



## Effect of Heat Treatment and Gamma Irradiation on Protein of Selected Millet Grains

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### *Authors' contributions*

*This work was carried out in collaboration among all authors. Author MS designed the study and managed the analyses of the study. Author TVH performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors KUD and NB managed the literature searches and corrected the thesis. All authors read and approved the final manuscript.*

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### ABSTRACT

The present investigation was conducted to find out the effects of heat or irradiation combined on protein. Sorghum, pearl millet, foxtail millet were used in the study. Whole (WC) and dehulled (DC) grains were treated either with heat (170°C) or irradiation at 1.0 kGy / 2.5 kGy and stored for 90 days. There was a significant ( $p < 0.05$ ) effect of treatments, storage and grain and their interactions on protein. The mean protein was 9.89 percent and there was a reduction in protein content of the grains due to treatments by 11.8% in DC and 8.8% in WC. Irradiation combination treatment could prevent the loss of protein in WC but not in DC over heat treatment. The mean loss of protein during storage was 3.56 percent. The loss of protein in heat treated grains was 6.0 and 5.4 percent in DC and WC. Heat treatment increased the losses by 6.0 and 5.4 percent which were reduced to 4.1 and 1.58 when irradiation was combined with 1.0 kGy dose in DC and WC. The losses reduced with dosages of 2.5 kGy to 2.97 and 2.5 percent in DC and WC.

*Keywords: Irradiation; whole grains; dehulled grains; protein.*

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## 1. INTRODUCTION

Irradiation is one of the treatment technologies currently available for the inactivation of microorganisms, and it has proven to be effective in ensuring food safety and extending the shelf life of food [1]. In 1999, the Food and Agriculture Organization of the United Nations (FAO)/International Atomic Energy Agency [2] (IAEA)/World Health Organization (WHO) Study Group and International Conference advocated that foods irradiated at any dose appropriate to meet technological goals should be both safe and nutritionally adequate. The irradiation process is, therefore, useful and desirable as an alternative in the preservation and processing of various fresh, perishable, and high-protein foods, with or without chemical additives or biological controls [3,4].

During storage there may be nutritional changes in cereals, although for dry grains these changes will be small even over a period of several months. If grains are stored with more than ideal water content, grains and microbial amylases can begin to break down the starch, leading to deterioration in grain quality. Several drying methods are used to reduce the moisture content to desirable levels. The effect of heating before irradiation is additive or slightly greater than the additive, the ionizing radiation applied before heating is highly synergistic in the inactivation of bacterial spores [5]. High temperatures applied before radiation sensitize insects to radiation and therefore allow the use of low doses [6]. The

chemical structure of irradiated food is less modified than heat-treated one and this technique avoids the use of potentially harmful chemicals [7].

## 2. MATERIALS AND METHODS

### 2.1 Procurement of Raw Materials

Sorghum and foxtail millet grains were collected from RARS, Nandyal, ANGRAU and pearl millet from RARS, Palem, PJTSAU.

### 2.2 Processing of the Grains

All the grains were cleaned to remove foreign material and stored in polythene bags until used under dry and cool conditions away from insects and pests.

### 2.3 Dehulling

The grains were dehulled in an abrasive dehuller (Gurunanak Engineering Co, Hyderabad) up to 17 percent removal of bran. Roller abrasive dehulling machine was used for dehulling the grains by adjusting the time. The dehulled grains were collected through bottom opening of the dehuller along with hull. The bran was separated from the dehulled grain through the sieve attached to the dehuller. The separated grain was further winnowed to ensure grain without any adhering bran (Plate 1).



Plate 1. Abrasive dehuller

**Grain treatment:****a) Heat treatment:**

In the present experiment electric rotary dryer (S k Engineering, New Delhi) was used which can be operated continuously for large quantity of grain. It has provision for adjusting the temperature and the rpm. For the present study 10 kg of whole and dehulled grains of all three millets were exposed to heat treatment at a temperature of 150-170°C for 1.5 min at 300 rpm. The treated grains were then packed in polythene pouches in desired quantity of 500 g for further use.

**b) Gamma irradiation:**

Irradiation is one of the processing technologies currently available for the inactivation of microorganisms, and it has proven successful in ensuring the safety and extending the shelf life of foods [1]. The millet grains were irradiated using cobalt – 60 gamma sources. Two different dosages 1.0 kGy and 2.5 kGy were used in the present study based on the previous studies. Grains of 500 g were packed in polythene pouches and exposed to the irradiation as guided by technical staff of the university.

Sorghum, pearl millet and foxtail millet grains of whole and dehulled grains were treated either by heat alone or heat and irradiation combination and stored for 90 days at ambient temperature (Table 1).

**Estimation of Protein:** 0.1 g of sample was weighed into a kjeldhal flask, 0.2 g of the digestion mixture ( $K_2SO_4$ , and  $CU_2SO_4 \cdot 5H_2O$ ) was added and digested in Kelplus – kjeldhal digester with 20 ml of concentrated  $H_2SO_4$  until all the organic matter was oxidized and uniform greenish – blue digest was obtained. The digest was cooled and volume was made up with 100 ml distilled water. An aliquot of 5 ml was taken

for steam distillation in kelpus distillation unit with excess of 40% NaOH solution (10 ml). The liberated ammonia was observed in 100ml of 2% boric acid containing a few drops of mixed indicator. This was titrated against N/70 HCl. A simultaneous standard (Anhydrous ammonium sulphate) was done to estimate the amount of nitrogen taken up by N/70 HCl. From the nitrogen content of the sample, the protein content of different samples was calculated by multiplying with a factor of 6.25.

$$\% \text{ of nitrogen present in given sample} = \frac{\text{Sample titre value} - \text{Blank titre normality of HCl} \times 14 \times 100}{\text{Sample Weight} \times 1000}$$

**3. RESULTS AND DISCUSSION**

The results of the protein content of different treatments, grains during 90 days storage are summarized and presented in Table 1. The mean protein content of the grains was 9.89 percent; foxtail millet had highest protein (12.0 and 12.4 in WC and DC) followed by sorghum (9.9 and 10.5 in WC and DC) and pearl millet (9.67 and 9.8 in WC and DC). In general there was a significant effect of treatments on the protein content in the study. As the millets are used as whole as well as dehulled forms, both were used as controls. Dehulling of the grain resulted in the increase of protein content by 3.41 percent from whole grain protein.

From Fig. 1, it can be observed that there was no difference in Protein content of Heat (HE) and Heat Irradiated (HEI) treated WC grains whereas only 1 percent increase was found in DC grains. When the grains were either heat treated or combined with irradiation there was a reduction in the protein content by 8.8 percent and 11.8 percent in whole grain (WC) and dehulled grains (DC) respectively. Higher reduction was observed in DC than in WC. Similar trend was observed with irradiation combination treatment, the percent reduction was 8.8 and 10.74 in WC and DC respectively.

**Table 1. The details of treatments used for the study**

S. no	Treatments (8)	Grains (3)	Storage period(4)
1	Control –Whole grain	Sorghum	0 Day
2	Control- Dehulled grain	Pearl Millet	30 <sup>th</sup> day
3	Heat treated –Whole grain	Foxtail Millet	60 <sup>th</sup> day
4	Heat treated –Dehulled grain		90 <sup>th</sup> day
5	Heat and 1.0kGy Irradiated -Whole grain		
6	Heat and 2.5kGy Irradiated - Whole grain		
7	Heat and 1.0kGy Irradiated -Dehulled grain		
8	Heat and 2.5kGy Irradiated - Dehulled grain		

From the results it is evident that compared to dehulled heat treated grains, irradiation of the grains at 1kGy significantly reduced the protein content in all grains ( $p < 0.05$ , 1.7%). However, irradiation at 2.5 kGy significantly enhanced the protein content ( $p < 0.05$ , 3.3%). In the similar fashion, compared to heat treated grain, in whole grain there was a reduction in protein by 1.97 percent at 1 kGy irradiation and, at 2.5 kGy irradiation the protein was increased by 1.14 percent. However the post hoc tests revealed that there was no statistically significant difference between Dehulled heat treated (DEHE) and Whole heat treated (WHE) ( $p > 0.05$ ) (Table 2).

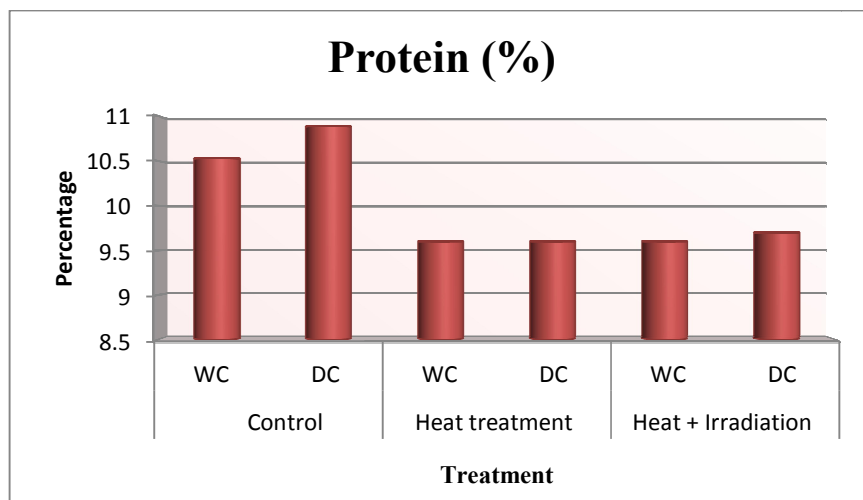
Among the grains the protein content was significantly high in foxtail millet, followed by sorghum and pearl millet (Table 2). Though the protein content is less in pearl millet as reported earlier it has better protein quality compared to other millets [8]. However earlier authors have reported higher protein content than in the present study. A protein content of 15.4, 14.8 and 16.3 percent was reported by Klopfenstein et al. [9] for gray, yellow and brown pearl millet, respectively. Local Sudanese cultivars investigated by Elyas et al. [10] gave a range of 10.8-14.9 percent protein [11]. Comparatively, lower protein content of pearl millet could be attributed to varietal differences [12].

The treated and untreated grains when they were stored for three months at ambient temperature in polyethylene bags there was a significant loss in the overall content of protein ( $p < 0.05$ ). The estimated loss was 1.68, 2.67 and 3.5 percent at

30, 60 and 90 days storage periods of the grains respectively. Even after storage the protein content in irradiated grains ranged from 8.8 to 12.15 g/100 g, which is appreciable as it, can meet up to 1/5<sup>th</sup> of the days.

Mean reduction in protein content ranged from 5.6 to 15.5 percent in dehulled grains, whilst in whole grain it was from 3.7 to 13.3 percent. Among dehulled grains maximum reduction in protein was observed in DHE1.0 of sorghum grain (17.14%), followed by DHE1.0 of foxtail millet (15.5%) and Pearl millet (14.28%) and DHEI 2.5 of sorghum. Among whole grains, maximum was in WHE of foxtail millet (13.3%) followed by WHEI 1.0 of pearl millet (12.92%) and sorghum (11.1%) (Fig. 2). Protein content was less affected by heat treatment in pearl millet while it affected differently in both whole and dehulled grains of sorghum.

Among the three grains studied maximum retention of protein was exhibited by foxtail millet (97.39%) followed by pearl millet (97.18%) and sorghum (94.75%) at the end of 90 days storage. The protein content was significantly affected by storage in all the treatments. Compared to the untreated, the protein content of treated grains significantly affected by storage. The percent protein retention at the end of 90 days was highest in DEHI2.5 grains (88.28%) followed by DHC (84.4%) and DHEI 1.0 grains (83.55 %). Similar trend was found in untreated whole grains also, where 89.43, 88.31 and 87.19 percent retention of protein in WHEI 2.5, WHE and WHEI 1.0 respectively.

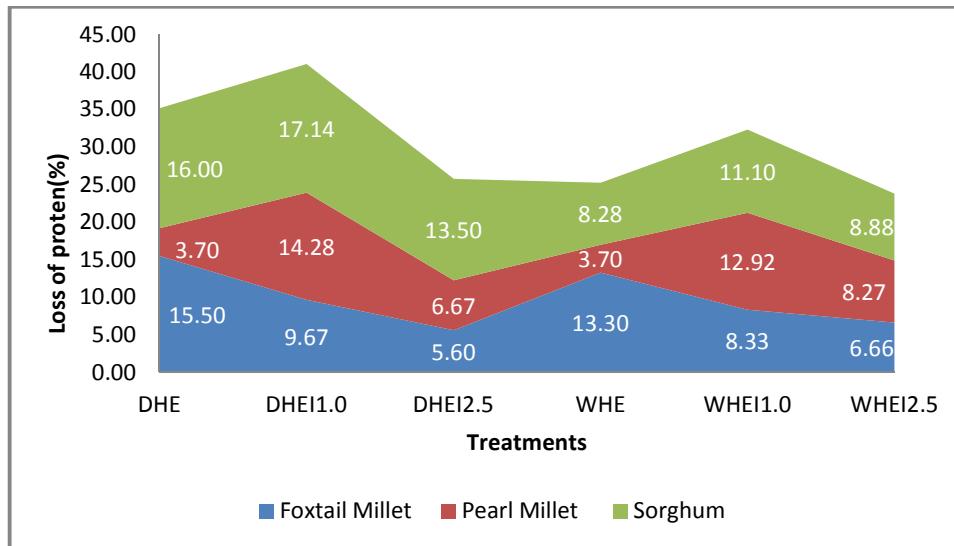


**Fig. 1. Protein content of millets as affected by heat and irradiation Combination treatments**  
 WC- Whole control, DC- Dehulled control

**Table 2. Effect of treatment, storage and grain type on protein**

Main Effects	Protein (%) ± SE
<b>Treatment</b>	
DC	10.89 ±0.01
DEHE	9.60 <sup>a</sup> ±0.02
DEHE1.0	9.44 ±0.03
DEHE2.5	9.92±0.01
WC	10.53±0.02
WHE	9.60) <sup>a</sup> ±0.01
WHE1.0	9.41 ±0.02
WHE2.5	9.71 ±0.01
<b>Storage</b>	
0 <sup>th</sup> day	10.09 ±0.02
30 <sup>th</sup> day	9.92 ±0.01
60 <sup>th</sup> day	9.82 ±0.03
90 <sup>th</sup> day	9.73 ±0.01
<b>Grains</b>	
Foxtail Millet	11.31±0.02
Sorghum	9.24±0.03
Pearl Millet	9.11±0.02

\*Significant at p=0.05, values with similar superscripts are not significantly different with each other (p>0.05)



**Fig. 2. Percent reduction of protein content in treated grains**  
 DHE - Dehulled heat treated, DHE1.0- Dehulled heat and irradiated (1kGy)  
 DHE2.5- Dehulled heat and irradiated (2.5kGy), WHE - Whole heat treated,  
 WHE1.0- Whole heat and irradiated (1.0kGy),  
 WHE2.5 – Whole heat and irradiated (2.5kGy)

**4. CONCLUSION**

Irradiation can be an effective alternative technology. Commercial scale use of radiation processing for food and feed commodities has been successful in several countries. Treating food with gamma radiations has been proven to be effective in

killing insects, reducing food borne microbial growth, increasing the shelf life of foods, changing their physical properties, selectively inactivating and removing anti nutritional factors like inhibitors of protease and amylase. The mean protein content of the grains was 9.88 percent. Among the three grains foxtail millet had significantly highest protein (11.31%) followed by

sorghum and pearl millet. Consumption of 100 g of foxtail millets can provide 6.36 g of protein which is equivalent to 1/10 of the daily requirement.

### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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