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Role of Durum Wheat Composition on the Quality of Pasta and Bread

Mike Sissons*

NSW Department of Primary Industries, Tamworth Agricultural Institute, 4 Marsden Park Road, Calala, NSW Australia 2340

Corresponding author: * **mike.sissons@dpi.nsw.gov.au**

ABSTRACT

Durum wheat (*Triticum turgidum* L.) is the preferred raw material for the production of pasta worldwide and some speciality bread common in parts of Italy and the Mediterranean region. The quality of such foods in terms of texture, colour, flavour and appearance are determined by raw material quality, processing methods and other ingredients. This review focuses on the raw material composition and how these influence the dough characteristics and the end product quality. Protein has been known as an important component having an influence on the quality of pasta and bread. The glutenin and gliadin proteins, the types present and their ratio have been shown to influence dough properties. Attempts to increase the number of high molecular weight glutenin subunits to obtain more varied dough properties has the potential to improve the breadmaking properties of durum flour. Starch is more than an inert filler and recent research has shown the affect of varying the amylose content and ratio of large to small starch granules on pasta quality can be significant. Potentially new durum germplasm could be created and used in new food products. Other minor components like non-starch carbohydrates and lipids have received less attention. The former can have a large impact on the water absorption of durum flours and alter dough properties. Enzymes like lipoxygenase and polyphenol oxidase together with the lipid yellow pigments strongly impact the appearance of pasta foods. The results of recent research about these components on both pasta and bread quality using durum wheat are discussed.

Keywords: gluten, glutenin, gliadin, starch, non-starch polysaccharides, semolina, spaghetti

Abbreviations: **AGP**, arabinogalactan peptide; **AX**, arabinoxylan; **DSC**, differential scanning calorimetry; **FN**, falling number; **GI**, gluten index; **HMW-GS**, high molecular weight glutenin subunit; **HPLC**, high performance liquid chromatography; **HT**, high temperature drying; **L**, alveograph length; **LMW-GS**, low molecular weight glutenin subunit; **LOX**, lipoxygenase; **LT**, low temperature drying; **LVP**, linear viscoelastic properties; **MPT**, mixograph peak time; **MWD**, molecular weight distribution; **NSP**, non-starch polysaccharides; **P**, alveograph tenacity; **PPO**, polyphenol oxidase; **RVA**, rapid visco-analyser; **SBE**, starch branching enzyme; **SDS-PAGE**, sodium dodecyl sulphate polyacrylamide gel electrophoresis; **SDSS**, sodium dodecyl sulphate sedimentation; **SSII**, starch synthase II; **W**, alveograph overall strength; **WE-AX**, water extractable arabinoxylan; **WU-AX**, water unextactable arabinoxylan

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INTRODUCTION

In this review the term "pasta" will be restricted to products made from durum wheat semolina. Pasta products made from gluten-free cereals such as rice, maize and sorghum will not be considered. The focus is on traditional dried pasta shapes because much of the research used this product although much of the discussion applies to fresh pasta products. For bread making, principally leavened bread, only that made from durum semolina or durum and common

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Invited Review

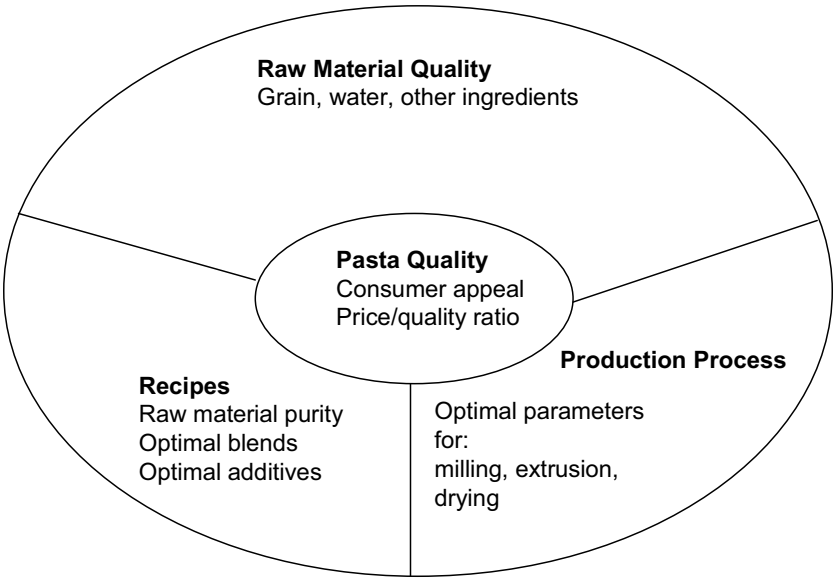

Fig. 1 Pasta quality determinants model.

wheat blends will be considered. The main focus of this review will be on the main chemical components of the durum wheat grain (starch, protein, non-starch carbohydrate and lipids) and how these influence the behaviour of semolina dough and the quality of pasta and bread. Nutritional benefits of the components will not be discussed.

USE OF DURUM WHEAT FOR MAKING PASTA

The wheat preferred for making pasta products is durum *Triticum turgidum* L. subsp. *turgidum* conv. *durum* (Desf. MacKey). Durum wheat, in contrast to common wheat *Triticum aestivum* L., which is used to make bread and oriental style noodles, is the hardest wheat and durum milling produces a coarse particle called semolina, ideal for making pasta and couscous. The key features of durum wheat include its hardness, intense yellow colour and nutty taste. After conversion to pasta, durum wheat produces products with good cooking quality and stability to overcooking with unmatchable eating quality. Several countries (Italy, France and Greece) have decreed that dried pasta be produced exclusively from durum wheat and that the use of other cereals not mentioned is considered a fraud (Italian law No. 580, 1967). Other countries such as Spain, United States, Canada and Australia traditionally consume by choice, pasta made from only durum wheat.

DURUM WHEAT FOR MAKING BREAD

Common wheat (*T. aestivum*) is commonly used to prepare leavened and flat breads. However, durum wheat is popular for bread making in homes in Southern Italy for which the bread has many names (Quaglia 1988). Durum is now being used in the Mediterranean regions for breads of all types and popularity is spreading to other countries (Quaglia 1988), although more as speciality bread. Although durum flours usually produce a smaller loaf volume than those of bread wheats, the durum bread has a yellowish colour, a characteristic taste and smell, a fine uniform crumb structure and more prolonged shelf-life, all of which appeal to consumers of this specialty type of bread (Liu *et al.* 1996). Durum bread has also been reported to have less gluten toxicity to people with gluten intolerance, another reason for making bread from durum wheat (Troncone and Auricchio 1991). Many of these breads are not produced industrially, so their production still exhibits artisan characteristics and consequently, they are more expensive to manufacture.

Breeding durum wheat suitable for both bread and pasta occurs in Italy and Canada because dual purpose durum wheats can be used in place of bread wheat or blends with high quality baking flour. Bread prepared from durum and spring wheat flour blends (60:40) using the sponge-dough baking method produced bread with similar loaf volume and external appearance to bread made from 100% spring wheat flour but with higher staling rates (Hareland and Puhr 1998). Acceptable bread was made using the straight dough method with durum and bread wheat flour blends (25:75) but only when the flour was enhanced with sodium stearoyl lactylate (Boyacioglu and D'Appolonia 1994).

CRITERIA FOR SELECTING DURUM FOR GOOD PASTA QUALITY

Pasta quality is determined by three main factors, the raw materials, the production recipe and the production process (Dawe 2001) (**Fig. 1**). In this review, the components that contribute to good dough properties which influence pasta quality will be discussed. For pasta making, dough properties are an important aspect of quality and it is the storage proteins of the wheat endosperm that are the main determinants of dough properties, such as dough strength, extensibility, and dough stability. Additional factors like starch and non-starch polysaccharides and non-gluten proteins can also play a role. The dough properties can be measured using traditional cereal chemistry instruments or using more modern equipment looking at fundamental rheology measurements.

Assessment of good potential to make pasta begins with the grain. Aspects of importance include visual appearance, test weight, weight of 1000 kernels, physical defects, vitreousness, moisture content, weather damage and grain protein percentage (Sissons 2004). For more details the reader is referred to a review on this subject (Sissons 2004). Protein content forms a part of the wheat payments to farmers in some countries (e.g. Australia, Canada) (Sissons 2004). High protein semolina from durum wheats of good physical condition will generally yield semolina of uniform particle size with a minimum number of starchy semolina particles, and will hydrate evenly during mixing to produce pasta that is strong and elastic. When cooked, the pasta will swell leaving minimal residue in the cooking water, remaining firm to the point of serving. Semolina with low protein will produce pasta products deficient in some or all of these characteristics. Typical values for protein in durum semolina range 11-16% (dwb) with the optimum determined by product desired and manufacturer (Turnbull 2001).

After the grain is milled into semolina, several criteria are used by pasta makers to assess the pasta quality potential. These include ash content, colour, speck count, particle size distribution, non-durum contamination and protein qua-

lity (Sissons 2004). The type of proteins present in the grain affects processing properties. Gluten strength is a term used to describe the ability of the proteins to form a satisfactory network that promotes good cooking quality. The continuity and strength of the protein matrix formed during dough mixing and extrusion is important in determining the textural characteristics of the pasta. Compared to weak gluten of the same protein level, strong gluten wheats exhibit less sticky dough with better extrusion properties and superior cooked textural characteristics (Dexter and Matsuo 1978; Autran *et al.* 1986; Matsuo *et al.* 1986; D'Egidio *et al.* 1993; Sissons *et al.* 2005b). Strength is particularly important for instant pastas since these have thinner walls and need more strength during processing. In contrast popular fresh pastas, require a more extensible dough and weaker gluten to improve sheeting properties (Marchylo *et al.* 2004). Thus, durum wheat or semolina specifications for gluten strength will vary depending on the type of final product being processed. In traditional pasta-consuming countries the consumer is concerned about the aroma, colour, appearance, texture, flavour and nutritional value of the pasta (D'Egidio and Nardi 1998). The pasta after cooking should maintain its texture and not become a thick, sticky mass. Mechanical texture is typically described by a range of terms (firmness, elasticity, stickiness, chewiness and bulkiness) and can be measured by a sensory panel or by objective tests (D'Egidio and Nardi 1998). Sensory evaluation is regarded as the ultimate test of pasta cooking quality and is the reference for which other methods are compared. However, some difficulties occur related to the different background and experience of the testers. To avoid subjectivity, various testing instruments have been developed to evaluate texture and all involve a means of deforming a sample and recording the force, time and compression rate. Another test involves measuring by chemical methods, the total amount of organic matter released from the cooked pasta after immersion in water for a fixed time. This test is highly correlated with sensory evaluation (D'Egidio *et al*. 1993).

CRITERIA FOR SELECTING DURUM FOR GOOD BREAD MAKING QUALITY

Durum wheat flour for bread making can be obtained by either re-milling semolina or through direct milling. Milling to make too fine a flour can increase starch damage, due to the extreme hardness of durum. This can cause problems by lowering the loaf volume, producing a wet and undercooked crumb and a dark crust colour (Dexter *et al.* 1994; Saperstein *et al.* 2007), thought to be related to the greater water absorption of damaged starch (Dexter *et al.* 1994). Smooth rolls impart more starch damage than fluted rolls. High protein content generally provides superior baking performance as does a short baking process but still does not achieve the loaf characteristics obtained using bread wheat flours (Dexter *et al.* 1994). A good bread making flour requires strong gluten capable of producing an extensive viscoelastic matrix during dough formation and that has good physical handling properties, such as high resistance to extension and moderate extensibility. Early durum wheat varieties were far too weak to make good bread (pre 1980). The development of stronger durum wheats in Canada and USA allowed durum wheats to approach but not match a good baking flour. Durum dough has been described as "mushy" or "firm" but not "tough" (Liu *et al.* 1996). In Italy it was found that durum varieties need gluten that is less elastic and more extensible. Traditional dough tests like farinograph, extensograph and alveograph have found durum wheat gluten to be too inelastic and very weak (Liu *et al.* 1996). The alveograph extends the dough sample under pressure and is deformed into a thin bubble, whose volume increases until its breaking point. Typically alveograms of durum wheat indicate very high tenacity (P) versus elasticity (L). Consequently the P/L ratio is >1.5. Quaglia (1988) concluded that to make durum bread, the semolina or flour should have a particle size range of 120-190 μm, less than 7-7.5% starch damage, protein >13% (dmb) and good gluten quality (alveograph P/L ratio >1.5 and energy (W) of ~200 J x 10^{-4}). Dough stability is important to ensure the dough reaches its optimal fermentation time. This degree of tolerance can be judged by the degree of softening obtained from farinograph analysis. Because of their high gluten content and tenacity, durum wheat flours have a high fermentation tolerance. Durum wheats typically have high falling numbers (sound grain) or low α-amylase activities making the dough less easy to develop properly and can produce a hard bread. Blending such flours with hard wheat, of higher amylolytic activity is used to overcome this problem. However, blends can cause uneven hydration which affects the bread due to differences in the water absorption and rate of hydration of the two flours (Quaglia 1988).

Traditionally wheat dough physical properties have been determined using dough mixing instruments. These tests provide indicators of relative dough strength, but do not provide fundamental linear viscoelastic properties (LVP). For polymers of high molecular weight like gluten, LVP is being increasingly viewed as a method of molecular characterisation due to the difficulties of solubilising and separating very large polymers. Mechanical tests like frequency sweeps and long time creep compliance tests performed in the linear viscoelastic region have been increasingly used to examine the mechanical properties of cereal dough (Rao *et al.* 2001). This approach is now allowing more definite conclusions about the chemical nature of the network structure in dough and its relationship to end product quality.

The remix-to-peak baking process has a relatively long fermentation time typical of the Italian bread making process (Kilborn and Tipples 1981). Fermentation time is critical. During fermentation the effects of acidity and the enzymatic and oxidation-reduction processes result in physical changes in dough properties called "mellowing". Short fermentation times promote better baking quality of durum loaves (Dexter *et al.* 1994). Stronger durum dough benefits more from shorter fermentations (Saperstein *et al.* 2007). The relatively poor fermentation tolerance of many durum genotypes encountered in the latter study was thought to be due to the absence of sufficient amount of high molecular weight glutenin subunits (HMW-GS) (Saperstein *et al.* 2007).

DOUGH

Pasta dough is made from semolina and water and forms upon the application of some stress (energy) under certain temperature and moisture conditions. As the wet mix of semolina and water passes from the mixer into the vacuum screw of the extruder, it is formed into a dough by the application of mechanical work (Dawe 2001). The dough is characterised by the formation of a gluten network where the semolina particles that contain protein exude proteinaceous fibrils, which interact to form a cohesive dough (Amend and Belitz 1989). The changes that follow during mixing and extrusion are called dough development. Ideas about what happens at the molecular level are not clear but a likely sequence has been proposed: The dough mixing with water blends the ingredients into a homogenous mass as the particles absorb water. Mixing aids this hydration by exposing new dry surfaces on flour particles to the water. Subsequently, further changes occur at the molecular level including interaction of gliadin and glutenin and the formation of disulphide bonds to form gluten, the viscoelastic matrix of the dough (Graveland *et al.* 1985). The development of a dough can be recorded using instruments like the farinograph and mixograph.

THE DOUGH MAKING PROCESS: OVERVIEW

The transformation of semolina into pasta involves wetting, mixing and extrusion. The resultant wet pasta has a network of protein that encapsulates the starch granules, to produce a structure that has the minimum of cracks and voids. In

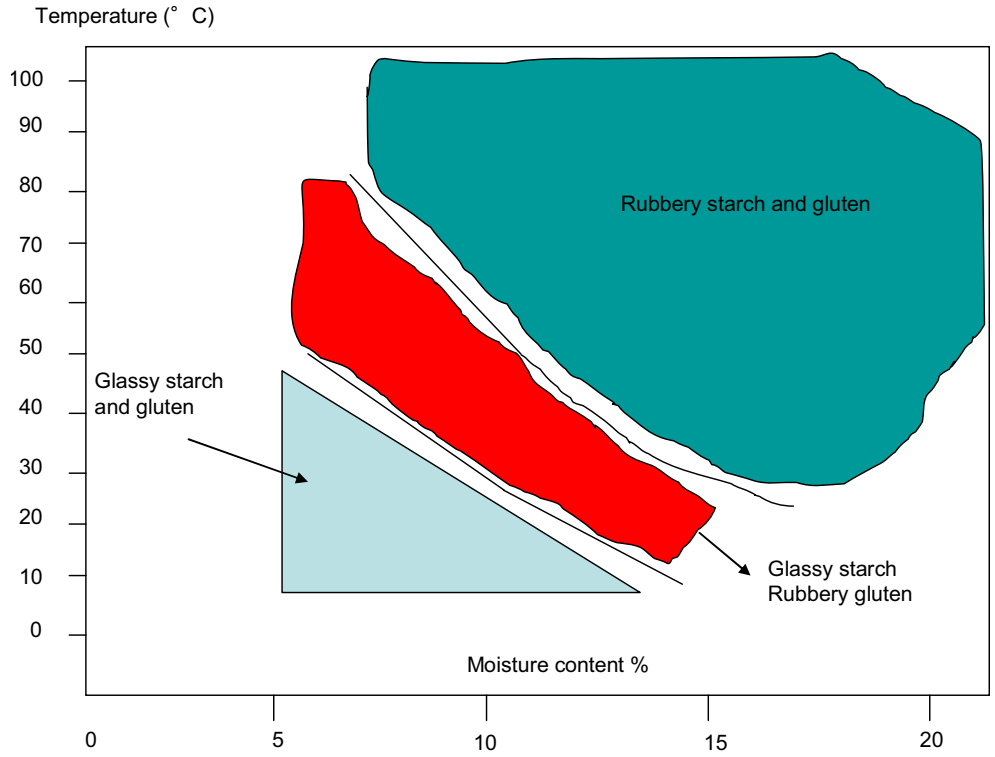

Fig. 2 Glass transitions of starch and gluten. (adapted from Blanshard 1995).

semolina the gluten is glassy but upon the addition of some water it becomes rubbery and elastic, acquiring the ability to form strands and sheets via inter-molecular bonds. This matrix helps to trap the starch granules in pasta and hold its shape during cooking. When the hydrated gluten is heated, irreversible protein-protein cross-links are formed. The starch behaves like a inert filler below about 55°C and cannot absorb much water. Upon heating, starch loses its rigid structure, becomes rubbery passing its glass transition (**Fig. 2**) and can readily absorb water. This causes an increase in viscosity as the granules swell and release soluble material from the granule.

At room temperatures (~25°C) and low moisture <12%, both the gluten and starch in the semolina behave as a glassy material. At slightly higher moisture, the gluten will behave as a rubbery material as it undergoes glass transition (**Fig. 2**) and as more water is added to around 33%, the gluten will flow under applied stress (mixing) (Blanshard 1995). During this phase the water is distributed among the dry semolina to produce an even moisture distribution. As the wet mix passes into the vacuum screw and to the extruder, it is formed into a dough and the application of mechanical work causes the protein to fuse and form the gluten network. If the temperature rises above 55°C, the gluten becomes increasingly tough and stiff and irreversibly forms a gel (Blanshard 1995). This process is undesirable in the extruder and any gluten in this condition will appear in the pasta as fragments of broken gel and these make the pasta strands weaker. Thus, maximum dough temperatures are held below 55°C. By forcing dough through a die under pressure, pasta of a desired shape can form. To give the pasta some resistance to overcooking, the protein network must not be adversely damaged. To avoid this, kneading (the homogenisation of the dough under pressure) of the dough in the screw must be gentle. It is the shearing forces that can damage the protein network.

Unless the pasta is to be sold as fresh, with a short shelf-life, the pasta has to be dried to remove excess moisture to a water activity at which microbial growth is impossible (a_w<0.65). Also, the use of high temperatures is needed to denature the gluten proteins to provide protein cross-linking which is desirable to form a network to entrap starch granules (for more details on pasta drying, see Sissons 2004).

DOUGH COMPONENTS THAT IMPACT ON PASTA AND DURUM BREAD QUALITY

In this part of the review recent literature will be used to highlight the impact of the different components (proteins, starch, non-starch polysaccharides and other minor components) of the dough that impact on the end product. The polypeptide composition of a wheat flour sample is determined by genotype effects (G) due to allelic composition of the glutenin and gliadin components. There is extensive evidence that the protein quality in durum wheat is governed primarily by chromosome 1B, due to the presence of glutenin and gliadin encoding loci (Payne *et al*. 1984; Peña *et al*. 1994; Liu *et al*. 1996; Vazquez *et al*. 1996; Porceddu *et al*. 1998). There is extensive polymorphism at the *Glu-B3* loci in durum and at each of the *Gli-1* loci. With many allele combinations possible, there is potential for diversity. In addition, there is the effect of growing conditions on the wheat and the expression levels (E) and the different sensitivities of the expression levels on the individual genes (GxE). The complexity of relating protein composition to quality requires an investigation at different levels: protein content, composition, ratio of glutenin to gliadin, ratio of HMW-GS to low molecular weight glutenin sub-units (LMW-GS). The polymeric protein (glutenin) is mainly responsible for the elasticity of the dough, whereas the monomeric gliadins are the extensibility-related characters. Thus the ratio of glutenin to gliadin can be directly related to the balance of dough strength and extensibility (Wrigley *et al*. 2006) while the effect of variation in the ratio of HMW-GS to LMW-GS is less clear (see later).

Much of our knowledge about the effects of specific proteins on the functional properties of dough is based on correlative studies. For this the quality of every member of a population of samples is measured. By correlating quality with differences in genetic composition, relationships can be built. A major limitation of this approach is that the statistical evaluation is performed on populations where the effects of several compositional variations eg, protein content and composition are superimposed on each other (Skerritt 1998). Even with quite large sample sets, variations caused by experimental error can easily conspire to produce conflicting results in different sample populations. The classical technique of reconstitution provides insight into the effects of flour components on flour quality by directly altering the

chemical composition of the flour. Using this approach, the main components of flour are isolated and recombined in various ways for direct measurement on the reconstituted samples.

Gluten

The group of proteins in wheat which exert the most influence on the strength and elastic properties of dough, are the glutenins and gliadins. The polypeptide complex composed of glutenin, gliadin and lipid is defined as the visco elastic mass remaining after removal of the starch (Miflin *et al.* 1983). Gluten has a different role in making bread or pasta and also the process is different. In bread making, gluten must ensure extensibility and elasticity of the dough which expands and retains carbon dioxide that is formed during fermentation and baking (Liu *et al.* 1996). In pasta making gluten must be tenacious enough to retain the gelatinized starch granules during pasta cooking. In addition, water absorption of the pasta dough is around 31-35% compared to a bread dough of 60% (Liu *et al.* 1996).

Gluten quantity

Durum wheat breeding programs generally focus on quality factors associated with pasta and in some cases, bread. Gluten quantity and composition are the predominant factors associated with superior pasta texture. The protein matrix holds the starch granules during cooking to decrease the loss of solids in the cooking water and thereby reduce surface stickiness. With very low levels of protein extremely fragile spaghetti is produced with low firmness. High protein durum wheat allows spaghetti to swell when cooked (affects mouthfeel), reduces cooking loss and allows retention of firmness with overcooking which is also associated with less stickiness (Dexter *et al.* 1983; **Table 1**). Protein content has been noted as a primary factor associated with superior pasta quality (Feillet and Dexter 1996) with protein quality being less important. D'Egidio *et al.* (1990) showed that with low temperature (<40°C) pasta drying, protein content and gluten strength assumed equal importance in determining pasta quality. Whereas for higher temperature (>70°C) and ultra-high (>90°C) temperature drying, protein content was more important. These results have been supported by others (Matsuo *et al.* 1982; Autran *et al.* 1986; Edwards *et al.* 1993). At low drying temperatures, intrinsic differences in quality are reflected in both surface characteristics associated with stickiness and firmness, whereas at high temperatures only differences in firmness are apparent (D'Egidio *et al.* 1993).

Breeding programs have continued to make protein content one of several objectives because of its importance in marketing and quality determination of pasta (Sissons 2004). This also applies to breeding durum for bread making where a higher protein is generally more desirable. Protein in grain and semolina can be measured rapidly using near-infrared spectroscopy and calibrations are maintained against reference nitrogen determination using Dumas combustion analysis (Sissons *et al.* 2006).

Table 1 Effect of protein content on solids lost during cooking and firmness (peak force) of durum wheat pasta (adapted from Edwards *et al.* 1993).

	Protein content (%)		
	10.3	**13.5**	**17.6**
Optimum cooking			
Cooking loss (%)	9.4	8.9	7.1
Firmness (kg/mm)	1.10	1.44	2.02
Overcooked			
Cooking loss (%)	15.5	13.7	13.3
Firmness (kg/mm)	0.83	0.97	1.32

Gluten composition

Protein quality is difficult to define and has been characterised by a range of tests. One important aspect is gluten strength, an indicator of the gluten viscosity and elasticity. It is accepted that weak and inelastic gluten promoted poor pasta cooking quality but how much strength is optimal is not known. To increase gluten strength in weak semolina, blending with higher strength semolina is a commonly employed strategy by millers in Italy which enhances pasta texture (Marchylo *et al.* 2004).

Glutenin is a polymer whose molecular mass can exceed 100 million and when reduced is separated into subunits of different molecular size (Shewry *et al.* 2002) (**Fig. 3**). The majority of the subunits (60-80% of the glutenin) are LMW-GS of size 30,000-50,000 (Oak and Dexter 2006). These have been subdivided into B and C subunits, based on their molecular weight and isoelectric points (Oak and Dexter 2006). LMW-GS are encoded by genes at the *Glu-A3*, *Glu-B3* and *Glu-B2* loci on chromosome 1. The other larger size subunits are HMW-GS of size 80,000-120,000 encoded by genes at the *Glu-1* loci on chromosome 1. Four HMW-GS genes are present in durum wheat, but due to gene silencing, most genotypes only possess one to three subunits (Oak and Dexter 2006). The other main group of proteins in gluten are the gliadins (25,000-75,000) encoded by genes at the *Gli-1* loci on chromosome 1 and *Gli-2* on chromosome 6 (Skerritt 1998). They only contain intra disulphide bonds and interact with the gluten polymer via non-covalent forces (Oak and Dexter 2006). There are four types of gliadins classified on the basis of their mobility in acid polyacrylamide gel electrophoresis (α, β, γ, ω). Most durum wheats have either γ-45 or γ-42 gliadin bands and a few rare γ-gliadins have been reported (Oak and Dexter 2006). For more details on the gene structure of the glutenins and gliadins the reader is referred to reviews (Skerritt 1998; Shewry *et al.* 2002).

There is evidence of a quantitative linkage between LMW-GS and durum wheat quality. Two types of LMW-GS were described in early work, designated LMW-1 and LMW-2 (Damidaux *et al.* 1978). The LMW-1 is associated with γ-42 gliadin and LMW-2 with γ-45 gliadin due to a genetic linkage (Payne *et al.* 1984). Carrillo *et al.* (1990) showed that there are different LMW-1 and LMW-2 types: LMW 1, 1^-, 2, 2^-, and 2* with the LMW-2 and LMW-2^- showing higher gluten strength but with overlap in the range of strength measurements for the three LMW-2 types. The two LMW patterns are distinguished mainly by the presence of a strongly expressed protein band (42,000) in the LMW-2 type pattern (Masci *et al.* 2000; **Fig. 3**). This protein may contribute to the quality characteristics of durum possessing the LMW-2 type pattern. LMW-2 genotypes range in strength from moderate to high due to the diversity of alleles at the *Glu-B3* locus (Brites and Carillo 2001). The description in terms of LMW-1 and LMW-2 are imprecise because LMW-GS patterns are composed of different subunits encoded at different loci. There are several different B-LMW-GS polypeptide patterns distinguished by SDS-PAGE present in durum wheats and their allelic designation has been proposed as a better system to classify the LMW-GS in durum wheat (Nieto-Taladriz *et al.* 1997). The separate effects of allelic variants at *Glu-1*, *Glu-A3*, *Glu-B3* and *Glu-B2* on quality are providing more precise information about the role of these subunits (Vazquez *et al.* 1996; Brites and Carrillo 2001).

The impact of these components on dough properties and pasta quality has been studied for many years. Reports showed associations between glutenin allelic composition and gluten strength in durum wheat (Du Cros 1987; Pogna *et al.* 1990; Ammar *et al.* 2000; Brites and Carrillo 2001; Sissons *et al.* 2005a). Variation at the LMW-GS loci is associated with significant differences in the dough strength of durum wheat (for a review see D'Ovidio and Masci 2004). The allelic variation in LMW-GS has been suggested as a biochemical marker in breeding programs. However, Sis-

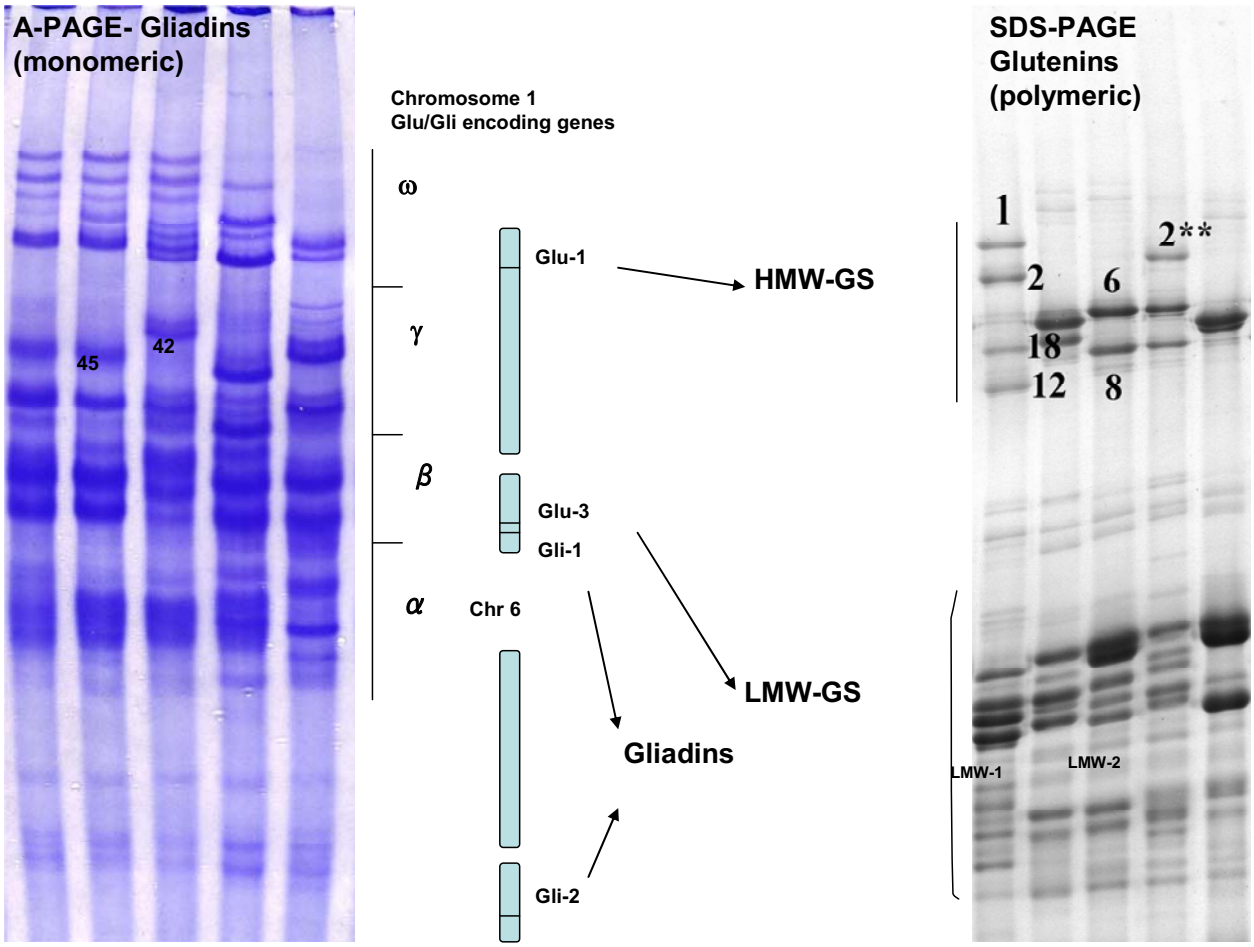

Fig. 3 Major glutenin and gliadin coding loci and separation on SDS-PAGE showing polymeric high molecular weight (HMW-GS) and low molecular weight (LMW-GS) glutenin subunits and by acid-PAGE showing monomeric gliadin components. Samples are a range of durum genotypes. Data from the author's laboratory.

sons *et al.* (2005a) pointed out that the use of this technique in the absence of a physical test for gluten strength, like gluten index or mixograph for screening is not recommended. This is because seasonal conditions can alter the MWD of the gluten polymer resulting in lower gluten index than expected for the specific glutenin allelic composition. Similarly, Edwards *et al.* (2007) found a wide range in gluten strength among HMW-GS groupings suggesting that particular allelic patterns indicate the potential for superior performance, but do not guarantee that level of performance. Brites and Carrillo (2001) found variation in dough properties measured using mixograph, SDSS, GI and alveograph could be partly explained by variation in glutenin composition controlled by *Glu-B1* and *Glu-B3* loci. They suggested avoiding crossing with durum wheats containing HMW-GS 20 and LMW-GS *b* and *k*. They suggested making intraspecific crosses with genotypes containing HMW-GS 14+15 with LMW-GS *c* or *j*. Other work supports the use of genotypes having HMW-GS 14+15 which were associated with a high SDSS (Oak *et al.* 2004). In other cases the presence of rare HMW-GS 2* and 3* subunits at the *Glu-A1* locus seem to offer higher SDSS and mixogram score than genotypes with the null allele, although the number of these unusual genotypes was only a handful (Raciti *et al.* 2003). Another unusual HMW-GS subunit pair found in a seed collection was 6+17 which had significantly higher SDSS than *Glu-B1* subunit 20 and 7+8. The influence of HMW-GS 1 on dough properties has shown a positive affect on gluten quality (Brites and Carrillo 2001; Martinez *et al.* 2005). Sissons *et al.* 2005a evaluated over 300 genotypes for LMW-GS and strength, and recommended that breeders avoid advancing lines with LMW patterns with *b* or *c* alleles at *Glu-A3*, with *b* or *f* alleles at *Glu-B3*, or with the *a* allele at *Glu-B2*. Edwards *et al.* (2003), using reconstitution, showed that the LMW-2 protein strengthened the dough more than LMW-1 protein.

Gluten strength is thought to relate to the balance between viscosity and elasticity (Shewry *et al.* 2002). It has generally been accepted that semolina from extra strong durum varieties is thought to produce firmer pasta and consequently gluten strength has become sought after in many markets where a higher price can be commanded (Marchylo *et al.* 2001). However, the scientific validity of this claim has not been proven and the exact cause of the gluten strength and the optimum level for firm pasta is not clear. A number of methods have been developed to measure gluten strength. The gluten index (GI) test (which is a ratio) has gained wide acceptance as a means of determining durum gluten strength (Cubadda *et al.* 1992) and is used in international trade specifications. However it is not a definitive test and other methods can be equally as effective in monitoring the gluten strength of durum wheats. For example, traditional methods for measuring bread wheat dough strength such as farinograph, mixograph and alveograph, have been adapted for assessment of durum semolima dough strength (Irvine *et al.* 1961; Quick and Donnelly 1980; Walle and Trentesaux 1980). The alveograph has become widely used by commercial wheat processors and is sometimes included in international trade specifications. While these methods correlate to a degree, they all measure different aspects. Nevertheless, they can rank cultivars equally with diverse strength (Marchylo *et al.* 2001; Edwards *et al.* 2007).

Sopiwnyk (1999) investigated seven durum wheat varieties with varying strength, gluten index (GI) range 2-84 with a narrow range in protein content (0.8%). They found no relationship between gluten strength and pasta cooking quality assessed using sensory and viscoelasticity of spaghetti and cooked firmness. Ames *et al.* (2003) tested 10

Table 2 Pasta-related physical dough properties and pasta texture for durum wheat varieties of variable gluten strength (adapted from Rao *et al*. 2001).

Property	Strong		Moderate		Weak	
	Pathfinder	**Navigator**	**Morse**	**Avonlea**	**Wascana**	**Stewart 63**
Semolina protein (%)	11.9	12.0	12.0	12.8	12.2	12.0
Gluten index (%)	87	89	62	38	40	27
Farinograph						
DDT (min)	6.5	11.0	7.3	5.5	4.5	3.5
Maximum consistency	490	440	500	560	560	560
Bandwidth (BU)	160	190	130	120	90	60
Alveograph						
P/L	1.6	2.0	0.8	0.5	0.4	0.5
Work (W)	301	268	179	145	80	49
Spaghetti texture						
Firmness (g)	393	406	422	430	378	318

Table 3 Baking-related physical dough properties and bread-making quality for durum wheat varieties of variable gluten strength (adapted from Rao *et al*. 2001).

Property	Strong		Moderate		Weak	
	Pathfinder	**Navigator**	**Morse**	**Avonlea**	**Wascana**	**Stewart 63**
Mixograph						
MT (min)	4.5	5.2	4	4.3	3.3	2.3
Bandwidth at peak (AU)	30.7	20.9	22.1	20.8	19.8	18.1
Work input (WI)	194	196	140	142	117	80
Remix-to-peak bread						
Absorption (%)	60	59	56	54	52	50
Remix energy (whr/kg)	3.3	2.4	2.3	2.0	1.3	0.5
Remix time (min)	3.0	2.6	2.2	2.0	1.3	0.7
Loaf volume (cm^3)	715	610	610	550	470	390
Baking strength index	92	78	78	66	59	50
Short process bread						
Absorption (%)	61	61	59	58	58	58
Mixing energy (whr/kg)	10.1	9.9	7.9	7.3	4.8	3.3
Mixing time (min)	10.0	10.2	7.9	7.3	5.5	3.9
Loaf volume (cm^3)	875	820	830	795	760	680

durum wheats varying widely in gluten index (GI 9-77) with a wide protein content (12.5-15.1%) and found inconsistent relationships between measures of pasta quality and gluten strength. Kovacs *et al.* (1997) found stronger significant correlations (r^2 0.5-0.8) between mixograph characteristics of dough (a measure of gluten strength) in 12 genotypes grown over three seasons and sensory characteristics of cooked pasta. However, protein content varied widely (12.7-15.0%) and this could have contributed to the relationships observed. In another study where a weak durum semolina was blended with increasing amounts of semolina from a extra strong Canadian durum, gluten strength increased markedly (GI 2-87) and pasta firmness increased from 696-1029 g measured using a texture analyser (Schlichting *et al.* 1999). An alternative approach to controlling variation in protein content which impacts on dough and texture measurements is to maintain this constant using the reconstitution method. In this way the effect of varying the gluten composition and strength at constant protein on pasta quality can be evaluated. Sissons *et al.* (2005b) found that varying the gluten composition in reconstituted dough by using gluten from diverse sources, produced a wide range in dough strength measured by mixograph and Kieffer rig attached to a texture analyser. It was found that mixograph development time was strongly correlated to pasta firmness, while maximal resistance to extension showed a weaker but significant correlation to firmness. The relationship between pasta texture and gluten strength in different durum wheats is well established (**Table 2**). In general, stronger gluten varieties give longer farinograph mixing time, wider bandwidth than weaker varieties. The alveograph which is performed at 50% water absorption and the W value is a good indicator of gluten strength. There was a clear differentiation across the various strength types in W values. The dough of strong varieties is less extensible, as indicated by higher ratios of tenacity (P; 1.1x height of the curve) to length of curve (L), which according to Quaglia (1988), limits the loaf volume potential of Italian durum wheat cultivars with strong gluten. Also, the W values are lower than found in strong bread wheat dough (Ammar *et al.* 2000). Only the very weak cultivar, Stewart had lower cooked pasta firmness, while Wascana was less firm than the extra strong varieties. The extra strength did not impart significantly firmer pasta than the moderately strong varieties but the extra strong varieties did have higher loaf volumes (**Table 3**). In more recent work, Sissons *et al*. (2007) found no clear relationship between gluten strength and cooked pasta firmness. While gluten strength is desired by wheat traders the scientific evidence does not support the contention that a stronger dough will make firmer pasta except when the gluten is very weak.

Typically many durum wheats have inferior gluten strength needed in the fermentation process to produce bread (Boyacioglu and D'Appolonia 1994). The dough tends to be inextensible and this can reduce oven spring in bread and therefore reduce loaf volume. In farinograph tests durum flours have higher water absorption than bread wheat flours due to the higher starch damage during milling, especially when semolina is re-ground into flour (Saperstein *et al.* 2007). Also, farinograph development times are often shorter than bread wheat flours and durum flours have unsuitable doughs for bread making when measured using the extensograph and alveograph (Boyacioglu and D'Appolonia 1994). The inextensibility typically associated with durum wheat is one reason thought to explain why durum bread has lower loaf volume than when made from hexaploid wheat (Ammar *et al.* 2000). Typical dough characteristics of durum wheats varying in dough strength used for baking are shown in **Table 3**. Mixograph mixing curves showed wide variation in dough mixing among the cultivars. The extra strong varieties have longer mixing times and this trend agrees with the mixing energy and mixing time data obtained during mixing using either baking process. The baking strength index (an indicator of loaf volume potential at a given protein) values obtained are lower than for common wheat, typically of 100. The strong durum exhibited

higher P/L ratios than common wheat, indicative of a less extensible dough. In addition, a less extensible dough is not desirable because of dough handling (sheeting) properties are inferior. A dough can be firm but also needs elasticity to avoid a doughy product. Durum with weak gluten exhibits more viscous and less elastic dough than bread wheat flours. This can be shown by a low extensibility using the alveograph (L) or extensograph. But the alveograph pressure and overall strength also tends to be too low in durum. To achieve an improvement in loaf volume a better balance of P/L and W is needed. By increasing the gluten strength, better loaf volumes and fermentation tolerance were obtained but they still fall short of loaves made from bread wheat (Marchylo *et al.* 2001). This deficiency may be due to the absence of the chromosome 1D encoded proteins thought necessary for bread dough's elasticity and extensibility (Redaelli *et al.* 1997). Efforts are being made to improve the baking potential of durum wheat by incorporating genes for proteins encoded by 1D (Lafiandra *et al.* 2000). Other approaches are to blend durum and bread wheat flours.

The lower alveograph W of durum dough compared to bread wheat dough could be explained by (a) durum wheats have a smaller percentage of HMW-GS compared to bread wheats (absence of D genome) which form more intermolecular S-S links; (b) the presence of more LMW-GS in durum wheat compared to common wheat means that LMW-GS mainly form linear polymers; and (c) length of the repeat sequence in LMW-GS is shorter than in HMW-GS. Gluten strength increases with increased average length of the glutenin polymer (Southan and Mac Ritchie 1999). Increasing the number of HMW-GS increases the glutenin polymer size. Therefore increasing the number of HMW-GS genes in durum wheat should increase dough strength. Replacement of the silent gene at the *Glu-A1* locus with allelic forms expressing both x- and y- types (eg 5+10, 2+12) has been achieved and these have larger amounts of polymeric gluten. Liu *et al.* (1996) prepared a set of D-genome substitution lines in a durum wheat Langdon cultivar and demonstrated a major impact on rheological properties. Replacing chromosome 1A with 1D increased the amount of glutenin and resulted in increased dough strength. This was the first demonstration of a positive effect of D-genome encoded proteins in durum wheat. Genetic transformation has been used to introduce a gene encoding subunit 1Dx5 into durum wheat which increased dough mixing time (He *et al.* 1999). Alternatively, chromosome engineering has been used to introduce various translocation lines into the Italian cultivars Svevo, Simeto and Lara. These lines contain chromosome with *Glu-D1* genes encoding HMW-GS 5+10 and 2+12 that replace the null allele present in the *Glu-A1* locus (Ceoloni *et al.* 2003). The same principal would apply in achieving a high loaf volume made from only durum flour, the stronger and more elastic the gluten, the higher the loaf volume that can be obtained as suggested by Saperstein *et al.* (2007).

In durum wheat *Glu-B1* and *Glu-B3* encoded proteins have a greater effect on gluten strength, spaghetti cooking quality and on bread making than any other genes (Boggini and Pogna 1989; Pogna *et al.* 1990). HMW-GS 7+8 combined with LMW-GS type 2 were associated with durum breads with greater loaf volume than those having LMW-GS type 1 and HMW-GS 20 (Peña *et al.* 1994). In contrast, Ammar *et al.* 2000 noted that genotypes with 6+8 had better overall bread making quality than those with 7+8 and 20. Therefore, the use of *Glu-B1* encoded HMW-GS alleles as markers to select for improved bread making quality in durum wheat might not be justified due to these discrepancies. The differences are probably cultivar background dependent. Breeding efforts to improve baking performance of durum wheat have been directed to manipulating the gluten composition. Durum wheats typically have weak gluten that is less extensible, characteristics detrimental to baking good loaves and have been found to be related to a reduced proportion of SDS-extractable polymer (Ammar *et al.* 2000). There is a range in bread making quality among different durum genotypes. Better baking performance was generally associated with greater dough extensibility and protein content while gluten strength was less important.

Glutenin subunits (LMW-GS and HMW-GS)

HMW-GS appear to have less critical effects on the gluten strength of durum wheat (Du Cros 1987; Porceddu *et al.* 1998) but this has not been clearly established due to limited genetic variability at the *Glu-1* loci present in modern durum wheat cultivars used in published studies. There is evidence that durum wheat with HMW-GS 20 tended to be weak (Ammar *et al.* 2000; Brites and Carillo 2001; Oak *et al.* 2004; Sissons *et al.* 2005a). Shewry *et al.* (2003) used wheat transformation to substantiate the association of HMW-GS 20 with inferior dough strength. The lower overall strength of this subunit is thought to be due to a lower density of intermolecular disulphide bonds (fewer cysteine residues at the N-terminus), resulting in lower polymeric protein content. The association of other HMW-GS in durum wheat to gluten strength are less clear with conflicting results and different methods to assess strength working with different populations (DuCros 1987; Pogna *et al.* 1990; Ammar *et al.* 2000; Brites and Carillo 2001; Sissons *et al.* 2005a). Edwards *et al.* (2007) found comparable dough strength in genotypes with 6+8, 7+8 and 7+16. This makes them of limited value as markers for selecting higher gluten strength in breeding programs.

There is limited information about how variation in the ratio of HMW-GS to LMW-GS influences pasta quality. One recent study (Sissons *et al.* 2007) showed that the augmentation of HMW-GS in semolina dough greatly increased dough strength but did not affect cooked pasta firmness. In that study, the HMW-GS were added in a reduced form and not reoxidised which may have prevented sufficient incorporation of the added HMW-GS into the gluten polymer because of insufficient available free thiol groups (Antes and Wiser 2001). While an increase in dough strength was still observed, this may not have affected the pasta structure sufficiently to alter its resistance to deformation (firmness). Edwards *et al.* (2007) found significant negative correlations between the HMW-GS/LMW-GS ratio and alveograph P, L and W and mixograph mixing time and bandwidth at peak, albeit the correlations were small in magnitude (r^2 0.08-0.20). Also the data was unevenly distributed which could bias the correlations obtained. This information showed that as the proportion of HMW-GS increased, dough strength decreased and that is in contrast with the findings of Sissons *et al.* (2007) using a different approach. The formation of a well developed network for durum wheat would preferentially involve LMW-GS over HMW-GS. Data from Edwards *et al.* 2003 showed that durum gluten does not form entanglement network involving HMW-GS as the primary contributor to gluten strength, but is more likely to be based on an associative polymer type of structure involving LMW-GS, where shorter chain lengths result in greater density of cross links for a given volume and therefore impart greater strength.

The association of HMW-GS with baking quality of durum wheats is better defined in bread making (Liu *et al.* 1996; Palumbo *et al.* 2002). The *Glu-B1* coded HMW-GS 20 is associated with inferior baking quality with a ranking for other *Glu-B1* alleles suggested to be 7+8>20>6+8. This is similar to the effect observed in hexaploid wheat (Liu *et al.* 1996). However, other work has suggested that the 7+8 pair were too tenacious for bread making while the presence of any subunit encoded by *Glu-A1* were favourable for bread making (Boggini *et al.* 1994). Ciaffi *et al.* (1995) showed that LMW-GS type 2 proteins are associated with higher loaf volumes. There is still the issue of a narrow range in genetic variability for HMW-GS and this needs to be expanded.

Glutenin to gliadin ratio

Early studies found a correlation between a high glutenin to gliadin ratio and strong dough (Wasik and Bushuk 1975; Dexter and Matsuo 1978). Introducing D genome proteins (HMW-GS 5+10) into durum increases dough strength and the glutenin/gliadin ratio (Liu *et al.* 1994; Lafiandra *et al.* 2000). Edwards *et al.* (2003) found that by adding a glutenin-rich fraction (consisting of HMW-GS and LMW-GS) to base semolina increased mixograph dough strength. Sissons *et al.* (2005b) found that increasing the glutenin to gliadin ratio of base semolina improved dough strength and the percentage of unextractable polymeric protein but with variable affects on pasta quality. Using reconstitution, this group showed that by increasing the amount of a glutenin-rich fraction, dough strength was increased while additional gliadin and LMW-GS decreased strength. These changes had no impact on spaghetti texture (Sissons *et al.* 2007). Increasing the glutenin content causes a shift to higher molecular weight in the gluten molecular weight distribution (an increase in the unextractable polymeric protein). This is measured using size-exclusion HPLC or field flow fractionation or multistacking SDS-PAGE. The molecular weight distribution influences dough properties (Southan and MacRitchie 1999). Increasing the glutenin to gliadin (polymeric to monomeric) ratio by addition of glutenin enriched fraction to base flour strengthened the dough (Uthayakumaran *et al.* 1999; Edwards *et al.* 2003; Grabberger *et al.* 2003; Sissons *et al.* 2005b). However, Edwards *et al.* (2007) found using genotypes with widely varying dough strength there was only a poor correlation between the glutenin to gliadin (polymeric to monomeric) ratio and measures of dough strength. This may be related to variation in the LMW-GS in the population studied. Another problem encountered was a significant correlation between the glutenin to gliadin ratio and grain protein content.

Non-gluten proteins

The high molecular weight albumins have been detected in wheat flour and are covalently bound into the glutenin matrix (Peruffo *et al.* 1996). Some of these are β-amylases and they tend to polymerise via disulphide bonds and link with LMW-GS. The presence of these correlates with poor dough properties (Krattiger *et al.* 1991).

Grain hardness is a key determinant of milling performance for semolina production. Proteins are responsible for determining grain texture (hardness) of wheat grain and these have been called friabilin, grain-softness protein and puroindolines. All are related and indeed friabilin is a puroindoline. However, durum wheat does not contain the genes for the puroindolines and their absence is the reason for the very hard texture of durum wheats (Morris 2002).

During preharvest sprouting endoproteinases can be released and these can degrade gluten affecting the baking quality (Kruger and Matsuo 1982). A proteinase purified from germinated durum wheat that preferentially hydrolysed hordeins been described (Bottari *et al.* 1996). It is likely that in sound grain, proteases are probably not important in pasta quality only in highly sprouted grain. Insect proteases have been detected in wheat that can cause preharvest damage by injecting proteinase enzymes into the kernel which attack the storage proteins (Sivri *et al.* 2004). In my knowledge there are no studies published describing this in durum wheat.

Enzymes

α-Amylases: α-Amylases are endo-splitting enzymes that hydrolyse the α-1,4 glycosidic bonds of starch. They are absent in mature grain but are produced during germination (Feillet 1988). Therefore, they have an important role only when preharvest sprouted grain is used in pasta manufacture. Even in sprouted grain some of the enzyme is removed from the semolina during milling because the enzyme is located in the embryo and further losses in enzyme activity occur during HT drying and what remains only survives short periods of cooking (Kruger and Matsuo 1982). Its presence in the grain can result in higher sugar levels, which during HT drying, leads to increased pasta redness. Sprout damage is known to be detrimental to wheat bread making quality, primarily due to elevated levels of α-amylase. Dexter *et al.* (1990) prepared durum wheat composites to give falling numbers (FN, an indicator of sprouting damage to the semolina) of 60-520 s at constant wheat protein content. The most highly sprouted sample exhibited slight checking in the pasta, which became more pronounced following 3 months storage. None of the other samples, including 12 with wheat FN below 150 s, exhibited checking. Cooked spaghetti stickiness, firmness, and resilience were not related to semolina α-amylase activity. There was no evidence that high α-amylase activity was detrimental to spaghetti storage stability as measured by strand strength and spaghetti cooking quality. In bread making some α-and β-amylase is required for effective fermentation. Durum wheat grown under dry and hot conditions generally has high FN 460-660 s indicating extremely low amylolytic activity. Such wheats favour the end-quality of pasta products, but in bread making they must be blended with higher amylolytic activity wheats or be supplemented with malt or pure enzymatic α-amylase, otherwise they would lead to heavy underdeveloped hard bread with low keeping quality (Josephides 1996).

Lipoxygenase: Lipoxygenase (LOX) refers to a group of enzymes that are highly specific for the oxidation by oxygen of fatty acids that contain a *cis*-*cis*-1,4-pentadiene unit, to produce conjugated hydroperoxidiene derivatives. Lipoxygenase activity varies widely among durum wheats and there exists several isoenzymes (Yemenicioglu and Ercan 1999). The bright yellow colour of durum wheat products is the result of the natural carotenoid pigment content (Irvine and Anderson 1953) and of their oxidative degradation by LOX. The extent of LOX degradation depends on several factors, among which the intrinsic quality of the semolina and the processing conditions are considered to be the most important. The phase mainly responsible for pigment loss is pasta processing, particularly the dough mixing stage, when a substantial decrease in pigment content occurs. Pigment loss is highly correlated with semolina LOX activity. Semolina LOX reaction could be inhibited by beta-carotene and a lower semolina bleaching was observed in samples having a higher carotenoid content (Trono *et al.* 1999). As semolina LOX appears to be inhibited by endogenous carotenoids, it is suggested that a high carotenoid content of semolina is desirable to impart a good yellow colour and possibly to partially prevent carotenoid bleaching during pasta processing. Changes in LOX activity during pasta processing showed slight decrease in activity during processing into semolina, and significantly during processing into macaroni, so that almost no LOX activity was left in macaroni obtained from cultivars with intrinsically low LOX activity. LOX was stable at 50°C, but was rapidly inactivated when heated at 65° and 75°C (Yemenicioglu and Ercan 1999). Therefore, a high pigment content, located in the interior of the whole grain, and a lower LOX activity in semolina must be the selection characteristics by which breeding programs obtain a bright yellow pasta.

Peroxidase and Polyphenol Oxidase: Both peroxidase and polyphenol oxidase enzymes occur in durum wheat, the latter being at much lower levels than in common wheat (Lamkin *et al.* 1981). Peroxidases are enzymes that catalyse the general reaction (Feillet 2000):

$$ROOH + H_2O_2 \rightarrow ROH + H_2O + 1/2O_2$$

and are not specific in their reaction, catalysing the oxidation of a large number of phenols which occur in the grain. Polyphenol oxidases (PPO) catalyse the oxidation of phenolic compounds in the presence of molecular oxygen. They occur widely in plants and cause the enzymatic browning in food material through an initial oxidation of phenols into quinones. Quinones readily undergo self-polymerisation or

condensation with amino acids or proteins via their amino groups to form complex brown polymers.

Both these enzymes are formed early in grain development and reside mainly in the pericarp and grain layers. As the grain develops, these enzymes decrease to low levels in mature grain (Kruger and LaBerge 1974). Pasta products from durum wheat varieties with a high peroxidase activity develop a brownish colour during processing, the brown colour tends to mask the yellow colour when it reaches substantial levels (Kobrehel and Gautier 1974). Therefore, reducing browning is desirable in durum wheat. Durum wheat cultivars vary in their peroxidase and PPO activity but more recent work has not found a relationship between peroxidase activity and semolina brownness (Delcros *et al*. 1998). Due to the low level of PPO in semolina, its role in pasta brownness is unlikely but it may be the cause of the inherent brownness in the semolina because this could have formed during grain maturation when PPO levels are much higher where they can oxidise the abundant phenols present in immature wheat (Kruger 1976). However, since the source of PPO is the bran layers, excess bran in the semolina arising from poor purification could produce brownness in the pasta made from such semolina.

Starch

Starch is deposited in the plastid of higher plants in the form of granules within membrane-bound organelles called amyloplasts in cereals and comprises ~70% w/w of the endosperm in wheat (Stone 1996). Starch is a polymer of α–linked glucose residues and is comprised of two molecules, amylose and amylopectin. Amylose is a lightly branched polymer, with molecular weight 10^5-10^6. In wheat it typically represents about 25% of the starch granule but in some genotypes this can vary greatly from 0-40%. Amylopectin is a highly branched polymer with MW 10^7-10^8. The amylose polymer can form complexes with lipids. This amylose-lipid complex resists leaching from the starch granule and also prevents entry of water into the granule. Native starch granules also contain small amounts of proteins, lipids and minerals (2% of granule mass) (Chibbar *et al*. 2005).

Durum and other wheats have biphasic granule distributions. The large A-type granules are lenticular in shape with diameters 20-25 μm. The B-granules are roughly spherical in shape and average 5-6 μm (**Fig. 4**). Starch granules exhibit concentric ring structures, that are visible in sections though granules.

When starch is heated in excess water it gelatinises, which is defined as the collapse of the molecular order within the granule and can be measured using a differential scanning calorimeter (DSC). During heating the starch absorbs water which causes the granules to swell and eventually rupture. At the gelatinisation temperature of starch (~50-70°C, **Table 4**) energy is absorbed and an endotherm can be measured (△H J/g). The loss of crystallinity leads to a rapid swelling of the granule, dissolution of starch and exudation of components from the granules and a rapid increase in viscosity is produced. The latter can be measured using a rapid visco analyser (RVA). Amylose leaches from the granule but amylopectin remains associated. With further heating the granules become distorted, soluble starch is released into the solution and eventually total disruption of the granule occurs resulting in a decrease in viscosity. As the paste is cooled and stirred, amylose retrogrades quickly and the viscosity of the paste increases to reach a final value (**Table 5**).

Wheat flour dough is a composite material in which gluten forms the continuous matrix and starch granules act as a filler within the matrix. Edwards *et al.* (2002) used a model system consisting of gluten-starch dough where glass beads (of similar particle size distribution to the durum starch) was substituted for the starch on a volume basis from 0-100%. Dough linear viscoelastic properties were weakened on substitution of starch with glass beads. Coating the beads with bovine serum albumin to provide H bonding sites reduced the friction contact between the beads and reduced dough moduli and increased compliance of the gluten in comparison to non-coated filler particles. This result confirmed the importance of protein-starch bonding on durum dough linear viscoelastic behaviour. Other work has shown that by removing the starch surface lipids and proteins the starch has altered DSC and RVA properties but this had no affect on the starch-gluten behaviour in pasta (Delcour *et al.* 2000). These authors concluded that the starch interaction behaviour in uncooked pasta is mainly by physical inclusion.

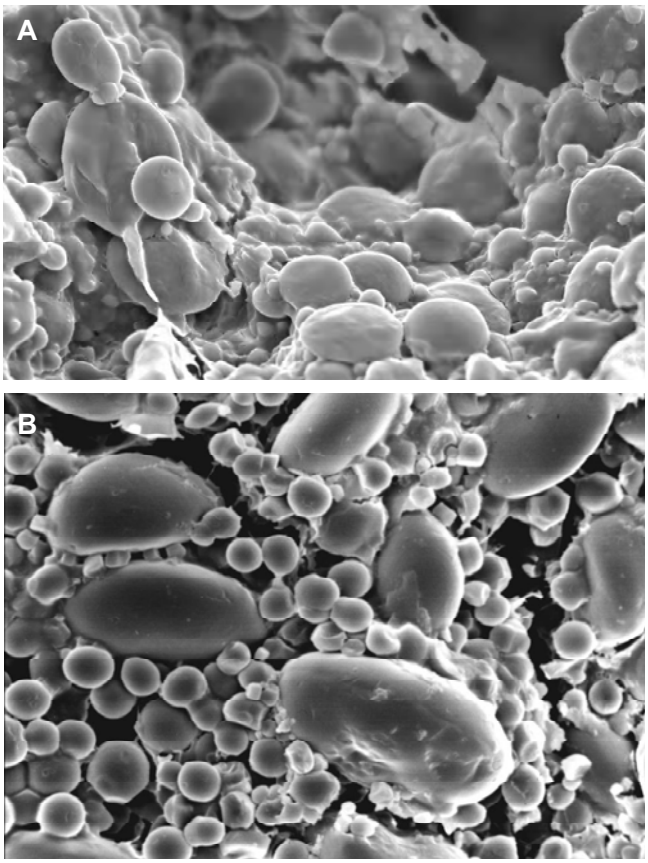

Fig. 4 Starch granules from (**A**) uncooked spaghetti made from durum semolina, cross section, 1000X (**B**) fractured endosperm from durum wheat starch, 1000X. Scanning electron micrographs for (**A**) by NSW DPI author's laboratory and (**B**) courtesy of Dr M Turner, University of Sydney, Plant Breeding Institute, Narrabri, Australia, by scanning electron microscopy

Amylose to amylopectin ratio

The composition of starch is controlled by comparatively few genes and naturally occurring alleles of these genes exist in cereals, leading to phenotypes having starches ranging from almost no amylose (the waxy starches) through to elevated amylose. The waxy character is caused by a mutation in the waxy locus that affects the production of amylose. Therefore, waxy mutants do not produce amylose and their starches are basically 100% amylopectin. Waxy mutants, lacking the functional waxy protein (granule-bound starch synthase), have been identified in durum wheat. All three possible waxy mutations in durum wheat (*Wx-A1*, or null 4A and *Wx-B1* or null 7A and the double null form) have been discovered (Urbano *et al.* 2002). Partial waxy, *Wx-B1*durum wheats lines had about 5% lower amylose content than the wild type (Sharma *et al.* 2002). Starches from partial and full waxy durum wheats have higher RVA peak viscosities and breakdown compared to normal lines (**Table 5**). The high peak viscosity may be due to a decrease in amylose because amylose suppresses swelling and maintains starch granule integrity so a reduction will encourage

Table 4 Starch properties of non-waxy, partial waxy and waxy durum wheats (adapted from Chakraborty *et al*. 2004).

Genotype	Wheat type	Starch (%)	Amylose (%)	Crystallinity (%)	Starch gelatinisation[a]			
					T_o (°C)	T_p (°C)	T_c (°C)	△H (J/g)
Ben	Durum	67.2	29.2	10.1	51.7	59.8	70.2	7.7
Parshall	Partial waxy	68.3	22.3	11.1	51.1	62.2	72.5	9.6
WD1	Waxy durum	64.1	2.6	15.3	55.9	67.4	79.8	12.1
WD2		67.1	2.4	15.3	55.9	67.3	80.8	11.2
WD3		66.0	2.3	15.3	55.2	67.4	79.7	11.3
WD4		66.0	2.3					
LSD		1.3	0.5	1.7	3.0	1.2	1.7	3.3

[a] T_o = onset temperature of starch gelatinisation; T_p = peak temperature; T_c = completion tempereature; △H= enthalpy change

Table 5 Starch properties of non-waxy, partial waxy and waxy durum wheats (adapted from Chakraborty *et al*. 2004).

Genotype	Wheat type	RVA Pasting (RVU)[a]				Texture
		PV	BKD	STB	PT	Firmness g
Ben	Durum	201	70	147	9.1	54.0
Parshall	Partial waxy	286	182	106	9.1	42.0
WD1	Waxy durum	265	183	43	4.8	6.9
WD2		269	187	45	4.9	7.8
WD3		258	179	43	4.9	7.3
LSD		4.6	4.3	2.7	0.1	2.0

[a] PV = peak viscosity; BKD = breakdown; STB = stability; PT = peak time (min).

high peak viscosities. This result was consistent with those in waxy hexaploid wheats (Kiribuchi-Otobe *et al.* 1997). In full waxy durum (no amylose) the isolated starch had higher onset and peak gelatinization temperatures and enthalpy of gelatinisation (**Table 4**). Pasta made from the full waxy durum was unacceptably softer (**Table 5**). Further work found full waxy durum wheat made pasta with shorter cooking times, much lower cooked yellowness and much softer in texture than the non-waxy durum (Grant *et al.* 2004).

Durum starch was substituted with waxy hexaploid wheat starch to obtain amylose contents of 0.7-22.9% (Gianibelli *et al.* 2005). As the amylose content was decreased by increasing the percentage of waxy starch in the flour blends, cooking time and cooked pasta firmness decreased while stickiness increased. Soluble carbohydrate, like amylose exuding from the starch granules during cooking is a probable cause of pasta stickiness (Grant *et al.* 1993) so how can stickiness be increased with reduction in amylose? Carbohydrate in the form of amylopectin or amylopectin fragments, not amylose, will be released in the waxy wheat pasta and contribute to the stickiness on the pasta surface.

Little is known about the characteristics of high amylose wheat starch, especially in durum. The effect of increased amylose content above normal levels ~24-28% on pasta technological quality has not been reported due to the absence of durum wheat with elevated amylose content. Using reconstituted flours Soh *et al*. 2006 showed that by progressively replacing the durum starch with high amylose maize starch (amylose 28.5-74.1%) resulted in much higher flour water absorption, which could be due to a higher fibre content coming from the Hi-maize® starch. Dough extensibility showed a progressive decrease with increase in amylose, consistent with a reduced dough elasticity reported in high amylose wheat flour (Hung *et al.* 2005). There was a tendency for pasta firmness to increase as amylose content increased. In high amylose starches, the granules are more tightly packed and on swelling have more resistance to rupture and deformation. This might explain the increased tendency to produce firmer pasta. Pasta water uptake decreased and cooking loss increased with elevation of amylose content (Soh *et al.* 2006). The ability of amylose to restrict swelling probably contributed to this drop in water uptake. The increase in cooking loss is probably a result of an increased availability of amylose to leach during cooking. This suggests that developing durum wheat with slightly enhanced amylose content would provide new starch types with potential food applications. Other benefits of higher amylose durum pasta are related to the affect on the food glycaemic index. Hospers *et al.* (1994) fed humans pasta with ~40% amylose compared to the control pasta of 25.9% amylose and noted significantly lower postprandial blood glucose and insulin levels.

There have been attempts to create durum wheats with elevated amylose content. Two starch branching enzyme isoforms (SBE) are present in most cereals. In wheat genetic elimination of starch synthase II (SSII) has been reported to increase apparent amylose concentrations. SDS-PAGE screening of wheat germplasm identified wheat lines lacking SSII derived from A and B genomes. Such a null has been crossed with tetraploid wheat to obtain a durum with elevated (32-35%) amylose content in the starch. However, genetic analysis of many hundreds of lines showed that amylose concentration in wheat is more complex and governed by pleiotropic effects (Chibbar *et al.* 2005).

The effect of reduced amylose content on durum bread and couscous quality is not known. Although the addition of waxy durum flour to a strong bread flour at 10-30% resulted in a decrease in firming of bread. This softening was more effective than using shortenings, suggesting a possible use of waxy durum flour as a low fat replacement for shortening in baked products (Bhattacharya *et al.* 2002). For traditional bread making using hexaploid waxy wheat their benefits have been described in a bakery for retarding staling and extending shelf life and formation of new texture of breads with soft, viscous and glutinous breadcrumbs (Hayakawa *et al.* 2004). These benefits have not been researched in durum bread.

Proportion of A:B starch granules

When the percentage of B-granules are increased in durum starch, the dough absorbs more water because the smaller B-granules have a higher surface to volume ratio and are able to hydrate and swell more efficiently and bind more water than A-granules. Therefore, increased B-granule content should increase flour water absorption (Soh *et al.* 2006). This result is supported from other work with common wheat flours (Kulp 1973; Stoddard 1999; Yun *et al.* 2000; Chiotelli and Le Meste 2002). The high demand for water by B-granule starch might create an imbalance of water distribution in the dough resulting in weaker dough. Evidence from the literature is unclear. Soh *et al.* (2006) found that an increased dough resistance up to 32% B-granules, followed by a decrease at B-granule above 40% whereas in another study using granules with sizes 6.5-19.5 μm decreased resistance to extension (Sebecic and Sebecic 1995, 1999). Park *et al.* (2005) proposed that more small granules could interact more intimately as filler particles with the continuous gluten phase in dough, which causes a corresponding increase in resistance to mixing.

There has only been one study reporting the affect of varying B-granule content on pasta quality. Soh *et al.* (2006) found that spaghetti made from samples exhibited higher cooked firmness and lower stickiness at 32-40% B-granules compared with the control reconstituted sample (22.7% B-granules). Pasta cooking loss decreased with elevated B-granule content which is a positive feature. Smaller granules have a greater surface area so increasing the percentage of these might be expected to extend the interactions between the starch granules and gluten and this may decrease the loss of amylose, reducing cooking loss (Vasanthan and Bhatty 1996). This might also explain a reduced stickiness in some of the samples since both measurements are a reflection of the leaching of amylose from starch granules. Chen *et al*. (2003) using potato starches, found that noodles containing more small granules have firmer cutting properties.

Damaged starch

The amount of damaged starch in semolina granulars and re-grounds is much more important in baking bread. To ensure adequate gassing during fermentation, sufficient damaged starch is needed. Under harsh grinding, the hard texture of durum can result in excessive damage starch that can be detrimental to baking quality (Dexter *et al.* 1994). Reduction of the particle size of semolina granulars increases starch damage and gassing power, increases farinograph water absorption and decreases development time. As particles become finer, dough stability decreases (Saperstein *et al.* 2007). This did not result in any clear trend in loaf volume using the Remix-to-peak baking process with different effects of grinding granulars to finer particles dependent on the cultivar.

Using semolina of too fine a granulation (<210 μm) can lead to high starch damage which can increase cooking loss in bran-rich pasta (Gauthier *et al.* 2006) and lower firmness and water absorption in whole wheat pasta (Manthey and Schorno 2002). This is not supported in another study in bran-free pasta where the use of semolina regrounds had no affect on pasta quality (Grant *et al.* 1993). The use of fine semolina can increase the amount of reducing sugars in the dough mixture allowing the action of endogenous α-amylase to produce reducing sugars. These are converted to Maillard products during high temperature pasta drying (Sensidoni *et al.* 2003) producing undesirable colour in the final product.

Non-starch polysaccharides (NSP)

The non-starch polysaccharides found in hexaploid wheat (with similar values in durum wheat) account for 3-8% of the grain and consist of cellulose (2.7% of dry weight), β-glucans (1%), arabinogalactan-peptides (AGP) and arabinoxylans (7.6%) (Stone 1996). These mostly have a structural role and make up about 75% of the endosperm cell walls. Although the arabinoxylans (AX) represent a minor component of the grain compared to starch and protein, they can have major effects on the use of the cereal grain due to their hydration properties and ability to form viscous solutions and will be discussed in more detail. Wheat is not recognised as a source of β-glucan because of its much lower content compared to barley and oats and as such will not be discussed further. However, it should be noted that β-glucan enriched milling fractions derived from barley have been added to pasta to increase its soluble fibre content for health benefits, however, there was deterioration in pasta quality (Cleary and Brennan 2006). There is potential to use other non-starch polysaccharides as fibre ingredients with functional food value in pasta and bread.

Arabinoxylans

The main polymer in wheat cell walls is AX and this has been classified into water extractable (WE-AX) and water unextactable (WU-AX) forms, with the latter type being strongly embedded in the cell wall network (Courtin and Delcour 2002). The AX have a xylan backbone consisting of β-1,4 D-xylopyranose units some of which are substituted by α-L-arabinose side chains. Durum wheat AX contents ranged between 4-6% (dry basis) in a study of five French varieties and in semolina between 0.58 and 3.0% (Lempereur *et al.* 1997) where about 25% is WE-AX. A more recent study has confirmed this range on 90 breeding lines (Saulnier *et al.* 2007). Since AX have a high water binding capacity (Courtin and Delcour 2002), these components are expected to have an impact on pasta dough properties. When durum semolina WE-AX were interchanged with HRS wheat WE-AX, macaroni samples experienced a decrease in cooked firmness (Sheu *et al.* 1967). During dough formation and pasta making a significant amount of the total AX is solubilised. This is not due to the action of endogenous enzymes, since these are hardly detectable during the processing but due to solublisation caused by mechanical forces during pasta preparation (Ingelbrecht *et al.* 2001). Only minor amounts of WE-AX were released from the pasta during cooking but this increased with overcooking. Addition of crude WE-AX at 1 and 2% to pasta dough had only small effects on pasta texture (Edwards *et al.* 1995). This was supported by more recent work using highly purified WE-AX added to durum dough at 0.125-2.0% with minimal impact on pasta texture but causing a large increase in dough water absorption (12%) and dough weakening (Turner *et al.* 2008).

The addition of microbial endoxylanases to semolina dough reduced the farinograph maximal consistency to different degrees because these enzymes convert some of the WU-AX into WE-AX and are also inhibited by endogenous inhibitors in the semolina (Ingelbrecht *et al.* 2000). Endoxylanases (EC 3.2.1.8) are enzymes that hydrolyse the xylan backbone in AX in a random manner. Further, these workers also showed a reduction in pressure at the extrusion die using added microbial endoxylanases during pasta processing with minimal affect on pasta quality (Ingelbrecht *et al.* 2001). These enzymes converted WU-AX to WE-AX and a reduction in the molecular weight of the WE-AX. At high enzyme doses, these soluble AX molecules were not readily leached from the pasta during cooking and overcooking, thus retaining the soluble fibre benefits. Endoxylanase functionality is determined largely by its preference for hydrolysis of WE-AX or WU-AX and the addition of such enzymes in processing can impact on the product. Further work where two endoxylanses with different substrate specificities were added to pasta dough, one that preferentially hydrolyses WE-AX (XBS) and the other that preferentially hydrolyses WU-AX (XAA), showed minimal impact on pasta colour, cooking time and firmness but with the benefit of higher soluble fibre which was largely retained in the cooked pasta (Brijs *et al.* 2004).

Lipids

Lipids are important components of wheat despite being only 1-3% (dry matter) of the grain. There are the starch bound and non-starch lipids in semolina. The former interact with the amylose helix during biosynthesis and exist as amylose-inclusion complexes in the starch granules (Morrison 1978). Non-starch lipids refer to all other lipids in the grain and these are divided further into free (soluble in non-polar solvents) and bound (soluble in cold polar solvent mixtures). In durum semolina free lipids represent 64% of total lipids. These components affect pasta and bread making quality which have been reviewed several years ago (Laignelet 1983; MacRitchie 1983). Lipids are important in determining the colour of pasta due to pigments and lipoxygenase (discussed previously). The pigments found in the endosperm consist of primarily xanthophylls. Lutein is the main xanthophyll in durum wheat. A bright yellow colour is desired in pasta products which arises from the pigments in the endosperm although some reduction in colour can occur

during pasta processing. This is due to the oxidation of yellow pigments catalysed by the enzyme lipoxygenase present in semolina, mainly during hydration, mixing, extrusion and the early lower temperature drying phase. The yellow pigments are easily oxidised by LOX to catalyse the reaction. High temperature drying (>70°C) and short mixing times (<1 min; Polymatic systems) minimise the impacts of LOX. Other lipids in durum wheat include hydrocarbons, sterols, glycerides, fatty acids, glycolipids and phospolipids (for a review see Youngs 1988). Hydrocarbons are a minor component (0.0036% of dry weight). Free sterols have been measured in durum wheat at levels of 25-38 mg/100 g wheat, mainly consisting of sitosterol and campesterol (Youngs 1988). The triglycerides are the main lipid in durum wheat and the reader is referred to the review of Youngs (1988).

There is less known about the changes in lipid composition during pasta processing than during bread making. Lipid content does not appear to decrease during pasta processing but it is less easily extracted from pasta than semolina, suggesting that under mechanical stress of extrusion, lipids undergo chemical changes or are complexed with proteins and carbohydrates (Laignelet 1983). Monoglycerides with saturated fatty acids are effective in complexing amylose in solution and when added to dough, pasta stickiness decreased and overcooking tolerance was improved (Laignelet 1983; Matsuo *et al.* 1986). The commercial use of monoglycerides is the subject of many patents. During the dough mixing process, free lipids interact with other flour components, especially proteins to provide a beneficial influence on gluten strength. The mixing process accelerates the formation of hydrophobic bonding of non-polar lipids with acid soluble components such as glutenin, gliadin, albumin and nitrogenous non-proteins. Whilst, polar lipids interact mainly with glutenins (Chung *et al.* 1978; Chung 1986), free polar lipids can also bind to gliadin by hydrophilic bonds. These bonds enhance protein interactions, which provide a better structural support for the gluten network (Chung *et al.* 1978). The interaction of free polar lipids with protein in common wheat have been found to have beneficial effects on quality. These interactions improve surface tension of dough gas cells and improve stability of dough foam structures, which increases loaf volume (MacRitchie 1983). Removing lipids from flour causes a strengthening of the defatted dough compared to the undefatted dough and changes its colour by removal of pigments. This can be restored by adding back the extracted lipids. The lipid appears to modify the interactions between proteins (MacRitchie 1983). Little is known about the influence of lipids on pasta quality. Matsuo *et al.* (1986) found that removal of total lipids and non-polar lipids had detrimental affects on pasta quality such as an increase in stickiness of pasta and cooking loss. More research is required to better understand the role of lipids in both pasta and breadmaking from durum.

FUTURE DIRECTIONS

It has been suggested that durum flour has inextensible dough and this is the main reason why loaf volumes are inferior to bread made from common wheat. The quantity of HMW-GS impacts directly on dough strength but further work in identifying the gluten proteins that are most desirable for optimum dough properties is needed. The introduction of D genome associated proteins is a worthwhile goal to broaden the extensibility of durum dough. This can be done by cytogenetic (for example the introduction of chromosome 1D into durum wheat) or genetic transformation methods (the expression of additional genes encoding HMW-GS in durum wheat). For pasta, it is not clear what role dough extensibility has in cooking quality.

More work is needed to extend the natural variation in starch composition of durum wheats through breeding. Recent research has shown that starch composition can influence pasta quality while the impact on durum breadmaking quality is unknown. The waxy durum wheats might have applications in food since waxy starch has unique properties. Similarly, the development of higher amylose durum wheat with different swelling properties and enhanced soluble fibre content could provide pasta with improved nutritional value. The creation of durum with widely varying swelling power and B-granule content would also be worthwhile.

More research is needed on the impact of lipid composition on both pasta and durum breadmaking quality. Techniques like reconstitution could be used to elucidate the role of specific lipid components on quality. More research is needed on the use of arabinoxylan degrading enzymes and lipases in pasta production and on the benefits of adding more non-starch soluble fibre on quality and nutritional value of pasta.

Pasta represents an excellent base food for improving its human health value. The development of pasta with added soluble and insoluble fibre, antioxidants, resistant starch, for example, would improve pasta nutritional value. However, to provide a pasta of acceptable eating quality, more research is needed to develop such pasta of similar quality to durum only pasta in addition to proving by human feeding trials the benefits to health, like lowering pasta glycaemic index, reducing cancer and diabetes risk.

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