

# EFFECTS OF CHIA SEED ON FERMENTATION AND ANTIOXIDANT ACTIVITY OF GOAT YOGURT

– Research paper –

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**Abstract:** Goat milk is considered a valuable alternative to cow milk due to its high digestibility, rich nutrient profile, and lower allergenicity. However, its consumer acceptability is limited by the presence of specific flavor compounds, such as caprylic, decanoic, and hexanoic acids. This study aimed to enhance the flavor and nutritional value of fermented goat milk through the incorporation of a Direct-Vat-Set (DVS) starter culture and chia seeds. Through a single-factor experimental design, we investigated the effects of fermentation time, chia seed particle size, inoculation level of the DVS starter culture, and concentration of chia seed powder on the antioxidant activity (measured by DPPH radical scavenging ability and ferric ion reducing antioxidant power), acidity, and pH of the product. The results demonstrated that fermentation at 42 °C with the addition of 1% 60-mesh chia seed powder and an inoculation level of 0.004% DVS culture significantly enhanced the product flavor, shortened the fermentation time, and boosted the antioxidant activity of the goat yogurt.

**Keywords:** chia seed; goat milk; yogurt; antioxidant properties

## INTRODUCTION

Goat milk is scientifically recognized as a valuable alternative to cow milk due to its lower allergenicity and higher digestibility (García et al., 2014). It is rich in essential vitamins, minerals, and contains higher concentrations of several essential amino acids than cow milk (Zervas and Tsiplakou, 2013). Furthermore, goat milk contains numerous bioactive growth factors, reinforcing its nutritional profile (Sharifi et al., 2017). Despite these advantages, its goaty smell, characterized by compounds such as caprylic and decanoic acids, limits widespread consumer acceptance (Ranadheera et al., 2018).

To enhance the functional properties of goat milk, the incorporation of probiotics has been widely explored. Probiotic-fermented dairy products offer some health benefits, including improved gut health and enhanced immune function (Hou et al., 2019). The fermentation process also generates bioactive peptides, increasing the bioactivity of the milk (Enrica P and Simona C, 2016). However, the addition of probiotics alone does not effectively mask the characteristic "goaty" flavor, which remains a significant barrier to market expansion.

The use of flavor-modifying functional ingredients, such as fruits or seeds, presents a promising strategy to address this issue. Chia seeds (*Salvia hispanica* L.) are rich in vitamins (da Silva et al., 2017), minerals (Ding et al., 2017), polyphenols (Rahman et al., 2017), and fatty acids (Kaur et al., 2014). They exhibit significant antioxidant capacity, including a high DPPH radical scavenging ability (Aparna et al., 2023), and can inhibit lipid and protein oxidation (Ullah et al., 2016). Critically for dairy applications, studies have shown that adding chia seeds to yogurt can increase dietary fiber content and improve the viability of probiotics during storage (Ribes et al., 2021; Pop et al., 2015; Espinaco, et al., 2025). While chia seeds have been approved for use in various food categories and have been shown to boost the nutritional value of products like bread (Belén et al., 2021) and beef burgers (Antonini et al., 2020), their potential in combination with probiotics in goat milk yogurt remains underexplored.

Building upon previous findings from our laboratory on the antioxidant properties of fermented goat milk, this study aims to systematically investigate the effects of combining probiotics and chia seeds. We focus specifically on how chia seeds influence the fermentation characteristics (e.g., fermentation time, acidity), antioxidant activity, and sensory properties of goat

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milk yogurt.  
The outcomes of this research are expected to provide a scientific basis for developing a novel,

functional goat dairy product with enhanced acceptability and health benefits.

## MATERIALS AND METHODS

### Preparation of Chia Seed Powder

Chia seeds were obtained from Golden Valley Grain and Oil Food Co. (China). The chia seeds were dried in a blast drying oven at 60 °C for 30 minutes and cooled to room temperature. The dried seeds were then ground using a laboratory pulverizer. The resulting powder was sieved through a series of standard sieves (20, 40, 60, 80, and 100 mesh) to obtain fractions of different particle sizes, which were stored for subsequent use.

### Preparation of goat yogurt with chia seed

Preparation of Chia-fortified Fermented Goat Milk  
Goat milk powder was provided by Shaanxi Yatai Dairy Co., Ltd. (China). A DVS starter culture was obtained from the School of Food Science and Engineering, Shaanxi University of Science and Technology.

Reconstituted goat milk (12.5%, w/w) was pasteurized at 90 °C for 15 min and then cooled to the fermentation temperature. A specified amount of chia seed powder was added to the cooled milk. The mixture was then inoculated with 0.005% (w/v) of a DVS starter culture and gently stirred to ensure even distribution. Finally, it was incubated in a water bath at 42 °C for 4.5 hours to obtain the final product.

### Preparation of whey samples

The fermented yogurt sample was adjusted to pH 3.4-3.6 using 1 mol/L HCl. The sample was then centrifuged at 8000 × g for 15 min. The resulting supernatant was collected, and its pH was readjusted to approximately 8.3 using 1 mol/L NaOH. This supernatant was centrifuged again under the same conditions. The final supernatant was collected as the whey sample for subsequent analysis.

### Optimization of Fermentation Conditions

Based on preliminary experiments, the basic fermentation conditions were set as follows: chia seed powder addition 1%, particle size 60 mesh, DVS inoculum size 0.005%, fermentation time 4 h, and temperature 42 °C. A single-factor experimental design was employed to investigate the individual effects of fermentation time (1, 2, 3, 4, 5, 6, 7 h), chia seed powder concentration (0.5, 1, 1.5, 2, 2.5%), chia seed particle size (20, 40, 60, 80, 100 mesh), and DVS inoculum size (0.002, 0.004, 0.006, 0.008, 0.010, 0.012%) on the physicochemical properties of the yogurt. The evaluation indicators included pH, acidity, sensory score, DPPH radical scavenging rate, and ferric ion reducing antioxidant power (FRAP).

### pH and Acidity Determination

The pH of the yogurt samples was measured at room temperature using a calibrated pHs-3C meter (Shanghai INESA Instruments Co., Ltd). Acidity was determined as titratable acidity according to the method described by Shu et al. (2017).

### DPPH Radical Scavenging Activity

The DPPH radical scavenging activity was determined according to Zeng et al. (2012). Briefly, the sample was mixed with a freshly prepared ethanolic DPPH solution. The mixture was incubated in the dark for 20 min, and the absorbance was measured at 517 nm. Ethanol was used as the control. The DPPH scavenging rate was calculated using the following formula:

$$\text{DPPH Scavenging Rate (\%)} = \left(1 - \frac{A_i}{A_0}\right) \times 100$$

where  $A_0$  is the absorbance of the control, and  $A_i$  is the absorbance of the sample.

### Ferric Reducing Antioxidant Power (FRAP)

The FRAP was determined as described by Damien et al. (2004). A 2.5 mL aliquot of the sample was mixed with 2.5 mL of 0.2 mol/L phosphate buffer (pH 6.6), 2.5 mL of 1% potassium ferricyanide solution, and 2.5 mL of 10% trichloroacetic acid solution. The mixture was vortexed and centrifuged at 3000 × g for 10 min. Then, 2.5 mL of the supernatant was mixed with 2.5 mL of distilled water and 0.5 mL of 0.1% ferric chloride solution. The absorbance was measured at 700 nm after thorough mixing. A higher absorbance indicates a stronger reducing power.

### Sensory Analysis

Sensory evaluation was performed by a trained panel (n=10) according to the criteria established by Chen et al. (2016). Panelists scored the products based on surface appearance, texture, taste, odor, and whey separation. The specific scoring criteria are presented in Table 1.

### Statistical Analysis

All experiments were conducted in triplicate. Data are presented as mean ± standard deviation (SD). Data processing and graphing were performed using Origin 2022 and Microsoft Excel 2010. One-way analysis of variance (ANOVA) was carried out using IBM SPSS Statistics 25 to determine significant differences between means, with a significance level defined at  $p < 0.05$ .

## RESULTS

### Effect of time on the fermentation of goat milk with chia seed

Reconstituted goat milk fortified with 1% 60-mesh chia seed powder and 0.005% of a DVS starter culture was fermented at 42°C for different durations (1-7 h) to investigate the impact of chia seeds on the acidity and pH development. As shown in Figure 1, the pH of the yogurt progressively decreased while the acidity increased with fermentation time in both the control and chia seed-fortified groups. Notably, the sample with chia seeds reached an acidity of 78 °T by 4 h, meeting the benchmark of  $\geq 70$  °T for the fermentation endpoint. Consequently, 4 hours was selected as the optimal fermentation time for subsequent experiments.

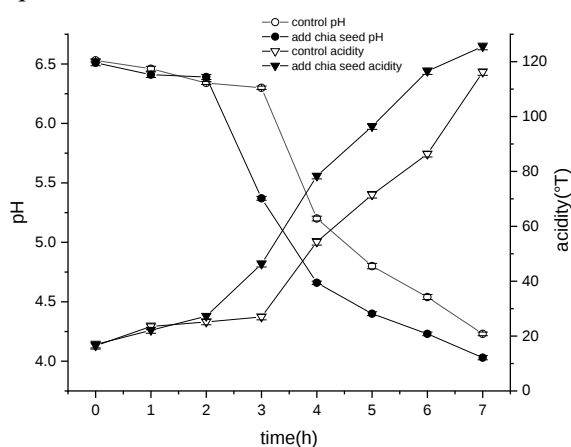


Figure 1. Effect of fermentation time on chia seed goat yogurt

### Effect of DVS inoculum size on fermentation and antioxidant activity of goat yogurt with chia seed

The inoculum size of the DVS starter culture significantly influenced both the antioxidant activity and fermentation characteristics of the chia seed goat yogurt. As shown in Figure 2a, the DPPH radical scavenging rate and sensory score varied significantly with inoculum size, both reaching their optimal values (62.38% and 80.3, respectively) at 0.006%. Furthermore, the highest FRAP values were observed at inoculum sizes of 0.006% and 0.008%, with no significant difference between them. This enhancement in antioxidant activity is likely attributed to the ability of lactic acid bacteria to produce antioxidant peptides.

Regarding the fermentation parameters (Fig. 2b), an inoculum size of 0.004% resulted in a pH close to 4.5 and an acidity greater than 70 °T, indicating a completed fermentation. As the inoculum size increased beyond this point, the acidity continued to rise while the pH decreases stabilized, showing no significant differences between the 0.006%-0.008% and 0.01%-0.012% groups.

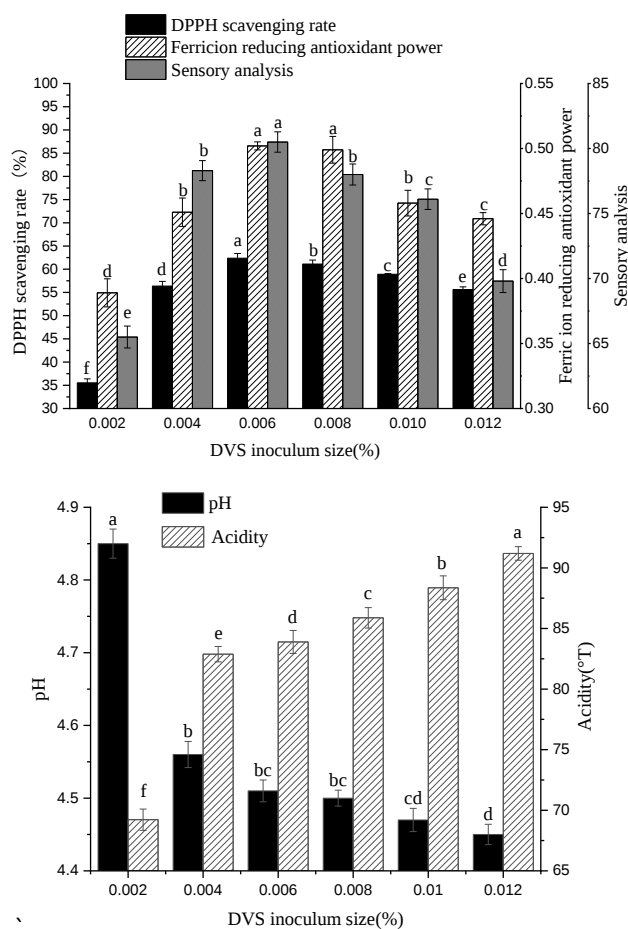


Figure 2. Effect of DVS inoculum size on DPPH scavenging rate, FRAP sensory analysis (a), pH, acidity (b) of goat yogurt with chia seed

### Effect of particle size on fermentation and antioxidant activity of goat yogurt with chia seed

Chia seed particle size exerted a more pronounced effect on the sensory properties than on the antioxidant activity or fermentation parameters of the yogurt. As illustrated in Fig. 3a, the sensory score increased significantly with finer particle sizes, reaching a maximum of 83.3 at 100 mesh. This improvement is attributed to the smoother and more uniform texture resulting from smaller particles. In contrast, particle size had a limited impact on antioxidant activity. Although a statistically significant increasing trend in DPPH scavenging was observed with larger particles, the absolute difference was minor and likely of little practical relevance. No significant effect on FRAP was detected. Furthermore, as shown in Fig. 3b, particle size did not significantly alter the fermentation endpoint, with the pH remaining stable between 4.5 and 4.6 across all treatments, and acidity showing only minor variations.

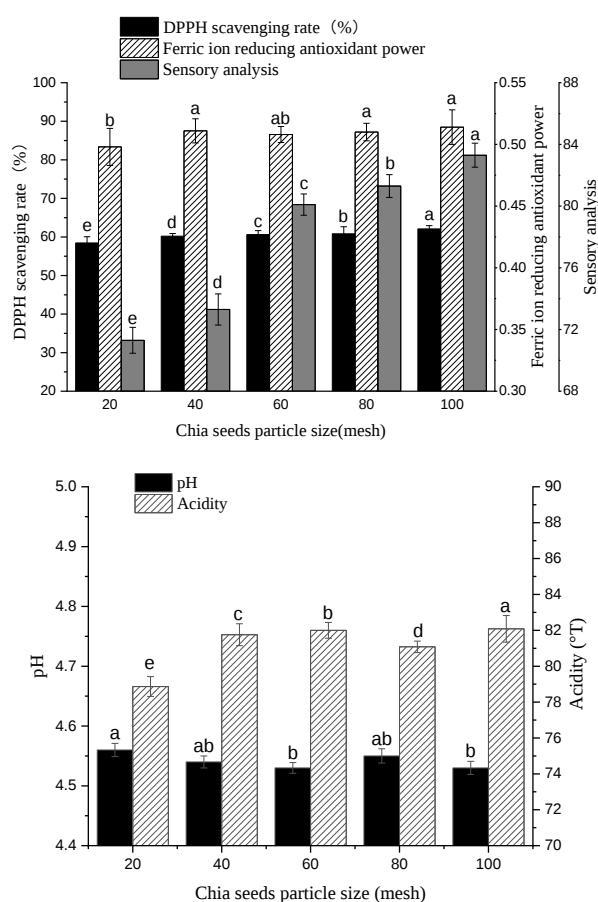


Figure 3. Effect of particle sizes on DPPH scavenging rate, FRAP, sensory analysis (a), pH and acidity (b) of goat yogurt with chia seed

### Effect of chia seeds addition on fermentation and antioxidant activity of goat yogurt with chia seed

The concentration of chia seeds significantly influenced the product's properties, revealing a critical trade-off between antioxidant activity and sensory quality. The DPPH scavenging rate and FRAP value increased with chia seed addition, peaking at 81.56% and 0.717, respectively, at the highest concentration of 2.5%. However, this maximum antioxidant capacity coincided with the

lowest sensory score (64.5), attributed to the high polysaccharide content in chia seeds. These polysaccharides absorb water and swell, leading to excessive viscosity and elasticity that adversely affect the texture and overall acceptability. In Fig. 4(b), as the concentration of chia seeds increased, the pH of the fermented goat milk initially decreased and then increased, while the acidity showed an inverse trend. The pH reached its minimum value of 4.52 at a 1% addition level, coinciding with the peak acidity of 84.06 °T.

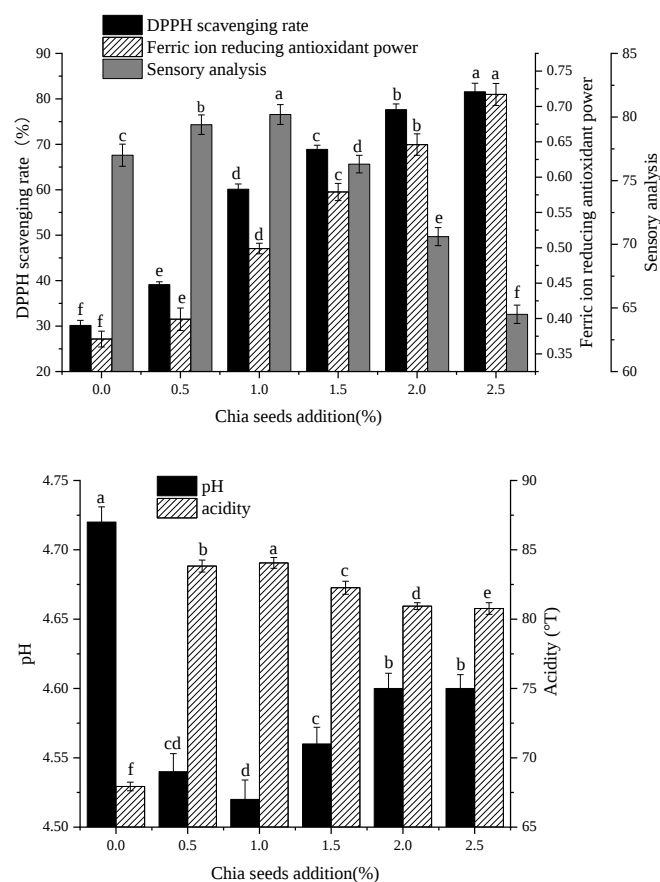


Figure 4. Effect of chia seeds addition on DPPH scavenging rate, FRAP, sensory analysis (a), pH and acidity (b) of goat yogurt with chia seed

## DISCUSSION

This study demonstrates that the incorporation of chia seeds significantly enhances the functionality of goat milk yogurt, primarily through three mechanisms: accelerating fermentation, boosting antioxidant activity, and modulating sensory properties. The observed reduction in fermentation time can be attributed to the rich polyphenolic compounds in chia seeds, such as chlorogenic acid and caffeic acid (Pellegrini et al., 2018; Munoz et al., 2013), which are known to promote the growth of lactic acid bacteria (LAB) (Pandey, K R et al., 2015). This prebiotic-like effect is consistent with

the findings of Kwon et al. (2019), who reported that chia seed extracts accelerated LAB growth and fermentation kinetics, thereby validating the results of our experimental study.

The enhanced antioxidant activity, reflected in the increased DPPH scavenging rate and FRAP values, results from a synergistic combination of factors. Firstly, chia seeds themselves are a direct source of antioxidants (Agarwal et al., 2023). Secondly, as supported by the work of Azadikhah et al. (2025), the metabolic activity of LAB during fermentation generates bioactive peptides with potent antioxidant

properties. Therefore, the combination of moderate DVS inoculum size and chia seed addition creates an environment conducive to the production of these beneficial compounds.

Regarding sensory characteristics, both particle size and concentration of chia seeds were determining factors. Finer particle sizes (e.g., 100 mesh) yielded a superior sensory score, which is directly linked to the smoother texture and absence of grittiness. Conversely, the relationship between chia seed concentration and sensory acceptance was non-linear. While low to moderate levels were acceptable, higher concentrations led to a sharp decline in scores. This is due to the high polysaccharide content in chia seeds, which upon hydration, excessively increases the product's viscosity and elasticity, creating a gelatinous texture that is perceived negatively. This trade-off between functionality and palatability is a common

challenge in functional food development, as similarly observed with high levels of flaxseed (Mousavi, M et al., 2018) and chia seeds in other yogurt formulations (Kowaleski et al., 2020).

Overall, the key innovation of this research lies in identifying the optimal balance between these competing factors. While maximum antioxidant activity was achieved at 2.5% chia seed addition, the optimal overall quality—considering enhanced fermentation, significant antioxidant boost, and high sensory acceptability—was determined to be at a lower concentration (approximately 1%) with a fine particle size. This work confirms that chia seeds are a highly effective multi-functional ingredient for goat yogurt, but their application must be carefully optimized to navigate the intrinsic compromise between health benefit enhancement and sensory appeal.

## CONCLUSIONS

This study systematically investigated the effects of key processing parameters on the quality of chia seed-fortified goat yogurt. The results demonstrated that chia seed incorporation significantly shortened the fermentation time and enhanced the antioxidant activity (DPPH and FRAP) of the product, while its particle size and concentration critically influenced the sensory acceptability. An optimal formulation was identified that effectively balances these functional and sensory properties: using a DVS inoculum size of 0.004%, a chia seed concentration of 1.0% with a particle size of 60 mesh, and

fermenting at 42 °C for 4.5 hours. This combination successfully leverages the prebiotic effect of chia seeds to accelerate fermentation and the synergistic action between seed-derived antioxidants and LAB-generated bioactive peptides to boost antioxidant capacity, without compromising the product's texture and overall palatability. Therefore, this work confirms the feasibility of using chia seeds as a multi-functional ingredient in goat yogurt and provides a theoretical foundation for developing novel, value-added functional dairy products.

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