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DEVELOPING AND EVALUATING NOVEL METHODS TO ESTIMATE BREAD
BAKING QUALITY OF WINTER AND SPRING WHEAT CULTIVARS

BY

BIPIN RAJPUROHIT

A thesis submitted in partial fulfillment of the requirements for the

Master of Science

Major in Biological Sciences

Specialization in Food Science

South Dakota State University

2020

THESIS ACCEPTANCE PAGE

Bipin Rajpurohit

This thesis is approved as a creditable and independent investigation by a candidate for the master's degree and is acceptable for meeting the thesis requirements for this degree.

Acceptance of this does not imply that the conclusions reached by the candidate are necessarily the conclusions of the major department.

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Date

I dedicate this thesis to:

My parents - Without their affection and encouragement, it would not have been possible to achieve any success in life.

Vimmy - My lovely wife, for her continuous support and encouragement.

Naveen and Vishal - My beloved brothers. Without their patience, understanding, support and love, I would not have completed my studies.

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CONTENTS

ABBREVIATIONS	x
LIST OF FIGURES	xii
LIST OF TABLES	xv
ABSTRACT	xix
CHAPTER 1. INTRODUCTION	1
1.1 Introduction.....	1
1.2 Literature Review.....	3
1.2.1 Rheological tests	4
1.2.1.1 The Farinograph.....	5
1.2.1.2 The Mixolab.....	6
1.2.2 Other wheat quality tests.....	8
1.2.2.1 Sodium dodecyl sulfate (SDS) sedimentation test.....	8
1.2.2.2 Solvent retention Capacity (SRC) test	8
1.2.3 Dough expansion studies	9
1.2.3.1 The Alveograph	12
1.3 Objectives of the Study.....	15
1.4 Hypotheses.....	16
CHAPTER 2. MATERIALS AND METHODS	17
2.1 Sample selection	19

2.1.1 Preparation of flour blends from pastry flour	19
2.1.2 Cleaning, tempering and milling of wheat.....	21
2.2 Analysis of flour and dough.....	21
2.2.1 Flour tests.....	21
2.2.1.1 Moisture analysis	21
2.2.1.2 Gluten analysis.....	22
2.2.1.3 Protein determination.....	23
2.2.1.4 The hybrid SDS-SRC method.....	24
2.2.2 Dough tests.....	25
2.2.2.1 Mixolab	25
2.2.2.2 The vacuum dough expansion system (VDES)	26
2.2.2.2.1 Automated mode.....	27
2.2.2.2.2 Manual mode	27
2.2.2.2.3 Mixing of dough for Vacuum expansion: Utilizing the Swanson 200 g mixer	28
2.2.2.2.4 Vacuum dough expansion.....	29
2.2.3 Bread baking	30
2.2.3.1 Bread loaf volume measurement	31
2.2.3.2 C-Cell analysis	31
2.3 Statistical analysis.....	32
CHAPTER 3. RESULTS AND DISCUSSIONS.....	34

3.1 Mean values and range of occurrence of wheat quality traits in 24 cultivars of HRW wheat and 33 cultivars of HRS wheat.	35
3.2 Preliminary investigation on the Vacuum Dough Expansion System	43
3.2.1 Dough expansion experiments utilizing flour blends made from pastry flour: Experiment 1	43
3.2.2 Dough expansion experiments utilizing flour blends made from pastry flour: Experiment 2	50
3.3 Loaf volume predictors in hard red winter (HRW) wheat cultivars	58
3.3.1 Dough expansion height as a predictor of baked loaf volume in HRW wheat 62	
3.3.1.1 Correlation between dough expansion height (from the VDES test) and other flour and dough quality parameters	62
3.3.2 Hybrid SDS-SRC sedimentation test as a predictor of loaf volume in HRW wheat	64
3.3.2.1 Correlation between hybrid SDS-SRC sedimentation method and other flour and dough quality parameters in HRW wheat	67
3.3.3 Other variables as predictor of bread loaf volume in HRW wheat.....	68
3.3.3.1 Grain protein and flour protein	68
3.3.3.2 Variables from the Gluten Analysis Tests (Glutomatic Analyzer).....	68
3.3.3.3 Variables from the Mixolab	69
3.3.4 Multiple regression models developed through stepwise procedure for predicting bread loaf volume in HRW wheat	70

3.3.5 Multiple regression models developed by combining weight value and dough expansion height for predicting bread loaf volume	71
3.4 Loaf volume predictors in hard red spring (HRS) wheat cultivars	74
3.4.1 Dough expansion height as a predictor of baked loaf volume in HRS wheat .	76
3.4.1.1 Correlation between dough expansion height and other flour and dough quality parameters in HRS wheat	80
3.4.2 Hybrid SDS-SRC sedimentation test as a predictor of loaf volume in HRS wheat	82
3.4.2.1 Correlation between hybrid SDS-SRC sedimentation method and other flour and dough quality parameters in HRS wheat	86
3.4.3 Other variables as predictor of baked loaf volume in HRS wheat.....	88
3.4.3.1 Grain protein and flour protein	88
3.4.3.2 Variables from the Glutomatic test	88
3.4.3.3 Variables from the Mixograph.....	89
3.4.3.4 Variables from the Farinograph	90
3.4.4 Multiple regression models developed through stepwise procedure for predicting bread loaf volume in HRS wheat.....	90
3.4.5 Multiple regression models developed by combining weight value and dough expansion height for predicting bread loaf volume in HRS wheat.....	93
CHAPTER 4. CONCLUSIONS	94
4.1 Dough expansion attributes as indicator of baking quality.....	94
4.1.1 Recommendations for improving the VDES	96

4.1.1.1 Integrate the mixing and resting function in the VDES.....	96
4.2 The hybrid SDS-SRC sedimentation test as an indicator of baking quality	97
4.3 Combining the weight value and dough expansion height in multiple regression equation to predict bread loaf volume	98
APPENDIX.....	99
REFERENCES	106

ABBREVIATIONS

AACCI	American Association of Cereal Chemists International
AYT	Advanced yield trials
AC	Air cells
BU	Brabender Units
CA	Cells per unit area
cc	cubic centimeter
CD	Cell diameter
cm	centimeter
CPT	Crop performance trials
FDDT	Farinograph dough development time
FG	Flour Gluten
FMTI	Farinograph mixing tolerance index
FP	Flour Protein
FQN	Farinograph quality number
FSTAB	Farinograph stability
FWA	Farinograph water absorption
g	gram
GP	Grain protein
GWG	Good wet gluten
HRS	Hard red spring
HRW	Hard red winter
In	Inch

LV	Loaf volume
m.b.	Moisture basis
mm	millimeter
MGA	Mixograph absorption
MGM	Mixograph mix time
MGPT	Mixograph peak time
MLA	Mixolab absorption
MLS	Mixolab stability
MTI	Mixing tolerance index
psi	per square inch
NIRS	Near Infrared Spectroscopy
SDS	Sodium dodecyl Sulfate
SRC	Solvent retention capacity
SLV	Specific loaf volume
TWG	Total wet gluten
VDEH	vacuum dough expansion height
VDES	Vacuum dough expansion system
VWG	Vital wheat gluten
WAC	Water absorption capacity
WT	Wall thickness
WV	Weight Value

LIST OF FIGURES

Figure 1.1 Figure showing a farinogram of strong gluten flour	6
Figure 1.2 Figure depicting a typical Mixolab curve.....	7
Figure 2.1 Flow chart of the experimental design used in the research.....	18
Figure 2.2 The Glutomatic system.....	22
Figure 2.3 Diagram depicting the process of gluten analysis on the Glutomatic system .	23
Figure 2.4 Flash EA 1112 N/Protein analyzer	24
Figure 2.5 The Chopin Mixolab.....	25
Figure 2.6 Mixing settings in Chopin+ protocol (Mixolab Handbook, 2012).....	26
Figure 2.7 The Vacuum Dough Expansion System.....	28
Figure 2.8 The 200g Swanson mixer	29
Figure 2.9 The C-Cell instrument	32
Figure 2.10 Image analysis of bread slice by C-Cell	32
Figure 3.1 Relationship between gluten concentration and bread loaf volume. The gluten concentration is expressed on 14 % moisture basis.	46
Figure 3.2 Bread baked from control weak flour (Pastry flour) and blends of pastry flour enriched with vital wheat gluten.....	47
Figure 3.3 Relationship between gluten concentration and dough expansion height. The gluten concentration is expressed on 14 % moisture basis.	48
Figure 3.4 Relationship between baked loaf volume and dough expansion height for weak dough system	49
Figure 3.5 Relationship between gluten content and baked loaf volume in control pastry flour and blends of pastry flour enriched with vital wheat gluten	54

Figure 3.6 Bread baked from control weak flour (Pastry flour) and blends of pastry flour enriched with vital wheat gluten.....	55
Figure 3.7 Relationship between dough expansion height and bread loaf volume for control pastry flour and blends of pastry flour enriched with vital wheat gluten	56
Figure 3.8 Relationship between gluten content and dough expansion in control pastry flour and blends of pastry flour enriched with vital wheat gluten.	57
Figure 3.9 Relationship between dough expansion height (VDEH) and specific loaf volume (SLV) for 24 cultivars of HRW wheat grown at Aurora growing location.	61
Figure 3.10 Relationship between weigh value and specific loaf volume for 24 cultivars of HRW wheat grown at Aurora growing location (N=24).....	65
Figure 3.11 Relationship between weigh value and specific loaf volume for 24 cultivars of HRW wheat grown at Onida growing location (N=24).	66
Figure 3.12 Relationship between dough expansion height and bread loaf volume for 33 cultivars of HRS wheat grown at three locations. The plot consists of data from all the three growing locations (N=99).....	77
Figure 3.13 Relationship between flour protein and dough expansion height. Data for 33 cultivars grown at three different growing locations are plotted in the same graph.....	78
Figure 3.14 Relationship between weigh value and bread loaf volume for 33 cultivars of HRS wheat grown at Brookings location (N=33).....	83
Figure 3.15 Relationship between weigh value and bread loaf volume for 33 cultivars of HRS wheat grown at Groton growing location (N=33).....	84
Figure 3.16 Relationship between weigh value and bread loaf volume for 33 cultivars of HRS wheat grown at Selby growing location (N=33).....	85

Figure 4.1 Dough expansion profile of bread flour procured from Costco 97

LIST OF TABLES

Table 2.1 Description of the control pastry flour and the gluten concentration of the blends prepared by incorporating Vital Wheat gluten in the pastry flour.....	19
Table 2.2 Description of the control pastry flour and the gluten concentration of the blends prepared by incorporating Vital Wheat gluten in the pastry flour.....	20
Table 2.3 Classes of wheat with the corresponding growing locations	21
Table 2.4 Formula used in the modified Basic Straight-Dough Bread-making Method ..	30
Table 3.1 Summary of quality parameters of the 24 cultivars of HRW wheat grown at Aurora and Onida growing locations	36
Table 3.2 Estimate of the variance components of routine flour and dough tests used for flour end use quality, the hybrid SDS-SRC Sedimentation test, and the dough expansion test for 24 cultivars of HRW wheat grown at two growing locations	38
Table 3.3 Summary of quality parameters of the 33 cultivars of HRS wheat grown at Brookings, Groton and Selby growing locations	41
Table 3.4 Estimate of the variance components of routine flour and dough tests used for flour end use quality, the hybrid SDS-SRC Sedimentation test, and the dough expansion test for 33 cultivars of HRS wheat grown at three growing locations	42
Table 3.5 Bivariate correlation coefficients (r) between variables of gluten content, dough expansion, baked loaf volume and C-Cell for control pastry flour and blends of pastry flour with increasing vital wheat gluten concentration. $N=5$	45
Table 3.6 Bivariate correlation coefficients (r) between variables of gluten content, dough expansion and baked loaf volume for control pastry flour and blends of pastry flour enriched with vital wheat gluten ($N = 12$ for All; $N = 6$ for mean).....	53

Table 3.7 Bivariate correlation coefficients (r) between specific loaf volume and variables from the VDES, the hybrid SDS-SRC and the conventional predictors of bread quality in HRW wheat cultivars	60
Table 3.8 Bivariate correlation coefficients (r) between the variables of grain and flour quality, dough mixing and quality and baked loaf characteristics for HRW wheat grown in Aurora growing location (N-24)	63
Table 3.9 Linear regression models developed through stepwise procedure	72
Table 3.10 Linear regression model developed by entering WV and VDEH as predictor variables with response to specific loaf volume for HRW wheat.....	73
Table 3.11 Bivariate correlation between baked loaf volume and variables of grain and flour quality, dough mixing and quality for HRS wheat grown at three growing locations	75
Table 3.12 Correlation coefficients between wheat quality parameters and dough expansion height in the three growing locations of HRS wheat	81
Table 3.13 Correlation coefficients between wheat quality parameters and weight value from the hybrid SDS-SRC sedimentation method in the three growing locations of HRS wheat cultivars	87
Table 3.14 Multiple regression models for HRS wheat samples developed through stepwise procedure by using multiple predictor variables with response to bread loaf volume.....	91
Table 3.15 Multiple regression models developed by entering WV and VDES as predictor variables with response to bread loaf volume for HRS wheat cultivars.....	92

Table A.1 Bivariate correlation coefficients (r) between the variables of grain and flour quality, dough mixing and quality, and baked loaf characteristics for HRS wheat grown in Brookings location (N-33)	99
Table A.2 Bivariate correlation coefficients (r) between the variables of grain and flour quality, dough mixing and quality and baked loaf characteristics for HRS wheat cultivars grown in Selby growing location (N- 33)	100
Table A.3 Bivariate correlation coefficients (r) between the variables of grain and flour quality, dough mixing and quality and baked loaf characteristics for HRS wheat cultivars grown in Groton location (N-33)	101
Table A.4 Bivariate correlation coefficients (r) between the variables of grain and flour quality, dough mixing and quality and baked loaf characteristics for HRS wheat cultivars grown in Brookings, Selby and Groton growing locations. The data from all the three locations was pooled to compute the correlations (N-99)	102
Table A.5 Bivariate correlation coefficients (r) between the variables of grain and flour quality, dough mixing and quality and baked loaf characteristics for HRS wheat grown in Brookings, Selby and Groton growing locations. The data for each variable for a cultivar was averaged across the three growing locations to compute the correlations (N-33)...	103
Table A.6 Bivariate correlation coefficients (r) between the variables of grain and flour quality, dough mixing and quality and baked loaf characteristics for HRW wheat cultivars grown in Aurora and Onida growing locations. The data from both the growing locations was pooled to compute the correlations (N-48)	104

Table A.7 Bivariate correlation coefficients (r) between the variables of grain and flour quality, dough mixing and quality and baked loaf characteristics for HRW wheat cultivars grown at Onida growing location (N-24).....	105
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ABSTRACT

DEVELOPING AND EVALUATING NOVEL METHODS TO ESTIMATE BREAD
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BIPIN RAJPUROHIT

2020

Bread loaf volume is a significant economic factor in the baking industry. Numerous flour and dough quality tests exist in estimating the functional traits of wheat. However, none of these tests accurately or fully predict bread loaf volume. The ultimate reliable method to determine the baking potential of wheat flours is a standard baking experiment which is time-consuming and effort-intensive. Therefore, there is a need to develop empirical methods to predict the baking potential of wheat flours that use small sample sizes and yet, are economical to implement in large throughput wheat breeding programs.

A Vacuum Dough Expansion System (VDES) was fabricated to expand optimally developed dough prepared from flour and water. Dough expansion was carried out in a chamber and the maximum height of the expanding dough was recorded. The VDES was evaluated for its ability to consistently expand dough and its applicability in predicting bread loaf volume.

The study also investigated a simple and rapid chemical test. One such test is the hybrid SDS-SRC sedimentation designed for evaluating winter wheat quality. This test was evaluated for applicability to hard red spring (HRS) wheat as well. The hybrid SDS-SRC test measures gluten proteins that precipitate on the addition of solvents and

detergents. Conventional flour tests, dough quality tests and baking experiments were also done in parallel to determine relationships between various tests.

Preliminary investigation to study the applicability of the VDES was performed on a low-protein pastry flour (7.3 % gluten content) which was further enriched with vital gluten (7.3 to 14.7% vital gluten). Dough expansion height was statistically significantly correlated with corresponding bread loaf volume ($r = 0.96$). Additionally, dough expansion height was statistically significantly correlated to gluten content in the blends ($r = 0.97$). Two validation studies were then conducted employing a total of 147 samples from two economic classes of wheat namely hard red spring (HRS) and hard red winter (HRW).

In the investigation on 24 HRW wheat cultivars, specific loaf volume (SLV; Loaf volume/weight) was statistically significantly correlated to dough expansion height ($r = 0.47$) and weight value (obtained from the hybrid SDS-SRC sedimentation test, $r = 0.54$). The best predictor of SLV in this sample set was flour protein content ($r = 0.62$).

In another investigation comprising 33 HRS wheat cultivars grown at three growing locations, loaf volume was found to be significantly correlated with dough expansion height ($r = 0.38$). However, in individual growing locations, predictability of bread loaf volume from dough expansion height was not seen. Statistically significant correlation was also found between weight value and bread loaf volume in all the three growing locations of HRS wheat samples. The correlation coefficient (r) between weight value and bread loaf volume was 0.50 in Brookings growing location, 0.59 in Groton growing location, and 0.41 in Selby growing location.

The research established a proof of concept for dough expansion and its potential applicability in measuring the baking quality of wheat. The VDES provided a linkage between true baking tests and other chemical indicators of wheat and dough quality. The effect of vital wheat gluten addition on dough expansion height as evaluated by the VDES was linear and incremental. The hybrid SDS-SRC sedimentation test was effective and consistent in predicting the loaf volume of HRW as well as HRS wheat cultivars. The test was also effective in predicting loaf volume in the environment affected by drought and cultivars with high protein.

CHAPTER 1. INTRODUCTION

1.1 Introduction

Wheat is one of the main human food crops worldwide. It is consumed in several foods such as bread, porridge, biscuits, muesli, cookies, cakes, donuts and pastries. In the United States, wheat has ranked 3rd among field crops in planted acreage as well as farm receipts during the last decade. The United States is the fifth largest producer of wheat in world and almost half of the wheat produced is exported. The estimated wheat production in United States for 2018/19 was 1.884 billion bushels. The 5 major classes of U.S. wheat are: hard red winter (HRW), hard red spring (HRS), soft red winter, white, and durum. Each class has different end uses. Hard red winter (HRW) and hard red spring (HRS) account for 40 % and 20 % of total production, respectively, and are used principally to make bread flour and specialty breads, respectively.

Bread is a staple food prepared from wheat flour and is popular around the world. The quality of bread depends on the bread baking quality of flour which may be affected by a number of factors such as gluten quality and quantity, starch quality and enzyme activity. The quality of bread is measured by parameters such as loaf volume and crumb texture. Bread loaf volume is the holy grail of the baking industry. Bread loaf volume can be measured easily without need for sophisticated instrumentation. The texture of a bread slice can be studied objectively by employing a C-Cell bread crumb analyzer.

An important aim in cereal science research has been the development of procedures to measure bread-making potential of wheat flours without going through actual baking. The essential requirements of such test are rapid, accuracy and the

requirement of very small amount of sample. Also, wheat breeders do not provide large sample size. The development of such tests will enable the wheat breeders to evaluate bread making quality of new wheat lines during breeding programs at the early generation stages (MacRitchie, 2014). Traditionally, it has been necessary to do actual baking to determine bread quality (loaf volume) and this may involve 3-4 hours/loaf.

Over the years, breeders have relied on a number of methods for predicting bread-making potential of wheat. Historically, they have relied on the fact that a moderate correlation exists between grain protein content and bread loaf volume. Other wheat functionality tests that have been used by hard winter wheat breeders are the American Association of Cereal Chemists International (AACCI) 56-70 sodium dodecyl sulfate (SDS) sedimentation method and the AACCI method 56-11 solvent retention capacity (SRC) test. Seabourn et al. (2012) developed a hybrid SDS- SRC method to develop a rapid, small-scale method to predict bread making potential of hard winter wheat. Results obtained from the hybrid method showed a higher correlation to bread volume as compared to the individual assays. The assay combined the solutions used in the SDS sedimentation method and the centrifugation process found within the SRC method. The hybrid method was performed in 66 % less time than the SDS methods and the in-house Hard Winter Wheat Quality laboratory (HWWQL) methods. Moreover, the hybrid SDS-SRC sedimentation assay can be performed utilizing merely 1 g of sample.

The Vacuum Dough Expansion System (VDES) is a simple device that employs vacuum to expand a piece of dough and may have ability to predict bread loaf volume. Dough is the intermediate stage between flour and bread, therefore testing the strength

and properties of dough may help in predicting quality of bread. Conventionally, the properties of dough have been studied using instruments such as Brabender Farinograph, Extensigraph and Alveograph. Expansion capacity is an inherent property of dough and may have potential to predict baking performance of dough (Gandikota & MacRitchie, 2005). VDES not only has the potential to predict baking performance of a flour but may also help to evaluate the effects of dough conditioners such as gluten, yeast, eggs, soy lecithin and ascorbic acid on bread loaf volume. Successful validation of a dough expansion method utilizing the VDES will be valuable for evaluating baking potential of wheat flours in breeding programs.

The purpose of this study was to develop a method of dough expansion employing the VDES and study its applicability in predicting bread loaf volume. Additionally, the study investigated applicability of the hybrid SDS-SRC sedimentation test for hard red spring (HRS) Wheat. The study also established relationships between the routine flour and dough tests used for flour end use quality, VDES data and baked loaf characteristics. The routine flour tests determine the flour protein, the Glutomatic test measures gluten quality and quantity. The dough testing instruments used in the study include the Mixograph, Farinograph and the Mixolab.

1.2 Literature Review

The literature review focused on the evolution of methodologies used for predicting bread-making quality of wheat. Focus was placed on the advancement of rapid chemical tests that determined gluten quantity and correlated with bread volume. Along with the flour and dough quality tests, the advancement and status of dough expansion

systems was also reviewed. The quality of bread is affected by several factors and quality evaluation is important at many levels. A number of tests are used to evaluate flour and dough quality, which may in turn, then predict end-product quality. However, the evaluation of the finished product, namely bread, is the ultimate quality test.

1.2.1 Rheological tests

Rheology is the study of flow and deformation of materials and involves the application of a well-defined deformation (strain) to material over a period of time. The response of the material (stress) is measured to provide additional information regarding the product. Dobraszczyk and Morgenstern (2003) stated that the objectives of rheological tests are “to obtain quantitative description of material’s mechanical properties, to obtain information related to molecular structure and composition of material, and to characterize and simulate material’s performance during processing and for quality control”. Traditionally, the subjective judgement of bakers is used to evaluate dough quality by checking dough stickiness. Bread is scored on a scale of 1 to 10, based on subjective evaluation.

The process of mixing is a critical step in the bread-making procedure. Mixing creates small gas bubbles in dough which expand during fermentation, causing dough to inflate. The whole structure is then set during the baking step. After baking, parameters such as bread volume and texture of the loaf are then used to determine the bread-making quality of the dough. In dough evaluation, a balance between extensibility of dough and resistance of dough to collapse is desired (MacRitchie, 2014). The behavior of the dough during the mixing stage may be used to predict quality of the end product. Measurement

of the mixing profile by dough recording mixers such as Farinograph, Mixograph or Mixolab is a popular way to determine dough quality. These instruments monitor the resistance of the dough against the mixing action of the blades and rheological changes taking place during mixing are known. The mixing curve comprises of two parts; the development stage and breakdown stage. The dough consistency increases during the development stage and the mixing curve reaches peak; this is known as dough development time. Mixing profile provides important information on gluten quality.

1.2.1.1 The Farinograph

One of the most widely used dough recording mixers used in industry is the Brabender Farinograph developed in 1930s. Figure 1.1 shows a farinogram obtained for a strong gluten flour. The important parameters measured from Farinograph device are the water absorption, the dough development time, the mixing stability and the mixing tolerance index (MTI). The water absorption quantifies the amount of water required to form a dough with optimum consistency and is expressed as percentage of flour weight. The optimum consistency of flour is realized when the mixing curve reaches the 500 Brabender units (BU) line. The dough development time or the peak time is the time required for dough to reach maximum consistency. Arrival time is the time when the top of the curve touches the 500 BU line. It indicates the rate of hydration. Departure time is the time when top of the curve leaves the 500 BU line. This indicates the time when dough begins to break. Stability time is the difference in time between the arrival time and the departure time. The stability time indicates the time the dough maintains maximum consistency and indicates dough strength. Mixing Tolerance Index (MTI) is the difference in BU units at the top of the curve at peak time and the value at top of the

curve five minutes after the peak time. It is expressed in Brabender Units and indicates the degree of dough softening during mixing. Parameters obtained from the Farinograph are used to determine amount of water required to make optimum dough, the stability of the dough and the amount of optimal mixing time to form the dough. The Farinograph can also be used to evaluate the effects of ingredients on dough development. The Farinograph results may be used to predict finished product quality attributes.

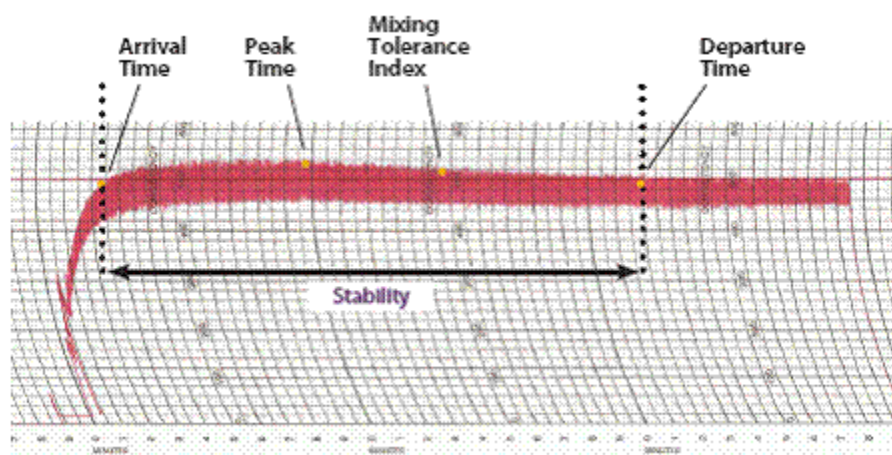


Figure 1.1 Figure showing a farinogram of strong gluten flour

1.2.1.2 The Mixolab

The Mixolab is a dough recording mixer developed by Chopin Technologies. It operates at variable temperatures unlike the Farinograph. In addition to mixing attributes, it evaluates pasting characteristics of dough when subjected to heating. It measures flour water absorption, dough development time, and dough mixing stability just like a Farinograph. Figure 1.2 shows a typical Mixolab output curve. A typical Mixolab output comprises of 5 stages. The first stage corresponds to dough formation(C1), the second stage corresponds to protein weakening (C2), the third stage corresponds to swelling of

starch granules (C3), the fourth stage measures alpha amylase activity (C4) and the fifth stage measures the starch retrogradation (C5).

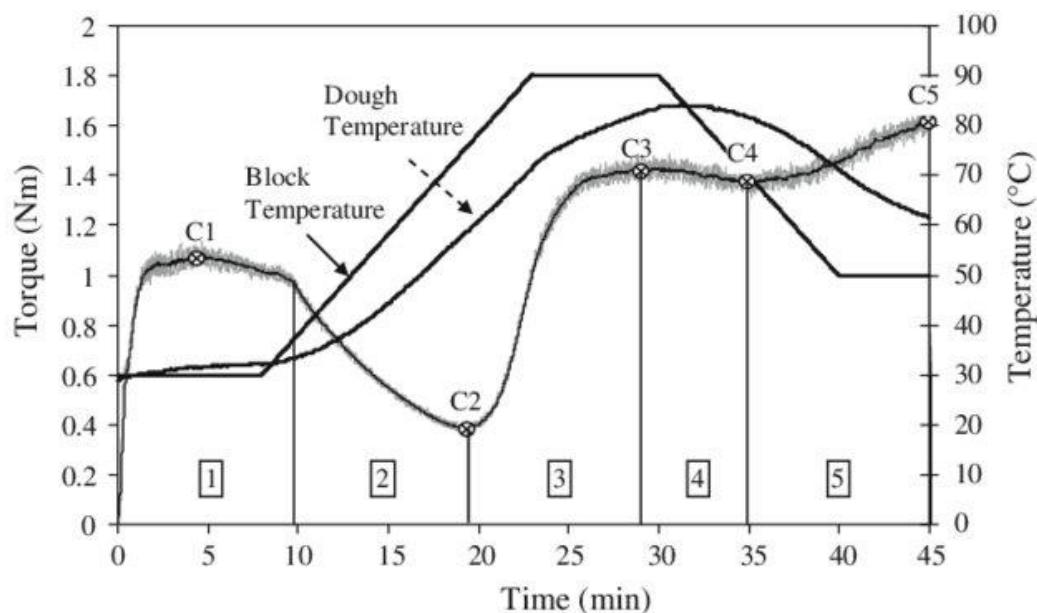


Figure 1.2 Figure depicting a typical Mixolab curve

There are limited number of studies using Mixolab to study dough rheology. A general agreement was found between Farinograph and Mixolab values according to Koksel et al. (2009). They also found correlation between bread loaf volume and C3($r=-0.53$), C4 ($r=-0.51$), C5 ($r=-0.61$) and difference between C4 and C5($r=-0.60$). Caffè-Treml et al. (2010) found statistically significant correlation between loaf volume and water absorption ($r=0.62$), dough development time ($r=0.63$), mixing stability ($r=0.70$), C2($r=-0.54$), beta ($r=0.51$) and C3 ($r=0.59$) for genotype means of 18 cultivars grown across 20 environments ($n = 18$). Thus, some of the mixing properties and rheology of the dough are related to end-product quality and may be used to predict finished end product quality. The Mixograph, Mixolab and Farinograph have been used traditionally

to study rheological properties of dough which is the intermediate stage between flour and bread. There is a logical expectation that the final loaf volume can be predicted from the rheological properties of the dough. However this expectation has not been realized (MacRitchie, 2014).

1.2.2 Other wheat quality tests

1.2.2.1 Sodium dodecyl sulfate (SDS) sedimentation test

For a given wheat cultivar, a linear relationship between protein content and loaf volume was found by Finney (1943) and Finney and Barmore (1948). Hard winter wheat breeders have relied on the American Association of Cereal Chemists International (AACCI) Method 56-70 (sodium dodecyl sulfate (SDS) sedimentation method) for predicting bread making potential of wheat. This procedure uses approximately 30 minutes to complete. The test is a good indicator of gluten quality and quantity which are in turn related to bread volume. Some of the limitations of the test include inability of the test to work on flours with protein levels greater than 14 % (Preston et al., 1982; Ayoub et al., 1993) and high probability of handling errors being introduced to results due to sensitivity of test to variables such as centrifuge time and shaking time.

1.2.2.2 Solvent retention Capacity (SRC) test

Another test that measures gluten quality is the Solvent Retention Capacity (SRC) test which involves a centrifugation step. The test has been successfully used to predict quality of soft wheat products and hard winter wheat bread volume ($r = 0.83$) (Xiao et al., 2006). However, the test takes 50 minutes to implement. Breeding programs require simple tests that are quick and efficient to be economical.

Due to the limitations of the SDS test and the SRC test, Seabourn et al. (2012) developed a hybrid SDS-SRC sedimentation protocol which can be performed in less than 10 minutes. The protocol combined the solutions used in the SDS method and the centrifugation process employed in the SRC method. The hybrid method yielded a higher correlation to bread volume ($r \geq 0.84$) than the AACC method on 53 hard winter wheat (HWW) varieties. The study by Seabourn et al. (2012) established a proof of concept for the hybrid test with further validation utilizing HWW varieties. However, further studies are required to see if the method works well for spring wheat and winter wheat with a larger population and greater range of variation of quality traits.

Karki et al. (2016) used weather data to predict loaf volume of bread using artificial neural network. Weather data was collected at various intervals and correlated with flour quality, dough quality and baked loaf volume. Neural network (NN) models were developed and compared to arrive at the best model that could predict bread loaf volume from selected input data. Best predictions of bread loaf volume were obtained from weather data obtained at 20 days after heading (DAH). The best predictors of bread loaf volume were found to be nighttime, minimum, and maximum temperatures. This finding underscores the effects of growing conditions that manifest as food product quality traits long after the crop has been harvested.

1.2.3 Dough expansion studies

Hankoczy (1920) and Chopin (1921) used a dough inflation to determine wheat gluten and bread flour dough extensibility. The concept has evolved over the years to

determine fracture and biaxial extensibility of wheat doughs, and to evaluate quality of wheat flour dough and gluten (Dobraszczyk and Roberts, 1994; Dobraszczyk, 1997).

The use of vacuum for dough expansion originated from work on gluten by Bungenberg de Jong (1956). Subsequently, MacRitchie (1976) used low pressure to expand dough pieces and correlated the expanded volumes to baked loaf volume. He also found that the expanded volumes correlated better than alveograms to bread loaf volume. Bell et al. (1981) used the concept of vacuum expansion on mechanically developed doughs at proof temperature to test whether the improved bread volume due to shortening were a result of temperature in oven. In the study by Bell et al. (1981), effect of shortening, of a liquid triglyceride, of flour type (weak, medium and strong) and condition both on dough expansion at proof temperature and on bread volume was studied. Dough expansion was carried out in glass tubes (about 25 cm X 5 cm ID) placed in a water bath at $38 \pm 1^\circ \text{C}$. The rate of pressure decrease (from approximately 29 In Hg to 2 In Hg) was controlled and slow (approximate time was 25 minutes) as compared to previous studies. Fully expanded dough heights increased with increasing dough strength. Additionally, the addition of shortening increased the mean height of expanded doughs.

In mixing dough, flour is mixed with water (an ingredient in bread baking). Gas bubbles are created during the mixing step of bread-making. Stability of the bubble wall against premature failure during fermentation and proofing is important for final bread volume and crumb structure (Dobraszczyk et al., 2000). An expanding gas cell during proof and baking is characterized by rheological conditions of biaxial extension, large strain and low strain rate. Tests which have the ability to simulate these conditions may

be appropriate to study rheological properties of dough (Dobraszczyk et al., 2003). The Stable Micro Systems dough inflation system was used by Dobraszczyk (1997) and Dobraszczyk et al. (2003) to study extensional rheology and stability of gas cells in relation to bread making performance. A good correlation was obtained between strain hardening measured at 50°C and baking volume (Dobraszczyk et al., 2003). Strain hardening is related to stability of gas cells and their ability to expand further. A high value of strain hardening index (n) corresponds to a high baking performance. Strain hardening capacity can be measured by extensibility tests. Kindelspire et al. (2015) developed a model to identify extensibility parameters contributing to strain hardening index. Three extensibility parameters, namely, dough strength (R_{max}), extensional delay, and initial slope of the curve (E_i) were identified as best indicators in a model contributing to n and correlating to baked loaf volume. Among the three extensibility parameters, the R_{max} had the largest contribution and was proportional to the dough strain hardening properties. The model was validated using 2 sample sets (19 genotypes, 12 environments). The strain hardening index was found to be a good indicator of baking performance.

Gandikota and MacRitchie (2005) developed an instrument for measuring expansion capacity of dough using a desiccator jar, vacuum pump and dough height tracker. They correlated expanded heights of dough developed from 9 hard and 10 soft wheat flours with corresponding height of baked loafs ($r=0.99$). The instrument was used to determine the effects of bromate addition and lipid removal from flour during processing. The longest step in the process was mixing. Vacuum expansion of dough

required less than 2 minutes. The simplicity of this process could be used to develop a semi-automatic instrument capable of predicting bread loaf volume (MacRitchie, 2014). The experiment established a proof of concept for determining baking potential of wheat flour by measuring dough expansion however the method was not validated by using a large diverse set of samples.

1.2.3.1 The Alveograph

Another device that measures dough strength by expansion is the Alveograph. Unlike the VDES, the Alveograph uses positive air pressure. It measures the force required to blow and break a dough bubble. A dough is first formed using 250 grams of flour and 5 circular disks of 4.5 cm diameter are cut. Each dough piece is individually expanded by blowing air; after resting at 25°C for 20 minutes. Results include P value, L value, W value. The P value is obtained in millimeters (mm) and is the force required to blow the bubble of dough. The L value is also obtained in millimeters (mm) and represents extensibility of dough. The W value represents area under the curve and is expressed in Joules. The results of the test are used commonly by flour millers and processors to maintain uniformity in products and processes (AACC method 54-30.02). Contrasting results were obtained during early attempts to correlate Alveograph parameters to loaf volume. Khattak et al. (1974) reported low and non-significant correlations between Alveograph parameters and loaf volume in his study on hard red spring (HRS). Weipert (1981) found that P and W values were less reliable predictors of baking potential of German flours. Chen and D' Appolonia (1985) reported a negative correlation of the P value with flour protein, wet gluten and loaf volume. Pomeranz et al. (1989) developed Alveograph algorithms using combinations of protein, hardness and

Alveograph values P, L and W to predict loaf volumes and specific volumes of bread. L and W values of Alveograph plus protein contributed in predicting loaf volume in soft wheats ($r=0.91$). For hard wheats, Alveograph L and protein content predicted loaf volume ($r= 0.95$). Bettge et al. (1988) utilized the Alveograph to evaluate baking potential of soft and hard wheat flours from Pacific Northwest. L was found to have a maximum correlation with loaf volume and specific loaf volume in both wheat flour types.

A Vacuum Dough Expansion System (VDES) developed by Padmanaban Krishnan, a Cereal Chemist at SDSU, was utilized by a master's student, Brijesh Merawat for his thesis on predicting loaf volume of bread using VDES. The VDES measured cross sectional area of raised dough brought by vacuum expansion which was then compared and correlated to true volume of baked dough. The dough expanded in the VDES was developed using the two basic ingredients, flour and water. The study was performed on 65 samples of HRS and HRW wheat grown in South Dakota in 2012. The aim of the study was to evaluate effectiveness of the VDES in predicting loaf volume of bread by vacuum expansion of dough. A correlation coefficient of 0.24 between vacuum expansion and loaf volume was obtained for HRS wheat whereas it was 0.15 for HRW wheat. The reason for the low correlation coefficients in the study performed by Merawat (2013) may be attributed to the small design of the VDES which allowed bread dough of 45 g to be expanded. Consequently 45 g pup loaves of bread were baked parallel for establishing a correlation. It is hypothesized that the proper development of the bread structure might not have been possible at such small scale which might be responsible for

the results. Therefore, it might be worthwhile to utilize a bigger version of the VDES which can expand dough made from 100 g of flour. Correspondingly, bread was baked from 100 g flour in this study as recommended by official methods prescribed by American Association of Cereal Chemists International (AACCI). A larger version of the VDES was used in the current study to bridge the gap found in the last study performed by Merawat (2013).

The VDES and the hybrid SDS-SRC Sedimentation test may have a potential to predict bread-making performance of wheat flours independently. Additionally, the power of prediction of bread loaf volume may be increased by utilizing dough expansion attribute (from VDES) and output of the hybrid SDS-SRC Sedimentation test in a multiple regression equation. This study investigated the two methods with the following objectives:

1.3 Objectives of the Study

1. To develop technique for dough expansion utilizing the Vacuum Dough Expansion System (VDES) and study the applicability of the VDES in measuring bread baking quality of wheat flours.
2. To study the applicability of the VDES for testing the effects of vital wheat gluten on dough expansion.
3. To study the relationship between the routine flour and dough tests used for flour end use quality, the hybrid SDS-SRC Sedimentation test, the VDES data and the baked loaf characteristics.
4. To combine the output of rheological test (VDES) with output of chemical test (hybrid SDS-SRC Sedimentation test) in a regression equation and to measure its effectiveness in predicting bread loaf volume

1.4 Hypotheses

1. **H₀**: There is no statistically significant correlation between dough expansion attributes and corresponding baked loaf volume. The height of dough expanding in a VDES is not statistically significantly related to bread loaf volume from actual baking test.

H₁: There is a statistically significant relationship between dough expansion attribute and corresponding baked loaf volume.

2. **H₀**: There is no effect of vital wheat gluten on dough expansion attribute. The effect of vital wheat gluten on dough expansion height is not incremental and linear as evaluated by VDES

H₁: The effect of vital wheat gluten addition on dough expansion height is incremental and linear as evaluated by the VDES.

3. **H₀**: There is no statistically significant correlation between the output of hybrid SDS-SRC Sedimentation test and baked loaf volume.

H₁: There is a statistically significant correlation between the output of hybrid SDS-SRC Sedimentation test and bread loaf volume.

4. **H₀**: Multiple regression equation containing dough expansion height and weight value from the hybrid SDS-SRC test doesn't explain the variation in baked loaf volume significantly.

H₁: Multiple regression equation containing dough expansion height and weight value from the hybrid SDS-SRC test output explains the variation in baked loaf volume significantly.

CHAPTER 2. MATERIALS AND METHODS

Figure 2.1 provides a flowchart of the experimental procedure used in this study. Preliminary investigation on developing a method of dough expansion and studying the response of different flour types to dough expansion was done on commercially acquired all-purpose flour, bread flour and pastry flour. For further investigation, a total of 147 wheat samples representing the two major economical classes of wheat (namely HRS and HRW), were used. The grain protein content of the wheat samples was determined utilizing the FOSS NIRS 6500 device (FOSS, Hilleroed, Denmark). The wheat samples were milled to obtain the flour. Flour tests, dough tests and baking test were performed on each flour sample. The flour tests included moisture analysis, gluten analysis, protein analysis and the hybrid SDS-SRC sedimentation test. The dough tests included Mixograph analysis (only on HRS wheat samples), Mixolab analysis (only on winter wheat samples), Farinograph analysis (only on HRS wheat samples) and the vacuum dough expansion test. Finally, baking tests were also conducted. The bread loaf volume was determined using mustard seed displacement method and the C-Cell instrument was used to perform bread crumb structure analysis. The detailed description of the materials and methods is presented in the following sections.

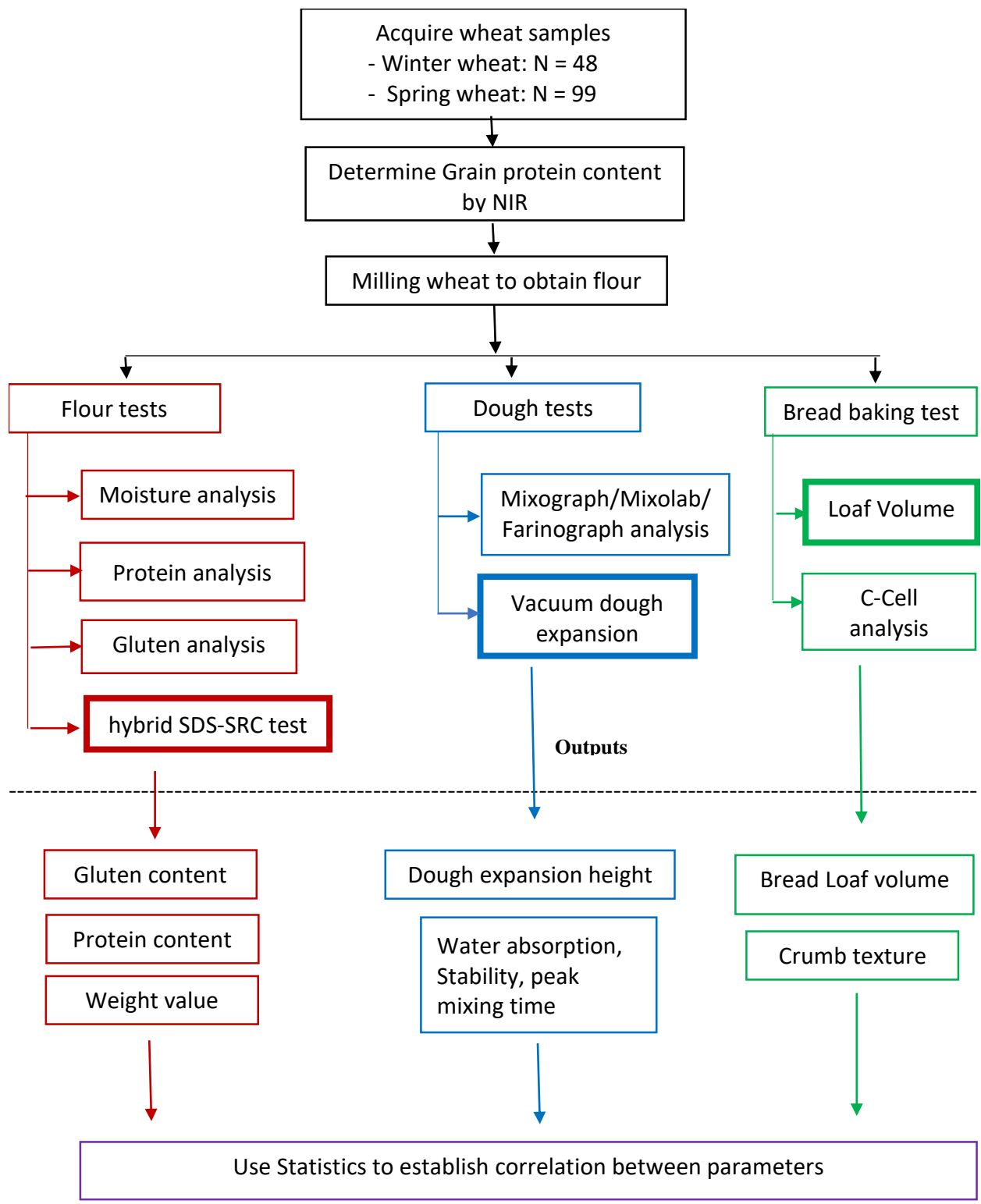


Figure 2.1 Flow chart of the experimental design used in the research

2.1 Sample selection

Bulk All Purpose Flour (APF) and Bread Flour (BF) marketed by Ardent mills was procured from Costco, Sioux Falls. These bulk flours were used primarily for preliminary investigations.

2.1.1 Preparation of flour blends from pastry flour

The response of various flour types to dough expansion was evaluated using the VDES. Since weak flours and strong flours are defined by their gluten content or gluten strength, a weak pastry flour was chosen for vital gluten fortification at several levels. Pastry flour, a weak flour, was acquired from Ardent Mills. Vital Wheat Gluten (VWG) was enriched to the pastry flours in varying levels to form flour blends with final gluten concentration ranging from 7.3 to 12.1 % (Table 2.1)

Table 2.1 Description of the control pastry flour and the gluten concentration of the blends prepared by incorporating Vital Wheat gluten in the pastry flour

Sample description	Vital Gluten percentage (14 % mb)
Control pastry flour	7.30
Blend 1	8.02
Blend 2	9.06
Blend 3	10.27
Blend 4	12.10

Gluten analysis of the blends was done using the Glutomatic system. Optimally developed dough from the flour blends was expanded in the VDES. Baking experiments were performed, and bread loaf volume was determined. C-Cell instrument was used to determine bread crumb texture analysis. The repeatability of the findings was further

investigated on flour blends with final gluten concentration ranging from 7.3 to 14.3 % (Table 2.2).

Table 2.2 Description of the control pastry flour and the gluten concentration of the blends prepared by incorporating Vital Wheat gluten in the pastry flour

Sample description	Gluten percentage (14 % mb)
Control pastry flour	7.30
Blend 1	9.06
Blend 2	10.5
Blend 3	12.10
Blend 4	12.27
Blend 5	14.73

For further investigation, a total number of 147 pure varieties of HRW and HRS wheat were acquired from the SDSU wheat breeding programs.

Table 2.3 summarizes the number of varieties and the growing location of each class of the wheat samples. For HRS wheat, 33 cultivars, each from 3 different growing locations, were procured. The growing locations were Brookings, Selby and Groton. These varieties had advanced to Advanced Yield Trials (AYT) for the 2016 Spring Wheat breeding program. For HRW wheat, 24 varieties, each from 2 different growing locations were included in the sample set. The growing locations were Aurora and Onida. These varieties had advanced to Crop Performance Trials (CPT) in the 2016 winter wheat breeding program. The HRW wheat samples were milled in the SDSU Seedhouse before processing further. The two classes of wheat were chosen as they are ideal bread baking flours.

Table 2.3 Classes of wheat with the corresponding growing locations

Wheat Class	Locations	Samples
Spring	Brookings	33
	Selby	33
	Groton	33
HRW	Aurora	24
	Onida	24

2.1.2 Cleaning, tempering and milling of wheat

The HRW wheat samples were cleaned manually by removing the visible contaminants such as stones, weeds, insect parts, husks and grains other than wheat. Diseased, damaged, shrunken and broken wheat unfit for milling were also removed. The cleaned grains were stored in mason jars for tempering. The moisture percentage of the cleaned grains was found by NIRS and amount of water to be added to attain 16 % target moisture level was calculated. The water was added sequentially, and the mason jars were shaken periodically to distribute water uniformly. The Quadrumat Junior Mill (Brabender Instruments, Inc South Hackensack, NJ) was used to mill the tempered wheat. The flour was stored in airtight Ziploc bags.

2.2 Analysis of flour and dough

2.2.1 Flour tests

2.2.1.1 Moisture analysis

Two to three grams of the flour sample was kept in forced-air convection Fisher Isotemp™ oven (Fisher Scientific, Pittsburgh, PA) for 1 hour at 130°C. Moisture content was determined by the loss of water as per AACCI method 44-15-A

2.2.1.2 *Gluten analysis*

The Glutomatic system (Perten Instruments, Waltham, MA) was used and the official AACCI method 38-12.02 was followed. Ten-gram flour sample was weighed. The flour was mixed with water to form a dough. The dough was washed with 2% NaCl solution to remove starch and to recover gluten.



Figure 2.2 The Glutomatic system

Figure 2.3 shows the steps of gluten analysis employing the Glutomatic system. Ten-gram flour sample was weighed. The sieve of the washing chamber was moistened to prevent flour loss. The measured flour was placed in the washing chamber which had a 88 μm polyester sieve. A 3.5 ml of 2% NaCl solution was added to the flour sample. The chamber was shaken gently to disperse the solution uniformly. The washing chamber was attached to the Glutomatic system below the plexiglass attachment. The instrument was turned on. The flour was mixed with the solution for twenty seconds to form a small dough ball. After termination of the mixing, washing started automatically and continued for five minutes. This removed starch from the dough leaving behind a gluten mass. The wet gluten piece was transferred to the centrifuge. Centrifugation was carried out at 6000 rpm. The portion of the gluten that passed through the sieve was scraped and weighed. The remaining portion was referred to as good wet gluten (GWG) in this study. The

GWG was collected and added to the balance to obtain total wet gluten weight. The wet gluten was dried on Glutork 2020 for four minutes and the weight of the dried gluten wafer was weighed. The dried gluten content was converted to gluten content equivalent in flour on a 14 % moisture basis.

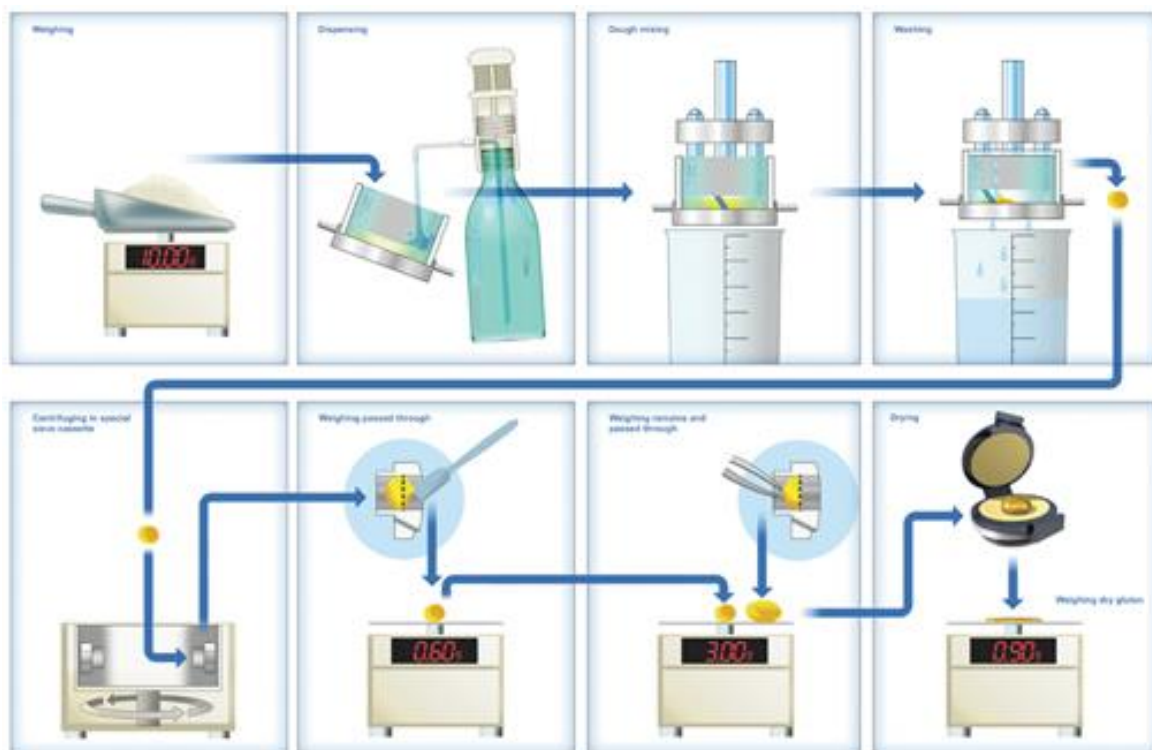


Figure 2.3 Diagram depicting the process of gluten analysis on the Glutomatic system

2.2.1.3 Protein determination

Protein content was determined as per AACCI method 46-30.01 employing the N/protein analyzer Flash EA 112 (CE Elantech, Lakewood, NJ). The analyzer combusted the sample (as per the modified Dumas method) of known mass at high temperature (900 °C) in presence of oxygen leading to release of CO₂, N₂ and H₂O. The instrument estimated Nitrogen content that was converted to protein by multiplying with a fudge factor. A factor of 5.7 was used for wheat flour samples.



Figure 2.4 Flash EA 1112 N/Protein analyzer

2.2.1.4 The hybrid SDS-SRC method

The protocol for the hybrid SDS-SRC method was developed by Seabourn et al. (2012). The method combines use of solutions used in the AACC Sodium dodecyl sulfate (SDS) sedimentation test with the centrifugation process found within the AACC Solvent retention capacity (SRC) method.

One-gram flour sample was weighed and placed in pre-weighted 50 ml polypropylene tube. Five ml of 0.47 % lactic acid was added to the tube. The tube was capped and shaken on a small vibratory mixer for six seconds. Next, 20 ml of 1.25 % SDS solution was added. The tube was capped and mixed again for six seconds. The tube was shaken on a platform shaker (MaxQ 4450, Thermo Scientific, Marietta, OH) at 300 rpm for four minutes. Next, the tubes were centrifuged at 3200 x g in a swinging bucket rotor (Eppendorf 5810R, Brinkmann Instrument Inc., Woburn, NY) for two minutes. After centrifugation, the supernatant was separated by decanting making sure not to disturb the precipitate. A paper towel was used to wipe away any visible foam inside the tube. The tube was capped and weighted to determine the sedimentation weight value as per the formula below. The weight value was used as the outcome of the hybrid SDS-

SRC sedimentation assay to compute correlations with parameters obtained from other flour and dough quality tests.

Weight value (%) = ((pellet weight/flour weight) × {[86/(100 – percent flour moisture)] – 1}) × 100.

2.2.2 Dough tests

2.2.2.1 Mixolab

The Chopin Mixolab (Chopin Technologies, Villeneuve-la-Garenne, France) was used to study dough mixing properties as per the AACCI method 54-60.01. Mixolab plots the torque (expressed in Nm), in real time, produced by dough between the two kneading arms when subjected to shear and temperature. Figure 2.6 shows the Chopin + protocol that was used.



Figure 2.5 The Chopin Mixolab

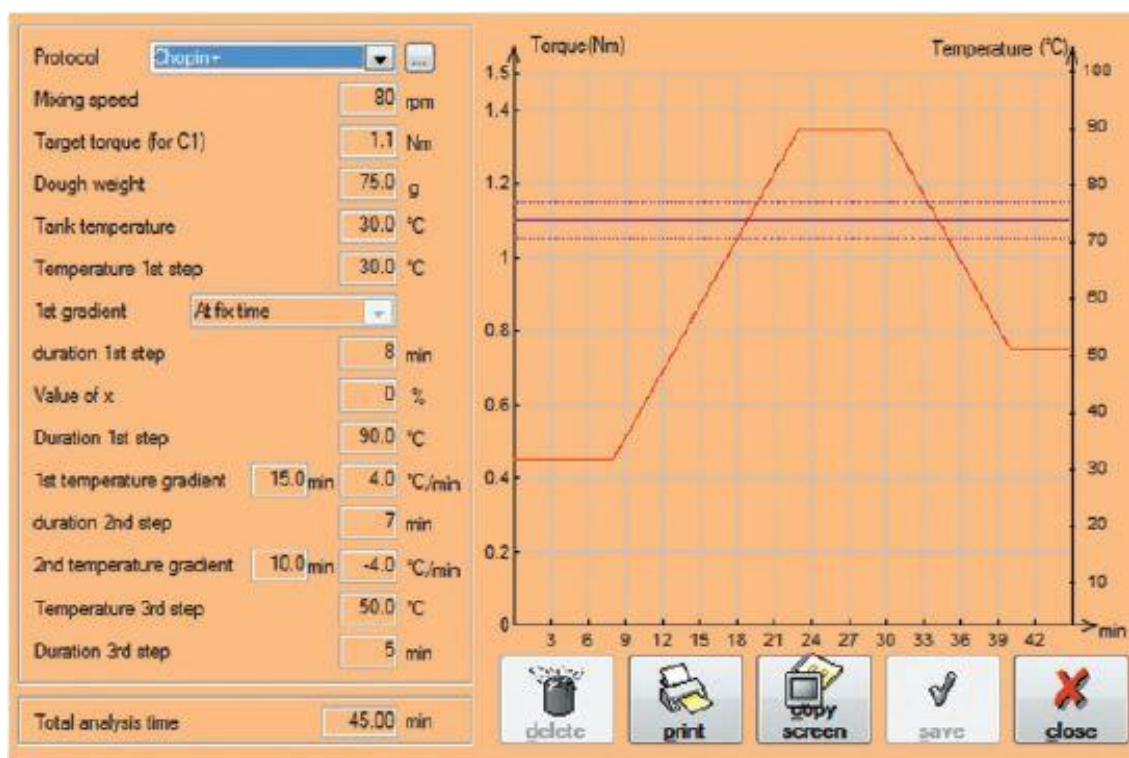


Figure 2.6 Mixing settings in Chopin+ protocol (Mixolab Handbook, 2012)

Parameters of interest obtained from the curve were the water absorption (%) or the percentage of water required for the dough to produce a torque of 1.1 ± 0.05 Nm; dough development time (min) or the time to reach the maximum torque at 30° C; and the dough stability (min).

2.2.2.2 The vacuum dough expansion system (VDES)

A VDES system was designed to expand well developed dough inside a vacuum chamber. The VDES comprised of a vacuum chamber made of Aluminum, a camera, a vacuum pump, a relay box for relay control of the pump and a computer system with a software. The vacuum chamber was equipped with pressure sensor and a temperature

sensor. The software handled the data acquisition, data processing and transporting results to excel spreadsheet.

The new vacuum dough expansion chamber was designed to work between 30 In Abs Hg (0% vacuum, 14.7 PSI) to 0.8 In Abs Hg (97.3% vacuum, 0.39 PSI). The new system was a bigger version of the original VDES and had the capacity to expand larger volumes of dough. Two modes of operation were researched for method development of dough expansion.

2.2.2.2.1 Automated mode

The expansion of the dough under vacuum was monitored and measured by a digital camera. This utilized the VDES camera and the software. Dough was expanded in 5-5/8" x 3-1/8" open top bread pan. Dough mass ranging from 150g to 200 g were expanded. The camera took images at stipulated pressures as the dough rose in the chamber. The pressures were 10, 1 and 0.8 In Hg. The software digitized the cross-sectional area of the expanding dough in pixels.

2.2.2.2.2 Manual mode

Expansion of 50-75 g dough was carried out in a cylindrical geometry (400 ml Kimax Kimble beaker). The pressure decreased from 29 In Hg to 0.8 In Hg. The time needed for the pressure drop was between 79-87 seconds. The maximum expansion of the dough was recorded as dough height in centimeters (cm).

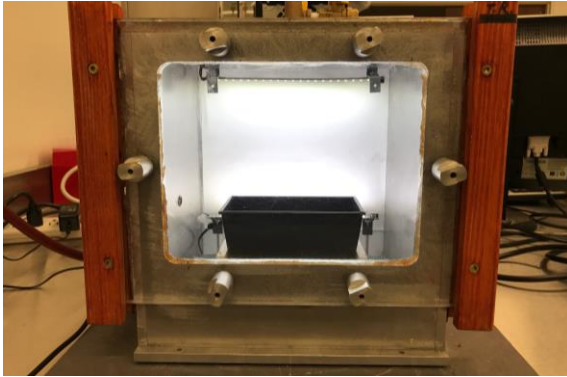


Figure 2.7 The Vacuum Dough Expansion System

2.2.2.2.3 *Mixing of dough for Vacuum expansion: Utilizing the Swanson 200 g mixer*

The mixing of flour and water to form a dough for vacuum expansion was carried out in a 200g Swanson mixer (National Mfg. Co., Lincoln, NE). This was done to remove human subjectivity and bias for determining optimum dough consistency. The amount of water to be added was determined by the Mixolab. The mixing time was determined from the real time mixing chart (Mixing was stopped after the mixing peak began to drop). Dough prepared by the Swanson mixer was split into desired mass and rested in a proofing cabinet at 87° F for 10 minutes. Dough was lightly greased to prevent moisture loss and drying.



Figure 2.8 The 200g Swanson mixer

2.2.2.2.4 Vacuum dough expansion

2.2.2.2.4.1 Dough expansion in a bread pan: Automation mode of the VDES

The desired dough mass was sheeted, molded and placed in a bread pan. The pan with the dough was rested in proofer at 87° F for ten minutes prior to transferring to vacuum chamber for dough expansion. As described earlier, the automated mode of the VDES was employed for dough expanding in bread pan.

2.2.2.2.4.2 Dough expansion in a beaker: Manual mode of the VDES

The desired dough masses (50 – 75 g) were split and rested in proofer at 87° F for ten minutes. The dough piece was then transferred to a 400 ml beaker with lightly greased walls. The dough was flattened on the bottom of the beaker with a kitchen silicone spatula. The manual mode of the VDES was employed to expand the dough and to record expanded dough height.

2.2.3 Bread baking

A modified official AACCI Basic straight-dough bread-making method 10-09 was used for making bread with 100g flour. The formula of bread was as follows:

Table 2.4 Formula used in the modified Basic Straight-Dough Bread-making Method

Ingredients	Flour basis (%)
Flour, 14% moisture basis	100
Salt, chemically pure NaCl	2
Yeast, compressed (active or instant dry)	3
Water	Variable
Sugar	5
Shortening	3

Flour, salt and shortening was dry mixed in 100g pin mixer (National MFG IN., Lincoln, NE.). The dry yeast was activated by adding it in warm water (amount required for mixing) with sugar (5g). The activated yeast-sugar solution was added to the ingredients in the pin mixer. The dough consistency was checked at 3 minutes and the dough was mixed till optimum consistency was obtained. The dough was then transferred to lightly greased fermentation bowls and covered with plastic wrap. Dough was fermented for 55 minutes in proofing cabinet (National MFG IN., Lincoln, NE.) at 87 F and 85% relative humidity. A single punch was performed. The dough was punched by passing the dough through sheeter utilizing 3-in. roll width and a spacing of 5/16 inches. The dough was then rolled and molded using a molder (National MFG IN., Lincoln, NE.). The dough was then placed in a lightly greased bread pan (5-5/8" x 3-1/8"), seam down. The dough was then proofed for another 55 minutes in the proofer. Baking was performed in electric rotary oven (National MFG IN., Lincoln, NE.) set at 420 F. The

bake time was 25 minutes. The baked loaves were cooled for an hour and the weight and loaf-volume was measured.

2.2.3.1 Bread loaf volume measurement

AACCI method 10-05.01 was used to measure the volume of the bread loaves. Prior to loaf volume measurement, the loaves were cooled for at least one hour. The loaf volume was measured using the mustard seed displacement method. The density of the mustard seeds was determined prior to the test. A 3 Quart capacity stainless steel bowl was used as the measuring container. The bowl was filled with mustard seeds till the top. The quantity of the mustard seeds required to fill the container was separated and used as the stock each time. Next, bread loaf was placed in the 3-Quart bowl, mustard seeds from the stock were then added till the bowl was filled. The stock of mustard seeds displaced by the bread loaf was weighed; volume was calculated from the known density of the mustard seeds.

2.2.3.2 C-Cell analysis

The texture of a bread slice was analyzed using C-Cell (Calibre Control Intl. Ltd., Warrington, UK). Objective information on the cell size, number of cells, cell wall thickness, uniformity and the overall shape of the bread slices was obtained.



Figure 2.9 The C-Cell instrument

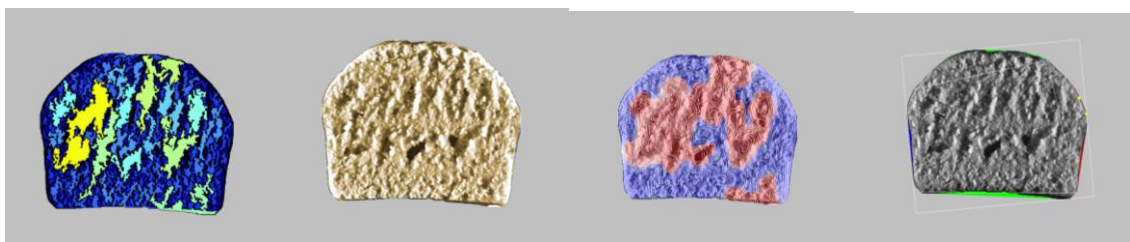


Figure 2.10 Image analysis of bread slice by C-Cell

2.3 Statistical analysis

For each sample, the data was collected for grain quality (grain protein), flour quality (flour protein, gluten content, weight value which was derived from the pellet weight of the hybrid SDS-SRC sedimentation test), dough quality (Mixograph parameters, Farinograph or Mixolab parameters, dough expansion height) and baked loaf characteristics (loaf volume).

Statistical analysis included computing bivariate correlations between the various quality parameters utilizing the SPSS software. Subsequently, multiple regression

equations were also developed to predict loaf volume from the various independent variables.

Variance components were estimated using the `lmm.jack` function in R Package ‘minque’ developed by Dr. Jixiang Wu of South Dakota State University (<https://cran.r-project.org/web/packages/minque/minque.pdf>), with genotypes (breeding lines and cultivars) and environments (sites within years) considered as random effects. Each variance component was reported as a proportion of total variance. `lmm.jack` is an R function for linear mixed model analysis with integration two linear mixed model approaches (Restricted maximum likelihood and MINQUE) and a jackknife technique.

CHAPTER 3. RESULTS AND DISCUSSIONS

In this section, first the results of mean and the range of occurrence of wheat quality traits in the 24 cultivars of HRW wheat and the 33 cultivars of HRS wheat are summarized and discussed. The effect of genotype and environment on the variability of different quality traits is discussed for both the HRS and HRW wheat cultivars.

Secondly, the results of the preliminary investigation to generate a proof of concept for the VDES is shared and discussed. The preliminary investigation on the VDES was performed on pastry flour and blends of pastry flour enriched with vital wheat gluten.

Thirdly, the results and discussions pertaining to best predictors of bread loaf volume for the HRW wheat cultivars are presented. The section starts with a general overview of the best predictor variables for bread loaf volume in HRW wheat. Then, it focuses on the results and discussions for the VDES and the hybrid SDS-SRC sedimentation method. This includes results and discussions on the correlation of each of the method (VDES and hybrid SDS-SRC sedimentation) with other quality traits. Next, the predictor variables other than the VDES and the weight value from the hybrid SDS-SRC sedimentation method are discussed. Finally, the results and discussion on developing multiple regression equation for predicting bread loaf volume is discussed for the HRW wheat cultivars.

Finally, the results and discussions for the HRS wheat cultivars are presented. The sub sections for the HRS wheat follow the same pattern as for the HRW wheat cultivars section.

3.1 Mean values and range of occurrence of wheat quality traits in 24 cultivars of HRW wheat and 33 cultivars of HRS wheat.

Tables 3.1 and 3.2 provide information of the protein content and protein functional qualities that describe the behavior of dough in mixing stages for HRW and HRS cultivars, respectively. The hybrid SDS-SRC sedimentation test is a chemical test that provides information on the water holding capacity of gluten proteins and potential for bread baking. VDES data yield information on dough expansion of doughs that were mixed to optimal conditions prescribed by rheological tests such as the Farinograph or Mixolab. Bread characteristics data provide information of actual bread baking trials for each of the flour samples milled from the wheat grain.

At the Aurora growing location of HRW cultivars, variations in grain protein content, flour protein content, gluten content, Mixolab absorption, weight value, loaf volume were 12.1 to 14.7 % (mean 13.3 %), 11.3 to 13.8 % (mean 12.5 %), 9.9 to 12.8 % (mean 11.1%), 53.7 to 57.2 % (mean 55.1 %), 267 to 379 (mean 330), and 616 to 937 cc (mean 795 cc), respectively. At Onida, the range of grain protein content, flour protein content, dry gluten content, Mixolab absorption, weight value, loaf volume were 12.9 to 15.3 % (mean 14.1 %), 12.9 to 15.1 % (mean 13.7 %), 10.8 to 14.2 % (mean 12.4 %), 54.8 to 58.8 % (mean 56.4 %), 276 to 425 (mean 355), and 580 to 943 cc (mean 743 cc), respectively.

Table 3.1 Summary of quality parameters of the 24 cultivars of HRW wheat grown at Aurora and Onida growing locations

Aurora			Onida		
Quality parameter	Mean	Range	Quality parameter	Mean	Range
<u>Wheat characteristics</u>			<u>Wheat characteristics</u>		
Grain protein content, 14% mb	13.3 ^a	12.1-14.7	Grain protein content, 14% mb	14.1 ^b	12.9-15.3
Flour protein content, 14% mb	12.5 ^a	11.3-13.8	Flour protein content, 14% mb	13.7 ^b	12.9-15.1
<u>Gluten characteristics</u>			<u>Gluten characteristics</u>		
Dry gluten content, 14% mb	11.1 ^a	9.9-12.8	Dry gluten content, 14% mb	12.4 ^b	10.8-14.2
Wet gluten content	3.13 ^a	2.68-3.68	Wet gluten content	3.57 ^b	3.17-4.16
Good wet gluten content	3 ^a	2.66-3.54	Good wet gluten content	3.27 ^b	2.98-3.72
Water absorption capacity	2.02 ^a	1.61-2.40	Water absorption capacity	2.3 ^b	1.51-2.75
<u>Mixolab characteristics</u>			<u>Mixolab characteristics</u>		
Mixolab absorption, %	55.1 ^a	53.7-57.2	Mixolab absorption, %	56.4 ^b	54.8-58.8
Mixolab Stability, min	10 ^a	4.47-12.33	Mixolab Stability, min	10.4 ^a	8.17-12.31
Peak time, min	3.7 ^a	1.55-9.19	Peak time, min	7.5 ^b	1.72-9.85
<u>Hybrid SDS-SRC sedimentation method</u>			<u>Hybrid SDS-SRC sedimentation method</u>		
Weight value	330 ^a	267-379	Weight value	355 ^b	276-425
<u>VDES</u>			<u>VDES</u>		
Dough expansion height, cm	7.1 ^a	5.5-8.5	Dough expansion height, cm	7.8 ^b	6.13-9.25
<u>Bread characteristics</u>			<u>Bread characteristics</u>		
Loaf volume, cc	795 ^a	616-937	Loaf volume, cc	743 ^a	580-943
Specific loaf volume, cc/g	5.82 ^a	4.48-6.87	Specific loaf volume, cc/g	5.63 ^a	4.32-7.24

Grain protein, flour protein and dry gluten content are expressed on a 14% moisture basis.

Specific loaf volume was calculated by dividing loaf volume with bread weight

Mean values for each variable with differing variable within rows are significantly different across the growing locations (p = 0.05)

The range of grain protein content (12.1 to 15.3 %) of HRW cultivars suggests that the samples belonged in the high protein range. Seabourn et. al., (2012) conducted validation studies on a set of 53 HRW varieties with a range of grain protein from 10.5 to 16.8 %. In the HRW wheat cultivars, growing location was a significant factor and influenced the variability of the chemical composition of the wheat. The mean grain protein content of the varieties in Onida growing location was statistically significantly higher than in Aurora growing location. Similarly, the mean flour protein content of varieties in Onida growing location was statistically significantly higher. The other quality parameters such as gluten and Mixolab characteristics also followed similar trend as expected. However, the mean specific loaf volume was not statistically significantly higher in Onida growing location.

Table 3.2 summarizes the results of relative proportion of the variance component for parameters from routine flour and dough tests, the hybrid SDS-SRC sedimentation test and the dough expansion test in the HRW wheat cultivars. For most quality traits, only environment was the statistically significant source of variability. Caffè-Treml and coworkers (2011) reported that both genotype and environment were statistically significant source of variation for quality parameters such as protein, Mixograph parameters and loaf volume. In our study, the significant effect of growing location may have been highlighted further by the fact that Onida growing location was affected by drought in 2016. Both environment and genotype were statistically significant sources of variation for the Mixolab parameters. Environment represented the largest source of variation for flour protein content, gluten content, total wet gluten, Mixolab

Table 3.2 Estimate of the variance components of routine flour and dough tests used for flour end use quality, the hybrid SDS-SRC Sedimentation test, and the dough expansion test for 24 cultivars of HRW wheat grown at two growing locations

Parameters	Relative proportion (%) of variance components and their significance				
	Genotype	Sig	Location	Sig	Residual
GP	08		37	***	55
FP	01		57	***	42
FG	10		47	***	43
TWG	07		60	***	33
GWG	09		39	***	52
WAC	02		36	*	62
WV	00		12	*	88
MLA	33	**	42	***	25
MLPT	18	*	47	***	45
MLS	57	***	02		41
SLV	00		00		100
VDEH	00		15	*	85

FG- Flour Gluten; FP- Flour Protein; GP- Grain Protein; GWG- Good Wet Gluten; MLA- Mixolab Absorption; MLPT- Mixolab Peak Time; MLS- Mixolab Stability; SLV- Specific Loaf Volume; TWG- Total Wet Gluten; VDEH- Vacuum Dough Expansion Height from Vacuum Dough Expansion System (VDES); WAC- Water Absorption Capacity; WV- Weight Value from hybrid SDS-SRC Sedimentation test

***, ** and * indicate significance at P = 0.001, 0.01 and 0.05 respectively.

absorption and Mixolab peak time. Genotype represented the largest source of variation for only one variable, the Mixolab stability.

Table 3.3 summarizes the quality parameters of the 33 HRS wheat cultivars grown at three growing locations. Across the three growing locations, the range of grain protein content, flour protein content, gluten content, Farinograph absorption, and loaf volume were 12.5 to 15.9 % (mean 14.4%), 11.2 to 15.8 % (mean 13.6 %), 10.1 to 14.4 % (mean 12.9 %), 54.5 to 62.7 % (mean 62.5 %), and 150 to 214 cc (mean 190 cc), respectively. As with HRW cultivars, the cultivars in HRS were found to be in the high protein range. Additionally, location was a significant factor and statistically significantly affected the mean chemical composition of the varieties across the growing locations. The mean grain protein content and mean flour protein content of each growing location was statistically significantly different than the other two growing locations. Even though, the flour protein content was different in all three growing locations, the wet gluten content in Brookings and Selby growing locations were not statistically significantly different. This means that even if the protein quantity is similar in two varieties or growing locations, the protein quality in terms of gluten strength may be significantly different.

Table 3.4 shows the contribution of genotype, environment and residual to the variability in the parameters obtained from routine flour and dough tests, the hybrid SDS-SRC test and the dough expansion test in HRS cultivars. Unlike in the HRW cultivars, for the HRS wheat cultivars both genotype and environment were statistically significant sources of variation except for Farinograph water absorption. Environment represented

the dominant source of variation for grain protein content, flour protein content, gluten content, good wet gluten, weight value, Mixograph absorption, farinograph dough development time, Farinograph quality number, dough expansion height and loaf volume. Genotype represented the largest source of variation for total wet gluten, Mixograph mix time, Mixograph peak time and Farinograph water absorption. Genotype x environment interaction confounded with residual represented the main source of variation for Farinograph dough development time, Farinograph stability, and Farinograph mixing tolerance index.

Table 3.3 Summary of quality parameters of the 33 cultivars of HRS wheat grown at Brookings, Groton and Selby growing locations

Brookings			Groton			Selby		
Quality parameter	Mean	Range	Quality parameter	Mean	Range	Quality parameter	Mean	Range
<u>Wheat characteristics</u>			<u>Wheat characteristics</u>			<u>Wheat characteristics</u>		
Grain protein content, %	14.3 ^a	12.5-15.2	Grain protein content, %	14.7 ^b	13.6-15.4	Grain protein content, %	15.7 ^c	13.4-15.9
Flour protein content, %	12.9 ^a	11.2-13.7	Flour protein content, %	13.4 ^b	12-14.4	Flour protein content, %	14.5 ^c	12.3-15.8
<u>Gluten characteristics</u>			<u>Gluten characteristics</u>			<u>Gluten characteristics</u>		
Dry gluten content, %	12 ^a	10.1-13.0	Dry gluten content, %	13.9 ^b	11.4-14.4	Dry gluten content, %	12.7 ^c	10.9-13.6
Wet gluten content, g	3.45 ^a	2.84-3.77	Wet gluten content, g	3.75 ^b	3.06-4.2	Wet gluten content, g	3.5 ^a	3.05-3.79
Good wet gluten content, g	3.23 ^a	2.48-3.53	Good wet gluten content, g	3.61 ^b	3-4.03	Good wet gluten content, g	3.27 ^a	2.82-3.59
<u>Mixograph characteristics</u>			<u>Mixograph characteristics</u>			<u>Mixograph characteristics</u>		
Absorption, %	59.7 ^a	56.9-62.1	Absorption, %	57.7 ^b	55.3-62.7	Absorption, %	56.3 ^c	54.5-58.5
Mix time, min	3.7 ^a	2-5.8	Mix time, min	3.2 ^b	1.8-4.5	Mix time, min	2.8 ^c	1.5-4.5
Peak time, min	7.9 ^a	4.1-14.4	Peak time, min	6.3 ^b	3.1-9.4	Peak time, min	5.7 ^c	3.2-9.1
<u>Farinograph characteristics</u>			<u>Farinograph characteristics</u>			<u>Farinograph characteristics</u>		
Absorption, %	62.2 ^a	58.2-66.4	Absorption, %	62.5 ^{ab}	59-65.9	Absorption, %	62.8 ^c	58.6-68.4
Dough development time, min	10.8 ^a	2.1-16.3	Dough development time, min	14.9 ^b	6- 24	dough development time, min	16.8 ^c	6.6-29.2
Stability, min	13.4 ^a	5.9-21.9	Stability, min	16.8 ^b	7.5-29	Stability, min	19.2 ^c	3.5-30.5
Mixing tolerance index, BU	22 ^a	6- 59	Mixing tolerance index, BU	20 ^a	9- 43	Mixing tolerance index, BU	12 ^c	1- 33
Farinograph quality number, mm	184 ^a	98-259	Farinograph quality number, mm	265 ^b	110-386	Farinograph quality number, mm	290 ^c	93-427
<u>Hybrid SDS-SRC sedimentation method</u>			<u>Hybrid SDS-SRC sedimentation method</u>			<u>Hybrid SDS-SRC sedimentation method</u>		
Weight value	365 ^a	282-407	Weight value	398 ^b	342-454	Weight value	472 ^c	388-536
<u>VDES</u>			<u>VDES</u>			<u>VDES</u>		
Dough expansion height, cm	8.3 ^a	6.8-9.5	Dough expansion height, cm	9.9 ^b	8.6-11.4	Dough expansion height, cm	9.5 ^c	8.1-11
<u>Bread characteristics</u>			<u>Bread characteristics</u>			<u>Bread characteristics</u>		
Loaf volume, cc	180 ^a	150-203	Loaf volume, cc	202 ^b	181-214	Loaf volume, cc	187 ^c	172-199

Grain protein, flour protein and dry gluten content are expressed on a 14% moisture basis. Mean values for each variable with differing variable within rows are significantly different across the growing locations (p = 0.05)

Table 3.4 Estimate of the variance components of routine flour and dough tests used for flour end use quality, the hybrid SDS-SRC Sedimentation test, and the dough expansion test for 33 cultivars of HRS wheat grown at three growing locations

Parameters	Relative proportion (%) of variance components and their significance				
	Genotype	Sig	Location	Sig	Residual
GP	27.8	***	59.3	***	12.9
FP	24.6	***	58.8	***	16.6
FG	22.1	***	57.4	***	20.5
TWG	41.4	***	29.3	***	29.3
GWG	28.3	***	48.1	***	23.6
WV	13	***	78	***	9
MGM	48.4	***	25.1	***	26.5
MGA	13.6	***	63.2	***	23.2
MGPT	46.1	***	36.2	***	17.7
FDDT	24.3	***	32.8	***	42.9
FWA	78.2	***	1.5		20.3
FSTAB	34.3	***	23.3	***	42.4
FMTI	30.3	***	20.5	***	49.2
FQN	29.1	***	44.9	***	26
VDEH	12	**	52.3	***	35.7
LV	14.3	***	59.4	***	26.3

FDDT- Farinograph Dough Development Time; FG- Flour Gluten; FMTI- Farinograph Mixing Tolerance Index; FP- Flour Protein; FQN- Farinograph Quality Number; FWA- Farinograph Water Absorption; FSTAB- Farinograph Stability; GP- Grain Protein; GWG- Good Wet Gluten; MGA- Mixograph absorption; MGM- Mixograph Mix time; MGPT- Mixograph Peak time; TWG- Total Wet Gluten; VDEH- Dough Expansion height from Vacuum Dough Expansion System (VDES); WV- Weight Value from hybrid SDS-SRC Sedimentation test

***, ** and * indicate significance at P = 0.001, 0.01 and 0.05, respectively.

3.2 Preliminary investigation on the Vacuum Dough Expansion System

3.2.1 Dough expansion experiments utilizing flour blends made from pastry flour:

Experiment 1

The response of various flour types to dough expansion was evaluated using the VDES. Since weak flours and strong flours are defined by their gluten content or gluten strength, a weak pastry flour was chosen for vital gluten fortification. Vital wheat Gluten (VWG) was added to the pastry flour in varying levels to form flour blends with a final gluten concentration ranging from 7.3 to 12.1 % (Table 2.1). The total number of samples were 5 including the control pastry flour.

Table 3.5 provides results of bivariate correlations between parameters of flour test, dough expansion height, bread loaf volume and C-Cell parameters. Figure 3.1 shows the relationship between flour gluten content and bread loaf volume. Statistically significant high correlation was observed between gluten content and loaf volume ($r = 0.94$). Figure 3.2 shows the pictures of the bread baked from the control pastry flour and the different blends of pastry flour enriched with vital wheat gluten.

Figure 3.3 shows the relationship between gluten content in flour and dough expansion height. A statistically significant and high correlation was observed between gluten content and dough expansion height ($r = 0.98$). The correlation coefficient (r) between good wet gluten and dough expansion height was found to be 0.99. The data provided evidence that the dough expansion height responded linearly with concentration of vital wheat gluten in the bread flour. These results provided evidence that the vacuum

dough expansion system may have practical applications for evaluating dough response to bread ingredients.

Table 3.5 Bivariate correlation coefficients (r) between variables of gluten content, dough expansion, baked loaf volume and C-Cell for control pastry flour and blends of pastry flour with increasing vital wheat gluten concentration. N=5

	Gluten	TWG	GWG	LV	VDEH	SA	AC	WT	CD	CA
Gluten	1.00									
TWG	0.96**	1.00								
GWG	0.98**	0.92*	1.00							
LV	0.94*	0.93*	0.93*	1.00						
VDEH	0.98**	.93*	0.99**	0.94*	1.00					
SA	0.94*	.93*	0.95*	0.97**	0.97**	1.00				
AC	0.74	0.82	0.74	0.86	0.78	0.90*	1.00			
WT	0.13	-0.09	0.18	-0.06	0.11	-0.10	-0.52	1.00		
CD	0.77	0.61	0.79	0.61	0.74	0.58	0.18	0.73	1.00	
CA	-0.85	-0.71	0.88**	-0.76	-0.85	-0.73	-0.36	-0.60	-0.97**	1.00

AC- Air cells; CA- Cells per unit area; CD- Cell Diameter; GWG- Good wet gluten; LV- Loaf volume; SA- Slice area; TWG- Total wet gluten; VDEH- Vacuum Dough Expansion Height; WT- Wall thickness

** and * indicate significance at P = 0.01 and 0.05, respectively

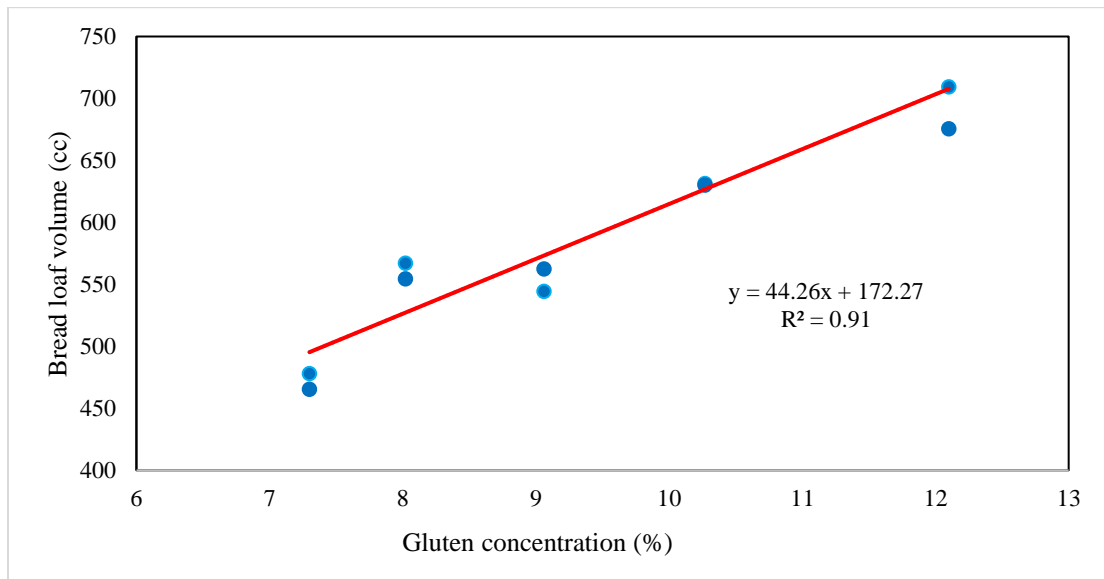


Figure 3.1 Relationship between gluten concentration and bread loaf volume. The gluten concentration is expressed on 14 % moisture basis.



Figure 3.2 Bread baked from control weak flour (Pastry flour) and blends of pastry flour enriched with vital wheat gluten

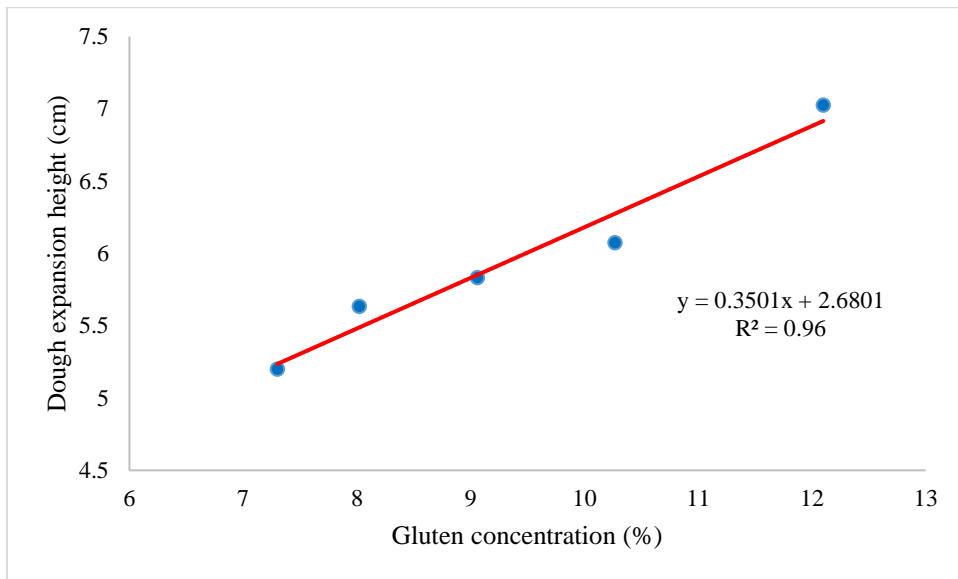


Figure 3.3 Relationship between gluten concentration and dough expansion height. The gluten concentration is expressed on 14 % moisture basis.

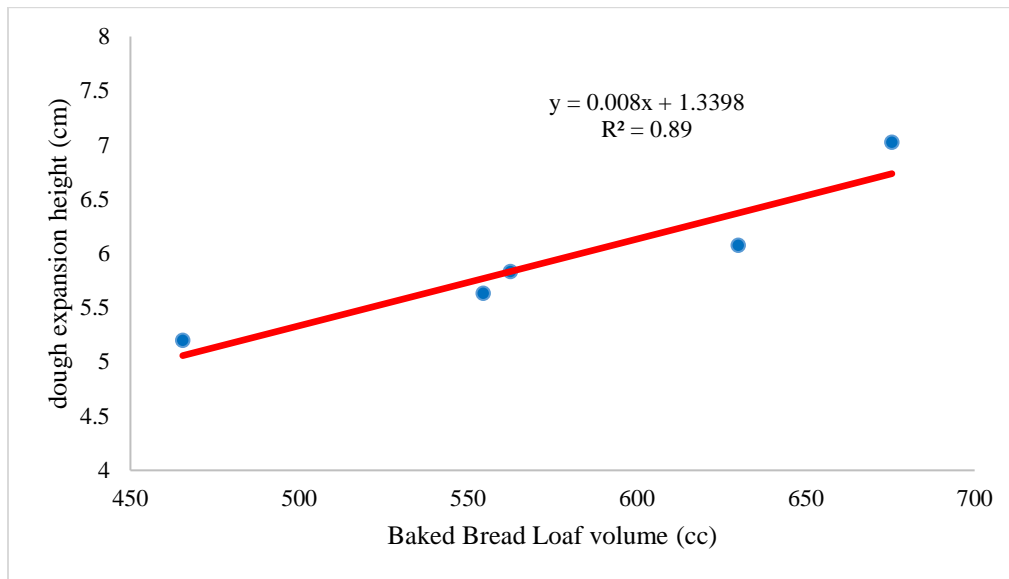


Figure 3.4 Relationship between baked loaf volume and dough expansion height for weak dough system

Figure 3.4 shows the relationship between bread loaf volume and dough expansion height. A statistically significant and high correlation was also observed between dough expansion height and loaf volume ($r = 0.94$). This provided evidence of the applicability of VDES to predict loaf volume. These findings were further investigated, and the results are discussed in the following subsection.

3.2.2 Dough expansion experiments utilizing flour blends made from pastry flour:

Experiment 2

The experiment in the section 3.2.1 was further investigated. The maximum gluten concentration of the flour blends in this experiment was increased to 14.7 % from 12.1 % (Table 2.2). Additionally, in this study, the hybrid SDS-SRC sedimentation test was also performed on the samples. Data from the hybrid SDS-SRC test are expressed as weight value (WV) and reflect the swelling behavior and water holding behavior of gluten proteins that have strong implications for bread baking potential. Table 3.6 provides bivariate correlation coefficients (r) that were calculated between parameters from the Glutomatic test, dough expansion, hybrid SDS-SRC and baked loaf characteristics. The Glutomatic test reveals the vital gluten content of flour and expresses gluten as a percentage of the flour weight. Figure 3.5 shows the relationship between dry gluten content and bread loaf volume. A statistically significant and high correlation was found between gluten content and bread loaf volume ($r = 0.98$). Figure 3.6 shows the picture of the bread loaves that were baked from the control pastry flour and the various flour blends. A statistically significant and high correlation was observed between total wet gluten and bread loaf volume ($r = 0.96$). A statistically significant and high

correlation was observed between good wet gluten and bread loaf volume ($r = 0.89$). The results were expected and provided evidence that increasing the gluten content of flour increased the bread loaf volume.

Figure 3.7 shows the relationship between dough expansion height and the bread loaf volume. Statistically significant and high correlation was observed between dough expansion height and baked loaf volume ($r = 0.98$). The data provided evidence that strong relationship existed between dough expansion attributes and bread loaf volume. The results of the above two experiment provided significant evidence to establish a proof of concept for applicability of the dough expansion system in predicting baked loaf volume.

From Table 3.6, a significant finding from the experiment was the linear response of the dough gluten strength on the dough expansion. Figure 3.8 shows the relationship between gluten content and dough expansion height. Statistically significant and high correlations were observed between dough expansion height and dry gluten content ($r = 0.98$). The dough expansion height was significantly and highly correlated to total wet gluten content ($r = 0.96$). Additionally, the dough expansion height was significantly and highly correlated to good wet gluten content ($r = 0.89$). This means that the dough expansion system may be a valuable tool to measure the quality of ingredients used as dough modifiers. A statistically significant and high correlation was found between weight value and bread loaf volume ($r = 0.99$). This was the first evidence of the applicability of the hybrid SDS-SRC sedimentation method in predicting bread loaf volume. Based on the encouraging results from the preliminary investigations, real world

HRS and HRW wheat samples were utilized to further study the applicability of the dough expansion system and the hybrid SDS-SRC sedimentation method in estimating the baking quality of HRS and HRW wheat cultivars.

Table 3.6 Bivariate correlation coefficients (r) between variables of gluten content, dough expansion and baked loaf volume for control pastry flour and blends of pastry flour enriched with vital wheat gluten (N = 12 for All; N = 6 for mean)

	GL		TWG		GWG		LV		VDEH		WV
	All	Mean	All	Mean	All	Mean	All	Mean	All	Mean	All
GL	1	1									
TWG	0.96**	0.97**	1	1							
GWG	0.85**	0.89*	0.74**	0.79	1	1					
LV	0.98**	0.98**	0.95**	0.96**	0.86**	0.89*	1	1			
VDES	0.97**	0.99**	0.97**	0.97**	0.80**	0.87*	0.97**	0.98**	1	1	
WV	0.97**	0.99**	0.98**	0.99**	0.89**	0.99**	0.97**	0.99**	0.98**	0.98*	1

GL- Gluten content; GWG- Good Wet Gluten; LV- Loaf Volume; TWG- Total Wet Gluten; VDEH- Dough Expansion height from vacuum Dough Expansion System (VDES); WV- Weight Value from hybrid SDS-SRC Sedimentation test

** and * indicate significance at P = 0.01 and 0.05, respectively

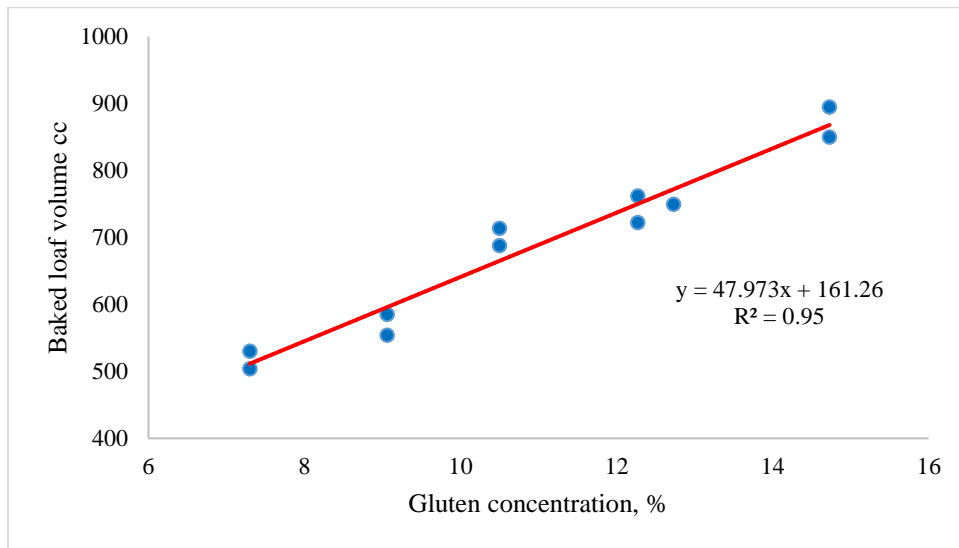


Figure 3.5 Relationship between gluten content and baked loaf volume in control pastry flour and blends of pastry flour enriched with vital wheat gluten



Figure 3.6 Bread baked from control weak flour (Pastry flour) and blends of pastry flour enriched with vital wheat gluten

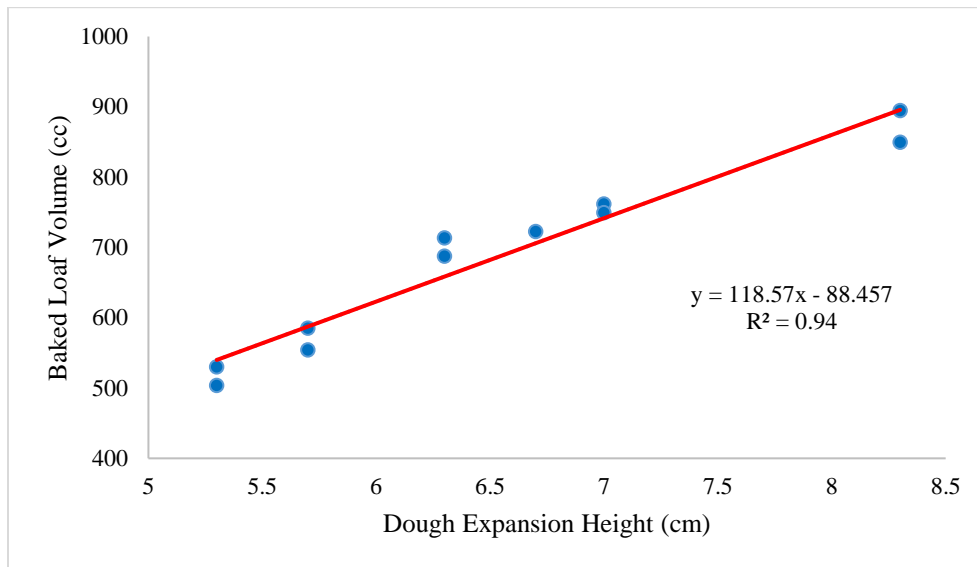


Figure 3.7 Relationship between dough expansion height and bread loaf volume for control pastry flour and blends of pastry flour enriched with vital wheat gluten

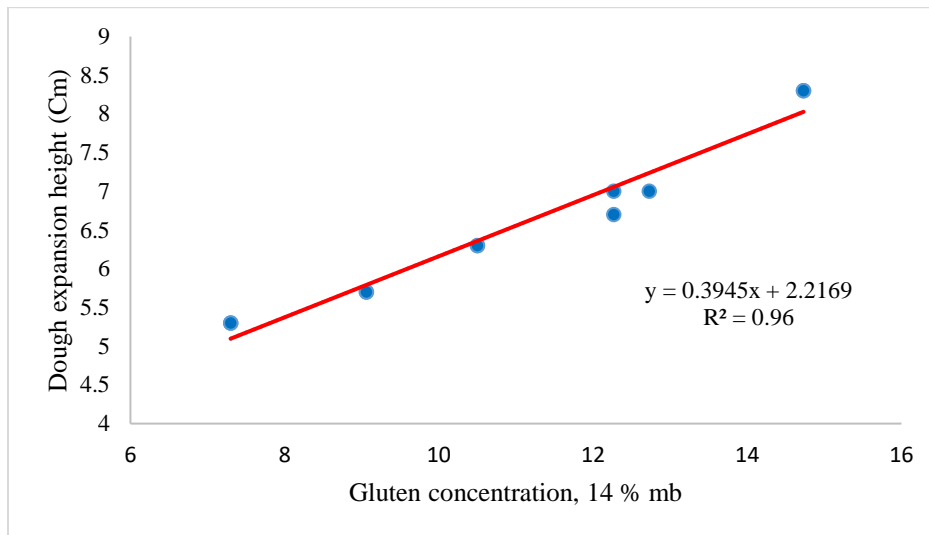


Figure 3.8 Relationship between gluten content and dough expansion in control pastry flour and blends of pastry flour enriched with vital wheat gluten.

3.3 Loaf volume predictors in hard red winter (HRW) wheat cultivars

Table 3.7 summarizes the results of prediction of baked loaf volume in HRW wheat from the various predictor variables obtained from the various grain, flour, dough tests, gluten tests and dough expansion test. The predictor variables comprised of parameters from routine flour and dough tests used for flour end-use quality, the hybrid SDS-SRC Sedimentation test, and the dough expansion test. The predictor variable from whole grain was the Grain Protein (GP) content. The predictor variables from flour were flour protein (FP) content and flour gluten (FG) content. The gluten protein was estimated from the Glutomatic instrument and was expressed as percentage in flour. The predictor variables from the Glutomatic test were the total wet gluten (TWG), good wet gluten (GWG) and water absorption capacity (WAC). The predictor variable from the hybrid SDS-SRC sedimentation assay was the weight value (WV). The predictor variables from the Mixolab were the Mixolab absorption (MLA) and the Mixolab stability (MLS). The predictor variable from the VDES was the vacuum dough expansion height (VDEH). Additionally, in the HRW wheat data, a parameter of bread loaf density called the specific loaf volume (SLV) was utilized to compute the correlations instead of bread loaf volume (LV). The specific loaf volume was calculated by dividing the baked loaf volume with loaf weight. In relating chemical composition to qualities of baked bread, specific volume (cc/g) is sometime a better parameter to measure instead of absolute volume in cubic centimeters.

From Table 3.7, when the data from the two growing locations was combined, the best predictor of baked loaf volume was found to be good wet gluten ($r = 0.39$) followed

by weight value ($r = 0.37$). This showed that protein moieties were directly linked to baking functionality or baking potential of flours.

Growing location Aurora yielded wheat that showed significant and high correlation coefficients linking bread loaf volume to grain protein, flour protein, gluten proteins, sedimentation tests and Vacuum dough height. As noted from Table 3.7, for the Aurora growing location, the best predictor of bread loaf volume was flour protein ($r = 0.62$). Additionally, the specific loaf volume was statistically significantly correlated to flour gluten content ($r = 0.60$). The specific loaf volume was statistically significantly correlated to good wet gluten ($r = 0.59$). Moreover, the specific loaf volume was statistically significantly correlated to total wet gluten ($r = 0.58$). Overall, in the Aurora growing location, the data suggested predictability of loaf volume by measuring flour protein contents and gluten related quality parameters such as total wet gluten and good wet gluten. An important observation was that the specific loaf volume was statistically significantly correlated to dough expansion height ($r = 0.49$) from the VDES test. Even though the correlation coefficient was only 0.49, it was statistically significant ($p = 0.05$).

The Onida growing location was affected by drought in the year 2016. From this location however, none of the conventional predictors of loaf volume such as the protein or wet gluten content were significantly correlated with loaf volume. From Table 3.7, in the Onida growing location, the best predictor of baked loaf volume was weight value from the hybrid SDS-SRC sedimentation test ($r = 0.48$). Weight value was the only parameter that was statistically significantly correlated to specific loaf volume in bread produced from wheat grown in this location.

Table 3.7 Bivariate correlation coefficients (r) between specific loaf volume and variables from the VDES, the hybrid SDS-SRC and the conventional predictors of bread quality in HRW wheat cultivars

Location	N	GP	FP	FG	TWG	GWG	WAC	WV	MLA	MLS	VDEH
All	48	0.25	0.27	0.26	0.17	0.39**	0.05	0.37*	-0.04	0.26	0.02
Aurora	24	0.58**	0.62**	0.60**	0.58**	0.59**	0.52*	0.48*	-0.03	0.43*	0.49*
Onida	24	0.11	0.33	0.11	-0.04	0.37	-0.20	0.48*	0.30	-0.07	-0.37

FG- Flour Gluten; FP- Flour Protein; GP- Grain Protein; GWG- Good Wet Gluten; MLA- Mixolab Absorption; MLS- Mixolab Stability; TWG- Total Wet Gluten; VDEH- Dough Expansion height from vacuum Dough Expansion System (VDES); WAC- Water Absorption Capacity; WV- Weight Value from hybrid SDS-SRC Sedimentation test

** and * indicate significance at $P = 0.01$ and 0.05 , respectively

- The complete table for each location is shared in the Appendix section at end of the document

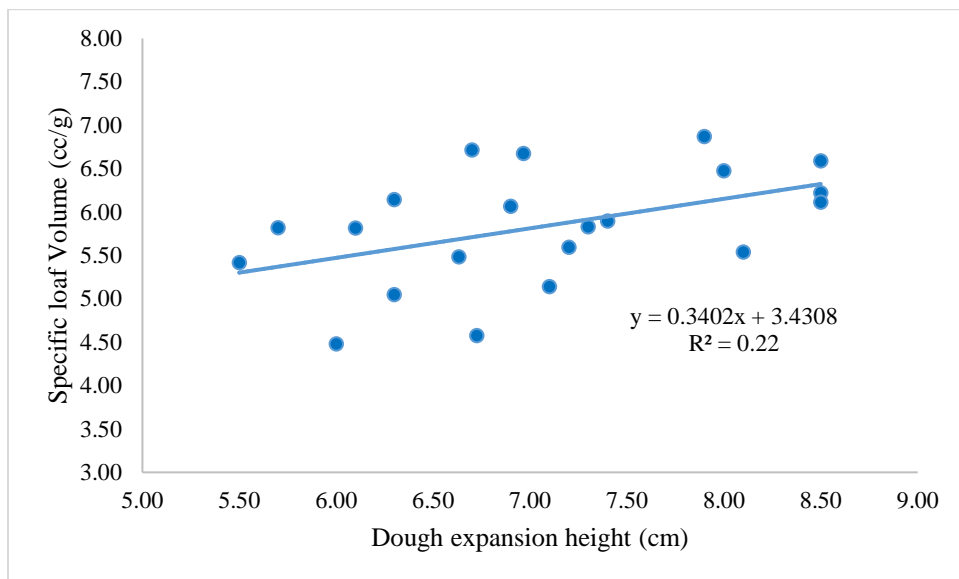


Figure 3.9 Relationship between dough expansion height (VDEH) and specific loaf volume (SLV) for 24 cultivars of HRW wheat grown at Aurora growing location.

3.3.1 Dough expansion height as a predictor of baked loaf volume in HRW wheat

From Table 3.7, in the Aurora growing location, a statistically significant correlation was observed between dough expansion height and specific loaf volume ($r = 0.49$, $p = 0.05$). Figure 3.9 shows the scatter plot between dough expansion height and the specific loaf volume for HRW wheat varieties grown at Aurora growing location. The findings were significant as the data provided validation of proof of concept of dough expansion. When the data from the two growing locations of HRW wheat was combined, there was no statistically significant correlation between dough expansion height and specific loaf volume. As noted above, the Onida growing location was affected by drought and thus combining data from Onida growing location with Aurora growing location might have diluted the relationships.

3.3.1.1 Correlation between dough expansion height (from the VDES test) and other flour and dough quality parameters

In Table 3.8, the statistically significant correlations coefficients between vacuum dough expansion height (VDEH) and the parameters from other flour and dough tests are highlighted in bold. The dough expansion height was statistically significantly correlated with parameters obtained from the Glutomatic test. The dough expansion height was statistically significantly correlated to water absorption capacity ($r = 0.71$, $p = 0.01$). Additionally, the dough expansion height was statistically significantly correlated to total wet gluten ($r = 0.66$, $p = 0.01$). The dough expansion height was statistically significantly correlated to good wet gluten ($r = 0.59$, $p = 0.01$) and to flour gluten content ($r = 0.53$, $p = 0.05$). Overall, the dough expansion height was significantly related to gluten strength

Table 3.8 Bivariate correlation coefficients (r) between the variables of grain and flour quality, dough mixing and quality and baked loaf characteristics for HRW wheat grown in Aurora growing location (N-24)

	GP	FP	FG	TWG	GWG	WAC	WV	MLA	MLPT	MLS	VDEH	SLV
GP	1.00											
FP	0.80**	1.00										
FG	0.87**	0.81**	1.00									
TWG	0.75**	0.59**	0.87**	1.00								
GWG	0.75**	0.67**	0.85**	0.84**	1.00							
WAC	0.73**	0.54**	0.83**	0.98**	0.82**	1.00						
WV	0.60**	0.57**	0.56**	0.34	0.54**	0.25	1.00					
MLA	0.13	0.23	0.26	0.39	0.30	0.38	-0.04	1.00				
MLPT	0.42*	0.30	0.56**	0.48*	0.32	0.43*	0.39	0.40	1.00			
MLS	0.54**	0.42*	0.47*	0.50*	0.53*	0.49*	0.36	0.32	0.44*	1.00		
VDEH	0.35	0.11	0.53*	0.66**	0.59**	0.71**	0.15	0.08	0.23	0.35	1.00	
SLV	0.58**	0.62**	0.60**	0.58**	0.59**	0.52*	0.48*	-0.03	0.09	0.43*	0.49*	1.00

FG- Flour Gluten; FP- Flour Protein; GP- Grain Protein; GWG- Good Wet Gluten; MLA- Mixolab Absorption; MLPT- Mixolab Peak Time; MLS- Mixolab Stability; SLV- Specific Loaf Volume; TWG- Total Wet Gluten; VDEH- Dough Expansion height from Vacuum Dough Expansion System (VDES); WAC- Water Absorption Capacity; WV- Weight Value from hybrid SDS-SRC Sedimentation test

** and * indicate significance at P = 0.01 and 0.05, respectively

and the water absorption capacity. This means that the gluten strength may be predicted by the VDES.

3.3.2 Hybrid SDS-SRC sedimentation test as a predictor of loaf volume in HRW wheat

From Table 3.7, when the data from the two growing locations of HRW wheat were combined, a statistically significant correlation was observed between the weight value (from the hybrid SDS-SRC sedimentation test) and specific loaf volume ($r=0.37$, $p = 0.01$) from the bread baking test. Statistically significant correlations between weight value and baked loaf volume were also obtained in the individual growing locations namely Aurora ($r = 0.48$, $p = 0.05$) (Figure 3.10) and Onida ($r = 0.48$, $p = 0.05$) (Figure 3.11). From the literature, Seabourn and coworkers (2012) found statistically significant correlation between weight value and bread loaf volume for 53 hard winter wheat cultivars that were selected randomly from 600 breeding lines harvested in 2006 ($r=0.84$) (Seabourn, Xiao, Tilley, Herald, & Park, 2012). The selected samples in the study were provided by the Hard Winter Wheat Quality Laboratory (Manhattan, KS) (HWWQL). In our study, the hybrid SDS-SRC sedimentation method consistently predicted bread loaf volume with statistical significance particularly in HRW wheat. Out of all the predictor variables studied in this research for HRW wheat, the weight value (from the hybrid SDS-SRC test) is the only predictor variable that had statistically significant correlation with specific loaf volume in all three data sets of HRW wheat (Table 3.7). As mentioned earlier, the Onida growing location was affected by drought in the year 2016. It is worth noting that despite the drought conditions, weigh value served as a good predictor of loaf volume in the Onida growing location. These findings have considerable significance and

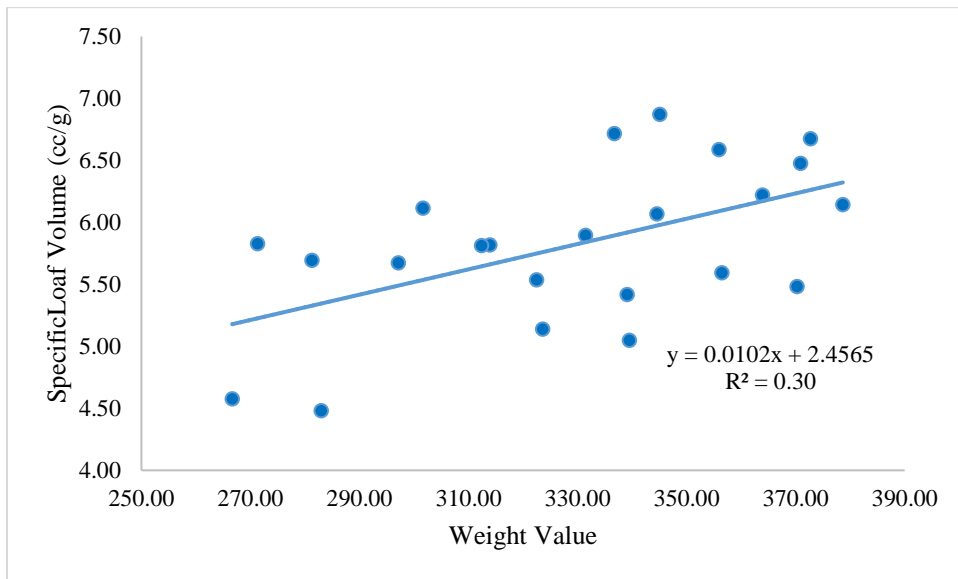


Figure 3.10 Relationship between weigh value and specific loaf volume for 24 cultivars of HRW wheat grown at Aurora growing location (N=24)

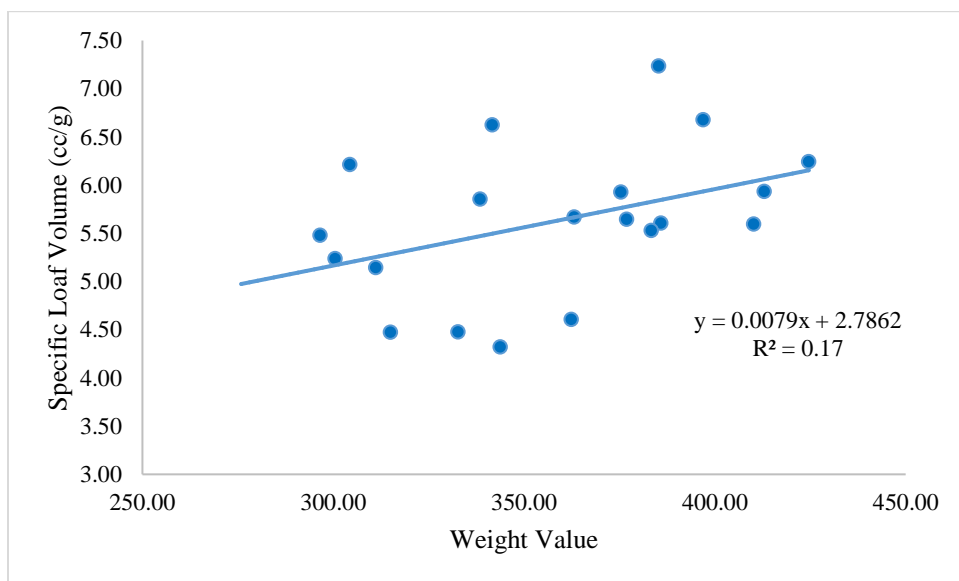


Figure 3.11 Relationship between weigh value and specific loaf volume for 24 cultivars of HRW wheat grown at Onida growing location (N=24).

provide evidence for robustness and reliability of the hybrid SDS-SRC sedimentation method even in growing environments that experience stress.

3.3.2.1 Correlation between hybrid SDS-SRC sedimentation method and other flour and dough quality parameters in HRW wheat

From the Table 3.8, the correlations between weight value and other parameters of flour and dough quality can be seen in Aurora location. The weight value obtained from the hybrid SDS-SRC sedimentation was positively correlated with flour protein content and the gluten constituents. A statistically significant correlation was observed between weight value and grain protein content ($r = 0.60$, $p = 0.01$). Additionally, weight value statistically significantly correlated to flour protein content ($r = 0.57$, $p = 0.01$). Seabourn et. al., (2012) reported similar correlation between weight value and flour protein content ($r = 0.60$ to $r = 0.65$). A statistically significant correlation was observed between weight value and flour gluten content ($r = 0.56$, $p = 0.01$). A statistically significant correlation was observed between weight value and good wet gluten ($r = 0.54$, $p = 0.01$). This means that the protein constituents in the flour and more specifically the gluten constituents contribute significantly to the weight value obtained by the hybrid SDS-SRC sedimentation method.

3.3.3 Other variables as predictor of bread loaf volume in HRW wheat

3.3.3.1 Grain protein and flour protein

Table 3.7 shows that a statistically significant correlation was observed between grain protein content and specific loaf volume for HRW wheat varieties grown in the Aurora location ($r = 0.58$). Seabourn et. al (2012) reported statistically significant correlation between wheat meal protein content and bread loaf volume for 53 hard winter wheat varieties ($r=0.64$). The correlation coefficients reported by Seabourn and coworkers (2012) agreed with the values we obtained. However, Gabriel et. al. (2017) stated that the practice of using exclusively protein content as an indicator for baking quality is questionable. Gabriel and coworkers evaluated the relationship between grain protein content and bread loaf volume using 600 winter wheat samples. They found that the correlation between grain protein content and bread loaf volume was low in samples above protein content of 12 % ($R^2 = 0.15$). Additionally, the baking quality of wheat depends on the protein quantity as well as the protein quality. A high protein content doesn't necessarily mean high quality attributes that provide baking functionality.

Table 3.7 also showed that a statistically significant correlation was observed between flour protein and specific loaf volume in the Aurora location ($r = 0.62$). Seabourn et. al (2012) reported similar statistically significant correlation between wheat flour protein and bread loaf volume for 53 hard winter wheat varieties ($r=0.66$).

3.3.3.2 Variables from the Gluten Analysis Tests (Glutomatic Analyzer)

The variables related to gluten content in dough that were investigated were the dry gluten content, total wet gluten, the good wet gluten and the water absorption

capacity. Table 3.7 showed that in the Aurora growing location, all the four variables statistically significantly correlated with specific loaf volume (a bread quality trait). The correlation coefficients were $r = 0.60$, $r = 0.58$, $r = 0.59$ and $r = 0.52$ for dry gluten content, total wet gluten, good wet gluten and water absorption capacity, respectively. It is generally accepted in the scientific literature that the quantity and quality of gluten protein fractions in wheat flour are positively correlated to bread loaf volume. Quality parameter such as the wet gluten content combine physical constituent with functionality such as the water holding capacity. However, the correlation coefficients did not increase when wet gluten content was correlated to bread loaf volume as compared to correlation coefficient between dry gluten content and bread loaf volume. Overall, the quality parameters obtained from the Glutomatic tests were positively correlated to bread loaf volume. It is worth noting that the Glutomatic test can be performed in an estimated 10 minutes and thus provides a rapid method to estimate baking quality of wheat flours.

3.3.3.3 Variables from the Mixolab

Table 3.7 shows that for the HRW wheat grown in the Aurora growing location, a statistically significant correlation was observed between Mixolab stability and Specific loaf volume ($r=0.43$). None of the other variables obtained from Mixolab namely Mixolab peak time and Mixolab absorption were found to be significantly correlated with specific loaf volume. Koxsel and coworkers (2009) used the Mixolab to study the quality of 16 bread wheat samples obtained from Fields Crop Research Center, Ankara, Turkey. Unlike our study, statistically significant correlation was not observed between Mixolab stability and Specific loaf volume in their study. Caffè-Treml and coworkers (2010) reported that the correlations between bread loaf volume and Mixolab parameters within

environments were not consistent. Additionally, the Mixolab requires 45 minutes to generate the mixing profile of one sample. Therefore, it is not suitable as a rapid tool to estimate baking quality of wheat flours.

3.3.4 Multiple regression models developed through stepwise procedure for predicting bread loaf volume in HRW wheat

Table 3.9 summarizes the results of multiple regression models that were developed by using the same predictor variables (as discussed in earlier sections) with response to bread loaf volume utilizing the HRW wheat samples. For HRW wheat varieties grown at Aurora location, regression equation comprising of Mixolab Stability and flour protein were the best predictors of specific loaf volume ($R^2 = 0.52$). The second-best model comprised of Mixolab stability and explained 38 % of the variation in specific loaf volume ($R^2 = 0.38$). Since several predictors variables were directly related to protein content or its functionality such as the water absorption capacity, they were resulting in multicollinearity between the predictor variables. In multiple regression equations, predictor variables that exhibit multicollinearity are not retained; only the best and statistically significant ones are. The multiple regression equation selected predictor variable obtained from the Mixolab namely the Mixolab stability. However, as discussed earlier, the Mixolab test requires a longer time to operate and is not conducive for large throughput analysis.

3.3.5 Multiple regression models developed by combining weight value and dough expansion height for predicting bread loaf volume

Table 3.10 summarizes the results of multiple linear regression models that were developed by combining weight value and the dough expansion height to predict specific loaf volume. It was hypothesized that combining both the variables may increase the power of prediction of baked loaf volume. For HRW wheat varieties grown in Aurora growing location, combining weight value and dough expansion height in regression increased the power of prediction of specific loaf volume ($R^2 = 0.44$). Additionally, both the variables were statistically significant ($p = 0.05$). The results are significant because both the predictor variables namely, weigh value and dough expansion height can be estimated rapidly and combining them in multiple regression equation increased the power of prediction of bread loaf volume.

Table 3.9 Linear regression models developed through stepwise procedure by using various predictor variables with response to specific loaf volume for HRW wheat

Location/Dataset	Best Predictors	R-Square
Aurora	MLS*, FP*	0.52
Aurora	MLS**	0.38

MLS- Mixolab Stability; FP- Flour Protein

** and * indicate significance at $P = 0.01$ and 0.05 , respectively.

Table 3.10 Linear regression model developed by entering WV and VDEH as predictor variables with response to specific loaf volume for HRW wheat

Location/Dataset	Predictors	R-Square
Aurora	WV*, VDEH*	0.44

WV- Weight Value; VDEH- Dough expansion height from VDES

** and * indicate significance at $P = 0.01$ and 0.05 , respectively.

3.4 Loaf volume predictors in hard red spring (HRS) wheat cultivars

Table 3.11 summarizes the results of prediction of bread loaf volume in spring wheat employing the various predictor variables. The predictor variables comprised of parameters from routine flour and dough tests used for flour end use quality, the hybrid SDS-SRC Sedimentation test, and the dough expansion test. The predictor variable from whole grain was the Grain Protein (GP) content. The predictor variables from flour were flour protein (FP) content and flour gluten (FG) content. The predictor variables from the Glutomatic test were the total wet gluten (TWG) and good wet gluten (GWG). The predictor variable from the hybrid SDS-SRC sedimentation assay was the weight value (WV). The predictor variables from the Mixograph were the Mixograph mix time (MGM), the Mixograph absorption (MGA) and the Mixograph peak time (MGPT). The predictor variables from the Farinograph were the Farinograph water absorption (FWA), Farinograph dough development time (FDDT), Farinograph stability (FSTAB), Farinograph mixing tolerance index (FMTI) and the Farinograph quality number (FQN). The predictor variable from the VDES was the vacuum dough expansion height (VDEH). The predictor variable obtained from the baking experiment was bread loaf volume (LV) and not specific loaf volume. When the data from the three growing locations was combined and considered in total, the best predictor of baked loaf volume was found to be good wet gluten ($r = 0.61$), followed by dry gluten content ($r = 0.58$) and grain protein ($r = 0.54$). In the data set, where mean for each predictor variable was computed across the three location for each cultivar, the best predictor of baked loaf volume was the weight value from the hybrid SDS – SRC sedimentation test ($r = 0.62$) followed by grain protein ($r = 0.55$) and

Table 3.11 Bivariate correlation between baked loaf volume and variables of grain and flour quality, dough mixing and quality for HRS wheat grown at three growing locations

Location	N	GP	FP	FG	TWG	GWG	WV	MGM	MGA	MGPT	FDDT	FWA	FSTAB	FMTI	FQN	VDEH
All	99	0.54**	0.28**	0.58**	0.39**	0.61**	0.31**	0.07	-0.09	-0.11	0.35**	0.07	0.19	0.01	0.40**	0.38**
Average	33	0.55**	0.55**	0.26	0.09	0.49**	0.62**	0.33	0.51**	0.22	0.36*	0.35*	0.08	-0.29	0.40*	-0.23
Brookings	33	0.24	0.39*	0.00	0.02	0.25	0.50**	0.14	0.37*	0.14	0.37*	0.22	0.12	-0.25	0.36*	0.01
Groton	33	0.53**	0.56**	0.50**	0.31	0.52**	0.59**	0.36*	0.30	0.09	0.19	0.18	0.00	-0.26	0.21	-0.15
Selby	33	0.42*	0.30	0.08	-0.08	0.42*	0.41*	0.27	0.13	0.21	0.34	-0.01	0.13	0.07	0.27	-0.09

FDDT- Farinograph Dough Development Time; FG- Flour Gluten; FMTI- Farinograph Mixing Tolerance Index; FP- Flour Protein; FQN- Farinograph Quality Number; FWA- Farinograph Water Absorption; FSTAB- Farinograph Stability; GP- Grain Protein; GWG- Good Wet Gluten; MGA- Mixograph absorption; MGM- Mixograph Mix time; MGPT- Mixograph Peak time; TWG- Total Wet Gluten; VDEH- Dough Expansion height from vacuum Dough Expansion System (VDES); WV- Weight Value from hybrid SDS-SRC Sedimentation test

** and * indicate significance at P = 0.01 and 0.05, respectively.

- The complete table for each location is shared in the Appendix section at end of the document

flour protein ($r = 0.55$). For the Brookings growing location, the best predictor of baked loaf volume was weight value ($r = 0.50$). For Groton growing location, the best predictor of baked loaf volume was weight value ($r = 0.59$). For Selby growing location, the best predictor of baked loaf volume was good wet gluten ($r = 0.42$). No single variable stood out as the major influencer of bread loaf volume across the three growing locations.

Weight value was the only predictor variable that was statistically significantly correlated with bread loaf volume in all three growing locations of HRS wheat.

3.4.1 Dough expansion height as a predictor of baked loaf volume in HRS wheat

Figure 3.12 shows relationship between dough expansion height and bread loaf volume when data from three growing locations was pooled together. A statistically significant correlation was observed between dough expansion height and baked loaf volume ($r = 0.38$, $p = 0.01$). However, Table 3.11 shows that the relationship between bread loaf volume and dough expansion height was lost in the data set where means for each predictor variable was computed across the three location for each cultivar. This means that the effect of genotype on the variance was not significant. Similarly, a statistically significant correlation between dough expansion height and loaf volume was not observed in the individual growing locations of HRS wheat unlike the Aurora growing location of HRW wheat. The reason for the observations may be explained by referring to Figure 3.16. Here, the relationship between flour protein and dough expansion height is plotted for 33 cultivars of HRS wheat grown at three different growing locations. As already mentioned earlier, Location played a significant factor and affected the flour protein content across the locations. Table 3.3

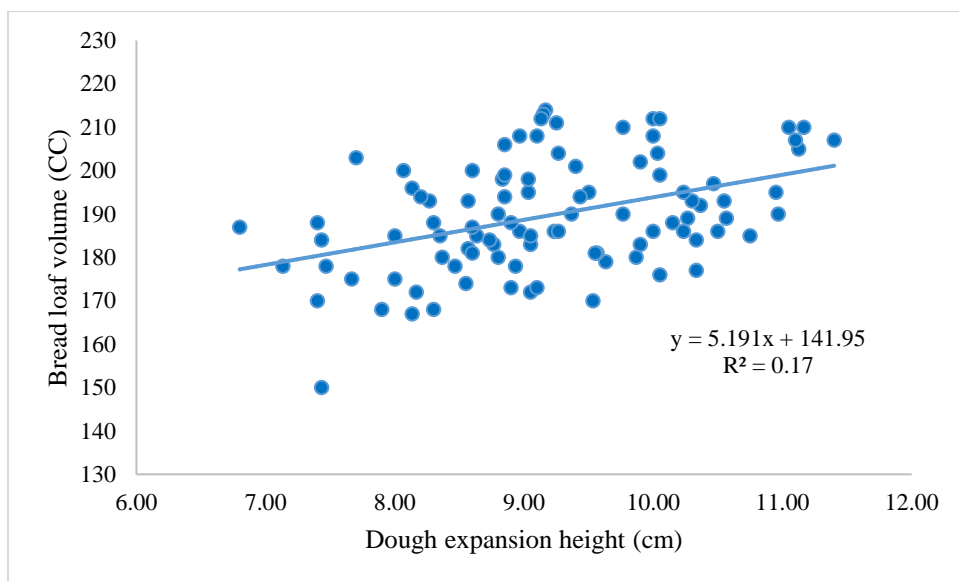


Figure 3.12 Relationship between dough expansion height and bread loaf volume for 33 cultivars of HRS wheat grown at three locations. The plot consists of data from all the three growing locations (N=99)

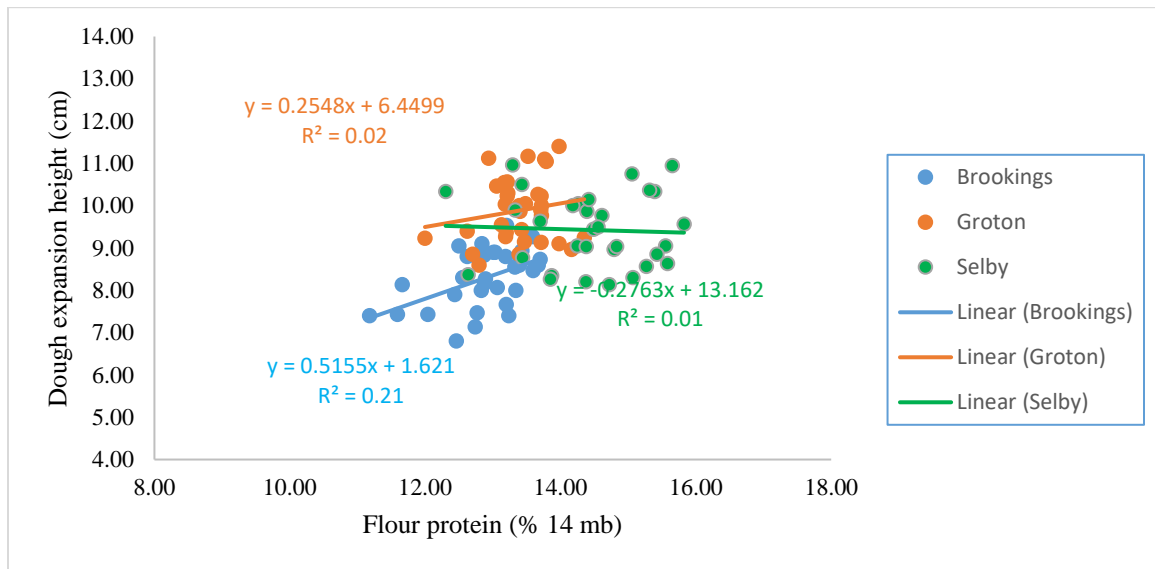


Figure 3.13 Relationship between flour protein and dough expansion height. Data for 33 cultivars grown at three different growing locations are plotted in the same graph

shows that the mean flour protein content increased across the growing locations from Brookings, Groton and Selby and was 12.9, 13.4 and 14.5 %, respectively. Figure 3.13 shows that there is a linear and statistically significant correlation between flour protein and dough expansion height in Brookings location ($R^2 = 0.21$). In the Groton location, there is no statistically significant correlation between flour protein and dough expansion; although, the trendline has a positive slope. In the Selby location, where the mean flour protein is very high, there is no statistically significant correlation between flour protein and the dough expansion height. Additionally, the trend line showed a negative slope. These observations indicate that with the increase in flour protein content in range of 11.2 to 13.7 % (mean 12.9 %), the dough expansion height also increased, as observed in the Brookings growing location ($R^2 = 0.21$). As the mean flour protein further increased (mean flour protein of 13.4 %), as in the Groton growing location, the R^2 between flour protein content and dough expansion height dropped to 0.01 (from $R^2 = 0.21$ in Brookings growing location). At very high flour protein content in the range of 12.3 to 15.8 % (mean 14.5 %), as observed in the Selby growing location, R^2 between flour protein content and dough expansion height was yet again not significant ($R^2 = 0.01$) and the slope of the curve was negative. This means low and moderate protein content supports dough expansion, and as protein content increases, the dough (and gluten strength) increases in strength and resists expansion. The reason for not observing statistically significant correlation between dough expansion height and bread loaf volume in individual growing locations of HRS wheat samples might be because of lack of samples with flour protein content in the range of 8 to 12 %. This also means that the

applicability of the dough expansion system in predicting bread loaf volume may be limited to flours with low and medium protein content.

3.4.1.1 Correlation between dough expansion height and other flour and dough quality parameters in HRS wheat

From Table 3.12, the correlation coefficients between vacuum dough expansion height (VDEH) and other parameters of flour and dough quality can be seen for all the three growing locations of HRS wheat. The dough expansion height was statistically significantly correlated to dry gluten content in Brookings growing location ($r = 0.70$, $p = 0.01$) and Groton growing location ($r = 0.35$, $p = 0.05$). The dough expansion height was statistically significantly correlated to wet gluten content in Brookings growing location ($r = 0.51$, $p = 0.01$) and Groton growing location ($r = 0.48$, $p = 0.01$). This means that the dough expansion system can be used to predict gluten quality. In other words, the dough expansion capacity of dough may be attributed to the gluten proteins. A statistically significant correlation was exhibited between dough expansion height and weight value obtained from the hybrid SDS-SRC sedimentation method in Brookings growing location ($r = 0.44$, $p = 0.05$) and Groton growing location ($r = -0.37$, $p = 0.05$).

Table 3.12 Correlation coefficients between wheat quality parameters and dough expansion height in the three growing locations of HRS wheat

Quality parameter	Brookings	Groton	Selby
<u>Wheat characteristics</u>			
Grain protein content	0.25	0.03	-0.06
Flour protein content	0.40*	0.09	-0.02
<u>Gluten characteristics</u>			
Dry gluten content	0.70**	0.35*	0.19
Wet gluten content	0.51**	0.48**	0.31
Good wet gluten content	0.40*	0.17	0.02
<u>Mixograph characteristics</u>			
Absorption	0.15	-0.17	-0.28
Mix time	-0.51**	-0.52**	-0.55**
Peak time	-0.48**	-0.44**	-0.42*
<u>Farinograph characteristics</u>			
Absorption	0.10	-0.19	-0.16
dough development time	-0.15	-0.41*	-0.41*
Stability	-0.18	-0.40*	-0.19
Mixing tolerance index	0.01	0.50**	-0.18
Farinograph quality number	-0.09	-0.51**	-0.30
<u>hybrid SDS-SRC sedimentation method</u>			
Weight value	0.44*	-0.37*	-0.30
<u>Bread characteristics</u>			
Loaf volume	0.01	-0.15	-0.09

** and * indicate significance at $P = 0.01$ and 0.05 , respectively.

3.4.2 Hybrid SDS-SRC sedimentation test as a predictor of loaf volume in HRS wheat

Table 3.11 shows that when the data from the three growing locations of HRS wheat was combined, a statistically significant correlation was observed between the weight value and baked loaf volume ($r = 0.31$, $p = 0.01$). When the weight value was averaged for each cultivar across the three locations, a statistically significant correlation was again obtained with baked loaf volume ($r = 0.62$, $p = 0.01$). Figure 3.14, Figure 3.15 and Figure 3.16 show relationship between weight value obtained from the hybrid SDS-SRC sedimentation test and bread loaf volume in Brookings, Groton and Selby growing locations, respectively. Statistically significant correlations between weight value and bread loaf volume were also obtained in the individual growing locations namely Brookings ($r=0.50$, $p = 0.01$), Groton ($r = 0.59$, $p = 0.01$) and Selby ($r = 0.41$, $p = 0.05$). As observed in the HRW wheat cultivars, the hybrid SDS-SRC sedimentation method consistently predicted baked loaf volume with statistical significance in the HRS wheat also. From Table 3.11, we can see that out of all the predictor variables studied in this research for HRS wheat, the weight value is the only predictor variable that had statistically significant correlation with baked loaf volume in all five data sets. These findings are significant as there is no published study on the application of the hybrid SDS-SRC sedimentation method for determining baking quality of HRS wheat. Additionally, for both the classes of wheat studied in this research, the weigh value significantly and consistently correlated with baked loaf volume. The hybrid SDS-SRC method may have the potential to be used as a universal method to estimate baking quality of wheat flours.

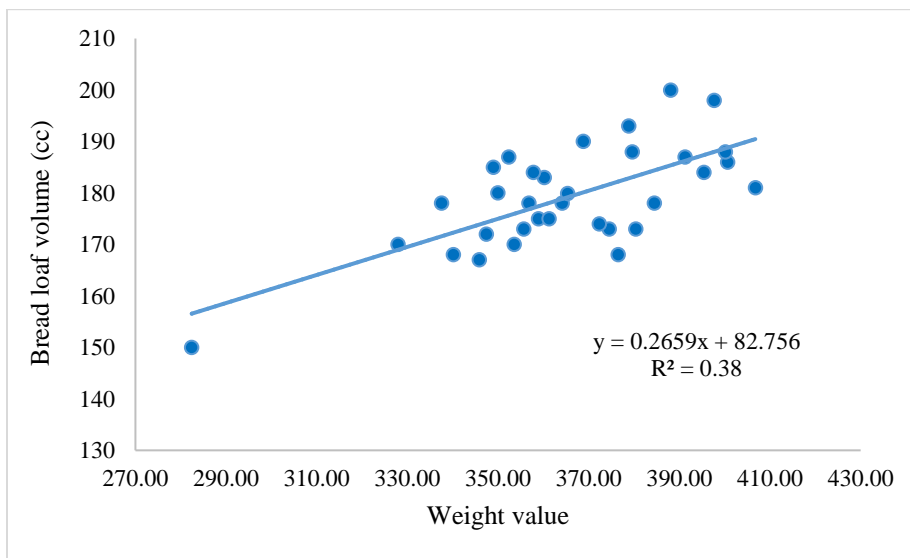


Figure 3.14 Relationship between weigh value and bread loaf volume for 33 cultivars of HRS wheat grown at Brookings location (N=33)

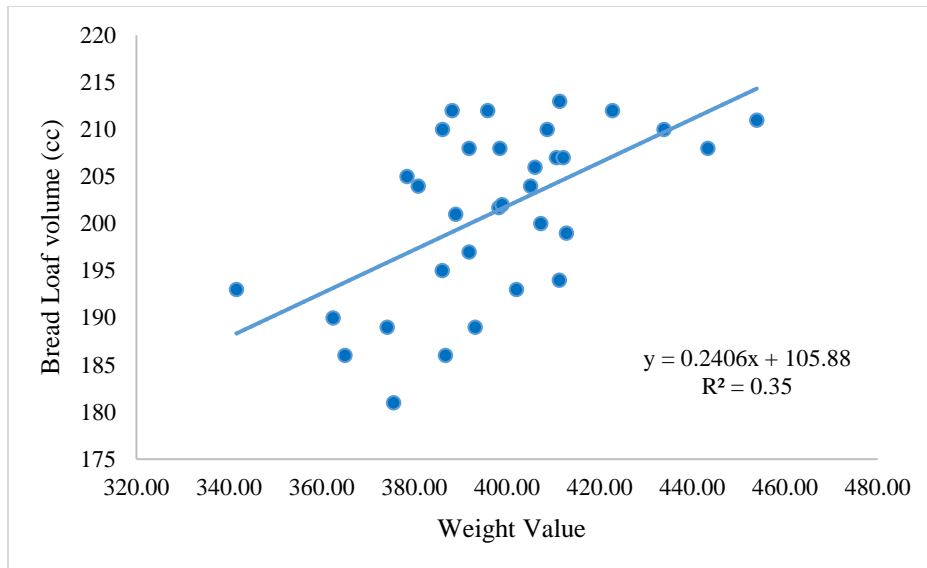


Figure 3.15 Relationship between weigh value and bread loaf volume for 33 cultivars of HRS wheat grown at Groton growing location (N=33)

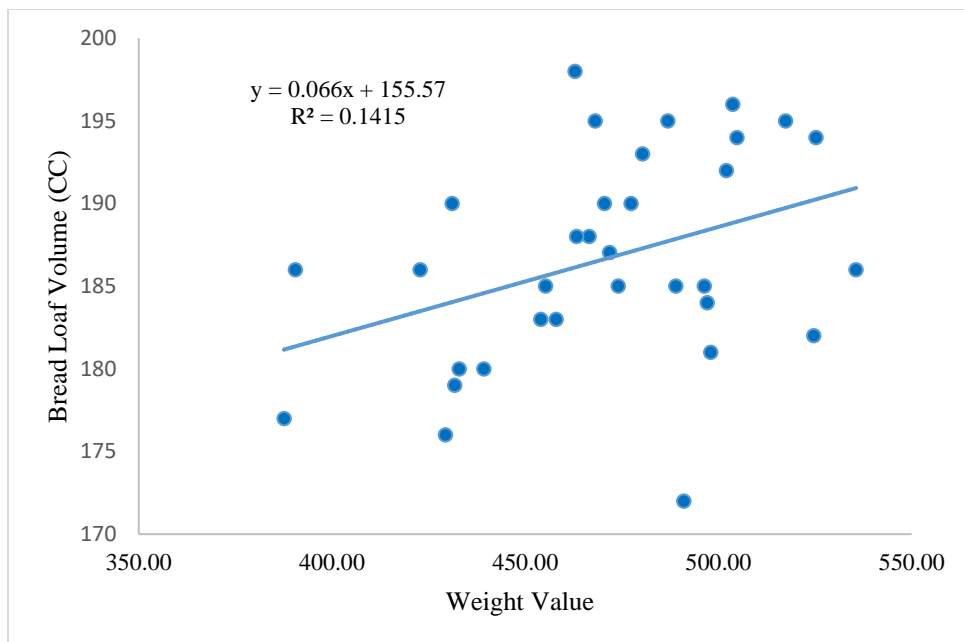


Figure 3.16 Relationship between weigh value and bread loaf volume for 33 cultivars of HRS wheat grown at Selby growing location (N=33).

3.4.2.1 Correlation between hybrid SDS-SRC sedimentation method and other flour and dough quality parameters in HRS wheat

From Table 3.13, the correlations between weight value (WV) obtained from the hybrid SDS-SRC test and other parameters of flour and dough quality can be seen for the three growing locations of HRS wheat samples. Statistically significant correlations were exhibited between weight value and flour protein constituents in Brookings ($r = 0.49$, $p = 0.01$), Groton ($r = 0.50$, $p = 0.01$), and Selby growing locations ($r = 0.75$, $p = 0.01$). Statistically significant correlation was observed between weight value and good wet gluten content in Brookings ($r = 0.73$, $p = 0.01$) and Selby growing locations ($r = 0.73$, $p = 0.01$). This means the hybrid SDS-SRC sedimentation may be applicable to predict good wet gluten of wheat flour samples. The weight value was statistically significantly correlated with Farinograph quality number in Brookings ($r = 0.46$, $p = 0.01$) and Groton ($r = 0.59$, $p = 0.01$) growing locations. The statistically significant correlations with other quality parameters particularly good wet gluten and Farinograph quality number further validates that the hybrid SDS-SRC method may be a very reliable and robust method to estimate baking quality of wheat flours.

Table 3.13 Correlation coefficients between wheat quality parameters and weight value from the hybrid SDS-SRC sedimentation method in the three growing locations of HRS wheat cultivars

Quality parameter	Brookings	Groton	Selby
<u>Wheat characteristics</u>			
Grain protein content	0.21	0.45**	46**
Flour protein content	0.49**	0.50**	0.75**
<u>Gluten characteristics</u>			
Dry gluten content	0.47**	0.35*	0.03
Wet gluten content	0.19	0.08	-0.14
Good wet gluten content	0.73**	0.45**	0.34
<u>Mixograph characteristics</u>			
Absorption	0.39*	0.30	0.29
Mix time	-0.04	0.64**	0.27
Peak time	0.08	0.46**	0.09
<u>Farinograph characteristics</u>			
Absorption	0.31	0.07	0.08
dough development time	0.32	0.42*	0.22
Stability	0.35*	0.38*	0.21
Mixing tolerance index	-0.40*	-0.15	-0.18
Farinograph quality number	0.46**	0.59**	0.22
<u>VDES</u>			
Dough expansion height	0.44*	-0.37*	-0.30
<u>Bread characteristics</u>			
Loaf volume	0.50**	0.59**	0.41*

** and * indicate significance at $P = 0.01$ and 0.05 , respectively.

3.4.3 Other variables as predictor of baked loaf volume in HRS wheat

3.4.3.1 Grain protein and flour protein

Table 3.11 shows that a statistically significant correlation was observed between grain protein and baked loaf volume in four out of the five data sets of HRS wheat. The correlation coefficients were in the range from $r = 0.42$ to $r = 0.55$. The results obtained in our study agreed with the other earlier studies. Relationship between bread loaf volume and grain protein was studied by Gabriel et. al. (2017). These researchers found a moderately strong correlation between bread volume and grain protein with coefficient of determination (R^2) of 0.59. For samples with more than 12 % protein content, protein content was a poor indicator of loaf volume with R^2 of 0.15. However, for samples with less than 12 % protein, the predictive power of protein content was much higher ($R^2 = 0.64$) (Gabriel et al., 2017).

Table 3.11 shows that a statistically significant correlation was observed between flour protein and baked loaf volume in four out of the five data sets of HRS wheat as expected. The correlations were in the range from $r = 0.28$ to $r = 0.56$. Dhaka et. al. (2012) reported statistically significant correlation between flour protein content and specific loaf volume for 15 wheat varieties in the protein range of 8.61 to 14.7 % that were grown at different wheat research stations and agricultural universities in India ($r = 0.60$).

3.4.3.2 Variables from the Glutomatic test

The variables related to gluten content in dough that were investigated were the dry gluten content, total wet gluten and the good wet gluten. Among these, the most

reliable and consistent predictor of the baked loaf volume was the good wet gluten. Table 3.11 shows that a statistically significant correlation between good wet gluten and the baked loaf volume was observed in four out of the five data sets in HRS wheat. The correlation coefficients were in the range from $r = 0.42$ to $r = 0.61$. The results obtained in this study agreed with other studies. Yaming Lu (2017) studied the predictability of the parameters obtained from the Glutomatic test in predicting bread loaf volume. For 48 genotypes of spring wheat, grown at three growing locations of South Dakota in the year 2013, good wet gluten statistically significantly correlated with bread loaf volume ($r=0.29$). Dry gluten statistically significantly correlated with bread loaf volume ($r=0.36$) (mean of the parameter were computed across the three growing location to compute the correlations). For the same genotypes grown at the same locations in the year 2015, good wet gluten significantly correlated with bread loaf volume ($r=0.48$). Based on this study and the past studies, there is significant evidence to support that good wet gluten is reliable predictor of bread loaf volume. However, in terms of the sample size and the analysis time, the hybrid SDS-SRC sedimentation test may be a better alternative to the Glutomatic test.

3.4.3.3 Variables from the Mixograph

Table 3.11 shows that statistically significant correlations between Mixograph water absorption and baked loaf volume were observed in two out of five data sets. The correlation coefficients between Mixograph water absorption and bread loaf volume were low and ranged from $r = 0.37$ to $r = 0.51$. There was no consistency in the correlations between the three growing locations.

3.4.3.4 Variables from the Farinograph

Table 3.11 shows that a statistically significant correlation between Farinograph dough development time and baked loaf volume was observed in three out of five data sets. The correlation coefficients were low and ranged from $r = 0.35$ to $r = 0.37$. Similarly, a statistically significant correlation between the Farinograph quality number and baked loaf volume was observed in the same three data sets. The correlation coefficients were low and ranged from $r = 0.36$ to $r = 0.40$.

3.4.4 Multiple regression models developed through stepwise procedure for predicting bread loaf volume in HRS wheat

Table 3.14 summarizes the results of linear regression models that were developed by using the various predictor variables with response to baked loaf volume. The stepwise procedure of regression development was used. When the data from the three growing locations was combined, the best predictors of baked loaf volume were found to be good wet gluten and the Farinograph quality number. The two predictors in combination explained 49 % of the variation in baked loaf volume ($R^2 = 0.49$). In data set, where mean for each predictor variable was computed across the three location for each cultivar, the best predictor of baked loaf volume was the weight value from the hybrid SDS – SRC sedimentation test ($R^2 = 0.46$). For Brookings location, the best predictor of baked loaf volume was again weight value ($R^2 = 0.21$). For Groton location, the best predictor of baked loaf volume was weight value ($R^2 = 0.35$). For Selby location, the best predictor of baked loaf volume was grain protein ($R^2 = 0.22$).

Table 3.14 Multiple regression models for HRS wheat samples developed through stepwise procedure by using multiple predictor variables with response to bread loaf volume

Location/Dataset	Best Predictors	R-Square
All	GWG***, FQN***	0.49
Averaged	WV***	0.46
Brookings	WV*	0.21
Selby	GP**	0.22
Groton	WV***	0.35

GWG- Good Wet Gluten; GP- Grain Protein; FQN- Farinograph Quality Number; WV- Weight Value

***, ** and * indicate significance at P= 0.001, 0.01 and 0.05, respectively

Table 3.15 Multiple regression models developed by entering WV and VDES as predictor variables with response to bread loaf volume for HRS wheat cultivars

Location/Dataset	Predictors	R-Square
All	WV, VDES	0.15
Averaged	WV***, VDES	0.47
Brookings	WV**, VDES	0.28
Selby	WV*, VDES	0.16
Groton	WV***, VDES	0.37

VDES- Dough expansion height from VDES; WV- Weight Value

***, ** and * indicate significance at P= 0.001, 0.01 and 0.05, respectively.

3.4.5 Multiple regression models developed by combining weight value and dough expansion height for predicting bread loaf volume in HRS wheat

Table 3.15 summarizes the results of multiple linear regression models that were developed by combining weight value and the dough expansion height to predict baked loaf volume. It was hypothesized that combining both the variables may increase the power of prediction of baked loaf volume. Additionally, both weight value and dough expansion height can be determined in relatively less time as compared to the conventional predictors of bread loaf volume. However, in none of the data set, the dough expansion height was found to be statistically significant in combination with weight value (unlike findings noted in HRW wheat). One of the reasons for this observation may be multicollinearity between the weight value and the dough expansion height. From Table 3.13, it is evident that there was statistically significant correlation between weight value and dough expansion height in Brookings ($r = 0.44$) and Groton ($r = -0.37$) growing locations. When two predictor variables provide redundant information, one of them is eliminated from the multiple regression equation and only the best predictor variable is retained in the equation, namely weight value, in two out of three data sets (Table 3.14).

CHAPTER 4. CONCLUSIONS

The expression of bread loaf volume depends on the combination and interaction of several constituents. Gluten strength is a significant factor that determines the rheological, processing and end-product quality (bread loaf volume) of wheat flour. Variations in the composition of wheat is largely determined by genotype however, environment and its interaction with genotype plays an important role in determining the phenotype of wheat. Evaluation of the baking quality is of utmost importance along the value chain of wheat. Several analytical methods are available to determine the baking quality of wheat.

Two rapid methods were evaluated for their accuracy and effectiveness in predicting bread loaf volume. Overall, quality parameters measuring protein constituents or functionality of protein such as the water holding capacity were moderately correlated to bread loaf volume.

4.1 Dough expansion attributes as indicator of baking quality

In the current study, the dough expansion system was investigated as a rapid tool to measure baking quality of wheat flours. The VDES successfully and consistently expanded dough pieces with an acceptable coefficient of variation (CV). In the preliminary investigation that was carried on pastry flour blends enriched with vital wheat gluten, we found evidence of a statistically significant relationship between dough expansion height and bread loaf volume. Further investigation on HRW wheat varieties grown at Aurora location found that there was a statistically significant correlation between dough expansion height and loaf volume. For HRS wheat samples, statistically

significant correlation was observed when all the samples were pooled together irrespective of growing location. Based on these findings, we reject the null hypothesis number 1 and accept the alternative hypothesis that a statistically significant correlation exists between dough expansion attribute and corresponding bread loaf volume. The study established proof of concept of applicability of VDES and further validated the concept utilizing true cultivars representing two major classes of wheat. Another significant finding was non-linearity between dough expansion height and bread loaf in samples with high protein content. Generally, acceptable predictability of baking performance from VDES was observed in weak dough systems (flour protein 8 to 12.5 %).

Another potential application of the VDES may be to study the effect of dough modifying ingredients on dough rheology and final baked product. The baking industry is going through the clean label movement. There is significant research being undertaken to replace conventional ingredients in bread with more acceptable and natural ingredients. Currently, the most reliable and accurate method to study the effect of new formulations on dough properties and final baked product is to carry standard baking test. This is time consuming. The VDES offers a rapid and reliable alternative to baking test for the purpose of testing new formulations. Our study which utilized vital wheat gluten (VWG) in pastry flour demonstrated that dough expansion was significantly related to the VWG content in the new formulations. Hence, we reject the null hypothesis number 2 and accept the alternative hypothesis that the effect of vital wheat gluten addition on dough expansion height is incremental and linear as evaluated by the VDES. We recommend

further testing of the VDES to see the response of other commonly used dough modifiers on dough expansion.

4.1.1 Recommendations for improving the VDES

Based on the current study, we have come up with some recommendations to further improve capabilities of the VDES.

4.1.1.1 Integrate the mixing and resting function in the VDES

In the current investigation, to prove a concept for the VDES, data from other devices was used. Mixolab was utilized to determine flour water absorption and Swanson mixer was used to form a dough prior to expanding it on the VDES. Our hypothesis is that using multiple instruments may have resulted in adding up of errors.

We propose to develop an instrument that first determines water absorption of a flour samples. Once the water absorption is determined, the flour should be mixed with the determined amount of water to form a dough with optimum consistency. This would generate a mixing profile. Finally, the optimally developed dough can then be expanded after giving a resting time of 10 minutes. Additionally, unlike in the current VDES where maximum dough expansion was measured, the proposal is to acquire complete dough expansion profile as shown in Figure 4.1. The advantages of such an instrument would be manifold.

1. The instrument may negate the need to perform multiple quality tests utilizing multiple instruments.
2. The proposed instrument will generate a mixing profile just as the Mixograph.

3. The instrument will expand dough piece to predict loaf volume and generate expansion profile to provide more information on the flour quality.

4. The hypothesized instrument may negate the need for performing other quality tests such as the Glutomatic and hybrid SDS-SRC sedimentation test. In the current study, it was evident that dough expansion was statistically correlated with gluten content.

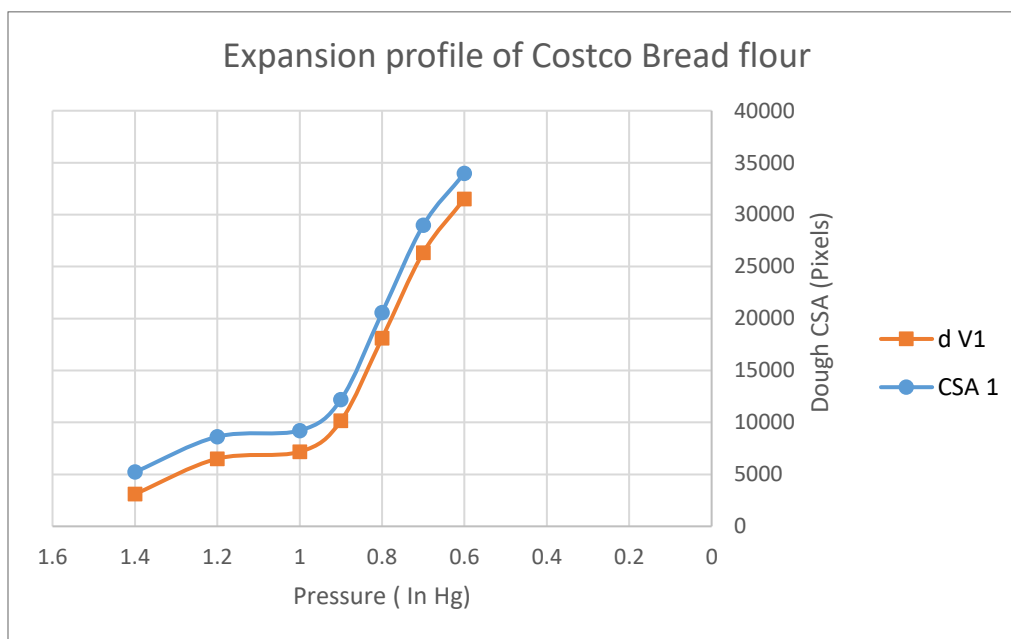


Figure 4.1 Dough expansion profile of bread flour procured from Costco

4.2 The hybrid SDS-SRC sedimentation test as an indicator of baking quality

In the current study, applicability of the hybrid SDS- SRC sedimentation test in predicting loaf volume was investigated. We got enough evidence that the weight value obtained from the hybrid SDS-SRC sedimentation test was statistically significantly correlated to bread loaf volume. Thus, we reject the null hypothesis number 3 and accept the alternative hypothesis that there is a statistically significant correlation between

weight value and bread loaf volume. The test which was developed for measuring baking quality of winter wheat cultivars grown in Kansas was found to be applicable for HRS wheat as well as HRW wheat cultivars grown in South Dakota. The findings are significant as the test is rapid and may be utilized by the winter and spring breeding programs in South Dakota. The relationship between the weight value and bread loaf volume was not lost in environment affected by stress. Furthermore, the method predicted bread loaf volume in growing locations with very high protein. The hybrid method provides significant time saving as compared to the individual SDS and the SRC methods; it can be performed in 10 minutes. Additionally, the test requires only 1-gram sample. We recommend further investigation on validation of the test with samples acquired from different growing years. Additionally, developing an automated instrument for the test may decrease testing time further.

4.3 Combining the weight value and dough expansion height in multiple regression equation to predict bread loaf volume

It was hypothesized that combining dough expansion height and weight value in a multiple regression equation would increase the power of prediction of bread loaf volume. In HRW wheat, combining weight value and dough expansion height increased the power of prediction of bread loaf volume. However, in the HRS wheat, the application of dough expansion height was not found to be statistically significant in combination with weight value in a multiple regression equation.

APPENDIX

Table A.1 Bivariate correlation coefficients (r) between the variables of grain and flour quality, dough mixing and quality, and baked loaf characteristics for HRS wheat grown in Brookings location (N-33)

	GP	FP	FG	TWG	GWG	WV	MGM	MGA	MGPT	FDDT	FWA	FSTAB	FMTI	FQN	VDEH
GP	1.00														
FP	0.82**	1.00													
FG	0.36	0.63**	1.00												
TWG	0.58**	0.72**	0.72**	1.00											
GWG	0.55**	0.55**	0.55**	0.36*	1.00										
WV	0.21	0.49**	0.47**	0.19	0.73**	1.00									
MGM	-0.47*	-0.38*	-0.55**	-0.67**	-0.23	-0.04	1.00								
MGA	0.23	0.25	0.17	0.13	0.19	0.39*	-0.16	1.00							
MGPT	-0.35	-0.21	-0.47**	-0.52**	-0.08	0.08	0.83**	-0.27	1.00						
FDDT	0.06	0.28	-0.10	-0.12	0.01	0.32	0.34	0.06	0.50**	1.00					
FWA	0.39*	0.31	0.09	0.07	0.25	0.31	-0.25	0.55**	-0.31	0.19	1.00				
FSTAB	-0.09	0.06	-0.08	-0.18	0.14	0.35*	0.41*	0.01	0.66**	0.65**	-0.01	1.00			
FMTI	0.06	-0.12	-0.09	0.05	-0.22	-0.40*	-0.33	-0.03	-0.37*	-0.56**	0.04	-0.75**	1.00		
FQN	-0.09	0.17	-0.09	-0.16	0.07	0.46**	0.37*	0.08	0.60**	0.83**	0.09	0.85**	-0.76**	1.00	
VDEH	0.25	0.40*	0.70**	0.51**	0.40*	0.44*	-0.51**	0.15	-0.48**	-0.15	0.10	-0.18	0.01	-0.09	1.00
LV	0.24	0.39*	0.00	0.02	0.25	0.50**	0.14	0.37*	0.14	0.37*	0.22	0.12	-0.25	0.36*	0.01

FDDT- Farinograph Dough Development Time; FG- Flour Gluten; FMTI- Farinograph Mixing Tolerance Index; FP- Flour Protein; FQN- Farinograph Quality Number; FWA- Farinograph Water Absorption; FSTAB- Farinograph Stability; GP- Grain Protein; GWG- Good Wet Gluten; MGA- Mixograph absorption; MGM- Mixograph Mix time; MGPT- Mixograph Peak time; TWG- Total Wet Gluten; VDEH- Dough Expansion height from Vacuum Dough Expansion System (VDES); WV- Weight Value from hybrid SDS-SRC Sedimentation test

** and * indicate significance at P = 0.01 and 0.05, respectively.

Table A.2 Bivariate correlation coefficients (r) between the variables of grain and flour quality, dough mixing and quality and baked loaf characteristics for HRS wheat cultivars grown in Selby growing location (N- 33)

	GP	FP	FG	TWG	GWG	WV	MGM	MGA	MGPT	FDDT	FWA	FSTAB	FMTI	FQN	VDEH
GP	1.00														
FP	0.70**	1.00													
Gluten	0.44*	0.35*	1.00												
TWG	0.37*	0.33	0.74**	1.00											
GWG	0.44*	0.49**	0.51**	0.20	1.00										
WV	0.46**	0.75**	0.03	-0.14	0.34	1.00									
MGM	-0.16	-0.16	-0.34	-0.64**	-0.02	0.27	1.00								
MGA	0.21	0.13	0.14	-0.19	0.44*	0.29	0.24	1.00							
MGPT	-0.31	-0.32	-0.32	-0.71**	-0.06	0.09	0.83**	0.21	1.00						
FDDT	0.06	-0.07	-0.27	-0.52**	0.00	0.22	0.42*	0.19	0.53**	1.00					
FWA	0.19	0.10	0.19	0.08	0.35*	0.08	-0.09	0.57**	-0.13	0.06	1.00				
FSTAB	-0.02	-0.10	-0.17	-0.42*	-0.02	0.21	0.39*	0.02	0.39*	0.41*	-0.37*	1.00			
FMTI	0.02	-0.09	0.05	0.18	0.07	-0.18	-0.13	-0.08	-0.08	0.03	-0.17	-0.24	1.00		
FQN	0.13	-0.06	-0.02	-0.49**	0.10	0.22	0.49**	0.24	0.57**	0.69**	-0.11	0.81**	-0.23	1.00	
VDEH	-0.06	-0.02	0.19	0.31	0.02	-0.30	-0.55**	-0.28	-0.42*	-0.41*	-0.16	-0.19	-0.18	-0.30	1.00
LV	0.42*	0.30	0.08	-0.08	0.42*	0.41*	0.27	0.13	0.21	0.34	-0.01	0.13	0.07	0.27	-0.09

FDDT- Farinograph Dough Development Time; FG- Flour Gluten; FMTI- Farinograph Mixing Tolerance Index; FP- Flour Protein; FQN- Farinograph Quality Number; FWA- Farinograph Water Absorption; FSTAB- Farinograph Stability; GP- Grain Protein; GWG- Good Wet Gluten; MGA- Mixograph absorption; MGM- Mixograph Mix time; MGPT- Mixograph Peak time; TWG- Total Wet Gluten; VDEH- Dough Expansion height from Vacuum Dough Expansion System (VDES); WV- Weight Value from hybrid SDS-SRC Sedimentation test

** and * indicate significance at P = 0.01 and 0.05, respectively

Table A.3 Bivariate correlation coefficients (r) between the variables of grain and flour quality, dough mixing and quality and baked loaf characteristics for HRS wheat cultivars grown in Groton location (N-33)

	GP	FP	FG	TWG	GWG	WV	MGM	MGA	MGPT	FDDT	FWA	FSTAB	FMTI	FQN	VDEH
GP	1.00														
FP	0.89**	1.00													
FG	0.65**	0.71**	1.00												
TWG	0.60**	0.63**	0.88**	1.00											
GWG	0.65**	0.69**	0.89**	0.84**	1.00										
WV	0.45**	0.50**	0.35*	0.08	0.45**	1.00									
MGM	0.13	0.08	-0.02	-0.26	0.16	0.64**	1.00								
MGA	0.14	0.21	0.36*	0.30	0.29	0.30	-0.06	1.00							
MGPT	-0.01	-0.12	-0.33	-0.49**	-0.15	0.46**	0.80**	-0.30	1.00						
FDDT	0.01	0.16	-0.04	-0.22	0.06	0.42*	0.38*	0.28	0.28	1.00					
FWA	0.23	0.27	-0.02	0.05	0.10	0.07	-0.18	0.23	-0.24	0.31	1.00				
FSTAB	-0.05	-0.03	-0.20	-0.32	0.00	0.38*	0.55**	-0.14	0.50**	0.47**	-0.08	1.00			
FMTI	0.11	0.04	0.21	0.26	0.09	-0.15	-0.19	-0.07	-0.11	-0.42*	-0.26	-0.44*	1.00		
FQN	0.04	0.15	-0.11	-0.34	0.02	0.59**	0.54**	0.13	0.48**	0.82**	0.06	0.79**	-0.44*	1.00	
VDEH	0.03	0.09	0.35*	0.48**	0.17	-0.37*	-0.52**	-0.17	-0.44**	-0.41*	-0.19	-0.40*	0.50**	-0.51**	1.00
LV	0.53**	0.56**	0.50**	0.31	0.52**	0.59**	0.36*	0.30	0.09	0.19	0.18	0.00	-0.26	0.21	-0.15

FDDT- Farinograph Dough Development Time; FG- Flour Gluten; FMTI- Farinograph Mixing Tolerance Index; FP- Flour Protein; FQN- Farinograph Quality Number; FWA- Farinograph Water Absorption; FSTAB- Farinograph Stability; GP- Grain Protein; GWG- Good Wet Gluten; MGA- Mixograph absorption; MGM- Mixograph Mix time; MGPT- Mixograph Peak time; TWG- Total Wet Gluten; VDEH- Dough Expansion height from Vacuum Dough Expansion System (VDES); WV- Weight Value from hybrid SDS-SRC Sedimentation test

** and * indicate significance at P = 0.01 and 0.05, respectively

Table A.4 Bivariate correlation coefficients (r) between the variables of grain and flour quality, dough mixing and quality and baked loaf characteristics for HRS wheat cultivars grown in Brookings, Selby and Groton growing locations. The data from all the three locations was pooled to compute the correlations (N=99)

	GP	FP	FG	TWG	GWG	WV	MGM	MGA	MGPT	FDDT	FWA	FSTAB	FMTI	FQN	VDEH
GP	1.00														
FP	0.60**	1.00													
FG	0.67**	0.46**	1.00												
TWG	0.62**	0.35**	0.83**	1.00											
GWG	0.62**	0.21*	0.77**	0.65**	1.00										
WV	0.35**	0.81**	0.32**	0.05	0.14	1.00									
MGM	-0.19	-0.39**	-0.30**	-0.35**	0.02	-0.22*	1.00								
MGA	-0.08	-0.40**	-0.15	0.02	0.15	-0.51**	0.34**	1.00							
MGPT	-0.31**	-0.48**	-0.44**	-0.43**	-0.10	-0.33**	0.86**	0.33**	1.00						
FDDT	0.19	0.41**	0.12	-0.13	0.10	0.57**	0.09	-0.28**	0.03	1.00					
FWA	0.23*	0.26**	0.11	0.08	0.19	0.18	-0.21*	0.20*	-0.25*	0.23*	1.00				
FSTAB	0.04	0.26**	0.03	-0.21*	0.04	0.53**	0.18	-0.36**	0.14	0.62**	-0.10	1.00			
FMTI	0.10	-0.321**	0.09	0.24*	0.16	-0.49**	-0.01	0.30**	0.04	-0.40**	-0.10	-0.57**	1.00		
FQN	0.23*	0.46**	0.24*	-0.10	0.15	0.70**	0.06	-0.41**	-0.02	0.85**	0.05	0.85**	-0.52**	1.00	
VDEH	0.31**	0.33**	0.66**	0.51**	0.38**	0.30**	-0.53**	-0.45**	-0.55**	0.10	-0.02	0.04	-0.03	0.22*	1.00
LV	0.54**	0.28**	0.58**	0.39**	0.61**	0.31**	0.07	-0.09	-0.11	0.35**	0.07	0.19	0.01	0.40**	0.38**

FDDT- Farinograph Dough Development Time; FG- Flour Gluten; FMTI- Farinograph Mixing Tolerance Index; FP- Flour Protein; FQN- Farinograph Quality Number; FWA- Farinograph Water Absorption; FSTAB- Farinograph Stability; GP- Grain Protein; GWG- Good Wet Gluten; MGA- Mixograph absorption; MGM- Mixograph Mix time; MGPT- Mixograph Peak time; TWG- Total Wet Gluten; VDEH- Dough Expansion height from vacuum Dough Expansion System (VDES); WV- Weight Value from hybrid SDS-SRC Sedimentation test

** and * indicate significance at P = 0.01 and 0.05, respectively

Table A.5 Bivariate correlation coefficients (r) between the variables of grain and flour quality, dough mixing and quality and baked loaf characteristics for HRS wheat grown in Brookings, Selby and Groton growing locations. The data for each variable for a cultivar was averaged across the three growing locations to compute the correlations (N=33)

	GP	FP	FG	TWG	GWG	WV	MGM	MGA	MGPT	FDDT	FWA	FSTAB	FMTI	FQN	VDEH
GP	1.00														
FP	0.62**	1.00													
FG	0.56**	0.82**	1.00												
TWG	0.39*	0.66**	0.84**	1.00											
GWG	0.72**	0.87**	0.79**	0.59**	1.00										
WV	0.42*	0.63**	0.34	0.03	0.65**	1.00									
MGM	0.00	-0.07	-0.36*	-0.66**	-0.04	.43*	1.00								
MGA	0.33	0.27	0.20	0.03	0.33	0.47**	0.08	1.00							
MGPT	-0.04	-0.24	-.50**	-0.68**	-0.16	0.18	0.86**	-0.12	1.00						
FDDT	0.30	0.16	-0.03	-0.24	0.24	0.32	0.49**	0.36*	0.56**	1.00					
FWA	0.32	0.27	0.12	0.13	0.29	0.24	-0.20	0.61**	-0.24	0.16	1.00				
FSTAB	-0.05	-0.17	-0.27	-0.56**	-0.01	0.21	0.72**	0.08	0.69**	0.50**	-0.04	1.00			
FMTI	-0.09	-0.02	0.18	0.39*	-0.17	-0.33	-0.57**	-0.17	-0.53**	-0.56**	-0.13	-.732**	1.00		
FQN	0.20	0.03	-0.17	-0.50**	0.14	0.41*	0.75**	0.25	0.75**	0.74**	0.05	0.80**	-0.80**	1.00	
VDEH	0.28	0.25	0.53**	0.58**	0.26	-0.14	-0.64**	-0.28	-0.55**	-0.32	-0.19	-0.45**	0.37*	-0.41*	1.00
LV	0.55**	0.55**	0.26	0.09	0.49**	0.62**	0.33	0.51**	0.22	0.36*	0.35*	0.08	-0.29	0.40*	-0.23

FDDT- Farinograph Dough Development Time; FG- Flour Gluten; FMTI- Farinograph Mixing Tolerance Index; FP- Flour Protein; FQN- Farinograph Quality Number; FWA- Farinograph Water Absorption; FSTAB- Farinograph Stability; GP- Grain Protein; GWG- Good Wet Gluten; MGA- Mixograph absorption; MGM- Mixograph Mix time; MGPT- Mixograph Peak time; TWG- Total Wet Gluten; VDES- Dough Expansion height from Vacuum Dough Expansion System (VDES); WV- Weight Value from hybrid SDS-SRC Sedimentation test

** and * indicate significance at P = 0.01 and 0.05, respectively

Table A.6 Bivariate correlation coefficients (r) between the variables of grain and flour quality, dough mixing and quality and baked loaf characteristics for HRW wheat cultivars grown in Aurora and Onida growing locations. The data from both the growing locations was pooled to compute the correlations (N=48)

	GP	FP	FG	TWG	GWG	WAC	WV	MLA	MLS	VDEH	SLV
GP	1.00										
FP	0.84**	1.00									
FG	0.84**	0.88**	1.00								
TWG	0.75**	0.80**	0.89**	1.00							
GWG	0.78**	0.81**	0.80**	0.71**	1.00						
WAC	0.65**	0.71**	0.75**	0.95**	0.63**	1.00					
WV	0.40**	0.51**	0.39**	0.25	0.67**	0.23	1.00				
MLA	0.30	0.43**	0.40*	0.47**	0.40*	0.49**	0.18	1.00			
MLS	0.29	0.23	0.33*	0.26	0.41**	0.24	0.40*	0.17	1.00		
VDEH	0.20	0.20	0.42**	0.52**	0.32*	0.49**	0.03	0.24	0.10	1.00	
SLV	0.25	0.27	0.26	0.17	0.39**	0.05	0.37*	-0.04	0.26	0.02	1.00

FG- Flour Gluten; FP- Flour Protein; GP- Grain Protein; GWG- Good Wet Gluten; MLA- Mixolab Absorption; MLS- Mixolab Stability; SLV- Specific Loaf Volume; TWG- Total Wet Gluten; VDEH- Dough Expansion height from Vacuum Dough Expansion System (VDES); WAC- Water Absorption Capacity; WV- Weight Value from hybrid SDS-SRC Sedimentation test

** and * indicate significance at P = 0.01 and 0.05, respectively

Table A.7 Bivariate correlation coefficients (r) between the variables of grain and flour quality, dough mixing and quality and baked loaf characteristics for HRW wheat cultivars grown at Onida growing location (N-24)

	GP	FP	FG	TWG	GWG	WAC	WV	MLA	MLPT	MLS	VDEH	SLV
GP	1.00											
FP	0.85**	1.00										
FG	0.73**	0.68**	1.00									
TWG	0.56**	0.58**	0.70**	1.00								
GWG	0.62**	0.71**	0.51*	0.37	1.00							
WAC	0.38	0.44*	0.45*	0.93**	0.22	1.00						
WV	0.16	0.43*	0.12	-0.02	0.74**	-0.05	1.00					
MLA	0.11	0.02	0.03	0.03	0.18	-0.01	0.03	1.00				
MLPT	-0.17	-0.07	-0.10	0.03	-.003	0.07	0.01	0.60*	1.00			
MLS	-0.05	0.05	0.21	0.06	0.21	-0.08	0.45	-0.05	0.25	1.00		
VDEH	-0.26	-0.25	0.07	0.24	-0.17	0.30	-0.33	0.05	-0.05	-0.29	1.00	
SLV	0.11	0.33	0.11	-0.04	0.37	-0.20	0.48*	0.30	0.16	-0.07	-0.37	1.00

FG- Flour Gluten; FP- Flour Protein; GP- Grain Protein; GWG- Good Wet Gluten; MLA- Mixolab Absorption; MLPT- Mixolab Peak Time; MLS- Mixolab Stability; SLV- Specific Loaf Volume; TWG- Total Wet Gluten; VDEH- Dough Expansion height from Vacuum Dough Expansion System (VDES); WAC- Water Absorption Capacity; WV- Weight Value from hybrid SDS-SRC Sedimentation test

** and * indicate significance at P = 0.01 and 0.05, respectively.

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