Comparative Effects of Traditional Processing Techniques on the Phytic Acid Reduction in Commonly Consumed Pulses in Pakistan Households

Fazia Ghaffar

Department of Food & Nutrition Sciences, College of Home Economics, University of Peshawar Corresponding author email: faziaghaffar@uop.edu.pk

Abstract

Pulses are a vital source of essential dietary nutrients, contributing significantly to global and local food economies. These nutrient-dense legumes are rich in protein, fiber, vitamins, and minerals, enhancing diet quality while providing key health benefits. However, their inherent phytic acid content acts as an antinutrient, potentially hindering mineral absorption. Various cost-effective domestic processing methods can be employed to degrade phytic acid to improve its nutritional utility. The present study involves four common pulse cultivars: Red Lentils (Lens culinaris), Split Mung pulse (Vigna radiata), Urad pulse (Vigna mungo), and Split Bengal Gram pulse (Cicer arietinum) to investigate the diverse soaking and cooking techniques for their efficacy in reducing phytate content. Results demonstrated that soaking pulses for an hour led to a 40.6% reduction in phytic acid. In comparison, a six-hour soaking yielded a higher degradation of 44.5%, with extended soaking (overnight) showing variable effects due to saturation. Boiling pulses until tender resulted in a 43% reduction in phytates, whereas water absorption boiling led to a lower 35.7% degradation. Pressure cooking methods exhibited variable efficacy, with cooking without bicarbonate soda leading to a greater phytate reduction (above 36.3%) than cooking with soda. These findings highlight the importance of traditional household processing techniques in enhancing the bioavailability of nutrients in pulses. By optimizing soaking and cooking conditions, the nutritional quality of pulses can be improved, making them more effective in supporting dietary health and overall well-being.

Keywords: Pulses, Percent Phytate content, Percent phytate degradation, soaking, boiling, pressure cooking Highlights

- Raw pulses have higher concentrations of phytic acid.
- Soaking pulses leads to significant phytate degradation. •
- Cooking under vacuum (pressure cooking) results in greater phytate degradation. •
- As expected, adding bicarbonate with prolonged cooking times leads to comparatively less phytate degradation.

1. Introduction

Pulses, also called legumes, are considered a significant source of plant protein. They are regarded as smart crops for human consumption and cropping systems. In the human diet, they are a source of protein, minerals, vitamins, and fiber. They contribute to maintaining soil biodiversity by contributing nitrogen to the soil. Globally, pulses contribute about 33% of the human population's global dietary protein requirements (Ullah et al., 2020). Pulses are the most important source of vegetable protein in Pakistan, being cultivated in 5% of the total cropped area. Being the "poor man's meat", their uses range from producing baby foods to delicacies of the rich and major food of the poor (AARI, 2024).

The term pulses and legumes are used interchangeably. Leguminosae's pulse family consists of plants that produce pods with seeds. The word "pulses" has been described as crops cultivated solely for dry seeds of the leguminous plants. Commonly edible pulses include dry peas, lentils, chickpeas, green mung beans, etc. Pulses are reportedly part of human nutrition throughout 5500 B. C. in many civilizations and are considered the oldest crops cultivated by humanity. They are valued as the major meal of low-income people and an alternative to meats (Soomro et al., 2020; Kouris-Blazos & Belski, 2016). Pulses are considered an excellent source of protein, minerals, vitamins, fiber, and starchy carbohydrates with health promotion benefits as they are likely to lower the risk of many non-communicable diseases with a significant variation in proteins, carbohydrates, and the daily minerals' requirements can be met by consuming 100-200 g pulses daily (Langyan et al., 2022, Gowda et al., 2015). Besides being abundant in macronutrients (except lipids) and fiber, pulses are rich in many nutrients (Tharanathen & Mahadevamma, 2003; Besseda et al., 2019). The protein quality of pulses is considered to be of good biological value (Siddhuraj et al., 2002; Escurdero et al., 2006). As reported, regular consumption of 3-4 times per week and at least once a week can help fight against cardiovascular diseases (Flight & Clifton, 2006). As food crops, pulses and cereals are considered to significantly contribute to providing dietary energy sources for many populations (Mal, 1992; Sidduraj et al., 2000; Kalidaas & Mohan, 2011). It is also a rich source of highquality protein and a substantial source of minerals and water-soluble vitamins (Friedman, 1996).

Besides being rich in macronutrients, pulses have essential micronutrients: iron, potassium, magnesium, zinc, and B complex vitamins (Langvan et al., 2022). Pulses manufacture isoflavones, natural secondary metabolites with excellent hypocholesterolemic properties (Oomah & Mazza, 2000). Pulses contain a variety of isoflavones such as genistein,



glycitein, daidzein, genistin, daidzin, protensin, biochanin A, and formononetin which are protective against cardiovascular diseases. However, the distribution of isoflavones across different pulses has been explored in limited studies. Bengal gram (channa dhal), green mung, and black gram(mash dhal)have been reported to contain higher isoflavone content (Chibbar et al.,2010; Suneja et al., 2011). Besides isoflavones, pulses contain lectins, enzyme inhibitors, oxalates, oligosaccharides, phytic acid, and phenolic compounds, which are some other naturally occurring bioactive compounds that are available from pulses both as whole grains and split pulses (Henchion et al., 2017). The primary polyphenolic compounds in pulses include tannins, flavonoids, and phenolic acids. They are abundant in black gram, lentils, and some others. Pulses contain antioxidant compounds such as tocopherols, ascorbic acid, and isoflavones. These polyphenols may act as "preventive" antioxidants by chelating the catalytic transition metals such as copper and iron, rendering them unavailable for Fenton-type reactions. They may also act as "scavenger antioxidants" by providing an electron to highly reactive free radicals and quenching the radicals' activity by delocalizing ion-paired electrons on the phenol ring (Singh & Basu, 2012). Until recently, phenolic compounds were thought to be non-nutritive; however, it is reported that high levels of phenolics, especially tannins, can pose negative repercussions on iron absorption by inhibiting and blocking the digestive enzymes in the gastrointestinal tract (Singh & Basu, 2012).

The provision of many nutrients and bioactive compounds and their preventive role in Non-communicable diseases make them an essential component of the diet. The secondary metabolites have also been proven to be anti-cancer against many cancers (Basker et al., 2010). The presence of lycopene in the chickpea has been reported to have a role in reducing the risk of prostate cancer. Carotenoids and isoflavone biochanin in peas have been found to reduce the risk of lung cancer and suppress cell growth in stomach cancer. At the same time, saponins help suppress pre-neoplastic colon lesions (Mittal et al., 2009). Pulses contain high levels of amylases, resistant starch that is antidiabetic, reduce the risk of thrombosis in people with diabetes, help regulate serum glucose, and improve insulin sensitivity and glucose tolerance, thereby reducing the risks of diabetes, cancers, and cardiovascular diseases (Basker et al., 2016; Pittaway et al., 2007; Jenkin et al., 2012; and Osorio-Diaz et al., 2008). These fibre-rich pulses can play an essential role in lowering plasma cholesterol and BMI while promoting faster satiety. The β-carotene increases folic acid uptake, reducing homocysteine concentrations by 13.4-51.7% (Crujeiras et al., 2007; Jukanti et al., 2012).

Owing to their rich nutrient composition, the presence of some plant components can affect the bioavailability of nutrients, specifically the micronutrients. The in vitro bio-accessibility of minerals depends upon the mineral and the type of food matrix. Generally, pulses are the best source of iron and zinc (Joanna and Zhigniew, 2011). Still, the limited bio-availability has been attributed to the presence of phytic acid that reduces their bioavailability to 5-15% (Das et al., 2011), which is challenging from a nutritional perspective, specifically in the context of developing countries like Pakistan which already has a serious problem of undernutrition, anemia, and other nutritional deficiency disorders.

The phytic acid is a myoinositol, 1, 2, 3, 4, 5, 6-hexyl hexakis dihydrogen phosphate. It is the primary storage form of phosphorus, comprising 1-5% by weight in cereals (Vats & Banerjee, 2004), and represents 50-85% of the total phosphorus in plants (Reddy et al., 1982). During the ripening period, it is rapidly accumulated and stored in the globoid crystal within the protein bodies (Erdman, 19790. Monogastric animals and humans cannot metabolize phytic acid due to the lack of sufficient phytases in the GI tract (Wodzinski & Ullah, 1996; Boling et al., 2000; Singh et al., 2011) and bind to iron, zinc, and calcium as insoluble salts making them unavailable (Urbano et al., 2000 and Feil, 2000). The post-harvest phytic acid strategy will help ensure enhanced micronutrient availability. The conventional methods of phytic acid reduction, such as soaking and cooking, have been shown to degrade phytic acid. However, an in-depth analysis of domestic processes needs to be explored. The current study aimed to analyze the effects of conventional domestic techniques on the phytate degradation and mineral solubility of the commonly cultivated pulses, which are widely used in Pakistan before and during cooking.

2. Materials and Methods

2.1. Sample Collection

The sample was comprised of pulses, namely Red Lentils (*Lens culinaris*), Split mung (*Vigna radiata*), urad (*Vigna mungo*), and Split Bengal gram (*Cicer arietinum*), procured from the agricultural fields of the Nuclear Institute of Food & Agriculture and the Agricultural Research Institute, Tarnab, Peshawar.

2.2. Sample Preparation

The four types of pulses and eight processing methods are used in the preparatory and cooking methods applied to them. These categories were viz, i) Raw, ii) 1-hour soaking, iii) 6-hour soaking, iv) overnight soaking, v) boiling until the whole water is absorbed, vi) boiling until the sample is tender, vii) Pressure cooking with soda, and viii) Pressure cooking without soda. The samples in the first category were kept raw (controlled) for ease of comparison.

2.3. Types of Samples

2.3.1. Raw Pulses

The raw seeds were mixed thoroughly, and 12 samples were used to estimate the phytic acid content of raw seeds in triplicate. The pulses were grounded before phytic acid determination. The grinding was done with a mechanical grinder.

2.3.2. One (1) Hour -Soaked Pulses

The seeds of the respective pulses were soaked by submerging them in tap water in a container for analysis for one hour. The excess water was removed with the help of a sieve, and then the adherent moisture was removed by gently rolling them on a thick absorbent cloth. Afterwards, sieved samples were placed in an oven to remove excess moisture at 70 degrees Celsius. After drying, the samples were then subjected to chemical analyses.

2.3.3. Six (6) Hour-Soaked Pulses

As the study is based on preparatory and cooking methods used commonly in Pakistan, all the pulses were soaked for 6 hrs, which is also a standard pre-cooking procedure in Pakistani cooking practices. The pulses were soaked for 6 hrs, and excess water was drained through a sieve and then placed in an oven to remove the excess moisture. After drying, the samples were ground with the help of an electric grinder. The samples were then subjected to different lab analyses.

2.3.4. Overnight Soaking

Another common practice in Pakistani households is the overnight steeping of pulses. All the pulses were soaked overnight in tap water, sieved in the morning, oven-dried, and subjected to laboratory analyses.

2.3.5. Boiling Until the Whole Moisture was Fully Absorbed

Boiling is the most common cooking method in Pakistan; the samples were boiled at and above 100°C until the sample was tender and the water was completely absorbed. The samples were then placed in an oven to remove the excess moisture. After drying, the samples were ground with the help of an electric grinder and stored in air-tight jars for analysis.

2.3.6. Boiling Until the Sample Was Tender

Boiling in sufficient water is also another common practice, so the samples were boiled in boiling water at 100°C until tender. Afterwards, the excess water was removed by sieving and oven-dried, ground, kept in air-tight jars, and tested for different parameters.

2.3.7. Pressure Cooking with Bicarbonate Soda

Pulses were pressure-cooked by adding a pinch of bicarbonate soda to tap water. The water was filtered with the help of a sieve. The excess water was drained, and the samples were placed in an oven until a static weight. After drying, the samples were ground, stored, and analyzed.

2.3.8. Pressure Cooking Without Bicarbonate Soda

Pulses were pressure-cooked each time per the manufacturer's directions. The cooled samples were drained in a sieve and oven-dried for laboratory analyses.

2.4. Determination of Phytic Acid

The phytic acid content of the samples was determined using the method of Haug W and Lantzch H (1983) under the principle of spectrophotometry.

2.4.1. Principle

The sample extract, prepared with 0.2 NH₄Cl, was heated with an acidic Iron (III), or ferric iron (Fe³⁺) solution of known iron content. The decrease in iron content, determined colorimetrically using 2,2'-bipyridine in the supernatant, was used to measure the phytate phosphorus.

2.4.2. Phytate Reference Solutions

The sodium salt of phytic acid type V, with 97% purity and containing approximately 15% water, was obtained from Sigma (NO.P–5756). It was used without further purification. The actual content of phytate needed to be determined once for each new purchase using a direct method. Stock solutions were prepared by dissolving 0.15 g of sodium phytate in 100.0 mL of distilled water. Since phytase was absent, these stock solutions remained stable.

2.4.3. Preparation of a Series of Reference Solutions and a Standard Curve

A series of reference solutions was prepared by diluting stock solutions with HCl within a range of 3 to 30 mg/mL phytate phosphorus. The dilution involved adding approximately 1.2–11.7 mL of stock solution to 100.0 mL of final volume. The HCl concentration in the reference solution was adjusted to 0.2 N. Reference Solutions Concentrations at $3\mu g/mL$, $6\mu g/mL$, $9\mu g/mL$, $12\mu g/mL$, $15\mu g/mL$, $18\mu g/mL$, $21\mu g/mL$, $24\mu g/mL$, $27\mu g/mL$, $30\mu g/mL$ were prepared and respective calculations were done for the preparation of standard curve for the estimation within the samples. Iron standard solution was prepared by dissolving 0.2 g of ammonium iron (III) sulphate 12 H₂O (Merck Art. 3776) in 100.0 mL 2N HCl and making up to 1000.0 mL with distilled water. The 2,2'-bipyridine solution was prepared by dissolving 10.0 g of 2,2'-bipyridine (Merck art. 3098) and 100.0 mL of thioglycolic acid (Merck art. 700) in distilled water and making up to 1000.0 mL. The Standard Curves were developed by taking 0.5 mL of each standard solution (Phytic acid and iron

solutions) in a test tube. After heating in a boiling water bath for 30 minutes (with test tubes remaining covered with a stopper for the first 5 minutes). The tubes are cooled under tap water and then iced water for 15 minutes. Upon attaining room temperature, 2.0 mL of 2,2'-bipyridine solution was added. The optical density was measured within 0.5-1 minute at 519 nm with the help of a spectrophotometer. The optical density against concentration was plotted as the standard curve. It was linear between 0 & 35 mg/g concentrations of phytic acid.

2.5. Quantification of Phytic Acid in Pulses

A 0.06g sample was placed in three different tubes. About 10.0 mL of 0.2 N HCl was added to each tube and shaken using an electric shaker for 30 minutes. Subsequently, 0.5 mL of the supernatant was transferred into another tube, and 0.1 mL of Fe³⁺ (ammonium iron (III) sulfate) was added to each tube. The samples were placed into a boiling water bath for 30 minutes at 37°C. The samples were then removed from excess water with the help of a sieve. After cooling the tubes in an ice bath for 15 minutes, the samples were allowed to adjust to room temperature. A 2.0 mL of 2,2'-bipyridine was added to each tube and was immediately measured for optical density (O.D.) using a spectrophotometer at 519 nm. The following formulae (i and ii) were used to determine the phosphorus phytate and percentage of phytic acid.

Y= 0.0156X + 1.2811 ----- (i)

X = Y - 1.2811 / 0.0156 (regression line)

Where; X = Conc.of Phosphorus phytate in μ g/ mL; Y = Optical Density or Absorbance; Wt. = weight of sample taken **Phytic Acid = phytate phosphorus x 4.97** / **1000 x 0.06** ----- (ii)

2.6. Statistical Analysis

Data were assessed for mean, standard deviation, percent decline, and analysis of variance (ANOVA) on IBM SPSS version 19. The Duncan multiple range test was used to separate the means. Dunnett's multiple ANOVA comparisons were used to create confidence intervals for differences between the mean phytate content of each processing method against the mean of raw pulses as a control. Significance was accepted at $P \le 0.05$.

3. Results and Discussion

3.1. Effect of Processing Techniques on the Phytate Content (g/100g dry weight) of the Pulses

The phytic acid content and percentage degradation in Red Lentils (Lens culinaris) under different domestic processing methods (Table 1) showed that raw red lentils had the highest phytic acid content ($0.76\% \pm 0.01$), serving as the baseline (0% degradation). Soaking had varying effects on phytic acid reduction. A 1-hour soak reduced it to $0.59\% \pm 0.02$ (22.37% degradation), while a 6-hour soak led to a lesser reduction at $0.71\% \pm 0.02$ (6.58% degradation). However, a 12hour soak was significantly more effective, reducing phytic acid to $0.41\% \pm 0.07$ (46.05% degradation). This suggests that prolonged soaking enhances enzymatic breakdown and mineral availability. Boiling methods also contributed to phytic acid degradation. Boiling until tenderization reduced phytic acid to $0.38\% \pm 0.00$ (50.00% degradation) while boiling until moisture absorption slightly increased phytic acid content to 0.43 ± 0.04 (43.42% degradation) indicates that prolonged heat exposure and water absorption influence the degradation efficiency. Pressure cooking showed moderate effectiveness. Cooking with soda reduced phytic acid to $0.40\% \pm 0.11$ (47.37% degradation), while pressure cooking without soda resulted in slightly lower degradation at 39.47% ($0.46\% \pm 0.01$). It reflects that sodium bicarbonate slightly enhances phytic acid breakdown, but may not be as effective as boiling until tenderization. This study suggests that boiling until tenderization was the most effective method, followed by 12-hour soaking and pressure cooking with soda. These findings are similar to the degradation of phytates in Lens culinaris reported by Hefnawy (2011), emphasizing the importance of using appropriate processing techniques to enhance the nutritional quality and mineral bioavailability of Red Lentils. Table 1 Phytic Acid Contents & % Degradation in Lens culinaris (Red Lentils)

S. No.	Processing Method	Mean % ± SD	P Values	% Degradation
1	Raw Lens culinaris	$0.76{\pm}0.01$		0.0
2	1-hour Soaked Lens culinaris	0.59 ± 1.02	0.045ª	22.37
3	6-hour Soaked Lens culinaris	0.51 ± 2.02	0.072,	6.58
4	12-hour-Soaked Lens culinaris	$0.41{\pm}1.07$	$0.046^{\rm abc}, 0.044^{\rm abc},$	46.05
5	Boiling till Tender Lens culinaris	0.38 ± 0.75	0.038 ^a , 0.039 ^{abc} , 0.043 ^{abcd}	50.00
6	Boiling till Moisture Absorption Lens culinaris	0.43 ± 2.04	$0.006^{a}, 0.021^{ab}, 0.001^{abc}, 0.042^{abcd}$	43.42
7	Pressure Cooking with Soda Lens culinaris	0.40±0.11	$0.007^{a}, 0.026^{ab}, 0.006^{abcd}.$ $0.002^{abcds}, 0.056^{abcde}$	47.37
8	Pressure Cooking without Soda Lens culinaris	0.46±0.01	$0.003^{a}, 0.0048^{ab}, 0.043^{abc}, 0.032^{abcd}, 0.009^{abcde}, 0.029^{abcde}$	39.47

The letters in the superscript indicate the significantly different values at P \leq 0.05

The phytic acid content and percentage degradation (Table 2) in Split mung pulse (*Vigna radita*) under different domestic processing methods showed that raw split mung dal contained the highest phytic acid content (1.90% \pm 0.00), serving as

the baseline (0% degradation). Soaking significantly reduced phytic acid levels, with 1-hour soaking decreasing it to 0.64 \pm 0.01 (66.3% degradation), 6-hour soaking lowering it to 0.77 \pm 0.09 (59.5%), and 12-hour soaking reducing it further to 0.66% \pm 0.01 (65.3%). It indicates that short-duration soaking (1 hour) is more effective in degrading phytic acid than prolonged soaking. Boiling methods showed moderate effectiveness, with boiling until tenderization reducing phytic acid to 0.83% \pm 0.01 (56.3%) and boiling until complete moisture absorption lowering it to 0.98% \pm 0.01 (48.4%). These results suggest that heat contributes to degradation, and moisture content is crucial to efficiency. Among all methods, pressure cooking without soda was the most effective, reducing phytic acid to 0.50% \pm 0.00 (73.7% degradation), reflecting that high-pressure cooking enhances the breakdown of phytic acid more than other methods. In contrast, pressure cooking with soda resulted in a slightly lower degradation rate of 53.2% (0.89% \pm 0.00), suggesting sodium bicarbonate may interfere with phytic acid breakdown. These findings are similar to the results of Kemal et al. (2025), who found a similar reduction in percent phytates, specifically when cooked through autoclaving.

S.No.	Processing Method	Mean % ± SD	P values	% Degradation
1	Raw Vigna radita	1.90±0		0.0
2	1-hour-soaked Vigna radita	$0.64{\pm}0.01$	0.000^{a}	66.3
3	6-hour-soaked Vigna radita	0.77 ± 0.09	$0.016^{a}, 0.042^{b}$	59.5
4	12-hour-soaked Vigna radita	0.66 ± 0.01	$0.003^{a}, 0.028^{ab}, 0.045^{abc}$	65.3
5	Boil till Tenderized, Vigna radita	0.83 ± 0.01	0.043 ^a , 0.038 ^{ab} , 0.026 ^{abc} , 0.009 ^{abcd}	56.3
6	Boiling till Moisture Absorption Vigna radita	0.98±0.01	$0.045^{a}, 0.023^{ab}, 0.008^{abc}, 0.045^{abcd}, 0.041^{abcde}$	48.4
7	Pressure Cooking with Soda Vigna radita	0.89±0.00	$0.026^{a}, 0.019^{ab}, 0.0032^{abc}, 0.031^{abcd}, 0.009^{abcde}$	53.2
8	Pressure Cooking without Vigna radita	0.50±0.00	$0.000^{a}, 0.001^{ab}, 0.000^{abc}, 0.019^{abcd}, 0.032^{abcde}, 0.005^{abcdef}, 0.007^{abcdefg}$	73.7

Table 2: Phytic Acid Content & % Degradation in Vigna radita (\$	Split Mung pulse)
--	-------------------

The letters in the superscript indicate the significantly different values at P \leq 0.05

Table (3) results illustrate the phytic acid content and percentage degradation in Urad pulse (*Vigna mungo*) after different domestic processing methods. The raw mash ki dal/urad dal contained the highest phytic acid content $(1.90\% \pm 0.01)$, serving as the baseline (0% degradation), meaning it retained its full antinutritional potential. Soaking significantly reduced phytic acid levels, with 6-hour soaking decreasing it to $0.70\% \pm 0.08$ (63.2% degradation), while 12-hour soaking further lowered it to $0.36\% \pm 0.00$ (81.1% degradation), indicating that prolonged soaking enhances enzymatic activity, improving nutrient bioavailability. Boiling methods also proved effective in boiling until tenderization, reducing phytic acid to $0.54\% \pm 0.01$ (71.6%), and boiling until complete moisture absorption, lowering it further to $0.40\% \pm 0.03$ (78.9%). This suggests that cooking is crucial in breaking down phytic acid and improving digestibility. Pressure cooking without soda was the most effective method, reducing phytic acid to $0.33\% \pm 0.00$ (82.6% degradation), meaning that high-temperature, high-pressure cooking maximizes phytic acid breakdown. In contrast, pressure cooking with soda led to a slightly lower degradation of 76.8% ($0.44\% \pm 0.00$), suggesting that the presence of sodium bicarbonate might interfere with enzymatic activity or alter pH conditions, affecting phytic acid breakdown. The current study's findings strongly agree with the results of Huma et al. (2008), who reported a similar reduction in the phytate concentrations in urad dal under different cooking procedures.

	Table 3: Ph	ytic Acid (Content &	% Degr	adation in	Vigna n	<i>ungo</i> (ura	d dal)
--	-------------	-------------	-----------	--------	------------	---------	------------------	--------

S.No.	Processing Method	Mean % ± SD	P value	% Degradation
1	Raw Vigna mungo	1.90 ± 0.01		0.0
2	6-hour-soaked Vigna mungo	0.70 ± 0.08	0.000^{a}	63.2
3	6-hour-soaked Vigna mungo	0.36 ± 0.00	$0.002^{a}, 0.001^{ab}$	81.1
4	12-hour soaked Vigna mungo	0.54 ± 0.01	$0.000^{a}, 0.004^{ab}, 0.009^{abc}$	71.6
5	Boil till tender Vigna mungo	0.44 ± 0.01	$0.000^{a}, 0.002^{ab}, 0.019^{abc}, 0.036^{abcd}$	76.8
6	Boiling till the moisture content was fully absorbed	0.40 ± 0.03	$0.000^{a}, 0.008^{ab}, 0.003^{abc}, 0.0026^{abcd}, 0.049^{abcde}$	78.9
7	Pressure cooking with soda (sodium bicarbonate)	0.44 ± 0.00	$0.000^{a}, 0.002^{ab}, 0.006^{abc}, 0.038^{abcd}, 0.68^{abc}, 0.051^{abce}$	76.8
8	Pressure cooking without soda	0.33 ± 0.00	$0.000^{a}, 0.000^{ab}, 0.002^{abc}, 0.002^{abcd}, 0.019^{abcde}, 0.013^{abcdef}, 0.017^{abcdefg}$	82.6

The letters in the superscript indicate the significantly different values at $P \le 0.05$.

The Phytic Acid Content & % Degradation in *Cicer arietinum* (Table 4) presents the effect of different processing methods on the phytic acid content. The raw sample has the highest phytic acid content at 0.71 ± 0.01 , serving as the reference with 0% degradation. Soaking treatments show significant reductions: 1-hour soaking decreases phytic acid to 0.41 ± 0.10 (42.3% degradation), 6-hour soaking to 0.49 ± 0.00 (31.0%), and 12-hour soaking to 0.65 ± 0.03 (8.5%). Boiling methods also lower phytic acid content, with boiling till tenderized, reducing it to 0.43 ± 0.08 (39.4%) and boiling until complete moisture absorption, decreasing it to 0.49 ± 0.03 (31.0%). Pressure cooking with soda results in 0.60 ± 0.09 (15.5%) degradation, whereas pressure cooking without soda leads to 0.65 ± 0.00 (8.5%) degradation. The findings indicate that 1-hour soaking is the most effective method, reducing phytic acid content by nearly half (42.3%), followed by boiling till tender (39.4%) and 6-hour soaking (31.0%). Mamiro et al. (2017) studied the effects of cooking methods on the antinutritional factors of different *Phaseolus vulgaris* (split pea dal) cultivars and concluded positive effects of steeping and cooking on the phytate degradation.

S. No.	Processing	Mean ± SD	P values	% Degradation
1	Raw	0.71 ± 0.01		0.0
2	1-hour soaking Cicer arietinum	0.41 ± 0.10	0.000a	42.3
3	6-hour soaking Cicer arietinum	0.49 ± 0.00	0.000a, 0.002ab	31.0
4	12-hour soaking Cicer arietinum	0.65 ± 0.03	0.023a, 0.000ab, 0.001abc	8.5
5	Boiling till tender Cicer arietinum	0.43 ± 0.08	0.000a, 0.045ab, 0.023ab, 0.000acd	39.4
6	Boiling till the whole moisture content was fully absorbed <i>Cicer arietinum</i>	0.49 ± 0.03	0.000a, 0.021ab, 0.003abd, 0.019	31.0
7	Pressure cooking with soda Cicer arietinum	0.60 ± 0.09	0.006a, 0.000ab, 0.001abc, 0.001abcd, 0.003abcde	15.5
8	Pressure cooking without soda Cicer arietinum	0.65 ± 0.00	0.023a, 0.003ab, 0.008abc, NS, 0.006abce, 0.052abc, 0.049abce	08.5

The letters in the superscript indicate the significantly different values at P \leq 0.05

3.2: Percent Phytic Acid Content & Percent Phytate Degradation in Pulses

The data regarding the phytic acid content and its percentage degradation in various pulses (Fig. 1-7), including *Lens culinaris, Vigna radiata, Vigna mungo*, and *Cicer arietinum* showed that raw *Vigna radiata* and *Vigna mungo* had the highest percent phytic acid, followed by *Lens culinaris* and *Cicer arietinum*. The percent degradation in 1-hour-soaked pulses (Fig. 1) was the highest percentage of degradation that occurred in *Vigna radiata*, followed by *Vigna mungo*. Among the pulses, *Vigna radiata* exhibited the highest phytic acid degradation of 66.3 percent, with a remaining phytic acid content of 0.64 percent. *Vigna mungo* followed closely, with a 63.2 percent degradation and a final phytic acid content of 0.70 percent. *Lens culinaris* showed a lower degradation percentage of 22.37 percent, retaining 0.59 percent phytic acid. *Cicer arietinum* had the lowest phytic acid content post-soaking at 0.41 percent, with a degradation percentage of 42.3 percent. The results indicate that soaking pulses for one hour significantly reduces phytic acid levels, though the extent of reduction varies among different pulses.





Figure (2) shows the phytic acid content and its percentage degradation in pulses soaked for 6 hours. Among the samples, mash ki dal/urad dal exhibited the highest phytic acid degradation at 81.1%, reducing the phytic acid content to 0.36%.

This was followed by Split mung dal with a 59.5% degradation and a final phytic acid content of 0.77%. Split Bengal gram showed moderate degradation at 31.0%, with a remaining phytic acid content of 0.49%. Red Lentils had the lowest reduction in phytic acid levels, with only 6.58% degradation, resulting in a phytic acid content of 0.71%. These findings indicate that 6-hour soaking is particularly effective in reducing phytic acid in mash ki dal/urad dal and Split mung dal. In contrast, its impact on Red Lentils is relatively limited.



Figure 2: Phytic Acid Degradation in 6-Hour-Soaked Pulses

Figure (3) presents the percentage degradation of phytic acid in various pulses after 12 hours of soaking. Mash ki dal/urad dal exhibited the highest phytic acid degradation at 71.6%, reducing its phytic acid content to 0.54%. Split mung dal followed with a degradation rate of 65.3%, leaving 0.66% of phytic acid. Red Lentils showed a moderate degradation of 46.05%, with a remaining phytic acid content of 0.41%. In contrast, Split Bengal gram had the lowest degradation rate at only 8.5%, retaining most of its phytic acid content at 0.65%.



Figure 3: Phytic Acid Degradation in 12-Hour-Soaked Pulses

Figure (4) illustrates the percent degradation of phytic acid in different pulses after boiling until tender. The results show that boiling significantly reduces phytic acid content. Among the samples, mash ki dal/urad dal exhibited the highest degradation at 76.8%, followed by Split mung dal at 56.3% and Red Lentils at 50.0%. This indicates that these pulses benefit greatly from boiling in terms of reduced antinutrient content. Split Bengal gram showed the lowest phytic acid degradation at 39.4%, suggesting that it retains more phytic acid even after boiling. This could indicate the need for additional processing techniques such as soaking or fermentation to further reduce its phytic acid content. Overall, boiling proves to be an effective method for reducing phytic acid in most legumes, thereby improving their nutritional quality and mineral absorption potential.



Figure 4: Phytic Acid Degradation in Boiling till Tenderized Pulses

Figure (5) represents the percentage phytic acid degradation in various pulses after boiling until complete water drying. Among the samples, mash ki dal/urad dal exhibited the highest degradation at 78.9%, with phytic acid content of 0.40%. The Split mung dal followed, with 48.4% degradation and 0.98% phytic acid content. Red Lentils had a moderate degradation rate of 43.4%, containing 0.43% phytic acid. Meanwhile, Split Bengal gram exhibited the lowest degradation at 31.0%, with a content of 0.49%. The data indicates that boiling pulses until complete water absorption significantly reduces phytic acid content, enhancing their nutritional value.



Figure 5: Phytic Acid Degradation in Boiling till Water Absorbs Pulses

The data on phytic acid degradation in various pulses (Figure 6) was subjected to pressure cooking with added soda. Among the samples, mash ki dal/urad dal + soda exhibited the highest degradation of 76.8%, reducing its phytic acid content to 0.44%. Split mung dal + soda, followed with a 53.2% degradation, retaining 0.89% phytic acid. Red Lentils + soda showed a moderate degradation of 47.3%, with a reduced phytic acid content of 0.40%. Meanwhile, Split Bengal gram + soda had the lowest degradation rate at 15.5%, maintaining a phytic acid content of 0.60%. These results highlight that pressure cooking with soda effectively reduces phytic acid levels. However, the extent of degradation varies among different pulses, with mash ki dal/urad dal showing the most significant reduction.



Figure 6: Phytic Acid Degradation in Pressure Cooking with Bicarbonate Soda

The phytic acid degradation observed in different pulses during pressure cooking without soda (Fig. 7) indicated that *Vigna mungo* exhibited the highest degradation at 82.6%, followed by *Vigna radiata* at 73.7%. Red Lentils showed a moderate reduction of 39.4%, whereas Split Bengal gram had the lowest degradation at 8.5%. This data highlights the variation in phytic acid reduction among pulses when cooked under pressure without soda.



Figure 7: Phytic Acid Degradation in Pulses during Pressure Cooking without Soda

The mean overall percent degradation of phytates in pulses under various domestic processing methods (Fig. 8) showed that soaking pulses for 1 hour led to a 40.6% reduction in phytic acid, while soaking for 6 hours resulted in a higher degradation at 44.5%. However, an extended 12-hour soak showed a slightly lower reduction of 39.5%, possibly due to a saturation effect. Boiling methods also played a crucial role; pulses boiled till tender resulted in a mean 43% reduction, whereas boiling until water absorption resulted in only 35.7% degradation. Pressure cooking showed varying effects depending on the use of soda. Cooking with soda led to a 36.3% reduction; without soda, the degradation was slightly higher at 40%. This indicates that while pressure cooking is practical, the addition of soda does not necessarily enhance phytic acid breakdown significantly. These findings are almost similar to many such studies (Mamiro et al., 2017; Iorgyer et al., 2009; Gibson et al., 2010; Kemal et al., 2025; Das et al., 2011)



Figure 8: Mean Effects of Different Processing on Percent Phytate Degradation

Conclusion

The current study concludes that conventional soaking and cooking methods effectively reduce phytate levels in locally consumed pulses. This reduction enhances the bioavailability of certain minerals, which are otherwise bound by phytates and rendered inaccessible for metabolic processes. These preparation practices are particularly beneficial for most Pakistani households, where pulses are a dietary staple consumed frequently, often on a daily basis, and play a significant role in mineral intake.

Acknowledgment

The authors acknowledge the technical and laboratory assistance of the staff of the Food Science Division, Nuclear Institute of Food & Agriculture (NIFA), Peshawar.

Conflict of Interest

Not applicable.

References

Ayub Agricultural Research Institute. Pulses. https://aari.punjab.gov.pk/pulses_cropvarities

- Baskar, V., Park, S. W., and Nile, S. H. (2016). An update on potential perspectives of glucosinolates on protection against microbial pathogens and endocrine dysfunctions in humans. Crit. Rev. Food Sci. Nutr. 56, 2231–2249. doi: 10.1080/10408398.2014.910748
- Bessada, S. M., Barreira, J. C., & Oliveira, M. B. P. (2019). Pulses and food security: Dietary protein, digestibility, bioactive and functional properties. Trends in Food Science & Technology, 93, 53-68.
- Boling SD, Douglas MW, Johnson ML, Wang X, Parsons CM, Koelkebeck KW. The effects of dietary available phosphorus levels and phytase performance of young and older laying hens. Poult Sci. 2000; 79:224 230. doi: 10.1093/ps/79.2.224.
- Chibbar, R. N., Ambigaipalan, P., and Hoover, R. (2010). Molecular diversity in pulse seed starch and complex carbohydrates and its role in human nutrition and health. Cereal Chem. 87, 342–352. doi: 10.1094/CCHEM-87-4-0342
- Crujeiras, A. B., Parra, D., Abete, I., and Martínez, J. A. (2007). A hypocaloric diet enriched in legumes specifically mitigates lipid peroxidation in obese subjects. Free Radic. Res. 41, 498–506. doi: 10.1080/10715760601131935
- Das A, Raychaudhuri U, Chakraborty R. Cereal based functional food of Indian subcontinent: a review. J Food Sci Tech. 2011 doi: 10.1007/s13197-011-0474-1
- Escudero, N. L., Zirulnik, F., Gomez, N. N., Mucciarelli, S. I., Mucciarelli, S. I., & Giménez, M. S. (2006). Influence of a protein concentrate from Amaranthus cruentus seeds on lipid metabolism. Experimental Biology and Medicine, 231(1), 50-59

Feil B. Phytic acid. J New Seeds. 2001; 3:1–35. doi: 10.1300/J153v03n03_01.

- Flight, I., & Clifton, P. (2006). Cereal grains and legumes in the prevention of coronary heart disease and stroke: a review of the literature. European Journal of Clinical Nutrition, 60(10), 1145-1159.
- Friedman, M. (1996). Nutritional value of proteins from different food sources. A review. Journal of Agricultural and Food Chemistry, 44(1), 6-29.

- Gibson RS, Bailey KB, Gibbs M, Ferguson EL (2010). A review of phytate, iron, zinc, and calcium concentrations in plant-based complementary foods used in low-income countries and implications for bioavailability. Food Nutr. Bull. 31: S134-S146.
- Gowda, C. L., Chaturvedi, S. K., Gaur, P. M., Sameer Kumar, C. V., and Jukanti, A. K. (2015). "Pulses research and development strategies for India," in Pulses Handbook (2015), 17–33.
- Hefnawy H (2011). Effect of processing methods on nutritional composition and antinutritional factors in lentils (Lens culinaris), Annals of Agricultural Sciences, 56(2): 57-61
- Huma, N., Anjum, M., Sehar, S., Issa Khan, M. and Hussain, S. (2008), "Effect of soaking and cooking on nutritional quality and safety of legumes", Nutrition & Food Science. 38(6): 570-577. <u>https://doi.org/10.1108/00346650810920187</u>
- Iorgyer MI, Adeka IA, Ikondo ND, Okoh JJ (2009). The impact of boiling periods on proximate composition and level of some antinutritional factors in pigeon pea (Cajanus cajan) seeds. Prod. Agric. Technol. 5(1):92-102.
- Jenkins, D. J., Kendall, C. W., Augustin, L. S., Mitchell, S., Sahye-Pudaruth, S., Mejia, S. B., et al. (2012). Effect of legumes as part of a low glycemic index diet on glycemic control and cardiovascular risk factors in type 2 diabetes mellitus: a randomized controlled trial. Arch. Intern. Med. 172, 1653–1660. doi: 10.1001/2013.jamainternmed 70
- Joanna S, Zbigniew K. Evaluation of the content and bioaccessibility of iron, zinc, calcium and magnesium from groats, rice, leguminous grains and nuts. J Food Sci Tech. 2011 doi: 10.1007/s13197-011-0535-5
- Jukanti, A. K., Gaur, P. M., Gowda, C. L. L., and Chibbar, R. N. (2012). Nutritional quality and health benefits of chickpea (Cicer arietinum L.): a review. Br. J. Nutr. 108, S11–S26. doi: 10.1017/S0007114512000797
- Kalidass, C., & Mohan, V. R. (2011). Genetic resources of under exploited legumes/tribal pulses of Western Ghats, Tamil Nadu. Journal of Economic Taxonomy Botany, 35, 241-248
- Kemal M, Tilahun A. Teka, Dibaba K, Urugo M M (2025)Effects of processing methods on nutrient and antinutrient composition, techno-functional and antioxidant properties of mungbean (Vigna radiata (L.) R. Wilczek) varieties. LWT, 222: 117663. <u>https://doi.org/10.1016/j.lwt.2025.117663</u>
- Khattab RY, Arntfield SD (2009). Nutritional quality of legume seeds as affected by some physical treatments 2. Antinutritional factors. Food Sci. Technol. 42(6):1113-1118
- Kouris-Blazos, A., & Belski, R. (2016). Health benefits of legumes and pulses with a focus on Australian sweet lupins. Asia Pacific Journal of Clinical Nutrition, 25(1), 1-17.
- Langyan S, Yadava P, Khan F N, Bhardwaj R, Tripathi K, Bhardwaj V, Bhardwaj R, Gautam R K, Kumar A(2022). Nutritional and Food Composition Survey of Major Pulses Toward Healthy, Sustainable, and Biofortified Diets. Frontiers in Sustainable Food Systems. 6. RL=https://www.frontiersin.org/journals/sustainable-foodsystems/articles/10.3389/fsufs.2022.878269. DOI=10.3389/fsufs.2022.878269
- Mal, B. (1992). Biodiversity utilization and conservation in underutilized plants: Indian perspective. Indian Journal of Plant Genetic Resources, 5(1), 1-22.
- Mamiro, P S, Mwanri, AW, Mongi, R J, Chivaghula, T J, Nyagaya M, Ntwenya (2017). Effect of cooking on tannin and phytate content in different bean (Phaseolus vulgaris) varieties grown in Tanzania. African Journal of Biotechnology. 16(20): 186-119. DOI: 10.5897/AJB2016.15657
- Oomah, B. D., and Mazza, G. (2000). Bioactive components of flaxseed: occurrence. Phytochem. Phytopharm. 106, 105–112.
- Osorio-Diaz, P., Agama-Acevedo, E., Mendoza-Vinalay, M., Tovar, J., and Bello-Perez, L. A. (2008). pasta added with chickpea flour: chemical composition, in vitro starch digestibility and predicted glycemic index pasta adicionada con harina de garbanzo: composición química, digestibilidad in vitro del almidóny predicción del índice glucémico. Cyta J. Food 6, 6–12. doi: 10.1080/11358120809487621
- Pittaway, J. K., Ahuja, K. D., Robertson, I. K., and Ball, M. J. (2007). Effects of a controlled diet supplemented with chickpeas on serum lipids, glucose tolerance, satiety and bowel function. J. Am. Coll. Nutr. 26, 334–340. doi: 10.1080/07315724.2007.10719620
- Reddy NR, Sathe SK, Salunkhe DK. Phytases in legumes and cereals. Adv Food Res. 1982; 82:1–92. doi: 10.1016/S0065-2628(08)60110-X
- Siddhuraju, P., Becker, K., & Makkar, H. P. S. (2000). Studies on the nutritional composition and antinutritional factors of three different germplasm seed materials of an under-utilized tropical legume, Mucuna p ruriens Var. U tilis. Journal of Agricultural and Food Chemistry, 48(12), 6048-6060.
- Siddhuraju, P., Becker, K., & Makkar, H. P. S. (2002). Chemical composition, protein fractionation, essential amino acid potential and antimetabolic constituents of an unconventional legume, Gila bean (Entada phaseoloides Merrill) seed kernel. Journal of the Science of Food and Agriculture, 82(2), 192-202

- Singh B, Kunze G, Satyanarayana T. Developments in biochemical aspects and biotechnological applications of microbial phytases. Biotechnol Mol Bio Rev. 2011; 6:69 87.
- Singh, U. B., Malviya, D., Singh, S., Singh, P., Ghatak, A., Imran, M., et al. (2021). Salt-tolerant compatible microbial inoculants modulate physio-biochemical responses enhance plant growth, Zn biofortification and yield of wheat grown in saline-sodic soil. Int. J. Environ. Res. Public Health 18, 9936. doi: 10.3390/ijerph18189936
- Soomro H, Soomro AH, Haider MS, Tarar OM (2020). Evaluation of nutritional profile of pulses consumed in Pakistan. Journal of Pure and Applied Agriculture (2020) 5(4): 27-33
- Suneja, Y., Grewal, S., Gupta, A., and Kaur, N. (2011). Levels of nutritional constituents and antinutritional factors in black gram (Vigna mungo L. Hepper). Food. Res. 44. 621–628. doi: 10.1016/j.foodres.2010.12.020
- Tharanathan, R. N., & Mahadevamma, S. (2003). Grain legumes—a boon to human nutrition. Trends in Food Science & Technology, 14(12), 507-518
- Ullah, A., Shah, T.M. & Farooq, M. Pulses Production in Pakistan: Status, Constraints and Opportunities. Int. J. Plant Prod. 14, 549–569 (2020). <u>https://doi.org/10.1007/s42106-020-00108-2</u>
- Urbano G, Lopez-Jurado M, Aranda P, Vidal-Valverde C, Tenorio E, Porres J (2000). The role of phytic acid in legumes: antinutrient or beneficial function? J Physiol Biochem.56:283 294. doi: 10.1007/BF03179796.
- Vats P, Banerjee UC. Production studies and catalytic properties of phytases (myo-inositol-hexakis-phosphate phosphohydrolases): an overview. Enzyme Microb Technol. 2004; 35:3–14. doi: 10.1016/j.enzmictec.2004.03.010.

Wodzinski RJ, Ullah AH. Phytase. Adv Appl Microbiol. 1996; 42:263 301. doi: 10.1016/S0065-2164(08)70375-7

Received: April 3rd, 2025.

Accepted: May 4th, 2025