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DON reduction of wheat grain without compromising the lab-scale milling properties of flour

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ABSTRACT

Wheat bran was investigated to be the most commonly contaminated raw material by mycotoxins. However, there are no economical and practical pretreatment methods for industrial on-line application until now. The effect of light debranning on deoxynivalenol (DON) removal, polyphenol oxidase (PPO) activity and flour quality from lab-scale milling were performed. For on-line production, the DON concentration in wheat decreased 15.89% at debranning ratio of 1.2%. For lab experiment, the maximum DON removal for wheat and flour was 23.35% and 21.95%, respectively. However, the PPO activity, browning of dough sheet and flour qualities in lab scale exhibited no significant variation. Light debranning (1.2%) prior to milling could be efficiently applied to on-line wheat production.

1. Introduction

Wheat bran, as the peripheral tissue of kernel, is composed of six layers with various composition and efficacies [1]. The outer bran (pericarp and testa) is essentially consisted of cellulose and lignin with the primary role of protection, it can't be digested and absorbed completely [2]. Thus, wheat bran should be removed during milling. Researches have been reported that wheat bran containing significant amounts of dietary fiber and phenolic polymer could be utilized to enhance food nutrients and improve digestibility [3], such as preventing gradual increment in visceral fat [4], mediating insulin resistance and inflammation [5], and protecting against cardiovascular diseases and cancers [6].

However, wheat was recently determined to be the most commonly contaminated crop by mycotoxins. Considering DON could inhibit protein synthesis and disturb cell function [7], several strategies were applied to counteract or alleviate possible deleterious effects of DON, such as hydrothermal treatment [8], ultraviolet irradiation [9], and ozonation [10]. However, several problems, such as decomposition of cell structure, degradation of nutrient components during pretreatment, excessive equipment investment increasing production cost, restricted their commercialization in on-line milling process.

The dehulling process was proved to be efficient for DON reduction [11], but high bran elimination was required [3]. In addition, wheat flour from dehulling pretreatment showed undesirable properties as compared with untreated flour in terms of enzyme activity, antioxidant activity (AOA), pentosan and phenolic content, and flour yield [12]. Therefore, a novel treatment method should be explored to particularly solve these challenges. To the best of our knowledge, no studies have been conducted on on-line production. The objective of the presented work was to study the effect of different light debranning ratios (0–1.2%) using innovative machine on DON removal and flour qualities from on-line and lab scales.

2. Materials and methods

2.1. Materials

The on-line untreated and treated samples were obtained from manufacturing workshop of Jinshahe Flour Group. The two commercial wheat samples of naturally contaminated E and H were supplied by Hanzhong Sanyi Science and Technology Co. Ltd. The debranning treatment was conducted using an industrial scale single cylinder (SC) dehusking machine FBGM (Hanzhong Sanyi Science and Technology Co. Ltd., Hanzhong, China). The controlled removal degrees of bran were accomplished by manually adjusting the angle and gap between the carborundum scraper and inner wall. The four debranning levels of 0 (D0), 0.40% (DI), 0.80% (DII), and 1.20% (DIII) were performed. The samples of E and H were subsequently milled using a Bühler MUL-202. The Bühler mill consisted of three breaks (TB) and three reductions (TR). As illustrated in Fig. 1, the TR streams were mixed as F1 flour, the TB streams were

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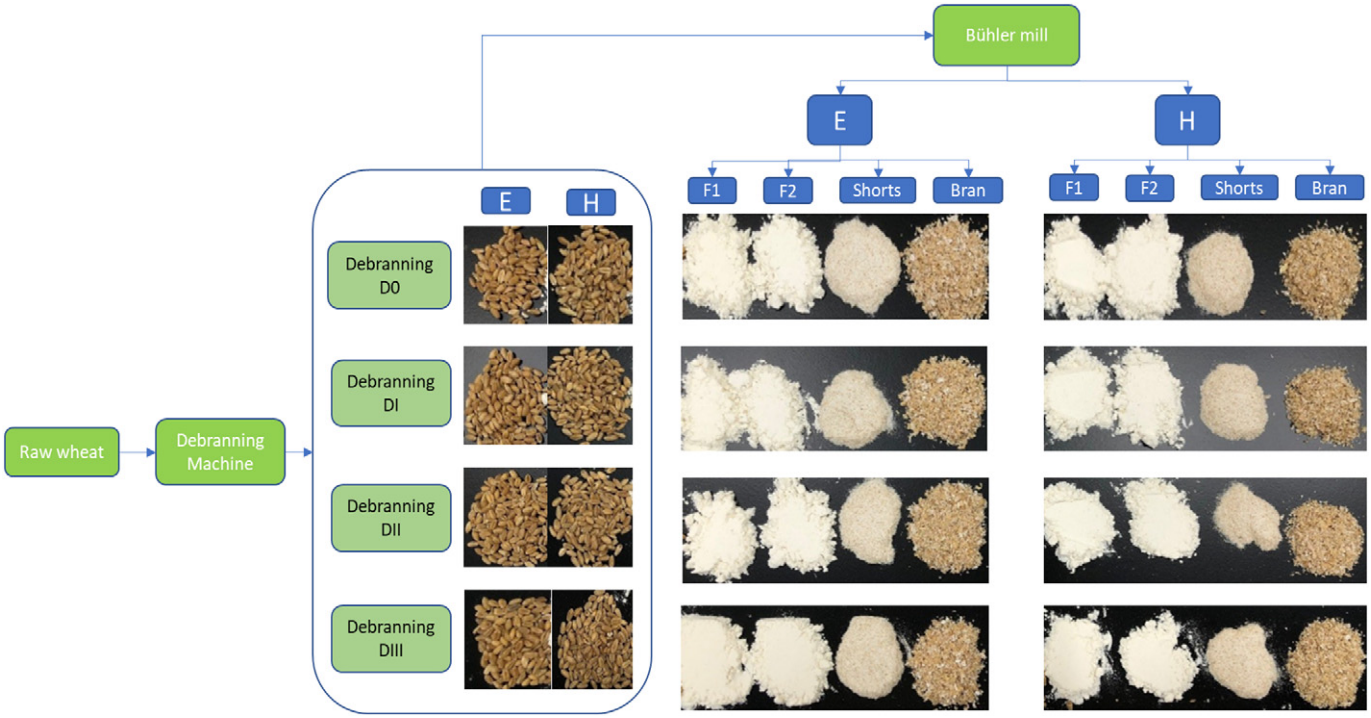

Fig. 1. Schematic of experimental procedure.

collected as F2 flour, byproducts were labeled as shorts (fine bran) and coarse bran, respectively (Fig. 1).

2.2. Moisture, ash content and breakage of wheat kernel

The moisture and ash content of wheat were determined according to AACC method 44-15A and AACCI Method 08-01.01, respectively. Broken kernels from samples were separated manually [13].

2.3. DON, APC and EE content of wheat kernel and flour

Detection of DON was performed using a self-assembly enzyme-linked immunosorbent assay (ELISA) kit [14]. As a fundamental immunoassay, ELISA has been widely developed for detecting DON, given its efficiency and specificity [15,16]. Aerobic Plate Count (APC) was measured following AACCI method 42-11.01. Enumeration of Enterobacteriaceae (EE) was determined according to the Chinese National Standard GB4789.41-2016, the data was reported using most probable number (MPN).

2.4. Enzymatic activity and flour quality

PPO activity of wheat flour was tested by AACCI method 22-85.01. The bran speck and black spot content in flour were measured by Flourscan F 2000 (Branscan Co. Ltd., Tokyo, Japan), parameters were shown as the ratio of area of bran speck and black spot to total area of flour, respectively. Dough sheet browning was tested by the Minolta CR-400 Chroma Meter (Minolta Co. Ltd., Japan). The dough sheet was prepared according to the noodle processing technology. The first step was to form the dough, flour (100 g, 14% mb) and distilled water (32 mL), which were mixed for 5 min using a CS-B5A mixer (Tongxin Machine Group Co. Ltd., Guangzhou, China), and followed by putting the dough into sealed container at 25 °C for 20 min. Then, the dough was sheeted with an electric noodle-making machine (DMT-10A, Fuxing Machinery Co. Ltd., Shandong, China). In this procedure, the roll gaps of the noodle making machine were adjusted [17], finally the dough sheet was packed in sealed plastic bags and placed in refrigerator at 4 °C for 24 h. The final color shown (W24, L*, a*, b*) was an average of four replicates.

2.5. Data analysis

Data were analyzed using GraphPad Prism 8 (GraphPad Software, San Diego, CA). Differences among the values were analyzed using one-way analysis of variance (ANOVA). Mean comparisons were conducted using Tukey adjustment at $P < 0.05$. All experimental results were at least performed in three replicates.

3. Results and discussions

3.1. Effect of debranning on wheat kernel from on-line processing

To verify whether the light debranning process could be used on-line milling process. The untreated and treated samples from milling company were testified. As illustrated in Table 1, differences in ash, DON, APC, and EE contents between the untreated and treated were significant ($P < 0.05$), while the grain temperature and broken wheat rate were unchanged. The ash content of wheat after debranning pretreatment decreased 0.04%–

Table 1
Ash, broken wheat, DON, APC, EE of the untreated and treated wheat grains.

Sample	Temperature (°C)		Ash content (%)		Broken Wheat (%)		DON (ng/g)		APC (cfu/g)		EE (MPN/g)	
	SC	DC	SC	DC	SC	DC	SC	DC	SC	DC	SC	DC
J-untreated	35.1 ±0.1^{a}	34.9 ± 0.1^{a}	1.98 ± 0.01^{b}	1.97 ± 0.00^{b}	2.5 ± 0.0^{a}	2.4 ± 0.1^{a}	916 ± 37^{b}	969 ± 28^{b}	13500	14000	36	43
J-treated	35.1 ±0.0^{a}	34.9 ± 0.1^{a}	1.91 ± 0.02^{a}	1.93 ± 0.01^{a}	2.5 ± 0.0^{a}	2.4 ± 0.0^{a}	827 ± 16^{a}	815 ± 20^{a}	3700	805	7.4	23

Note: Means in the same column with different letters are significantly different ($P < 0.05$). SC: single cylinder; DC: double cylinder; J: sample from milling company.

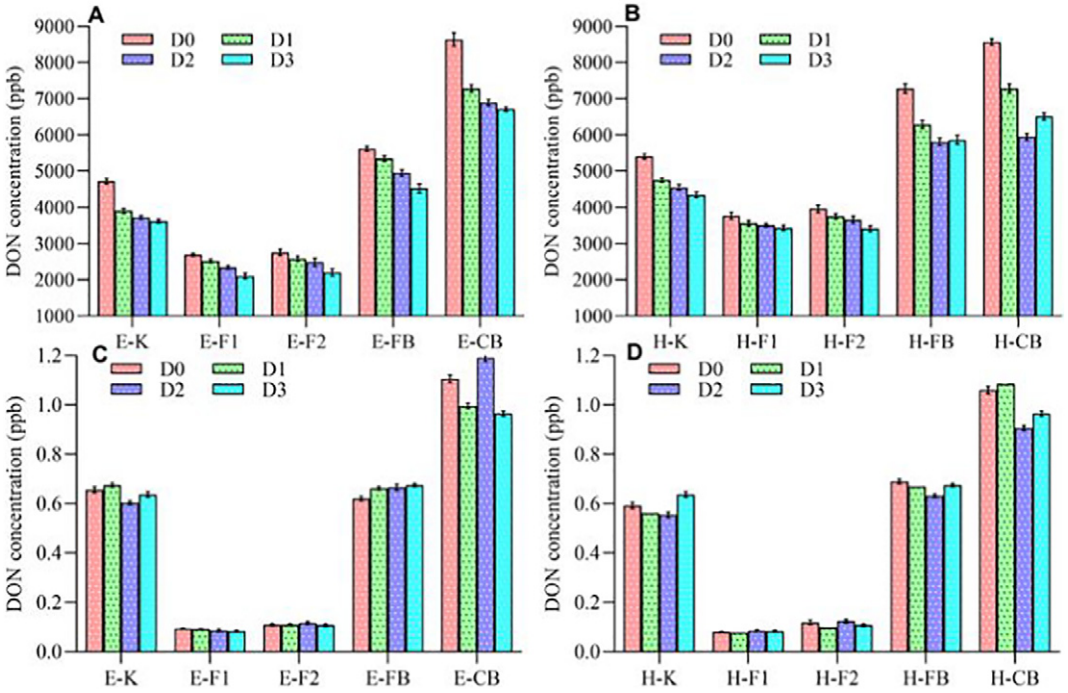

Fig. 2. Distribution of DON content (A, B), PPO activity (C, D) of wheat kernels and flours with different debranning levels. K: kernel of sample; F1: flour from three reductions; F2: flour from three breaks; FB and CB were the abbreviation of fine bran and coarse bran, respectively.

0.07%, which was attributed to partial removal of wheat bran. Similar effect of extended debranning on ash content has been reported earlier [18]. Remarkably, as debranning ratio increased to 1.20%, the contents of DON, APC, and EE decreased by 89–154 ng/g (9.72%–15.89%), 9800–13,195 cfu/g (72.59%–94.25%), 20–28.6 MPN/g, respectively, which is consistent with other findings that one of the benefits of debranning on flour was to remove the microbial population and mold [19]. These changes demonstrate that light debranning could be used as bacteria scavenger on industrial cleaning of wheat.

3.2. Effect of debranning on DON content and PPO activity

Two naturally contaminated wheat samples (E and H) were employed to evaluate the effect of light debranning on DON and PPO activity of wheat kernels and flours. Fig. 2A and B showed similar DON reduction trends as debranning level increased except H-FB and H-CB. As debranning ratio increased from 0 to 1.2%, DON concentrations of E-K, E-F1, E-F2, E-FB, and E-CB decreased significantly from 4723 to 3620 ng/g, 2697 to 2105 ng/g, 2765 to 2202 ng/g, 5623 to 4527 ng/g, and 8634 to 6707 ng/g corresponding to the removal rate of 23.35%, 21.95%, 20.36%, 19.49%, and 22.32% (Fig. 2A), respectively. Similarly, DON concentrations of H-K, H-F1, H-F2, H-FB, H-CB reduced with increment of debranning degrees from 5408 to 4352 ng/g, 3769 to 3437 ng/g, 3965 to 3410 ng/g, 7284 to 5866 ng/g, and 8571 to 6507 ng/g with the elimination percentage of 19.53%, 8.81%, 14.00%, 19.47%, and 24.08%, respectively (Fig. 2B). These changes strongly indicate that light debranning prior to milling is an effective method to alleviate the DON content in wheat. DON content of CB was higher than FB (Fig. 2A and B), which is agreement with other study showing a higher concentration of mycotoxins in the outer layers of wheat grain [20]. The milling process, even at the optimum condition, could not completely separate the starchy endosperm from bran, fine bran particles could still pass through the sifter into flour streams [21]. Therefore, the cleaning of wheat kernels is critical to prevent flour from being contaminated by bacteria. Besides, the DON content of F1 for both cultivars was slightly higher than that of F2, which also indicates that the purity of flour in the reduction streams was higher than break streams.

PPO, known as catechol oxidase or metalloprotease, can oxidize phenols or polyphenols to form corresponding quinones. The enzymatic browning reaction caused by PPO has a negative or undesirable effect on the processing of fruits, vegetables [22], and even flour products [23,24]. Fig. 2C and D showed that the debranning process had no considerable influence on the PPO activity of wheat kernels and flour. However, PPO activity in bran was significantly higher than flour, which demonstrates that PPO is highly present in the bran fraction of wheat [25,26].

3.3. Effect of debranning on APC content of wheat kernel and flour

The determination of aerobic plate count was conducted on E and H samples. The measurement of total bacterial count was usually used to evaluate the degree of bacterial contamination on food due to it reflects whether the food meets the sanitary requirements during processing. Thus, making an appropriate evaluation of the tested samples is necessary. Table 2 illustrated that debranning treatment had no trend influence on the total number of bacterial colonies in samples.

3.4. Effect of debranning on browning of dough sheet and flour property

The colors (W24, L*, a*, b*) of dough sheet could be utilized to reflect the activity of polyphenol oxidase. Table 3 showed that the light

Table 2
APC content of the wheat kernel and F1, F2 flours with different debranning levels.

Sample	Debranning (%)	APC($\times 10^4$ cfu/g)		
		Wheat kernel	F1	F2
E	0.00	51.0	1.2	5.4
	0.40	9.3	2.1	2.2
	0.80	14.0	1.9	2.7
	1.20	12.0	4.8	3.6
H	0.00	4.6	0.6	3.5
	0.40	7.9	2.3	4.4
	0.80	5.3	1.5	5.4
	1.20	61.0	1.8	1.4

Table 3
Browning of dough sheet with different debranning levels.

Sample	Debranning (%)	F1				F2			
		W24	L*	a*	b*	W24	L*	a*	b*
E	0.00	62.53 ± 0.24^{c}	14.61 ± 0.23^{b}	-0.60 ± 0.03^{b}	0.49 ± 0.04^{b}	58.09 ± 0.37^{c}	15.63 ± 0.34^{a}	-0.73 ± 0.05^{b}	1.59 ± 0.17^{b}
	0.40	61.97 ± 0.38^{bc}	14.98 ± 0.34^{b}	-0.60 ± 0.05^{b}	0.47 ± 0.02^{b}	57.70 ± 0.42^{c}	15.78 ± 0.51^{a}	-0.79 ± 0.04^{b}	1.43 ± 0.09^{b}
	0.80	61.82 ± 0.42^{b}	14.76 ± 0.19^{b}	-0.53 ± 0.03^{b}	0.41 ± 0.05^{ab}	57.64 ± 0.36^{c}	15.84 ± 0.37^{a}	-0.76 ± 0.06^{b}	1.41 ± 0.14^{b}
	1.20	63.31 ± 0.30^{d}	14.43 ± 0.36^{ab}	-0.55 ± 0.04^{b}	0.50 ± 0.03^{b}	57.92 ± 0.39^{c}	15.69 ± 0.40^{a}	-0.72 ± 0.05^{a}	1.50 ± 0.11^{b}
H	0.00	60.00 ± 0.36^{a}	14.96 ± 0.29^{b}	-0.44 ± 0.02^{a}	0.43 ± 0.03^{ab}	54.44 ± 0.28^{a}	17.01 ± 0.43^{b}	-0.66 ± 0.03^{a}	0.95 ± 0.15^{a}
	0.40	61.21 ± 0.54^{b}	14.33 ± 0.33^{ab}	-0.43 ± 0.03^{a}	0.47 ± 0.04^{ab}	54.42 ± 0.31^{a}	16.74 ± 0.36^{b}	-0.68 ± 0.04^{ab}	0.87 ± 0.06^{a}
	0.80	60.73 ± 0.41^{ab}	14.17 ± 0.20^{a}	-0.40 ± 0.03^{a}	0.41 ± 0.03^{a}	55.09 ± 0.42^{ab}	16.30 ± 0.44^{ab}	-0.65 ± 0.02^{a}	0.85 ± 0.10^{a}
	1.20	61.89 ± 0.39^{b}	14.55 ± 0.31^{ab}	-0.45 ± 0.02^{a}	0.45 ± 0.02^{ab}	55.66 ± 0.35^{b}	17.24 ± 0.52^{b}	-0.64 ± 0.04^{a}	0.84 ± 0.05^{a}

Note: Means in the same column with different letters were significantly different ($P < 0.05$).

Table 4
Milling property of flours with different debranning levels

Sample	Debranning (%)	Ash Content (%)		Bran Speck (%)		Black Spot (%)		Whiteness	
		F1	F2	F1	F2	F1	F2	F1	F2
E	0.00	0.60 ± 0.03^{ab}	0.70 ± 0.02^{a}	9.97 ± 0.15^{a}	13.69 ± 0.10^{bc}	2.02 ± 0.02^{a}	2.19 ± 0.03^{bc}	75.15 ± 0.26^{b}	74.39 ± 0.30^{b}
	0.40	0.64 ± 0.04^{bc}	0.71 ± 0.03^{a}	9.99 ± 0.11^{a}	13.42 ± 0.09^{a}	2.03 ± 0.02^{a}	2.25 ± 0.04^{c}	75.33 ± 0.32^{b}	74.58 ± 0.25^{b}
	0.80	0.65 ± 0.02^{bc}	0.73 ± 0.02^{a}	10.15 ± 0.13^{a}	13.86 ± 0.14^{c}	2.07 ± 0.03^{a}	2.08 ± 0.02^{a}	74.91 ± 0.34^{b}	74.54 ± 0.24^{b}
	1.20	0.57 ± 0.01^{a}	0.68 ± 0.04^{a}	10.07 ± 0.09^{a}	13.58 ± 0.13^{ab}	2.06 ± 0.02^{a}	2.14 ± 0.05^{ab}	75.43 ± 0.37^{b}	74.59 ± 0.27^{b}
H	0.00	0.63 ± 0.02^{bc}	0.86 ± 0.05^{b}	15.18 ± 0.12^{c}	23.32 ± 0.15^{e}	2.30 ± 0.05^{b}	5.19 ± 0.11^{d}	72.83 ± 0.28^{a}	70.56 ± 0.19^{a}
	0.40	0.63 ± 0.03^{bc}	0.85 ± 0.03^{b}	14.85 ± 0.10^{b}	22.95 ± 0.21^{d}	2.27 ± 0.03^{b}	5.49 ± 0.09^{e}	72.84 ± 0.16^{a}	70.88 ± 0.16^{a}
	0.80	0.69 ± 0.05^{c}	0.86 ± 0.04^{b}	14.95 ± 0.16^{bc}	22.93 ± 0.19^{d}	2.24 ± 0.03^{b}	5.34 ± 0.13^{de}	72.82 ± 0.21^{a}	70.54 ± 0.23^{a}
	1.20	0.63 ± 0.02^{bc}	0.81 ± 0.04^{b}	14.89 ± 0.17^{bc}	23.03 ± 0.22^{de}	2.32 ± 0.06^{b}	5.09 ± 0.16^{d}	72.94 ± 0.22^{a}	70.93 ± 0.31^{a}

Note: Means in the same column with different letters were significantly different ($P < 0.05$).

debranning process had no obviously influence on the surface browning of dough sheet, which is in agreement with light debranning only removed nonfunctional components of outer bran without affecting the chemical and structural features of aleurone layer [27].

The milling parameters (bran speck, black spot, whiteness) of E and H flours gave no pronounced differences with the incremental debranning degrees (Table 4), which indicates that the outer bran layers were removed partially in the process of debranning, starchy endosperm remained constant. Based on the results exhibited above, light debranning, which eliminated partial contaminated bran layers from wheat grains [21], could be a promising candidate for industrial application.

4. Conclusions

Light debranning prior to milling has potential as an efficient method to reduce DON content of contaminated wheat grains in the course of industrial production. For on-line production, the DON concentration decreased 15.89% at debranning ratio of 1.2%. For lab scale, the highest DON removal for E-K (23.35%) and H-CB (24.08%) were achieved, respectively. However, the PPO activity, surface browning of dough sheet and lab-scale milling quality of wheat and flour showed no significant changes.

Conflicts of interest

All the authors confidently declare that there is no conflict of interest.

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