https://doi.org/10.17221/64/2024-CJFS

The relationship between large deformation rheology of wheat flour dough with protein quantity and aggregate stretching degree of milling streams flour based on regression analysis

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Citation: Dong Z., Su T., Dai M., Don C., Guo B., Du S., Zhang B. (2024): The relationship between large deformation rheology of wheat flour dough with protein quantity and aggregate stretching degree of milling streams flour based on regression analysis. Czech J. Food Sci., 42: 353–363.

Abstract: The aim of this study was to compare the role of protein quantity and aggregate stretching degree in predicting dough stability and extensibility using the regression analysis, and to explore a more effective way of conducting the prediction. Flours from 28 milling streams of the wheat cultivar Shiluan 02-1 were collected as experimental material. Using the value of (ash content/L^*) (L^* – lightness), we sorted the milling streams flour from the inner layer to the outer layer of wheat kernel, which was divided into early reduction, later reduction, and break flours. Three regression models, quantity-based, stretching-degree-based and (quantity × stretching-degree)-based model for predicting dough stability and extensibility were evaluated in each category of milling streams through their coefficient of determination (R^2). Certain patterns were observed in physicochemical properties of flour from different categories of milling streams. Despite those considerable changes, the quantity-based model broadly produced greater R^2 values than the stretching-degree-based model, and the (quantity × stretching-degree)-based model could in general provide higher R^2 values than the other two models on predicting dough stability and extensibility. The results suggest that measuring the protein quantity and aggregate stretching degree at the same time is of practical improvement in dough rheology evaluation, compared to focusing on either factor alone.

Keywords: regression analysis; dough; rheology; protein; quantity; stretching degree

Supported by Earmarked Fund for China Agriculture Research System (CARS-03), Agricultural Science and Technology Innovation Program, Institute of Food Science and Technology, Chinese Academy of Agricultural Sciences (CAAS-ASTIP-2022-IFST), and Financial Fund of Institute of Food Science, Technology, Nutrition and Health (Cangzhou), Chinese Academy of Agricultural Sciences (CAAS-IFSTNH-CZ-2022-01).

https://doi.org/10.17221/64/2024-CJFS

Wheat production totalled over 800 million tons in 2022 worldwide, and 70% of wheat has been used to produce food such as bread and noodles, serving as the staple food for about one-third of the world population (FAOSTAT 2024). Mixing wheat flour with water can form elastic and malleable dough, which is a fundamental step in making most wheat-based products, and the performance of wheat flour dough largely determines wheat end-use quality (Rosell 2011; Delcour et al. 2012; Singh et al. 2018). Dough mixing is usually conducted at 30 °C, which is a much lower temperature than pasting temperature of starch, hence it is generally believed that protein properties are more relevant to dough rheological properties. In the wheat processing industry, dough consistency and extensional properties are mainly measured by the devices like farinograph and extensograph, which examine the variation of consistency throughout dough development and breakdown, and that corresponds to large deformation rheology (Singh et al. 2011; Singh et al. 2018). In particular, dough stability has been considered as an important parameter in several national standards in China (GB/T 14614-2019; GB/T 17320-2013; GB/T 17893-1999), while dough extensibility plays a crucial role in making traditional Chinese food, such as noodles and dumpling wrappers.

An analogy with polymer chemistry has often been sought to explain the increased elasticity of the dough as the polymers stretch, get longer, crosslink and so result in a strong viscoelastic dough at peak development (Singh and MacRitchie 2001; Ortolan and Steel 2017; Yang et al. 2022). The quantity of protein in wheat flour and quality of gluten development have been suggested to influence the amount and intensity of entanglements in the gluten network, thus determining the variation of dough consistency (Barak et al. 2013; Delcour et al. 2012). Protein aggregates in dough mainly refer to gluten, which is the storage protein in wheat grains, accounting for about 80% of the total protein. Wheat gluten consists of monomeric gliadins and polymeric glutenins of protein types: α-, γ-, and ω-gliadins and the high-molecular-weight and low-molecular-weight glutenin subunits (HMW-GS and LMW-GS, respectively) (Shewry 2023). A large-size fraction of wheat glutenin, namely glutenin macropolymer (GMP), can be obtained by suspending defatted flour in 1.5% sodium dodecyl sulphate (SDS) and collecting the gel layer on top of the sediment after centrifugation (Wieser et al. 2023). The flour protein content and content of protein fractions are often used to predict dough rheological properties, whose correlations have widely been reported to varying degrees (Barak et al. 2013; Hasniza et al. 2014; Yue et al. 2020). In fact, most of the wheat grains of different varieties used in related studies not only had different protein quantity, but also they showed varying gluten strength and the ability to form the network structure due to different genetic backgrounds. Therefore among different studies, the correlations between protein quantity and dough rheological properties were quite different and difficult to compare. But it is worth noting that Singh and Singh (2013) studied the farinograph characteristics of 12 wheat varieties (protein content 8.3–10.1%) with different gluten strength, and they found that the correlation coefficients of dough development time and stability with GMP content in flour were 0.921 and 0.798, respectively, which were higher than those with flour protein content, glutenin content or gliadin to glutenin ratio, indicating that GMP might be the key to understanding the unique rheology of wheat flour dough (Don 2022).

Meanwhile, to quantitatively determine the degree of gluten development in dough formation the method is somewhat scanty: Bernklau et al. (2016) developed a new approach to quantifying the wheat dough microstructure that determines not only the protein area, but also the number of junctions and structure regularities for describing the strength of the network. However, such a method is still unfavourable to the small- and medium-sized wheat processing industries to evaluate the quality of flour, for its requirements of high-precision measurements performed by the staff with related background. For wheat processing industries to conduct quality control more conveniently in a holistic manner, aggregate stretching degree is adopted to express the degree of gluten development in the dough system. It is defined as the volume of protein per unit mass, which can be calculated from the water-holding capacity of gluten and GMP storage modulus per unit mass. The higher aggregate stretching degree indicates the larger volume of protein per unit mass, which facilitates crosslinking within and among protein aggregates, thus increasing dough consistency macroscopically. Gluten water-holding capacity (*WHC*) is the ability of gluten proteins to prevent water from being released or expelled from their network structure (Haque et al. 2016). This property is based on the direct interaction of protein molecules with water, which mostly involves hydrogen bonding (Aryee et al. 2018). The size of the gel network can affect the gluten *WHC*, for example the large-pore network usually shows the lower *WHC* value due to the lack of capillary ef-

https://doi.org/10.17221/64/2024-CJFS

fect that is seen more commonly in the small-size network (Aryee et al. 2018). According to Veraverbeke and Delcour (2002), a size distribution of glutenin polymers with more large-size polymers would result in stronger and more elastic doughs. It was also suggested that the average particle size of GMP had a parallel with the plateau values of its storage modulus (Don et al. 2003a, b). In addition, gluten water-holding capacity and GMP storage modulus have been reported to be in positive correlation with dough stability and dough development time (Don et al. 2006; Zheng et al. 2020), respectively.

The aim of this study was to compare the role of protein quantity and aggregate stretching degree in predicting dough stability and extensibility using regression analysis, and to explore a more effective way of conducting the prediction through the coefficient of determination of regression models. Milling streams flour from the wheat cultivar Shiluan 02-1 was used as our test material, which ensures the same genetic and environmental factors of the samples. In addition, we divided the total milling streams into 3 categories which corresponded to different parts of wheat kernel, and compared the physicochemical properties and their performance by regression analysis.

MATERIAL AND METHODS

Materials. The wheat cultivar Shiluan 02-1 was harvested in 2019 and then cleaned and conditioned in an industrial manner. The wheat kernels were milled in an industrial plant (800 t per day; Buhler, China). The plant consists of 7 break (1B, 2B, 3B, 3BF, 4B, 4BF, 5B) passages, 11 reduction (1M1, 1M2, 1M3, 2M1, 2M2, 3M1, 3M2, 4M2, 5M1, 5M2, 6M1) passages, 2 sizing (1S, 2S) passages, 6 resifting (D1, D2, D3, DS, D4, DF) passages, 1 tail passage (1T), and 1 suction passage (XF4). Flour was obtained in each passage mentioned above, using a container to get the sample of 5 kg, respectively, and the yield was calculated with the time recorded. According to Dai et al. (2020), (ash content/L^*) (L^* – lightness) is able to characterise the order of milling streams flour from the inner to the outer layer of wheat kernel. And the value of (ash content/L^*) was used to sort each milling streams flour, with the order of 1 to 28 (Table 1). The flours from 28 milling streams were then divided into 3 categories: early reduction flours (order: 1–10; yield: 0–55%), later reduction flours (order: 11–20; yield: 55–79%) and break flours (order: 21–28; yield: 79–90%). Therefore, the number of samples from total milling streams, early reduction, later reduction and break streams was 28, 10, 10, and 8, respectively.

Protein content. Based on the Dumas combustion method described in ISO/TS 16634-2:2016 and the method described by Hu et al. (2022), the sample was burned at a high temperature in a pure oxygen environment, and its impurities were absorbed by the reducing agent, and the released nitrogen was detected by a thermal conductivity detector (TCD) to obtain the total nitrogen content, which was multiplied by the corresponding conversion factor to obtain the protein content. In this study, the protein content (calculated as nitrogen × 5.7) was determined using the Dumas Nitrogen Analyser (DN2100; Nordtech, China) with aspartic acid as the standard.

Dry gluten content and gluten water-holding capacity. Content of wet and dry gluten was measured according to the AACC 38-12.02 procedure using a gluten instrument (Perten Instruments AB, Sweden). Gluten water-holding capacity was calculated as follows:

$$Q_1 = \frac{w_1 - w_2}{w_2} \times 100\% \quad (1)$$

where: Q_1 (%) – gluten water-holding capacity; w_1 (%) – wet gluten content; w_2 (%) – dry gluten content.

Glutenin macropolymer extraction and flour sodium dodecyl sulphate retention capacity. The method used to extract GMP was modified from Don et al. (2003a). First, a 1.4 g flour sample was dispersed in 28 mL of 1.5% (*w*/*v*) SDS and then centrifuged at 20 000 g for 30 min at 20 °C in a high-speed centrifuge (3-30K; SIGMA, Germany). Subsequently, the supernatant was decanted, 1 mL of which was dried for 48 h at 50 °C. Finally, the SDS-soluble protein content was measured by Dumas Nitrogen Analyser (DN2100; Nordtech, China). The corresponding formulas used to calculate the protein content were as follows (Li et al. 2020):

$$P_2 = \frac{C \times M_1 \times M_3}{1.40 \times M_2} \quad (2)$$

$$P_3 = P_2 - P_1 \quad (3)$$

where: P_1 (g·g^{-1} flour) – SDS-soluble protein content per unit amount of flour; P_2 (g·g^{-1} flour) – protein content per unit amount of flour; M_1 (g) – total weight of the supernatant; M_2 (g) – weight of 1 mL supernatant; M_3 (g) – weight of dried 1 mL supernatant; C (%) – flour protein content; 1.40 (g) – weight of flour; P_3 (g·g^{-1} flour) – GMP content.

https://doi.org/10.17221/64/2024-CJFS

Table 1. Yield and categories of milling streams flour for the wheat cultivar Shiluan 02-1 [in ascending order of (ash content/L^*)]

Milling streams	Yield (%)	Ash content (%)	L^*	Ash content/L^* (%)	Order from the inner to the outer layer of wheat kernel**	Milling streams category
2M1	12.80	0.38	92.95	0.0041	1	early reduction flours
1M2	13.80	0.41	92.76	0.0044	2	
1M3	3.60	0.42	92.54	0.0045	3	
3M1	7.80	0.43	91.97	0.0047	4	
1M1	4.90	0.45	92.72	0.0049	5	
2M2	6.40	0.47	91.97	0.0051	6	
DS	0.10	0.51	90.84	0.0056	7	
1S	0.70	0.58	91.41	0.0063	8	
5M1	1.10	0.58	90.81	0.0064	9	
3M2	4.50	0.62	89.99	0.0069	10	
D3	6.80	0.62	90.43	0.0069	11	later reduction flours
D2	3.90	0.65	89.89	0.0072	12	
4M2	3.50	0.66	90.88	0.0073	13	
3B	3.70	0.67	90.14	0.0074	14	
2S	1.00	0.69	90.29	0.0076	15	
5M2	0.30	0.72	89.84	0.0080	16	
2B	2.20	0.73	89.46	0.0082	17	
D4	1.10	0.74	88.39	0.0084	18	
1T	0.60	0.80	89.14	0.0090	19	
4B	1.00	0.81	89.45	0.0091	20	
D1	2.50	0.80	88.18	0.0091	21	break flours
3BF	0.20	0.83	89.54	0.0093	22	
4BF	0.40	0.90	88.12	0.0102	23	
1B	1.60	0.91	87.68	0.0104	24	
5B	0.60	0.93	87.12	0.0107	25	
DF	0.90	0.95	88.12	0.0108	26	
XF4	2.20	1.24	86.05	0.0144	27	
6M1	1.80	1.36	88.74	0.0153	28	

**According to (ash content/L^*), flour sample of milling streams was sorted from the inner layer to the outer layer of wheat kernel, with the order increasing from 1 to 28; L^* – lightness; 1B, 2B, 3B, 3BF, 4B, 4BF, 5B – break passage; 1M1, 1M2, 1M3, 2M1, 2M2, 3M1, 3M2, 4M2, 5M1, 5M2, 6M1 – reduction passage; 1S, 2S – sizing passage; D1, D2, D3, DS, D4, DF – resifting passage; 1T – tail passage; XF4 – suction passage

Flour SDS retention capacity was calculated as follows:

$$\text{SDS } \textit{retention capacity} = \left(\frac{W_1}{1.40} \times \frac{100-14}{100-T_1} - 1 \right) \times 100\% \quad (4)$$

where: SDS – sodium dodecyl sulphate; W_1 (g) – weight of GMP gel after centrifugation; 1.40 (g) – weight of the sample; 14 – standard wet-based water content; T_1 – water content of the sample.

Dynamic rheology of glutenin macropolymer gels. The rheology of GMP gels was determined as described by Don et al. (2003a) with some modifications. The gel-like layer was present on the top of the starch after decanting the supernatant and was collected as GMP. Then, 1 g sample of GMP was carefully collected from the top of the three remaining precipitates and measured on a rotational rheometer (MCR 502; Anton Paar, Austria) with two parallel plates (d = 25 mm), where the gap between the plates was 1 mm. To reduce the loss of water during dispersion, a cover was used during the analysis. Measurements were conducted at 20 °C in a strain sweep mode at 0.01–100% with a fixed frequency of 1 Hz.

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GMP storage modulus per unit mass was calculated as follows:

$$G_p = \frac{G'}{m} \quad (5)$$

where: G_p (Pa·g^{-1}) – GMP storage modulus per unit mass; G' – storage modulus of GMP; m (g) – protein weight of the tested GMP gels.

Large deformation rheology of wheat flour dough. Dough rheological behaviour was examined using Farinograph 827504 and Extensograph 860033 (Brabender, Germany) according to the AACC 54-21 and AACC 54-10 methods, respectively. Although several results were obtained, we chose dough stability produced in farinograph and dough extensibility in extensograph, which were popular and often necessary among wheat processing industries as the parameters to conduct regression analysis.

Statistical analysis. All measurements were performed in triplicates. Microsoft Excel (version 2010) was used to process data and tables and SPSS (version 22.0) was used to conduct regression analysis.

Dough stability and extensibility were taken as the dependent variables, respectively. Protein content, dry gluten content, and GMP content were taken as the independent variables in a quantity-based model; gluten water-holding capacity and GMP storage modulus per unit mass were taken as the independent variables in a stretching-degree-based model; flour SDS retention capacity, (dry gluten content) × (gluten water-holding capacity) and GMP storage modulus were taken as the independent variables in a (quantity × stretching-degree)-based model. The regression analysis was conducted in terms of total milling streams flour, early reduction flours, later reduction flours and break flours, respectively, using the three types of models mentioned above. The coefficient of determination (R^2) values of different models were compared in the same category of milling streams to evaluate the relative accuracy of each model to predict dough stability or extensibility, and here the significance of the model was considered less important. For example, higher R^2 value indicates a relatively accurate model, even if the model is not significant.

RESULTS

Protein quantity, aggregate stretching degree, dough stability, and dough extensibility of milling streams flour. Protein quantity parameters, including protein content, dry gluten content and GMP content, ranged between 9.34–18.28%, 9.96–19.00%, and 1.89–4.20%, respectively (Table 2), which all exhibited a gradual increase from early reduction streams to later reduction streams, and then to break streams. Aggregate stretching degree parameters, including gluten water-holding capacity and GMP storage modulus per unit mass, ranged from 167.31% to 201.79% and 20.22 Pa·g^{-1} to 73.71 Pa·g^{-1}, respectively, in which the latter showed larger variation than the former. However, gluten *WHC* and GMP storage modulus per unit mass decreased across the milling streams.

(Quantity × stretching-degree) parameters, including flour SDS retention capacity, (dry gluten content) × (gluten water-holding capacity) and GMP storage modulus, ranged between 288.61–455.59%, 1 890–3 250‱, and 55.94–141.52 Pa, respectively. Dough rheological parameters, including dough stability and extensibility, ranged from 2.3 min to 32.8 min and 120 mm to 194 mm, respectively, with dough stability showing the highest coefficient of variation (*CV*) of all parameters.

Regression analysis on predicting dough stability. Within each category of milling streams, R^2 was calculated for 3 types of regression models, including quantity-based, stretching-degree-based and (quantity × stretching-degree)-based model to measure the relative contribution of each source to the total variance in dough stability (Table 3). For 28 flour samples from total milling streams, the quantity-based and stretching-degree-based model showed R^2 values of 0.182 and 0.059, respectively, in predicting dough stability. A higher R^2 value of 0.417 was observed in the (quantity × stretching-degree)-based model, which was significant at the 0.01 probability level.

For 10 early reduction flours, R^2 values were 0.703 and 0.547 for quantity-based and stretching-degree-based model, respectively, while the (quantity × stretching-degree)-based model had a R^2 value of 0.701. Results for 10 later reduction flours were quite similar to those of early reduction flour, with the R^2 value of the (quantity × stretching-degree)-based model exceeding that of the other two models. For 8 break flours, in particular, the (quantity × stretching-degree)-based model exhibited a high R^2 value of 0.984, which was significant at the 0.001 probability level. In summary, for all the categories of milling streams, the R^2 values from (quantity × stretching-degree)-based model for predicting dough stability were higher than those from quantity-based or stretching-degree-based model.

Regression analysis on predicting dough extensibility. Comparable results were obtained in the regression analysis for predicting dough extensibility (Table 4). For to-

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Table 2. Mean and range for physicochemical properties of milling streams flour for the wheat cultivar Shiluan 02-1

Quality parameters	Early reduction flours		Later reduction flours		Break flours		Average	*CV* (%)
	mean	range	mean	range	mean	range		
Protein quantity								
Protein content (%)	10.26	9.34–11.45	12.92	10.63–15.65	14.27	11.74–18.28	12.36	18.5
Dry gluten content (%)	10.83	9.96–11.70	13.20	11.30–16.20	13.66	11.60–19.00	12.48	16.5
GMP content (%)	2.29	1.89–2.49	2.84	2.24–3.71	3.01	2.11–4.20	2.69	21.5
Aggregate stretching degree								
Gluten water holding capacity (%)	191.34	183.78–201.79	181.21	174.45–189.74	181.14	167.31–192.24	184.81	4.3
GMP storage modulus per unit mass ($Pa \cdot g^{-1}$)	53.08	39.52–73.71	37.64	29.07–47.83	28.93	20.22–38.26	40.67	31.8
Quantity × stretching-degree								
Flour SDS retention capacity (%)	306.29	288.61–339.85	361.00	310.95–433.07	367.12	317.87–455.59	343.21	12.5
(Dry gluten content) × (gluten water holding capacity) (‰)	2 071.40	1 890.00–2 260.00	2 389.00	2 100.00–2 930.00	2 462.50	2 100.00–3 250.00	2 296.57	13.5
GMP storage modulus (Pa)	120.37	92.13–141.52	105.13	84.73–127.37	84.92	55.94–114.68	104.80	21.5
Dough rheological properties								
Dough stability (min)	21.77	5.90–32.80	18.80	2.80–29.20	19.09	2.30–29.50	19.94	45.9
Dough extensibility (mm)	160.70	135.00–184.00	155.90	130.00–194.00	151.63	120.00–178.00	156.39	11.9

CV – coefficient of variation; GMP – glutenin macropolymer; SDS – sodium dodecyl sulphate

https://doi.org/10.17221/64/2024-CJFS

Table 3. Regression analysis of different categories of milling streams using three types of models predicting dough stability

Milling streams category	Yield (%)	Order from the inner to the outer layer of wheat kernel	Model	Regression equation	R^2	F	Significance
Total milling streams flour	0–90	1–28	quantity-based model	dough stability = 7.300 + 2.634 × protein content + – 2.111 × dry gluten content + 2.569 × GMP content	0.182	1.785	0.177
			stretching-degree-based model	dough stability = 66.506 – 0.275 × gluten water holding capacity + – 0.115 × GMP storage modulus per unit mass	0.059	0.783	0.468
			(quantity × stretching-degree)-based model	dough stability = –29.536 –0.086 × flour SDS retention capacity + – 0.242 × (dry gluten content) × (gluten water holding capacity) + + 0.024 × GMP storage modulus	0.417	5.723	0.004
Early reduction flours	0–55	1–10	quantity-based model	dough stability = –106.334 + 8.211 × protein content + + 1.244 × dry gluten content + 13.242 × GMP content	0.703	4.737	0.050
			stretching-degree-based model	dough stability = 55.292 + 0.001 × gluten water holding capacity + – 0.634 × GMP storage modulus per unit mass	0.547	4.230	0.062
			(quantity × stretching-degree)-based model	dough stability = –44.969 + 0.193 × flour SDS retention capacity + – 0.244 × (dry gluten content) × (gluten water holding capacity) + + 0.019 × GMP storage modulus	0.701	4.689	0.051
Later reduction flours	55–79	11–20	quantity-based model	dough stability = 3.385 + 5.876 × protein content – 0.332 × dry gluten content – 19.774 × GMP content	0.346	1.057	0.434
			stretching-degree-based model	dough stability = 165.156 – 0.913 × gluten water holding capacity + + 0.509 × GMP storage modulus per unit mass	0.405	2.387	0.162
			(quantity × stretching-degree)-based model	dough stability = –30.675+0.544 × flour SDS retention capacity + + 0.054 × (dry gluten content) × (gluten water holding capacity) + – 0.065 × GMP storage modulus	0.553	2.476	0.159
Break flours	79–90	21–28	quantity-based model	dough stability = –34.161 + 7.786 × protein content + – 3.333 × dry gluten content – 4.113 × GMP content	0.788	4.965	0.078
			stretching-degree-based model	dough stability = 122.701 – 0.609 × gluten water holding capacity + + 0.233 × GMP storage modulus per unit mass	0.394	1.628	0.285
			(quantity × stretching-degree)-based model	dough stability = –16.494 – 0.203 × flour SDS retention capacity + + 0.555 × (dry gluten content) × (gluten water holding capacity) + + 0.024 × GMP storage modulus	0.984	82.714	< 0.001

R^2 – coefficient of determination; F – ratio of two variances; GMP – glutenin macropolymer; SDS – sodium dodecyl sulphate

https://doi.org/10.17221/64/2024-CJFS

Table 4. Regression analysis of different categories of milling streams using three types of models predicting dough extensibility

Milling streams category	Yield (%)	Order from the inner to the outer layer of wheat kernel	Model	Regression equation	R^2	F	Significance
Total milling streams flour	0–90	1–28	quantity-based model	dough extensibility = 85.803 + 7.686 × protein content + – 1.104 × dry gluten content – 3.939 × GMP content	0.531	9.070	< 0.001
			stretching-degree-based model	dough extensibility = 361.494 –1.018 × gluten water holding capacity + – 0.419 × GMP storage modulus per unit mass	0.376	7.520	0.003
			(quantity × stretching-degree)-based model	dough extensibility = –46.160 + 40.146 × flour SDS retention capacity + – 0.084 × (dry gluten content) × (gluten water holding capacity) + + 0.023 × GMP storage modulus	0.542	9.470	< 0.001
Early reduction flours	0–55	1–10	quantity-based model	dough extensibility = 138.678 + 3.368 × protein content + – 5.847 × dry gluten content + 22.139 × GMP content	0.045	0.095	0.960
			stretching-degree-based model	dough extensibility = 562.940 –2.067 × gluten water holding capacity + – 0.125 × GMP storage modulus per unit mass	0.494	3.421	0.092
			(quantity × stretching-degree)-based model	dough extensibility = 186.847 + 1.067 × flour SDS retention capacity + – 0.474 × (dry gluten content) × (gluten water holding capacity) + – 0.151 × GMP storage modulus	0.375	1.200	0.387
Later reduction flours	55–79	11–20	quantity-based model	dough extensibility = 19.645 + 3.867 × protein content + + 13.870 × dry gluten content – 34.116 × GMP content	0.697	4.604	0.053
			stretching-degree-based model	dough extensibility = 591.443-2.488 × gluten water holding capacity + + 0.409 × GMP storage modulus per unit mass	0.304	1.525	0.282
			(quantity × stretching-degree)-based model	dough extensibility = –66.379 + 1.220 × flour SDS retention capacity + + 0.276 × (dry gluten content) × (gluten water holding capacity) + – 0.106 × GMP storage modulus	0.748	5.938	0.031
Break flours	79–90	21–28	quantity-based model	dough extensibility = 26.065 + 20.371 × protein content + – 5.220 × dry gluten content – 31.235 × GMP content	0.927	16.924	0.010
			stretching-degree-based model	dough extensibility = 171.847 – 0.171 × gluten water holding capacity + + 0.369 × GMP storage modulus per unit mass	0.033	0.086	0.919
			(quantity × stretching-degree)-based model	dough extensibility = 79.370 – 0.443 × flour SDS retention capacity + + 0.678 × (dry gluten content) × (gluten water holding capacity) + + 0.069 × GMP storage modulus	0.840	7.000	0.045

R^2 – coefficient of determination; F – ratio of two variances; GMP – glutenin macropolymer; SDS – sodium dodecyl sulphate

https://doi.org/10.17221/64/2024-CJFS

tal milling streams flour, the quantity-based and (quantity × stretching-degree)-based model had higher R^2 values of 0.531 and 0.542, respectively which were significant at the 0.001 probability level, compared to the stretching-degree-based model with the R^2 value of 0.376 that was only significant at the 0.01 probability level.

For early reduction flours, the quantity-based and stretching-degree-based model showed R^2 values of 0.045 and 0.494, respectively, while the R^2 value of the (quantity × stretching-degree)-based model was 0.375. For later reduction flours, the (quantity × stretching-degree)-based model exhibited a R^2 value of 0.748, which was higher than that of the quantity-based and stretching-degree-based model, but at the same significant level with the quantity-based model. In break flours, the quantity-based and (quantity × stretching-degree)-based model showed R^2 values of 0.927 and 0.840, respectively, at the same significant level and both were higher than the R^2 value of the stretching-degree-based model. To sum up, higher R^2 values from (quantity × stretching-degree)-based model for predicting dough extensibility were observed in total milling streams flour and later reduction flours, compared to those from quantity-based and stretching-degree-based model.

DISCUSSION

Although the amount of protein and protein fraction has long been confirmed to correlate with dough rheological parameters, there is also a certain limitation when it comes to comparing wheat samples with genetic and environmental difference, including weather conditions and soil in different locations, fertilisation, irrigation and mutual interaction between those factors (Wieser et al. 2023). Using milling streams flour from one wheat cultivar could provide the samples with different protein content and varying degree of gluten strength, while ensuring the same genetic background, storage conditions and other relevant treatments of the wheat grains before milling.

The variation in the protein quantity of flour from different milling streams in this study was consistent with the reports of Sutton and Simmons (2006), where higher flour protein content and SDS-insoluble glutenin were seen in break streams of a pilot-scale flour mill, compared to those of reduction streams. They have studied the relationship between protein quantity and dough processing quality for 2 strong wheat cultivars and 2 weak ones, suggesting that farinograph dough stability was in positive correlation with SDS-insoluble glutenin for 2 strong cultivars. A significant positive relationship of flour protein content with mixograph peak time and peak width was shown in all 4 cultivars.

Gluten water-holding capacity is a measure of the amount of water that is absorbed per gram of gluten protein. In this study, the changes in gluten *WHC* of flour from different milling streams were in agreement with the results of Dai et al. (2020). In their study, flour fractions from the outer layer to the inner layer of wheat kernels were obtained using a pearling mill, and the *WHC* was seen highest in the fraction close to the inner layer of wheat kernels, while the lowest *WHC* came from the outer layer fraction. In addition, our flour samples from 28 milling streams were sorted using the value of (ash content/L^*), and the flours lower in ash content and higher in L^* were classified into early reduction streams, which corresponded to the characteristics of inner-layer fractions for wheat kernels.

To characterise the degree of gluten development during hydration, flour solvent retention capacity (*SRC*) is also a popular parameter. In *SRC* measurement, selected diagnostic solvents are independently used to produce *SRC* values, for example 5% lactic acid is used to produce the *SRC* value associated with the swelling behaviour of the glutenin polymer network (Kweon et al. 2011). However, *SRC* value might be a combination of protein quantity and stretching degree, that is, the more protein in flour or the more stretched the protein aggregate, the higher *SRC* value may be produced. If the *SRC* value is divided by the amount of protein, *SRC* of protein per unit mass would then be obtained, which reflects the average swelling degree of protein. A higher *SRC* of protein per unit mass indicates the protein network that is more stretched and expanding. After comparing the relationship of *SRC* versus dough rheological parameters (in farinograph) with *SRC* of protein per unit mass versus those parameters, a general decrease of the correlation was seen in the latter for several studies (Hammed et al. 2015; Singh et al. 2018; Magallanes López and Simsek 2021), corroborating our idea that both protein quantity and aggregate stretching degree play a role in *SRC*, which encouraged us to further explore whether the idea also applies to predicting dough rheological parameters.

In our regression analysis, 3 types of models were established, including quantity-based, stretching-degree-based and (quantity × stretching-degree)-based model, to predict dough stability and extensibility. In the third model, flour SDS retention capacity was considered as a parameter that reflected both protein quantity and aggregate stretching degree at the level of wheat flour, and (dry gluten content) × (gluten water-holding ca-

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pacity) and GMP storage modulus were the parameters at the level of gluten and glutenin fraction, respectively. R^2 values of regression models were used to evaluate the relative accuracy of each model to predict dough stability or extensibility, and the higher R^2 value indicated a relatively accurate model, even if the model was not significant. Our results showed that for total milling streams flour and its 3 divided categories, the (quantity × stretching-degree)-based model outperformed both the quantity-based and the stretching-degree-based model in dough stability prediction; the advantage of the (quantity × stretching-degree)-based model was also seen in predicting dough extensibility for total milling streams flour and later reduction flours. The variation of R^2 values in different categories could be attributed to the difference in protein composition. Wang et al. (2007) indicated that the gliadin percentage in total flour protein increased from break streams to reduction streams, using flours produced from a laboratory experimental mill. And the percentage of polymeric glutenin was also reported to increase dramatically within break streams, and then decrease within reduction streams. Sutton and Simmons (2006) also found higher thiol content in later reduction and break streams due to more molecular disruption caused by intensive grinding, which could then affect the oxidation of thiol groups into disulphide bonds during flour storage. However, in spite of those considerable differences across the milling streams which resulted in dough stability and extensibility that varied in a wide range, our established (quantity × stretching-degree)-based model showed higher R^2 values than the quantity-based model and the stretching-degree-based model in six cases out of eight. The results suggest that measuring protein quantity and aggregate stretching degree at the same time is of practical improvement in dough stability and extensibility prediction, compared to focusing on either factor alone.

CONCLUSION

Flours from 28 milling streams of the wheat cultivar Shiluan 02-1 were sorted using the value of (ash content/L^*) from the inner to the outer layer of wheat kernel, and then divided into early reduction flours, later reduction flours and break flours as experimental material. Dough rheological properties are associated with the quantity of protein in wheat flour and quality of gluten development, and the 'aggregate stretching degree', representing the volume of protein per unit mass, was proposed to quantitatively characterise the degree of gluten development in the dough system. Parameters relating to protein quantity, aggregate stretching degree and (quantity × stretching-degree) were examined for all the flour samples. Increase in protein quantity and decrease in aggregate stretching degree have been observed from early reduction to later reduction streams and then to break streams.

To figure out the relationship between large deformation rheology of wheat flour dough with protein quantity and aggregate stretching degree, the R^2 values of three types of regression models, including quantity-based, stretching-degree-based, and (quantity × stretching-degree)-based model for predicting dough stability and extensibility, were compared in each category of milling streams. The quantity-based model broadly produced greater R^2 values than the stretching-degree-based model. The fact that the (quantity × stretching-degree)-based model could in general provide higher R^2 values than the other two models on predicting dough stability and extensibility indicates that protein quantity and aggregate stretching degree are both involved in determining these dough properties, and that we can better evaluate dough rheology combining protein quantity and aggregate stretching degree. The findings can help wheat processing industries to evaluate and control flour quality more accurately and conveniently.

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Received: April 4, 2024
Accepted: August 20, 2024
Published online: September 19, 2024