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Impact of hydrothermal processing on the mineral, antinutritional, antioxidant and functional characteristics of proso millet (*Panicum miliaceum*)

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Abstract

This study explored the effects of several hydrothermal treatments such as steeping, blanching, steaming, roasting, and pressure cooking on proso millet grain. Both untreated and treated samples were analyzed for nutritional composition and mineral content. Anti-nutritional factors like tannin and phytic acid were also quantified illustrating 46.09% reduction in tannin levels and 31.94% in phytic acid levels after hydrothermal treatments. Antioxidant activity was assessed using various methods, such as 2,2-diphenyl-1-picrylhydrazyl (DPPH%), Total Phenolic Content (TPC), Total Flavonoid Content (TFC), and Ferric Reducing Antioxidant Power (FRAP) assays. The DPPH% and TFC content was enhanced 41.12% and 34.54%, respectively after the hydrothermal treatments. Additionally, the study evaluated the functional and pasting properties of the millet to assess its performance during processing and potential applications in food products. The findings highlight how hydrothermal treatments can optimize the nutritional profile and functional characteristics of proso millet, making it a valuable option for addressing dietary needs.

Keywords Hydrothermal, Millets, Antioxidant, Tannin, Pasting properties

Introduction

Millets are often referred to as "coarse grains" with current status as "nutri-cereals." Millets are anticipated to be "future crops" due to their resilience against a wide range of pests and diseases, as well as their ability to thrive in the challenging conditions of arid and semi-arid regions in Asia and Africa. Millets are categorized as small-seeded grains, with the most prevalent and significant varieties for consumption being sorghum (*Sorghum bicolor* L.), finger millet (*Eleusine carocana*), pearl millet (*Pennisetum glaucum*), kodo millet (*Paspalum scrobiculatum*), teff (*Eragrostis tef*), proso millet (*Panicum*

miliaceum), foxtail millet (*Setaria italica*), little millet (*Panicum sumatrense*), and fonio (*Digitaria exilis*) [7].

Proso millet (*Panicum miliaceum*) represents one of the earliest cultivated millet crops and serves as a staple food for a large portion of the population residing in arid and semi-arid tropical regions around the globe, including Asia, Africa, and parts of Europe [27]. Various studies on Proso millet have indicated its superior nutritional value compared to other primary cereals [39]. Proso millet boasts a rich content of protein, minerals, vitamins, carbohydrates, and lipids that align closely with the recommended proportions, surpassing other common cereals [19]. This particular millet variety stands out as a top source of phosphorus, sodium, iron, zinc, calcium, and potassium among cereals. Moreover, the amino acid profile of proso millet surpasses that of wheat, containing all essential amino acids while being deficient in lysine at 189 mg/g [19]. It also showcases a significant presence of

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phytonutrients like polyphenolic compounds, flavonoids, tannins, phytates, and saponins, which deliver antioxidative, anti-inflammatory, and anti-carcinogenic effects through diverse pathways. Despite its exceptional nutritional and functional attributes, proso millet has regrettably been undervalued and underutilized. Urbanization and modernization led to the decline of proso millet from dietary practices due to its coarse texture and the absence of ideal processing conditions, despite its commendable nutritional profile comprising dietary fiber, high-quality proteins, low glycemic index, diverse mineral content, and gluten-free protein composition [55]. Nonetheless, the presence of anti-nutritional elements poses challenges to the digestibility of proteins and starch, along with diminishing the absorption and availability of specific minerals.

Millet's necessitate processing prior to consumption for the elimination of indigestible components, elongation of shelf life, and enhancement of nutritional and sensory characteristics. Various hydrothermal treatments such as steeping, blanching, boiling, steaming, roasting, and pressure cooking are commonly used not only in scientific research but also as basic cooking methods in daily life. Each method introduces variations in heat, moisture, and time, influencing the millet's chemical composition and functional properties. Hydrothermal techniques change the structure and behavior of whole-grain flours and starches extracted from cereals, millets and pulses. When exposed to higher temperatures, water becomes more active and penetrates deeper, helping to break down components and trigger significant changes during processing [26]. These changes not only improve how the ingredients function in recipes but also make them easier to digest, expanding their usefulness in a variety of food products [40].

The utilization of these hydrothermal treatments has demonstrated a beneficial influence on the nutritional quality of millets. These methods disintegrate intricate carbohydrates, thereby rendering the nutrients in proso millet more accessible for absorption within the body. The hydrothermal treatment procedure for millets encompasses diverse techniques, all geared towards amplifying their nutritional value and functional attributes [8]. These treatments play a pivotal role in diminishing antinutrient compounds, ameliorating product quality through intercomponent interactions, and positioning millet products as functional foods based on biochemical analyses [1]. Moreover, hydrothermal processing has the potential to escalate bioavailability of mineral content, enhance the bioaccessibility of iron and zinc, and diminish total phenolic content, thereby ultimately enriching the comprehensive nutritional profile of millets [43]. Additionally, hydrothermal treatments can

lead to decreased rates of starch digestion, lowered glycemic impact, and improved structural attributes of millet grains, rendering them more suitable for consumption and storage purposes. Hence, the primary objective of the current investigation is to assess the influence of various hydrothermal treatments such as steeping, blanching, boiling, steaming, roasting, and pressure cooking on the nutritional, antinutritional, antioxidant and functional quality of proso millet.

Materials and methods

Sample preparation

The proso millet (*Panicum miliaceum*) sample was procured from local market of Prayagraj city. The millet grains underwent treatments like washing, air-drying and subsequent storage in a refrigerator before use. All the chemicals employed for chemical analysis and extraction were of analytical grade.

Hydrothermal treatments

Hydrothermal treatment is applied to cereal grains to maximize the yield of milled grain while minimizing breakage. This process also induces beneficial nutritional changes, such as the gelatinization of starch within the grains. Typically, hydrothermal treatment includes soaking the grains in water, steaming them, and then drying them to a specific moisture content suitable for milling. The samples were given selected hydrothermal treatment such as soaked proso millet (SOPM) where 100 g proso millet grains were steeped in a 1:3 water ratio at 30 °C for 16 h in a water bath (Metrex Scientific Instruments Ltd.) followed by draining in a colander; blanched proso millet (BLPM) in which the millet grains (100 g) were soaked in water for 30 min at 30 °C, followed by hot water blanching at 100 °C for 10 min; steamed proso millet (STPM) sample was prepared by soaking millet grains (100 g) in hot water (60 °C) for 2 h followed by steam-cooking for 10 min. Roasted samples (ROPM) were prepared by roasting in an open pan at 80 °C, kept for 10 min to cool; pressure cooked proso millet (PCPM) where proso millet grain was soaked in hot water at 60 °C for 2 h, after which it was sieved, pressure cooked for 20 min. All the treated samples were dried in a hot air oven dryer (Narang Scientific works Ltd.) at 60 °C till constant weight was achieved. The dried samples were ground and stored in an air tight container till further use. Raw sample, without any treatment was taken as control (RPM).

Nutritional composition and mineral content

The proso millet samples were analyzed for moisture, protein, fat, carbohydrate, ash, and crude fiber by methods specified in AOAC [2]. Estimation of minerals was done by Inductively coupled plasma-optical emission

spectrometry (ICP-OES) method. A standard solution of selected minerals (100 µg/mL) was purchased from Agilent Technologies (Standard ICM 101). The standard solution curve was prepared in 10, 50, 100, 150, and 200 µg/mL concentrations by diluting a standard solution (100 µg/L) with deionized water. Electronic grade hydrochloric acid (35%), nitric acid, and hydrogen peroxide were purchased from Merck (USA). Accurately weigh a sufficient amount of the thoroughly blended material in a tared silica dish. Firstly, heat over a low Bunsen flame to evaporate the organic ash. Place the dish in a muffle furnace at 420 °C until all of the carbon has stopped glowing. Take the dish out of the muffle, let it cool, add 1 or 2 mL of concentrated HNO₃, let it evaporate until it is completely dry, and then heat it in the muffle for an additional hour or so if it seems that not all of the carbon has been oxidized. Take the ash out of the furnace, let it cool, then weigh it if necessary. Using a pipette, carefully add 40–50 ml of diluted HCL (1 + 1) to the dish after covering it with a watch glass. After 30 min of heating over a water bath, remove the lid and rinse. To dehydrate silica, keep cooking for an additional 30 min. Filter through Whatman filter paper into a 100 ml volumetric flask. Use diluted HCl to wash the residue in the basin once or twice. The insoluble residues and used filter paper were transferred into the crucible and then repeated using the same procedure of drying ash as the tested samples. A small amount of water and 2 mL HCl (12 N) were added into the crucible and then each sample was diluted with 5 mL water. The sample was heated in water bath and filtered into 100 mL volumetric flask [24]. The sample solution was adjusted to exactly 100 mL in volume with water and analyzed for the determination of minerals by ICP-OES (Agilent Technologies, USA).

Antinutritional content

Tannin content

The tannin content was determined following the procedure outlined by [13], a commonly used method for assessing tannin levels in samples. This method relies on the color change observed in the presence of tannins, which is then measured using a spectrophotometer for quantification. Initially, a sample weighing 0.5 g was boiled with 75 ml of water for approximately 30 min. After centrifugation at 2000 rpm for 20 min, the supernatant was collected. Folin Denis reagent (Sigma-Aldrich) and sodium carbonate (CDH) were added to the sample extract, and the resulting solution was diluted to a total volume of 100 mL with water. After allowing the solution to stand for 30 min, its absorbance was measured at a wavelength of 700 nm. To create a calibration curve, a series of standard solutions of Tannic acid (ranging from

10 to 70 µg/mL) were prepared following the same procedure mentioned above. The total tannin content was then calculated and expressed in terms of milligrams of Tannic Acid Equivalents (TAE) per gram of extract.

Phytate content

The phytate content of the samples was analyzed following the method outlined by Sadasivam and Manickam [44]. Initially, 5 g of the samples were used for phytate extraction using 3% trichloroacetic acid (CDH), and the phytate was then precipitated by ferric chloride (CDH) into ferric salt. The iron content of the ferric salt was determined by dissolving the precipitate in hot nitric acid and diluting it accordingly. Additionally, 20 mL of potassium thiocyanate (Sigma-Aldrich) was added to develop the color of the diluted extract, and the absorbance of the resulting brick-red color was immediately measured at 480 nm using a UV-visible spectrophotometer (Thermo Scientific, model-Evolution 600). The optical density of the iron content was utilized to calculate the phytate phosphorus content, assuming a constant molecular ratio of 4 Fe to 6 P as described by Sadasivam and Manickam [44].

Antioxidant activity

Total Phenolic Content (TPC)

The Folin Ciocalteu technique was employed to ascertain the TPC, as per the guidelines established in [53]. A 0.2 mL segment of the extract was utilized. This segment was mixed with 1.5 mL of Folin-Ciocalteu reagent procured from Sigma-Aldrich. The concoction was left to sit at ambient temperature for 5 min. Subsequently, 1.5 ml of a 6% solution of sodium carbonate (CDH) was introduced to the mixture. The specimen was permitted to undergo incubation at room temperature for 1.5 h. Post incubation, the absorbance of the sample was gauged at 725 nm wavelength through a spectrophotometer. The calibration curve was constructed by utilizing 1 mL portions of 50, 100, 150, 200, 250, 300, 350, 400, and 450 mg/mL gallic acid dilutions (Sigma-Aldrich). The primary phenolic composition found in millets is gallic acid, recognized as 3,4,5-trihydroxy benzoic acid (C₆H₂(OH)₃CO₂H), hence the outcomes were quantified and expressed as gallic acid equivalent per 100 g of Dry Weight (DW). This approach is widely employed for evaluating the total phenolic content in specimens and acts as a crucial gauge of their antioxidant power.

Estimation of total flavonoid content

A colorimetric method [22] with certain modifications was employed for the quantification of overall flavonoid content. To elaborate, 25 µl of a diluted sample were

combined with 125 μ l of double-distilled H_2O . Following this, 7.5 μ l of 5% $NaNO_2$ (CDH) were introduced to the mixture and allowed to incubate for 5 min, after which 15 μ l of 10% $AlCl_3$ (CDH) were added. The amalgam was left to incubate at room temperature (25 °C) for an additional 5 min, and then 50 μ l of 1 M NaOH was added. Subsequently, the mixture was promptly diluted by the addition of 27.5 μ l of dd H_2O , and the absorbance of the solution was measured at 510 nm in comparison to a blank solution prepared using dd H_2O .

DPPH free radical scavenging assay

The assessment of the DPPH free radical scavenging capability of the sample was conducted through the observation of the decline in absorbance of an ethanolic DPPH (2,2-diphenyl- 1-picrylhydrazyl) solution at a wavelength of 517 nm [31]. A primary DPPH solution (Sigma-Aldrich) was prepared at a concentration of 0.1 mM (mm). Subsequently, the prepared DPPH solution was left undisturbed for 30 min. Following the incubation period of 30 min, the absorbance of the solution was measured at a wavelength of 517 nm.

Determination of reducing power

The millet extract's Ferric Reducing Antioxidant Power (FRAP) was evaluated according to the method outlined by Sreeramulu et al. [51]. Initially, samples were combined with potassium ferricyanide (Fe^{3+}) to convert it to potassium ferrocyanide (Fe^{2+}). Subsequently, the resulting potassium ferrocyanide reacted with ferric chloride, forming a complex called the ferric ferrous complex. Each sample underwent a process where 0.1 mL was mixed with 0.9 mL of 96% ethanol, 5 mL of distilled water, 1.5 mL of 1 M Hydrochloric acid (HCl), 1.5 mL of 1% potassium ferricyanide $K_3 [Fe (CN)_6]$, 0.5 mL of 1% sodium dodecyl sulphate (SDS), and 0.5 mL of 0.2% ferric chloride ($FeCl_2$) (Sigma-Aldrich). The mixture was then heated in a water bath at 50 °C for 20 min, followed by rapid cooling and thorough mixing. The absorbance of each sample was measured at a wavelength of 700 nm using a spectrophotometer, with ascorbic acid serving as the standard. The calibration curve was plotted by 1 mL aliquots of 20, 40, 60, 80, 100, 120, 140, and 160 mg/mL ascorbic acid dilution.

Functional characteristics

Bulk density

Bulk density was estimated according to the method given by Chau and Huang [6].

Water Absorption Capacity (WAC)

The water activity capacity (WAC) was measured using Beuchat's method [3]. One gram of sample was mixed with 10 mL of distilled water in a vortex tube. The mixture was then centrifuged at a speed of 3000–5000 X g for 20–25 min. Once separated, the volume of the liquid portion (supernatant) was noted to determine water absorption, which was expressed as grams per milliliter of the sample.

Oil-Holding Capacity (OHC)

One gram of sample was mixed with vegetable oil (1:10) and stirred on vortex for 30 min followed by centrifugation at 3000–5000 $\times$ g for 30–45 min. After the separation of the content, the volume of the supernatant was recorded and used to determine water absorption. The OHC was expressed as a milliliter of vegetable oil held per gram of sample [6].

Gelation temperature

The gelation temperature was determined using Narayana and Narasinga's method [35]. In a 250 mL beaker, 10 grams of the starch sample was suspended in distilled water, and the volume was 100 mL. A thermometer was clamped with a stand in such a way that its bulb got submerged in the suspension. With a magnetic stirrer, the suspension was continuously stirred and heated. This continued until the suspension began to gel and the corresponding temperature was recorded.

Pasting properties

The viscoamylographic properties of the samples were performed with a Rapid Visco Analyser (Perten Equipments, USA) using 3 g of sample in 25 mL of water. The parameters paste temperature, peak viscosity, breakdown viscosity, final viscosity, and setback viscosity were measured from the viscoamylographs.

Results and Discussion

Proximate composition

The contents of proximate components in raw and hydrothermally treated proso millets, including protein, crude fat, ash, and dietary fiber fractions are reported in Table 1. The studied samples were raw proso millet (RPM), soaked proso millet (SOPM), blanched proso millet (BLPM), steamed proso millet (STPM), roasted proso millet (ROPM), and pressure-cooked proso millet (PCPM). Moisture contents ranged from 5.20% to 8.46%, significantly higher moisture content was recorded in SOPM (8.46%), while lowest moisture content was recorded in ROPM (5.00%). Zhang et al. [58] recorded a high moisture content ranging from 7.96% to 9.63% in

Table 1 Proximate Composition of raw and treated Proso millet (g/100 g)

Sample	Moisture	Ash	Protein	Fat	Fibre	Carbohydrate
RPM	5.20 ± 0.81 ^a	3.89 ± 0.50 ^c	13.94 ± 7.55 ^b	4.52 ± 0.58 ^c	2.75 ± 0.49 ^d	73.59 ± 1.70 ^c
SOPM	8.46 ± 0.23 ^d	2.00 ± 0.86 ^a	12.04 ± 8.20 ^a	2.98 ± 0.96 ^b	2.03 ± 0.69 ^a	71.49 ± 1.03 ^a
BLPM	6.40 ± 0.34 ^b	3.33 ± 0.50 ^b	13.64 ± 0.30 ^b	2.41 ± 0.79 ^a	2.22 ± 0.42 ^b	71.33 ± 0.78 ^a
STPM	6.13 ± 0.41 ^b	3.50 ± 0.76 ^c	16.18 ± 0.31 ^c	4.68 ± 0.50 ^c	2.44 ± 0.19 ^c	76.11 ± 1.02 ^d
ROPM	5.00 ± 0.34 ^a	4.16 ± 1.60 ^d	16.68 ± 0.45 ^c	2.77 ± 0.26 ^b	2.00 ± 0.17 ^a	73.05 ± 0.36 ^b
PCPM	7.46 ± 0.23 ^c	3.16 ± 0.28 ^b	18.39 ± 0.61 ^d	4.74 ± 0.46 ^d	2.40 ± 1.73 ^c	73.45 ± 0.98 ^c

Values expressed are mean ± standard deviation

RPM Raw Proso Millet, SOPM Soaked Proso Millet, BLPM Blanched Proso Millet, STPM Steamed Proso Millet, ROMP Roasted Proso Millet, PCPM Pressure Cooked Proso Millet

Data of different alphabets in the same column were different with statistical significance ($p < 0.05$)

proso millet cultivars. In another research, an analysis of proso millet varieties for moisture content showed a significant difference at a 5% level, with moisture content ranging from 8.04–8.34% [20]. Between the raw and hydrothermally treated proso millets, ash content ranged from 2.00 to 4.16 g/100 g. ROMP recorded significantly higher ash content (4.16 g) and SOPM recorded lower (2.00 g) ash content. This may happen because the moisture content and ash content in food are inversely related. When the grains have high moisture content, the ash content is usually lower as the water dilutes the mineral concentration. Mineral leaching that occurs during soaking of grains was previously noted by Kumar et al. [23]. A similar reduction in the ash content was observed in hydrothermally-treated millets by Iralepatil et al. [15]. It has been observed that the protein content of the proso millet was significantly affected by the hydrothermal process, specifically, the roasting and pressure cooking as shown in Table 1. The RPM was found to have a protein content of 13.94 g/100 g. Protein content was found to decrease with the soaking time in SOPM. The decrease in protein may be due to the prolonged soaking time in which the protein molecules get separated and sunk inside the gelatinized starch and become less available for extraction [28]. Similar results were reported in the study made by Rajasekhar et al. [42] and Prashanth et al. [40]. Dharmaraj and Malleshi [10] have also reported that alterations in the composition of the constituent protein fractions could potentially arise from the process of protein bodies' denaturation, as well as the plausible occurrence of protein-starch and protein-polyphenols complexes. However, the highest retention of protein was observed in PCPM (18.39 g/100 g) as in pressure cooking the proteins get denatured, thus making them more easily digestible. It also improves protein availability by breaking down some of the antinutrients like phytates.

The fat content of the samples showed a decreasing trend in soaking, blanching, and roasting. This decrease can be attributed to its complexing with the amylose component of the millet starch [40], [10]. Messina et al. [32] reported that hydrothermal treatment could break fat globules located in the germ and aleurone layers of the grains, initiating a redistribution and /or migration of fat to the neighbouring endosperm structure. This might be the reason for the changes in fat content in the sample. Mahadevappa and Desikachar [29] also reported the destruction of the globular structure of fat molecules in parboiled paddy. They reported that it might have occurred due to the expansion of the endosperm cells during parboiling. However, an increasing trend has been observed in steaming and pressure cooking. The highest value (4.74 g/100 g) of fat was observed in pressure-cooked samples. This may happen because pressure cooking can help retain the natural fat content of proso millet, as it uses high pressure and temperature to cook the grain quickly. It also redistributes the fat content within the grain, making it more evenly distributed and increasing its bioavailability. In similar research, the fat content of proso millet was found in the range of 2.92 and 3.35 g/100 g [20]. In one study [58], fat content was reported to be 3.64 to 4.40 g which was found by the present study. A slight reduction in the fiber content was also observed in hydrothermally-treated proso millet samples. Pressure cooking also had a less significant effect on fiber content. The results were reported in previous research [20, 40]. Similarly, the effect of hydrothermal treatment on the total carbohydrate content of proso millet was found to be statistically insignificant, except for STPM (76.11 g/100 g). Carbohydrate content in STPM increased significantly as steaming can help retain the natural carbohydrate content of proso millet. This could happen because steaming uses minimal water and does not involve high temperatures. Steaming can also gelatinize some of the starch, making it more easily

Table 2 Mineral Composition of raw and treated Proso millet (mg/L)

Sample	Ba	Mg	Zn	Se	Cr	Cu	Fe	K	Mn	Sr
RPM	0.59 ± 0.54 ^a	2802.08 ± 1.82 ^a	8.85 ± 0.67 ^a	0.334 ± 0.09 ^c	7.49 ± 0.88 ^f	0.77 ± 0.05 ^a	60.80 ± 1.12 ^d	4256.11 ± 2.18 ^a	0.33 ± 0.05 ^a	0.51 ± 0.09 ^b
SOPM	1.61 ± 0.89 ^c	5222.32 ± 2.84 ^e	11.74 ± 1.16 ^d	0.158 ± 0.06 ^a	3.06 ± 0.96 ^c	1.09 ± 0.87 ^b	42.01 ± 1.35 ^c	4838.63 ± 1.67 ^c	3.37 ± 0.94 ^b	1.22 ± 0.24 ^d
BLPM	0.74 ± 0.11 ^b	4585.04 ± 2.16 ^c	22.27 ± 1.14 ^f	0.470 ± 0.04 ^e	3.31 ± 0.74 ^d	1.23 ± 0.68 ^d	40.28 ± 0.96 ^c	5115.83 ± 1.13 ^d	3.68 ± 0.83 ^c	1.31 ± 0.35 ^d
STPM	1.85 ± 0.42 ^d	5321.72 ± 1.49 ^f	14.48 ± 0.98 ^e	0.381 ± 0.08 ^d	4.15 ± 0.81 ^e	1.14 ± 0.74 ^c	30.85 ± 1.06 ^b	4702.68 ± 1.05 ^b	7.73 ± 0.79 ^e	3.08 ± 0.31 ^e
ROPM	0.65 ± 0.07 ^a	4400.82 ± 1.53 ^b	10.42 ± 1.21 ^c	0.277 ± 0.08 ^b	2.61 ± 0.62 ^b	1.15 ± 0.83 ^c	120.44 ± 2.11 ^e	7120.02 ± 1.08 ^f	4.02 ± 0.92 ^d	0.37 ± 0.04 ^a
PCPM	0.65 ± 0.06 ^a	4857.67 ± 0.89 ^d	9.79 ± 1.32 ^b	0.476 ± 0.06 ^e	0.54 ± 0.06 ^a	1.16 ± 0.64 ^c	21.03 ± 1.13 ^a	5276.12 ± 1.24 ^e	3.26 ± 0.87 ^b	0.76 ± 0.06 ^c

Values expressed are mean ± standard deviation

RPM Raw Proso Millet, SOPM Soaked Proso Millet, BLPM Blanched Proso Millet, STPM Steamed Proso Millet, ROPM Roasted Proso Millet, PCPM Pressure Cooked Proso Millet
Data of different alphabets in the same column were different with statistical significance ($p < 0.05$)

accessible to enzymes. A slight decrease in carbohydrate content was observed in all the other treatments such as blanching and soaking, in which loss of carbohydrate content may occur due to extraction of carbohydrates in blanching water, while roasting can caramelize some of the carbohydrates, leading to a slight reduction in carbohydrate content. Similar results were obtained in the study of Thilagavathi et al. [52] who reported that moisture, fat, fiber, ash, and total carbohydrate content were 11.75 g, 3.78 g, 4.87 g, and 64.84 g respectively.

Mineral content

Minerals are necessary nutrients for human health and the function of the body is critical to their proper functioning [54]. Minerals serve a variety of purposes in the body. Minerals make up about 4–5% of the weight of an adult human, with the skeleton containing the majority of these minerals and the remainder in other organs [30]. The mineral analysis was done by Inductively coupled plasma optical emission spectrometry (ICP-OES) method and the result is given in Table 2. In comparison to major cereal grains, proso millet is a good source of minerals like phosphorus, potassium, magnesium, manganese, iron, and zinc. Except for chromium, an increase in all the mineral content (Ba, Mg, Zn, Se, Cu, Fe, K, and Mn) was observed in hydrothermally-treated proso millet samples. The highest iron content was found in ROPM sample (60.80 mg/L). Similar observations on the extrusion processing of other grains have been also reported. Singh et al. [48] reported an increase in iron (3.45–3.91 mg/100 g) as well as calcium (337.31–341.24 mg/100 g) in finger millet subjected to roasting, suggesting research probability for newer insights into these changes. Roasting grains can increase iron bioavailability because it reduces phytates, which are natural compounds that inhibit iron absorption, and enhances the release of phenolic acids, which can also improve iron absorption. Obadina et al. [36] studied the nutritional profile of roasted pearl millet at varying temperatures and time combinations (120 °C, 140 °C for 5, 10, and 15 min and 160 °C, 180 °C for 3, 5, and 10 min). This resulted in up to 274% increase in iron content. Increased contents of iron and zinc were reported in fonio millets after hydrothermal treatment [8]. However, a decrease in iron content was observed in SOPM, BLPM, STPM, and PCPM samples. During soaking, outward leaching of minerals into the soaking medium can occur [5]. The reduction in iron content from 43.23 to 38.51 ppm in pearl millet within 12 h of soaking was attributed to leaching in the water used for soaking [47]. A significant decrease in iron was also observed after pressure cooking (43.2 to 36.30 ppm) for 10 min and steaming (43.23 to 36.79 ppm) for 20 min [8].

Conversely, a decrease in the level of chromium concentration was observed in all the hydrothermally-treated proso millet samples. Dharmaraj and Malleshi [10] also reported decrease in the levels of copper, phosphorous, and total mineral content in hydro-thermally treated finger millet. In this study, copper and manganese content of hydrothermally-treated proso millet ranged from 1.09 mg/L to 1.23 mg/L and 3.26 mg/L to 7.73 mg/L respectively, which is higher than that of RPM. These values were slightly higher than the values reported by Gopalan et al. (2012) for copper (1.06 mg/100 g) and manganese (1.15 mg/100 g) as evaluated in pearl millet. There was a significant difference in the manganese content between RPM and hydrothermally-treated samples. Hydrothermal processing did not cause significant changes in the nutrient contents of the millet. A slight decrease in the content of some of the nutrients could be due to the loss during steeping.

Antinutritional and antioxidant properties

The primary phosphorus-storing component of the majority of seeds and cereal grains is called phytate (myoinositol hexa-phosphoric acid), which may make up more than 70% of the total [14]. The effect of hydrothermal treatment on the major antinutrients, tannins, TFC, TPC, and phytates present in proso millet has been investigated and the findings have been presented in this section. Tannins are water-soluble polyphenolic compounds [37]. The tannin content in the hydrothermally-treated samples ranged from 0.152 to 0.161 mg TAE/g. Tannins were significantly reduced by soaking treatment. The reduction in tannins can be attributed to the leaching of the polyphenols into water [16]. Pressure cooking at higher temperatures with less time also shows a significant reduction. The decrease in these compounds can be ascribed to their water-soluble and heat-sensitive characteristics. These compounds degrade at elevated temperatures and subsequently leach into the liquid medium [17]. Similar results were observed by Wu et al. [56] in their study on the effects of temperature on the tannin and phytate contents of sorghum grains. As compared to the raw sample, the phytic acid also decreased in all the hydrothermally-treated samples and ranged from 2.128 to 3.115 mg GAE/g. Pressure cooking was found to have a significant effect on the phytic acid content ($P < 0.05$). At high temperatures and decreased time, a significant retention of the phytic acid was observed. The variations in the total phytic acid could be attributed to the influence of the phytase enzyme, which is responsible for breaking down phytates. At lower temperatures (BLPM), extended soaking periods (SOPM), and reduced steaming time (STPM), the enzyme becomes highly active, thus reducing phytates [38]. On the contrary, if

Table 3 Antioxidant and Antinutritional properties of raw and treated Proso millet

Sample	DPPH%	TFC (mgQE/g)	TPC (mgGAE/g)	FRAP (mmol of Fe(II)/g)	Tannin (mgTAE/g)	Phytic acid (mg/g)
RPM	12.757 ± 0.83 ^a	2.559 ± 0.41 ^b	2.551 ± 0.85 ^b	0.424 ± 0.08 ^a	0.282 ± 0.07 ^f	3.124 ± 0.91 ^b
SOPM	15.572 ± 0.71 ^c	2.488 ± 0.52 ^a	1.548 ± 0.63 ^a	0.498 ± 0.09 ^b	0.161 ± 0.09 ^b	2.215 ± 0.63 ^a
BLPM	14.720 ± 1.21 ^b	2.335 ± 0.17 ^a	1.870 ± 0.49 ^a	0.501 ± 0.11 ^b	0.173 ± 0.08 ^c	2.128 ± 0.29 ^a
STPM	15.103 ± 0.98 ^c	2.521 ± 0.33 ^b	1.753 ± 0.57 ^a	0.566 ± 0.09 ^c	0.197 ± 0.04 ^e	2.126 ± 0.74 ^a
ROPM	18.003 ± 0.88 ^e	3.433 ± 0.67 ^d	2.634 ± 0.53 ^b	0.766 ± 0.09 ^e	0.181 ± 0.10 ^d	3.107 ± 0.62 ^b
PCPM	17.165 ± 0.79 ^d	3.130 ± 0.94 ^c	2.662 ± 0.24 ^b	0.653 ± 0.10 ^d	0.152 ± 0.09 ^a	3.115 ± 0.38 ^b

Values expressed are mean ± standard deviation

RPM Raw Proso Millet, SOPM Soaked Proso Millet, BLPM Blanched Proso Millet, STPM Steamed Proso Millet, ROMP Roasted Proso Millet, PCPM Pressure Cooked Proso Millet

Data of different alphabets in the same column were different with statistical significance ($p < 0.05$)

higher temperatures (ROPM and PCPM) are applied, the enzyme can be denatured, resulting in the retention of phytates [40]. These findings aligned with a similar study conducted by Sihag et al. [47] that examined the impact of domestic processing methods on the levels of phytic acid and polyphenols in pearl millet. The effect of hydrothermal treatment on the TPC of proso millet grains has been listed in Table 3. TPC was found to be not affected significantly ($p < 0.05$) with the increased temperature as in the case of roasting and pressure cooking. For soaking, blanching, and steaming, TPC was reduced. The highest TPC (2.662 mg GAE/g) was observed for the PCPM samples. The possible cause for this phenomenon could be attributed to the influence of the polyphenol oxidase enzyme. Under elevated temperatures, it is plausible that the enzyme undergoes denaturation. Furthermore, at higher temperatures, there is an enhanced liberation of phenolics due to the disintegration of cellular structures [8]. Consequently, this leads to an increase in the total phenolic content. Correspondingly, Azad et al. [18] also reported comparable findings in their investigations on the impact of four different hydrothermal processes on the functional characteristics of broomcorn millet flour. Similarly, TFC was found to increase significantly ($p < 0.05$) with the increased temperature in case of ROMP and PCPM samples. Azad et al. [18] studied the effects of four hydrothermal processes on the functional properties of broomcorn (*Panicum miliaceum* L.) millet flour. TFC were observed to be highest in the roasted samples followed by steamed, puffed, extruded, and untreated control samples. The effect of roasting and popping on the TPC, TFC of two cultivars of amaranth seeds was studied by Muyonga et al. [33]. A slight increase in TPC was observed upon roasting. TFC also increased in the roasted and popped samples compared to the raw samples. This increase in the TPC and TFC were attributed to the release of bound phenolics, and flavonoids, caused

by the disruption of the cell walls upon application of high temperature.

The values for total antioxidant capacity for proso millet sample by DPPH (2, 2 diphenyl-1-picryl-hydrazyl) radical scavenging assay and FRAP assay were found to be highest in roasted samples, while RPM samples showed lowest value of both DPPH and FRAP. Since flavonoids along with other phenolics mostly contribute to the antioxidant activity in plants, increase in these compounds could be the reason for the enhanced antioxidant activity. Roasting has been reported to improve phenolic content in grains by disrupting cell wall structures [18]. In a study by Siroha and Sandhu [49] on pearl millet cultivars, the effect of toasting, and cooking on the antioxidant activity (AOA) of flours was investigated. Mild oven toasting at 115 ± 5 °C with prior conditioning of samples till 10% moisture content resulted in a significant increase in AOA of pearl millet cultivars. This was attributed to the release of bound phenolics due to the disintegration of cellular constituents. Enhanced AOA was also attributed to specific melanoid compounds formed on processing.

Functional properties

Functional properties of raw and processed cereal grains are mostly associated with the status of their most abundant, and versatile hygroscopic component, starch, and the relatively abundant proteins. Products made from dough and batter need higher water absorption capacity of the flour, which can be attained by hydrothermal treatments. The functional properties of raw and hydrothermally treated proso millet samples are presented in Table 4. The bulk density (BD) of the control sample (RPM) was found to be 0.55 g/mL. Hydrothermally treated proso millet flour was found to have higher bulk densities compared to raw proso millet samples. It was observed that bulk density was maximum (0.70 g/mL) for

Table 4 Functional Properties of raw and treated Proso millet

Sample	BD (g/mL)	WAC (g/mL)	OAC (g/mL)
RPM	0.55 ± 0.03 ^a	1.28 ± 0.12 ^a	1.60 ± 0.15 ^a
SOPM	0.70 ± 0.09 ^c	1.43 ± 0.16 ^b	1.77 ± 0.54 ^b
BLPM	0.61 ± 0.05 ^b	1.40 ± 0.29 ^b	1.72 ± 0.27 ^b
STPM	0.50 ± 0.04 ^a	1.52 ± 0.13 ^c	1.73 ± 0.21 ^b
ROPM	0.59 ± 0.05 ^b	1.44 ± 0.12 ^b	1.75 ± 0.14 ^b
PCPM	0.66 ± 0.08 ^c	1.55 ± 0.24 ^c	1.87 ± 0.18 ^c

Values expressed are mean ± standard deviation

RPM Raw Proso Millet, SOPM Soaked Proso Millet, BLPM Blanched Proso Millet, STPM Steamed Proso Millet, ROPM Roasted Proso Millet, PCPM Pressure Cooked Proso Millet

Data of different alphabets in the same column were different with statistical significance ($p < 0.05$).

the soaking treatment. Similar results were reported by Prashanth et al. [41] and Yadav et al. [57] for pearl millet. This property dramatically influences the packaging properties, which can be an advantage to pack a higher amount of hydrothermally treated flour than normal flour within a constant volume. A decrease in bulk density has also been observed for samples that have been exposed to prolonged steam treatment. This phenomenon can be explained by the extended exposure to steam, which likely caused starch gelatinization and subsequent reduction in bulk density [57]. Soaking temperature and steaming time predominantly had a higher positive effect on the bulk density of the samples.

Hydrothermally processed products attain a moisture-induced consistency. Therefore, water absorption capacity (WAC) of raw materials and ingredients are critical for processability. For cereal grains, starch is the major and largely hygroscopic fraction. Therefore, starch primarily relates to the WAC. It generally represents the structural integrity of starch in its aqueous dispersion and is often used as an index for starch gelatinization [34]. Interaction of starch with non-starch components in a processed food matrix can remarkably affect WAC. The WAC of

RPM was found to be 1.28 g/mL which significantly increased in hydrothermally treated samples. The highest WAC (1.55 g/mL) was found in PCPM. In one study [52], WAC of different millet flours were found to be within the range of 74.08–76.83 mL/100 g against 74.08–78.83 mL/100 g for wheat and 58.17–60.02 mL/100 g for soybean. Similarly, hydrothermally treated millet flour can show higher oil absorption capacity (OAC). This was attributed to the treatment-induced lipophilicity of millet starch. Intrinsic factors like the composition of amino acids, conformation of protein, and surface hydrophobicity govern OAC. Nonpolar amino acid chains of hydrophobic proteins bind with the paraffin chains of fats. Generally, both higher WAC and OAC also represent better organoleptic properties. The OAC of RPM was found to be 1.60 g/mL which significantly increased after hydrothermal treatment. The highest OAC was observed in PCPM (1.87 g/mL). This may be due to the fact that binding of the lipid depends on the surface availability of hydrophobic amino acids [9, 50]. The oil absorption capacity of any food material is important because it relies mainly on its capacity to physically entrap oil by a complex capillary attraction process. Fat acts as a flavour retainer, a consistent trait and an enhancer of mouth feel [21]. In a research, HMT pearl millet starch samples showed significantly higher OAC which was almost twice compared to the native starch [46]. When oil absorption capacity (OAC) was assessed, finger millet, and pearl millet flours showed values of 1.93 and 1.60 g/g, respectively [12].

Pasting properties

The pasting properties can indicate quality parameters such as pasting, degradation and starch expansion in flour. As shown in Table 5, a slight difference in pasting temperature and peak time for all samples ($p > 0.05$) was observed. Peak time ranged from 0.00 min at the minimum to 7.00 min at the maximum. However, there

Table 5 Pasting Properties of raw and treated Proso millet

Sample	Peak Viscosity (cP)	Through Viscosity (cP)	Breakdown (cP)	Final Viscosity (cP)	Set Back (cP)	Peak time (min)	Pasting Temp (°C)
RPM	276.00 ± 1.47 ^d	206.00 ± 2.45 ^c	61.00 ± 1.07 ^d	1552.33 ± 3.02 ^f	1340.66 ± 3.03 ^e	7.00 ± 0.97 ^a	92.46 ± 0.27 ^a
SOPM	361.33 ± 2.18 ^f	285.00 ± 2.17 ^d	72.33 ± 2.16 ^e	574.66 ± 2.50 ^e	280.00 ± 2.58 ^d	6.26 ± 0.88 ^a	92.12 ± 0.94 ^a
BLPM	61.33 ± 1.14 ^c	38.00 ± 1.16 ^b	26.00 ± 1.56 ^b	142.33 ± 2.87 ^c	105.66 ± 2.68 ^c	6.75 ± 0.69 ^a	95.96 ± 0.80 ^b
STPM	352.66 ± 1.78 ^e	323.00 ± 1.72 ^e	31.33 ± 1.05 ^c	435.00 ± 1.52 ^d	100.00 ± 2.71 ^c	7.40 ± 1.05 ^c	96.15 ± 0.76 ^b
ROPM	19.33 ± 1.02 ^a	16.00 ± 0.98 ^a	15.66 ± 1.08 ^a	37.66 ± 1.16 ^a	31.00 ± 1.58 ^a	7.10 ± 1.11 ^b	97.32 ± 1.14 ^c
PCPM	28.33 ± 1.13 ^b	17.00 ± 1.05 ^a	15.33 ± 1.89 ^a	80.00 ± 1.35 ^b	66.00 ± 1.52 ^b	7.15 ± 0.93 ^b	96.06 ± 0.93 ^b

Values expressed are mean ± standard deviation

RPM Raw Proso Millet, SOPM Soaked Proso Millet, BLPM Blanched Proso Millet, STPM Steamed Proso Millet, ROPM Roasted Proso Millet, PCPM Pressure Cooked Proso Millet

Data of different alphabets in the same column were different with statistical significance ($p < 0.05$).

were significant variations in all of the RVA viscosity parameters. The range for peak viscosity was found to be 19.33 to 361.33 cP. The three derivative parameters were, in order, 15.66 to 72.33 cP, 31.00 to 1340.66 cP, and 37.66 to 1552.33 cP for breakdown viscosity (BD), setback viscosity (SB), and final viscosity (Table 5). Final

viscosity showed the greatest variation among these six viscosity parameters [25]. Due to hydrothermal processing, a significant decrease in peak viscosity was observed in blanched, pressure cooked, and roasted samples (Fig. 1). It was assumed that break down viscosity indicates the ease with which swollen starch

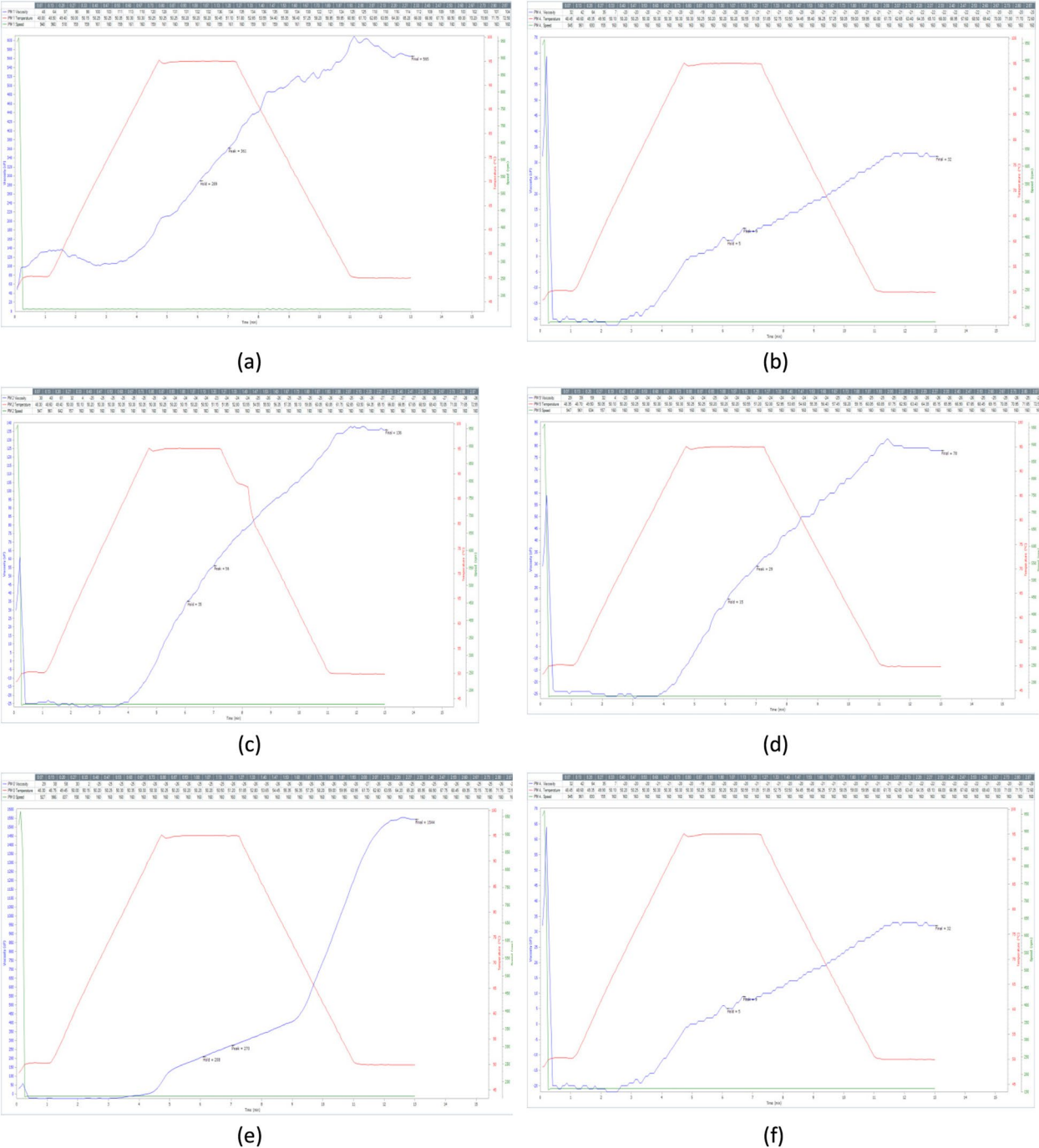


Fig. 1 Pasting properties of raw and treated Proso Millet. **a** RPM: Raw Proso Millet **(b)** SOPM: Soaked Proso Millet **(c)** BLPM: Blanched Proso Millet **(d)** STPM: Steamed Proso Millet **(e)** ROPM: Roasted Proso Millet **(f)** PCPM: Pressure Cooked Proso Millet

granule disintegrated or in other words the degree of its organization [11]; [4]. All these treated samples contain pregelatinized starch, which almost look like a homogeneous mass with least degree of organization. Peak viscosity is an important characteristic of any starch or flour. The reduced peak viscosity after hydrothermal processing may be due to limited starch swelling and structural disintegration. These observations are in line with the earlier report on hydrothermal treatment to finger millet starch by (10).

Cooking quality of flours is indicated by their pasting and rheological properties. Yadav et al. [57] reported a marked decrease in PV (1362.3–115.7 cP) and CPV (1938.7–432.0 cP) values with increased steaming duration in pearl millet. Granular degradation ideally causes drop in granular or paste swelling on heating and lowering the recorded PV. Severely broken-down glycosidic chains fail to retrograde efficiently, thereby giving such low CPV and SB. The pasting temperature showed an increase from 92.46 °C to 96.15 °C on steaming and 97.32 °C on roasting. Under controlled moisture content in HMT processes, the pasting temperature of hydrothermally treated millet starches tend to increase. This owes to the formation of cross-linkages and rearrangement of amorphous regions, therefore requiring more thermal energy for paste formation. In a work reported by Sharma et al. [45], native pearl millet starch showed a pasting temperature of 77.3 °C. Upon hydrothermal treatment at 25% and 30% moisture content (HMT 25 and HMT 30), pasting temperature increased by 5.3 °C and 6.5 °C.

Conclusion

This study demonstrates that various hydrothermal treatments can significantly enhance the nutritional profile and functional properties of proso millet. By applying techniques such as steeping, blanching, steaming, roasting, and pressure cooking, it is possible to reduce anti-nutritional factors like tannins and phytic acid, thereby improving the grain's overall nutritional value. The treatments also positively influenced the antioxidant activity, mineral content, and functional properties such as pasting behaviour, which are important for its use in food production. Roasting and pressure cooking had the most pronounced impact on all measured parameters compared to the other treatments. These findings indicate that optimizing processing techniques can enhance the nutritional value and versatility of proso millet, positioning it as a more effective grain for tackling global dietary challenges.

Future research directions

Despite providing valuable insights, this study has certain limitations that need consideration. The scope of hydrothermal treatments can be extended to processing methods such as germination, fermentation, or extrusion that may have distinct or synergistic effects on the nutritional and functional properties of proso millet. Additionally, to evaluate the bioavailability of nutrients and antioxidants in vivo studies can be done. Varietal differences in millets can significantly influence processing outcomes. These limitations suggest the need for future studies that may incorporate a wider range of processing methods, multiple millet genotypes, and in vivo assessments to better understand and optimize the health and functional potential of proso millet.

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Authors' contributions

TJ: Research work, formal analysis, data curation; SY: First draft writing, Methodology; MA: Final draft checking, Software, Reviewing; PS: Conceptualization, Supervision, Final reviewing.

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