmacopeial requirements for effervescent tablets typically require disintegration within 5 minutes in water therefore, a 40-second is well within acceptable limits. This rapid breakdown ensures the quick release and dispersion of the probiotic cultures into the liquid medium, making it ready for immediate consumption (Figure 4).

Viable count of probiotics after preparation

After the tablets were formulated, a viable count was performed using serial dilution to determine the number of living probiotics (colony-forming units) per table 4.

Table 4. Observation.

Dilution	df	cfu/ml	Average
10^6	10^6	101	[(101+460+1800)/3] x 10^6 = 7.87 x 10^8
10 ^7	10 ^7	46	
10 ^8	10 ^8	18	

As tablet is diluted in 200 ml d/w and then 1 ml is taken for dilution, the viable count = 200 x 7.87 x 10^8 = 1.574 X 10^11

Figure 4. Evaluation of the disintegration/dissolution time of the effervescent tablet in water.

The viability of the probiotics was measured to assess the survival rate after the stresses of tablet manufacturing. The resulting viable count of 1.574 X 10^11 is exceptionally high. A final count in the 10^11 range indicates that the formulation parameters and excipients used were highly protective of the microbial culture, and the probiotics successfully survived the tableting process, when got nutritional content (Media) showed rapid growth (Figure 5).

Figure 5. Enumeration of viable probiotic bacterial colonies on Plate Count Agar (PCA) across serial dilutions (10-6, 10-7, and 10-8).

Microbial limit test for prebiotic and probiotic effervescence tablets

To ensure the safety formulated probiotic effervescent tablets (1 day old and 3 months old) were subjected to microbial limit testing, the objective was to confirm the absence of specified objectionable microorganisms. There were no visible colonies or characteristic growth on the plates of MacConkey agar, Cetrimide agar, or xylose-lysine deoxycholate agar, the absence of growth on these media

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indicates that the sample was free from Escherichia coli, Pseudomonas aeruginosa, Salmonella spp., and Shigella spp. microbial growth was observed on the Salt Mannitol agar plate only. To verify the identity of colonies from SMA plates, gram staining and microscopic examination were performed, the isolated colonies from the SMA plates appeared as distinct purple-stained under oil immersion (100X) and the morphology was consistently identified as gram-positive rods (bacilli). No gram-positive cocci in clusters were observed during the microscopic survey, which confirmed the absence of Staphylococcus aureus. The experimental results confirmed that the probiotic effervescent tablet met the required microbiological safety standards which proved that the manufacturing process and the raw materials used were free from enteric pathogens and opportunistic contaminants (Figure 6 to Figure 10).

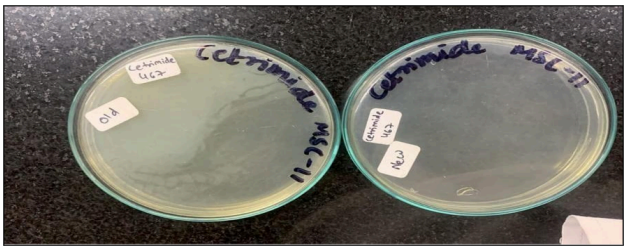

Figure 6. Microbial limit test on Cetrimide agar demonstrating the absence of Pseudomonas aeruginosa.

Figure 7. Microbial limit test on MacConkey's agar demonstrating the absence of Escherichia Coli.

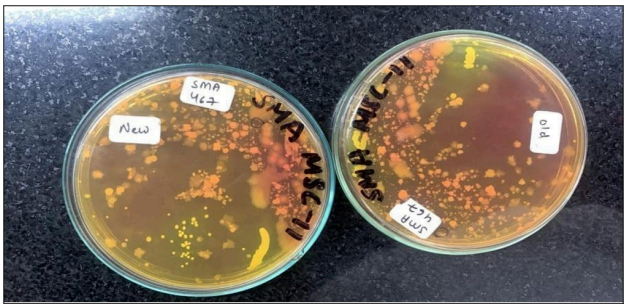

Figure 8. Microbial limit test on Mannitol Salt Agar (SMA) and differential agar showing absence of Staphylococcus Aureus.

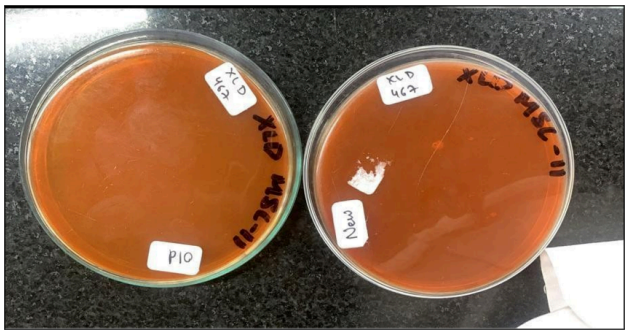

Figure 9. Microbial limit test on Xylose Lysine Deoxycholate agar (XLD) showing absence of Salmonella spps.

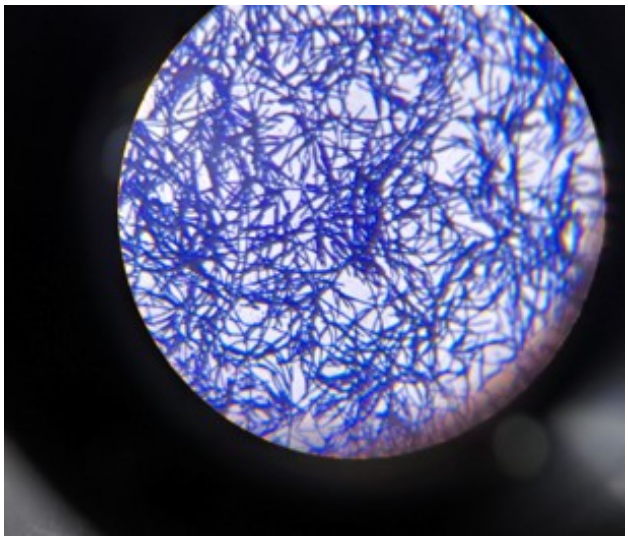

Figure 10. Microscopic Image of Colony Picked from SMA agar Plate Confirming the absence of S.aureus., Microscopic observation Indicates the gram nature and morphology of Probiotic Culture.

Conclusion

This study is about the formulation and assessment of a synbiotic effervescent tablet aimed at enhancing the delivery of particular probiotic strains and prebiotic fiber. The tablets were manufactured using a direct compression technique to minimize mechanical stress on the Probiotic culture, resulting in a robust dosage form that achieved rapid disintegration and maintained a physiologically compatible pH. Physical assessments confirmed high precision in tablet thickness and excellent uniformity of the active ingredients across all samples. Microbiological safety was verified through extensive limit testing, which confirmed the total absence of enteric pathogens.

Disclosure Statement

No potential conflict of interest was reported by the author.

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