

AN ABSTRACT OF THE THESIS OF

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Title: Wheat Flour Arabinoxylans in Soft Wheat End-Use Quality

Abstract approved:

Andrew S. Ross

Little is known about the effects of arabinoxylans (AX) on noodle quality. The aim of this study was to observe interrelationships between wheat flour AX, SRC tests, and noodle quality attributes, and to investigate the use of SRCs to predict cookie diameter. Cookie diameter is the most common index of overall soft wheat quality used in practice. Duplicate samples of 63 soft white wheat (*Triticum aestivum*) varieties and breeding lines grown at Corvallis in 2002 were selected to study the relationships between flour and noodle characteristics. Kernel hardness was positively correlated with starch damage, total AX and water-extractable AX (WEAX) content but negatively correlated with break flour yield. In this set of samples, despite significant correlations, the sodium carbonate and sucrose SRC tests were not considered to be reliable predictors of cookie diameter due to low numerical correlation coefficients.

A modified extraction method for WEAX-SE and WUAX-SE was optimized and reduced in scale. During method development, WUAX 1-SE and WUAX 2-SE fractions that had been treated with protease and amylase respectively were observed using SEHPLC. The equivalent fractions had been discarded in other studies. In this

study, AX was found to present in these fractions. A subset of 12 lines was used for further AX extraction. WUAX 2-SE had the highest molecular weight, followed by WUAX1-SE, and then WEAX-SE. The molecular weights of WEAX-SE ranged from approximately 411,305 and 447,282. However, molecular weight of WUAX 1-SE and WUAX 2-SE could not be specifically defined in this study. In addition, WEAX-SE contained a higher degree of substitution than WUAX 1-SE and WUAX 2-SE.

For the whole sample set, flour protein content was negatively correlated with t_{15A} cooked noodle hardness, adhesiveness and chewiness but positively correlated with springiness. At the very low flour protein contents of this sample set, protein composition, which related to lactic acid SRC, became more important for noodle texture. Both starch damage and sodium carbonate SRC were positively correlated with cooked noodle hardness and chewiness at t_0 and t_{15A} . Total AX and WUAX were positively correlated with adhesiveness at t_0 , which might result from gummy and sticky characteristics of AX.

Using the subset of 12 lines, described above, increased xylose and arabinose contents reflected overall higher AX abundance, and were related to harder kernel texture, poor milling properties. They were also related to higher water, carbonate and sucrose SRCs, and smaller cookie diameter. A/X ratios of WEAX-SE and WUAX 1-SE were positively correlated with flour yield and break flour yield. The WUAX 2-SE fraction seemed to behave different from the WEAX-SE and WUAX 1-SE fractions. The relationships between A/G ratio, and milling characteristics and SRC were opposite to

A/X ratios for all fractions. Decreased MW and increased abundance of WUAX in this sample set was related to poorer milling characteristics. There appeared to be no direct systematic relationships between AX and cooked noodle texture parameters in this study. However, AX content appeared to affect noodle texture indirectly, mediated through the effects of AX on kernel hardness, milling properties, starch damage, reduced FSV, and hence harder noodle texture. Kernel hardness index, flour yield, break flour yield and t_{15W} cooked noodle hardness were able to be predicted with some confidence using stepwise multiple regressions that used selected parameters from the WEAX, WUAX 1-SE and WUAX 2-SE fractions.

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Wheat Flour Arabinoxylans in Soft Wheat End-Use Quality

By
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I understand that my thesis will become part of the permanent collection of Oregon State University libraries. My signature below authorizes release of my thesis to any reader upon request.

Sunida Asawaprecha, Author

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TABLE OF CONTENTS

	<u>Page</u>
1 Chapter 1 Introduction.....	1
2 Chapter 2 Literature review.....	4
2.1 Wheat.....	4
2.2 Dry milling process.....	7
2.3 Soft wheat quality.....	8
2.3.1 Factors affecting soft wheat quality	9
2.3.1.1 Proteins.....	9
2.3.1.2 Starch.....	11
2.3.1.3 Lipids.....	12
2.3.1.4 Arabinoxylans (AX) structure and function.....	13
2.3.1.5 Xylanases.....	17
2.4 Solvent retention capacity.....	18
2.5 Noodles.....	19
2.6 Noodle processing.....	22
2.7 Noodle quality evaluations.....	25
2.7.1 Appearance.....	25
2.7.2 Texture.....	27
3 Chapter 3 Materials and methods.....	30
3.1 Material.....	30
3.1.1 Soft white wheat (SWW).....	30
3.1.2 Chemicals and ingredients.....	30
3.2 Methods.....	30
3.2.1 Single Kernel Characterization System.....	30
3.2.2 Milling.....	31
3.2.3 Moisture content.....	32
3.2.4 Flour ash content.....	33
3.2.5 Flour protein content.....	33
3.2.6 Flour swelling volume (FSV).....	33

TABLE OF CONTENTS (Continued)

	<u>Page</u>
3.2 Methods (Continued).....	30
3.2.7 Rapid Visco Analyzer (RVA).....	34
3.2.8 Cookie making.....	34
3.2.9 Solvent Retention Capacity test (SRC).....	35
3.2.10 Starch damage.....	36
3.2.11 AX content determination	38
3.2.12 Extraction procedure of AX.....	39
3.2.13 Soluble starch determination.....	41
3.2.14 Molecular weight distribution of AX using Size Exclusion HPLC (SEHPLC).....	41
3.2.15 Sugar analysis.....	42
3.2.16 Chinese white salted raw noodle making	43
3.2.16.1 Method of optimum water absorption determination.....	43
3.2.16.2 Production of Chinese white salted raw noodles.....	44
3.2.17 Noodle evaluation.....	45
3.2.17.1 Optimum cooking time.....	45
3.2.17.2 Water content of noodle	46
3.2.17.3 Cooking loss	46
3.2.17.4 Cooking yield	47
3.2.17.5 Texture evaluation	47
3.2.18 Statistical analysis.....	49
4 Chapter 4 Results and Discussion: Kernel, Milling and Analytical characteristics.....	50
5 Chapter 5 Results and Discussion: Extraction of AX fractions.....	58
6 Chapter 6 Results and Discussion: Chinese white salted raw noodles.....	71
7 Chapter 7 Results and Discussion: AX, Kernel and Milling Characteristics, and Noodles.....	83
8 Chapter 8 Conclusion.....	107
Bibliography	113

TABLE OF CONTENTS (Continued)

	<u>Page</u>
Appendices.....	120
Appendix A Pedigrees of the sixty three advance soft white wheat breeding lines used in this study. Each line had two replicates, with the exception of ORH011273.....	121
Appendix B Pedigrees of the subset of twelve soft white wheat breeding lines chosen for extraction of AX fractions. Six were chosen on the basis of having the highest total AX contents, and six, as they had the lowest. Each line was replicated twice.....	123

LIST OF FIGURES

<u>Figure</u>	<u>Page</u>
2.1 Longitudinal section of wheat kernel illustrating major anatomical parts.....	6
2.2 Representative structure of AX in wheat flour. n = finite number of polymer units; * = position at which substitution occurs.....	14
2.3 Flow diagram of processing steps used to manufacture six different styles of Asian noodles.....	24
2.4 Schematic diagram representing to CIE L*a*b* color space. L* values of 0 = pure black and 100 = pure white; + a* value (0°) = redness and -a* value (180°) = greenness; + b* value (90°) = yellowness and -b* value (270°) = blueness.....	26
2.5 Texture profile analysis time/force curve for white salted noodles obtained using a TA.XT plus®	27
3.1 Flow scheme of the extraction procedure used for isolation of WUAX and WEAX.....	40
4.1 Scatter plot of the relationship between sodium carbonate SRC and cookie diameter for 125 soft white wheat samples.....	56
4.2 Scatter plot of the relationship between sucrose SRC and cookie diameter for 125 soft white wheat samples.....	56
5.1 Flow scheme of extraction procedure used for isolation of WUAX and WEAX from 50 g of a commercial all-purpose flour.....	59
5.2 Gel permeation profiles of the WEAX fraction obtained from of a commercial all-purpose flour used during method development, before (---), and after (—) dialysis.....	60
5.3 Gel permeation profiles of WEAX (a), WUAX 1 (b) and WUAX 2 (c) fractions extracted from the 10 g (—) and 50 g (- -) extraction procedures using a commercial all-purpose flour.....	62
5.4 Gel permeation profiles of WEAX (····), WUAX 1 (---) and WUAX2 (—) fractions obtained from a commercial all-purpose flour.....	63

LIST OF FIGURES (Continued)

<u>Figure</u>	<u>Page</u>
5.5 Gel permeation profiles of (a) WEAX, (b) WUAX 1 and (c) WUAX 2 fractions obtained from flour samples with highest (ORH010377; —) and lowest (ORH010085; ---) AX content.....	64
5.6 MW standard curve derived from $K_{av} = (V_e - V_0) / (V_t - V_0)$ using Shodex P-82 pullulan standards with molecular weights of 40.4×10^4 , 21.2×10^4 , 11.2×10^4 , 4.73×10^4 , 2.28×10^4 , 1.18×10^4 and 0.59×10^4 . V_e = elution volume, V_0 = void volume and V_t = total bed volume.....	65
7.1 Comparison of wet chemical WEAX contents, and WEAX-SE contents. WEAX-SE contents were calculated from arabinose and xylose contents of the WEAX-SE fraction $[0.88 \times (\text{arabinose} + \text{xylose})]$ of the 6 breeding lines with highest and the 6 breeding lines with lowest AX contents as determined by the wet chemical method.....	83
7.2 Comparison of wet chemical WUAX contents, and WUAX 1-SE (a), WUAX 2-SE (b) and total WUAX-SE (c) content s. WUAX-SE contents were calculated from arabinose and xylose contents of WUAX-SE fraction $[0.88 \times (\text{arabinose} + \text{xylose})]$ of the 6 breeding lines with highest and the 6 breeding lines with lowest AX contents as determined by the wet chemical method.....	86
7.3 The relationships between actual and predicted values of hardness index (a), % flour yield (b) and % break flour yield (c) using stepwise linear regression of AX parameters obtained from the 6 breeding lines with highest, and the 6 breeding lines with lowest AX contents as determined by the wet chemical method	104
7.4 The relationship between actual and predicted values of hardness at t_{15w} using stepwise linear regression of AX parameters obtained from the 6 breeding lines with highest and the 6 breeding lines with lowest AX content as determined by the wet chemical method	106

LIST OF TABLES

<u>Table</u>	<u>Page</u>
2.1 Chemical composition of wheat endosperm.....	4
2.2 Physical and sensory definitions of the mechanical parameters of food texture..	28
4.1 Means and standard deviation values of hardness index, and kernel weight and diameter obtained from Single Kernel Characterization System	51
4.2 Means, standard deviations and ranges of wheat milling characteristics, and flour starch damage, ash, protein and AX contents of 63 soft white wheat breeding lines.....	52
4.3 Correlation coefficients between kernel texture, milling characteristics, solvent retention capacity tests, cookie diameter, and flour AX, starch damage and protein contents of 63 soft white wheat breeding lines.....	53
5.1 Monosaccharide composition and molecular weight (MW) data of WEAX extracted from the 6 breeding lines with highest AX content and the 6 breeding lines with lowest AX content as determined by wet chemical method.....	67
5.2 Monosaccharide composition and molecular weight (MW) data of WUAX 1 extracted from the 6 breeding lines with highest AX content and the 6 breeding lines with lowest AX content as determined by wet chemical method	68
5.3 Monosaccharide composition and molecular weight (MW) data of WUAX 2 extracted from the 6 breeding lines with highest AX content and the 6 breeding lines with lowest AX content as determined by wet chemical method	69
6.1 Correlation coefficients between flour protein, starch damage, FSV and RVA peak viscosity, and noodle cooking characteristics of 63 soft white wheat breeding lines.....	74
6.2 Correlation coefficients among TPA texture parameter of noodles cooked for a fixed cooking time of 5 min using 63 soft white wheat breeding lines	75
6.3 Means, ranges, and standard deviations of noodle texture parameters at fixed (5 min) and optimum (opt) cooking times from 63 soft white wheat breeding lines	76

LIST OF TABLES (Continued)

<u>Table</u>	<u>Page</u>
6.4 F-values from analysis of variance for noodle texture parameters comparing the fixed (5 min) and optimum cooking times using 63 soft white wheat breeding lines	78
6.5 Correlation coefficients between flour protein, AX contents, FSV, RVA peak viscosity, solvent retention capacity tests and noodle texture parameters at a fixed cooking time of 5min, using 63 soft white wheat breeding lines.....	82
7.1 Means, standard deviations and ranges of WEAX, WUAX 1 and WUAX 2 contents from size exclusion chromatography HPLC (SEHPLC), and total AX, WEAX and WUAX contents from wet chemical method of the 6 breeding lines with highest and the 6 breeding lines with lowest AX content as determined by wet chemical method.....	84
7.2 Correlation coefficients between xylose and arabinose contents of WEAX, WUAX 1 and WUAX 2, kernel texture, milling characteristics, solvent retention capacity tests, cookie diameter, and noodle texture parameters from the 6 breeding lines with highest and the 6 breeding lines with lowest AX content as determined by wet chemical method.....	89
7.3 Correlation coefficients between AX contents and A/X Ratio of WEAX, WUAX 1 and WUAX 2, kernel texture, milling characteristics, solvent retention capacity tests, cookie diameter, and noodle texture parameters from the 6 breeding lines with highest and the 6 breeding lines with lowest AX content as determined by wet chemical method.....	91
7.4 Correlation coefficients between AG contents and A/G ratio of WEAX, WUAX 1 and WUAX 2, kernel texture, milling characteristics, solvent retention capacity tests, cookie diameter, and noodle texture parameters from the 6 breeding lines with highest and the 6 breeding lines with lowest AX content as determined by wet chemical method.....	97
7.5 Correlation coefficients between size Exclusion Chromatography parameters of WUAX 1 and WUAX 2, kernel texture, milling characteristics, solvent retention capacity tests, cookie diameter, and noodle texture parameters from the 6 breeding lines with highest and the 6 breeding lines with lowest AX content as determined by wet chemical method	100

LIST OF TABLES (Continued)

<u>Table</u>	<u>Page</u>
7.6 Coefficients of determination (R^2) calculated by stepwise multiple linear regressions for kernel hardness, flour milling characteristics and cooked noodle texture at t_{15W} using AX parameters of WEAX, WUAX 1, and WUAX 2 obtained from the 6 breeding lines with highest and the 6 breeding lines with lowest AX content as determined by wet chemical method.....	103

Wheat Flour Arabinoxylans in Soft Wheat End-Use Quality

Chapter 1

Introduction

Wheat (*Triticum aestivum*) is the principle food grain of the world and is one of the USA's major exported products. There are six major classes of U.S. wheat: hard red winter, hard red spring, soft red winter, soft white, hard white, and durum. Almost half of wheat that is produced in the USA is exported. Exports go to many countries and many of these countries require high quality wheat. The US wheat industry is developing new varieties to meet market requirements of these countries. The goal is to become more competitive against other exporting countries such as Australia, Canada, the European Union (EU) and Argentina.

About one third of all US wheat is soft-grained. Of the soft wheat produced in the eastern growing states, about 50% is exported each year. In the Pacific Northwest, over 85% of the approximately 6 to 10 million ton crop were exported. One product produced from US soft wheat in Asia is noodles. Unfortunately, this market is dominated by Australia. Only 5% of US soft wheat is currently used for noodle making. The USA would like to increase the amount of soft wheat in this application. The flour required for noodles is generally white and finely granulated, has rapid water uptake, and low enzymatic activity (Hoseney et al, 1988). The basic formulation for noodles is wheat flour, salt, and water. For specific types of noodles, alkaline salts such as sodium and potassium carbonates can be used instead of, or with sodium

chloride. Both texture and appearance are significant factors for noodle quality but vary by noodle type. For example, udon noodles that are consumed in Japan should be bright and creamy, have a smooth surface, and have soft texture with springy resistance when chewed (Toyokawa, 1989a).

The most important factor in determining the quality of these end-use products is wheat flour. Wheat flour comes from the endosperm part of the wheat kernel and contains about 6 - 20% protein, 60 - 75% starch, 1.5-2.0% lipids, and 2-3% arabinoxylans (AX) (Hoseney, 1986; Halverson and Zeleny, 1988). Flour protein content and starch composition are the major factors affecting noodle texture. AX, the major component of nonstarch polysaccharides in flour, play an important role in quality of many end products. This occurs primarily through their effects on water holding capacity. AX consist of a xylose backbone with branches of one or two arabinosyl units. The molecules are considered linear and the commonly accepted model for conformation of AX is an extended rod. However, a recent study showed that AX may behave as random coil (Dervilly et al, 2000). Wheat AX are generally divided into two types: water-extractable arabinoxylans (WEAX) and water-unextractable arabinoxylans (WUAX). The unique properties of AX are that they are readily dispersible in water, form highly viscous solutions, and have high water holding capacities. After addition of oxidizing agents such as H_2O_2 and peroxidase, WEAX can also form solid gels by dimerization of ferulic acid residues linked to the O5 atom of an arabinosyl substituent (D' Appolonia and Kim, 1976).

WEAX increase loaf volume and give a fine structure to bread (Jeleca and Hlynka, 1971; Michniewicz et al, 1992). Notably, most studies deal with the quality of wheat flour for bread, cakes, and cookies, not noodles. As a result, there is limited knowledge of the effect of AX on noodle quality. Ingelbrecht et al (2001b) found low levels of AX loss in pasta cooking water, even after a high dose of endoxylanase was used. They also found that there was no change in arabinose substitution of WEAX during pasta processing.

There are many methods used to rapidly monitor the quality of wheat and characterize wheat flour or meal. Solvent retention capacity (SRC) tests were recently developed to monitor the ability of flour to retain a set of solvents. These solvent solutions are water, and aqueous solutions of 5% sodium carbonate, 50% sucrose, and 5% lactic acid. SRC tests have been shown to relate to specific flour components. This method is now used for evaluating multiple aspects of wheat quality: AX content, starch damage, gluten strength, and general water retention (Guttieri, 2001). SRC tests are proposed as useful and rapid methods of screening wheat samples for end-use products such as cookies, crackers, sponge and noodles.

The objectives of this study are to study the complex interrelationships between AX content and composition, molecular weight distribution of wheat flour AX, SRC tests, and noodle processing and quality attributes.

Chapter 2

Literature review

2.1 Wheat

Wheat (*Triticum aestivum*) is one of the major components in many diets. Wheat is widely grown because of its agronomic adaptability and ease of storage. It also produces a range of tasty and enjoyable foods and provides health benefits (Orth and Shellenberger, 1988).

A schematic diagram of a wheat kernel is shown in Figure 2.1. Wheat kernels weigh about 35 mg and are about 8 mm in length. However, the size of the kernel can vary depending on the cultivar and the location of the kernel in wheat head or spike (Hoseney, 1986). The kernel is composed of germ and a central endosperm surrounded by the pericarp. The major chemical components of the endosperm are protein, starch and lipid as shown in Table 2.1.

Table 2.1 Chemical composition of wheat endosperm (Hoseney, 1986; Halverson and Zeleny, 1988)

Chemical compositions	% Dry weight basis
Protein	6-20
Starch	60-75
Lipids	1.5-2.0

The aleurone layer is the outer layer of endosperm and contains high levels of ash, protein, polyphenol oxidase, total phosphorus, phytate phosphorus, fat, and niacin.

The aleurone layer is removed during the milling process along with the nucellar epidermis, seed coat and pericarp. These four components are called bran by flour millers.

Wheat can be divided by the texture of the kernel into two types: hard and soft.

Because of strong bonding between protein and starch, hard wheat requires more grinding energy than soft wheat to reduce the size of flour particles. Moreover, the starch granules of hard wheat are often broken before the rupture of protein-starch bonding resulting in a large amount of starch damage (Hoseney, 1986). The increased starch damage leads to increased water absorption of flour and is related to the water and sodium carbonate SRC tests (Guttieri, 2001).

Puroindolines are proteins associated with polar lipids and endosperm membranes.

Puroindoline gene has two main alleles (*pin-a* and *pin-b*). This gene also affects the hardness of wheat (Bettge and Morris, 2000). A mutation in *pin-b*, or the absence of *pin-a* results in hard wheat.

Besides the difference in hardness, kernel appearance is another important characteristic. Vitreousness results from the absence of air spaces in the kernel and has been associated with hardness and high protein content whereas opaqueness has been associated with softness and low protein (Hoseney, 1986).

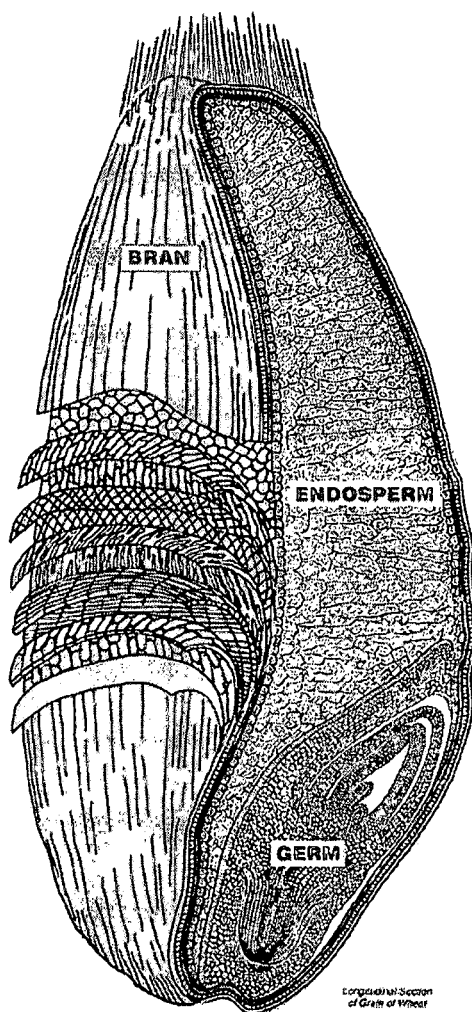


Figure 2.1 Longitudinal section of wheat kernel illustrating major anatomical parts (Permission from Kansas Wheat Commission).

2.2 Dry milling process

The dry milling process involves removal of pericarp, the seed coat, the nucellar epidermis, the aleurone layer and germ and makes the remaining endosperm as pure as possible. It is also used for reducing the particle size of the endosperm until it is sufficiently fine to be called flour. The germ is rich in oil and this oil can easily become rancid. This is why it is important to remove the germ during this process. After the milling process, the nutritional value of wheat flour is decreased because protein, vitamin B, minerals, and fat are removed with the bran and germ.

Wheat is cleaned and tempered before milling (Figure 2.2). Tempering involves adding water to dry grain and allowing it to rest for a period of time (about 8-24 hours) before it is milled. Usually, hard wheat requires a longer temper time and higher tempering moisture than soft wheat. Tempering toughens the bran and makes it resist breaking into small pieces during milling. Tempering also softens the endosperm and makes it easier to grind.

The next step is the grinding of grain. Flour mills have two distinct roll systems: break and reduction. Each set of rolls in a break system has a pair of corrugated rolls that rotate in the opposite directions. They are used to take out bran and germ from the endosperm. After that, broken endosperm passes through sifters and purifiers to remove light bran particles. The coarse endosperm is sent to reduction rolls to reduce particle size to flour fineness. Each set of rollers in a reduction system has a pair of smooth rolls that also rotate in the opposite directions. The ground endosperm is

passed through a purifier again to remove the last remaining particles of bran and germ. Generally, soft wheat flour is harder to sift than hard wheat flour because the small particles of soft wheat flour tend to interact with each other and are more difficult to pass through a flour sieve. This can be a major problem in soft wheat milling.

2.3 Soft wheat quality

Soft wheat is generally used for cookies, cakes, crackers and noodles. Cookies, cakes and crackers have a better appearance and better eating quality when they are made from soft instead of hard wheat flour. Hard wheat flour is usually used for bread and mostly contains higher percentage of starch damage than soft wheat. As a result, it is considered less suitable for cookies and noodles (Hoseney, 1986).

Soft wheats are classified depending on time of sowing and color of their seed coat. Winter and spring types are characterized based on time of sowing. Winter wheat varieties are sown in the fall whereas; spring wheats are sown in the spring. Both types are harvested in late summer. Winter wheat production represents 70-80% of total U.S. production. There are two seed coat colors in wheat, red and white, depending on the presence or absence of pigmentation in the seed coat. Soft red winter (SRW) wheat, accounting for 15 to 20% of total US wheat production, is grown primarily in states along the Mississippi River and in the Eastern States. Flour produced from SRW is used in the United States for cakes, cookies, and crackers. Soft white wheat (SFW) makes up approximately 10 to 15% of total production of US

wheat and is grown in Washington, Oregon, Idaho, Michigan, and New York. Its flour is used for crackers, cereals, and steamed breads and noodle products (Anonymous, 2000).

Cookie diameter is the most widely used index of overall soft wheat quality. Constant references to cookie diameter were made throughout the literature review and results and discussion. This is a result of the centrality of cookie diameter to soft wheat quality determinations.

2.3.1 Factors affecting soft wheat quality

2.3.1.1 Proteins

Protein content of wheat ranges from 6 to 20% depending on variety and growing condition (Halverson and Zeleny, 1988). Dry soil during crop growth tends to give wheat with high protein content, whereas wheat with low protein content results from adequate soil moisture during the period of kernel development. Each end product requires a different optimum quantity of flour protein. Lower-protein soft white wheat (7-10%) tends to be best for cake and cookie flours while high protein soft (10-14%) are used for noodles, Middle Eastern type flat breads and other foods.

For cookie making, soft wheat flours that contain lower protein content than hard wheat flour spread at faster rate during baking, giving large cookie diameters, and cookies that are thin and tender (Miller and Hosney, 1997). Protein content of wheat

has a positive relationship with hardness of noodles, but has a negative relationship with noodle brightness (Park et al, 2003).

Wheat flour proteins are commonly classified by their solubility into four types: albumins soluble in water, globulins soluble in dilute salt solution, gliadins soluble in 70% ethyl alcohol, and glutenins soluble in dilute acids and bases (Hoseney, 1986). The endosperm storage proteins that contribute to dough characteristics are the gliadins and glutenins. Gliadins are monomeric proteins with compact conformation. They form intra-molecular disulfide bonds and contribute to the viscous component of dough viscoelasticity. Glutenins, polymeric proteins, are extended in conformation and form the elastic component of visco-elastic wheat flour doughs through inter-molecular disulfide bonding between polypeptide subunits. High molecular weight glutenin subunit (HMWGS) alleles, coded at loci designated *Glu-A1*, *Glu-B1* and *Glu-D1*, have a large effect on the quality of wheat flour. Wesley et al (1999) showed that HMWGS 5+10, and specific LMWGS and gliadin were responsible for good protein quality for both breadmaking and noodlemaking. Weaker dough characteristics are commonly associated with best cookie quality. However, in one study, Huebner et al (1999) found that stronger wheats with the HMWGS pair 5+10 were able to make cookies with larger diameters than with wheat with the HMWGS pair 2+12, suggesting that HMWGS composition is not a primary factor in cookie quality.

2.3.1.2 Starch

Starch is commonly between 60% and 75% of dry weight of the wheat kernel (Hoseney, 1986). Starch exists in the endosperm as discrete granules that are partially crystalline and exhibit birefringence under polarized light. Soft wheat varieties generally have higher starch content than hard wheat varieties as a function of lower protein content (Lineback and Rasper, 1988). According to Donelson and Gaines (1998), soft wheat flour produced larger sugar-snap cookies diameter than hard wheat flour because of lower amount of starch damage.

The major components of the starch granule are amylose and amylopectin. Amylose is a linear polymer of α - (1 \rightarrow 4)-linked D-glucopyranose units with molecular weights ranging from 10^5 to 10^6 . Amylopectin is a highly branched polymer with many shorter α - (1 \rightarrow 4)-linked D-glucopyranose units and α - (1 \rightarrow 6) at branch points. Amylopectin has molecular weight typically ranging from 10^8 to 10^9 (Whistler and Daniel, 1984).

Swelling of starch granules in baked products is controlled by temperature, the amount of water available to the granule and by the formulation and other ingredients such as sugar and shortening (Lineback and Rasper, 1988). With increasing of temperature, starch begins to gelatinize and compete with other ingredients for available water.

With the large amounts of sucrose or shortening present in cookie dough, starch granules require more water for gelatinization. This results in the restriction of starch gelatinization (Hoseney, 1986).

Toyokawa et al (1989b) showed that high ratio of amylose to amylopectin resulted in poor texture of Japanese noodle. High levels of amylose were associated with the decrease of water holding capacity of flour at 75°C, decreased elasticity of cooked noodles, and increased firmness of cooked noodles. The ideal textural attributes for soft-bite noodles like udon, are related to the relative increase in the amount of amylopectin. Amylopectin is a highly branched molecule that swells more on hydration than amylose, leading to a more diffuse and softer gel structure that gives a unique soft and elastic mouthfeel.

2.3.1.3 Lipids

Wheat usually contains 1.5-2.0% lipids. Approximately, 64-71% of the fatty acids in these lipids are polyunsaturated. Lipids differ in each part of the wheat kernel. Whole wheat contains about 70% nonpolar lipids, 20% glycolipids, and 10% phospholipid (Hoseney, 1986). The germ contains the greatest amount of phospholipids. Bran contains more phospholipids than glycolipid. Endosperm generally contains more glycolipids than phospholipids. In starchy endosperm, lipids can be divided into nonstarch lipids and starch lipids (Hoseney, 1986). Nonstarch lipids are commonly composed of 60% nonpolar lipids, 25% glycolipids, and 15% phospholipids while starch lipids are composed of 9% nonpolar lipids, 5% glycolipids, and 86% phospholipids (Hoseney, 1986).

Lipids are more susceptible to deterioration than other wheat grain components that contribute to baking quality (Morrison, 1988). Postharvest storage causes lipid

hydrolysis, lipid oxidation, release of volatile odor substances, particularly free fatty acids (FFA), so this affects the quality of wheat grain. It was found that cooling the grain to 8°C could reduce the deterioration of gluten and baking quality and the breakdown of polar lipids. At moisture level below 8%, autoxidation of lipids can occur and result in formation of off-flavors and loss of baking quality (Morrison, 1988).

Polar lipids have been shown to provide increased benefit for loaf volume of bread whereas nonpolar lipids gave negative effects (DeStefanis and Ponte, 1976). Cole et al (1960) reported that cookies became darker brown in color and had small diameter when baked from flours which had been extracted with water-saturated n-butanol. Cookie diameter became larger again when free lipids were restored. In noodles, it was found that surface firmness of cooked noodles decreased when free lipids were removed from hard wheat and soft wheat flours but noodles become firmer when free lipids were restored (Rho et al, 1989).

2.3.1.4 Arabinoxylans (AX) structure and function

The major polysaccharide components of wheat endosperm cell wall are AX, cellulose and β -glucans. AX make up approximately 88% of wheat endosperm cell wall material (Lineback and Rasper, 1988). AX are sometimes referred to as pentosans although pentosans also contain arabinogalactan as a minor component (Faurot et al, 1995). The remarkable property of AX is the ability to readily absorb water and form highly viscous solutions. Therefore, they are one of the factors affecting quality of end

products. It has also been found that endogenous secretions in the human digest tract cannot digest them, so they are also regarded as dietary fiber (Lu et al, 2000; Ingelbrecht et al, 2001a).

Wheat flour contains about 2-3% of AX (Jeleca and Hlynka, 1971). AX are divided into two types: water-extractable AX (WEAX) and water-unextractable AX (WUAX).

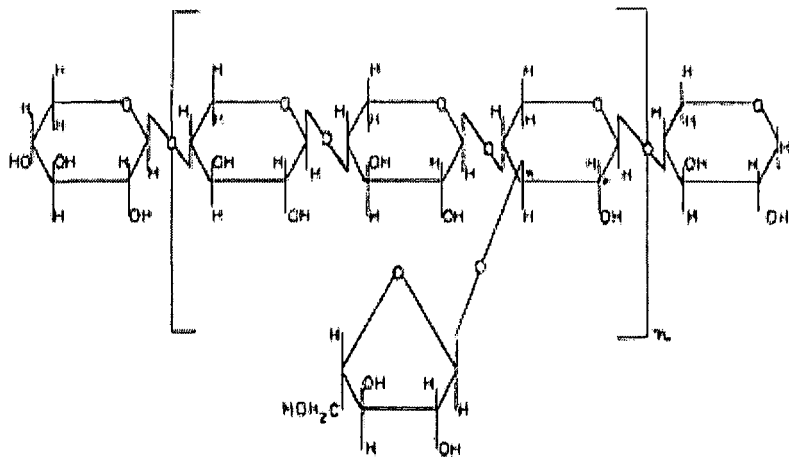


Figure 2.2 Representative structure of AX in wheat flour. n = finite number of polymer units; * = position at which substitution occurs (Hoseney, 1986).

AX consist of straight chains of anhydro-D-xylopyranosyl residues linked together by β -(1 \rightarrow 4) glycosidic bonds (Figure 2.3). The major substituents are α -L-arabinofuranosyl residues, which attach to 2- or 3- positions, or both the 2- and 3- position of xylosyl residues (Lineback and Rasper, 1988). ^1H -nuclear magnetic resonance (NMR) spectroscopy was used to study the structure of AX. It showed that AX chains contained unsubstituted-, mono- and disubstituted D-xylose residues (Cleemput et al, 1993). Ferulic acid is a minor substituent. It is generally linked to O5

atom of an arabinosyl substituent. It is related to gel forming by dimerization of ferulic acid when H_2O_2 and peroxidase are added (Ciacco and D'Appolonia, 1982; Michniewicz et al, 1990). Without the substitution on arabinose residues, AX exist as a relatively flexible and fully extended twisted ribbon (Lineback and Rasper, 1988). The substitutions of arabinosyl residues make for more extended highly asymmetric rod conformations and also affect the solubility of AX. Recently, Dervilly et al (2000) using multiangle laser light scattering, found that structures of AX behaved as random coils instead of the previously reported extended rodlike conformation.

Molecular weights of wheat endosperm WEAX and WUAX range from 231,000-315,000, and 380,000-570,000, respectively (Cui, 2001). In addition, Courtin et al (1999) found the molecular weight of WEAX extracted from durum wheat flour was 501,000 to 530,000. WEAX and WUAX have very similar structures. However, WUAX from Canadian red spring, red winter, soft white spring, and durum wheats have been shown to have a higher degree of substitution with arabinosyl residues than WEAX (Michniewicz et al, 1990; Izydorczyk et al, 1991). Generally, the L-arabinose to D-xylose ratio of AX varied between 0.51-0.71 when analyzed by gas-liquid chromatography (Cleemput et al, 1993; Dervilly et al, 2000; Izydorczyk et al, 1991). Higher ratio of arabinose to xylose indicates a higher degree of arabinose substitution. It was found that when the ratio of L-arabinose to D-xylose decreased, the numbers of un-substituted xylose residues increased, di-substituted xylose residues decreased, and the numbers of mono-substituted xylose remained constant (Delcour et al, 1999).

WEAX, accounts for 0.5-0.8% (w/w) of wheat flour (Jelega and Hlynka, 1971).

WEAX make extremely viscous solutions, whereas WUAX have the ability to highly hydrate in water without actually going into solution (Jelega and Hlynka, 1971; Dervilley et al, 2000). It was reported that the supplementation of flour with WEAX is more effective in increasing the water binding properties of wheat flour than the increase of starch damage (Michniewicz et al, 1992).

WEAX are considered to be beneficial in breadmaking. WEAX had a positive effect in loaf volume and bread quality when 2% WEAX were added, whereas WUAX were detrimental for breadmaking when added at the same level (Faurot et al, 1995; Courtin and Delcour, 2001). WEAX also gave softer crumb structure and finer grain of bread crumb (Michniewicz et al, 1992; Faurot et al, 1995; Courtin and Delcour, 1998). Moreover, it was reported that WEAX retard the rate of retrogradation of starch gels and the staling rate of bread by forming H-bond with amylose and also gave high moisture content to the system to reduce the firmness of bread (D'Applonia and Kim, 1976; Michniewicz et al, 1992).

WUAX are normally insoluble in water and they become soluble when treated with alkaline or enzyme (Courtin et al, 1999). WUAX cannot directly change the viscosity of dough but they trap a large amount of water and contribute to the water distribution in the dough (Courtin et al, 1999) and so affect dough viscosity indirectly. When WUAX are hydrolyzed, they lose their strong water-holding capacity (Courtin and Delcour, 2001). Because of the detrimental effect of WUAX on breadmaking, it was

reported that the removal of WUAX could improve loaf volume of bread (Courtin et al, 1999).

AX are highly hydrophilic and are correlated with smaller cookie diameter (Guttieri et al, 2001). Bettge and Morris (2000) reported a negative relationship between cookie diameter and WEAX, total flour AX, and flour protein in soft wheat flour. If the flour is less hydrophilic, then more water is available to the sugar to form syrup. Reduced concentration of sugar in the dough syrup leads to decreased dough viscosity during baking. Dough viscosity appears to control cookie-spread rate and affect final cookie diameter. The resulting dough spreads farther, producing larger diameter (Guttieri et al, 2001).

2.3.1.5 Xylanases

Endoxylanase- β -1, 4-xylanases (EC 3.2.1.8), also referred to as endoxylanases or xylanases, are widely use in bread making applications.

Xylanases are able to hydrolyze both WUAX and WEAX. Xylanases convert WUAX into medium and high molecular weight, enzyme-solubilized, AX (S-AX). S-AX are found in the water phase (Courtin and Delcour, 2001). Xylanase can also degrade WEAX to lower molecular weights (Courtin et al, 1999).

Xylanases are widely used to improve the rheological properties of dough in breadmaking. Xylanases degrade WUAX into S-AX. The high amount of WEAX and

S-AX in water phase increase the viscosity of dough aqueous phase, (Courtin and Delcour, 2001). It was found that xylanase provided beneficial effect on bread loaf volume by decreasing the amount of WUAX and increasing the amount of WEAX and S-AX (Courtin et al, 1999). However, excess amounts of xylanase can make doughs sticky. Excess stickiness can be remedied using oxidative enzymes like glucose oxidase or peroxidase that appear to act by changing water distribution and increasing water binding capacity in the dough (Hilhorst et al, 2002).

2.4 Solvent retention capacity

Traditionally, two AACC approved methods were used to predict baking quality of soft wheat flours. These included sugar-snap cookie method, 10-52 and the alkaline water retention capacity (AWRC) method, 56-10.

AWRC measures water holding capacity of flour under alkaline conditions and can be used to predict cookie baking quality of flour (Miller and Hosene, 1997). Cookie diameter was highly and negatively correlated with AWRC of the soft and hard wheat flour (Donelson and Gaines, 1998).

More recently new methods to predict cookie diameter were introduced. Solvent retention capacity (SRC) method was developed by Slade and Levine and approved by the Soft Wheat Flour Committee of the Approved Methods Committee of AACC for evaluating soft wheat quality (Gaines, 2000). The tests are a modification and extension of the AWRC.

SRC is the weight of solvent held by flour after centrifugation and expressed as a percent of flour weight (Guttieri, 2001). The SRC method uses four solvents for predicting the cookie baking performance (Gaines, 2000). The solvents are water, and aqueous solutions of 50% sucrose, 5% sodium carbonate, and 5% lactic acid

Generally, lactic acid SRC is associated with glutenin characteristics, sodium carbonate SRC is associated with levels of damaged starch, and sucrose SRC is associated with AX and gliadin characteristics. Water SRC is influenced by all of those flour constituents (Guttieri, 2001). SRC values relate to end-use performance of different products. For example, good quality cookie flour should have $\leq 51\%$ water, $\leq 89\%$ sucrose, $\geq 87\%$ lactic acid and $\leq 64\%$ Na_2CO_3 SRC values (Gaines, 2000).

Despite the usefulness of SRC tests for preliminary screening of soft wheats, and predicting the quality of cookies, their usefulness in noodle quality prediction is unknown.

2.5 Noodles

Noodles have been consumed in Asian countries for 1,000's years. In the last few decades they have also become popular in many countries outside Asia. Within Asia, noodles are a traditional staple food and each country has specific quality preferences. For example, Japanese and Korean people like salted noodles that have a creamy white color and a soft and elastic texture. Chinese people prefer noodles that are bright

creamy white for white salted noodles and bright yellow for alkaline noodles. A firm and chewy texture is desirable in both categories of Chinese noodles.

Noodles are made from wheat flour (hard and soft wheat (*Triticum aestivum*), water and salts. Common salts (sodium chloride), alkaline salts (kan sui, a mixture of sodium and potassium carbonates), or a combination of both may be used. Alkaline noodles have a yellow color. The presence of flavonoid compounds which are relatively stable and colorless at acidic pH, but yellow at high pH, are the major factors for the yellowness of noodles (Miskelly, 1984). Alkaline noodles also provide firm and elastic texture because of the strengthening effects of adding alkaline salts on dough characteristics.

Different ingredients can influence particular characteristics for the different types of noodles. For example, to make firm textured instant fried noodles that are easy to rehydrate on cooking, guar gum and other hydrocolloids can be used. Polyphosphates can also be used to provide better mouth-feel because of their ability to absorb water on surface of noodle. Native or modified potato starches, or other equivalent starches are frequently added to premium instant fried noodle formulations to provide a springy texture and improve steaming and cooking quality (Hou and Kruk, 1998). This is a result of the capacity of the added starches to reduce gelatinization temperature. Like many natural occurring gums, AX also have the ability to absorb copious amounts of water so that they may affect texture and appearance of noodles.

Important wheat flour components that affect noodle color and texture are protein, starch, lipid, ash, yellow pigment and polyphenol oxidase. Wheat protein content is important because it is positively related to noodle hardness (Park et al, 2003). The softer Japanese salted noodles require wheats of medium protein content, whereas the harder Chinese noodles require wheat with high protein content. Protein content is also negatively related to brightness of cooked noodles (Park et al, 2003).

Starch composition profoundly affects noodle texture. Superior quality Japanese and Korean salted noodles require high starch paste peak viscosity, low gelatinization temperature, high breakdown and high swelling volume (Konik et al, 1992; Baik and Lee, 2003). The key factor is the amylose to amylopectin ratio (addressed in section 2.3.1.2). High amylose content gives a harder texture quality. Due to linear conformation of amylose, it swells less than amylopectin, leading to a harder gel structure and provides harder noodles.

Lipid content also affects noodle quality. During storage of oil coated Japanese noodles (*tenobe-somen*), unsaturated fatty acids can form complexes with amylose. This makes amylose become insoluble and inhibits swelling of gelatinized starch. This process has been called the Yaku effect, which results in increased hardness and decreased cohesiveness during cooking (Niihara et al, 1996).

High ash content can affect noodle color. This is related to higher bran contamination at higher ash levels. To produce high quality noodles, ash levels should be as low as 0.35-

0.40% (Miskelly, 1996). Also, the presence of polyphenol oxidase (PPO) can make noodles become darker (Miskelly, 1996). PPO oxidizes phenols to quinones, which are consequently changed to melanoids and provide an undesirable dark color.

Changes in AX seem to have limited effects on pasta quality. Ingelbrecht et al, (2001a) added high amounts of endoxylanase. The addition of endoxylanase had no great effect on pasta quality, particularly color and surface condition. It provided high quality pasta with increased levels of solubilized AX, which are defined as soluble fiber, and were considered to be a health benefit by the authors. However, there are still no reports about the effects of AX on noodle quality.

2.6 Noodle processing

The processing steps for machine-made noodle are shown in Figure 2.4. The first step for making noodles is mixing all ingredients to make small and uniform particle sizes of crumbly dough. Horizontal mixers are more commonly used than vertical mixers because they give better mixing results. To get smoother and less streaky dough, the dough pieces are rested for 20-40 minutes after mixing step to distribute water evenly throughout dough particles. Crumbly dough pieces are passed through a pair of sheeting rolls to form a noodle dough sheet. The dough sheet is combined and then passed through sheeting rolls again. After that, the dough sheet is rested for 30-40 minutes. The purposes of resting steps are to distribute moisture to dough, to increase the formation of disulfide bonds, to make gluten and lipids form bonds, and to make gluten relax and ready for the sheeting step (Hou, 2001).

Dough sheeting is further continued by decreasing roll gaps on a series of 4-6 pairs of rolls. At this stage, sheeting speed and sheeting ratio become the important factors to obtain the optimum reduction in dough thickness. The sheeting speed shows how fast the dough passes through the rolls. The sheeting ratio is the thickness of the dough after sheeting, divided by the thickness of the dough before sheeting (Hoseney, 1986). The maximum reduction in thickness should be no more than one-third at any reduction pass. This helps to prevent the damage of gluten network and obtain the desired thickness of a dough sheet. After sheeting, the sheet is cut into noodle strands with desirable shape by using various cutting rolls such as square or round types. Noodle strands are cut into a desirable length by a cutter.

Cooking processes, which include parboiling, boiling, steaming and frying, are different depending on what types of noodles are being processed. For example, long-life precooked Japanese udon noodles, are boiled for 10-15 minutes, rinsed and cooled in running water, stepped in dilute acidic acid water before packaging, and further steam for more than 30 seconds in a pressurized steamer (Hou and Kruk, 1998). In contrast, instant noodles are passed through a traveling net conveyor resulting in wavy noodle-strands, and then steamed and fried before packaging (Hou and Kruk, 1998).

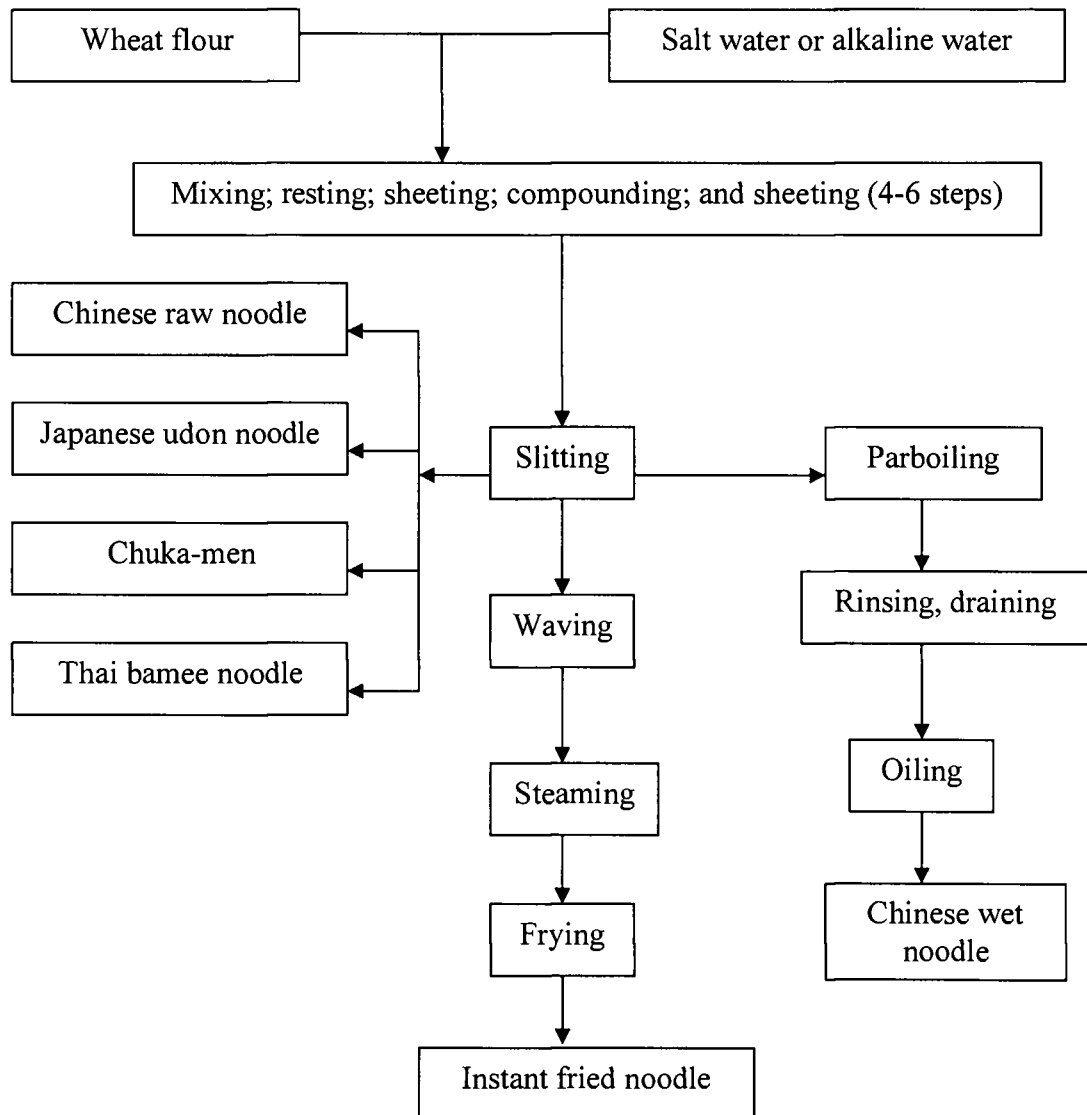


Figure 2.3 Flow diagram of processing steps used to manufacture six different styles of Asian noodles (Hou, 2001)

2.7 Noodle quality evaluations

Appearance and texture of the noodle are the main factors examined in quality evaluations. Each type of noodle has specific characteristics.

2.7.1 Appearance

Appearance is important factor for noodle quality because it is the first quality parameter perceived by customers. Usually, color measurements of raw noodles are performed immediately following noodle making, and again after 24 h of storage. Color is measured in two ways, sensory evaluations and instrumental assessments. Sensory evaluation requires many trained panelists and cannot test a large number of samples in one day. Therefore, the instrumental measurement can be more useful because of its precision and convenience. Instrumental measurements are more applicable where one needs to measure a lot of samples in one day. However, the disadvantage of instrumental measurement is that it cannot measure subtle characteristics of a sample as can be done by sensory evaluations.

Color is measured in many ways. The CIELAB (Commission Internationale de l'Eclairage), $L^*a^*b^*$ Color Space is used to provide color measurements in a variety of food products. In this system, L^* indicates lightness ($L = 100 = \text{white}$; $L = 0 = \text{black}$), $+a^*$ is redness (0°), $-a^*$ is the greenness (180°), $+b^*$ is yellowness (90°) and $-b^*$ is blueness (270°) as shown in figure 2.5.

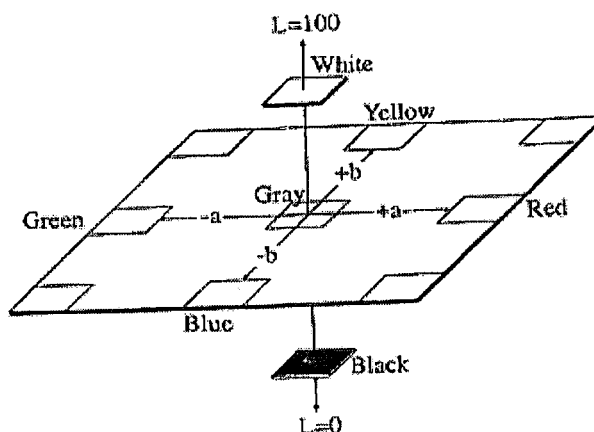


Figure 2.4 Schematic diagram representing to CIE $L^*a^*b^*$ color space. L^* values of 0 = pure black and 100 = pure white; $+a^*$ value (0°) = redness and $-a^*$ value (180°) = greenness; $+b^*$ value (90°) = yellowness and $-b^*$ value (270°) = blueness (Wrolstad, 2000)

Noodles should be bright. Colors (hue) can range from creamy white to yellow depending on noodle type. Park and Baik (2002) found that commercial white salted noodles dough had L^* values and a^* values of 81 to 82 and -0.3 to 0.3, respectively. Protein content of flour is negatively related to L^* values because the extra protein dilutes the amount of highly reflective, semi-crystalline, starch in the dough leading to duller appearance. High starch damage content and PPO also play an important role on the discoloration of noodle (Miskelly, 1996). Yellowness (b^* value) is largely affected by endosperm pigmentation. The presence of xanthophylls or carotenoids in flour is the primary factor determining the yellowness of salted noodles. The yellowness of alkaline noodles is affected both by the presence of xanthophylls, and also by the expression of yellow color by flavonoid compounds at high pH (Miskelly, 1984). Flavonoids are relatively stable and colorless at acidic pH but give a yellow color at high pH. This is the source of yellowness in alkaline noodles.

2.7.2 Texture

Texture is the other critical characteristic of noodle quality. Like color measurement, instrumental measurements are often more beneficial than sensory tests because they provide fast and precise comparisons of many samples. Instrumental measurements are also especially useful when an experiment varies formulations and processing protocols. Uniaxial testing machines (e.g. TATX 2 texture analyzer) are commonly used to determine noodle texture. Figure 2.6 shows a typical texture profile analysis (TPA) time-force curve. This technique is widely used with TATX texture analyzers.

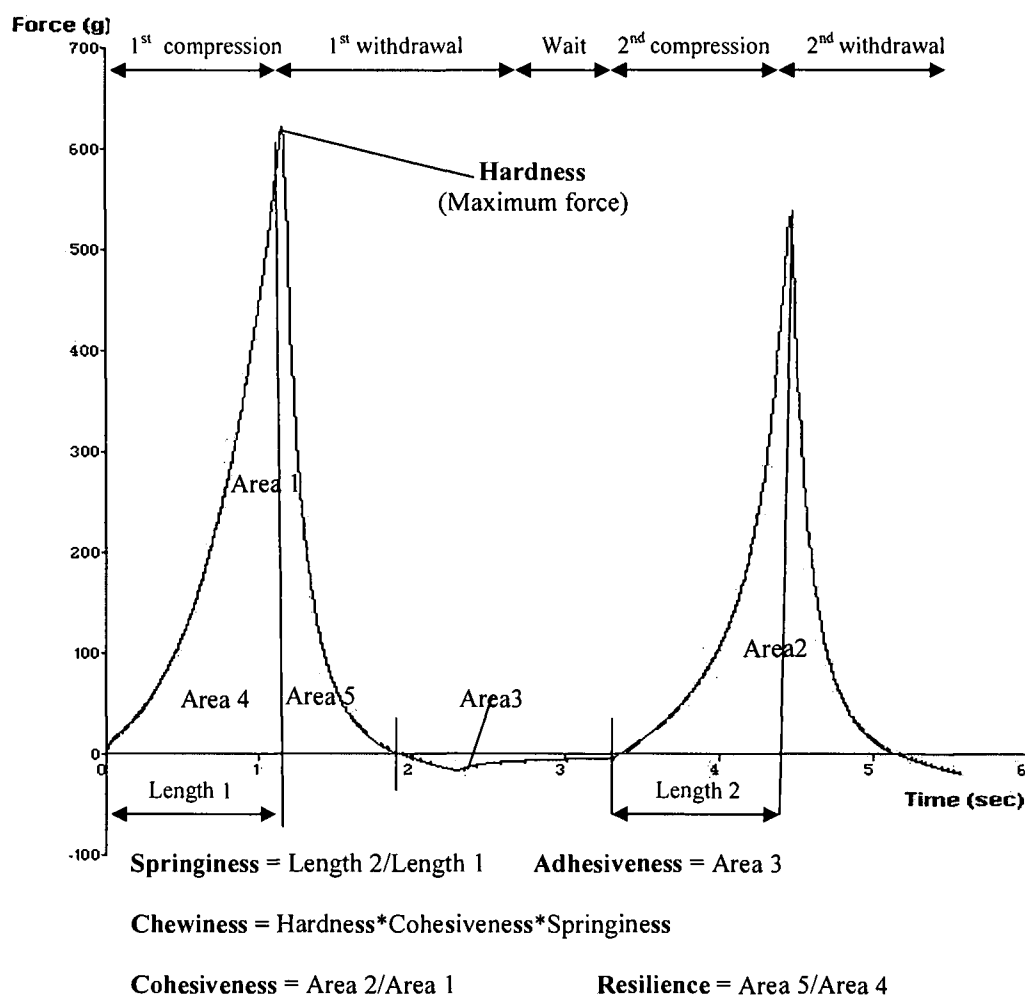


Figure 2.5 Texture profile analysis time/force curve for white salted noodles obtained using a TA.XT plus®

TPA was developed for determining food texture in a wide variety of foods.

Szczesniak (1998) developed both physical and sensory definitions of these texture characteristics: hardness, adhesiveness, springiness, cohesiveness and chewiness (Table 2.2).

Table 2.2 Physical and sensory definitions of the mechanical parameters of food texture (Szczesniak, 1998).

Parameter	Physical definition	Sensory definition
Hardness	Force necessary to attain a given deformation	Force required to compress a substance between molar teeth(solids) or the tongue and palate (semisolids)
Adhesiveness	Work necessary to overcome the attractive forces between the surface of the food and other surfaces with which the food come in contact	Force of the tongue required to remove the material that adheres to the mouth (generally the palate, but also lips, teeth, etc.) during the normal eating process
Springiness	Rate at which a deformed material goes back to its undeformed condition following removal of the deforming force	Degree and speed with which the material returns to its original height following partial compression with molar teeth
Cohesiveness	Strength of the internal bonds	Amount of sample deformation before rupture when biting with molar
Chewiness	Energy required masticating a solid food product to a state ready for swallowing	Number of chews required to masticate a sample at one chew per second and constant rate of force application to reduce it to a consistency suitable for swallowing

TPA has been shown to be effective in determining noodle texture. It was reported that cohesiveness, hardness and chewiness of TPA are significantly correlated with the eating quality of cooked noodles (Hou, 2001). Protein content, starch composition and lipid content of flour are the key factors that affect noodle texture as mentioned in section 2.5.

The objectives of this study are to study the complex interrelationships between AX content and composition, molecular weight distribution of wheat flour AX, SRC tests, and noodle processing and quality attributes.

Chapter 3

Materials and methods

3.1 Materials

3.1.1 Soft white wheat (SWW)

Sixty three advanced soft white wheat breeding lines and check Pacific Northwest varieties were obtained from the OSU wheat breeding program (Appendix A).

Samples were grown at two sites in 2002: Corvallis, OR and Pendleton, OR. Each site had two replications except for ORH011273 grown at Corvallis had one replication.

All samples were stored at room temperature in paper bag before testing.

3.1.2 Chemicals and ingredients

- All chemicals were analytical grade or equivalent.

3.2 Methods

3.2.1 Single Kernel Characterization System

Three hundred kernels from each sample were tested for hardness index, weight, diameter and moisture content using a Perten 4100 Single Kernel Characterization System (SKCS, Instruments, Huddinge, Sweden) according to American Association of Cereal Chemists approved method 55-31 (AACC, 2000). Grains were scooped out and placed into a plastic container. Broken and non-uniform kernels were discarded.

Grains were added to the SKCS hopper, located at the front of the instrument.

Hardness index, weight, diameter and moisture content data were collected and analyzed using an IBM compatible computer with the SKCS data analysis software supplied by the manufacturer.

3.2.2 Milling

Grain was tempered to 14.0% moisture content using deionized water. Grain samples and added water were shaken 50 times using a manual rotational mixer (Bioengineering Inversina, Switzerland). Moistened grain was then stored overnight in a plastic container, and milled with a Brabender Quadrumat® Senior experimental flour mill. Milling was performed using a modification of AACC approved method 26-50 (AACC, 2000).

Roll temperature was increased to 31°C by using a small electric fan heater and maintained at 31-32°C. Total weight of wheat sample was measured and recorded before milling. Grain (300 g) was laid on the sample feeder. Wheat was fed to the 1st break roll at a rate of 130-135 g/min. After all the grain passed through the break rolls (this was assumed to be when the grinding sound disappeared), the break rolls were cleaned by reversing for 3 sec and running forward again for an additional 3 sec. This sequence was repeated 3 times. The cleaning hatch was then opened and brushed 2 times. After the rolls were cleaned, the timer was set to 16 min to allow sufficient time for all flour to pass through the sieves. When the timer indicated that 11 min had elapsed, a pan was put under the spout for 5 min to catch the remaining flour (short). The reduction rolls were cleaned in the same way as the break rolls. After the completion of the 16 min reduction grinding, bran, short, break, and reduction flours were weighed and recorded. Reduction and break flours were mixed using a manual rotational mixer. Percentage flour yield and % break flour yield were calculated as follows:

$$\% \text{ Flour yield} = \frac{(\text{Weight of reduction flour} + \text{break flour})}{\text{Weight of reduction flour} + \text{break flour} + \text{short} + \text{bran}} \times 100$$

$$\% \text{ Break flour yield} = \frac{\text{Weight of break flour}}{\text{Weight of reduction flour} + \text{break flour} + \text{short} + \text{bran}} \times 100$$

3.2.3 Moisture content

Flour moisture content was determined in a Fisher Isotemp® air oven 200 series Model 230 F air oven according to AACC approved method 44-15 A (AACC 2000).

Moisture dishes and lids (5.5 x 1.5 cm) were dried for 1 hour at 130°C, cooled in a desiccator, and weighed. A 2- to 3 g flour (weighed accurately to ± 0.005 g) was measured into each dish. The dishes were uncovered and the lids were placed beside their containers. The samples were dried for exactly 60 min after the oven recovered its temperature. Samples were removed from the oven, rapidly covered, and transferred to desiccator as quickly as possible. Dish, lid and dry flour were weighed after 45 min. The replicate must be within 0.2% moisture; otherwise the determination was repeated. The loss in weight was determined as moisture. Percent moisture was calculated using formula:

$$\% \text{ Moisture} = \frac{\text{Moisture loss in grams}}{\text{Original weight of sample}} \times 100$$

3.2.4 Flour ash content

Flour ash content was determined using AACC approved method 08-01 (AACC, 2000). Testing was done by the USDA-ARS Western Wheat Quality Laboratory (Pullman, WA). A 4 g sample of flour was weighed into an ashing dish, placed in muffle furnace, and ignited at 550°C for 15 hours. The ashing dish plus dry flour were cooled in desiccator and weighed after reaching room temperature.

$$\% \text{ Ash} = \frac{\text{Weight of residue} \times 100}{\text{Sample weight}}$$

3.2.5 Flour protein content

Flour protein content was determined by near infrared spectroscopy (NIRS) using AACC approved method 39-11 (AACC, 2000) by USDA-ARS Western Wheat Quality Laboratory (Pullman, WA). The NIRS protein results were checked and minor corrections made by determining crude protein on about 10% of the samples using the reference combustion method (AACC approved method 46-30 Leco, model FP-428).

$$\text{Crude protein, \%} = \% \text{ N} \times 5.70$$

3.2.6 Flour swelling volume (FSV)

FSV was determined by USDA-ARS Western Wheat Quality Laboratory (Pullman, WA). Flour (0.45 g dry weight basis) was mixed with 12.5 ml of water. Samples were placed in 92.5 °C water bath and constantly inverted for thirty minutes. Then samples were rapidly cooled in ice water bath and followed by 5 minutes in 25°C water. After

that, the samples were centrifuged at 1,000 X g for 15 minutes. The height of the gel in mm was measured. The following conversion formula was used.

$$\text{Flour swelling volume (ml/g)} = [(\text{mm} \times 1.52) - 0.30\text{mL}] / 0.45\text{g}.$$

3.2.7 Rapid Visco Analyzer (RVA)

RVA analyses of flour were determined at USDA-ARS Western Wheat Quality Laboratory (Pullman, WA). A 3.5 ± 0.05 g of flour (at 14% moisture basis) was weighed into aluminum RVA containers. 25 ml of water was added to make a total weight of 28.5 g. The mixture was inserted into the RVA heating block (Newport Scientific, Warriewood, New South Wales, Australia). The RVA temperature was set at 60°C for 2 min, and then heated for 6 minutes at a constant rate to 93°C with a final hold at 93°C for 2 min. Total test time was 10 min. The peak viscosity was reported in RVA units (centipoise x 10).

3.2.8 Cookie making

Cookies were made at USDA-ARS Western Wheat Quality Laboratory (Pullman, WA) using AACC approved method 10-52 (AACC, 2000) with modifications. The ingredients were composed of 40 g flour, 25% water, 60% sugar, 30% emulsified shortening, 3% dry skim milk, 2.06% NaHCO₃, 0.68% NH₄Cl, 0.26% NaCl, and 0.24% emulsifier (distilled mono- and di-glycerides). All % additional calculated on a flour basis. Cookie diameter (CODI) was measured and reported in cm. Cookie diameter is the most widely used index of overall soft wheat quality and will be used as index in the results.

3.2.9 Solvent Retention Capacity test

Flour SRC tests were determined according to AACC approved method 56-11 (AACC, 2000) with a modification using 1 g of flour instead of 5 g.

Reagents preparation

1. Deionized water
2. Sucrose solution, 50 % (w/w). 500 g of reagent-grade sucrose was weighed into 1 L container. Water was added to make 1000g. Solution should be made 12 hours in advance, stored at room temperature, and used within 7 days.
3. Sodium carbonate solution, 5% (w/w). 500 g of reagent-grade anhydrous sodium carbonate was weighed into 1 L container. Water was added to make 1000 g. Solution should be used within a week.
4. Lactic acid solution, 5% (w/w). The weight of reagent required to give 50 g of lactic acid was calculated. For example, if lactic acid is 88.50% concentration, add $50/0.885$ (=56.497 g) lactic acid. The calculated amount of reagent was weighed into 1 L container. Water was added to make 1000 g. Solution should be used within 2-3 days.

Procedure

Conical 15 ml centrifuge tubes with screw caps were weighed. Then, 1.000 ± 0.050 g flour of known moisture content was weighed into each tube. Next, 5 ml of solvent was added to each tube containing flour. Caps were put on tubes and shaken vigorously to suspend flour. The timer was started. Tubes were shaken for 5 sec using a Labquake® shaker (Barnstead/Thermolyne) when the timer indicated that 5, 10, 15 and 20 min has elapsed. Tubes were transferred to a centrifuge (Beckman GS-15R)

and centrifuged at 1000 X g for 15 min. After that, the supernatant was decanted.

Tubes were drained at 90° angle for 10 min on a paper towel. Tube, cap, and pellet were weighed. Weight of gel was determined by subtracting weight of tube and cap from total weight of tube, cap, and gel. SRC value was calculated for each solvent.

$$\%SRC = \{[\text{gel weight/ flour weight}] \times [86 / (100 - \% \text{ flour moisture})] - 1\} \times 100$$

3.2.10 Starch damage

Starch damage was determined by using a starch damage assay kit (Megazyme, Ireland) according to AACC approved Method 76-31 (AACC, 2000).

Reagents

GOPOD reagent composed of glucose determination reagent and glucose reagent buffer (concentrated).

Glucose Determination Reagent

Reagent concentrations after dissolution in buffer:

Glucose oxidase > 12,000 U/liter

Peroxidase > 650 U/liter

4-Aminoantipyrine 0.4 mM

Both glucose determination reagent and glucose reagent buffer were mixed and diluted to 1 L with distilled water. The reagent after mixing was referred to GOPOD reagent.

Procedure

A 0.1 ± 0.01 g of flour was weighed into 14 ml-glass centrifuge tube and pre-equilibrated to 40°C for 2 to 5 min. At the same time, fungal α -amylase solution (50 U/ml) was pre-equilibrated to 40°C for 5 to 10 min. Then, 1 ml of pre-equilibrated fungal α -amylase was added to tubes containing flour. The tubes were vigorously and immediately stirred for 5 sec using a vortex mixer and then incubated at 40°C for exactly 10 min (from time of addition of enzyme). After 10 min, 0.2% (v/v) sulfuric acid was added to terminate the reaction, and the tubes were centrifuged at $1000 \times g$ for 5 min using a Beckman GS-15R centrifuge. Aliquots of 0.1 ml supernatant solution were carefully transferred to the bottom of the bottom of test tube. After that, 0.1 ml of amyloglucosidase solution (2 U) was added to each tube. The tubes were then incubated at 40°C for 10 min. Then, 4 ml of GOPOD reagent was added to each tube (including glucose standards and reagent blank tubes), and the tubes were incubated at 40°C for 20 min. The absorbance at 510 nm for each sample was recorded. Starch damage was calculated as follows.

$$\begin{aligned} \% \text{ Starch damage} &= \Delta E \times F \times 90 \times \frac{1}{1000} \times \frac{100}{W} \times \frac{162}{180} \\ &= \Delta E \times \frac{F}{W} \times 8.1 \end{aligned}$$

ΔE = Absorbance read against the reagent blank

$$F = \frac{150 \text{ (}\mu\text{g of glucose)}}{\text{absorbance of } 150 \mu\text{g of glucose}}$$

90 = volume correction (0.1 ml taken from 9 ml)

1/1000 = conversion from micrograms to milligrams

$100/W$ = Factor to express Starch damage as a percentage of flour weight

W = Weight in milligrams of flour analyzed

$162/180$ = Adjustment from free glucose to anhydro glucose (the form of glucose in starch)

3.2.11 AX content determination

AX content were determined according to Douglas (1981) and Bettge and Morris (2000) and expressed as % xylose equivalents, dry weight basis.

Extracting solution

1. glacial acetic acid 110 ml
2. concentrated HCl 2 ml
3. 20% w/v phloroglucinol in ethanol 5 ml
4. 1.75% w/v glucose 1 ml

Standard solution preparation

To prepare xylose standards, a 0.1 ± 0.001 g of D- (+)-xylose was weighed and mixed with distilled water in a 100 ml volumetric flask (solution A). A 10 ml of solution A was transferred to another 100 ml volumetric flask and made up to 100 ml with distilled water to make it 0.01g/100 ml (solution B). Then, 0.5, 1.0, 1.5 and 2.0 ml of solution B were made up to 2.0 ml with distilled water to give solution concentrations of 25, 50, 75, 100 $\mu\text{g/ml}$, respectively.

Sample preparation

For total AX content determination, 5 ± 0.5 mg of flour was weighed into stoppered glass tube. 2 ml of water was added to each tube.

For WEAX content determination, 0.1 ± 0.001 g of flour was weighed. 10 ml of water was added. The solution was mixed 3 times during a 30 min extraction time. Then, tubes were centrifuged at $3000 \times g$ for 10 min using a Beckman GS-15R centrifuge. 2 ml of supernatant solution was taken to determine WEAX content.

Procedure

Blank, standard solution, total AX solution and WEAX solution were added with 10 ml of extracting solution. Tubes were placed in vigorously boiling water for 25 min, then removed and rapidly cooled in flowing cold water. Absorbance at 510 and 552 nm were recorded using a spectrophotometer (Spectronic 1001, Milton Roy company, NY). $A_{552}-A_{510}$ of samples were compared with standard curve of difference between $A_{552}-A_{510}$ and xylose standards ($\mu\text{g/ml}$) to obtain total AX and WEAX content.

WUAX content was obtained from the subtraction of WEAX content from total AX content.

3.2.12 Extraction procedure of AX

Enzymes: Alcalase (P-4860; Lot 022K1864), amylase (A-7095; Lot 110K1350) and amyloglucosidase (A-3403; Lot 072K1623) were obtained from Sigma-Aldrich, Inc, MO.

Six high and six low AX content of SWW breeding lines were selected to extract AX (Appendix B). Extraction method of AX was modified according to Faurot et al (1995) and Loosveld et al (1997) as shown in figure 3.1.

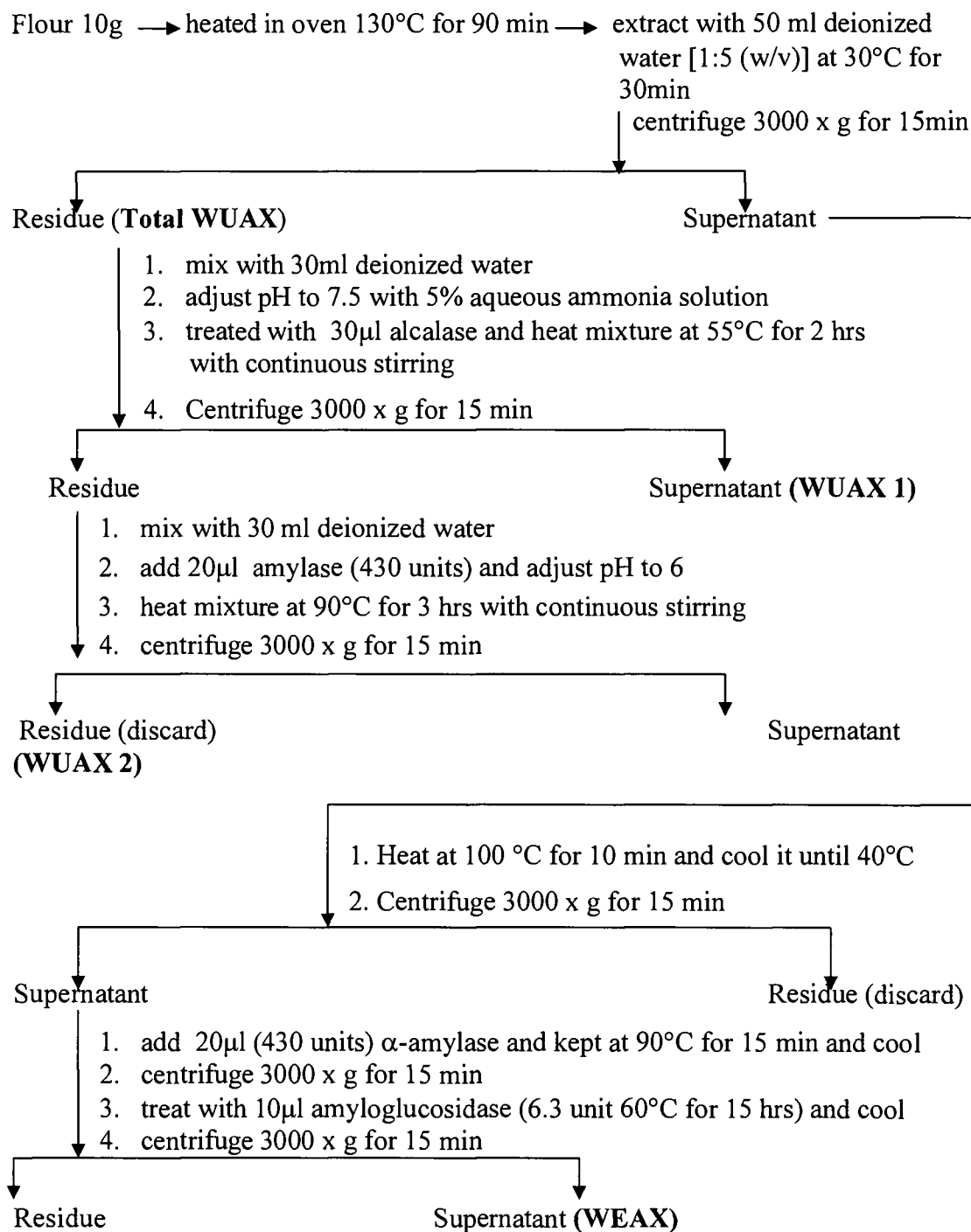


Figure 3.1 Flow scheme of the extraction procedure used for isolation of WUAX and WEAX

3.2.13 Soluble starch determination

According to Stoddard and Sarker (2000), iodine/potassium iodide solution was used to check for soluble starch in AX fraction solutions. 200 μL of each AX fraction was added to 800 μL of iodine solution (0.2 g of I_2 + 2 g of KI + 250 ml of distilled water). The solution was diluted with 9 ml of water. Then, solution was kept in a refrigerator for 20 min and the color observed compared to the blank.

3.2.14 Molecular weight distribution of AX using Size Exclusion HPLC (SEHPLC)

AX fractions were filtered with 0.45 μm membrane filter (Millipore, Ireland) into autosampler vials. AX fractions (20 μl) were separated using a Shodex OH pak SB-804 HQ column (30x0.8cm) (Showa Denko K.K., Tokyo, Japan) on a Waters 2695 HPLC (Waters, MA) equipped with a Waters 2414 refractive index detector (Waters, MA). AX fractions were eluted with 0.3% NaCl (1.0 ml/min at 30°C). The column was calibrated with Shodex standard P-82 pullulans (1 mg/ml) (Showa Denko K.K., Tokyo, Japan) with molecular weights of 78.8×10^4 , 40.4×10^4 , 21.2×10^4 , 11.2×10^4 , 4.73×10^4 , 2.28×10^4 , 1.18×10^4 , and 0.59×10^4 .

The chromatograms were collected and integrated manually. Peak area and peak height were calculated by the Empower program provided by HPLC manufacturer (Waters, MA).

3.2.15 Sugar analysis

Sugar standard: D-(+)-Glucose (glc), D-(+)-xylose (xyl), D-(+)-galactose (gal), D-(-)-arabinose (ara) and D-(+)-mannose (man) were obtained from Sigma-Aldrich, Inc, MO. Sugars were determined according to laboratory analytical procedure LAP-002 (Raymond and Ehrman, 1996) with modifications.

Sample preparation

1.5 ml of 72% (w/w) H_2SO_4 was mixed with 7.5 ml of AX fractions in 125 ml autoclavable bottles. Then, 34.5 ml of H_2O was added to make a final concentration of 4% acid solution. A series of sugar standards (predried at 45°C for 12 hours) in 4% H_2SO_4 were prepared in 125 ml autoclavable bottle. The bottles were closed with screw caps. Samples along with all sugar standards were put into an autoclave (model: AS-02L626-15-14, American sterilizer Co., PA) for 1 hour at $121 \pm 3^\circ\text{C}$. After completion of autoclaving, samples were cooled in flowing cold water until they reached room temperature. All hydrolyzate were transferred to 50 ml volumetric cylinders and adjusted 50 ml by adding distilled water. Then, 20 ml of hydrolyzate was transferred to an 50 ml Erlenmeyer flask and adjusted with CaCO_3 to a pH between 5 and 6. CaCO_3 was added slowly with frequent swirling to avoid foaming. The pH adjusted hydrolyzate was filtered with $0.2\mu\text{m}$ disposable nylon syringe filter (Alltech Associates, Inc., IL) into an autosampler vial. The portion of pH adjusted hydrolyzate was securely sealed and stored in a refrigerator for no longer than two weeks before HPLC testing. pH adjusted hydrolyzed standards and samples ($50\mu\text{l}$) were separated on a Biorad Aminex HPX-87P (300 mm x 7.8 mm) (Bio-Rad Laboratories, CA) equipped with de-ashing guard column (Bio-Rad Laboratories, CA)

using Waters 2695 HPLC. Samples were eluted with 18 MΩ deionized water (0.3 ml/min at 85°C) for 45 min and detected using a Waters 2414 refractive index detector held at 50°C. The chromatograms were collected and integrated manually. Peak area and peak height were calculated by the Empower program provided by HPLC manufacturer (Waters, MA). The percentage of sugar was calculated for each sample.

AX and AG content, and A/X and A/G ratio were calculated according to Cleemput et al (1993).

$$\text{AX content} = 0.88 \times (\% \text{ ara} + \% \text{ xyl})$$

$$\text{A/X} = \% \text{ ara to } \% \text{ xyl ratio}$$

$$\text{AG content} = 0.87 \times (\% \text{ ara} + \% \text{ gal})$$

$$\text{A/G} = \% \text{ ara to } \% \text{ gal ratio}$$

3.2.16 Chinese white salted raw noodle making

3.2.16.1 Method of optimum water absorption determination

Amounts of water content for noodle making varying above or below an estimated optimum water content were added to 10 ± 0.001 g of flour (14% moisture basis).

Flour was mixed with water in a 10 g pin mixer (National MFG. Co. Lincoln, NE) for 1 min. After that, adhering dough was scraped from the pins using a plastic scraper.

The mixture was mixed for further 2 min 30 sec. Total mixing time was 3 min 30 sec.

Crumbly dough was removed from the mixer and compressed between steel rollers using a 4 mm roll gap in an Ohtake noodle machine (Tokyo, Japan). This was repeated

3 times. The optimum water absorption was determined visually by observing the uniformity of water distribution in the dough sheet.

3.2.16.2 Production of Chinese white salted raw noodles

Sample preparation

1. A 150 g of flour (14% moisture basis) was weighed and placed into a 200 g mixing bowl of a 200 g pin mixer (National MFG. Co. Lincoln, NE).
2. A 1.2 % salt was weigh and dissolved in the optimum amount of deionized water

Procedure

Salt solution was added to the top of the dry flour. The mixture was mixed using the 200 g pin mixer for 1 min. After that, the mixer was stopped and adhering dough was scraped from the pins using a plastic scraper. The mixture was mixed for further 2 min 30 sec. Total mixing time was 3 min 30 sec. After mixing, the crumbly dough was removed from mixer, kept in a zip-lock bag, and rested for 30 min at room temperature.

Small green rubber strip was placed between Ohtake noodle machine rollers. The rollers were started and stopped until rubber strip blocked the gap between the rolls. The crumbly dough was poured into gap between the rolls. Then, the dough was compressed at 4 mm gap. After that, dough sheet was fold in half and resheeted three more times at 4 mm gap. Without folding sheet, dough sheet was passed through the

rollers one more time at 4 mm gap. The dough sheet was kept in a zip-lock bag and rested for 30 min at room temperature.

The dough sheet was passed through the rollers four times with progressively reduced roll gaps of 3.5, 3.0, 2.0 and 1.5 mm. A small piece of dough sheet was cut and passed through the last calibration roll. The dough sheet thickness was measured using a Peacock thickness gauge. The roll gap was adjusted to make the final sheeted dough thickness 1.2 ± 0.05 mm. Then, the remaining dough sheet was cut into strips with a #12 square type cutter (2.5 mm width). The noodle strands were cut into 6.5 cm length and store in a plastic bag for 24 h before the determination of optimum cooking time, cooking loss, cooking yield and texture evaluation

3.2.17 Noodle evaluation

3.2.17.1 Optimum cooking time

A 200 ml of deionized water was boiled rapidly on induction hotplate Model US-9000-15M (Iwatani International Corporation, Japan). The timer was started when 10 ± 0.5 g of noodles were added into boiling water. Water was brought back to boil and maintained a slow boil (medium) with continued stirring during cooking time. After 4 min of cooking, a strand of noodle was removed from cooking water, placed in ice water to arrest the cooking process, and placed on a plastic plate. Every 10 s, a strand of noodle was removed from cooking water, and followed the same step as indicated above until noodles had been cooked for 6 min. The noodle strands were squeezed

between plastic plates. The optimum cooking time was judged by the disappearance of the uncooked doughy core at the center of the noodle strands.

3.2.17.2 Water content of noodle

Aluminum dishes were weighed to 0.0001 g. Then, a 2 ± 0.0001 g of raw noodles were weighed out into the aluminum dishes, and put in an air oven. Raw noodles were evaporated at 105 ± 2 °C for 16 hours. After that, aluminum dish and dried noodles were cooled to room temperature (~ 60 min) in a desiccator, and weighed to 0.0001 g. Water content was calculated as follows:

$$\text{Water content (\%)} = \frac{a-b}{c} \times 100$$

a = weight of raw noodle and aluminum dish (g) (Before dried)

b= weight of raw noodle and aluminum dish (g) (After dried)

c= weight of noodle (g)

3.2.17.3 Cooking loss

A 10 ± 0.0001 g of raw noodle was weighed. A 200 ml of deionized water was boiled rapidly on an induction hotplate. The timer was started when noodles were added into boiling water. Water was brought back to boil and maintained a slow boil (medium) with continued stirring during cooking time. After completing of cooking time, the cooking water was poured into an aluminum dish, which was already weighed to 0.0001 g. Water was evaporated in an air oven at 105 ± 2 °C for 16 hours. Then, aluminum dish was cooled to room temperature (~ 60 min) in a desiccator and

weighed to 0.0001 g. Cooking loss was defined as weight of noodle residue in cooking water.

$$\text{Noodle residue in cooking water (NR)} = \frac{(a-b) \times 100}{\frac{[(100\% - W) \times d]}{100}}$$

a = weight of actual solids and aluminum dish

b = weight of aluminum dish

d = weight of raw noodle

W = water content in %

3.2.17.4 Cooking yield

Cooked noodle strands from cooking loss determination were drained in a wire screen for 30 sec. Then, all noodle strands were put on disposable absorbent tissue paper for 30 min and weighted to 0.0001 g

$$\text{Cooking yield} = \frac{\text{Weight of cooked noodle(g)} - \text{Weight of uncooked noodle(g)}}{\text{Weight of uncooked noodle (g)}} \times 100$$

3.2.17.5 Texture evaluation

Instrumental setup

A Texture Analyzer (TA, TA.XT plus®, Stablemicrosystems, UK) was used for TPA. The texture analyzer with a compression blade with a contact surface 5 mm wide was calibrated by using 2 kg standard weight. Probe distance from the bottom plate was set to 5 mm.

Instrumental settings:

Pretest speed	4.0 mm/sec
Test speed	1.0 mm/sec
Strain	70%
Time between compressions	1.0 sec
Trigger force	5 g
Trigger distance	2.0 mm

Testing

Noodles were tested by TPA with two cooking time methods: optimum and 5 min fixed cooking time. A 200 ml of deionized water was boiled rapidly on induction hotplate. The timer was started when 10 ± 0.5 g of noodles were added into boiling water. Water was brought back to boil and maintained a slow boil (medium) with continue stirring during cooking time. When the predetermined cooking time (fixed or optimum) was reached, noodles were rapidly drained into a sieve. Noodles were rinsed with deionized water for 1 min. Drained noodles were transferred to two closed plastic containers and one closed plastic containers containing RT water. The timer was set to 15 min. Noodles were tested at 0 min without water by placing three strands of noodles adjacent to each other under the center of compression blade. Noodles were compressed to 70% strain, allowed 1 sec pause between bites, and compressed again. The compression blade and bottom plate were cleaned before the next test. At 15 min, noodles stored with and without water were tested as described above.

Textural profile data was collected from the TA using an IBM compatible computer using the Texture Exponent software supplied with the TA. TPA parameters used were, hardness, adhesiveness, cohesiveness springiness, chewiness, and resilience.

Definitions and calculations of these parameters are explained in Figure 2.5 and Table 2.2.

3.2.18 Statistical analysis

Each of the breeding lines or varieties used, except for ORH011273, was represented by duplicate samples. Analytical and other tests were performed on each sample once. Mean values for each breeding line, or wheat variety, for all tests, were calculated using the individual results from each replicate. Statistical analysis of data was performed using the SAS software (Version 8e, SAS Institute. Cary, NC). Correlation procedure was performed using a simple linear least squares model. ANOVA was performed using the general linear model in SAS. Stepwise multiple regression procedure was used where indicated. Differences were considered significant at $P < 0.05$, unless otherwise specified.

Chapter 4

Results and Discussion: Kernel, Milling and Analytical characteristics

Table 4.1 shows hardness index, kernel weight and diameter of 251 samples of soft white wheat representing 63 breeding lines grown in duplicate locations at Corvallis and Pendleton in 2002(Except for ORH011273 grown at Corvallis had one replication). Visual examination showed that the grain from Pendleton was pinched and shrunken. This was a result of environmental stress late in the grain filling period. The stress occurred as a result of near drought conditions in Pendleton in 2002. The visual observation of the shrunken grain was confirmed by the low average kernel weight of the Pendleton samples (Table 4.1). It was considered that the milling characteristics of the Pendleton grain would have made it more poor and difficult to separate bran from flour. This would have introduced more bran sourced AX. Given the differences in characteristics of bran and endosperm sourced AX (Fincher and Stone, 1986), this was thought to be undesirable. As a result of the condition of the grain from Pendleton, it was decided to use only grain from the Corvallis site. Wheat kernels from the Corvallis site had overall higher hardness index value, and kernel weight and diameter (Table 4.1).

Table 4.1
Mean and standard deviation¹ values of hardness index, and kernel weight
and diameter obtained from Single Kernel Characterization System

Single Kernel Characteristics	Locations	
	Corvallis	Pendleton
Hardness index	41.24 ± 7.04	36.95 ± 8.10
Weight (mg)	48.54 ± 4.61	34.90 ± 4.42
Diameter (mm)	3.02 ± 0.18	2.37 ± 0.20

¹Standard deviation was calculated from 300 kernels.

Table 4.2 shows Means, standard deviations and ranges of wheat milling characteristics, and flour starch damage, ash, protein and AX contents. Straight grade flour is usually about 72% extraction (72% flour yield, see section 3.2.2). These milling results are similar. Wheat flour AX content has been reported to be about 2-3 % (Jeleva and Hlynka, 1971). In these results, flour contained only around 1.7 % AX. WEAX typically accounts for 0.5-0.8% (w/w) of wheat flour (Jeleva and Hlynka, 1971). These samples had a mean value of WEAX of 0.47%, which also somewhat lower than previous reports. It considered that both of these observations resulted from the use of soft wheats, which have been deliberately bred to contain lower amounts of AX than hard wheats. Ash content is an important measurement because it relates to bran contamination in flour. In this sample set, ash content was around 0.4 %, which was acceptable for milling quality from a Brabender® Quadramat Senior experimental flour mill.

Table 4.2
Means, standard deviations and ranges of wheat milling characteristics, and flour starch damage, ash, protein, and AX contents of 63 soft white wheat breeding lines

Quality Characteristics	Mean	Range	SD
Flour yield (%)	70.4	10.2	1.8
Break flour yield (%)	37.4	15.9	2.9
Starch damage (%) ¹	4.15	2.50	0.54
Flour ash (%) ¹	0.40	0.16	0.03
Flour protein (%) ¹	6.4	1.9	0.4
AX			
Total (%) ¹	1.74	0.93	0.19
Water extractable (%) ¹	0.47	0.33	0.08
Water unextractable (%) ¹	1.27	0.86	0.19

¹On a 14 % flour moisture basis (n = 125).

Table 4.3 shows linear correlations between kernel texture, milling characteristics, solvent retention capacity tests, cookie diameter, and flour AX, starch damage and protein contents of 63 soft white wheat breeding lines. Hardness index was positively correlated with total AX and WEAX. This agrees with the literature. Hong et al (1989) observed a positive correlation between WEAX and total AX contents and grain hardness in hard, soft and club wheats. Bettge and Morris (2000) also found a positive correlation between total and soluble grain AX with kernel hardness index. AX may associate with amyloplast membranes and act as cement between starch granules and storage proteins. Therefore, the higher degree of adhesion between starch granule and protein was related to higher amount of AX, and therefore to kernel hardness.

Table 4.3
Correlation coefficients between kernel texture, milling characteristics, solvent retention capacity tests, cookie diameter, and flour AX, starch damage and protein contents of soft white wheat breeding lines¹

Quality Characteristics	Hardness	Flour Yield	Break Flour	Solvent Retention Capacity				Arabinoxylan ²			Starch	Flour
	Index	(%)	Yield (%)	H ₂ O (%) ³	Na ₂ CO ₃ (%) ³	Sucrose (%) ³	Lactic acid (%) ³	Total (%) ³	WEAX (%) ³	WUAX (%) ³	Damage (%) ³	Protein (%) ³
Flour yield (%)	-0.268**											
Break flour yield (%)	-0.537***	0.728***										
Solvent retention capacity												
H ₂ O (%) ³	0.427***	NS	NS									
Na ₂ CO ₃ (%) ³	0.337**	NS	NS	0.686***								
Sucrose (%) ³	NS	NS	NS	0.610***	0.758***							
Lactic acid (%) ³	0.200*	-0.284**	-0.236**	NS	0.259**	0.279**						
Arabinoxylan ²												
Total (%) ³	0.236**	NS	NS	0.358***	0.438***	0.315**	NS					
WEAX (%) ³	0.209*	-0.339**	-0.209*	0.450***	0.660***	0.634***	0.356***	0.215*				
WUAX (%) ³	NS	NS	NS	0.179*	NS	NS	NS	0.919***	-0.187*			
Starch damage (%) ³	0.588***	NS	-0.343***	0.387***	0.233**	NS	NS	NS	NS	NS		
Flour protein (%) ³	NS	NS	NS	NS	NS	NS	0.259**	NS	NS	NS	NS	
Cookie diameter (cm)	-0.501***	NS	0.238**	-0.396***	-0.332**	-0.297**	-0.221*	-0.346***	-0.248**	-0.249**	-0.252**	NS

¹ *, ** and ***= Correlation coefficient is significant at P<0.05, 0.01 and 0.001, respectively. NS: P>0.05

²WEAX= water extractable arabinoxylan and WUAX= water unextractable arabinoxylan.

³On a 14 % flour moisture basis (n = 125)

Kernel hardness index was negatively correlated with break flour yield and positively correlated with starch damage. As hardness index was also correlated with total and WEAX, this helped to confirm the concept that the higher amount of AX, the harder the wheat kernel, as also observed by Hong et al (1989).

The highly significant positive correlation between starch damage and hardness index (Table 4.3) indicated that the bonding strength between protein and starch of softer samples was lower. As a result the soft wheats required less grinding energy to reduce the size of the flour particles and had lower amounts of starch damage than harder samples. Flour yield was positively related to break flour yield. Break flour yield was negatively correlated with WEAX and starch damage. Overall, softer wheats gave higher amounts of break flour and had lower amount of WEAX and starch damage.

Starch damage, total AX, WEAX and WUAX were also highly, and negatively, correlated with cookie diameter (Table 4.3). This may be because damaged starch and AX can readily absorb high amounts of water or form entanglement networks. Both of these phenomena can restrict cookie spreading during baking.

SRC tests relate flour performance to specific flour components (Section 2.4). From table 4.3, sodium carbonate SRC, which is related to starch damage, showed positive correlations with total AX, WEAX and starch damage. Sucrose SRC, which has been related to AX content by other workers (e.g.; Guttieri et al 2001), also showed positive correlations with total AX and WEAX. Lactic acid SRC, which is related to both

protein content, and gluten composition and strength, showed no significant correlation with mixograph absorption ($r = 0.044$, NS) as might have been expected. Lactic acid SRC was positively correlated with flour protein content in this study.

Sodium carbonate SRC showed a highly positive correlation with sucrose SRC ($r = 0.758$, $p < 0.001$). A similar correlation was observed by Guttieri et al (2001). Guttieri et al (2001) also found positive relationships between sodium carbonate SRC, sucrose SRC and lactic acid SRC. These relationships were also observed in this study. Water contained in each solvent can influence the absorption of flour components other than the component targeted by the solute itself. As a result, there is a large overlap between different SRC tests. This may not be surprising.

All SRC tests showed negative correlations with cookie diameter. Water SRC showed the best correlation ($r = -0.396$, $p < 0.001$) with cookie diameter, but despite the statistical significance, it was a numerically weak relationship. Guttieri et al (2001) also observed relationships between cookie diameter and sodium carbonate SRC, sucrose SRC, and lactic acid SRC, but not water SRC. Correlation coefficients for these relationships varied from 0.55 to 0.78. However, the significant relationships observed in this study were far more scattered than those observed by Guttieri et al (2001) (Figures 4.1 and 4.2). This suggests that SRC tests should not be the only method that is used to predict the cookie baking quality of flour. Additional methods such as mixograph, flour swelling volume (FSV), SDS-sedimentation volume, Rapid Visco Analyzer (RVA), may be required for adequate prediction. Recently, Gaines

(2004) developed a simple regression model of sugar-snap cookie diameter using flours from a long flow milling system. The parameters used for the regression model were sucrose SRC, flour protein content, and break flour yield. This multifactor approach is likely to be useful for the prediction of cookie characteristics in the future.

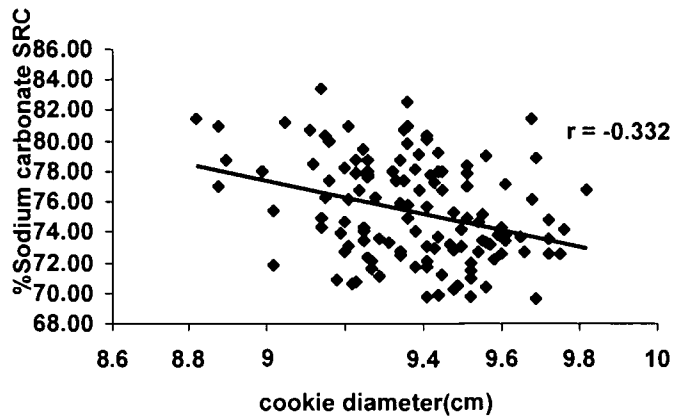


Figure 4.1 Scatter plot of the relationship between sodium carbonate SRC and cookie diameter for 125 soft white wheat samples

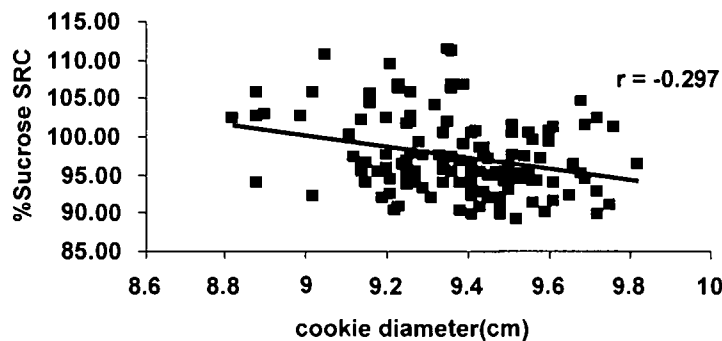


Figure 4.2 Scatter plot of the relationship between sucrose SRC and cookie diameter for 125 soft white wheat samples

Milling yield and break flour yield of Corvallis flour samples were in the ranges expected (Table 4.2). The correlations between milling and flour characteristics were similar to those found in the literature (Gaines, 2000; Guttieri et al, 2001; Table 4.3). In this study, SRC tests were assessed as predictors of the quality of specific end-products, in this case, cookies. However, it was found that sucrose and sodium carbonate SRC tests were not reliable predictors of cookie diameter because of their scattered correlations with this parameter. Total, WEAX, and WUAX contents were no better at predicting cookie diameter than were the SRC tests.

Chapter 5

Results and Discussion: Extraction of AX fractions

Development of AX extraction method

To achieve the desired extraction of AX, selected components of the AX extraction and fractionation methods of Faurot et al (1995) and Loosveld et al (1997) were combined. In addition, the extraction method was scaled down from a 50 g to 10 g of flour due to the limited amount of flour available.

The larger (50 g) scale extraction protocol is shown in Figure 5.1. Faurot et al (1995) and Loosveld et al (1997) found a WEAX fraction in the supernatant after extraction with distilled water. In this study, the equivalent WEAX fraction (Figure 3.1) was continuously dialyzed against water and analyzed using SEHPLC. However, it was observed that the dialysis caused the loss of some WEAX (Figure 5.2). As a result of the loss of WEAX, it was decided not to dialyze in the revised extraction procedure used here. Consequently, only crude WEAX fraction was obtained in this step.

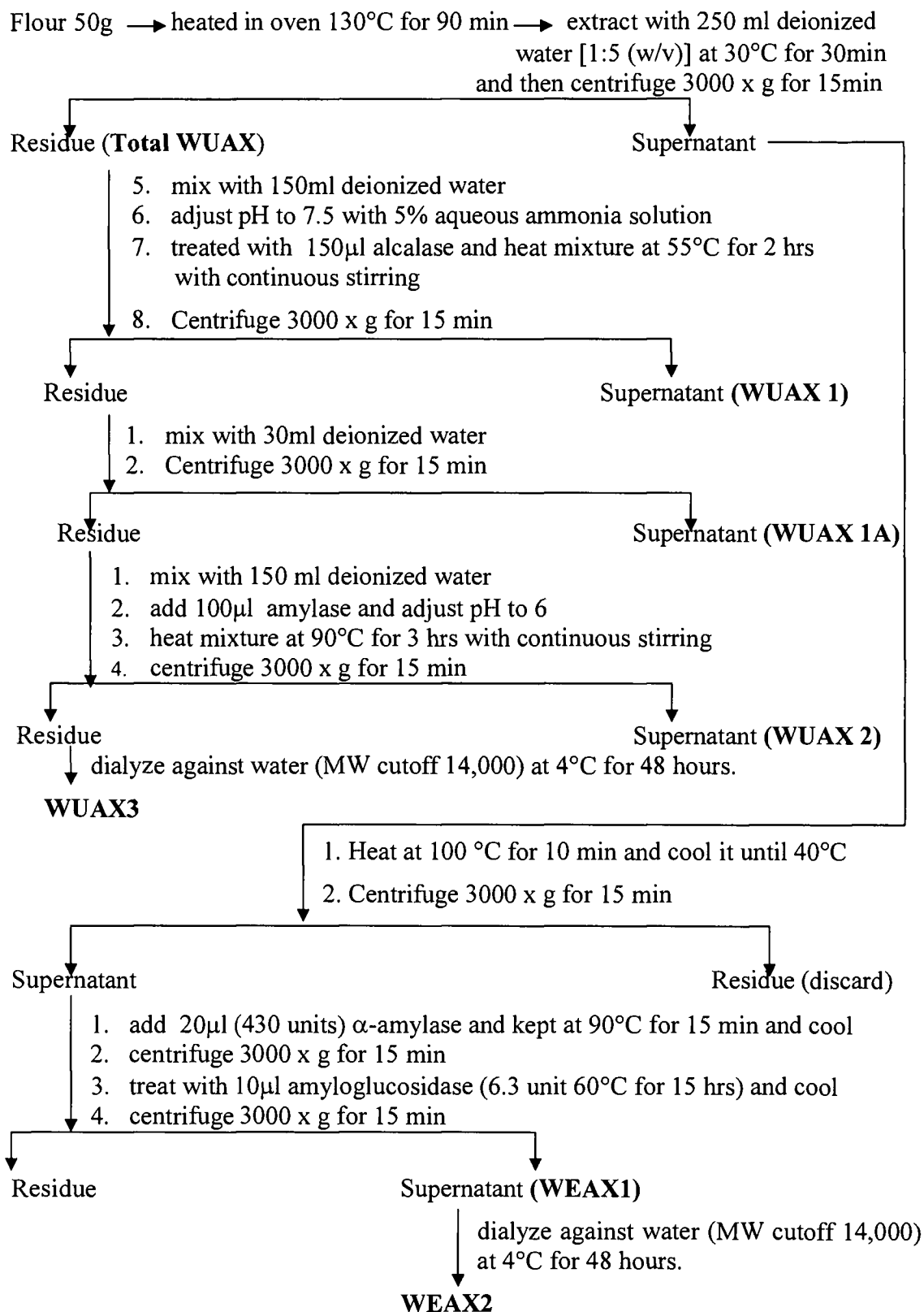


Figure 5.1 Flow scheme of extraction procedure used for isolation of WUAX and WEAX from 50 g of a commercial all-purpose flour

Faurot et al (1995) discarded the WUAX 1 and WUAX 2 fractions from Figure 5.1. These are the supernatant fractions that had been treated with protease and amylase, respectively. However, in this study these two fractions were observed using SEHPLC during the method development. A high molecular weight (HMW) component was found to be present in both these fractions. As a working hypothesis, it was initially assumed that this HMW fraction was AX. On the other hand, SEHPLC detected only very small amounts of AX in the WUAX 1A and WUAX 3 fractions (Figure 5.1). These fractions were not further analyzed.

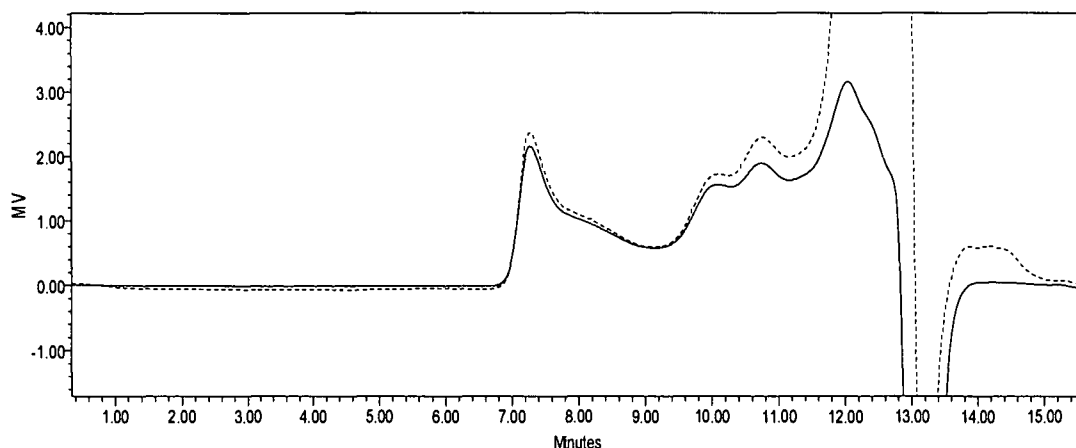


Figure 5.2 Gel permeation profiles of the WEAX fraction obtained from of a commercial all-purpose flour used during method development before (---) and after (—) dialysis

The solutions containing the WEAX, WUAX 1 and WUAX 2 fractions were tested with I₂/KI solution to detect potential soluble starch contamination according to method of Stoddard and Sarker (2000). It was observed that there was no difference in the yellow color of these three fractions compared to blank after I₂/KI addition. It was concluded that there was no soluble starch in all fractions. Accordingly, the HMW material detected in the WUAX 1 and WUAX 2 fractions was presumed to be mostly AX with a small amount of arabinogalactan (AG).

AX fractions produced at this stage were not purified. Loosveld et al (1997) purified AX by adding ethanol at 96%, 80%, and 60 % concentrations. This separated AX from other substances such as starch and AG. In the revised method used here, as a result of the use of α -amylase, it was thought that starch was unlikely to be present in AX fractions. Hence, only AG might be separated from AX by this fractionation method. Consequently, AX were not fractionated by ethanol. In addition, ethanol precipitated and freeze dried wheat flour AX are particularly difficult to resolubilize. The potential outcome is that the largest molecules would have been left out of solution, potentially making the molecular weight distribution analyses invalid. It was also considered that low molecular weight contaminating molecules would be effectively separated in the SEHPLC runs. As a result, the crude, unpurified, AX fractions were used to study molecular weight distributions. The complete modified extraction method for AX was developed and used as shown in figure 3.1.

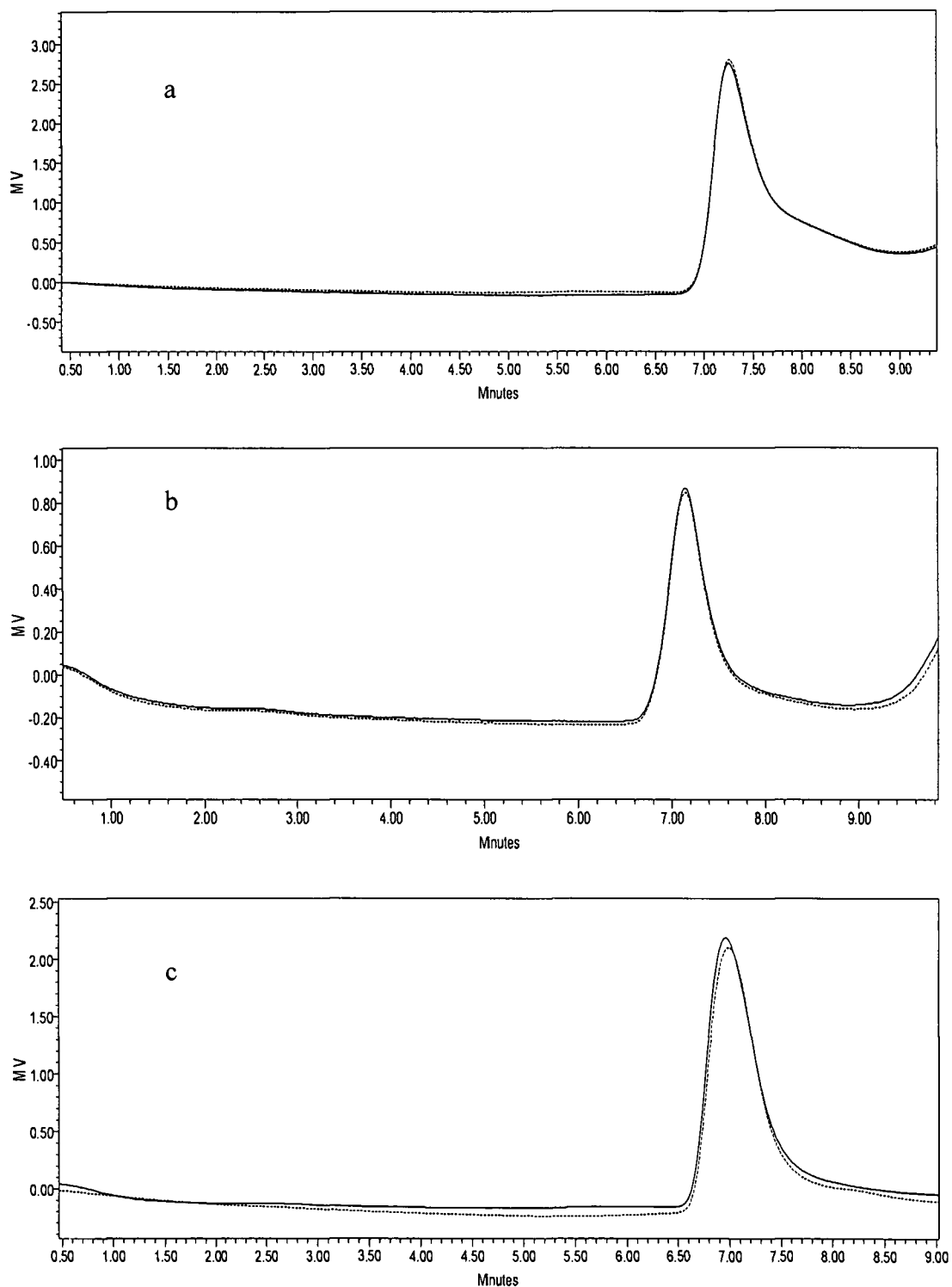


Figure 5.3 Gel permeation profiles of WEAX (a), WUAX 1 (b) and WUAX 2 (c) fractions extracted from the 10 g (—) and 50 g (- -) extraction procedures of a commercial all-purpose flour

In order to validate the revised small scale AX extraction, the fractions obtained were run on SEHPLC (Figure 5.3). Equivalent injection volumes from the large scale and small scale extracts were used. It was shown that there is very little difference in the yield of AX in all AX fractions (WEAX, WUAX1 and WUAX2) obtained from commercial all-purpose flour using either 10 g or 50 g of flour. Accordingly, it was considered appropriate to proceed using the revised 10 g extraction procedure (Figure 3.1) for the extraction of AX.

Six wheat flour samples with high and six wheat flour samples low AX content (Appendix B) were chosen to extract WEAX and WUAX. These were studied to determine their molecular weight distributions using SEHPLC. Figure 5.4 shows typical molecular weight distributions of AX fractions. WUAX 2 had the highest molecular weight, followed by WUAX1, and then WEAX. This may be as a result of a higher degree of substitution in WUAX (Izydorczyk et al, 1991). The higher molecular weight of the WUAX 2, in part, may explain its relative insolubility in water.

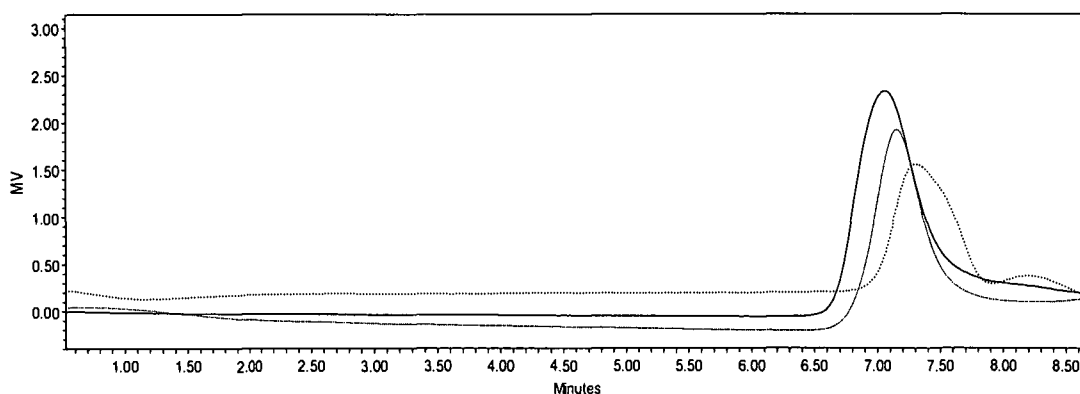


Figure 5.4 Gel permeation profiles of WEAX (····), WUAX 1 (---) and WUAX2 (—) fractions obtained from a commercial all-purpose flour

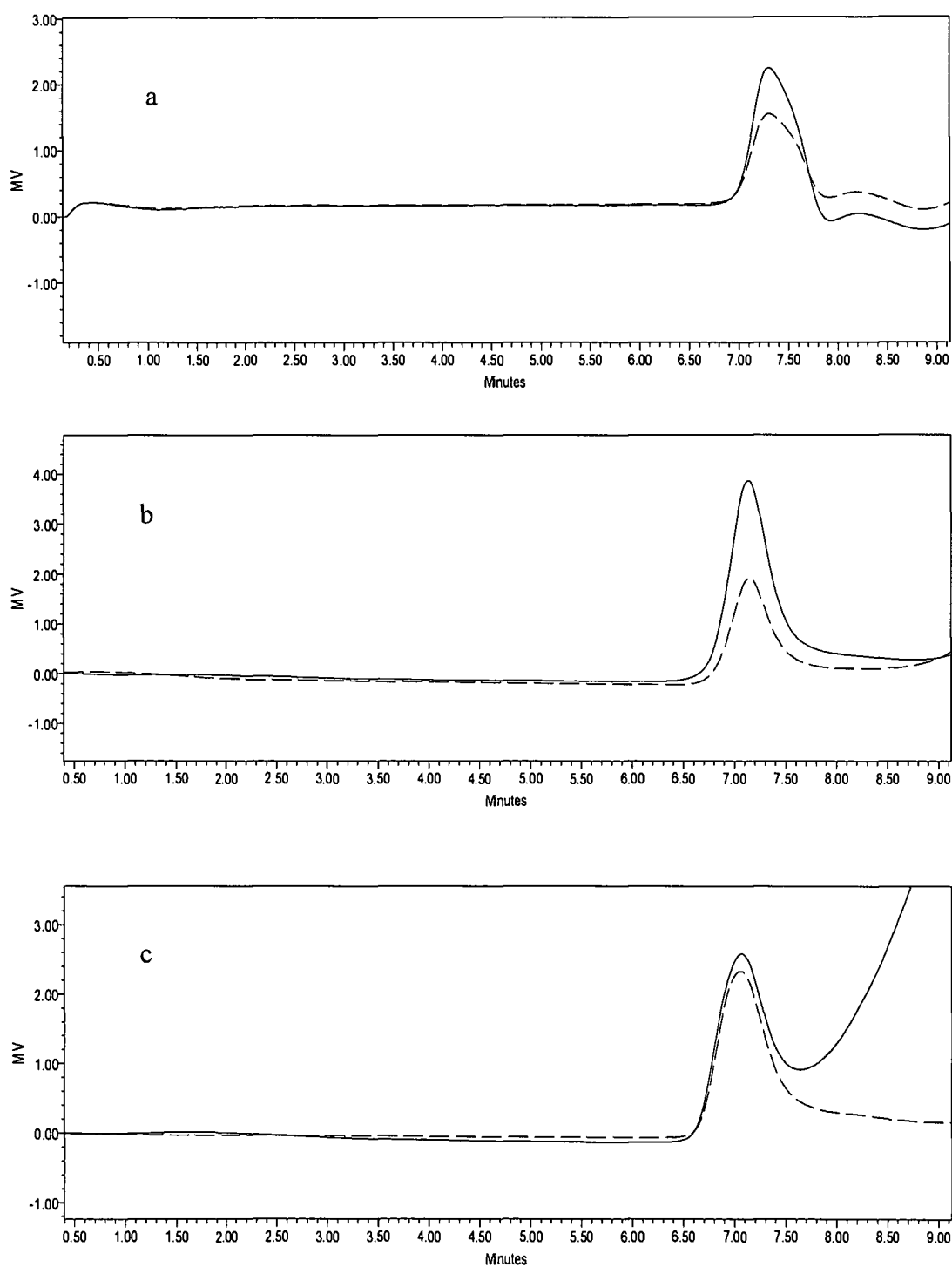


Figure 5.5 Gel permeation profiles of (a) WEAX, (b) WUAX 1 and (c) WUAX 2 fractions obtained from flour samples with highest (ORH010377; —) and lowest (ORH010085; ---) AX content

From Figure 5.5, it was shown that there were no major differences in molecular weight distributions between the samples with highest and lowest AX contents in all three fractions (WUAX 1, WUAX 2 and WEAX). However, samples with the highest total AX had higher abundances of WEAX, WUAX 1, and WUAX 2 than those with the lowest total AX, when separated on SEHPLC.

Tables 5.1, 5.2 and 5.3 show sugar compositions, AX content and A/X ratio of WEAX, WUAX1 and WUAX2 obtained from ion moderated partition column HPLC. High amounts of glucose were observed in every fraction. This glucose probably derived from starch originally present in AX fractions, especially the WUAX 2 fraction, which occurred after the amylase treatment (Figure 3.1). This agrees with Faurot et al (1995). In their extraction method, crude AX fractions also contained high amounts of glucose in AX fractions, especially in crude WEAX fraction (Figure 5.1). Therefore, in future it would be recommended that ethanol fractionation procedure should be used to purify AX fractions further before making monosaccharide analyses.

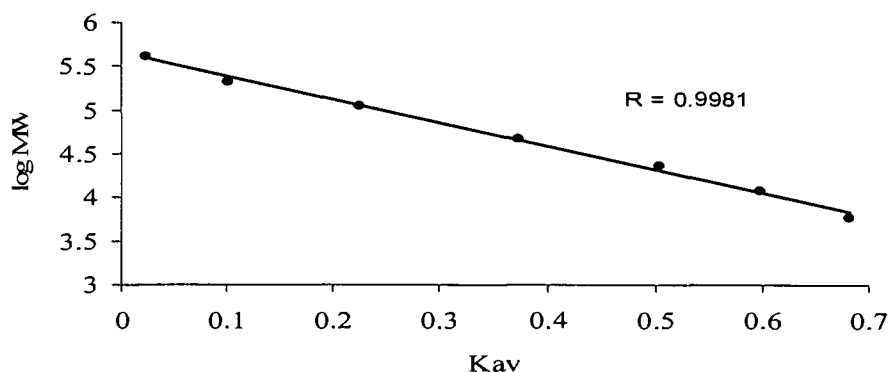


Figure 5.6 MW standard curve derived from $K_{av} = (V_e - V_o) / (V_t - V_o)$ using Shodex P-82 pullulan standards with molecular weights of 40.4×10^4 , 21.2×10^4 , 11.2×10^4 , 4.73×10^4 , 2.28×10^4 , 1.18×10^4 and 0.59×10^4 . V_e = elution volume, V_o = void volume and V_t = total bed volume

Figure 5.6 shows the correlation of the volumetric distribution coefficient (K_{av}) and Shodex standard P-82 pullulans that were used for molecular weight identification. As shown in tables 5.1, 5.2 and 5.3, molecular weights of WEAX ranged from 411,305 to 447,282. This was in some agreement with the results of Cui (2001) and Courtin et al (1999). Cui (2001) found that the molecular weight of endosperm WEAX was around 231,000 to 315,000. Courtin et al (1999) found the molecular weight of endosperm WEAX to be around 501,000 and 530,000. However, molecular weights of WUAX 1 and WUAX 2 could not be specifically defined in this study because their MWs were well above the MW of the largest pullulan standard (404,000) and extrapolation of the standard curve even further than was done for WEAX was not considered to be valid. In addition, the column exclusion limit of about 1,000,000 caused the WUAX fractions to elute very near the void volume. To identify molecular weights of WUAX 1 and WUAX 2, a column with a higher size exclusion limit should be used along with a larger set of MW standards. Use of a column with a larger exclusion was explored in this study, but time constraints precluded the incorporation of those results. It is recommended that these fractions be analyzed using SEHPLC with a column exclusion limit of at least 2,000,000.

Table 5.1
Monosaccharide composition and molecular weight (MW) of WEAX extracted from the 6 breeding lines with highest AX content
and the 6 breeding lines with lowest AX content as determined by wet chemical method

Variety	Glucose (%) ⁵	Xylose (%) ⁵	Galactose (%) ⁵	Arabinose (%) ⁵	AX ¹ (%) ⁵	A/X ²	AG ³ (%) ⁵	A/G ⁴	Retention peak time (min)	Peak area ($\mu\text{V} \cdot \text{sec}$)	MW
Low Total AX											
ORH010085	0.73	0.20	0.11	0.16	0.37	0.80	0.28	1.45	7.31	40122	424483
ORH011141	0.59	0.13	0.09	0.12	0.26	0.94	0.22	1.29	7.26	48797	447282
ORH010830	0.76	0.21	0.13	0.18	0.40	0.86	0.32	1.44	7.30	37459	425860
ORH010920	0.87	0.20	0.11	0.17	0.38	0.85	0.29	1.54	7.27	70962	440270
ORH010652	0.68	0.19	0.09	0.14	0.34	0.71	0.23	1.55	7.33	30330	411305
ORH010083	0.70	0.20	0.11	0.16	0.37	0.80	0.28	1.45	7.31	37429	420799
High Total AX											
ORH011481	0.70	0.20	0.11	0.17	0.37	0.82	0.29	1.49	7.31	28619	424231
ORH011090	0.59	0.19	0.10	0.14	0.33	0.72	0.24	1.43	7.32	33552	417398
ORH010385	0.81	0.29	0.11	0.19	0.49	0.66	0.31	1.82	7.30	27827	425242
ORH012183	0.61	0.25	0.09	0.16	0.42	0.63	0.25	1.85	7.32	20650	418723
ORH011450	0.74	0.21	0.11	0.16	0.38	0.75	0.28	1.49	7.29	59817	433604
ORH010377	0.66	0.25	0.09	0.15	0.41	0.61	0.25	1.71	7.31	37786	422071
LSD ⁶	NS	0.04	0.02	0.03	0.07	0.04	0.05	0.05	0.04	NS	17236

¹AX (Arabinoxylan, %)= 0.88 x (Arabinose + Xylose).

²A/X= Arabinose (%) /Xylose (%).

³AG (Arabinogalactan, %) = 0.87 x (Arabinose + Galactose)

⁴A/G= Arabinose (%) /Galactose (%)

⁵On a 14 % flour moisture basis (n = 24).

⁶LSD = Fisher's least significant difference (P < 0.05) for comparison of means within a column.

Table 5.2
Monosaccharide composition and molecular weight (MW) of WUAX 1 extracted from the 6 breeding lines with highest AX content
and the 6 breeding lines with lowest AX content as determined by wet chemical method

Variety	Glucose (%) ⁵	Xylose (%) ⁵	Galactose (%) ⁵	Arabinose (%) ⁵	Mannose (%) ⁵	AX ¹ (%) ⁵	A/X ²	AG ³ (%) ⁵	A/G ⁴	Retention peak time (min)	Peak area ($\mu\text{V}\cdot\text{sec}$)	MW
Low Total AX												
ORH010085	16.68	0.17	0.08	0.06	0.05	0.24	0.38	0.15	0.84	7.15	52304	>404000
ORH011141	15.97	0.10	0.07	0.06	0.38	0.16	0.59	0.14	0.79	7.14	35979	>404000
ORH010830	19.57	0.14	0.09	0.07	0.58	0.22	0.53	0.17	0.79	7.15	54122	>404000
ORH010920	14.90	0.12	0.11	0.08	0.25	0.20	0.63	0.19	0.81	7.14	53248	>404000
ORH010652	19.96	0.19	0.10	0.08	0.05	0.27	0.40	0.18	0.80	7.15	63309	>404000
ORH010083	21.27	0.19	0.10	0.07	0.05	0.27	0.38	0.17	0.75	7.15	54988	>404000
High Total AX												
ORH011481	20.17	0.19	0.10	0.11	0.04	0.30	0.58	0.22	1.06	7.16	98689	>404000
ORH011090	14.03	0.13	0.09	0.07	0.49	0.20	0.56	0.17	0.96	7.15	57622	>404000
ORH010385	18.99	0.27	0.10	0.11	0.05	0.39	0.41	0.21	1.16	7.16	109819	>404000
ORH012183	25.78	0.26	0.12	0.08	0.03	0.35	0.33	0.21	0.72	7.19	82991	>404000
ORH011450	18.65	0.16	0.09	0.08	0.67	0.24	0.52	0.17	0.92	7.14	78240	>404000
ORH010377	23.82	0.26	0.11	0.09	0.05	0.36	0.35	0.21	0.82	7.14	94537	>404000
LSD ⁶	4.80	0.03	NS	0.01	0.35	0.04	0.07	NS	0.12	0.02	14424	-

¹AX (Arabinoxylan, %)= 0.88 x (Arabinose + Xylose).

²A/X= Arabinose (%) /Xylose (%).

³AG (Arabinogalactan, %) = 0.87 x (Arabinose + Galactose)

⁴A/G= Arabinose (%) /Galactose (%)

⁵On a 14 % flour moisture basis (n =24).

⁶LSD = Fisher's least significant difference (P < 0.05) for comparison of means within a column.

Table 5.3
Monosaccharide composition and molecular weight (MW) of WUAX 2 extracted from the 6 breeding lines with highest AX content
and the 6 breeding lines with lowest AX content as determined by wet chemical method

Variety	Glucose (%) ⁵	Xylose (%) ⁵	Galactose (%) ⁵	Arabinose (%) ⁵	Mannose (%) ⁵	AX ¹ (%) ⁵	A/X ²	AG ³ (%) ⁵	A/G ⁴	Retention peak time (min)	Peak area ($\mu\text{v} \cdot \text{sec}$)	MW
Low Total AX												
ORH010085	24.65	0.10	0.12	0.04	0.04	0.14	0.35	0.16	0.30	7.05	749089	>404000
ORH011141	26.22	0.12	0.13	0.03	0.01	0.15	0.26	0.17	0.23	7.02	88529	>404000
ORH010830	24.00	0.12	0.12	0.04	0.01	0.16	0.34	0.17	0.32	7.03	105757	>404000
ORH010920	26.36	0.12	0.14	0.04	0.01	0.17	0.34	0.18	0.31	7.03	121915	>404000
ORH010652	26.92	0.11	0.12	0.03	0.04	0.15	0.28	0.16	0.27	7.08	64142	>404000
ORH010083	25.64	0.10	0.11	0.04	0.05	0.14	0.35	0.15	0.32	7.10	48296	>404000
High Total AX												
ORH011481	24.44	0.13	0.11	0.04	0.02	0.17	0.31	0.16	0.36	7.08	46888	>404000
ORH011090	26.66	0.13	0.12	0.04	0.01	0.17	0.29	0.16	0.31	7.03	98497	>404000
ORH010385	25.57	0.14	0.11	0.04	0.01	0.18	0.31	0.15	0.40	7.05	92016	>404000
ORH012183	23.77	0.12	0.10	0.04	0.06	0.16	0.33	0.14	0.38	7.12	65186	>404000
ORH011450	25.81	0.13	0.12	0.04	0.01	0.17	0.27	0.16	0.31	7.13	34261	>404000
ORH010377	23.88	0.12	0.10	0.04	0.06	0.16	0.31	0.15	0.38	7.14	49812	>404000
LSD ⁶	1.34	0.01	0.01	NS	0.02	0.02	0.06	0.01	0.09	0.07	23396	-

¹AX (Arabinoxylan, %)= 0.88 x (Arabinose + Xylose).

²A/X= Arabinose (%) /Xylose (%).

³AG (Arabinogalactan, %)= 0.87 x (Arabinose + Galactose)

⁴A/G= Arabinose (%) /Galactose (%)

⁵On a 14 % flour moisture basis (n = 24).

⁶LSD = Fisher's least significant difference (P < 0.05) for comparison of means within a column.

The arabinose-to-xylose ratio is related to degree of substitution of arabinose on xylose backbone chain. The degree of substitution is also related to the solubility of AX in water. In this sample set, WEAX contained higher degrees of substitution than WUAX1 and WUAX2 (Tables 5.1 to 5.3). These results agreed with Basic and Stone (1981). They found that AX extracted with water contained higher substitution of arabinosyl residues than AX extracted with alkali. These alkali extracted AX correspond to the WUAX fractions observed in this study. On the other hand, Mares and Stone (1973) found similar arabinose-to-xylose ratios of AX extracted with both water and alkali. However, these studies, and the results of this study conflicted with more recent AX studies. Recent studies mostly used hard wheat and observed higher degrees of substitution in WUAX (Michniewicz et al, 1990; Izydorczyk et al, 1991). However, soft white wheats, which have different genetic backgrounds to hard wheats, were used in this study. This difference may affect relative degrees of substitution of WEAX and WUAX. From previous reports, it appears that there is still disagreement on the degree of substitution of arabinose on xylose backbone chains of both WEAX and WUAX.

¹H-nuclear magnetic resonance (NMR) spectroscopy was used by Loosveld et al (1997) to characterize the substitution of arabinose on the xylose backbone. The structure of AX was not specifically addressed in this study. However, it will be interesting to use ¹H-NMR to distinguish the structure of soft wheat AX in the future.

Chapter 6

Results and Discussion: Chinese white salted raw noodles

Table 6.1 shows linear correlations between flour protein, starch damage, FSV and RVA peak viscosity, and noodle cooking characteristics of 63 soft white wheat breeding lines noodle cooking and flour characteristics. Flour protein was weakly and negatively correlated to optimum water absorption of noodle dough. When flour protein is higher, the dough develops more quickly and uses less water to achieve optimum handling characteristics. Park and Baik (2002) and Oh et al (1986) also found that higher protein content required more water for optimum noodle making characteristics.

Flour swelling volume (FSV) was negatively correlated to optimum cooking time of noodles (Table 6.1). FSV is related to the swelling of starch. When starch easily and speedily absorbs water during cooking, it swells more quickly, FSV is increased, and as a consequence, noodles should be softer at a fixed cooking time. FSV has been shown to be negatively related to the gelatinization temperature (Konik et al, 1992). This suggests that decreased gelatinization temperature was related to the decreased optimum cooking time of noodles observed here.

RVA and FSV tests can both be used to predict noodle texture. Superior quality of Japanese and Korean salted noodles is related to high starch paste peak viscosity, low gelatinization temperature, high breakdown and high swelling volume (Konik et al, 1992; Baik and Lee, 2003). WEAX were positively correlated with both FSV and

RVA peak viscosity. In the RVA at least, the WEAX compete for water and could make the starch suspension more concentrated. This effect, as well as the highly viscous nature of AX solutions may enhance RVA peak viscosity. The mechanistic impact of higher WEAX in the FSV test is not clear. However, as the FSV test is an excess water test and all components can hydrate fully, then it seems that AX themselves may swell to some extent and contribute to overall flour swelling. FSV was also negatively related to starch damage in this study. As increased RVA peak viscosity and FSV are influenced by damaged starch and WEAX, these components may interfere with the predictive capacity of these tests for noodle texture. Interestingly, RVA peak viscosity, which is usually related to starch characteristics, was positively related to lactic acid SRC and flour protein content in this study, indicating a contribution to hot paste viscosity by these components as well.

Noodle cooking loss showed highly negative correlations with lactic acid SRC, flour protein content, and RVA peak viscosity. Both high gluten strength and protein content are responsible for the strength and extent, respectively, of the gluten network formed during mixing and sheeting in the dough. If the gluten network was stronger (higher lactic SRC), soluble components and noodle fragments were lost in smaller amounts in cooking water. Moreover, high protein content had an influence on the extent of protein network available to trap these components in the noodle during cooking. At the very low protein contents of the flour used in this study, the relative importance of protein composition becomes greater. Lactic acid SRC, as an indicator of HMWGS composition, show that higher amounts of HMWGS are related to lower

cooking loss as a factor of protein network formation in the noodle sheet. RVA peak viscosity is related to starch characteristics. The higher RVA viscosity was related to the lower gelatinization temperature, and lower cooking loss. The more highly swollen starch granules from the high RVA peak viscosity flours may form a more continuous gel on the surface of the cooked noodle that restricts loss of solids. In addition, WEAX content was also negatively correlated with cooking loss. Because WEAX can readily hold high quantities of water this may also restrict the loss of materials such as amylose in water during cooking process

Table 6.1

**Correlation coefficients between flour protein, starch damage, FSV and RVA peak viscosity,
and noodle cooking characteristics of 63 soft white wheat breeding lines¹**

	Flour Swelling Volume (ml/g) ³	RVA ² Peak Viscosity (RVU) ³	Noodle Characteristics			
			Optimum Water Absorption (%) ³	Optimum Cooking Time (min)	Cooking Yield (%) ³	Cooking Loss (%) ³
Flour protein (%) ³	NS	0.182*	-0.214*	NS	NS	-0.452***
Starch damage (%) ³	-0.269**	NS	NS	NS	NS	NS
Arabinoxylan ⁴						
Total (%) ³	NS	NS	NS	NS	NS	NS
WEAX (%) ³	0.266**	0.230**	NS	NS	NS	-0.209*
WUAX (%) ³	NS	NS	NS	NS	NS	NS
Solvent retention capacity						
H ₂ O (%) ³	NS	NS	0.321**	NS	NS	NS
Na ₂ CO ₃ (%) ³	NS	NS	0.202*	NS	NS	NS
Sucrose (%) ³	NS	NS	NS	NS	NS	NS
Lactic acid (%) ³	NS	0.408***	NS	NS	NS	-0.511***
Flour swelling volume (ml/g) ³	-	0.360***	NS	-0.182*	NS	NS
RVA peak viscosity (RVU) ³	-	-	NS	NS	NS	-0.391***

¹ *, ** and ***= Correlation coefficient is significant at P<0.05, 0.01 and 0.001, respectively. NS: P>0.05

² RVA=Rapid Viscosity Analyzer

³ On a 14 % flour moisture basis (n = 125)

⁴ WEAX= water extractable arabinoxylan and WUAX= water unextractable arabinoxylan.

Table 6.2
Correlation coefficients among TPA texture parameter of noodles cooked for
a fixed cooking time of 5 min using 63 soft white wheat breeding lines¹

Texture Parameters	Hardness (g)	Adhesiveness (g*sec)²	Springiness	Cohesiveness	Chewiness
Adhesiveness (g*sec) ²	0.363***				
Springiness	NS	-0.677***			
Cohesiveness	NS	-0.231**	0.358***		
Chewiness	0.948***	0.180*	0.262**	0.444***	
Resilience	NS	-0.547***	0.547**	0.816***	0.366***

¹ *, ** and *** = Correlation coefficient is significant at P<0.05, 0.01 and 0.001, respectively.

NS: P>0.05 (n = 125)

² Multiplied by -1 to change from negative to positive values so that high values = more sticky

Table 6.2 shows linear correlations among TPA texture parameter of noodles cooked for a fixed cooking time of 5 min. Hardness was highly and positively correlated with adhesiveness and chewiness. For chewiness, this was not surprising because chewiness is the mathematical product of hardness, springiness, and cohesiveness. Generally, adhesiveness is used to measure the degree of stickiness of noodle and is expressed by the negative area of TPA profile (Figure 2.6). Adhesiveness was negatively correlated to springiness, cohesiveness and resilience but positively related to chewiness and hardness. From this, less adhesive noodles were more cohesive, springy and resilient, and less hard and chewy. Cohesiveness was highly correlated with resilience due to the similar ability of these parameters to indicate recovery of original shape after compression. The area 2 to area 1 ratio of cohesiveness gave almost identical information to the area 5 to area 4 ratio of resilience so that these two parameters were also strongly related to each other ($r = 0.816$, $p < 0.001$, Table 6.2).

Table 6.3
Means, ranges, and standard deviations of noodle texture parameters at
fixed (5 min) and optimum (opt) cooking times from 63 soft white wheat breeding lines

Noodle Texture Parameters	Mean		Range		SD	
	Fixed	Opt	Fixed	Opt	Fixed	Opt
t_0^1						
Hardness (g)	658.0	653.7	267.8	259.9	45.2	42.7
Adhesiveness (g*sec) ⁴	14.3	12.8	20.8	17.3	3.7	3.2
Springiness	1.0	1.0	0.1	0.1	0.01	0.01
Cohesiveness	0.7	0.7	0.07	0.1	0.01	0.01
Chewiness	414.9	413.8	189.8	191.0	32.0	30.6
Resilience	0.4	0.4	0.12	0.1	0.02	0.02
t_{15A}^2						
Hardness (g)	693.1	689.8	264.4	310.5	53.9	53.4
Adhesiveness (g*sec) ⁴	20.3	18.0	27.2	26.8	5.1	5.3
Springiness	0.9	0.9	0.07	0.07	0.01	0.01
Cohesiveness	0.6	0.6	0.1	0.1	0.02	0.02
Chewiness	402.2	401.2	186.1	179.7	35.2	35.5
Resilience	0.4	0.4	0.1	0.1	0.02	0.03
t_{15W}^3						
Hardness (g)	618.6	622.2	275.1	290.9	49.8	50.7
Adhesiveness (g*sec) ⁴	13.8	11.6	30.0	18.2	4.6	3.8
Springiness	1.0	1.0	0.3	0.1	0.03	0.02
Cohesiveness	0.6	0.6	0.2	0.1	0.02	0.02
Chewiness	356.5	363.2	211.1	187.3	38.2	35.1
Resilience	0.4	0.4	0.1	0.1	0.02	0.02

¹Test immediately after cooling 1 min in water following cooking (n =125)

²Kept in plastic container without water for 15 min before testing (n =125)

³Kept in plastic container containing water for 15 min before testing (n =125)

⁴Multiplied by -1 so that high values = more sticky

Table 6.3 shows Means, ranges, and standard deviations of noodle texture parameters at fixed (5 min) and optimum (opt) cooking times from 63 soft white wheat breeding lines. Fixed cooking time was 5 minutes. Average of optimum cooking time was 5 minutes and 15 seconds. Noodle texture values, especially hardness, adhesiveness and chewiness at t_{15A} were significantly higher than those at t_0 (Table 6.3). However, when noodles were kept in water for 15 min (t_{15W}), noodles were softer, less adhesive, and less chewy than both t_0 and t_{15A} . This was confirmed by the significant F-values from ANOVA for differences in storage methods for all the texture parameters (Table 6.4).

Table 6.4 also shows the effects of varieties and cooking methods on noodle texture parameters. Wheat varieties showed highly significant effects on all noodle texture parameters. Significant differences between optimum and fixed cooking time were observed only for adhesiveness, springiness and resilience. The varieties by storage methods interactions were significant for hardness, adhesiveness and springiness. The cooking methods by storage methods interactions were significant for cohesiveness, chewiness and resilience. This suggested that storage methods influenced noodle texture parameters. Consequently, storage methods should be appropriately chosen for the texture analysis based on the serving customs for different noodle types. The analysis of variance did not reveal significant interaction between varieties and cooking methods except for resilience. Due to the small difference between optimum and fixed cooking times, and for the sake of clarity, only noodle texture characteristics from the fixed cooking time experiments will be further discussed. It will be useful to use only fixed cooking time for noodle testing in the future.

Table 6.4
F-value from analysis for variance of noodle nexture parameters comparing
the fixed (5 min) and optimum cooking times using 63 soft white wheat breeding lines¹

Source of Variation	df ²	Noodle Texture					
		Hardness	Adhesiveness	Springiness	Cohesiveness	Chewiness	Resilience
Varieties (V)	59	34.5***	3.9***	1.44*	10.4***	36.1***	17.1***
Cooking Methods (C) ³	1	0.8	55.7***	7.0**	3.8	0.9	41***
Storage Methods(S) ⁴	2	454.6***	219.5***	60.7***	1579.3***	621.5***	1436.7***
V x C	59	0.9	1.2	0.7	1.2	1.0	1.6**
V x S	118	1.3*	2.0***	1.9***	0.7	1.0	1.3
C x S	2	1.8	0.77	0.3	3.7*	4.0*	4.3*

¹ *, ** and ***= F-value is significant at P<0.05, 0.01 and 0.001, respectively.

²df= degree of freedom (n = 125)

³Cooking methods = fixed and optimum cooking time

⁴Storage methods = t₀ (Test immediately after cooling 1 min in water following cooking), t_{15A} (Kept in plastic container without water for 15 min before testing) and t_{15W} (Kept in plastic container containing water for 15 min before testing)

Table 6.5 shows linear correlations between flour protein, AX contents, FSV, RVA peak viscosity, solvent retention capacity tests and noodle texture parameters at fixed cooking time (5min). In the literature, flour protein content has consistently been shown to be positively correlated with hardness of cooked noodles (Miskelly, 1984; Toyokawa et al, 1989a; Baik et al, 1994; Park et al, 2003). In this study, no significant correlations were observed between flour protein content and noodle texture parameters at t_0 except for a weak negative correlation with resilience. However, flour protein content was negatively correlated with hardness, adhesiveness and chewiness but positively correlated with springiness at t_{15A} . There were also weak negative correlations between flour protein content and springiness and resilience at t_{15W} . These results were different from previous reports and might have occurred as a result of the very low flour protein contents in this sample set (about 6.4%).

In relation to potential direct effects, AX parameters were almost entirely unrelated to cooked noodle texture. However, total AX and WUAX were positively correlated with adhesiveness at t_0 (Table 6.5). This could suggest that the aqueous phase may be “gummier” and stickier as AX contents increase.

High FSV and high RVA paste peak viscosity have been reported as desirable for texture of Japanese salted noodles. FSV was negatively correlated with adhesiveness at t_0 min and negatively correlated with hardness and chewiness at t_0 , t_{15A} and t_{15W} (Table 6.5). This indicated that noodles got softer and less sticky when FSV increased and the ratio of amylopectin to amylose in the starch increased (Toyokawa et al,

1989b). RVA peak viscosity was also negatively related to hardness at t_{15A} and t_{15w} and negatively related to adhesiveness at t_{15A} again suggesting that factors leading to higher paste viscosity and higher amylopectin content were related to softer and less sticky noodles (Table 6.5). With regard to the relationships between FSV, RVA and noodle texture, this sample set generally behaved as it was expected to. RVA and FSV gave similar results in predicting noodle texture. This agrees with the common use of either RVA or FSV for this task. Accordingly, it may not have been necessary to use both in screening for noodle texture potential of flours.

Sodium carbonate SRC had positive correlations with hardness and chewiness of cooked noodles at t_0 and t_{15A} (Table 6.5). Starch damage was also positively related to hardness and chewiness at t_0 and t_{15A} (Table 6.5). The starch damage result agreed with Elbers et al (1996). In addition, sodium carbonate SRC was significantly and positively related to starch damage ($r = 0.233$, $p < 0.01$, Table 4.3), although the relationship was weak. Carbonate SRC has been promoted as a predictor of starch damage by other workers (Slade and Levine, 1994; Gaines, 2000; Guttieri et al, 2001), although Guttieri et al (2001) did not actually compare carbonate SRC directly to starch damage. The predictive value of carbonate SRC for starch damage in this sample set was not adequate. Even though there was no direct correlation between sodium carbonate SRC and FSV, a negative correlation between starch damage and FSV was observed (Table 6.1), reflecting the poor predictive capacity of carbonate SRC for starch damage observed here. Increased starch damage was related to decreased FSV, but not necessarily in a causal fashion. However, it is plausible that

physically damaged starch granules swell very early in the cooking process, either in the FSV test, or in the noodles, and subsequently collapse due to thermal energy and shear forces, leading to reduced swelling volume and a less diffuse gel that is detected as harder and chewier noodle texture.

Lactic acid SRC was positively correlated with hardness, cohesiveness, chewiness and resilience at t_0 and t_{15w} but positively correlated with only cohesiveness and chewiness at t_{15A} (Table 6.5). Lactic acid SRC is commonly positively related to both protein content, and gluten strength and composition. Due to the low protein content of the flour used in this study, protein composition could become more important than protein content in determining noodle texture. HMWGS are responsible for strong gluten network formation and when protein content is limiting, as in this instance, then the effect of the HMWGS, as inferred by the lactic acid SRC values, becomes more evident. The stronger glutenin network formation was related to the harder, chewier, and more cohesive noodles.

In conclusion, cooked noodle hardness seemed to be related to protein composition and starch damage, rather than protein content. AX seemed to relate only cooked noodle adhesiveness, which might result from their sticky properties in an aqueous solution. Both FSV and RVA were useful for predicting noodle texture but it is not necessary to use both for noodle testing.

Table 6.5
Correlation coefficients between flour protein, AX contents, FSV, RVA peak viscosity, solvent retention capacity tests
and noodle texture parameters at a fixed cooking time of 5 min, using 63 soft white wheat breeding lines¹

Noodle Texture Parameters	Flour	Arabinoxylan ²			FSV	RVA	Starch	Solvent Retention Capacity	
	Protein	Total	WEAX	WUAX		PV ³	Damage	Na ₂ CO ₃	Lactic acid
	(%) ⁴	(%) ⁴	(%) ⁴	(%) ⁴	(ml/g) ⁴	(RVU) ⁴	(%) ⁴	(%) ⁴	(%) ⁴
t₀⁵									
Hardness (g)	NS	NS	NS	NS	-0.489***	NS	0.235**	0.186*	0.240**
Adhesiveness (g*sec) ⁸	NS	0.194*	NS	0.179*	-0.205*	NS	NS	NS	NS
Springiness	NS	NS	NS	NS	NS	NS	NS	NS	NS
Cohesiveness	NS	NS	NS	NS	NS	NS	NS	0.181*	0.336**
Chewiness	NS	NS	NS	NS	-0.421***	NS	0.239**	0.225*	0.312**
Resilience	-0.228*	NS	NS	NS	NS	NS	NS	NS	0.200*
t_{15A}⁶									
Hardness (g)	-0.378***	NS	NS	NS	-0.344***	-0.242**	0.273**	0.186*	NS
Adhesiveness (g*sec) ⁸	-0.204*	NS	NS	NS	NS	-0.202*	NS	NS	NS
Springiness	0.280**	NS	NS	NS	NS	NS	NS	NS	NS
Cohesiveness	NS	NS	NS	NS	NS	NS	NS	NS	0.370***
Chewiness	-0.345***	NS	NS	NS	-0.301**	NS	0.244**	0.185*	0.203*
Resilience	NS	NS	NS	NS	NS	NS	NS	NS	NS
t_{15W}⁷									
Hardness (g)	NS	NS	NS	NS	-0.499***	-0.206*	0.268**	NS	0.259**
Springiness	-0.201*	NS	NS	NS	NS	NS	NS	NS	NS
Cohesiveness	NS	NS	NS	NS	NS	NS	NS	NS	0.291**
Chewiness	NS	NS	NS	NS	-0.383***	NS	0.219*	NS	0.327**
Resilience	-0.276**	NS	NS	NS	NS	NS	NS	NS	0.208*

¹ *, ** and ***= Correlation coefficient is significant at P<0.05, 0.01 and 0.001, respectively. NS: P>0.05

²WEAX= water extractable arabinoxylan and WUAX= water unextractable arabinoxylan.

³RVA PV=Rapid Viscosity Analyzer peak viscosity

⁴On a 14 % flour moisture basis (n = 125)

⁵Test immediately after cooling 1 min in water following cooking

⁶Kept in plastic container without water for 15 min before testing

⁷Kept in plastic container containing water for 15 min before testing

⁸Multiplied by -1 to change from negative to positive values so that high values = more sticky

Chapter 7

Results and Discussion: AX, Kernel and Milling Characteristics, and Noodles

Table 7.1 shows means, SD and ranges of WEAX, WUAX 1 and WUAX 2 content from SEHPLC (WEAX-SE, WUAX 1-SE and WUAX 2-SE) and total AX, WEAX and WUAX from the wet chemical AX content determinations. Compared to the wet chemical method, almost all of WEAX-SE content was extracted when using the procedure described in section 3.2.11. There was also a highly significant correlation between WEAX-SE and WEAX ($r = 0.802$, $p < 0.001$). This provides a high degree of confidence in the WEAX-SE results as they are well aligned with the results of the wet chemical method (Figure 7.1).

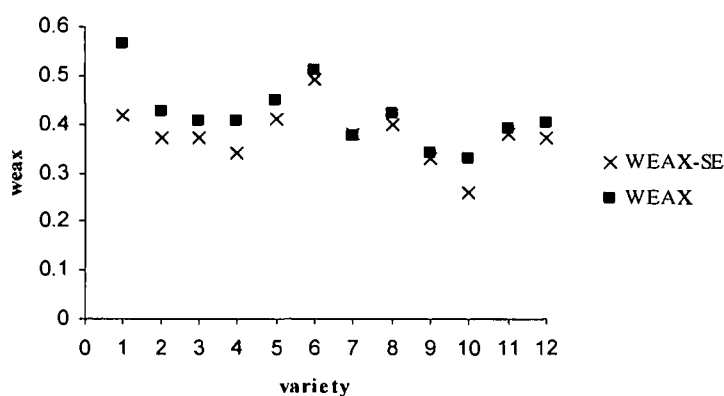


Figure 7.1 Comparison of wet chemical WEAX contents, and WEAX-SE contents. WEAX-SE contents were calculated from arabinose and xylose contents of the WEAX-SE fraction [$0.88 \times (\text{arabinose} + \text{xylose})$] of the 6 breeding lines with highest and the 6 breeding lines with lowest AX contents as determined by the wet chemical method

Table 7.1
Means, standard deviations and ranges of WEAX, WUAX 1 and WUAX 2 contents from
size exclusion chromatography HPLC (SEHPLC), and total AX, WEAX and WUAX contents
from wet chemical method of the 6 breeding lines with highest and the 6 breeding lines
with lowest AX content as determined by wet chemical method

Variety	SEHPLC			Wet Chemical Method		
	WEAX-SE ¹ (%) ²	WUAX 1-SE ¹ (%) ²	WUAX 2-SE ¹ (%) ²	Total AX (%) ²	WEAX (%) ²	WUAX (%) ²
Low Total AX						
ORH010085	0.37	0.24	0.14	1.33	0.40	0.75
ORH011141	0.26	0.16	0.15	1.40	0.33	0.89
ORH010830	0.40	0.22	0.16	1.46	0.42	0.85
ORH010920	0.38	0.20	0.17	1.48	0.38	0.91
ORH010652	0.34	0.27	0.15	1.52	0.41	0.91
ORH010083	0.37	0.27	0.14	1.52	0.43	0.89
High Total AX						
ORH011481	0.37	0.30	0.17	1.97	0.40	1.31
ORH011090	0.33	0.20	0.17	1.98	0.34	1.40
ORH010385	0.49	0.39	0.18	1.99	0.51	1.22
ORH012183	0.42	0.35	0.16	2.00	0.57	1.17
ORH011450	0.38	0.24	0.17	2.07	0.39	1.41
ORH010377	0.41	0.36	0.16	2.08	0.45	1.36
Mean	0.37	0.27	0.16	1.73	0.42	1.09
Range	0.23	0.23	0.04	0.75	0.24	0.66
SD	0.06	0.07	0.01	0.30	0.07	0.25

¹WEAX-SE = water extractable arabinoxylan from SEHPLC, WUAX 1-SE = water unextractable arabinoxylan 1 from SEHPLC, and WUAX 2-SE = water unextractable 2 from SEHPLC

²On a 14 % flour moisture basis (n =24)

On the other hand, combined yields of WUAX 1-SE and WUAX 2-SE were smaller than WUAX obtained from wet chemical method. In the wet chemical method, WUAX were calculated from the subtraction of WEAX content from total AX content. There was no significant correlation between WUAX 1-SE and WUAX ($r = 0.415$, ns) (Figure 7.2a). However, there was a significant correlation between WUAX 2-SE and WUAX ($r = 0.691$, $p < 0.05$) (Figure 7.2b). In this respect more confidence is placed on the WUAX 2-SE and WUAX from wet chemical method results than the WUAX 1-SE. However, the poor correlation between WUAX 1-SE and wet chemical WUAX

content seems to have resulted from the observation that in the WUAX 1-SE determinations, two lines (ORH011090 and ORH011450) appeared to be anomalous compared to the relationships of the SEHPLC and wet chemical methods for the other samples. Based on this observation, ORH011090 and ORH011450 were removed from this sample set and another correlation calculated. As a result, a significant correlation between WUAX 1-SE and WUAX was found ($r = 0.787$, $p < 0.01$). There was also no significant correlation between total WUAX-SE (calculated from the sum of WUAX 1- and 2-SE fractions) and WUAX ($r = 0.507$, ns) (Figure 7.2c). Again, it was found that the two lines (ORH011090 and ORH011450) were responsible for the poor correlation. If these two data points were removed again, a highly significant correlation between total WUAX-SE and WUAX was also found ($r = 0.826$, $p < 0.001$).

On the observation of plots of combined total WUAX-SE versus WUAX for all varieties, it was clear that the absolute results for WUAX-SE were lower than the WUAX from wet chemical method. However, the general ranking was still well aligned. The lower yield of WUAX-SE might be caused by the interference of glucose during monosaccharide analysis. This reinforced the idea expressed in chapter 5 that all AX fractions should be purified before monosaccharide analyses. There was insufficient time to repeat the wet chemical and monosaccharide analyses for ORH011090 and ORH011450 prior to the deadlines for this document. However, it is obvious that repeat analyses will need to be performed before attempting publication of these results.

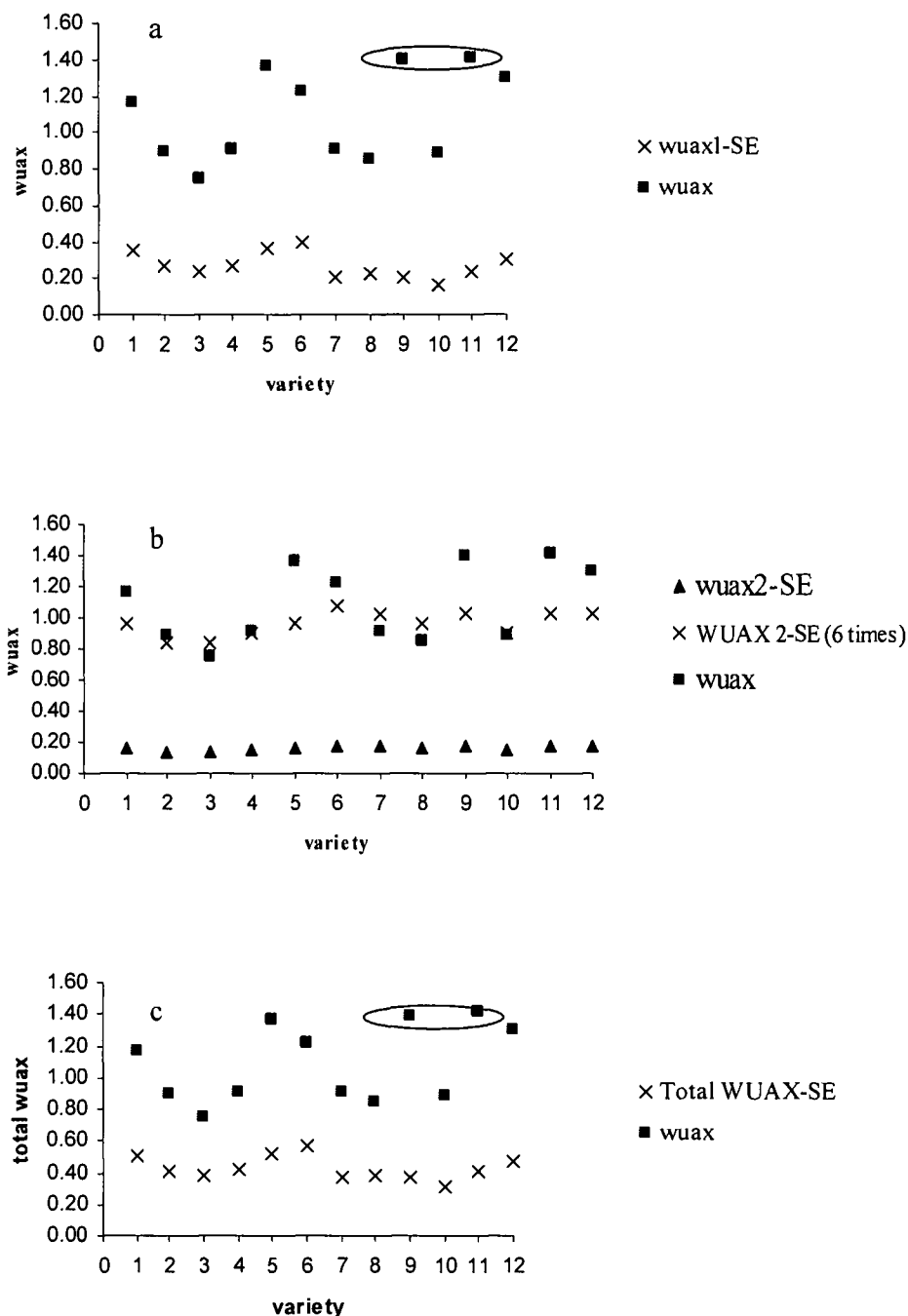


Figure 7.2 Comparison of wet chemical WUAX contents, and WUAX 1-SE (a), WUAX 2-SE (b) and total WUAX-SE (c) content s. WUAX-SE contents were calculated from arabinose and xylose contents of WUAX-SE fraction $[0.88 \times (\text{arabinose} + \text{xylose})]$ of the 6 breeding lines with highest and the 6 breeding lines with lowest AX contents as determined by the wet chemical method

Table 7.2 shows linear correlations between kernel and milling characteristics, SRCs, cookie diameter, noodle texture parameters, and xylose and arabinose contents of WEAX-SE, WUAX 1-SE and WUAX 2-SE fractions.

Xylose and arabinose contents of WEAX-SE and WUAX 1-SE fractions were positively correlated with kernel hardness index, and negatively correlated with flour yield and break flour yield (Table 7.2). Xylose content of the WUAX 2-SE fraction showed no correlation with hardness index or milling yields. However, arabinose content of the WUAX 2-SE fraction was positively correlated with kernel hardness index, and negatively correlated with flour yield and break flour yield. This repeated the pattern seen for arabinose content in the other two fractions. This suggested that higher xylose and arabinose contents simply reflected overall AX abundance and its effects on kernel texture and milling properties. The SEHPLC results are more aligned with the generally accepted model that equates higher AX abundance with harder kernel texture and poorer milling properties in soft wheats, than are the AX contents determined by wet chemistry.

Xylose and arabinose contents of WEAX-SE, WUAX 1-SE and WUAX 2-SE fractions only had significant correlations with water, sodium carbonate, and sucrose SRCs (Table 7.2). There were no significant correlations between xylose and arabinose contents of the three fractions and lactic acid SRC, as expected. Any significant relationships between xylose and arabinose contents of the three fractions and SRCs were positive. This is expected from the knowledge that pentosans absorb

large amounts of water. For WEAX-SE fraction, xylose content was more strongly related to water and carbonate SRCs than arabinose. In this fraction xylose and arabinose contents were not correlated with sucrose SRC. However, this might relate to the result showing that WEAX are simply less abundant than WUAX (Table 4.2).

For WUAX 1-SE, there was a relatively equal weighting between xylose and arabinose in their correlations with water and carbonate SRCs, but only arabinose content was related to sucrose SRC.

For WUAX 2-SE, weak but positive correlations with water and carbonate SRCs were observed only for xylose. However, in contrast to the other two fractions, xylose content was significantly and positively correlated with sucrose SRC.

The SRC tests are highly responsive to changes in kernel hardness and the attendant effects on milling. As a result, the interrelationships between AX content, xylose and arabinose contents, kernel hardness, and milling yields are further reflected in the relationship between xylose and arabinose contents and water, carbonate, and sucrose SRCs.

Table 7.2
Correlation coefficients between xylose and arabinose contents of WEAX, WUAX 1 and WUAX 2, kernel texture,
milling characteristics, solvent retention capacity tests, cookie diameter, and noodle texture parameters from
the 6 breeding lines with highest and the 6 breeding lines with lowest AX contents determined
by wet chemical method¹

Quality characteristics	WEAX-SE ²		WUAX 1-SE ²		WUAX 2-SE ²	
	Xylose (%) ⁴	Arabinose (%) ⁴	Xylose (%) ⁴	Arabinose (%) ⁴	Xylose (%) ⁴	Arabinose (%) ⁴
Hardness index	0.637**	0.670**	0.556**	0.623**	NS	0.436*
Flour yield(%)	-0.797***	-0.564**	-0.739***	-0.554**	NS	-0.430*
Break flour yield(%)	-0.672**	-0.494*	-0.600**	-0.470*	NS	-0.475*
Solvent Retention Capacity						
H ₂ O(%) ⁴	0.540**	NS	0.434*	0.592**	0.471*	NS
Na ₂ CO ₃ (%) ⁴	0.807***	0.434*	0.703**	0.604**	0.481*	NS
Sucrose (%) ⁴	NS	NS	NS	0.617**	0.689**	NS
Cookie diameter (cm)	NS	-0.481*	NS	-0.412*	-0.477*	NS
Noodle Texture Parameters						
t_0 ⁵						
Adhesiveness (g*sec) ⁷	NS	NS	0.411*	NS	NS	NS
Springiness	-0.441*	NS	-0.620**	NS	NS	NS
Resilience	NS	NS	NS	NS	NS	NS
t_{15A} ⁶						
Hardness (g)	NS	NS	NS	NS	-0.432*	NS
Resilience	NS	NS	NS	NS	-0.462*	NS

¹ *, ** and ***= Correlation coefficient is significant at P<0.05, 0.01 and 0.001, respectively.

NS: Correlation coefficient is not significant at P<0.05.

²WEAX-SE = water extractable arabinoxylan from SEHPLC, WUAX 1-SE = water unextractable arabinoxylan 1 from SEHPLC, and WUAX 2-SE = water unextractable arabinoxylan 2 from SEHPLC

³AX (Arabinoxylan, %)= 0.88 x (Arabinose + Xylose); A/X= Arabinose (%) /Xylose (%)

⁴On a 14 % flour moisture basis (n =24)

⁵Test immediately after cooling 1 min in water following cooking

⁶Kept in plastic bottle without water for 15 min before testing

⁷Multiplied by -1 to change from negative to positive values so that high values = more sticky

Arabinose contents of WEAX-SE and WUAX 1-SE fractions were weakly and negatively correlated with cookie diameter (Table 7.2). In contrast, it was the xylose content of the WUAX 2-SE fraction that was negatively related to cookie diameter. This contrast seems to be a repetition of the contrast between WUAX 1-SE and WUAX2-SE fractions for sucrose SRC. Overall, increased xylose or arabinose contents were related to harder kernel texture, poorer milling properties, and weakly related to smaller cookie diameter. However, depending on the fraction observed, the relative importance of xylose and arabinose differs (Table 7.2).

There were few significant relationships between xylose content and cooked noodle texture parameters (Table 7.2). In the few cases where significant correlations were observed there seemed to be no pattern to the relationships that might provide clues regarding any underlying causal or indirect factors. There was no significant relationship between any cooked noodle texture parameter and arabinose content in any fraction.

Table 7.3 shows linear correlations between kernel and milling characteristics, SRCs, cookie diameter, noodle texture parameters, and AX content and A/X ratio of WEAX-SE, WUAX 1-SE and WUAX 2-SE fractions.

Table 7.3
Correlation coefficients between AX contents and A/X Ratio of WEAX, WUAX 1 and WUAX 2, kernel texture, milling characteristics, solvent retention capacity tests, cookie diameter, and noodle texture parameters from the 6 breeding lines with highest and the 6 breeding lines with lowest AX content as determined by wet chemical method¹

	WEAX-SE ²		WUAX 1-SE ²		WUAX 2-SE ²	
	WEAX ³	A/X ³	WUAX1 ³	A/X ³	WUAX2 ³	A/X ³
Quality characteristics	(%) ⁴		(%) ⁴		(%) ⁴	
Hardness index	0.698**	NS	0.605**	NS	NS	NS
Flour yield(%)	-0.772***	0.602**	-0.740***	0.538**	NS	-0.455*
Break flour yield(%)	-0.658**	0.573**	-0.607**	0.468*	NS	-0.546**
Solvent Retention Capacity						
H ₂ O(%) ⁴	0.508*	-0.483*	0.493*	NS	0.469*	NS
Na ₂ CO ₃ (%) ⁴	0.720***	-0.746***	NS	-0.483*	0.483*	NS
Sucrose (%) ⁴	NS	-0.471*	0.452*	NS	0.637**	NS
Cookie diameter (cm)	-0.514*	NS	NS	NS	-0.503*	NS
Noodle Texture Parameters						
t_0^5						
Adhesiveness (g*sec) ⁷	NS	-0.595**	NS	NS	NS	NS
Springiness	NS	0.590**	-0.583**	0.542**	NS	NS
Resilience	NS	0.472*	NS	NS	NS	NS
t_{15A}^6						
Resilience	NS	NS	NS	NS	-0.405*	NS

¹ *, ** and ***= Correlation coefficient is significant at P<0.05, 0.01 and 0.001, respectively.

NS: Correlation coefficient is not significant at P<0.05.

²WEAX-SE = water extractable arabinoxylan from SEHPLC, WUAX 1-SE = water unextractable arabinoxylan 1 from SEHPLC, and WUAX 2-SE = water unextractable arabinoxylan 2 from SEHPLC

³AX (Arabinoxylan, %)= 0.88 x (Arabinose + Xylose); A/X= Arabinose (%) /Xylose (%)

⁴On a 14 % flour moisture basis (n =24)

⁵Test immediately after cooling 1 min in water following cooking

⁶Kept in plastic bottle without water for 15 min before testing

⁷Multiplied by -1 to change from negative to positive values so that high values = more sticky

AX contents WEAX-SE and WUAX 1-SE fractions were positively correlated with kernel hardness index, and negatively correlated with flour yield and break flour yield (Table 7.3). However, WUAX 2-SE content showed no correlations with hardness index, flour yield and break flour yield (Table 7.3). These trends were similar to xylose and arabinose contents, as mentioned above. These relationships were also similar to the correlations between wheat kernel and milling characteristics, and total AX and WEAX contents as measured by the wet chemical method (Table 4.3). Overall these results indicate that in this sample set, any measure of AX abundance, except in the case of the WUAX 2-SE fraction, was to some extent able to predict kernel hardness and milling properties.

WEAX-SE content was positively related to water and sodium carbonate SRCs (Table 7.3). This partly agreed with the previous results that found strong correlations between WEAX from wet chemical method and all SRCs (Table 4.3). This was also in alignment with the relationships between xylose content of WEAX- SE fraction and water and carbonate SRCs.

WUAX 1-SE content showed weak but positive correlations with water and sucrose SRC (Table 7.3). This doesn't line up with the arabinose and xylose results from the same fraction (Table 7.2).

WUAX 2-SE content was positively correlated with water, sodium carbonate and sucrose SRCs (Table 7.3). This conflicted with the previous result (Table 4.3), which

observed a correlation of WUAX determined by the wet chemical method with only water SRC. However, it agrees with the results of the xylose content from the same fraction. The general alignment of relationships between the xylose, arabinose, and combined AX contents from each fraction and the milling and SRC results, indicate that the measured xylose, arabinose contents reflect AX abundance rather than any factors related to polymer size.

Cookie diameter also showed negative correlations with the AX contents of the WEAX-SE and WUAX 2-SE fractions. This agreed with the previous results (Table 4.3) and helped to confirm the effect of WEAX and WUAX on cookie diameter (Bettge and Morris, 2000). However, it is interesting that it was the abundance of WUAX 2-SE fraction that was related to lower cookie diameter and not the WUAX 1-SE fraction.

There were few significant relationships between AX contents of the three fractions noodle texture parameters (Table 7.3). In the few cases where significant correlations were observed there again seemed to be no pattern to the relationships.

Arabinose-to-xylose (A/X) ratios of the WEAX-SE, WUAX 1-SE and WUAX 2-SE fractions showed no correlations with kernel hardness index (Table 7.3). However, A/X ratios of WEAX-SE and WUAX 1-SE fractions were positively correlated with flour yield and break flour yield. In contrast, A/X ratio of WUAX 2-SE fraction was negatively correlated with flour yield and break flour yield (Table 7.3). This suggested

that lower arabinose substitution of WEAX-SE and WUAX 1-SE was related to poorer milling characteristics. It also suggested, somewhat surprisingly, that higher arabinose substitution was related to poorer milling characteristics in the WUAX 2-SE fraction. This result tends to reflect the emerging pattern which suggests more similarity between the WEAX-SE and WUAX 1-SE fractions, than between the two WUAX-SE fractions. These results create more questions than answers.

A/X ratio of WEAX-SE fraction was negatively correlated with water, sodium carbonate and sucrose SRCs (Table 7.3). A/X ratio of WUAX 1-SE fraction was negatively correlated with only sodium carbonate SRC. A/X ratio of WUAX 2-SE fraction was not correlated with any SRC. In the cases of the WEAX-SE and WUAX 1-SE fractions, where there was a significant relationship, higher arabinose was related to lower solvent retention. The interrelationships between WEAX-SE and WUAX 1-SE AX contents, milling yields, and SRCs appear to be influenced by the degree of substitution of xylose by arabinose. Again, the WUAX 2-SE fraction shows divergent behavior with respect to the other two fractions. Arabinose substitution ratio showed no significant relationships with cookie diameter. This suggests that AX abundance, and maybe MW, have more influence on cookie diameter than arabinose substitution levels.

A/X ratio of WEAX-SE and WUAX 1-SE fractions showed positive correlations with springiness of cooked noodles at t_0 (Table 7.3). In addition, A/X ratio of WEAX-SE show negative correlation with adhesiveness of cooked noodles at t_0 (Table 7.3).

Table 7.4 shows linear correlations between kernel and milling characteristics, SRCs, cookie diameter, noodle texture parameters, and AG content and A/G ratio of WEAX-SE, WUAX 1-SE and WUAX 2-SE fractions.

AG contents of WEAX-SE and WUAX 1-SE fraction were positively correlated with kernel hardness index (Table 7.4). AG content of WUAX 1-SE fraction was also negatively correlated with flour yield and break flour yield (Table 7.4). This was somewhat similar to results seen for AX contents of WEAX-SE and WUAX 1-SE fractions. This suggested that AG abundance might be related to AX abundance, and so also to wheat kernel and milling characteristics. However, these relationships need to be studied more in the future.

AG content of WEAX-SE fraction had no correlation with any SRCs (Table 7.4). AG content of WUAX 1-SE fraction was positively correlated with water and sucrose SRC. This was similar to the results of AX content of WUAX 1-SE fraction (Table 7.3). AG content of WUAX 1-SE fractions seemed to act like the equivalent AX component in that fraction. AG content of WUAX 2-SE fraction was negatively correlated with sodium carbonate SRC (Table 7.4). This was opposite to the relationship between AX content of WUAX 2-SE fraction and carbonate SRC. It is

unclear why increased AG content in the WUAX 2-SE fraction would related to reduced carbonate SRC.

AG content of WEAX-SE fraction showed a positive correlation with cohesiveness and resilience of cooked noodles at t_0 min (Table 7.4). AG content of WUAX 1-SE fraction showed a negative relationship with springiness at t_0 . AG content of WUAX 2-SE fraction showed negative relationship with adhesiveness at t_0 min. It hard to understand if there is any practical significance in these relationships and they need to be further studied.

Arabinose-to-galactose (A/G) ratios of WEAX-SE and WUAX 1-SE fractions were positively correlated with kernel hardness index, and negatively correlated with flour yield and break flour yield (Table 7.4). On the other hand, A/G ratio of WUAX 2-SE fraction was negatively correlated with hardness index but positively correlated with flour yield and break flour yield (Table 7.4). Once again, the behavior of the WUAX 2-SE fraction is divergent from the other two fractions. This time is respect to A/G ratio.

Table 7.4
Correlation coefficients between AG contents and A/G ratio of WEAX, WUAX 1 and WUAX 2, kernel texture, milling characteristics, solvent retention capacity tests, cookie diameter, and noodle texture parameters from the 6 breeding lines with highest and the 6 breeding lines with lowest AX content as determined by wet chemical method¹

	WEAX-SE ²		WUAX 1-SE ²		WUAX 2-SE ²	
	AG ³ (%) ⁴	A/G ³	AG ³ (%) ⁴	A/G ³	AG ³ (%) ⁴	A/G ³
Quality characteristics						
Hardness index	0.572**	0.651**	0.480*	0.412*	NS	-0.441*
Flour yield(%)	NS	-0.762***	-0.546**	-0.689**	NS	0.632**
Break flour yield(%)	NS	-0.678**	-0.545**	-0.618**	NS	0.756***
Solvent Retention Capacity						
H ₂ O(%) ⁴	NS	0.555**	0.481*	NS	NS	NS
Na ₂ CO ₃ (%) ⁴	NS	0.806***	NS	0.543**	-0.552**	NS
Sucrose (%) ⁴	NS	NS	0.476*	NS	NS	NS
Cookie diameter (cm)	NS	-0.489*	NS	NS	NS	NS
Noodle Texture Parameters						
t₀⁵						
Adhesiveness (g*sec) ⁷	NS	NS	NS	0.494*	-0.502*	NS
Springiness	NS	NS	-0.474*	-0.677**	NS	NS
Cohesiveness	0.461*	NS	NS	NS	NS	NS
Resilience	0.450*	NS	NS	NS	NS	NS
t_{15A}⁶						
Hardness (g)	NS	NS	NS	NS	NS	-0.405*

¹ *, ** and ***= Correlation coefficient is significant at P<0.05, 0.01 and 0.001, respectively.

NS: Correlation coefficient is not significant at P<0.05.

²WEAX-SE = water extractable arabinoxylan from SEHPLC, WUAX 1-SE = water unextractable arabinoxylan 1 from SEHPLC, and WUAX 2-SE = water unextractable arabinoxylan 2 from SEHPLC

³ AG (Arabinogalactan, %) = 0.87 x (Arabinose + galactose); A/G= Arabinose (%) /Galactose (%).

⁴On a 14 % flour moisture basis (n=24)

⁵Test immediately after cooling 1 min in water following cooking

⁶Kept in plastic bottle without water for 15 min before testing

⁷Multipled by -1 to change from negative to positive values so that high values = more sticky

The relationships between A/G ratio and A/X ratio and both flour yield and break flour yield were opposite to each other for all fractions (Table 7.3). The positive correlations between A/G ratios of WEAX-SE and WUAX 1-SE fractions and kernel hardness, suggests that increased arabinose substitution is related to harder kernel texture. However, it is interesting that decreased arabinose substitution of AG in the WUAX 2-SE fraction is related harder kernel texture. The only possible mechanism that could be speculated is that the AG extracted in the WEAX and WUAX 1-SE fractions is located in different anatomical structures in the kernel of endosperm than the AG extracted with the WUAX 2-SE fraction. Also interesting that the level of arabinose substitution is differentially related to milling properties depending on whether xylose or galactose is the backbone monomer unit. These relationships need to be further studied.

A/G ratio of WEAX-SE fraction also showed a positive correlation with water and sodium carbonate SRCs (Table 7.4). This was contrast to A/X ratio of WEAX-SE fraction, which had a negative correlation with water, carbonate and sucrose SRCs. A/G ratio of WUAX 1-SE fraction showed a positive correlation only with sodium carbonate SRC (Table 7.2). This was contrast to A/X ratio of WUAX 1-SE fraction, which had a negative correlation with carbonate SRC. There were no significant relationships between WUAX 2-SE fractions and SRCs. Again, this highlighted the different characteristics of the A/G ratio in this fraction compared to the other two.

A/G ratio of the AG extracted with the WUAX 1-SE fraction was positively correlated with cooked noodle adhesiveness and negatively correlated with springiness at t_0 min (Table 7.4). This was opposite to A/X ratio of WUAX 1-SE fraction. A/G ratio of WUAX 2-SE fraction was negatively correlated with hardness of cooked noodles at t_{15A} . Again, any underlying factors affecting relationships between A/G ratios and noodle texture are not clear.

Tables 7.5 shows linear correlations between kernel and milling characteristics, SRCs, cookie diameter, noodle texture parameters, and SEHPLC parameters of WUAX 1-SE and WUAX 2-SE fractions.

There were no significant relationships between retention peak time, peak area, or MW of the WEAX-SE fraction and any of the response variables listed in Table 7.5.

Retention peak time of WUAX 1-SE fraction was negatively correlated with flour yield and break flour yield. Even though it was not possible to calculate the average molecular weight of the WUAX 1-SE fraction using the K_{av} of the standards because the MW of the WUAX 1-SE were far larger than the largest standard, the retention peak time does give an indication of relative MW. This suggests that decreased MW, as well as increased abundance of WUAX 1-SE in this sample set was related to poorer milling characteristics.

Table 7.5
Correlation coefficients between size exclusion chromatography parameters of WUAX 1 and WUAX 2, kernel texture, milling characteristics, solvent retention capacity tests, cookie diameter, and noodle texture parameters from the 6 breeding lines with highest and the 6 breeding lines with lowest AX content as determined by wet chemical method¹

Quality characteristics	WUAX 1-SE ²		WUAX 2-SE ²	
	Retention peak time (min)	Peak area ($\mu\text{v}\cdot\text{sec}$)	Retention peak time (min)	Peak area ($\mu\text{v}\cdot\text{sec}$)
Hardness index	NS	0.619**	0.449*	NS
Flour yield(%)	-0.457*	-0.593**	NS	NS
Break flour yield(%)	-0.453*	-0.531**	-0.558**	NS
Solvent Retention Capacity				
H ₂ O(%) ³	NS	0.644**	NS	NS
Na ₂ CO ₃ (%) ³	NS	0.739***	0.453*	NS
Sucrose (%) ³	NS	0.620**	NS	NS
Cookie diameter (cm)	NS	-0.427*	NS	NS
Noodle Texture Parameters				
t_0 ⁴				
Hardness (g)	NS	NS	0.634**	-0.550**
Adhesiveness (g*sec) ⁶	NS	NS	0.633**	-0.428*
Springiness	-0.437*	NS	NS	NS
Chewiness	NS	NS	0.529**	-0.475*
Resilience	-0.410*	NS	NS	NS
t_{15w} ⁵				
Hardness (g)	NS	NS	NS	-0.436*
Chewiness	NS	NS	NS	-0.438*

¹ *, ** and ***= Correlation coefficient is significant at P<0.05, 0.01 and 0.001, respectively.

NS: Correlation coefficient is not significant at P<0.05.

² WEAX-SE = water extractable arabinoxylan from SEHPLC, WUAX 1-SE = water unextractable arabinoxylan 1 from SEHPLC, and WUAX 2-SE = water unextractable arabinoxylan 2 from SEHPLC

³ On a 14 % flour moisture basis (n=24)

⁴ Test immediately after cooling 1 min in water following cooking

⁵ Kept in plastic bottle with water for 15 min before testing

⁶ Multiplied by -1 to change from negative to positive values so that high values = more sticky

Retention peak time of WUAX 2-SE fraction was positively correlated with hardness index and negatively correlated with break flour yield. These relationships were similar to the correlations between wheat kernel and milling characteristics, and total AX and WEAX contents (Table 4.3). This agreed with the results seen for WUAX 1-SE fraction retention times reported above. Suggesting again the unexpected outcome that lower molecular weight WUAX seem to be related to harder kernel texture.

Retention times of the WUAX 2-SE fraction were weakly and positively correlated with carbonate SRC. Retention times of all three fractions had no significant relationships with cookie diameter.

There were again few significant relationships between these AX parameters and cooked noodle texture. Retention peak time of WUAX 1-SE fraction was negatively correlated with springiness and resilience of cooked noodles at t_0 (Table 7.5).

Retention peak time of WUAX 2-SE fraction was positive correlated with hardness, adhesiveness and chewiness at t_0 (Table 7.5). If, as seems apparent, lower molecular weight of WUAX and higher AX contents are related to harder kernel texture, increased starch damage, and decreased FSV, then as an indirect result of increased AX content etc, it could have been the decreased FSV made noodles harder, more adhesive, less springy and less resilient. However, little confidence can be placed on any of the direct relationships between AX parameters and noodle texture.

Peak area of WUAX 1-SE fraction showed a positive correlation with kernel hardness index, and negative correlations with flour yield and break flour yield (Table 7.5). This was similar to WUAX 1-SE content and shows that peak area is probably another way of describing AX abundance in each fraction. This again supported the idea that the higher AX content, the harder wheat kernel. Peak area of WUAX 1-SE also showed positive correlation with water, sodium carbonate, and sucrose SRCs (Table 7.5). This was partly similar to Table 7.3, which found correlations between WUAX 1-SE AX content and water and sucrose SRCs. Peak area of WUAX 1-SE fraction was negatively correlated with cookie diameter (Table 7.5). This was similar to the previous results and also helped to reinforce the effect of AX content on cookie diameter (Table 4.3).

Peak area of WUAX 2-SE fraction was negatively correlated with cooked noodle hardness, adhesiveness, and chewiness at t_0 (Table 7.5). In addition, peak area of WUAX 2-SE fraction was also negatively correlated with hardness and chewiness at t_{15W} (Table 7.5). However, this conflicted with the previous result that found positive correlations between total AX and WUAX contents with only adhesiveness at t_0 (Table 6.6).

Table 7.6 shows stepwise multiple linear regression results between kernel, milling, and noodle parameters, and AX parameters of WEAX, WUAX 1-SE and WUAX 2-SE. Regressions with R^2 values ≥ 0.70 were considered to indicate a good fit to the

data and suggested that the dependent variables could be predicted with some confidence by the selected parameters.

Table 7.6

Coefficients of determination (R^2) calculated by stepwise multiple linear regressions for kernel hardness, flour milling characteristics and cooked noodle texture at t_{15w} using AX parameters of WEAX, WUAX 1, and WUAX 2 obtained from the 6 breeding lines with highest and the 6 breeding lines with lowest AX content as determined by wet chemical method

Dependent Variables	Selected Parameters¹	R^2 (n = 24)
Kernel hardness index	weax wu1re wu2pa were	0.813
Flour yield	wexl wu1ma wu2xl were	0.826
Break flour yield	wu2agr weagr wu2re wu2gl	0.742
Cooked noodle hardness at t_{15w}	wega mwwe wu2re wegl	0.709

¹mwwe = molecular weight of WEAX-SE fraction, weagr = Arabinose-to-galactose ratio of WEAX-SE fraction, weax = WEAX content of WEAX-SE fraction, wega = galactose content of WEAX-SE fraction, wegl = glucose content of WEAX-SE fraction, were = retention peak time of WEAX-SE fraction, wexl = xylose content of WEAX-SE fraction, wu1ma = mannose content of WUAX 1-SE fraction, wu1re = retention peak time of WUAX 1-SE fraction, wu2agr = arabinose-to-galactose ratio of WUAX 2-SE fraction, wu2gl = glucose content of WUAX 2-SE fraction, wu2pa = peak area of WUAX 2-SE fraction, wu2re = retention peak time of WUAX 2-SE fraction, wu2xl = xylose content of WUAX 2-SE fraction

Four selected parameters accounted for 81% of the variation in kernel hardness index (Figure 7.3a). The selected parameters were WEAX content of the WEAX-SE fraction (weax), retention peak time of the WUAX 1-SE fraction (wu1re), peak area of WUAX 2-SE fraction (wu2pa), and retention peak time of WEAX-SE fraction (were). The best fit regression model for predicting kernel hardness index was: Kernel hardness index = $1729.16 + 104.18(\text{weax}) - 126.58(\text{wu1re}) - 0.00011(\text{wu2pa}) - 111.56(\text{were})$. As has been discussed throughout this chapter and as this stepwise multiple regression supports, parameters related to AX content and molecular weight were dominant in determining kernel hardness.

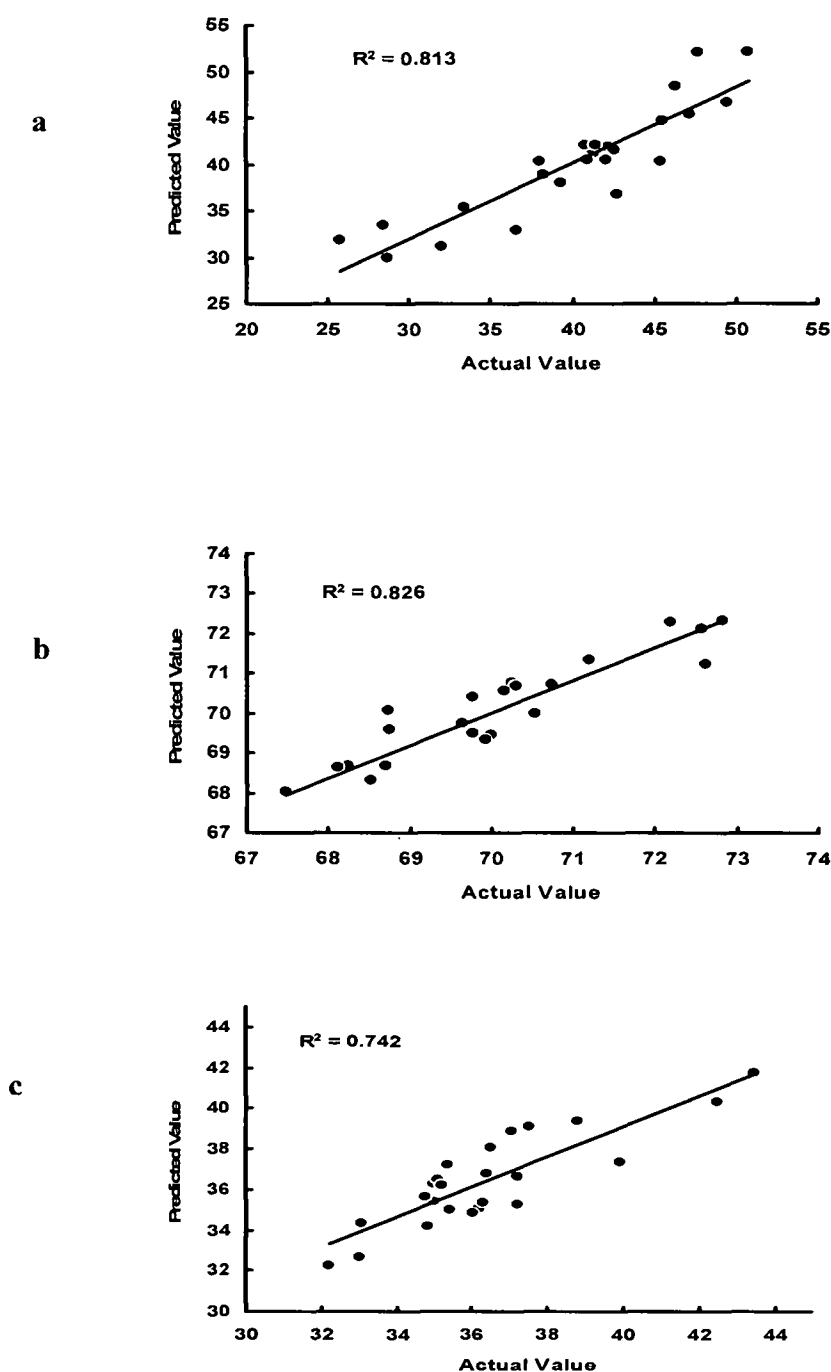


Figure 7.3 The relationships between actual and predicted values of hardness index (a), % flour yield (b) and % break flour yield (c) using stepwise linear regression of AX parameters obtained from the 6 breeding lines with highest, and the 6 breeding lines with lowest AX content as determined by wet chemical method

Four selected parameters accounted for 82% of the variation in flour yield. The selected parameters were xylose content of WEAX fraction (wexl), mannose content of WUAX 1 fraction (wu1ma), xylose content of WUAX 2 fraction (wu2xl), and retention peak time of WEAX fraction (were)(Figure 7.3b). The best fit regression model for predicting flour yield was: $\text{Flour yield} = -49.32 - 35.25(\text{wexl}) + 1.54(\text{wu1ma}) + 37.92(\text{wu2xl}) + 16.69(\text{were})$. As has been discussed throughout this chapter, and as this stepwise multiple regression supports, parameters related to xylose content and molecular weight were dominant in determining flour yield.

Four more selected parameters accounted for 74% of the variation in break flour yield (Figure 7.3c). The selected parameters were A/G ratio of WUAX 2 fraction (wu2agr), A/G ratio of WEAX fraction (weagr), retention peak time of WUAX 2 fraction (wu2re), and glucose content of WUAX 2 fraction (wu2gl). The best fit regression model for predicting break flour yield was: $\text{Break flour yield} = 113.48 + 16.09(\text{wu2agr}) - 16.02(\text{weagr}) - 13.12(\text{wu2re}) + 0.53(\text{wu2gl})$.

The parameters, galactose content of WEAX fraction (wega), molecular weight of WEAX fraction (mwwe), retention peak time of WUAX 2 fraction (wu2re), and glucose content of WEAX fraction (wegl) accounted for 71% of the variation in cooked noodle hardness at t_{15W} min (Figure 7.4). The best fit regression model for predicting cooked noodle hardness at t_{15W} min was: $\text{Cooked noodle hardness at } t_{15W} = -4471.91 + 2026.08(\text{wega}) + 0.0014(\text{mwwe}) + 618.85(\text{wu2re}) - 124.99(\text{wegl})$. As has been discussed throughout this chapter and as this stepwise multiple regression

supports, parameters related to molecular weight was key factor in determining cooked noodle hardness t_{15w} . However, as mentioned above, any relationships between AX parameters and noodle texture may be mediated through the indirect pathway, kernel hardness, starch damage, changes in FSV, and hence, changes in noodle texture and indeed, there may be no direct influence at all.

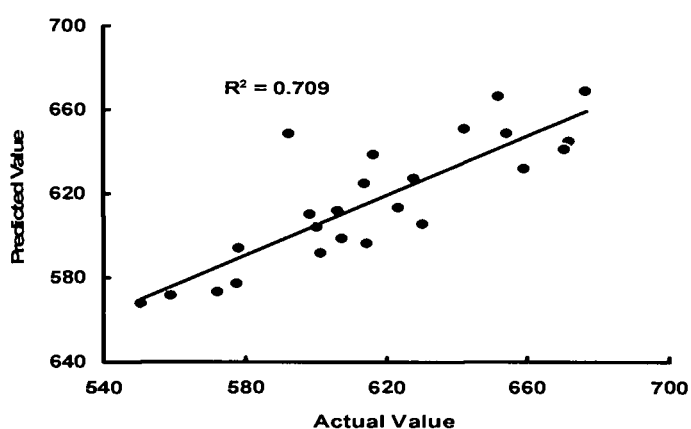


Figure 7.4 The relationship between actual and predicted values of hardness at t_{15w} using stepwise linear regression of AX parameters obtained from the 6 breeding lines with highest and the 6 breeding lines with lowest AX content as determined by wet chemical method.

However, these relationships between molecular weight distribution of all three AX fractions, flour characteristics, and noodle quality were evaluated only 12 varieties. To understand and predict the relationship of these parameters, a larger sample set should be used in the future.

Chapter 8

Conclusions

- Kernel hardness index was positively correlated with starch damage, and total AX and WEAX contents. Kernel hardness index was negatively correlated with break flour yield and total flour yield.
- Sodium carbonate SRC was positively correlated with total AX, and WEAX contents, and starch damage. Sucrose SRC, which has been related to AX content by other workers (e.g.; Guttieri et al 2001), also showed positive correlations with total AX and WEAX contents. Lactic acid SRC, which is reputedly related to both protein content, and gluten composition and strength, showed no significant correlation with mixograph absorption ($r = 0.044$, NS). However, at the very low protein contents of the flours (Table 4.2) used in this study, it is likely that the absence of a relationship between lactic acid SRC and Mixograph was a result of the Mixograph results.
- Sucrose and sodium carbonate SRC tests were correlated with cookie diameter. However, it was considered that they were not reliable predictors of cookie diameter because of the notably scattered correlations with this parameter (Figures 4.1 and 4.2).
- During AX method development, AX were found in WUAX 1-SE and WUAX 2-SE fractions (Figure 5.1) that had been treated with protease and amylase.

These two fractions were discarded in the previous report suggesting that Faurot et al (1995) had discarded some potentially valuable materials.

- Even though it not possible to quantify the absolute MWs (see below), the WUAX 2-SE fraction had the highest molecular weight, followed by the WUAX 1-SE, and then the WEAX-SE fractions (Figure 5.4).
- Molecular weights of WEAX-SE ranged from approximately 411,305 and 447,282. This was in some agreement with the results of Cui (2001) and Courtin et al (1999), which found the molecular weight of endosperm WEAX to be around 231,000 to 530,000.
- Molecular weights of WUAX 1-SE and WUAX 2-SE could not be specifically defined in this study because their MWs were well above the MW of the largest pullulan standard (404,000) and the column exclusion limit of about 1,000,000 caused the these two fractions elute very near the void volume.
- WEAX-SE contained a higher degree of substitution than WUAX 1-SE and WUAX 2-SE. These results were in agreement with Bacic and Stone (1981) but they were conflicted with more recent studies that observed higher degrees of substitution in WUAX (Gruppen et al, 1992; Courtin et al, 1999). Many of the more recent studies observed hard-common, or durum wheats, and the conflict in results with this study may be related to the different genetic

backgrounds of soft-wheats studied here. Nevertheless, there is still no conclusion about the degree of substitution of WEAX and WUAX.

- Flour protein content was negatively correlated with t_{15A} cooked noodle hardness, adhesiveness and chewiness, and positively correlated with t_{15A} cooked noodle springiness at (Table 6.6). These results conflicted with previous reports where flour protein content was positively correlated with cooked noodle hardness (Miskelly, 1984; Toyokawa et al, 1989a; Baik et al, 1994; Park et al, 2003). This might have occurred as a result of the very low flour protein contents in this sample set (about 6.4%). Therefore, protein composition, which related to lactic acid SRC, became more important for noodle texture.
- Total AX and WUAX were positively correlated with adhesiveness at t_0 (Table 6.6). This might result from characteristics of AX themselves, which are gummy and sticky. This lead to more adhesive noodles.
- Both starch damage and sodium carbonate SRC were positively correlated with cooked noodle hardness and chewiness at both t_0 and t_{15A} (Table 6.6). However, there was only a weak relationship between sodium carbonate SRC and starch damage, which indicated the poor predictive capacity of carbonate SRC for starch damage in this sample set.

- Both high FSV and high RVA peak viscosity were related to decreased optimum cooking time, decreased cooking loss of noodles, and softer and less adhesive noodles. (Table 6.1 and 6.6). Either FSV or RVA peak viscosity may be selected to predict noodle characteristics.
- There was a highly significant correlation between WEAX-SE and WEAX from wet chemical method ($r = 0.802$, $p < 0.001$).
- WUAX contents determined through monosaccharide analyses were lower than the WUAX contents determined by the wet chemical method. However, the general ranking was still well aligned. Two lines (ORH011090 and ORH011450) were responsible for the poor correlation of WUAX 1-SE and total WUAX-SE, and WUAX from wet chemical method (Figure 7.2).
- Increased xylose and arabinose contents appeared to simply reflect overall higher AX abundance, and they were related to harder kernel texture, poor milling properties, and water, carbonate and sucrose SRCs, and smaller cookie diameter (Table 7.2).
- Any measure of AX abundance, except in the case of the WUAX 2-SE fraction, was to some extent able to predict kernel hardness, milling properties and SRCs (Table 7.3). These trends were similar to xylose and arabinose contents.

- For A/X ratios of the WEAX-SE and WUAX 1-SE fractions, it was found that lower arabinose substitution was related to poor milling yields, and lower SRC values. Again, the WUAX 2-SE fraction shows divergent behavior with respect to the other two fractions in that higher arabinose substitution in this fraction was related to poor milling yields, and lower SRC values. (Table 7.3).
- The pattern of relationships between kernel and milling characteristics and AG contents of WEAX-SE and WUAX 1-SE fraction were somewhat similar to results seen for AX contents of the same fractions (Table 7.4). This suggested that AG abundance might be related to AX abundance.
- A/G ratios of WEAX-SE and WUAX 1-SE fractions were positively correlated with kernel hardness index, and negatively correlated with flour yield and break flour yield (Table 7.4). On the other hand, A/G ratio of WUAX 2-SE fraction behaved in the opposite fashion to the other two fractions.
- The relationships between A/G ratio and A/X ratio and flour yield, break flour yield, and SRCs were opposite to each other for all fractions (Table 7.3).
- Retention peak times of WUAX 1-SE and WUAX 2-SE fraction were negatively correlated with flour yield and break flour yield. The retention peak time give an indication of relative MW (Table 7.5). This suggests that

decreased MW, as well as increased abundance of WUAX in this sample set was related to poorer milling characteristics.

- Peak area of WUAX 1-SE fraction, again related to AX abundance, showed a positive correlation with kernel hardness index and SRCs. It also showed negative correlations with flour yield and break flour yield and cookie diameter (Table 7.5).
- There were few significant relationships between AX parameters and cooked noodle texture parameters (Table 7.2). It seemed to be no pattern to the relationships that might provide clues regarding any underlying causal or indirect factors.
- Kernel hardness index, flour yield, break flour yield and cooked noodle hardness at t_{15W} were able to be predicted with some confidence by using stepwise multiple regressions that utilized selected AX parameters of the WEAX, WUAX 1-SE and WUAX 2-SE fractions.
- AX content appeared affect noodle texture indirectly. The effects were mediated through the pathway of higher AX contents and low MW associated with harder kernels, poorer milling properties, increased starch damage, reduced FSV, and finally harder noodle texture. Surprisingly, low MW of WUAX in this sample set also appeared to be related to harder kernel textures.

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Appendices

Appendix A

Pedigrees of the sixty three advance soft white wheat breeding lines used in this study. Each line had two replicates, with the exception of ORH011273

Sample ID	ID	Pedigree
cv1-10-1	ORH012079	69-153/YMH//YMHDW
cv1-1-1	Stephens	STEPHENS
cv1-19-1	ORH012183	KVZ/3/HD/ON//BB/4/YPOPF/3/RBS1744//SU/GNS/5/SPN//AU/YMH
cv1-2-1	Madsen	MADSEN
cv1-3-1	OR939526	OR939526
cv1-36-1	ORH010013	(84X126VPM/M951//YMH/HYS//3518)/3/69-153/YMH//YMHDW
cv1-37-1	ORH010014	(84X126VPM/M951//YMH/HYS//3518)/3/69-153/YMH//YMHDW
cv1-42-1	ORH010083	DUSTY/ZGP-4074//((UNKNOWN))
cv1-43-1	ORH010085	DUSTY/ZGP-4074//((UNKNOWN))
cv1-50-1	ORH010142	NY80095-6//((UNKNOWN))
cv1-6-1	ORH012049	H73-F4-6210-3H-0P/BJY,F1/4/F1,TJB259//MHM/3/GLL/NAR
cv1-7-1	ORH012052	CLEO/PCH//ZZ,F1/4/F1,AVC/3/DJ/BEZ//WA520
cv1-75-1	ORH010361	7C/CNO//CAL/3/YMH/4/MacVicar
cv1-77-1	ORH010363	7C/CNO//CAL/3/YMH/4/MacVicar
cv1-78-1	ORH010364	7C/CNO//CAL/3/YMH/4/MacVicar
cv1-80-1	ORH010369	7C/CNO//CAL/3/YMH/4//((UNKNOWN))
cv2-12-1	ORH010404	((UNKNOWN))/4/6720/HYS//R37/GHL1,F1/3/SPN
cv2-19-1	ORH010447	6720/HYS//R37/GHL1,F1/3/SPN/4/NAD//SU92/BURT/3/CLP
cv2-25-1	ORH010486	7C/CNO//CAL/3/YMH/4/69-153/YMH//YMHDW
cv2-54-1	ORH010652	65-116-MBW//63-189-66-7/BEZO/4/SPN//AU/YMH/3/SPN
cv2-56-1	ORH010656	65-116-MBW//63-189-66-7/BEZO/3/FRENCH LINE E81FR
cv2-60-1	ORH010662	65-116-MBW//63-189-66-7/BEZO/3/FRENCH LINE E81FR
cv2-6-1	ORH010377	RMNF3-71/TORIM/4/6720/HYS//R37/GHL1,F1/3/SPN
cv2-63-1	ORH010685	AMD/HN4*2//FRENCH LINE E81FR
cv2-66-1	ORH010699	65-116-MBW//63-189-66-7/BEZO/4/HYS/YY/63-112-66-4/3/OR87065,H-281
cv2-7-1	ORH010382	RMNF3-71/TORIM/4/6720/HYS//R37/GHL1,F1/3/SPN
cv2-77-1	ORH010800	HYS/YY/63-112-66-4/3/OR87065,H-281/6/HN4/4/KT54A/N10B//KT554B/3/NAR/5/PLJ/7C
cv2-80-1	ORH010823	HYS/YY/63-112-66-4/3/OR87065,H-281/4/FRENCH LINE E81FR
cv2-8-1	ORH010385	RMNF3-71/TORIM/4/NAD//SU92/BURT/3/CLP
cv3-10-1	ORH010838	HYS/YY/63-112-66-4/3/OR87065,H-281/4/FRENCH LINE E81FR
cv3-1-1	Stephens	STEPHENS
cv3-2-1	Madsen	MADSEN
cv3-24-1	ORH010907	6720-11//MDA38/WRN/4/SPN//AU/YMH/3/SPN
cv3-29-1	ORH010917	6720-11//MDA38/WRN/3/FRENCH LINE E81FR
cv3-30-1	ORH010918	6720-11//MDA38/WRN/3/FRENCH LINE E81FR
cv3-3-1	OR939526	OR939526
cv3-32-1	ORH010920	6720-11//MDA38/WRN/3/FRENCH LINE E81FR
cv3-33-1	ORH010927	MRS/C114482//YMH/HYS/3/SPN//YMH/HYS
cv3-33-2	ORH010927	MRS/C114482//YMH/HYS/3/SPN//YMH/HYS
cv3-36-1	ORH010942	MRS/C114482//YMH/HYS/4/HYS/YY/63-112-66-4/3/OR87065,H-281
cv3-36-2	ORH010942	MRS/C114482//YMH/HYS/4/HYS/YY/63-112-66-4/3/OR87065,H-281
cv3-50-1	ORH011012	MACVICAR//((UNKNOWN))

Appendix A (Continued)

Sample ID	ID	Pedigree
cv3-55-1	ORH011040	MACVICAR/(UNKNOWN)
cv3-6-1	ORH010830	HYS/YY/63-112-66-4/3/OR87065,H-281/4/FRENCH LINE E81FR
cv3-68-1	ORH011090	KVZ/3/HD/ON//BB/4/YPOPF/3/RBS1744//SU/GNS/5/SPN//AU/YMH/6/YMH/HYS//VPM/MOS
cv3-69-1	ORH011091	KVZ/3/HD/ON//BB/4/YPOPF/3/RBS1744//SU/GNS/5/SPN//AU/YMH/6/YMH/HYS//VPM/MOS
cv3-7-1	ORH010834	HYS/YY/63-112-66-4/3/OR87065,H-281/4/FRENCH LINE E81FR
cv3-73-1	ORH011106	KVZ/3/ND/ON//BB/4/YPOPF/3/RBS1744//SU/CN/5/(UNKNOWN)
cv3-9-1	ORH010837	HYS/YY/63-112-66-4/3/OR87065,H-281/4/FRENCH LINE E81FR
cv4-30-1	ORH011220	7C/CNO//CAL/3/YMH/4/CER/YMH/HYS
cv4-31-1	ORH011226	7C/CNO//CAL/3/YMH/4/MacVicar
cv4-36-2	ORH011273	YMH/HYS//VPM/MOS4-2-16-1-7/4/SPN//AU/YMH/3/SPN
cv4-39-1	ORH011297	CER/YMH/HYS/3/69-153/YMH//YMHWDW
cv4-4-1	ORH011141	H73-F4-6210-3H-0P/BJY,F1/4/F1,TJB259//MHM/3/GLL/NAR/5/67-109/NUG//RBS/P101
cv4-41-1	ORH011300	CER/YMH/HYS/4/SPN//AU/YMH/3/SPN
cv4-44-1	ORH011327	(UNKNOWN)//SPN/QLP
cv4-55-1	ORH011450	HYS/YY/63-112-66-4/3/OR87065,H-281/5/YMH/HYS//VPM/MOS,F1/4/F1,CER/3/SPN//AU/ HYS
cv4-59-1	ORH011481	6720-11//MDA38/WRN/3/FRENCH LINE E81FR
cv4-60-1	ORH011482	6720-11//MDA38/WRN/3/FRENCH LINE E81FR
cv4-61-1	ORH011483	6720-11//MDA38/WRN/3/FRENCH LINE E81FR
cv4-64-1	ORH011587	(84X126VPM/M951//YMH/HYS//3518)/3/67-109/NUG//RBS/P101
cv4-68-1	ORH011618	(UNKNOWN)/3/HIM//KAL/BB
cv4-7-1	ORH011147	H73-F4-6210-3H-0P/BJY,F1/4/F1,TJB259//MHM/3/GLL/NAR/5/YMH/HYS/4/MRS/3/YMH//RBS/NCO
cv4-71-1	ORH011684	MACVICAR/5/H73-F4-6210-3H-0P/BJY,F1/4/F1,TJB259//MHM/3/GLL/NAR
cv4-78-1	ORH011828	98-967

Appendix B

Pedigrees of the subset of twelve soft white wheat breeding lines chosen for extraction of AX fractions. Six were chosen on the basis of having the highest total AX contents, and six, as they had the lowest. Each line was replicated twice.

Sample ID	ID	Pedigree
Low total AX		
1.43.1	ORH010085	DUSTY/ZGP-4074/(UNKNOWN)
4.4.1	ORH011141	H73-F4-6210-3H-0P/BJY,F1/4/F1,TJB259//MHM/3/GLL/NAR/5/67-109/NUG//RBS/PI01
3.6.1	ORH010830	HYS/YY/63-112-66-4/3/OR87065,H-281/4/FRENCH LINE E81FR
3.32.1	ORH010920	6720-11//MDA38/WRN/3/FRENCH LINE E81FR
2.54.1	ORH010652	65-116-MBW//63-189-66-7/BEZO/4/SPN//AU/YMH/3/SPN
1.42.1	ORH010083	DUSTY/ZGP-4074/(UNKNOWN)
High total AX		
4.59.1	ORH011481	6720-11//MDA38/WRN/3/FRENCH LINE E81FR
3.68.1	ORH011090	KVZ/3/HD/ON//BB/4/YPOPF/3/RBS1744//SU/GNS/5/SPN//AU/YMH/6/YMH/HYS//VPM/MOS
2.8.1	ORH010385	RMNF3-71/TORIM/4/NAD//SU92/BURT/3/CLP
1.19.1	ORH012183	KVZ/3/HD/ON//BB/4/YPOPF/3/RBS1744//SU/GNS/5/SPN//AU/YMH
4.55.1	ORH011450	HYS/YY/63-112-66-4/3/OR87065,H-281/5/YMH/HYS//VPM/MOS,F1/4/F1,CER/3/SPN//AU/ HYS
2.6.1	ORH010377	RMNF3-71/TORIM/4/6720/HYS//R37/GHL1,F1/3/SPN