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# Effect of incorporating hydrothermal, kilned and defatted oats on antioxidant and chapatti making properties of wheat flour

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ABSTRACT

Oats were subjected to treatments like defatting, hydrothermal cooking and kilning, milled into flour and then the control and treated flours were incorporated into wheat flour at 25% and 50% levels and chapatti making behaviour and antioxidant properties were studied. The treatments significantly affected the antioxidant properties of oats. Incorporating oat flours to wheat increased total phenolic content but lowered the antioxidant activity however both were decreased significantly upon baking. The reducing power of the oat blended flour was higher than the wheat flours and ranged from 8.0 to 15.5 μmol AAE/g and was further increased upon baking. The metal chelating activity of flour blends varied from 62.0% to 73.8% and further increased upon baking. After baking the total flavonoid content was lowered and ranged from 308 to 389 μg CE/g. The non-enzymatic browning index significantly increased up to 27.6% upon baking.



## 1. Introduction

In India almost 1.0 lakh hectares of land is under oat cultivation as a fodder crop and very little is utilised in human foods (Gujral, Sharma, and Singh (2011). Today oats are receiving increased interest because of their excellent health related properties. They are a rich source of soluble fibre, balanced proteins, vitamins and minerals, which are essential for human health. When used as a human food they are rolled or crushed into oatmeal and eaten as porridge or ground into fine oat flour and used in a variety of baked goods, such as oatcakes, oatmeal cookies, and oat flour for baby foods. Oats contain high content of dietary fibre β-glucan, minerals and antioxidants (tocols, phytic acids, phenolic compounds and avenanthramides). High content of β-glucan in oats make it a functional and nutraceutical food as it has been reported to be effective in reducing serum cholesterol concentration and postprandial blood glucose level. Phenolic compounds are important phytochemicals in grains and function as free radical scavengers and are involved in reducing the risk of atherosclerosis, prevent some forms of cancer and coronary heart disease (Emmons & Peterson, 1999).

Majority of the wheat grown in India i.e., nearly 80.7 million tonnes is consumed mainly in the form of unleavened flat bread known as chapatti (Gujral & Pathak, 2002; Gujral & Gaur, 2002). Chapatti is usually prepared from whole wheat flour and the desired quality parameters in chapatti are greater pliability, soft texture, light creamish brown colour, slight chewiness and baked wheat aroma (Gujral, 2010). Oat flour could be incorporated into wheat flour to make chapatties thus promoting consumption of oat and also increasing the nutraceutical potential of chapatti.

However the high fat content in oats make it particularly sensitive to oxidation, resulting in a rancid flavor caused by the non-volatile free fatty acids (Welch, 1995). The problem related to the high lipid content may be overcome by defatting the oat before processing. Alternatively, kilning and hydrothermal treatment may also be given to the oat before milling of groats to inactivate the lipase enzyme. Thus oats could be subjected to treatments like defatting, hydrothermal treatment or kilning and the oat flour could be blended with wheat flour to make chapatti and chapatti used as a delivery vehicle for the bioactive components especially oat glucans and antioxidants. The objectives of the present investigation were to study the effect of incorporation of oat flour subjected to different treatments on chapatti making behaviour and antioxidant properties.

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## 2. Materials and methods

### 2.1. Samples and chemicals

The oat variety OL-125 was collected from Punjab Agricultural University, Ludhiana, Punjab, India. The grain was cleaned before dehulling. The wheat flour of Ashirwad brand (ITC ltd, India) was procured from local market.

Standard ferulic acid, 2,2-diphenyl-1-picrylhydrazyl (DPPH), ferrozine, protease (from *Streptomyces griseus*) and catechin were procured from Sigma–Aldrich (Steinheim, Germany). L-ascorbic acid, potassium ferricyanide, ferric chloride, ferrous chloride, trichloroacetic acid, sodium carbonate and Folin Ciocalteu's reagent were procured from Loba Chemie, Mumbai, India. All chemicals were of analytical grade. Each test was performed in triplicates on dry weight basis. The Milli Q water (Millipore, France) was used for all analytical tests.

### 2.2. Defatted oat flour

The dehulling of oat was carried out in an impact dehuller (Lab Impact 1, Creative India, Mohali, Punjab) as described by Gujral et al. (2011). Groats were separated from husk manually. The oat groats were coarsely ground to form grit in a Newport Super Mill. Grit was mixed with hexane (1:5) in volumetric flask and placed overnight on a shaker. The contents were allowed to settle and the supernatant layer of solvent discarded, followed by a washing with the solvent (1:2) The defatted grit was dried in oven at 50 °C to remove solvent and then ground to flour in Super Mill 1500 (Newport Scientific, Australia) and passed through 52 (BSS) sieve. Any fraction retained on the sieve was reground till all of it passed through the sieve so as to obtain a flour of 100% extraction and the flour was packed in airtight bags and stored at 4 °C.

### 2.3. Hydrothermal treated flour

The oats were conditioned to a moisture content of 25%, and then steamed at atmospheric pressure for 45 min in an autoclave (Narang Scientific, New Delhi, India). The steamed oats were then dried at 50 °C to remove moisture, dehulled in impact mill and then the groats milled into flour.

### 2.4. Kilned oat flour

The oats were conditioned to a moisture content of 25%, then kilned at 120 °C for 1 h in a hot air oven. The oats were cooled to room temperature and then dehulled in the impact mill and then the groats ground into flour as described above.

### 2.5. Preparation of chapatti

The flour blends were prepared by replacing wheat flour with control or treated flour at levels of 25% and 50%. Preliminary trials were carried out to determine the amount of water to be added to the flour to develop non sticky viscoelastic dough that could be easily rolled and sheeted to make a chapatti. The flour was mixed with optimum water for three minutes in a laboratory mixer (National Manufacturing Company, Lincoln, USA). The dough was left to rest for half an hour. Dough ball (50 g) was rounded and then placed on a rolling board and was sheeted. The dough was rolled in one direction, inverted, and then rolled in a perpendicular direction, placed on an electric hot plate at 280 ± 5 °C and baked into chapatti. The baking time was 60 s for control, 73 s for defatted, 89 s for hydrothermal treated and 75 s for kilned oats. The chapatti was allowed to cool for 10 min at 25 °C and then freeze dried (Sim International, USA) finely ground and packed in polyethylene pouches and placed in an air tight container and stored at 4 °C till further analysis.

### 2.6. Total phenolic content (TPC)

The total phenolic content was determined according the Folin–Ciocalteu specterophotometric method explained by Gao, Wang, Oomah, and Mazza (2002). Samples (200 mg) were extracted with 4 ml acidified methanol (HCl/methanol/water, 1:80:10, v/v/v) at room temperature (25 °C) for 2 h using wrist action shaker (Narang Scientific, Delhi, India). The mixture was centrifuged at 3000 g for 10 min on a centrifuge (Eltek, RC 4100 F, Mumbai, India). The supernatant was used for determination of total phenolic content. Aliquot of extract (200 μl) was added to 1.5 ml freshly diluted (10-fold) Folin–Ciocalteu reagent. The mixture was allowed to equilibrate for 5 min and then mixed with 1.5 ml of sodium carbonate solution (60 g/l). After incubation at room temperature (25 °C) for 90 min, the absorbance of the mixture was read at 725 nm (Shimadzu, UV-1800, Kyoto, Japan). Acidified methanol was used as a blank. The results were expressed as μg of ferulic acid equivalents per gram of flour.

### 2.7. Antioxidant activity (AOA)

Antioxidant activity was measured using a modified version of the method explained by Brand-Williams, Cuvelier, and Berset (1995). This involved the use of free radical 2,2-diphenyl-1-picrylhydrazyl (DPPH) solution in the methanol. Samples (100 mg) were extracted with 1 ml methanol for 2 h and centrifuged at 3000*g* for 10 min. The supernatant (100 μl) was reacted with 3.9 ml of a $6 \times 10^{-5}$ mol/L of DPPH solution. Absorbance (A) at 515 nm was read at 0 and 30 min using a methanol blank. Antioxidant activity was calculated as% discoloration.

$$\%\text{Antioxidant activity} = (1 - (\text{A of sample t} = 30/\text{A of control t} = 0)) \times 100$$

### 2.8. Reducing power

The reducing power was measured as described by Zhao et al. (2008). Flour blends (0.5 g) was extracted with 80% methanol on wrist action shaker for 2 h. The supernatant was collected after centrifugation at 10000*g* for 10 min. The supernatant (1 ml) was mixed with phosphate buffer (0.2 mol/l, pH 6.6) and 2.5 ml potassium ferricyanide (1%) was added. The mixture was allowed to stand for 20 min at 50 °C in an incubator. After incubation, 2.5 ml of trichloroacetic acid solution was added to mixture, which was then centrifuged at 10000*g* for 10 min. The upper layer of solution was mixed with 2.5 ml deionized water and 0.5 ml ferric chloride (0.1%). The absorbance of the mixture was measured at 700 nm. Increased absorbance of the mixture indicated increased reducing power. A standard curve was prepared using various concentration of ascorbic acid. The results were expressed as μmol ascorbic acid equivalents/g.

### 2.9. Metal chelating ($Fe^{+2}$) activity

The metal chelating activity of extract was measured as reported by Dinis, Madeira, and Almeidam (1994). The extract was mixed with 50 μl of ferrous chloride (2 mM/l) and 1.6 ml distilled water was added. After 5 min, the reaction was initiated by the addition of ferrozine (100 μl) and the mixture was shaken on vortex. Further the mixture was incubated at room temperature (25 °C) for 10 min. Absorbance of solution was measured at