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Functional characteristics and storage stability of hot air assisted radio frequency treated pearl millet

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Abstract

Nutricereals, pearl millet flour consumption is hardly limited to a few specific regions of the world owing to the development of early rancidity on storage. Hot air assisted radio frequency technology (HARF) was used to improve the storage stability of pearl millet flour. Pearl millets at different moisture levels of 10.5±0.5, 12 & 15% were subjected to HARF for 5-, 10- & 15-min exposure period under fixed electrode position. The results revealed that significant reduction in peroxide value and free fatty acid values of flours from treated pearl millet was found compared to decorticated raw flour; thereby, the storage stability enhancement of pearl millet flour up to 180 days was achieved. No significant difference was found for emulsifying, foaming and cooking properties between treated pearl millet at 15% moisture, 15 min exposure and decorticated raw one. In contrast, the bulk density of flour from treated pearl millet was found higher. Significant increase in hardness of cooked grains after the treatment was observed while the springiness, cohesiveness and gumminess values did not vary. In FTIR analysis, no difference was observed between the peaks of whole and decorticated pearl millet flours in both untreated and treated samples.

Keywords Hot air assisted radio frequency, Functional properties, Storage stability, Pearl millet

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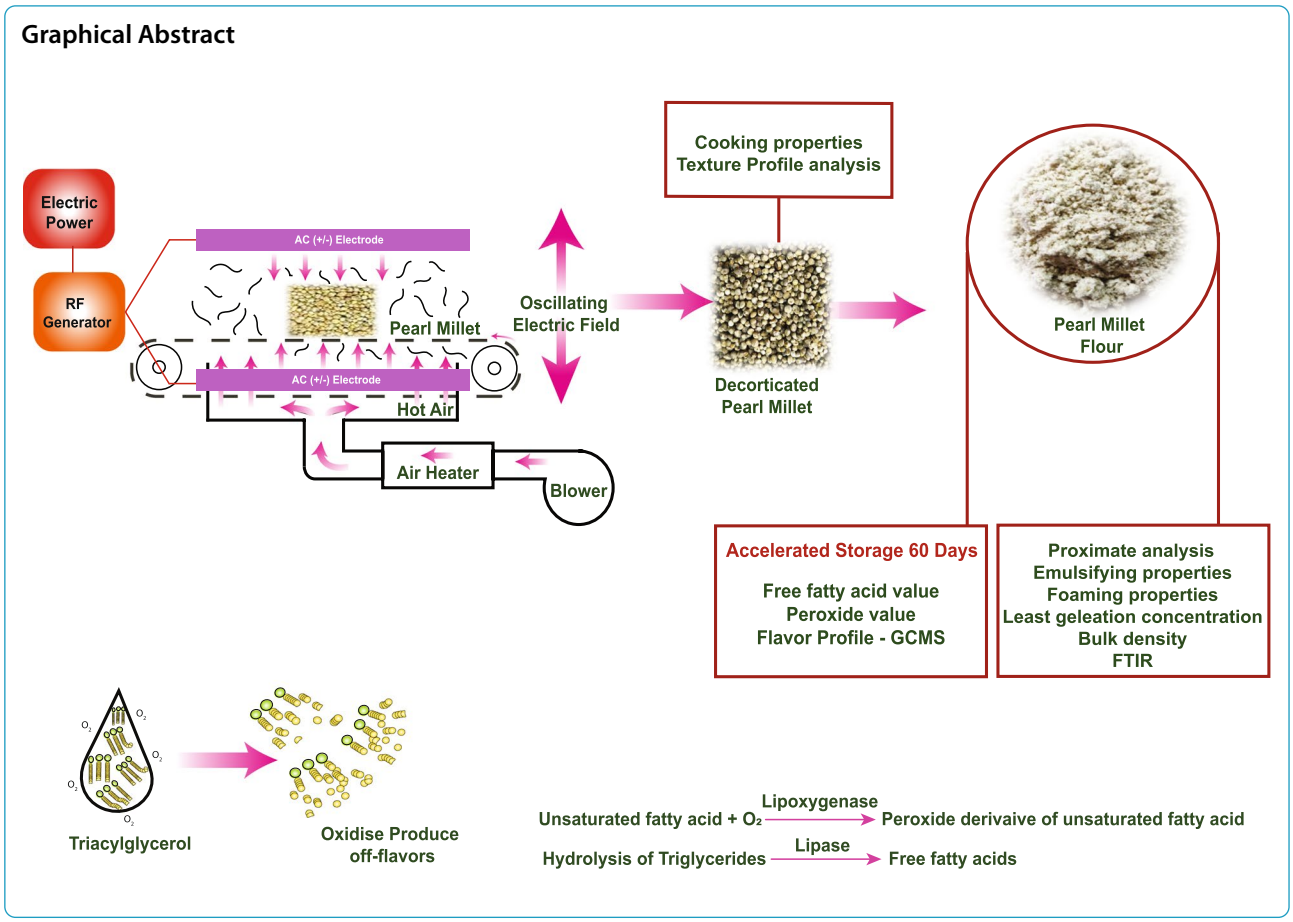

Introduction

Increasing consumers' concern worldwide to reduce disease risks is compelling to focus not only on the nutritional but also the functional roles of foods rather than curative treatments. Incorporating millets into one's diet can effectively reduce malnutrition, obesity, and celiac disease (Goswami et al. 2020). Notably, their consumption lowers the risk of chronic diseases like diabetes, cancer and cardiovascular diseases (Reynolds et al. 2019). Pearl millet contains high protein, fat, energy, minerals (calcium, magnesium, phosphorous, iron and zinc), phytochemicals, vitamins, and crude fibers (Goswami et al. 2020). Pearl millet being a climate-resilient crop, ensure food and nutritional security in changing climatic scenarios (Rani et al. 2018), and India is the largest producer with an average of 8.61 million tons of production (Directorate of Millets Development, 2020). Despite its nutritional superiority, pearl millet flour consumption is hardly limited to a few specific regions of the world owing to its poor storability (lower than ten days after milling) (Goswami et al. 2020) and early rancidity or unpleasant off-flavours development on storage (Akinola et al. 2017). Hydrolytic rancidity occurs due to high-fat content where lipase enzymes hydrolyze triglycerides to free fatty acids. Oxidative rancidity leads to develop the hydroperoxides and thereby generating the off-odor causing secondary metabolites such as aldehydes and ketones (Rani et al. 2018). The quality of the processed product is influenced by the physical and functional characteristics of flour. Therefore, stabilization of the flour is essential and the variations of functional and physical attributes due to the treatment need to be revealed to assess the ultimate quality of the food product (Chandra et al. 2015). In addition, the application of stabilized pearl millet flour in various food preparations can considerably enhance its economic importance.

Different methods have been investigated for pearl millet flour stabilization via lipase inactivation. These methods include hydrothermal (Yadav et al. 2012a), microwave (Yadav et al. 2012b) and HARF (Yarrakula et al. 2021). Radio frequency (RF) heating involves the utilization of electromagnetic energy with a frequency between 3 kHz to 300 MHz and initiates volumetric heating owing to frictional interaction between molecules. The advantageous deeper penetration and higher heating rates of HARF induce uniform heating, shorter processing time;

thereby less energy consumption (Yarrakula et al. 2021). HARF technique may have great potential to stabilize the pearl millet flour rapidly at a commercial level and improve the functional properties of flours. Therefore, the research was planned to investigate the functional characteristics of flours from HARF treated pearl millet along with accelerated storage stability.

Materials and methods

Raw material

Pearl millets were procured from the local market, Thanjavur, Tamil Nadu, India and passed through 0.50 mm sieve to clean and separate uniform sized pearl millets. The estimated moisture level of market sample was 10.5±0.5% w.b. The millets obtained were packed in plastic laminated packaging bags for further analysis and protect them from external factors.

Processing of pearl millets for storage stability

HARF processing

Considering the preliminary experiments, procured pearl millets were tempered to different moisture levels by following the method (Yadav et al. 2012a), the moisture level of 1 kg pearl millet was adjusted to 12 and 15%, w.b. from 10.5 ± 0.5% initial value. The tempered grains were subjected to HARF (10 kW, 40.68 MHz ± 0.5%) treatment for 5, 10 and 15-min duration and then decorticated by a laboratory steel polisher (Model: TM 05, Stake corporation, Japan). The decorticated grains were milled into flour by a hammer mill (Almech enterprise technologies, Coimbatore, India). The flour produced with less than 1 mm particle size was packed in airtight zip-lock polyethylene bags, labelled and stored in refrigerated condition (3-4 °C). Further, changes in quality attributes were studied for the flours obtained from treated pearl millets and decorticated raw pearl millet flour as control. Importantly, the accelerated storage stability of flours from decorticated raw (control) and HARF treated pearl millet was conducted.

Proximate composition

The chemical composition of flours from HARF treated and decorticated raw pearl millet (control) was determined. The proximate analysis was conducted by following AOAC (2005).

Storage studies

To conduct the shelf life test, flour from the HARF treated pearl millets at 15%, 15 min and the decorticated raw flour as control were selected, as the maximum of 97.3% lipase inactivation was found at 15% moisture for 15 min in our previous study (Yarrakula et al. 2021).

The accelerated storage stability of flours from HARF treated pearl millets (15%, 15 min) and decorticated raw pearl millet as control was conducted in dark temperature controllable incubator at 45℃, 70 ±2% RH for 60 days to simulate about 6 months under ambient storage conditions (35°C, 60-70% RH). Samples were withdrawn from incubator at an interval of 10 days for assessing the peroxide value and free fatty acid content. On the basis of Q_{10} value of 3.4 for lipid rancidity, the time of storage was determined (Ling et al. 2020). It is common to use the parameter Q_{10} to obtain the rate of quality changes (k) of food products as the temperature (T, °C) influences the quality of food products during storage and is estimated using the equation below

$$Q_{10}^{(T_1-T_2)/10} = \frac{\theta_s(T_1)}{\theta_s(T_2)}$$

Where T_1, and T_2 are normal, and accelerated storage temperatures (°C), respectively, and θ_s is the storage time (days).

Free fatty acid content

Free fatty acid (FFA) values were determined using a titration method according to Bai et al. (2017). Lipids were extracted using hexane and the oils (1g each) were added to hot neutralized ethanol (7 mL) with phenolphthalein (2 mL) as indicator and then titrated with 0.1 M NaOH (95% ethanol as solvent). The FFA was expressed as % oleic acid.

Peroxide value

Peroxide value (PV) was determined by titration method according to Bai et al. (2017). The oil samples (1g each) were placed into conical flask (50 mL). Then, 5 mL of $CH_3COOH/CHCl_3$ (3:2) was dissolved in oil by agitating gently. Saturated KI solution (0.1 mL) was added, allowed for 1 min shaking then 6 mL of deionized water was added to the mixture. The mixture was then slowly titrated with 0.01 M $Na_2S_20_3$. The PV was expressed as mill equivalents of O_2 kg^{-1} oil.

Effect of HARF processing on functional, cooking and textural attributes of flours

Emulsifying properties

Emulsion activity (EA) was determined by following the method of Siroha et al. (2016). Pearl millet flour of 3.5 g in 50 mL distilled water was homogenized for 30 s in a homogenizer (Model T18D, IKA, Germany) at 14000 rpm. Sunflower oil of 25 mL was added to the mixture and homogenized for 30 s. Additional 25 mL of sunflower oil was added and homogenized for 90 s. The two 50 mL centrifuge tubes that have shared the emulsion evenly, were centrifuged at 3100 rpm for 5 min.

The emulsion stability (ES) was estimated by following the procedure (Makri et al. 2005). The retained emulsion in the centrifuge tube was heated in hot water bath (Model RWB 6, Remi Elektrotechnik LTD, Vasai, India) at 80℃ for 30 min, cooled under tap water for 15 min, and then centrifugation at 4200 rpm for 15 min. EA, and ES were calculated as follows:

$$EA, \% = \frac{Volume\ of\ emulsified\ layer}{Volume\ of\ emulsion\ before\ centrifugation} \times 100$$
$$ES, \% = \frac{Height\ of\ emulsifiedlayer}{Total\ height\ of\ mixture} \times 100$$

Foaming properties

Foaming capacity (FC) and stability (FS) of pearl millet flours were determined (Siroha et al. 2016). In the method, the dispersion of pearl millet flour (3% w/v) in 50 mL distilled water were homogenized using homogenizer (Model T18D, IKA, Germany) at 14000 rpm for 2–3 min and then the blend was immediately taken into a graduated cylinder. The distilled water (10 mL) used to rinse the homogenizer cup was also added to the graduated cylinder. The volume of foam before and after whipping was assessed. FC was calculated using the below equation as:

$$Foaming\ capacity, \% = \frac{V_b - V_a}{V_a} \times 100$$

Where, V_a= Foam volume before whipping, mL, V_b = Foam volume after whipping, mL

FS is the changes in volume of foam in the graduated cylinder observed for 30, 60, 90 and 120 min of storage.

$$Foam\ stability, \% = \frac{V_{ft} - V_b}{V_b} \times 100$$

Where, V_{ft}= Foam volume after 120 min, mL

Least gelation concentration

Least gelation concentration (LGC) of pearl millets was found as per the method (Kaushal et al. 2012) in which the samples of 5 mL each were prepared in different concentrations (2, 4, 6, 8, 10, 12, 14, 16, 18, and 20 g/100 mL) and transferred into test tubes. Then, the tubes were subjected to heating in the water bath (Model RWB 6, Remi Elektrotechnik LTD, Vasai, India) at 80°C for an hour followed by rapid cooling under running tap water. Further, the tubes were cooled under refrigerated condition for 2 h. LGC was expressed as the concentration above which pearl millets remained when test tubes were inverted.

Bulk density

Bulk density was assessed by finding the ratio of weight of flour to its total volume. Flour sample was filled into 10 mL graduated cylinder. Void spaces in flour were avoided by gentle tapping at the bottom of the cylinder several times (Omowaye-Taiwo et al. 2015).

Cooking time

The decorticated grains of about 10g were taken in beaker with 100-mL boiling water and continued heating of grains. A few grains at an interval of 10 min were withdrawn from the beaker and pressed between the glass plates. The translucency of constant diameter of the spread was considered as cooking time.

Colour values of cooked pearl millet grains

Using Hunter lab colorimeter (Colourflex EZ model: 4510), the color values of cooked pearl millet grains were found in terms of L^*, a^* and b^*. Triplicates were recorded for each sample.

Texture Profile Analysis (TPA)

The TPA of cooked pearl millet grains was conducted to determine the hardness, springiness, cohesiveness, and gumminess of cooked grains using the Texture Analyser (Stable Micro Systems, Model: TA HD PLUS) interfaced with Exponent 5.1.1.0 software. The instrument was fitted with a load cell of 35 kg. The selected program of two cycle compression versus time was run so that the selected probe compressed the samples till 90% of the thickness of cooked grain, returned to the original position and then compressed again for each test. A cylindrical probe of 35 mm with 1mm/s test velocity and 1 mm penetration distance in the sample was used to compress 3 grains.

FTIR analysis

The identification of functional groups of pearl millet flours was carried out by FT-IR spectroscopy. The spectrum was obtained using IR Affinity-1S spectrometer with DLATGS detector by Labsolution IR software. The spectrum was collected at a resolution in the range of 4000–400 cm^{-1} at a resolution of 4 cm^{-1}.

Statistical analysis

The variations among the dependent variables between the different levels of the independent variables were analyzed and correlated using Multivariate analysis of Variance (MANOVA). Triplicate readings of all the experiments were recorded in order to obtain the data variability by estimating the standard deviation (SD). Using the tool, SPSS, version 25.0 (IBM Corp, USA), statistical analysis of experimental data was conducted. MANOVA and Pearson correlation coefficient (r) were employed to find the significant changes ($P<0.05$) in

proximate, functional, cooking and textural properties of pearl millet.

Flavor profile analysis of pearl millet flour using GC-MS

The flavor profile of control and treated pearl millet flour was analyzed using GC-MS on 0^{th} and 60^{th} day of storage to identify the compounds responsible for rancid flavor of pearl millet. In lipid extraction, 3 ml of methanol was added to the samples and lipids were extracted by shaking at room temperature for 15 min. The lipid extraction was carried out twice as the collected lipid layer was lower. The lipid extracts were filtered and dried under a stream of nitrogen gas. The flavor compounds in the pearl millet were extracted with methanol and analyzed. Agilent - 8890 GC coupled with 5977 MSD with Rtx-5MS column (5% Diphenyl / 95% Dimethyl poly siloxane) of 30m x 0.25mm length and 0.25 µm thickness was used for analysis. The 2 µL of sample was injected (10:1) in split mode. The following GC program was used for analysis, oven temperature program was 110 °C with 3.5 minutes holding time, then increased to 200 °C at a rate of 10 °C/minute and then increased to 280 °C at the rate of 5 °C/minute with 12 minutes holding time. The injector temperature was 280 °C, carrier gas flow rate of 1 mL/minute with total GC run time of 40.5 minutes. The used MS program was, inlet temperature 290 °C, source temperature 250 °C, electron energy 70 eV, and mass scan (m/z) 50-500 amu with total MS run time of 40.5 minutes. The NIST library of 2020 version was used for the identification of compounds (Johnson et al. 2022).

Results and discussion

Chemical composition of pearl millet flours

Table 1 represents the chemical composition of decorticated raw and HARF treated pearl millets. The protein content of HARF treated pearl millets varied from 5.11 to 7.11%. The increased moisture level and exposure time caused a significant ($P<0.05$) increase of protein content in treated pearl millet. Higher heating rates of flours might have obtained the variations in the nitrogen content after the exposure to the HARF technique (Wani et al. 2017). In contrast, the fat content was in reverse trend compared to decorticated raw pearl millet. Low amount of lipids will assure the shelf life enhancement of flours by reducing the chances of rancidity development (Sade, 2009). HARF treatment showed an increase in ash content with the enhancement of moisture levels of pearl millet. At 12 and 15% moisture levels, no significant ($P<0.05$) variation was observed. The ash content indicates a rough assessment of the mineral content of the material (Sade, 2009). Though, the carbohydrate content of HARF treated pearl millet was slightly reduced with the increase of moisture (10.5-15%), it was comparatively higher than the decorticated raw sample. The high carbohydrate content of the flour indicated that these millets would be served as energy rich source. These results are in agreement for microwave-treated little millet. The nutritional profile of millet flours depends on geographical regions, growth conditions and variety (Kumar et al. 2020).

Effect of HARF processing on storage stability

Flours from HARF-treated (15 % moisture, 15 min) and decorticated raw pearl millet were investigated for the changes in terms of peroxide value (PV) and free fatty acids (FFA) during accelerated storage at ambient conditions (45°C, 70–85% RH) (Fig. 1a & b). PV signifying the storage changes in lipids varied significantly with the storage duration (Fig. 1a). Significant ($P<0.05$) increase in PV of stored flours during the entire storage duration

Table 1 Proximate analysis and functional characteristics of decorticated raw (control) and HARF treated pearl millet at different processing conditions

Processing conditions		Proximate analysis				Functional properties			
		Protein	Fat (%)	Ash (%)	Carbohydrate (%)	Emulsion activity (%)	Emulsion stability (%)	Foam capacity (%)	Foam stability (%)
Decorticated raw pearl millet		6.13 ± 0.02^{a}	5.91 ± 0.01^{a}	0.04 ± 0.00^{a}	87.90 ± 0.02^{a}	43.5 ± 0.7^{a}	41.5 ± 0.7^{a}	7.55 ± 0.07^{a}	3.30 ± 0.00^{a}
HARF treated pearl millet at different processing conditions	10.5%, 10 min.	5.12 ± 0.03^{b}	3.83 ± 0.02^{b}	0.01 ± 0.00^{b}	91.03 ± 0.00^{b}	35.5 ± 0.7^{b}	34.5 ± 0.7^{b}	8.35 ± 0.07^{b}	3.25 ± 0.07^{a}
	10.5%, 15 min.	5.28 ± 002^{c}	4.23 ± 0.02^{c}	0.01 ± 0.00^{b}	90.47 ± 0.04^{c}	35.0 ± 1.4^{b}	35.0 ± 1.4^{b}	10.75 ± 0.07^{c}	10.50 ± 0.70^{b}
	12%, 10 min.	5.47 ± 0.04^{d}	3.06 ± 0.01^{d}	0.02 ± 0.00^{c}	91.45 ± 0.02^{d}	44.5 ± 0.7^{a}	43.5 ± 0.7^{c}	8.25 ± 0.07^{b}	6.30 ± 0.00^{c}
	12%, 15 min.	5.73 ± 0.02^{e}	3.27 ± 0.02^{e}	0.02 ± 0.00^{c}	90.97 ± 0.00^{e}	49.0 ± 0.0^{c}	37.5 ± 0.7^{d}	18.45 ± 0.21^{d}	8.45 ± 0.21^{d}
	15%, 10 min.	7.12 ± 0.03^{f}	2.08 ± 0.02^{f}	0.02 ± 0.00^{c}	90.76 ± 0.01^{f}	46.5 ± 0.7^{d}	45.5 ± 0.7^{c}	23.35 ± 0.07^{e}	10.05 ± 0.07^{e}
	15%, 15 min.	7.11 ± 0.03^{f}	2.22 ± 0.03^{g}	0.02 ± 0.00^{c}	90.63 ± 0.00^{g}	44.5 ± 0.7^{a}	44.5 ± 0.7^{c}	7.52 ± 0.21^{a}	3.30 ± 0.00^{a}

Superscripts with same letter in column are not statistically different

Superscripts represent the significant variation in samples

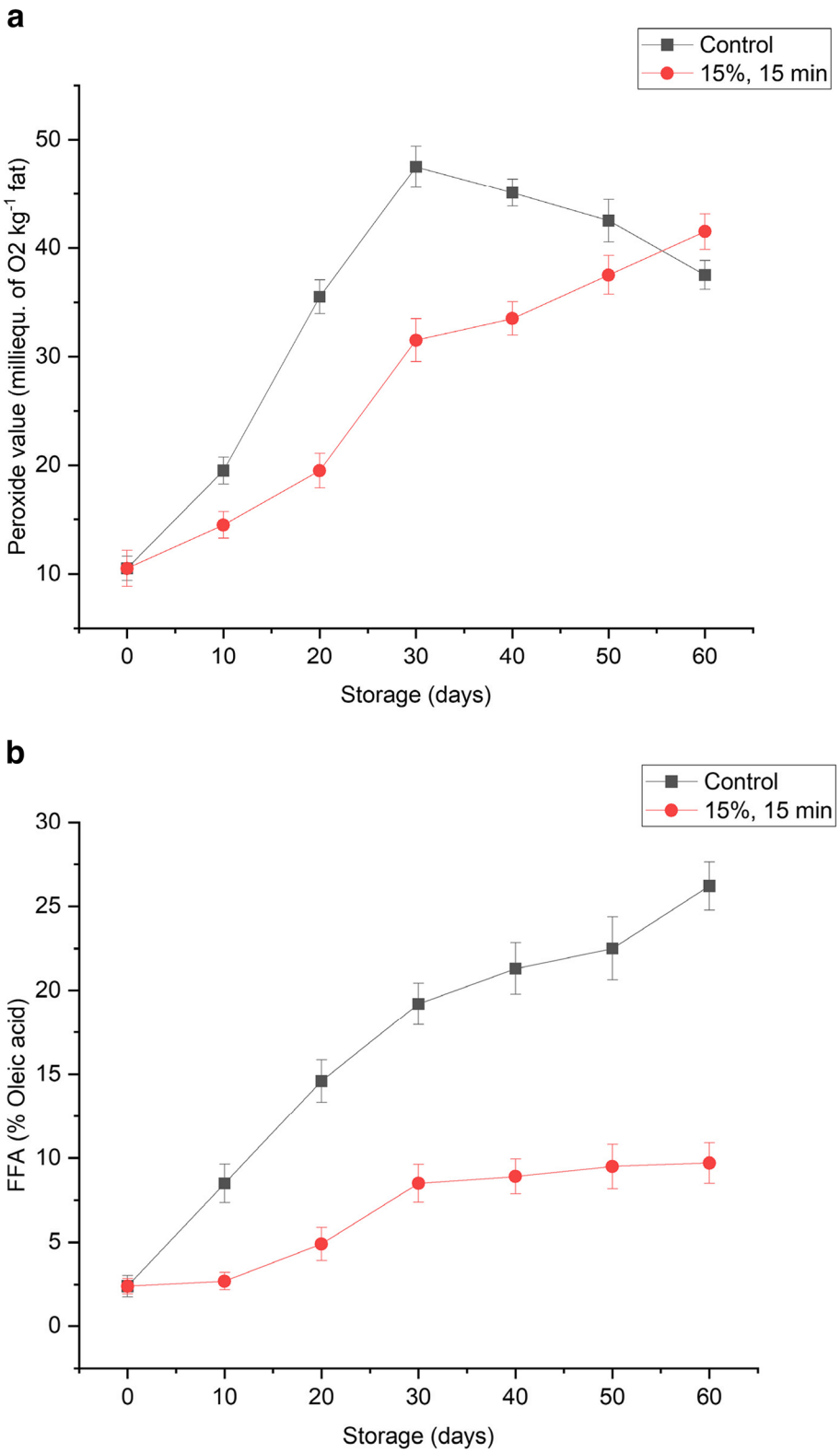

Fig. 1 **a** Peroxide values of pearl millet flour (milled from untreated and HARF treated pearl millet at 15%, 15 min) during accelerated storage at 45 ºC. **b** Free fatty acid (FFA) values of pearl millet flour (milled from untreated and HARF treated pearl millet at 15%, 15 min) during accelerated storage at 45 ºC

was observed. PV of the flour from HARF treated grains (10.5-42.5 meq of O_2 kg^{-1} fat) were significantly ($P<0.05$) lower than the flour from decorticated raw pearl millet (10.5-44.1 meq of O_2 kg^{-1} fat) and this reduction indicates that lower amounts of hydroperoxides were formed in treated one. The decorticated raw pearl millet flour might contain secondary metabolites such as ketones, and aldehydes (Rani et al. 2018), responsible for the bitter and mousy odour. Similar findings were obtained for microwave (Yadav et al. 2012a) and hydrothermally treated pearl millet (Yadav et al. 2012b).

During the entire storage duration, FFA of flours from decorticated raw and HARF treated pearl millet varied significantly ($P<0.05$) with the storage duration (Fig. 1b). Significant ($P<0.05$) increase in FFA was observed for both the flours. The level of FFA content in the flour from decorticated raw pearl millet (5.92-28.22%) was higher than the flour from HARF treated grains (2.25-9.94%) at each storage interval. A high value of FFA is mainly due to hydrolytic changes associated with the action of lipolytic enzymes (Rani et al. 2018). Further, an enhancement in lipase activity during the entire storage duration might be responsible for significantly higher FFA value in the flour obtained from decorticated raw pearl millet compared to the flour obtained from HARF-treated grains (Staniszewska et al. 2021). Similar findings were obtained for microwave (Yadav et al. 2012b) and hydrothermally treated pearl millet (Yadav et al. 2012a).

Effect of HARF processing on functional, cooking and textural properties of pearl millet flour

Emulsifying properties

The hydrophobic and polar groups of the protein present interface of oil and water reduce free energy and interface tension (Jiang et al. 2018). The effect of HARF treatment on emulsifying properties is shown in Table 1. HARF treatment showed a significant ($P<0.05$) effect in EA of pearl millet compared to decorticated raw one. With the increase of moisture levels (10.5 to 12%), the exposure of HARF showed a significant ($P<0.05$) increase in EA from 35.5 ± 0.7 to 49.0 ± 0.0%. Though the EA of pearl millet at 15% moisture level was reduced at different exposure periods (10 &15 min), there was no significant difference between the EA of HARF treated pearl millet at 15% moisture for 15 min and the untreated decorticated raw pearl millet. During the HARF treatment, the change of spatial structure of protein might have led to a slight decrease in its solubility which resulted in reduction in EA (Yarrakul et al. 2021). Previous studies also revealed that the emulsifying properties of RF treated soybean were similar to those of untreated soybean (Jiang et al. 2018).

During the HARF treatment of pearl millet at different processing conditions, ES was increased significantly ($P<0.05$) from 34.5±0.7 to 44.5±0.7% (Table 1). An emulsion with higher EA and ES is suitable in foods such as comminuted meat products, mayonnaise, frozen desserts and salad dressing (Khan et al. 2011). The ES of HARF treated pearl millet at low moisture levels (10.5 & 12 %), and different exposure periods was lower than 15% moisturized one. In addition, the ES of pearl millet (15% moisture) treated for 10 and 15 min was higher than untreated raw decorticated pearl millet. Similar results were reported for maize flour (Eltayeb, 2012).

Foaming properties

Table shows the effect of HARF treatment on the foaming properties of pearl millets. HARF treatment caused a significant ($P<0.05$) increase in foam capacity of pearl millet varied from 8.35 ± 0.07 to 23.25 ± 0.07%, and foam stability from 3.25 ±0.07% to 10.05 ±0.07% for different moisture and exposure period combinations (up to 15%, 10 min). The foaming capacity is owed to hydrophobic group of the protein molecule, that can cause the rapid spread and distribution at the interface (Jiang et al. 2018). In contrast, foaming capacity and stability reduced to 7.52 ± 0.27% and 3.3 ± 0.0%, respectively, at the 15% moisture, 15 min HARF exposure (lowest lipase activity) where the sample temperature was 78.3°C. The reduction in foaming properties might be due to protein denaturation and the occurrence of insoluble aggregates, which decreased the dispersion of protein (Devisetti et al. 2014). Foaming properties of flour mainly depends on carbohydrate and protein composition in the product. The foam formation and its stability mainly depend on interfacial film produced by protein which helps in maintaining air bubbles with a lower coalescence rate, whereas, carbohydrates enhance the viscosity of the suspension medium (Bhatt et al. 2017). Our findings showed that HARF treatment of pearl millet at 15%, 15 min causes no changes in both foaming capacity and stability compared to the untreated decorticated raw pearl millet.

Least gelation concentration

The effect of HARF treatment at different processing conditions is represented in Table 2. The lower concentration for gel formation is 2% for HARF treated and decorticated raw pearl millet samples. Lower the LGC value, better the gelation ability of protein, and swelling capacity of the flour (Chandra et al. 2015). Here, the HARF treatment did not lower the gelation ability of protein ingredient. The gel formation mainly depends on hydration, and swelling behavior of amorphous regions of starch granules (Devisetti et al. 2014).

Table 2 Least gelation concentration of decorticated raw (control) and HARF treated pearl millet at different processing conditions

Least gelation concentration (%)	Decorticated raw pearl millet	HARF treated pearl millet at different processing conditions					
		10.5%, 10 min.	10.5%, 15 min.	12%, 10 min.	12%, 15 min.	15%, 10 min.	15%, 15 min.
2	-	-	-	+	+	+	+
4	+	+	+	+	+	+	+
6	++	++	++	++	++	++	++
8	++	++	++	++	++	++	++
10	++	++	++	++	++	++	++
12	++	++	++	++	++	++	++
14	++	++	++	++	++	++	++
16	++	++	++	++	++	++	++
18	++	++	++	++	++	++	++
20	++	++	++	++	++	++	++

- No gel formation, + gel formation, ++ firm gel

The carbohydrates such as lactose, maltose, and sucrose are reported to have the capacity to decrease the thermodynamic affinity of the protein in for an aqueous solution and increases the magnitude of the interaction between protein molecules, thus improving the gelling ability (Adebowale et al. 2009). These properties help in the formulation of various food products including sausage emulsions, custard puddings, and sauces, which require thickening and gelling.

Bulk density

Significant ($P<0.05$) changes were observed in the bulk densities of flours from treated pearl millets. The bulk densities of flours from treated pearl millet at different moisture levels for 15 min showed significant ($P<0.05$) increase. The bulk density of flours was varied from 0.746 to 0.776 g/mL. When compared to the bulk density of flour from decorticated raw pearl millet (0.764 kg/m3), the bulk density of flour from treated pearl millet (15%, 15 min) was higher.

The grain moisture content is the responsible factor for the change in bulk density of millet flours. Flour with high bulk density is used in food preparations (liquids, semisolids, or solids), and also as a thickener in food products (Hasmadi et al. 2020). In contrast, low-density flour is used to prepare weaning food formulations (Devisetti et al. 2014).

Cooking time

The effect of HARF treatment on the cooking time of pearl millet is presented in Table 3. The cooking time of untreated and HARF treated whole pearl millets (15%, 15 min) was found as 30 and 30 min, respectively, indicating that HARF treatment did not show any significant variation between the cooking time of treated and untreated whole pearl millet grains. Nevertheless, the decortication process reduced the time of cooking from 30 to 20

Table 3 Colour values and Texture profile analysis (TPA) of cooked pearl millets of whole, decorticated, HARF treated (whole) and HARF treated-decorticated pearl millets at 15% for 15 min

Parameters	WPM	DPM	TWPM	TDPM
Cooking time (min)	30	20	30	20
L*	27.21 ± 0.02^{a}	37.42 ± 0.01^{b}	29.12 ± 0.04^{c}	37.64 ± 0.02^{d}
a*	2.12 ± 0.02^{a}	0.86 ± 0.03^{b}	2.42 ± 0.02^{c}	0.94 ± 0.02^{d}
b*	8.11 ± 0.02^{a}	6.65 ± 0.03^{b}	8.52 ± 0.02^{c}	7.43 ± 0.02^{d}
Hardness	2044.0 ± 199.0^{ab}	586.0 ± 356.0^{b}	2711.0 ± 1188.0^{a}	730.3 ± 86.8^{b}
Springiness	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}	1.00 ± 0.00^{a}
Cohesiveness	7.69 ± 1.62^{a}	8.98 ± 6.09^{a}	4.99 ± 2.65^{a}	5.56 ± 3.62^{a}
Gumminess	15737 ± 3547^{a}	4944 ± 2989^{a}	15259 ± 13666^{a}	4108 ± 2877^{a}

Values are mean±standard deviations of triplicate observations. Superscripts with same letters in row are not significant

WPM Whole pearl millet, *DPM* Decorticated pearl millet, *TWPM* HARF Treated whole pearl millet (15%, 15 min) and TDPM- HARF Treated decorticated pearl millet (15%, 15 min)

min. The time of cooking of decorticated pearl millets (untreated and HARF treated) was assessed as 20 min.

Colour of cooked grains

The colour values of whole, decorticated, whole HARF treated (15%, 15 min) and HARF treated-decorticated pearl millet grains were shown in Table 3. Significant ($P<0.05$) variation in the colour values (L*, a* and b*) was noticed between the whole and decorticated pearl millets. In fact, the L*, a* and b* values of whole pearl millets were greater ($P<0.05$) than the decorticated grains. Pigments such as flavonoids removal during the decortication process leads to the reduction of colour values of decorticated grains (Dias-Martins et al. 2018).

HARF treatment of pearl millets (15%, 15 min) lowered the whiteness of decorticated grains for the same cooking time. However, it was very small and not even perceptible to the human eye. In contrast, redness and yellowness of HARF treated (whole and decorticated) grains increased significantly in comparison with untreated (whole and decorticated) grains. Gavahian et al. (2019) reported that the lightness (L*) of microwave and conventionally-cooked grains did not vary significantly ($P<0.05$) for the same cooking time.

Textural properties of cooked grains

Texture profile analysis (TPA) of cooked grains in terms of hardness, springiness, cohesiveness and gumminess of cooked grains was given in Table 3. Hardness indicates the required maximum compressive force of the sample and the most essential attribute for assessing the quality of a cooked cereal. Hardness of whole pearl millet grains was greater than decorticated millet grains. It is due to the greater rigidity of the pericarp as the grain contains proteins, lipids and fiber. The decortication removes pericarp there by promotes better water absorption capacity and consequently, lower hardness (Dias-Martins et al. 2018). HARF processing (15%, 15 min) increased the hardness of decorticated grains; however, the variation was insignificant ($P<0.05$). No significant ($P<0.05$) variation in springiness of grains was found (Table 3). The decortication and HARF treatment did not promote any variation in the springiness of pearl millet grains. Dias-Martins et al. (2018) stated a springiness value of 0.30 for decorticated-conventionally cooked pearl millets. Cohesiveness is the rate that food disintegration takes place under mechanical force and it can serve as a reliable indicator of how the sample will keep together after being cooked. Greater mechanical force is needed to disintegrate whole grains than decorticated grains; however, decortication and HARF treatment did not promote any significant ($P<0.005$) difference in pearl millet grains (Table 3). The gumminess values of whole (15737 N) and HARF treated (15259 N) pearl millet grains were higher than decorticated (4944 N) and HARF-decorticated (4108 N) grains, respectively, due to the greater hardness of whole pearl millet grains. Dias-Martins et al. (2018) reported a gumminess value of 9.77 N for conventionally cooked pearl millet grains, which is less than the obtained finding.

FTIR analysis

The FTIR analysis was carried out to identify the change in structural properties of pearl millet. The IR spectrum of pearl millet was given in Fig. 2. The bands 3846 – 3738 cm^{-1} corresponds to the -O-H bond of carboxyl group, the bands 3693 – 3570 cm^{-1} corresponds to the -N-H bond of amide group. The vibrations 2980 – 2167 cm^{-1} specifies asymmetrical -C-H stretching. The vibrations observed in the range of 1743 – 1710 cm^{-1} corresponds to carbonyl stretch (Vinutha et al. 2022). The peaks observed between 1656 – 1641 cm^{-1} corresponds to the amide I band. The peaks observed around 1630 – 1640 cm^{-1} indicates dense water present in the starch, and these were absent in treated samples which may be due to the reduction of water in treated sample due to heat generation. The peak around 1460 cm^{-1}indicates the tightly bound moisture content of the pearl millet flour. The peaks observed around 1381 – 1076 cm^{-1} and 1149 – 993 cm^{-1} represents the hydroxyl and carboxylic acid groups of the starch present in the pearl millet (Aruna & Parimalavalli, 2022). The characteristic peaks of 1076 cm^{-1} and 1149 cm^{-1} corresponds to C-O stretch and C-O-H bonding. The peaks around 968 – 993 cm^{-1} corresponds to the -C-N bond of amine group. The peaks observed around 570 cm^{-1} indicates the -C-I bond of aliphatic iodo group. No difference was observed between the peaks of whole and decorticated pearl millet flour in both untreated and treated samples. The treated samples were observed with extra peaks at around 2167 cm^{-1} and 1022 cm^{-1}indicating the disruption of starch crystallinity due to heat generation (Ambigaipalan et al. 2014). The difference in the FTIR spectrum between treated and untreated samples was significantly lower indicating the minimal effect of RF treatment on structural properties.

Flavor profile of pearl millet

The aldehydes, ketones, alcohols, and heterocyclic compounds are responsible for the contribution of flavor to pearl millet. Thermal processing usually leads to formation of new flavors or evaporation and degradation of existing flavor compounds owed to their heat sensitive nature. The control (untreated) pearl millet showed 14 compounds on 0^{th} day (Table 4) whereas, treated pearl millet showed only 11 compounds

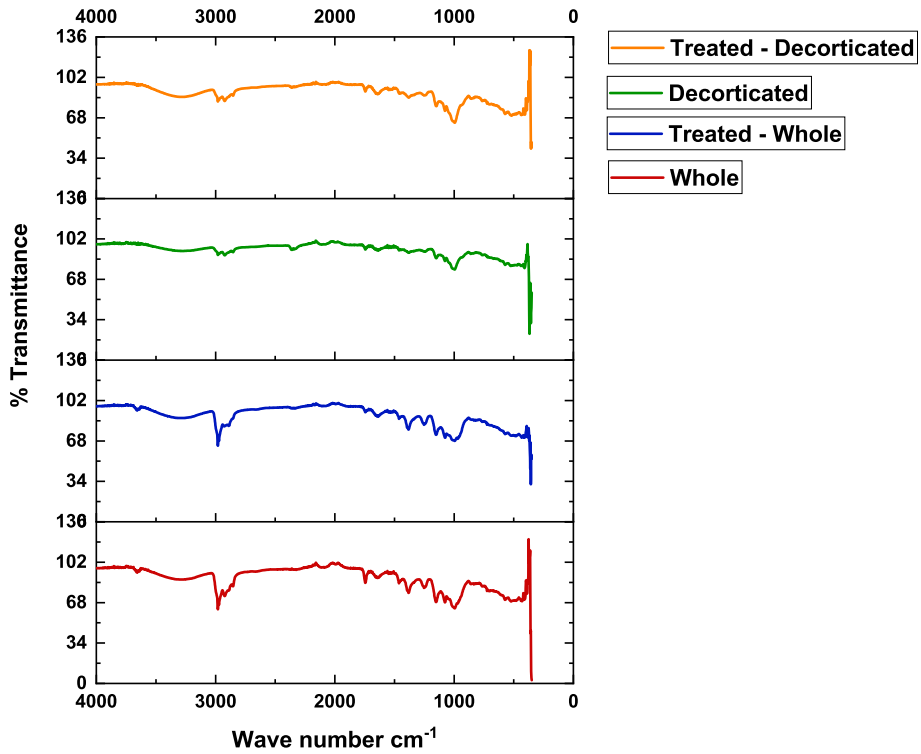

Fig. 2 FTIR spectrum of pearl millet flours

(Table 5). The compounds 2-Deoxy-2-fluoro-1,6-anhydro-β-d-glucopyranose, Methyl 4-nitrohexanoate, DL-Leucine, N-glycyl-, trans-13-Octadecenoic acid were observed to be newly formed compounds in treated pearl millet sample on 0^{th} day. The 2-Deoxy-2-fluoro-1, 6-anhydro-β-d-glucopyranose is produced due to protein-carbohydrate interaction during maillard reaction occur due to thermal treatment (Huonnic and Linclau, 2022). The Methyl 4-nitrohexanoate is formed as a product of Michael addition reaction between nitropropane and methyl acrylate. The increase in compounds was observed on storage up to 60 days (Tables 4 and 5). The result after 60 days of storage showed increase in various fatty acids indicating the various oxidative and lipolytic rancid reactions. The increase in various secondary oxidation products such as aldehydes and ketones were observed which are responsible for the development of off odors on storage. The fatty acid on oxidation gets converted into saturated fatty acids. The long chain fatty acids get degraded into various short chain saturated fatty acids which produces undesirable flavors to the products and makes the unacceptable for consumption (Vinutha et al. 2022). Apart from the fatty acids phytosterols also gets affected due to the oxidation process. The unsaturated bonds present in Sitosterol present on 0^{th} days gets converted into Stigmasterol through a series of enzyme catalyzed mevalonate pathway. The phytosterol also gets effected due to oxidation and produces new compounds such as Stigmasterol, Campesterol and Stigmastanol (Kmiecik et al. 2021). The oxidized products of phytosterols causes undesirable changes and shows potential toxicity (Kim et al. 2014).

Following a 60-day storage duration, a notable observation was the presence of newly formed compounds, specifically, peroxide derivatives of lipids and free fatty acids. These compounds are recognized byproducts of lipid oxidation processes (Aher et al. 2022). Investigating into existing literature further strengthens the connection, affirming that such compounds are frequently involved in rancidity and are, therefore, identified as key contributors to the development of the rancid flavor. The number of secondary oxidation products formed in the treated sample are lower compared to control sample after 60 days of accelerated storage indicating the lower oxidation in treated sample owed to inhibition of lipase activity in treated sample. The treated millet flour did not produce any off flavors up to 60 days of accelerated storage and has acceptable

Table 4 Flavour compounds found in flour from decorticated raw pearl millet grains (control) on 0^{th} and 60^{th} day of accelerated storage

Compound	Retention time (min)	Formula	Molecular Weight (g mol^{-1})	Peak area (%)
Control 0^{th} Day				
2,4,4-trimethylpentyl ethylphosphonofluoridate	8.122	$C_{10}H_{22}FO_2P$	224.1	2.47
Sucrose	9.069	$C_{12}H_{22}O_{11}$	342.3	5.22
Melezitose	9.086	$C_{18}H_{32}O_{16}$	504.4	4.96
2-Hydroxy-3-methylsuccinic acid	9.974	$C_5H_8O_5$	148.11	3.79
2,4-Di-tert-butylphenol	10.217	$C_{14}H_{22}O$	206.32	0.49
n-Hexadecanoic acid	15.676	$C_{16}H_{32}O_2$	256.42	13.42
7,10-Octadecadienoic acid, methyl ester	17.54	$C_{19}H_{34}O_2$	294.5	0.51
9,12-Octadecadienoic acid (Z,Z)	18.085	$C_{18}H_{32}O_2$	280.4	18.47
cis-13-Octadecenoic acid	18.157	$C_{18}H_{34}O_2$	282.5	21.49
Dasycarpidan-1-methanol, acetate (ester)	18.434	$C_{20}H_{26}N_2O_2$	326.4	0.58
Butyl 9,12-octadecadienoate	20.859	$C_{22}H_{40}O_2$	336.6	0.42
9,12-Octadecadienoic acid (Z,Z)-, 2-hydroxy-1-(hydroxymethy) ethyl ester	26.054	$C_{21}H_{38}O_4$	354.5240	3.27
Ethyl iso-allocholate	35.086	$C_{26}H_{44}O_5$	436.6	2.68
γ-Sitosterol	37.475	$C_{29}H_{50}O$	414.7067	8.07
Control 60^{th} Day				
Thymine	3.8219	$C_5H_6N_2O_2$	126	0.72
2,4,4-trimethylpentyl ethylphosphonofluoridate	8.1020	$C_{10}H_{22}FO_2P$	224.1	0.45
d-Glycero-d-ido-heptose	9.2750	$C_7H_{14}O_7$	210.1	2.86
2,4-Di-tert-butylphenol	10.2020	$C_{14}H_{22}O$	206.2	0.93
Hexadecanoic acid, methyl ester	15.2031	$C_{17}H_{34}O_2$	270.3	0.25
Palmitoleic acid	15.4205	$C_{16}H_{30}O_2$	254.2	0.36
n-Hexadecanoic acid	15.7753	$C_{16}H_{32}O_2$	256.2	19.91
9,12-Octadecadienoic acid (Z,Z)-, methyl ester	17.5148	$C_{19}H_{34}O_2$	294.3	0.63
11-Octadecenoic acid, methyl ester	17.5891	$C_{19}H_{36}O_2$	296.3	0.41
9,12-Octadecadienoic acid (Z,Z)-	18.2872	$C_{18}H_{32}O_2$	280.2	46.47
9-Octadecenoic acid, (E)-	18.3330	$C_{18}H_{34}O_2$	282.3	15.12
Octadecanoic acid	18.5333	$C_{18}H_{36}O_2$	284.3	1.75
[1,1′-Biphenyl]-2,3′-diol, 3,4′,5,6′-tetrakis(1,1dimethylethyl)-	21.4401	$C_{28}H_{42}O_2$	410.3	0.33
Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester	23.4771	$C_{19}H_{38}O_4$	330.3	0.80
9,12-Octadecadienoic acid (Z,Z)-, 2-hydroxy-1-(hydroxymethyl)ethyl ester	26.0448	$C_{21}H_{38}O_4$	354.3	2.72
γ-Tocopherol	31.3735	$C_{28}H_{48}O_2$	416.4	0.89
Campesterol	35.0013	$C_{28}H_{48}O$	400.4	1.06
Stigmasterol	35.8310	$C_{29}H_{48}O$	412.4	0.46
γ-Sitosterol	37.4046	$C_{29}H_{50}O$	414.4	3.53

sensory properties (data not shown) indicating the shelf stability of treated millet flour.

Conclusion

The storage stability of flour from treated pearl millet at 15%, 15 min was enhanced as compared to untreated decorticated raw one. During the entire accelerated storage (60 days), peroxide value and free fatty acid values of flour from HARF treated one were always lower than flour from decorticated raw pearl millets. HARF processing (15% moisture, 15 min exposure) retained the functional attributes of pearl millet appreciably. The difference in the FTIR spectrum between treated and untreated samples was significantly lower indicating the minimal effect of RF treatment on structural properties. The number of secondary oxidation products formed in the treated sample were lower compared to control after 60 days of accelerated storage indicating the lower

Table 5 Flavour compounds found flours from HARF treated pearl millet grains (15%, 15 min) on 0^{th} and 60^{th} day of accelerated storage

Compound	Retention time (min)	Formula	Molecular Weight (g mol^{-1})	Peak area (%)
Treated 0^{th} Day				
2-Deoxy-2-fluoro-1,6-anhydro-β-d-glucopyranose	5.025	C_6H_9FO	164.13	0.85
Methyl 4-nitrohexanoate	8.118	$C_7H_{13}NO_4$	175.18	3.66
Sucrose	9.036	$C_{12}H_{22}O_{11}$	342.3	12.84
DL-Leucine, N-glycyl	9.963	$C_8H_{16}N_2O_3$	188.22	6.4
2,4-Di-tert-butylphenol	10.218	$C_{14}H_{22}O$	206.32	1.16
n-Hexadecanoic acid	15.664	$C_{16}H_{32}O_2$	256.42	14.53
9,12-Octadecadienoic acid (Z,Z)	18.065	$C_{18}H_{32}O_2$	280.4	15.4
trans-13-Octadecenoic acid	18.142	$C_{19}H_{36}O_2$	282.5	22.01
Butyl 9,12-octadecadienoate	26.056	$C_{22}H_{40}O_2$	336.6	2.6
Ethyl iso-allocholate	35.085	$C_{26}H_{44}O_5$	436.6	4.34
γ-Sitosterol	37.46	$C_{29}H_{50}O$	414.7067	15.59
Treated 60^{th} Day				
d-Glycero-d-ido-heptose	9.2576	$C_7H_{14}O_7$	210.1	3.25
2,4-Di-tert-butylphenol	10.2017	$C_{14}H_{22}O$	206.2	0.93
Hexadecanoic acid, methyl ester	15.2028	$C_{17}H_{34}O_2$	270.3	0.25
n-Hexadecanoic acid	15.7693	$C_{16}H_{32}O_2$	256.2	20.06
9,12-Octadecadienoic acid (Z,Z)	18.2583	$C_{18}H_{32}O_2$	280.2	46.55
9-Octadecenoic acid, (E)	18.3213	$C_{18}H_{34}O_2$	282.3	15.04
Octadecanoic acid	18.5158	$C_{18}H_{36}O_2$	284.3	1.29
Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester	23.4711	$C_{19}H_{38}O_4$	330.3	0.80
9-Octadecenoic acid (Z)-, 2,3-dihydroxypropyl ester	26.0689	$C_{21}H_{40}O_4$	356.3	1.88
γ-Tocopherol	31.3675	$C_{28}H_{48}O_2$	416.4	0.84
Stigmasterol	35.8307	$C_{29}H_{48}O$	412.4	0.46
γ-Sitosterol	37.3986	$C_{29}H_{50}O$	414.4	3.57

oxidation in treated sample. Therefore, continuous HARF technology could be used for pearl millet flour storage stabilization with better functional attributes retention. The present study helps in value addition and promotes the consumption of nutricereals, which are underutilized due to the rapid rancidity development.

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Authors' contributions
Srinivas Yarrakula: Conceptualization, Designed the Study, Performed all the experiments, Writing Original draft, Gopinath Mummaleti: Writing, Review and Editing, Kavya Sree Toleti: Writing, Shanmugasundaram Saravanan: Supervision.

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