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Review Article

Whole Wheat Flour Stability: An Insight

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Abstract

In the current scenario, millers as well as food Industries face many challenges in relation to the whole-wheat flour due to its limited stability in comparisons to refined flour. Lipid degradation by the action of enzymatic as was well non-enzymatic pathways seems to be a one of the leading causes of whole wheat flour stability. Milling technique is also of paramount importance to produce whole Wheat flour. To address the stability issues, retentions of available antioxidant during milling and inactivation of enzymes can be explored along with wheat bran stabilization. The purpose of this review paper is to address these key issues in the milling, shelf-life, key biomarkers of whole wheat flour and finally discuss the strategies to overcome existing challenges.

Keywords: Whole Wheat Flour; Flour; Rancidity; Bran; Germ; Oxidation; Milling; Stone Mill; Roller Mill; Milling Techniques; Jet Milling; Enzymes; Refined Flour; Stability

Whole Wheat Flour Milling: Issues and Challenges

AACC International has defined whole wheat flour as being prepared from wheat (other than durum) such that the proportions of the intact grain, the bran, germ, and endosperm remain unaltered. Whole wheat flour contains substantially more vitamins, minerals, antioxidants and other nutrients than regular wheat flour, since these compounds are concentrated in the outer portions of the grain. Some of these nutrients are replaced in the enrichment process of wheat flour, which is mandatory in 64 countries around the world [1] although many nutritional components are still lower, especially minerals and dietary fiber [2].

Whole wheat flour possesses several unique challenges to the milling and related industries. For instance, whereas milling procedures for traditional flours have been well-established, whole grain flours are produced by a variety of techniques and result in flours with widely different particle sizes and functionalities [3]. Furthermore, whole wheat flour contains more enzymatic activity [4], lipids [5,6], and antioxidants [7] than wheat flour, which can affect end-use [8] and storage properties [9-12].

Studies on the Whole-Wheat Degradation

In the past decade, enough work has been carried out in the area of whole wheat degradation. In this section, we will try to discuss the research studies in the area of whole grain stability.

In one of the past work the effect of storage temperature (4, 20, and 30°C) on whole grain flour (WGF) storage for up to 12 months was studied [13]. Authors found significantly higher fat acidity caused by (partial) hydrolysis of triacylglycerols (TAG) yielding free fatty acids (FFA) in samples stored at higher temperature as compared to samples stored at 4°C.

Within the context of whole wheat flour storage, the presence of nonpolar lipids is one of the important factor to consider [14]. Nonpolar lipids predominantly occur in the germ and aleurone tissues of the wheat kernel and consist of FFA, monoacylglycerols (MAG), diacylglycerols (DAG), and, mainly, TAG [15]. Wheat milling transfers approximately 50% of the TAG of germ to flour [16]. During WGF storage, lipase (EC 3.1.1.3) hydrolyzes part of the TAG lipids [2], yielding DAG, MAG, and FFA. The latter can be responsible for what is referred to as acid rancidity. Acid rancidity can decrease WGF functional properties and affect the product such as noodles leading to onset of rancidity. In addition, FFA occurring as such are more susceptible to enzymatic oxidative conversion than when they are esterified to glycerol and, hence, present as part of TAG [17]. Released polyunsaturated FFA can be oxidized by wheat lipoxygenase (EC 1.13.11.12), which is mainly located in the germ and bran. It has been observed that the oxidation of lipid during whole wheat flour storage is relatively slower than the preceding lipid hydrolysis, because of the fact that whole wheat flour contains higher levels of antioxidants such as vitamin E which protect the lipid oxidation [8]. With reference to the loss in the sensory acceptability, it has been stated that FFA\acid value are the major cause that leads to typical off flavour due to lipid rancidity. Indeed, accumulation of FFA and volatile compounds related to subsequent lipid oxidation often gives it an off taste [18,19].

Within a similar context, oat processors execute groat roasting, kilning, or steaming treatments before further processing to control the lipid oxidation. The lipid content of oat groats varies from as low as 3.1% to as high as 11.6%, with most cultivars containing 5 - 9% lipids [20]. As is the case for whole wheat flour, the main factor limiting the storage and handling possibilities of raw oats is lipolysis followed by oxidation, which leads to rancid off-flavors and, hence, inadequate shelf life [21]. Heat inactivates enzymes in the kernel, such as lipase, lipoxygenase, and peroxidase [22]. By implementing oat heat processing technology into wheat flour production systems, it may well be possible to control rancidity through inactivation of lipase. Some of the studies related to the heat processing are discussed below.

In one of the work, wheat bran was roasted at 175°C for 40 minutes. Authors noticed that lipase, lipoxygenase, and protease activities were reduced by 40, 100, and 50%, respectively. Similar work was carried out to compare the effect of dry heat, steam, and microwave treatments of wheat bran on both its lipase. It was found that all treatments effectively decreased lipase activity. Optimal conditions (sample size, 203 g of bran), in as much as further heating did not decrease lipase activity, were 25 minutes of dry heating at 175°C, 60s of microwaving (1,000 W), or 60s of steaming. Microwave and steam treatments were more effective at decreasing lipase activity than dry heat (93 and 96%, versus 74% lipase activity decrease, respectively).

During storage of whole wheat flour containing heat-treated bran, lower levels of free fatty acid (FFA) were found than in whole wheat flour containing control bran, with the rates of FFA release in each of the treated samples correlating well with the measured lipase activity.

In one of the studies, rate of hydrolytic and oxidative degradation of lipids during whole wheat flour storage was evaluated [23]. The rancidity was measured in terms of the rate of oxygen uptake. It was found that there was linear relationship between the rate of oxygen uptake with storage at 20°C. This trend was positively correlated with the increases in unesterified fatty acids. Also the rate of oxygen uptake was found to be positively correlated with the increase in the relative humidity. The increase in relative humidity was not due to microbial growth.

In terms of oxygen uptake with different grain components, the rate of deterioration of bran over 30 days at 20°C was five-fold greater than that of a relatively pure germ fraction, but was only half the rate of a 5:1(w/w) blend of bran and germ. In conclusion it was found that increases in the rate of O_2 uptake and fatty acid content were highly correlated. Fine milling of bran, germ and blends increased the rate of deterioration and enhanced the observed synergistic effect between bran and germ on both 0_2-uptake and fatty acid increases. Lipid analyses of bran-germ blends supported the conclusion that, during storage ,there is a relatively slow (over several weeks) release of fatty acids, catalyzed by a lipolytic enzyme in the bran component of whole meal, but that lipid oxidation occurs rapidly (within minutes) when excess water is added ,facilitating lipoxygenase-catalyzed peroxidation of polyunsaturated fatty acids.

One of the unique observation that was found in this study was that when the flour was hydrated there was relatively rapid and extensive oxidation. Due to this oxidation, O2 depended lipoxygenase catalyzed degradation of polyunsaturated fatty acids was found out to occur. It was observed that even though reaction is initiated by TG acyl hydrolase (lipase) activity in the bran component of whole meal, degradation due to oxidation depends primarily upon the lipoxygenase activity, mainly in the germ, and on a supply of TG from germ, endosperm and outer layers of wheat grain.

Since the oxidation of lipids was found out to be more progressive in case of hydrated whole wheat flour, selection of key lipid oxidation products (e.g. peroxide value, thiobarbituric acid (TBA) test or 0_2-uptakeof dry materials) may not be suitable for the products that are hydrated before processing. Direct measurement of the TG lipase activity, measurement of unesterified fatty acids (FFA, fat acidity) or the most rapid and direct measurement of rates of 0_2-uptakeby aqueous suspensions are likely to be more useful predictors of potential rancidity. However this area need future investigation to establish firm relationship between the oxygen uptake and rancidity development.

Type of Milling and Impact on Whole Wheat Stability

In selection of the whole grain flour one of the prime consideration should be milling process. For example, milling technique may have a greater impact on whole wheat bread quality than the quality of wheat used for producing the flour or the formulation of the bread itself [24]. So the impact of milling is not only limited to the flour but also extend to the product derived from the raw material.

Currently stone and roller mill are the two predominant techniques for grinding whole grain flours. Whole grain flours could also notionally be produced with an impact or hammer mill, however these techniques are rarely used. In the recent past, work has been carried out to understand the effect of different milling methods on the whole wheat flour. In one such, effect of milling was studies on the chemical composition of whole wheat flour. In order to assess the effect of different types of milling methods on protein and lipid composition of whole wheat flour, two types of wheat varieties belonging to strong and weak wheat type were selected and milled in different mills such as plate, hammer, stone and roller mills. The temperatures generated during grinding of wheat in stone, plate, hammer and roller mills were 90, 85, 55 and 35°C respectively. The studies on SDS PAGE indicated degradation in proteins of whole wheat flour obtained from stone and plate mills, especially in the high molecular glutenin regions. Greater loss of total amino acids was also observed in the above milled flours when compared with that of hammer and roller milled samples. Free lipid content was lower in flours milled in stone and plate mills when compared with that of flours milled in other mills. Unsaturated fatty acid content, particularly linolenic acid, was lower in stone milled flour (1.3%) followed by plate mill (2.2%), hammer mill (2.8%) and roller flour mill (3.8%). The trends in the above values as influenced by different milling methods remained similar both in the weak and strong wheat types.

In one of the recent studies, chapatti making quality of whole wheat flour (atta) obtained by various processing techniques was evaluated [25]. Wheat was processed in chakki (CM), hammer (HM), disk (DM), pin (PM) and roller mill (RM) with an objective of quality characterization of whole wheat flour (atta) in relation to chapatti making quality. Results indicated atta produced from RM was cooler and had retained more moisture. Ash content was not significantly influenced by different grinders; however, acid insoluble ash was higher at 0.063% for the atta produced from CM. Variation in damaged starch was observed which was 15.99, 13.76, 11.76, 10.16 and 9.1% for CM, HM, DM, PM and RM ground atta, respectively. Farinograph water absorption of CM ground atta was highest at 85% and least for RM ground atta (71.5%). Overall quality of chapatti prepared from CM atta scored higher and had better texture and desirable wheaty aroma. Studies revealed that Atta quality parameters and its chapatti making quality varied with processing techniques.

Stone Milling

Stone mills are the oldest attrition mills used for making whole grain flours, which simultaneously use compression, shear, and abrasion to grind wheat kernels between two stones and produce a theoretical extraction rate of 100% [24]. Traditional stone (Chakki) Mills consisting of 2 dressed stone discs (one stationary, other rotating). Stone mills generate considerable heat due to friction Due to this it undergoes higher maintenance cost (high abrasion) and subsequently lower shelf life.

It has been found that the high abrasion in the stone mill leads to considerable damage to starch, protein, and unsaturated fatty acids in comparison with other milling techniques [26]. This leads to the low stability of the whole wheat flour using the stone mill. Breakdown of the unsaturated fatty acids in the whole wheat flour normally results into rancidity. It has been observed that there is higher breakdown of the unsaturated fatty acids in the stone mill as compared to the roller flour mills. This could be possibly due to different types of forces (Compression/Shear/Attrition) on the acting during the process of grinding.

There are certain reports that describes the effect of type of milling on the mycotoxins content of the wheat flour. In one of published work it was found that stone milling reduced vomitoxin and zearalenone content in flours, compared with the use of the roller-mill system.

Roller Milling

The process of roller milling involves separation of the endosperm from the bran and germ followed by gradual size reduction of endosperm. In this process, wheat is passed through a series of corrugated and smooth rollers accompanied by sifting between stages. Producing flour that fulfills the requirement for being whole grain is achieved by blending bran and germ back with the endosperm flour in the naturally-occurring proportions. Feeding the bran and germ milling streams with the endosperm flour stream is most often achieved in a continuous process, rather than collecting all fractions in separate bins and recombining at the end of milling. In this case, production of whole wheat flour would not involve additional capital expense beyond what is required for regular roller milling. Sometimes whole wheat flour is made by physically separating flour millstreams and then recombining at the end of the milling process. This is usually done when the bran will undergo some post-milling such as ultra fine grinding or heating. In these cases, capital costs would be required for the post-milling, plus equipment for recombining the fractions. When producing whole wheat flour on roller mills, a number of conditions are different from those used for wheat flour [27].

First, conditioning (tempering) is less important when milling whole wheat flour. While wheat flour relies on proper conditioning to facilitate endosperm and bran separation, this is not required for whole wheat milling. Thus, in theory no conditioning should be required, although many mills will add 1e2% moisture to soften the grain and improve efficiency in terms of the energy required to produce the flour. Efficiency can also be improved by tightening the roll gap and using more open scalp covers to increase the break release, as well as changing some of the smooth rolls to corrugate during reduction [26].

The purifier air valves should also be adjusted so that the bran and germ are not rejected but are returned to the reduction system. There are several noteworthy advantages of making whole grain flour from roller mills as opposed to stone mills. First, the amount of grinding and reduction at each roll can be adjusted to accommodate variations in raw materials, which makes roller milling both economical and flexible. Second, the use of selective corrugations and differential speeds subjects the endosperm fraction to minimal shear and compressive forces during the grinding and reduction, which allows less heat to build on reduction rolls and results in less destruction to chemical components in the flour. A third advantage of making whole grain flours from roller mills is that wheat bran and germ can be separated from the endosperm fraction and subjected to further processing such as heating or fine grinding to affect the storage or functional properties of the flour.

Jet Milling

Nowadays, alternative milling procedures and micronizing technologies are tested in order to produce flours with enhanced functional properties, which are suitable for making new edible products or for improving the properties of the current ones. Milling technologies focused on producing finer flours with improved properties are getting increased attention [28,29]. Jet milling is one such latest technological development that aims at the production of super fine flours by accelerating the particles in a high-velocity air stream, the size reduction being the result of inter-particle collisions or impacts against solid surface [29,30].

The particles impact at high velocities produces superfine powders and reduces the size of all aggregates. It is a fluid energy impact-milling technique, commonly used to produce particle sizes lower than 40 μm, which are greatly appreciated in the chemical, pharmaceutical and mineral industry. In food applications, smaller particle size results in faster starch digestion. Small particles have high surface-to-volume ratio increasing the access of enzymes to the interior of the particle taking advantage of the absence of intact cell walls. An increased surface area of food materials could increase the rate of water absorption of materials, improving solubility of dry products and increase site accessibility for chemical reactions (e.g. oxidation, digestion, flavor release, catalyst, and enzyme activity). Jet milling combined with air classification has been successfully used to separate starch from protein in order to produce starch-rich fine flours.

Furthermore, differential scanning calorimetry showed lower gelatinization enthalpy values for the doughs (flour: water, 60:40) of fine flours than their coarse flour counterparts (6.76 - 7.09 and 9.92 - 10.12 mJ/mg respectively. Overall, particle size of wheat flour seems to have an impact on dough mechanical and starch gelatinization properties. Therefore, there is a consensus that particle size reduction promotes changes in the majority of physicochemical properties due to the increase of a particle's surface area, although it must be assessed if there is a critical point that leads to an increase of damaged starch.

The higher the specific surface area per weight unit, the higher the rate of hydration and water absorption is. Generally, starch granules become physically injured with milling's shearing and scrapping, i.e., starch damage occurs which could also increase water holding capacity. Moreover the production of ultrafine powders from cereals flours may present benefits to human health. Jet milling may be useful for modifying or improving functionality and availability of bioactive compounds. However studies relating to the impact of jet milling on the storage stability of whole wheat flour is scare. This area need future investigation.

Distribution of Enzymes in Wheat Flour Mill

Wheat flour contains several technologically important enzymes such as amylases, proteases, lipoxygenase, polyphenol oxidase and peroxidase [4]. In the wheat grain alpha-amylase is located mainly in the pericarp with small quantities present in the aleurone layer and the seed coat1. Protease is concentrated in the endosperm, germ and aleurone layer [31,32]. The scutellum and embryo are rich in lipoxygenase. Polyphenol oxidase and per-oxidase are predominant in bran layers. Although these enzymes are inactive during storage of grain and flour, when water is added they become active and play a significant role in determining the functional attributes of flour.

The aim of roller flour milling is the gradual reduction of the wheat kernel through a series of break and reduction rolls. This results in the production of different types of flour streams containing endosperm, bran and germ in varying proportions. Thus the knowledge about the enzyme distribution in the different steams of whole mill can possibly help to design appropriate treatment of different streams so as to minimize the overall enzymatic activity thereby resulting to higher flour stability.

The levels of activity of various enzymes differ in different flour mill streams and hence their functional properties are also different. Several reports are available on the protein and ash contents in different flour mill streams [33-35] and a few reports are available on the suitability of various mill streams for different bakery products. Some information is available on the activity of individual enzymes in mill streams. It has been reported in the past regarding the distribution of alpha-amylase in various mill streams of soft winter wheat's [36]. It has also been reported that proteolytic activity was found to be higher in tail end reduction streams as compared to other flour streams during the wheat milling operations [37]. In one of the report, polyphenol oxidase activity was found out to be linearly correlated with ash content and it was also reported that PPO was most active in bran- and germ-rich milling fractions. The distribution of lipoxygenase and peroxidase in flour streams has also been reported. Knowledge about the distribution of the enzymes can be useful in the preparation of blends either by selecting or omitting particular stream/streams for the preparation of specific mill fractions for use in different product category. This area need to be explored in the near future.

Differentiation of Whole Grain from Refined Wheat

Differentiation of a whole grain product from a refined grain product can be very challenging. This is also important with the context of identification of the key makers that can differentiate between the whole grain flour and refined flour. Since the food labels can often be misleading, the whole Grains Council has created an official packaging symbol, the Whole Grain Stamp, to help consumers. The 100% whole grain stamp assures you that a food contains a full serving or more of whole grain in each labelled serving and that all the grain used is whole grain, whereas the basic Whole Grain Stamp indicates the products contain at least 8 g of whole grain per labelled serving.

In one of the recent studies, differentiation of whole grain from refined wheat (*T. aestivum*) flour was done using Lipid Profile of Wheat Bran, Germ, and Endosperm with UHPLC-HRAM Mass Spectrometry [38]. A comprehensive analysis of wheat lipids from milling fractions of bran, germ, and endosperm was performed using ultrahigh-performance liquid chromatography−high-resolution accurate-mass multistage mass spectrometry with electrospray ionization (ESI) and atmospheric pressure chemical ionization (APCI) in both positive and negative modes. About 155 lipid compounds, including free fatty acids (FA), oxylipins, alk(en)ylresorcinols (ARs), γ-oryzanol, sphingolipids, triglycerides (TGs), diglycerides (DGs), phospholipids, and galactolipids were characterized from the three milling fractions. Galactolipids and phospholipids were proposed to be potential discriminatory compounds for refined flour, whereas γ-oryzanols, ARs, TGs, and DGs could distinguish whole wheat flour from a refined one based on principal component analysis (PCA). These key compounds could be also measured during the whole wheat flour storage.

In one of the similar work, differentiation of bread made with whole grain and refined wheat (*T. aestivum*) flour was done using LC/MS-based chromatographic fingerprinting and chemometric approaches [39] fuzzy chromatography mass spectrometric (FCMS) fingerprinting method combined with chemometric analysis was established to differentiate between breads made from whole wheat (WW) flour and refined wheat (RW) flour. The chemical compositions of the bread samples were profiled using ultra high performance liquid chromatography with high-resolution accurate-mass multistage mass spectrometry with atmospheric pressure chemical ionization (APCI) in positive ionization mode Principal component analysis (PCA) and soft independent modelling of class analogy (SIMCA) of the FCMS fingerprints revealed the components responsible for the chemical differences between WW and RW flour/bread samples. Alk(en)ylresorcinols (ARs) have been demonstrated to be the most important markers for differentiation between WW and RW flour/breads. Diglycerides (DGs), and phosphatidylethanolamine (PE) also been shown contributed significantly to the classification. In this particular there was no significant difference observed between the bread crumb and crust. It was concluded that SIMCA, using WW modelling, could be a potent and robust tool for authentication of WW breads.

Biochemical Markers: Efficient Tools for the Assessment of Wheat Grain Tissue Proportions in Milling Fractions

Identification of the key biomarkers in the whole grains would be enormously helpful in differentiating the whole-wheat flour from refined flour. Also, these biomarkers could be tracked during the whole-wheat storage so as to understand the changes within whole wheat flour. This sections discus some of the recent studies in relation to the biomarkers in whole grain tissues.

Numerous epidemiological studies have demonstrated the health benefits of consuming more whole-grain foods [40-42]. However, all the wheat grain parts are not health-promoting, for example the outermost parts have been shown to concentrate the majority of the grain contaminants, like microorganisms, mycotoxins, pesticide residues and heavy metals [43]. On the other hand, the wheat aleurone layer has been shown to have great nutritional interest, and to concentrate most of the minerals and vitamins of the wheat grain. It has been reported that wheat aleurone layer contains interesting proportions of proteins, β-Glucan, phenolic compounds and other phytochemicals (lignans, sterols).

In the past, few of the authors have also pointed out that that antioxidant, including phenolic acids, are concentrated in the aleurone layer of wheat bran, and one of the authors showed that the higher the proportion of aleurone material in wheat fractions, the higher the antioxidant capacity observed for these fractions [44]. In one of the similar research authors found that that aleurone-rich fractions exhibited better *in vitro* digestibility and colonic fermentability than wheat bran. Few others have observed that the digestibility of minerals, protein and non-starch polysaccharides is much higher in bran fractions rich in aleurone than in fractions rich in pericarp and testa. These studies suggest that it could be interesting to produce aleurone-rich fractions for use as food ingredients. As a consequence, new processes are required to be developed in order to exploit all the nutritional benefits of whole grain and to produce new wheat foods and wheat-based ingredients with enhanced nutritional quality. For example, depending on the desired product, a process can aim at discarding the pericarp to obtain whole grains containing less contaminants, while other processes may be developed in order to produce bran fractions highly concentrated in aleurone material It is often difficult to exactly monitor the distribution of the different grain tissues among fractions during processing, as no simple method exists to quantify the respective proportions of these tissues in fractions. However, the monitoring of tissue proportions in the different fractions is essential, as it allows to control the quality of the products and to consequently adapt the processes. Therefore quantitative tools are needed. Different compounds can be measured to evaluate bran contamination in wheat flours and fractions. Ash content and measurement of flour colour are widely used in the milling industry as indicators of flour purity. The amino acid composition of the various tissues was also studied but did not allow the quantification of the grain tissues in milling fractions.

Some authors suggested using the concentration of ferulic acid to quantify bran in flours and semolinas, and alkylresorcinols have more recently been shown to be good markers of wheat bran content in foods. Such analyses may be useful to evaluate the total outer layer content but they do not allow to distinguish between the different outer tissues. Another way to evaluate the different grain tissues in wheat fractions consists of the use of specific fluorescence properties of the outer layers. Indeed, the aleurone cell walls display blue fluorescence under UV-light, due to the presence of ferulic acid, whereas the pericarp shows green fluorescence under blue light.

Based on these fluorescence properties, commercial equipment has been developed to determine the amount of aleurone in flours. Multispectral fluorescence image analysis of grain sections coupled with classification techniques have also been developed to more precisely quantify the proportions of the different parts of the grain, but have not yet been applied to powdery samples. These imaging methods can be good tools for on-line use, but their main disadvantage would be their lack of specificity, as they do not allow to quantify other tissues than aleurone and pericarp. Moreover, all these methods allow determination of bran proportions in flours during milling, but they may not be adaptable to other fractionation systems (such as progressive abrasion or bran fractionation). In the past work, biochemical analyses of isolated wheat grain tissues were carried out and the differences in chemical composition between these tissues to assess the histological composition of technological fractions were determined. They used compounds such as phenolic acids, phytic acid, and starch as biochemical markers. Indeed, these compounds were shown to be either present exclusively in one part of the grain (starch in starchy endosperm), or present in greater amounts in one particular tissue (phytic acid in aleurone cell contents and some phenolic acids in the cell walls). These biochemical markers were used to determine the amounts of aleurone layer and pericarp in flours and other milling fractions, and in the samples obtained from a wheat bran fractionation process.

In one of the earlier work, authors used different markers for aleurone cell walls and aleurone cell contents to assess the histological composition of bran fractions and to evaluate the dissociation and the accessibility of aleurone cellular components. This method was reported to provide accurate quantification of the histological composition of samples and to be versatile, as it can be refined and adapted depending on the type of sample analyzed, from either bran or whole-grain fractionation. However, it did not allow the quantification of testa (this tissue was deduced by subtraction and thus was perhaps overestimated), and it neither allowed the detection of the germ. Having a marker for wheat germ would nevertheless be very useful as it is either a part of the grain that needs to be excluded to avoid lipid oxidation and rancidity, or a nutritionally interesting byproduct that could be followed during fractionation processes in order to get germ-rich fractions.

Current Methods Employed to Enhance Wheat Flour Stability

Antioxidants

During the storage of whole wheat flour, development of rancidity has been observed as early as 2 - 14 days subsequently after the milling operation. Also, some authors have suggested in the past the limits for whole-wheat flour storage of 15 - 60 days. In the recent work it has been reported that high levels of vitamin E (tocopherols, tocotrienols) in whole-wheat flour provide antioxidant protection during the first 22 days of storage and efficiently contrast the peroxidation process, but a longer storage period (10 months) induced the degradation of up to 24% of the total vitamin E amount. The study suggested the important role of antioxidant compounds, such as polyphenols, to preserve nutritional properties of whole-wheat flour during storage. In the production of wholegrain flour, the selection of the milling process is a key factor as it exerts a great impact on the quality of the final whole-wheat flour. Thus the milling operation has to be optimized produce the whole wheat flour with higher antioxidant activity.

Cereals have been known to contain a high amount of hydroxycinnamic acid (HCA) derivatives that render potential health benefits [8,45]. Commercial processing of cereals may lead to products with low value fractions such as hulls and polish waste. In general, hulls are removed prior to food production. However, these low value fractions may serve as potential sources of natural antioxidants at relatively high concentrations [44]. In oats antioxidant compounds are mostly concentrated in the bran as compared to that in the endosperm as shown by in vitro assays. It has been demonstrated in the past that oat pearling fractions containing different levels of bran layers have higher antioxidant activity and total phenolic content (TPC) in compared to those of the flour extracts. Also it has been shown that bran extracts of different wheat varieties exhibits significant antioxidant properties against free radical scavenging and metal ion chelation. Moreover, in the fish model system, different varieties of whole grains exhibits antioxidant activity against lipid peroxidation. For Example, the 'Akron' variety of wheat was found out to be highly effective in scavenging 2, 2-diphenyl-1-picrylhydrazyl (DPPH) radical and chelating Fe (II) in one of the research work. Few other researchers have reported the relationship between growing conditions and the antioxidant activities of the wheat grown at different locations. These authors reported that TPC, scavenging of DPPH radical and chelation of Fe (II) were significantly influenced by agronomic practices and environmental conditions. The antioxidant properties of whole grains, bran and aleurone layer of a Swiss red wheat variety was studied using free radical scavenging and metal ion chelation capacity. Thus, aleurone, bran and grains differed significantly in their antioxidant potential, TPC and phenolic acid composition. Moreover, the aleurone layer exhibited the highest antioxidant activity, TPC and content of phenolic acids.

Ferulic acid was reported to be the predominant phenolic acid accounting for approximately 57 - 77% of total phenolic acids present in wheat on a dry weight basis [44]. Ferulic acid content was positively correlated with scavenging of free radicals and TPC and hence may be used as a potential marker of wheat antioxidants. Plant phenolic compounds including phenolic acids, flavonoids and anthocyanins, among others, have also been recognized as conferring stability against autoxidation of vegetable oils. There is much interest in the use of crude phenolic extracts from fruits, herbs, vegetables, cereals and other plant materials in the food and supplement industry because they have been shown to retard oxidative degradation processes, especially those of lipids thereby improving the quality and nutritional value of food.

In of the studies, Antioxidant and free radical scavenging activities of whole wheat and milling fractions was done [46]. It was found that the milling of wheat afforded several fractions, namely bran, flour, shorts and feed flour. In addition, semolina was the end-product of durum wheat milling. Among different milling fractions the bran had the highest phenolic content while the endosperm possessed the lowest amount and this was also reflected in free radical and reactive oxygen species (ROS) scavenging capacity, reducing power and iron (II) chelation capacity of different milling fractions in the two cultivars. This study demonstrated the importance of bran in the antioxidant activity of wheat, hence consumption of whole wheat grain may render beneficial health effects.

Irradiation

Cereals and cereal products, like semolina (rawa), refined (maida) and whole-wheat flour (atta), commonly in pre-packed form, are sold in the retail market. However, their shelf-life is restricted to 6 ± 8 weeks because of insect infestation. In tropical countries, like India, adverse climatic conditions of high temperature and humidity result in insect proliferation, even in sealed pouches. The conventional method of fumigation that is used for disinfestation of grains is not suitable for sealed pouches because of non-penetration of the fumigant through the packaging material. Extensive work done at Bhabha Atomic Research Centre has shown the effectiveness of low-dose gamma radiation (up to 1.00 kGy) to achieve insect disinfestation of wheat, Basmati rice, rawa, etc.

Gamma radiation (0.2 ± 1.00 kGy) destroys all the metamorphic stages of insects and sterilizes the adults of 32 known granary insects. As irradiated food is wholesome and nutritionally adequate, the FAO/WHO/IAEA Joint Expert Committee on Food Irradiation has unconditionally cleared foods irradiated up to 10 kGy as safe for human consumption. Insect disinfestation of wheat and wheat products by gamma irradiation (1.00 kGy) has been approved in eight countries. In India, draft rules amending the Prevention of Food Adulteration Act (PFA) 1954 to permit irradiation of 14 food products including rawa, atta and maida have been modified by the Government, for the commercial application of the technology. In one of the recent studies, Extension of shelf-life of whole-wheat flour by gamma radiation was done.

In one of the recent work, the effect of low-dose gamma irradiation (0.25 ± 1.00 kGy) on pre-packed whole-wheat flour (atta) was assessed in terms of physico-chemical properties, nutritional quality, chapatti-making quality and sensory attributes. Semi-pilot scale storage studies on irradiated pre-packed whole-wheat flour revealed that there was no adverse effect of irradiation and storage up to 6 months of whole-wheat flour treated at doses up to 1.00 kGy on total proteins, fat, carbohydrates, vitamin B1 and B2 content, colour index, sedimentation value, dough properties, total bacterial and mould count. Storage of wheat flour resulted in slight increase in moisture, free fatty acids, damaged starch, reducing sugars and slight decrease in gelatinization viscosity. However, irradiation as such had no effect on any of these parameters. Irradiation at 0.25 kGy was sufficient to extend the shelf-life of atta up to 6 months without any significant change in the nutritional, functional attributes. Chapattis made from irradiated atta (0.25 kGy) were preferred even after 6 months storage, compared with the control [47,48].

Effects of Different Milling Processes on Whole Wheat Flour Stability

Selecting the milling process that will be used is the key consideration in producing whole grain flour. The four predominant techniques for grinding whole grain flours are stone mill (SM), roller mill (RM), ultra-fine mill (UM) and hammer mill (HM). The hammer mill causes the product to be heated up and to lose moisture. Stone mills generate considerable heat due to friction, resulting in damage to starch, protein, and unsaturated fatty acids. The process of roller milling involves separation of the endosperm from the bran and germ followed by gradual size reduction of endosperm. Producing flour that fulfills the requirement for being whole grain is achieved by blending bran and germ back with the endosperm flour in the naturally-occurring proportions. In comparison with stone mills, roller milling is more economical and flexible, less heat production and thus less destruction to chemical components. A third advantage of making whole grain flours from roller mills is that wheat bran and germ can be separated from the endosperm fraction and subjected to further processing or post-milling such as heating or ultrafine grinding to affect the storage or functional properties of the flour . On the other hand, there are considerate studies about the post-milling processes for whole wheat flour, which including the twin-screw extrusion, the heat treatment of the bran and germ and the ultra-fine grinding processes.

Pre Conditioning with Additives

When whole wheat flour is stored, lipase hydrolyses lipids into non-esterified fatty acids (NEFA). NEFA are oxidized non-enzymatically during storage or enzymatically by lipoxygenase when the flour is mixed with water. These processes result in a decrease in the nutritional value and functional and sensory characteristics of WWF. More stable WWF would be desirable because it would not require such careful control of storage time and conditions and may result in higher quality bakery products. Researchers have focused on inhibiting lipase to stabilize WWF. Heat treatments are employed most commonly; however, this would require elevated costs in a large-scale operation, and the exposure to high temperature can initiate autoxidation and lead to non-enzymatic spoilage. Another strategy to stabilize WWF could involve the addition of lipase inhibitors. Metal salts are known to affect the activity of lipases from other cereals and oilseeds. In semi-purified extracts from wheat bran or rice bran, lipolytic activity has been reduced or activated by $CaCl_2$, FeNa–ethylenediaminetetraacetic acid (FeNa–EDTA), and NaCl.

Wheat is commonly conditioned by adding water prior to milling (approved method 26-10.02; AACC International, 2012). Others have replaced water with solutions to impart certain desirable characteristics. For instance, in one of the past work, tempering water was replaced with ozonated water or acetic acid solution (1 %) to reduce microbial load in flour. It was hypothesized that adding the salts in this fashion would allow the metal ions to diffuse into the kernel (along with the water) and interact and ultimately inhibit the lipase enzyme more readily and more practically than spraying the solutions on the bran after milling. Therefore, the objective of this research was to determine if salts added during wheat conditioning at levels that are typically found in baked goods formulations could substantively inhibit lipase and thus improve WWF functionality during storage. It was found that while no salt treatments completely inhibited lipase, salt solutions applied during wheat conditioning significantly influenced activity of the enzyme. This dictated the salt's effectiveness in stabilizing lipids and maintaining functionality during storage. Thus the application of the salt during conditioning could be probed for the inactivation of the enzymes.

Heat/Steaming Treatment

It has been found that the heat treatment can improve the whole wheat flour stability. In one of studies, heat treatment was applied to the whole wheat flour. Enhanced Lipid Stability in Whole Wheat Flour by Lipase Inactivation and Antioxidant Retention The purpose of this study was to determine the effectiveness of dry heat, steam, and microwave treatments in decreasing lipase activity, while retaining antioxidant activity, to stabilize whole wheat flour against lipid degradation during storage. Bran was heat-treated in 230-g batches using four levels (exposure times) for each of the three treatment methods. Lipase activity and antioxidant activity were quantified for all treatment combinations. None of the treatments significantly decreased antioxidant activity; the levels determined to be optimal, inasmuch as further heating did not significantly decrease lipase activity, were 25 minutes of dry heat, 60 sec of microwave (1000W), and 60 sec of steam. These treatments effectively decreased lipase activity by 74, 93, and 96 %, respectively. Optimum treatments were evaluated for acceptance using a consumer sensory panel during a 12-month storage period. No significant differences in acceptance were found between the control and any of the samples either at baseline or after storage. This suggests that whole wheat flour can be stabilized against lipolysis by utilizing the treatments described in this study without decreasing antioxidant activity, and that manufacturers may utilize these treatments without risking decreased consumer acceptance.

In one of the paper, effect of wheat grain steaming and washing on Lipase Activity in Whole Grain flour was evaluated. The present results demonstrated that lipase activity in WGF can be reduced effectively by steam treating wheat grains prior to milling. Steaming grains for 180 s sufficiently inactivated lipase, peroxidase, endoxylanase, and part of the α-amylase without altering the WGF gelatinization profile. Moreover, treatment of separate WGF fractions demonstrated that the (free) lipase activity is mainly, if not only, present in the bran fraction and offers another possible solution for obtaining WGF with longer shelf life. Finally, because washing grains did not reduce the lipase activity in WGF, we concluded that lipase is mainly located within the bran rather than associated with the kernel outside.

Flameless Catalytic Infrared Treatment

Flameless catalytic infrared radiation (FCIR) is a new technology developed by catalytic drying technologies. In flameless infrared emitters, propane or natural gas chemically react at the surface of a platinum catalyst, below gas ignition temperatures, delivering peak radiant energy in the range of 2.8–7 mm. The water molecule shows peak radiation absorption bands at 3, 4.5, and 6 mm [38]. The characteristic of water absorbing infrared radiation directly in the range of 3 - 7 mm has been applied in rapid drying of cereal commodities19 and in deactivating peroxidase in carrots [49]. To date, FCIR has not been employed in stabilizing WG or other cereal brans for extending their shelf life. In one of the study, Effect of flameless catalytic infrared treatment on rancidity and bioactive compounds in wheat germ oil was done/(2015 paper) [50]. Flameless catalytic infrared (FCIR) technology was used to inhibit lipase and lipoxygenase activities of wheat germ (WG) in this study to extend its shelf life. Moreover, the influence of FCIR heating on some quality characteristics of wheat germ oil was assessed. Results reflect that FCIR treatment could effectively decrease the water activity (Aw) and exert a damping effect on lipase and lipoxygenase activities of WG within a short time. The WG sample heated 35 cm below the emitter for 6 minutes with FCIR obtained an excellent stabilization effect. Under this condition, the residual water content and Aw were 3.18% and 0.186, respectively, and the corresponding relative lipase and lipoxygenase activities were 7.94 % and 14.33 %, respectively. The free fatty acid content and peroxide value of this WG sample at 40C remained below 4.65% and 3.15 meq. O_2 per kg WGO respectively for 60 days. The optimal Aw for WG storage is about 0.186. No significant change in main fatty acid but a significant decrease in tocopherol content and oxidative stability was observed when compared to raw WGO. In addition, no significant darkening was observed in WGO extracted from all treated WG samples.

Recent Advances in Wheat Bran and Strategies of Stabilization

The use of wheat bran in the food and feed industry has increased distinctly and visibly over the last decade [51-53]. Since the last decade there have been large number of new product development calming the incorporation of wheat bran in the food sector. Wheat bran is used as ingredient in different food products and its percentage may vary widely.

Annually, data from different sources suggest that wheat bran result as a byproduct of the milling industry and are mainly used in the application of animal feed. Since this approach has a lower valorization potential than applications in food, either directly or after tailored processing, the milling industry is increasingly interested in finding new strategies of wheat bran utilization.

Bran is rich in natural antioxidants, phytoestrogens and lignans, which may exert many beneficial effects to various body functions. However, native or even mechanically pretreated bran poses several disadvantages, which complicate its application in food systems. Among these, negative sensory attributes like the bitter taste are an important issue contributing to low consumer acceptance Moreover, its pronounced water-holding capacity negatively affects rheological properties of dough, thus leading to bread of low volumes and low elasticity. For these reasons, technologies (Figure 1) need to be developed to overcome the observed problem.

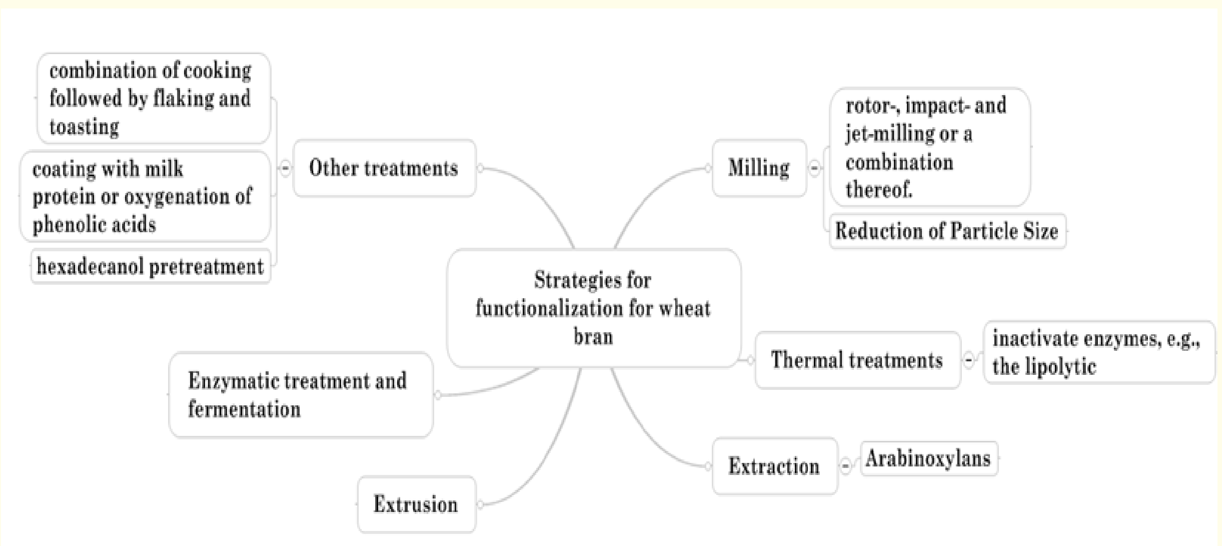

Figure 1: Different Strategies for Functionalization of Wheat Bran.

Various treatments such as milling, heating, extraction, extrusion and fermentation seem to offer some potential to improve the applicability of bran. In principle, the milling step is very effective as the reduction of the particle size disrupts the stable fiber matrix resulting in a significantly improved mouth-feel. However, a bigger surface also enables the development of hydrolytic and oxidation processes thereby leading to reduced shelf-life.

Thermal treatments including extrusion procedures may aid in the improvement of shelf-life due to the inactivation of endogenous enzymes and contaminating microorganisms. There is, however, some side-effect as the color darkens, and the hardness of the extrudate necessitates an additional milling step. Solutions are to be sought to reduce the level of undesired components in order to allow enhanced application of bran in baked goods.

For example, the use of enzymes or fermentation cultures to modify these parameters is optional. Fermentation may induce an anti-staling effect, retarded starch digestibility and further improves the bio-accessibility of minerals and bioactive compounds. On the other hand, increased water activity necessary for solid state fermentation may lead to fungal spoilage. Thus, a subsequent drying step is necessary to ensure sufficient shelf-life [54-67].

Conclusion

It is known that there are many challenges which the food industries as well as millers faces related to whole wheat flour stability. Lipid degradation by the action of enzymatic as was well non-enzymatic pathways seems to be a one of the leading causes of whole wheat flour stability. Among the available techniques of milling, producing whole wheat flour using roller mill seems to be advantageous as opposed to stone mill due to lower temperature as well as better control over entire operations. Jet milling also seems to be promising. However additional research has to be carried out to explore its potential. Among the available method to enhance whole wheat flour stability, retention of antioxidant during milling as well as heat treatment to inactivate enzymes seems to be promising. Flameless catalytic infrared radiation (FCIR) seems to be a promising technology however its need future investigation. Different Strategies for Functionalization of Wheat Bran needs to be explored in the future to develop whole wheat flour with addition of stabilized bran.

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Volume 2 Issue 3 March 2018