Journal of Advances in Biology & Biotechnology

Volume 27, Issue 11, Page 1408-1417, 2024; Article no.JABB.127625
ISSN: 2394-1081

Optimization of Soaking Conditions to Enhance Protein and Reduce Phytate Content in Foxtail Millet (*Setaria italica*)

Aiswarya Velekat Santhosh [a++*], Malashree L [a#], Harinivenugopal [b†], Rajunaik B [c†], Manjunatha H [d‡] and Ramachandra B [a^]

[a] Department of Dairy Microbiology, Dairy Science College, Hebbal, Bengaluru, KVAFSU, India.
[b] Department of Dairy Technology, Dairy Science College, Hebbal, Bengaluru, KVAFSU, India.
[c] Department of Dairy Engineering, Dairy Science College, Hebbal, Bengaluru, KVAFSU, India.
[d] Dairy Science College, Mahagaon Cross, Kalaburagi, KVAFSU, India.

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Article Information

DOI: https://doi.org/10.9734/jabb/2024/v27i111728

Open Peer Review History:
This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: https://www.sdiarticle5.com/review-history/127625

Original Research Article

Received: 05/11/2024
Accepted: 27/11/2024
Published: 29/11/2024

ABSTRACT

Aims: The study aimed to evaluate the effect of millet-to-water ratio of soaking and time-temperature combination used for soaking on the protein and phytate content of foxtail millet.

[++]M.Tech Scholar;
[#]Assistant Professor;
[†]Associate Professor;
[‡]Dean;
[^]Professor and Head;
**Corresponding author: E-mail: aisuvelekat@gmail.com;*

Cite as: *Santhosh, Aiswarya Velekat, Malashree L, Harinivenugopal, Rajunaik B, Manjunatha H, and Ramachandra B. 2024. "Optimization of Soaking Conditions to Enhance Protein and Reduce Phytate Content in Foxtail Millet (Setaria Italica)". Journal of Advances in Biology & Biotechnology 27 (11):1408-17. https://doi.org/10.9734/jabb/2024/v27i111728.*

Study Design: This was an experimental, laboratory-based study.
Place and Duration of Study: The study was conducted at the Department of Dairy Microbiology, Dairy Science College, Hebbal, Bengaluru, Karnataka, India, between January 2024 and October 2024.
Methodology: Foxtail millet (*Setaria italica*) was soaked in potable water at varying millet-to-water ratios (1:1, 1:1.5, 1:2) under ambient conditions (27 ± 1°C) for 6 hours and at different time-temperature combinations (20°C, 25°C, 30°C for 6, 12, 18, and 24 hours). The protein content was determined using the micro-Kjeldahl method, while phytate content was estimated via titration with ferric chloride. Statistical analysis was performed using ANOVA, with a critical difference (CD) at a 5% significance level used to determine significance among treatments.
Results: The 1:2 millet-to-water ratio showed the highest protein content (10.68%) and the lowest phytate content (3.07 mol/kg) after 6 hours of soaking under ambient conditions, with these differences being statistically significant (*P =.05*). Increasing the soaking temperature and duration further enhanced protein content and reduced phytate levels, with the most pronounced effect observed at 30°C for 24 hours. This condition yielded the highest protein content (10.99%) and the lowest phytate content (1.79 mol/kg), both significantly improved (*P =.05*) compared to unsoaked millet (10.50% protein, 4.96 mol/kg phytate).
Conclusion: The study concluded that soaking foxtail millet at a 1:2 millet-to-water ratio and at 30°C for 24 hours are effective strategies to enhance its nutritional quality. These findings provide a scientific basis for optimizing soaking conditions to improve the protein content and reduce antinutritional factors in millet.

Keywords: Foxtail millet; soaking; millet-to-water ratio; protein content; phytate reduction; time-temperature combinations; antinutritional factors; nutrient bioavailability.

ABBREVIATIONS

FAOSTAT : Food and Agriculture Organization Corporate Statistical Database;
FSSAI : Food Safety and Standards Authority of India.

1. INTRODUCTION

Millets are nutrient-dense grains, rich in proteins, dietary fiber, and essential micronutrients, making them a crucial component of healthy diets and a sustainable option for global food security. Despite their high nutritional value, the presence of antinutritional factors like phytates limits the bioavailability of these nutrients. Soaking has emerged as an effective method to enhance the nutritional quality of grains by reducing antinutrients and improving digestibility (Samuel and Peerkhan, 2020).

According to FAOSTAT (2022), global millet production was estimated at 30.86 million tonnes, with India contributing the largest share at 11.85 million tonnes. Currently, over half of millet production is used for purposes other than staple food consumption. However, millets hold significant potential in combating malnutrition and overcoming agricultural challenges in areas with harsh climates. Their resilience to shallow, less fertile soils and tolerance for a broad pH range (4.5-8.0) make them well-suited to various environments (Kumar et al., 2018).

Millets require very little water over their relatively short growing period and are known for their drought resistance. When stored as whole grains, they can be preserved for long durations (Devi et al., 2014). This makes them crucial for ensuring food security, as they can flourish even in challenging agro-climatic conditions (Balli et al., 2023).

Soaking improves protein digestibility by breaking down antinutritional factors that bind to proteins and inhibit enzyme activity. Hasan et al. (2019) reported that soaking increases protein digestibility by eliminating these antinutrients, which impede protein and starch digestion. Similarly, Samuel and Peerkhan (2020) observed enhanced starch digestibility post-soaking, attributed to the removal of inhibitors affecting amylase enzymes. These findings underscore the role of soaking in improving overall nutrient bioavailability.

The millet-to-water ratio during soaking significantly influences nutrient retention and antinutrient reduction. Thorat et al. (2017) demonstrated that soaking pearl millet at a 1:2

ratio effectively reduced phytate content, balancing phytate removal with minimal nutrient leaching. Sow (2023) similarly observed reductions in phytate levels in millet soaked at a 1:3 ratio, supporting the preference for moderate water ratios. However, excessive water ratios, as shown by Abioye et al. (2022) in finger millet, can lead to protein loss, highlighting the importance of optimizing soaking conditions.

Temperature and duration of soaking are critical factors influencing the enhancement of the nutritional profile of grains. Soaking at higher temperatures for extended durations has been shown to effectively improve protein content while significantly reducing antinutritional factors. Sharma et al. (2018) demonstrated this effect in foxtail millet, highlighting the dual benefit of increased protein levels and reduced phytates. Similar findings have been reported for other nutrient-rich grains, including amaranth, quinoa, and buckwheat, where prolonged soaking resulted in improved nutrient bioavailability and reduced antinutritional factors (Thakur et al., 2021). These observations underscore the importance of optimizing soaking conditions to maximize the nutritional potential of grains.

Recent commercial applications of millets further emphasize their versatility and functionality. For instance, Yadagouda and Ravindra (2022) developed a foxtail millet composite flour that was germinated, milled, and combined with groundnut and green gram. The composite mix, inoculated with *Lactobacillus acidophilus*, showed excellent viability (8.51 $\log_{10}$ cfu/ml) and retained stability for a month at room temperature without yeast or mold growth. Similarly, Rubavathi et al. (2022) studied probiotic foxtail millet laddus containing microencapsulated and lyophilized *Lactobacillus acidophilus*, observing better viability of microencapsulated probiotics compared to lyophilized forms after two months of storage at room (25°C) and refrigeration (4°C) temperatures. Additionally, Geetha et al. (2021) demonstrated the stability of *Lacticaseibacillus rhamnosus GG* in a millet-based health mix stored at room temperature for three months without a decline in viable counts.

Santhosh et al. (2024) demonstrated that foxtail millet stands out among various millet species for its superior nutritional profile, particularly its high protein and crude fiber content. Their study revealed that foxtail millet contains the highest protein content at 10.50%, followed by proso millet at 9.94%, while finger millet has the lowest at 7.24%. In terms of crude fiber, foxtail millet also ranks highly, with a content of 3.21%, second only to finger millet, which has 3.61%. Due to its relatively higher levels of protein and fiber, foxtail millet was selected for this study.

This research investigates the impact of different millet-to-water ratios and soaking time-temperature conditions on the protein and phytate content of foxtail millet. Building on previous studies and leveraging insights from commercial applications, the goal is to determine the optimal soaking parameters to improve the nutritional quality of foxtail millet, thereby supporting its potential use in health-oriented and functional food products.

2. MATERIALS AND METHODS

2.1 Materials

Foxtail Millet (*Setaria italica)* was procured from a reputed local market in Bengaluru, Karnataka, India.

2.2 Methods and Methodology

The foxtail millet was soaked in potable water at varying millet-to-water ratios of 1:1, 1:1.5, and 1:2 at ambient temperature (27 ± 1°C) for a duration of 6 hours. The primary aim was to determine the millet-to-water ratio that resulted in the highest protein content and the lowest phytate content. After assessing the protein and phytate levels at each ratio, the optimal millet-to-water ratio was selected based on these nutritional parameters.

Following the identification of the optimal ratio, the millets were soaked at this selected ratio and subjected to different temperature conditions (20°C, 25°C, and 30°C) for soaking durations of 6, 12, 18, and 24 hours. After soaking, excess water was drained off, and the protein and phytate content of the millet samples were analyzed.

The optimum time-temperature combination for soaking was determined by evaluating the reduction in phytate content and the increase in protein content. The combination that exhibited the lowest phytate levels and the highest protein content was selected as the ideal condition for further processing and optimization of the millet's nutritional profile.

2.3 Estimation of Protein Content

The protein content was determined using the micro-Kjeldahl technique, which involves several precise steps. For the digestion step, 0.5 g of a thoroughly mixed sample was weighed and transferred into a Kjeldahl digestion tube. To this, 0.1 ml of 5% copper sulfate solution, 1.5 g of potassium sulfate, and 5–10 boiling aids were added. Next, 15 ml of concentrated sulfuric acid was introduced along the tube's wall, and the contents were gently mixed. The mixture was digested until it became clear and free of any residual matter, indicating the complete breakdown of organic material. The digest was then cooled to 25 ± 2°C in preparation for distillation.

Then an indicator solution was prepared by dissolving 0.1 g of methyl red in 95% (v/v) ethanol and diluting it to 50 ml with ethanol. Similarly, 0.5 g of bromocresol green was dissolved in 95% (v/v) ethanol and diluted to 250 ml with ethanol. The indicator solution was then prepared by mixing one part of the methyl red solution with five parts of the bromocresol green solution or by combining both solutions entirely. A boric acid solution was prepared by dissolving 40 g of boric acid in hot water, cooling the solution, and diluting it to 1 L. To this solution, 3 ml of the indicator solution was added to enable the detection of ammonia during the process.

Then the cooled digest was diluted with 100 ml of distilled water and transferred to an automatic distillation unit. During the distillation process, 50% sodium hydroxide was added to liberate ammonia from the digest. The released ammonia was captured in 25 ml of the prepared 4% boric acid solution containing the indicator, forming a complex that enabled subsequent quantification.

Finally, 25 ml of the distillate was titrated with 0.02N hydrochloric acid until the solution changed to a pink color, signaling the endpoint. To account for procedural accuracy, blank test was simultaneously carried out by following the procedure as described above taking all the reagents and replacing the sample with 1 ml water and about 0.17 g of sucrose. The protein content was then calculated using the titration values of the sample and the blank, ensuring precise quantification of the protein present in the sample. The total nitrogen and crude protein percentages were then calculated using the formula provided below (FSSAI 03.017:2023).

$$W_n = \frac{1.4007 \times (V_S - V_B) \times N}{W}$$

$$\text{Crude protein (\%)} = W_n \times 6.25$$

Where,

W_n = Nitrogen content of sample, expressed as % by mass
V_S = Volume in ml of the standard HCl used for sample
V_B = Volume in ml of the standard HCl used for blank test
N = Normality of standard HCl
W = Mass of test portion in g

2.4 Estimation of Phytate Content

About 4.0 g of grounded sample was steeped in 100 ml of 2% hydrochloric acid for 3h before being filtered using Whatman No. 1 filter paper. 25ml of this filtrate and 5ml of 0.3% ammonium thiocyanate solution as an indicator were taken and 53.5ml of distilled water was added after setting it to proper acidity and titrated against standard ferric chloride solution (0.00195g of iron per ml) until brownish yellow colour appeared which persisted for 5 minutes (Sharma et al., 2016).

$$\text{Phytate content (mol/kg)} = \frac{T \times 564.11}{M}$$

Where,

T = Titre value
M = Molar mass of phytate in kg

2.5 Statistical Analysis

The data was analysed using R software [R. version 4.1.2 copyright] for statistical computing. Data on the response variables was collected for three replications of the trails and the ANOVA tables was prepared to analyse the data. The critical difference was calculated (*P=.05*), where the F value was significant, and used to identify whether significant differences existed and indicated in the table using superscripts.

$$\text{Critical difference (CD)} = \frac{\sqrt{2} \times MSS(E) \times t\alpha}{r}$$

Where,

MSS (E) = Mean Sum of squares of the error
r = number of replications
tα = table t value of the α level of significance

3. RESULTS AND DISCUSSION

3.1 Effect of Millet-to-water Ratio of Soaking on the Protein and Phytate Content of Foxtail Millet

The impact of varying millet-to-water ratios on the protein and phytate content of foxtail millet during soaking was evaluated under ambient conditions (27 ± 1°C) for 6 hours. The results demonstrated that both protein content and phytate levels were significantly affected by the soaking ratios, with the 1:2 ratio showing the most pronounced improvements in nutritional quality. The findings are presented in Table 1, which provides a detailed comparison across the treatments.

3.1.1 Protein content

The protein content of foxtail millet increased progressively with higher millet-to-water ratios during soaking. The control (unsoaked millet) exhibited the lowest protein content of 10.50%, serving as a baseline. Soaking the millet at a 1:1 ratio slightly increased the protein content to 10.55%, which was statistically significant from the control (*P = .05*). Further increasing the ratio to 1:1.5 resulted in a protein content of 10.62%, while the highest protein content of 10.68% was observed at the 1:2 ratio. The critical difference (CD) at a 5% significance level for protein content was 0.016, confirming the statistical significance of the observed differences.

These findings align with existing research. Abioye et al. (2022) reported a decline in protein content from 9.47% to 9.20% in finger millet soaked at a 1:5 ratio, attributing the loss to nutrient leaching. Conversely, de Lima et al. (2014) highlighted that a 1:1.5 ratio effectively minimized protein loss while achieving substantial reductions in phytate content in soybeans. They also found that a low ratio of soybeans to soaking medium, such as 1:1.5, reduced protein leaching, underscoring the importance of ratio optimization. The results of this study reaffirm the importance of using optimal soaking ratios to enhance protein content without compromising nutrient integrity.

3.1.2 Phytate content

Phytate, a significant antinutritional factor, decreased progressively with increasing millet-to-water ratios. The control sample exhibited the highest phytate content of 4.96 mol/kg, significantly higher than all soaked samples. Soaking at a 1:1 ratio reduced the phytate content to 4.06 mol/kg, while soaking at a 1:1.5 ratio further reduced it to 3.58 mol/kg. The lowest phytate content of 3.07 mol/kg was achieved at the 1:2 ratio, representing the most significant reduction among all treatments. The CD at a 5% significance level for phytate content was 0.067, confirming the statistical significance of these reductions.

The observed reductions in phytate levels are consistent with previous studies. Sow (2023) reported a 5.4% reduction in phytate content in millet soaked at a 1:3 ratio. Similarly, Thorat et al. (2017) documented a reduction in phytate content in pearl millet from 868 mg/100g in raw form to 548 mg/100g at a 1:2 ratio and 545 mg/100g at a 1:5 ratio after 12 hours of soaking. Notably, Thorat et al. found no significant difference between the 1:2 and 1:5 ratios but highlighted increased nutrient leaching at higher water ratios, reinforcing the preference for the 1:2 ratio as an optimal balance between phytate reduction and nutrient preservation. de Lima et al. (2014) similarly observed reduced phytate loss when using a 1:1.5 ratio for soybean soaking, further validating the approach of using optimal ratios for reducing antinutrient factors while maintaining nutritional quality.

Table 1. Effect of millet-to-water ratio of soaking on the protein and phytate content of foxtail millet

Soaking parameters (Foxtail millet: water ratio)	Protein content (%)	Phytate content (mol/kg)
Control	10.50^{d}	4.96^{a}
1:1	10.55^{c}	4.06^{b}
1:1.5	10.62^{b}	3.58^{c}
1:2	10.68^{a}	3.07^{d}
CD (*P=.05*)	0.016	0.067

Note: CD = Critical Difference and control is unsoaked foxtail millet and all the values are average of three trials (n=3) and same superscript indicate non-significance while different, indicate statistically significant difference at P=.05

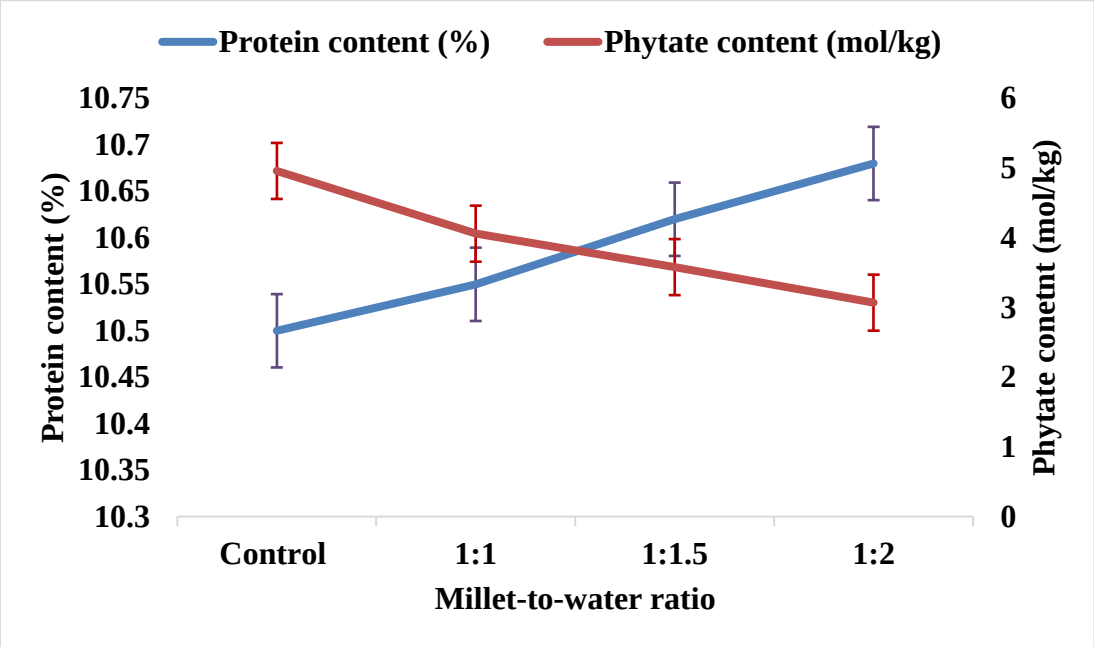

Fig. 1. Effect of millet-to-water ratio of soaking on the protein and phytate content of foxtail millet

The findings demonstrate that soaking foxtail millet at a 1:2 millet-to-water ratio significantly enhances protein content while effectively reducing phytate levels, making this ratio the most suitable for optimizing the nutritional profile of foxtail millet.

3.2 Effect of Time-temperature Combinations of Soaking on the Protein and Phytate Content of Foxtail Millet

The impact of various time-temperature combinations during soaking on the protein and phytate content of foxtail millet was evaluated, with the unsoaked millet serving as the control. The initial protein content of foxtail millet was 10.50%, while the phytate content was 4.96 mol/kg, providing a baseline for comparison. Soaking led to significant changes in both protein and phytate levels, with the combination of 30°C for 24 hours emerging as the most effective.

3.2.1 Protein content

The protein content of foxtail millet showed a significant increase with longer soaking times and higher temperatures. Initially, the protein content of unsoaked foxtail millet was 10.50%. After soaking at 20°C for 6 hours, the protein content increased to 10.58%. After 24 hours of soaking at 20°C, the protein content further increased to 10.84%, which was significantly higher than the control ($P = 0.05$). Similarly, at 25°C, the protein content rose from 10.59% after 6 hours to 10.88% at 24 hours. The most notable increase occurred at 30°C, where the protein content increased from 10.67% after 6 hours to 10.99% after 24 hours, representing the highest protein content observed among all treatments. The critical difference (CD) at a 5% significance level for protein content was 0.159, confirming the statistical significance of these results.

These findings are consistent with the study by Sharma et al. (2018), which reported an increase in protein content during high-pressure soaking of foxtail millet. In their study, protein content increased from 13.55% to 13.70% after soaking for 30 minutes at 40°C, and when the soaking time was extended to 120 minutes, protein content increased from 13.54% to 13.70%. Similarly, Handa et al. (2017) found that soaking horsegram flour for 18 hours increased the protein content from 22.60% at 0 hours to 28.77%. These studies highlight the beneficial effects of prolonged soaking times and higher temperatures on protein enhancement.

Additionally, Shashego (2019) observed a decrease in protein content when soybeans were soaked at higher temperatures (40°C), with a reduction from 50.76% to 49.16%. This study further supports the idea that soaking at

moderate temperatures and times, such as those used in this study, is beneficial for increasing protein content in millets. In this study, the most effective condition was 30°C for 24 hours, which resulted in the highest protein content of 10.99%.

3.2.2 Phytate Content

Phytate content, an important antinutrient factor, decreased significantly with increased soaking time and temperature. The initial phytate content

Table 2. Effect of millet-to-water ratio of soaking on the protein and phytate content of foxtail millet

Soaking parameters		Protein content (%)	Phytate content (mol/kg)
Temperature (°C)	Time (hours)		
Control	0	10.50^{d}	4.96^{a}
20	6	10.58^{cd}	3.76^{ab}
	12	10.64^{bcd}	3.07^{bcd}
	18	10.79^{abc}	2.73^{bcd}
	24	10.84^{abc}	2.56^{bcd}
25	6	10.59^{cd}	3.42^{bc}
	12	10.72^{abcd}	2.82^{bcd}
	18	10.80^{abc}	2.47^{bcd}
	24	10.88^{ab}	2.30^{cd}
30	6	10.67^{bcd}	2.73^{bcd}
	12	10.75^{abcd}	2.22^{cd}
	18	10.84^{abc}	1.96^{d}
	24	10.99^{a}	1.79^{d}
CD (*P=.05*)		0.159	0.753

Note: CD = Critical Difference and control is unsoaked foxtail millet and all the values are average of three trials (n=3) and same superscript indicate non-significance while different, indicate statistically significant difference at P=.05

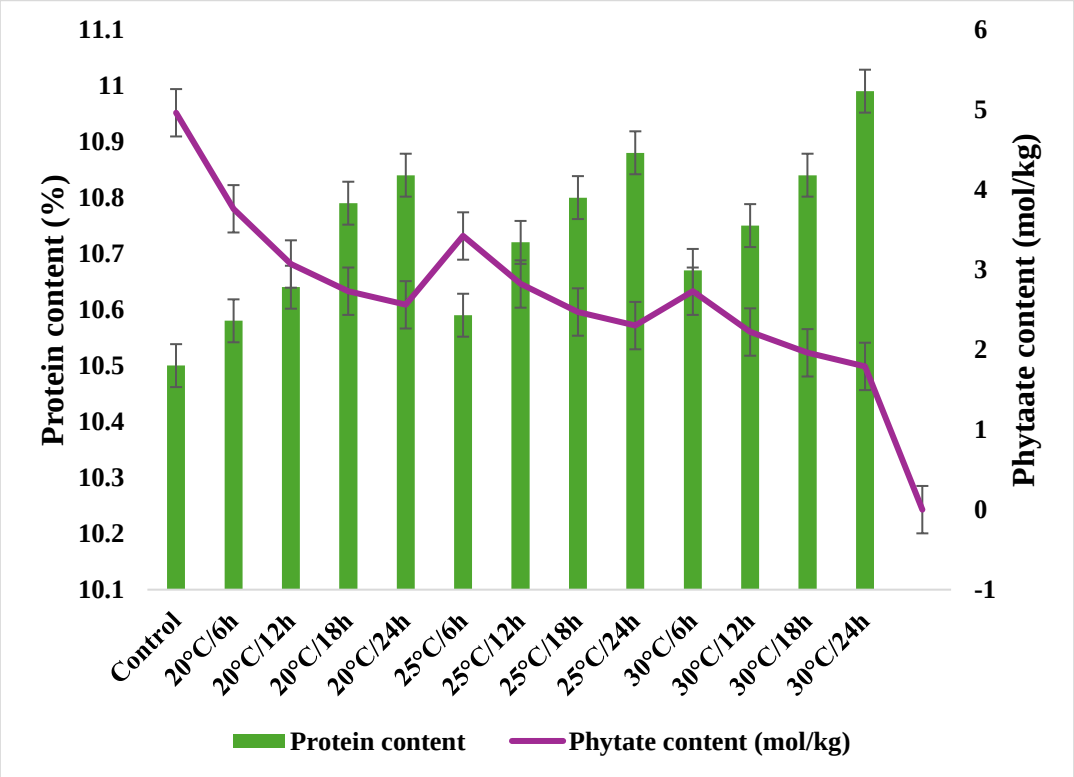

Fig. 2. Effect of time-temperature combinations of soaking on the protein and phytate content of foxtail millet

of unsoaked foxtail millet was 4.96 mol/kg. After soaking at 20°C for 6 hours, phytate content reduced to 3.76 mol/kg, and after 24 hours, it decreased further to 2.56 mol/kg. At 25°C, phytate content dropped from 3.42 mol/kg after 6 hours to 2.30 mol/kg after 24 hours. The most significant reduction occurred at 30°C, where phytate content decreased from 2.73 mol/kg after 6 hours to 1.79 mol/kg after 24 hours. This represents the lowest phytate content among all treatments, confirming that higher temperatures lead to greater reductions in phytate levels. The critical difference (CD) for phytate content at a 5% significance level was 0.753, validating the statistical significance of these reductions.

These results are consistent with the findings of Sharma et al. (2018), who demonstrated similar reductions in antinutritional factors, including phytates, with increased soaking time and temperature. Additionally, Thakur et al. (2021) observed decreases in phytate content in amaranth, quinoa, and buckwheat after soaking, further supporting the beneficial effects of soaking on phytate reduction. Shigihalli et al. (2018) also reported a significant reduction in phytate concentration in finger millet during soaking, with levels decreasing from 250 mg/100g at 12 hours to 221 mg/100g at 48 hours. The researchers attributed the decrease to the direct hydrolysis of phytate by seed phytase, reinforcing the idea that soaking induces enzymatic activity that breaks down phytates.

The current study's findings reinforce the effectiveness of soaking foxtail millet at 30°C for 24 hours, which not only reduces phytate content but also preserves and enhances protein levels. Thus, the optimal soaking condition of 30°C for 24 hours emerged as the most effective combination for increasing protein content (10.99%) and minimizing phytate levels (1.79 mol/kg) in foxtail millet. This combination significantly improved the nutritional profile of foxtail millet compared to the unsoaked samples, making it a suitable candidate for further processing into health-focused and functional food products.

4. CONCLUSION

The study demonstrated that both the millet-to-water ratio and time-temperature combinations significantly influence the protein and phytate content of foxtail millet during soaking. Among the tested millet-to-water ratios, the 1:2 ratio emerged as the optimal condition, resulting in the highest protein content (10.68%) and the lowest phytate content (3.07 mol/kg) after 6 hours of soaking under ambient conditions (27 ± 1°C). This ratio provided a balance between nutrient preservation and antinutritional factor reduction, aligning with findings in existing literature, which highlight the importance of maintaining a controlled water ratio to minimize nutrient leaching.

The time-temperature combinations of soaking further enhanced the nutritional profile of foxtail millet. Higher soaking temperatures and longer durations consistently improved protein content while reducing phytate levels. Soaking at 30°C for 24 hours proved to be the most effective, yielding the highest protein content of 10.99% and the lowest phytate level of 1.79 mol/kg. This combination demonstrated a significant improvement compared to the unsoaked sample (10.50% protein, 4.96 mol/kg phytate). These findings confirm the role of controlled soaking conditions in enhancing the nutritional quality of millet.

The results align with previous research emphasizing the synergistic effects of optimized soaking conditions on protein enhancement and phytate reduction in cereals and legumes. While higher temperatures and prolonged soaking periods can enhance nutritional outcomes, careful optimization is necessary to avoid potential nutrient loss or quality degradation.

In conclusion, soaking foxtail millet at a 1:2 millet-to-water ratio and at 30°C for 24 hours under time-temperature combinations, are recommended as effective strategies for improving the nutritional quality of foxtail millet. These findings provide valuable insights for food processing industries and health-conscious consumers seeking to maximize the nutritional benefits of millet-based diets.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

ACKNOWLEDGEMENTS

The authors would like to express their gratitude to the entire faculty, both teaching and non-

teaching at the Department of Dairy Microbiology, Dairy Chemistry, Dairy Technology and Dairy Engineering in Dairy Science College, Bengaluru, KVAFSU for their unwavering support in carrying out this research.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

Samuel, K. S., & Peerkhan, N. (2020). Pearl millet protein bar: nutritional, organoleptic, textural characterization, and in-vitro protein and starch digestibility. Journal of Food Science and Technology, 57(9), 3467-3473.

Food and Agriculture Organization. (2022). FAOSTAT: Food and Agricultural data. Available:https://www.fao.org/faostat/en/#home

Kumar, A., Tomer, V., Kaur, A., Kumar, V., & Gupta, K. (2018). Millets: a solution to agrarian and nutritional challenges. Agriculture & food security, 7(1), 1-15.

Devi, P. B., Vijayabharathi, R., Sathyabama, S., Malleshi, N. G., & Priyadarisini, V. B. (2014). Health benefits of finger millet (Eleusine coracana L.) polyphenols and dietary fiber: a review. Journal of food science and technology, 51, 1021-1040.

Balli, D., Cecchi, L., Pieraccini, G., Venturi, M., Galli, V., Reggio, M., et al. (2023). Millet Fermented by Different Combinations of Yeasts and Lactobacilli: Effects on Phenolic Composition, Starch, Mineral Content and Prebiotic Activity. Foods, 12(4), 748.

Hasan, M., Maheshwari, C., Garg, N. K., & Kumar, M. (2019). Millets: nutri-cereals. Biotech Express, 6, 18-21.

Thorat, A., Rawool, S., Ajbani, S., Salvi, A., Chheda, S., Ghugre, P. S., et al. (2017). Effect of processing on total iron and zinc, ionizable iron, extractable zinc, phytate and phytate: mineral ratios in pearl millet. Adv. Food Sci. Eng, 1, 129-143.

Sow, M. S. (2023). Effect of Soaking in Neutral and Acidic Media on The Phytate Content of Souna Millet. JN food sci tech, 4(1), 1-4.

Abioye, V. F., Babarinde, G. O., Ogunlakin, G. O., Adejuyitan, J. A., Olatunde, S. J., & Abioye, A. O. (2022). Varietal and processing influence on nutritional and phytochemical properties of finger millet: A review. Heliyon, 8(12).

Sharma, N., Goyal, S. K., Alam, T., Fatma, S., Chaoruangrit, A., & Niranjan, K. (2018). Effect of high pressure soaking on water absorption, gelatinization, and biochemical properties of germinated and non-germinated foxtail millet grains. Journal of Cereal Science, 83, 162-170.

Thakur, P., Kumar, K., Ahmed, N., Chauhan, D., Rizvi, Q. U. E. H., Jan, S., et al. (2021). Effect of soaking and germination treatments on nutritional, anti-nutritional, and bioactive properties of amaranth (*Amaranthus hypochondriacus* L.), quinoa (Chenopodium quinoa L.), and buckwheat (*Fagopyrum esculentum* L.). Current Research in Food Science, 4, 917-925.

Yadagouda, V. R., & Ravindra, U. (2022). Formulation and Evaluation of Foxtail Millet Composite Mix for Probiotication. Mysore Journal of Agricultural Sciences, 56(1), 417-425.

Rubavathi, S., Ganesan, A., & Harini, T. (2022). Formulation and Validation of Probioticated Foxtail Millet Laddu as a Source of Antioxidant for Biological System. Research Square, Preprint Version 1, 2-24.

Geetha, K., Banu, K. S. P., & Ramasamy, D. P. (2021). Evaluation of functional and sensory characteristics of probiotic incorporated millet based instant health mix. The Pharma Innovation Journal, 10(1), 289-292.

Santhosh, A. V., Ramachandra, B., Manjunatha, H., Malashree, L., Rajunaik, B., & Harinivenugopal. (2024). Exploring Nutritional Diversity in Millets: A Comparative Study on Protein and Crude Fiber Content. Journal of Scientific Research and Reports, 30(11), 458-465.

Food Safety and Standards Authority of India. (2023). Manual of methods of analysis of foods. Cereal and Cereal Products. Ministry of Health and Family Welfare, GOI, New Delhi, India.

Sharma, S., Saxena, D. C., & Riar, C. S. (2016). Analysing the effect of germination on phenolics, dietary fibres, minerals and γ-amino butyric acid contents of barnyard millet (*Echinochloa frumentaceae*). Food Bioscience, 13, 60-68.

de Lima, F. S., Kurozawa, L. E., & Ida, E. I. (2014). The effects of soybean soaking on grain properties and isoflavones loss.

LWT-Food Science and Technology, 59(2), 1274-1282.

Handa, V., Kumar, V., Panghal, A., Suri, S., & Kaur, J. (2017). Effect of soaking and germination on physicochemical and functional attributes of horsegram flour. Journal of food science and technology, 54, 4229-4239.

Shashego, Z. (2019). Effects of soaking time and temperature on the nutritional content and sensory quality of soybean flour and milk. Global Journal of Science Frontier Research: D Agriculture and Veterinary, 1(3), 25-38.

Shigihalli, S., Ravindra, U., & Ravishankar, P. (2018). Effect of processing methods on phytic acid content in selected white finger millet varieties. International Journal of Current Microbiology and Applied Sciences, 7(2), 1829-1835.

Peer-review history:
The peer review history for this paper can be accessed here:
https://www.sdiarticle5.com/review-history/127625