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GRAIN STRUCTURE AND END-USE PROPERTIES

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Abstract

Practical implications of grain structure relate to every step from grain development and production through marketing to processing, utilization, and consumption. The structure and adherence of the hulls may contribute to protection of grain during germination or malting and protection against insect infestations. Germ retention during threshing and separation during processing depend on the germ structure and location in the kernel. The subaleurone and central endosperm layers differ in cell size, shape, and structure and in composition, especially with regard to protein contents and quality. The main factors in grain hardness are the intrinsic hardness of the main components, the strength of interaction within the cell, and the interaction of individual cells to produce overall grain structure.

Endosperm structure and hardness is related to wheat conditioning, to breakage in milling, and to the structure and composition of the milled flour particles. Milling quality is governed by morphological characteristics of the wheat kernel and its mechano-physical properties and by the methods of grinding and separation. Reducing changes in texture and structure during drying of maize and rice are important in minimizing breakage during handling, storage, and transportation, dust formation, and infestation. Differences in grain structure are expressed in differences in composition, gradients of components in grain tissues, and end-use properties. Those differences have important nutritional implications. New microscopic methods to determine grain structure, composition, and end-use properties have the potential of contributing to improved nutritional quality and utilization of cereals by modifying-restructuring grain morphology through classical plant breeding and genetic engineering.

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Introduction

Grain structure is an expression of grain composition as it reflects properties from the standpoint of plant physiology. The plant does not synthesize or incorporate components into structures unless they have a specific function in preservation or propagation of the species. Cereal chemists and technologists, on the other hand, are interested in another set of properties --the function the grain or its fractions can perform in the production of nutritious foods, that have good shelf life, and are acceptable to the consumer.

Thus, in a way, grain structure forms the link between composition that is the source of our basic knowledge of biological systems and utilization of those components in food production. For optimum utilization of cereal grains, knowledge of their structures and compositions is required. The practical implications of kernel structure are numerous. They relate to the various steps of grain production, harvest, storage, marketing, and utilization. Some of the implications are listed in Table 1. Discussed here, in detail, are primarily studies that deal with wheat and barley. Other cereal grains are reviewed briefly.

Kernel Structure - General

The cereal grain is a one-seeded fruit called a caryopsis, in which the fruit coat is adherent to the seed. As the fruit ripens, the pericarp (fruit wall) becomes firmly attached to the wall of the seed proper. The pericarp, seed coats, nucellus, and aleurone cells form the bran. The embryo occupies only a small part of the seed. The bulk of the seed is taken up by the endosperm, which constitutes a food reservoir.

The floral envelopes (modified leaves known as lemma and palea), or chaffy parts, within which the caryopsis develops, persist to maturity in the grass family (MacMasters, 1962). If the chaffy structures envelope the caryopsis so closely that they remain attached to it when the grain is threshed (as in rice and most varieties of oats and barley), the grain is considered to be covered. However, if the caryopsis readily separates from the floral envelopes on threshing,

Table 1. Some Implications of Kernel Structure

Significance in	Parameter	Effect	Commodity
Threshing	Germ damage or skinning	Reduced germinability, impaired storability	All cereal grains
Drying	Cracks, fissures, and breakage; hardening	Reduced commercial value; lowered grade, impaired storability, dust formation, reduced starch yield	Mainly corn and rice
	Discoloration	Reduced commercial value, lowered grade	Mainly rice
Marketing	Breakage	Reduced commercial value in food processing	Mainly corn and rice
General use	High husk: caryopsis ratio or high pericarp: endosperm ratio	Reduced nutritional value--as food or feed	All cereal grains
General use	Kernel shape and dimensions, proportions of tissues in the kernel, distribution of nutrients in the tissues	Yield of food products; nutritional value of cereal (or cereal products) as food or feed	All cereal grains
Malting	Germ damage, skinning, or inadequate husk adherence	Reduced germinability, uneven malting	Mainly barley
Milling	Uneven surface, deep crease or uneven aleurone	Reduced milling yield	Mainly wheat and rice
Milling	Steely texture	Increased power requirements, starch damage, high water absorption, difficulty in air-classification	Wheat and malt milling
Germination-Malting	Starch granule size	Uneven degradation	All cereal grains
Consumption-Nutrition	Distribution and composition of protein	Change in nutritional value	All cereal grains

as with common wheats, rye, hull-less barleys, and the common varieties of corn, the grain is considered to be naked.

The structure of the wheat kernel is shown in Fig. 1. The dorsal side of the wheat grain is rounded, while the ventral side has a deep groove or crease along the entire longitudinal axis. At the apex or small end (stigmatic end) of the grain is a cluster of short, fine hairs known as brush hairs. The pericarp, or dry fruit coat, consists of four layers: the epidermis, hypodermis, cross cells, and tube cells. The remaining tissues of the grain are the inner bran (seed coat and nucellar tissue), endosperm, and embryo (germ). The aleurone layer consists of large rectangular, heavy-walled, cells. Botanically, the aleurone is the outer layer of the endosperm, but as it tends to remain attached to the outer coats during wheat milling, it is considered by millers as the innermost bran layer.

The embryonic axis consists of the plumule and radicle, which are connected by the mesocotyl. The scutellum serves as an organ for food storage. The outer layer of the scutellum, the epithelium, may function either as a secretory or as an absorption organ. In a well-filled wheat kernel, the germ comprises about 2-3% of the kernel, the bran 13-17%, and the starchy endosperm the remainder. The inner bran layer (the aleurone) is high in protein, fat, and minerals, whereas, the outer bran layers (pericarp, seed coats and nucellus) are high in cellulose, hemicelluloses, and minerals. The germ is high in proteins, lipids, sugars (chiefly sucrose), and minerals;

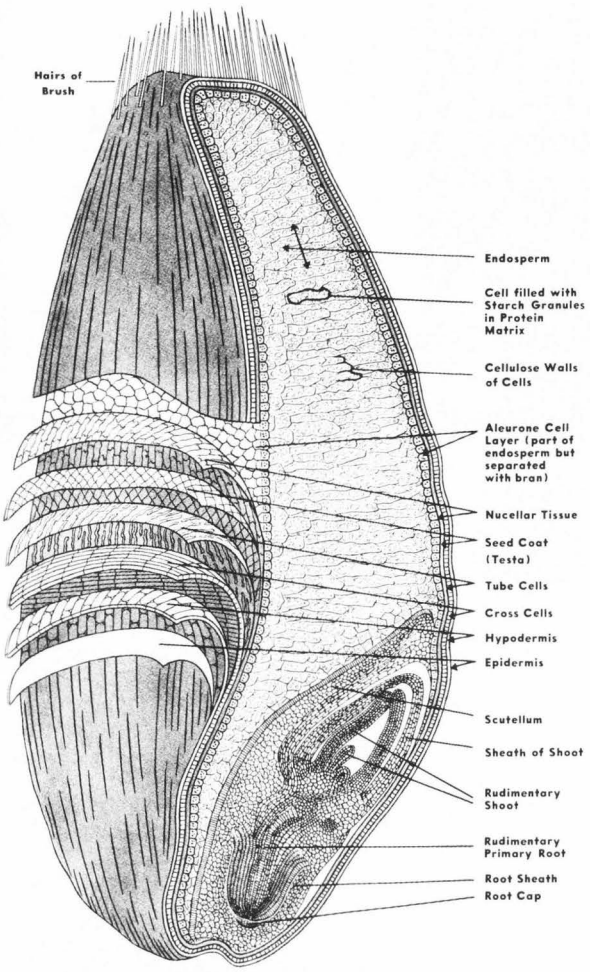

Fig. 1. Longitudinal section of a wheat kernel enlarged approximately 35 times.

the starchy endosperm consists largely of starch grains surrounded by protein.

Grains of other cereals are similar in structure to wheat. The corn grain is the largest of all cereals. The kernel is flattened, wedge-shaped, and broader at the apex than at its attachment to the cob. The aleurone cells contain much protein and oil and also contain the pigments that make certain varieties appear blue, black, or purple. Two types of starchy endosperms-horny and floury-are found beneath the aleurone layer (MacMasters, 1962). The horny endosperm is harder and contains a higher level of protein. In dent corn varieties, the horny endosperm is found on the sides and back of the kernel and bulges in toward the center at the sides. The floury endosperm fills the crown (upper part) of the kernel, extends downward to surround the germ, and shrinks as corn matures.

In a typical dent corn, the pericarp comprises 6%, the germ 11%, and the endosperm 83% of the kernel. Flint corn varieties contain more horny than floury endosperm.

The common varieties of oats have the fruit (caryopsis) enveloped by a hull composed of certain floral envelopes. In light thin oats, hulls may comprise as much as 45% of the grain; in very heavy or plump oats, they may represent only 20%. The hull normally makes up ∿30% of the grain.

Rice is a covered cereal; in the threshed grain (or rough rice), the kernel is enclosed in a tough siliceous hull, which renders it unsuitable for human consumption. When this hull is removed during milling, the kernel (or caryopsis), comprised of the pericarp (outer bran) and the seed proper (inner bran, endosperm, and germ), is known as brown rice or sometimes as unpolished rice. Brown rice is in little demand as a food. Unless stored under very favorable conditions, it tends to become rancid and is more subject to insect infestation than the various forms of milled white rice. When brown rice is subjected to further milling processes, the bran and germ are removed and the purified endosperms are marketed as white rice or polished rice.

Hull and Bran Layers

The structure and adherence of the hull are important in protecting the germinating grain and in the malting process. One reason that barley is uniquely suited for malting is that a cementing layer is present between the hull and the caryopsis. The hull restricts excessive seedling growth without adversely affecting the desirable enzymic degradation of insoluble high molecular weight materials. The adhering hull also protects the seedling from mechanical damage during turning of the malt, and provides a filtration bed during the extraction of soluble malt components in the mashing process.

The hull, as such or as a result of the high concentration of silica, can slow the attack of storage insects on rice and barley. The palea and lemma in barley are held together by two hook-like structures (Fig. 2 a & b). In rice, the ability of these structures to hold the palea and lemma together without gaps is probably variety dependent. Varieties of rice that had many gaps and separations had greater insect infestations than did varieties with tight husks.

Apparently the bran (pericarp, seed coats, nucellus and aleurone) affords little protection against insect infestations, because more insects consistently develop in brown rice than in either rough rice or milled rice. Lack of resistance to infestation is probably due to the thinness of the bran, which allows easy insect penetration, and to the large quantities of lipid and protein present in the aleurone which provide nourishment to the insects.

The outer pericarp layers of wheat (epidermis and hypodermis) have no intercellular spaces and are composed of closely adhering, thick-walled cells. The inner layers of the pericarp, on the other hand, consist of thinner-walled cells and often contain intercellular spaces, through which water can move rapidly and in which molds are commonly found. Similarly, molds can enter through the large intercellular spaces at the base of the kernel where the grain was detached from the plant at harvest and where there

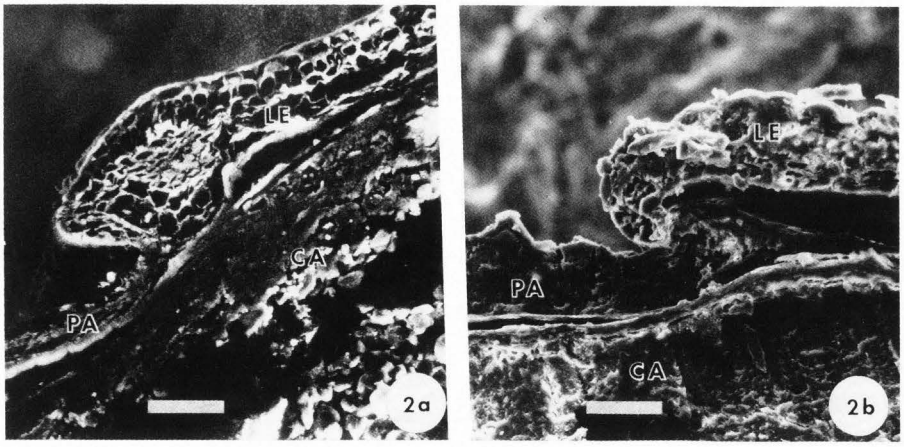

Fig. 2. Scanning Electron micrograph (SEM) transsection through the palea(PA), lemma(LE), and caryopsis(CA) of a) barley and b) rice (100 μm markers).

is no protective epidermis.

An intact grain stores much better than a damaged or ground grain; deteriorative changes (i.e., rancidity, off-flavors, etc.) occur slowly in the whole grain but quite rapidly in ground grain. The hull, apparently, prevents rancidity by protecting the bran layers from mechanical damage during harvesting and subsequent handling.

Once rough rice is dehulled, it rapidly becomes rancid, primarily because of the oxidation of free fatty acids released by the action of lipase. The lipids and lipase are normally compartmentalized in the aleurone and germ cells. Cell disruption may cause mixing of the cellular constituents. Possibly, the dehulling process and subsequent handling of dehulled rice disrupt the aleurone cells and allow the rice to become rancid. The hull, therefore, appears to be necessary to prevent cell disruption during harvesting, storage, and handling.

The Germ

The site of the germ in the kernel and the extent to which the germ is protected by adjacent layers determine whether it will be retained intact during threshing and, thus, the usefulness of the grain for seeding or malting. The ease with which the germ is removed from the caryopsis during milling depends on several factors.

The germ is a separate structure and generally can be easily separated from the rest of the cereal grain. However, the scutellar epithelium (located next to the endosperm) has fingerlike cells, which in wheat are attached to one another for about one-third of their length. The free ends protrude toward the adjacent starchy endosperm cells. The protruding epithelial cells may secrete an amorphous cementing material between germ and endosperm. If some of this material projects into the spaces between the fingerlike cells of the scutellar epithelium and into the folds of the scutellar structure, it may be difficult to separate the germ from the endosperm unless the cementing material is softened. The softening may be accomplished by steeping, as in corn wet milling, or by conditioning, as in wheat milling (MacMasters, 1962). In rice, a layer of crushed cells separating the scutellar epithelium from the starchy endosperm provides a line of easy fracture; hence the germ can be removed intact with minimum effort.

Germ separation is also facilitated by the fact that the germ takes up water faster and swells more readily than the endosperm. The strains resulting from differential swelling contribute to easy separation in milling.

The Subaleurone Layer

The uniqueness of the subaleurone layer in wheat was studied intensively by Kent and coworkers. The subaleurone endosperm in wheat consists of a region of distinctive starchy endosperm cells, one or more layers deep, adjacent to the aleurone cells on the outside and to the inner endosperm on the inside. In hard wheats, the subaleurone layer forms a fairly complete shell around the inner endosperm (except in the regions of the scutellar epithelium and the base of the crease). In soft wheats, the shell is often discontinuous and at the points of discontinuity endosperm cells with typical inner endosperm characteristics extend out to the aleurone layer (Kent, 1966). The two types of endosperm (subaleurone and inner) differ in cell size and shape, size and abundance of starch granules, and proportion of protein. Subaleurone endosperm cells are generally small and cubical; those of the inner endosperm are larger and either needle-shaped (prismatic) of polyhedral (central endosperm).

The ratio of gluten to water soluble proteins is higher in the subaleurone than in the central endosperm layer (Simmonds, 1971). The distribution of starch granules also shows marked differences in different areas of the endosperm. In the subaleurone cells, the starch granules are intermediate in size with relatively few small or secondary granules. In contrast, the mid-endosperm cells are packed with large primary and small secondary granules with the storage protein forming a thin matrix between them.

The differences in structure and composition of various starchy endosperm layers have significant implications in milling. Stock from the first break rolls consists of a coarse fraction, semolina, derived primarily from the center of

the endosperm (Simmonds, 1971, 1972). On further grinding this yields, especially from high protein wheats, a first reduction flour of high starch content. Subsequent milling of the overtailings from the first break removes flour endosperm cells progressively closer to the aleurone layer, and the final break rolls yield a product of high protein content. The reason is the high protein content of the subaleurone layer. Subaleurone and inner endosperm in the coarse (over 35 μm) air-classified fractions of flour milled from hard red winter wheat had average protein contents of about 45 and 11%, respectively. Cells of subaleurone endosperm were reduced to particles below 35 μm size less readily than were cells of inner endosperm. Consequently, subaleurone endosperm cells concentrated in the coarse air-classified fractions and raised the protein content of the latter (Kent, 1966).

Kernel Hardness

Over the years, few subjects have been more controversial and enigmatic than the biochemistry of wheat hardness. The major factors involved in wheat hardness are the intrinsic hardness of the main components (starch and protein), the strength of the interaction with the cell, and the interaction of individual cells to produce the overall grain structure (Stenvert and Kingswood, 1977).

All immature wheat grains are vitreous; as maturation proceeds, some grains remain vitreous, while others become mealy. Endosperm cells in soft wheat contain starch granules embedded in a friable matrix which is readily crushed by the rollers during milling, releasing the starch granules cleanly and with little damage. The endosperm cells of hard wheats, on the other hand, tend to shatter rather than powder, breakage of both starch granules and protein matrix occurring. Thus, the degree of starch damage and the amount of protein matrix carried at the surface of individual kernels is higher in hard than in soft wheats (Simmonds, 1971). According to Kent (1969), proteins are largely responsible for hindering the disruption of the endosperm cell contents during roller milling of hard wheat and for conferring upon the endosperm a soft or hard texture. Proteins govern the manner in which the endosperm shatters during the milling process and the sizes and types of particles that result. Their mechanical properties, together with those of the starch granules, are paramount in the process of fine grinding and of protein shifting in flour by means of air classification. They play, of course, an essential role in breadmaking.

The endosperm of the mature wheat kernel has both plastic and brittle properties, the one or the other being exhibited during grinding according to the moisture content of the endosperm and the method of grinding. When grinding of endosperm is effected by means of a roller mill, a machine in which pressure is applied relatively slowly to the particles, so that compression takes a measurable time, the particles exhibit plasticity, and become less fragmented as moisture content of the material at the moment of grinding is increased; this is so because plasticity increases with moisture content. If the same material is pulverized in a pin mill, in which the particles are fragmented instantaneously, the particles are brittle, and fragmentation increases as moisture content at the moment of grinding is reduced (Kent, 1969). It seems probable that the protein, rather than the endosperm cell walls (which are flimsy and weak) or the starch granules (which remain intact during fragmentation of the endosperm), is responsible for the phenomena of brittleness and plasticity; the protein is thus important as a structural element in the endosperm cell, governing the manner in which the endosperm becomes fragmented during milling (Kent, 1969). Barlow et al. (1973) found no varietal differences in the hardness of storage protein fragments and starch granules. This led Simmonds et al. (1973) to postulate that differences in wheat hardness are due to the presence of a cementing agent between starch and protein. It was suggested that a layer around starch granules was responsible for adhesion and that this was the basis for differences in wheat hardness.

According to Stenvert and Kingswood (1977), however, it is not necessary to invoke an adhesion theory for wheat hardness even if the protein fragments and starch granules from different varieties are of equal hardness. If the protein matrix, as a whole, is not continuous a considerably weakened endosperm structure would result. The extent to which the endosperm structure is ordered could determine hardness. This would depend primarily on the state of the protein matrix which functions as the connecting matter within mature endosperm cells. A continuous protein matrix physically entrapping the starch granules would make difficult separating the starch granules from the protein matrix--as is characteristic in hard wheats. A discontinuous matrix structure would allow the ready release of starch granules as found with soft wheats.

Stenvert and Kingswood (1977) studied the influence of a range of factors on wheat hardness with particular reference to the physical structure of the endosperm protein matrix. Differences in hardness involved the continuity of the protein matrix and the strength with which it physically entrapped starch granules. The primary determinant of wheat hardness is genetically controlled and relates to factors that influence compactness of endosperm cell components.

Relation between physical structure and some chemical constituents and milling performance of bread and feed wheats were studied by Nierle and Elbaya (1978). Vitreous kernels of varieties Jubilar and Caribo were higher in protein content, sedimentation value, and wet gluten content than mealy grains. The ratio of soluble protein content to the total protein content and of gliadin to glutenin in vitreous kernels was lower than in mealy grains. Electrophoretic and gel chromatographic studies suggested higher hydrogen bonding forces between protein molecules of vitreous than of mealy kernels. Non starch lipids were more extractable in mealy kernels, presumably, because of the less compact nature of the protein matrix. Mealy grains produced higher amounts of break flour with lower mineral content than vitreous kernels. It is questionable, however, whether the reported differences are related to kernel

structure or to associated variations in protein contents and distribution.

Cellular Structure and Endosperm Breakage

Endosperm structure and hardness is related to breakage in milling and to the structure and composition of milled flour particles. Microscopy has demonstrated fundamental differences between the cellular structure of soft and hard wheats and flour particles obtained from such wheats (Greer and Hinton, 1950; Greer et al., 1951; Kent and Jones, 1952). Greer et al. (1951) suggested that whereas flour particles of soft wheats consist of broken cells, those of hard wheats consist of entire cells or groups of cells, each including its individual cell wall. Kent and Jones (1952) presented a method of characterizing a flour in terms of its cellular structure. The proportions of flour particles were classifed according to 1) the part of the wheat endosperm from which they are derived (peripheral, prismatic, or central), 2) the number of cell units comprising the particle, 3) the relative intactness of the cells(s), and the relative extent of cell wall covering, depending on the type of wheat (hard or soft), part of the milling system (break or reduction), and conditioning.

Schultze and MacMasters (1962) showed that endosperm breakage occurs across cell walls, not between the walls of adjacent cells. The cell-wall particles consistently were portions of walls of two adjacent cells, with the middle lamella which cements them. Breakage of the cell walls was invariably transverse, rather than along the middle lamella.

Moss et al. (1980) examined six wheat cultivars and their mill brans with the SEM. Endosperm removal (flour) from bran was related to the cleavage pattern of the grains. Good bran clean-up and high flour yield were associated with inter-cellular cleavage. When fracture took place through the contents of the endosperm cells more endosperm adhered to the bran. The hardness of the endosperm cells determined the nature of the cleavage pattern. In a hard wheat, a continuous protein matrix around all the cell contents resulted in the boundary between the cell wall and cell contents becoming a zone of weakness. Hence cleavage was intercellular. The cell contents could then act as a single entity and the whole cell could be removed from the bran by shear forces imparted during milling. Some forces were also redirected towards the bran, fragmenting it into small pieces. In a soft wheat, air spaces and discontinuities in the protein matrix made the cell fragile and the shear forces were not redirected, but passed through the cell rather than removing it cleanly from the bran. Increasing the water content of the wheat prior to milling favored intra-cellular cleavage. Thus, it was concluded, that the relationship between bran clean-up and bran fracturing can be optimized by balancing intrinsic hardness and grain moisture content; this is the basis of wheat conditioning.

Wheat Conditioning

Water absorption during wheat conditioning largely governs its milling behavior (Bradbury, et al., 1960). Moss (1973) studied varietal differences in grain morphology as they affect water penetration into the wheat, overall conditioning, and the milling process. It has been suggested that varietal differences are affected by thickness and composition of the outer cuticle and testa, the extent to which the outer epidermal and inner parenchymal cells have been compressed, and the number and size of protein masses in the subaleurone endosperm cells (Hinton, 1955).

According to Drews (1979), an important factor in conditioning and separation of the starchy endosperm is the amount of water absorbing and swelling components. The amount is highest in bran, lower in the aleurone, and lowest in the starchy endosperm. Attack of grain by microorganisms may degrade the cellulosic and semicellulosic materials in the outer layers and reduce their water binding and swelling capacities. Sprouting can have profound effects on degradation of those materials and significantly affect their milling properties. The presence of relatively large amounts of pentosans in the starchy endosperm of rye and triticale may significantly influence their milling properties.

According to Meuser and Klingler (1979), starch is the main wheat component that can be relatively easily modified in its physicochemical properties by milling. This is accomplished by varying the degree of starch damage and results in changes in water binding capacity and susceptibility to α-amylase degradation. Such damage is small in soft wheat because conventional milling separates easily the starch granules from the protein matrix. An increase in water binding capacity that accompanies increased severity of milling soft wheats, is due to an increase in the number of particles rather than to an increase in starch damage (Meuser and Klingler, 1979). Those findings are especially relevant in light of the studies by Evers and Lindley (1977) on particle size distribution of starch starch in the endosperm of 12 wheats. Granules below 10 μm diameter accounted for about 1/3 to 1/2 of the total weight or endosperm starch. As the samples were representative of wheats used in baking, it was suggested that small granules may be of great importance in determining flour and dough properties.

In a study of the size distribution of starch granules in endosperm of different sized kernels of the wheat cultivar Maris Huntsman, the number of starch granules was greater in large-plump than in small-plump or shrivelled kernels (Brocklehurst and Evers, 1977). In all three kernel types more than 1/3 of the total starch weight was contributed by granules less than 10 μm diameter (B type granules). The proportion of small granules was significantly greater in large-plump kernels than in the other two types.

Studies by Butcher and Stenvert (1973) determined by an autoradiographic technique that the rate of water penetration into the kernel differed among Australian wheat cultivars. Moss (1977) used an improved technique, applicable directly to whole sections. This enabled a more precise location of the conditioning water to be made than could be previously achieved using halves of labeled wheat grains. Within 1 hr the labeled water penetrated into the aleurone cells and in

many cases into the starchy endosperm to a depth of 50 to 60 μm. The embryo and scutellum also absorbed the water with great rapidity. Subsequent penetration into the starchy endosperm was delayed for several hours. The cells of the embryo and scutellum appeared to bind the water more strongly than the aleurone and after 48 hr rest time were still more heavily labeled than the other components of the wheat grain (Moss, 1977).

In a recent publication, based on the above findings, the relation between optimum conditioning, moisture level, and grain hardness was reported (Anon., 1977) (Table 2).

Table 2. Relation Between Wheat Type, Grain Hardness and Optimum Conditioning Moisture

Wheat Type	Grain Hardness (PSI)	Optimum Conditioning Moisture (%)
V. Hard	9-13	16.5-17.5
Hard	14-19	15.5-16.5
Intermediate	20-25	14.5-15.5
Soft	25-30	13.5-14.5

Factors which affect the rest time after sampling and before milling include: (a) initial moisture content, (b) protein content, and (c) grain hardness. An increase in moisture content from 8 to 12% can reduce rest time three-fold due to a reduction in the water binding potential of grain components at higher initial moisture contents, thus allowing a more rapid movement of water. An increase in wheat protein content decreases the rate of water penetration. This is due to protein retarding the moisture movement because of its water binding capacity and because it contributes to a more ordered endosperm structure. The latter relates to the role of grain hardness in slowing moisture movement (Anon., 1977).

Milling Score-Flour Yield

Milling quality is governed by morphological characteristics of the wheat kernel and its mechano-physical properties and by the methods of grinding and separation (Meuser and Klingler, 1979). The size and shape of the crease affect yield and composition of flour because the bran in the crease area is difficult to separate from the starchy endosperm (Fig. 3 a & b). The volume occupied by the crease has been calculated to range from 0.7 to 1.9% of the total grain volume and its relative size affects the milling process in terms of ability to extract flour and its potential for providing a hospice for fungal growth at grain maturity. Some interest is being shown in breeding grain without a crease (Kingswood, 1975). The flour yield from such grain would be greatly increased. Kosina (1979) determined, in a study of several European wheats, gross morphological differences in kernel structure that are likely to affect flour yield. The coefficients of variation were largest for the height of the endosperm cavity and lowest for the thickness of the aleurone layer (Table 3).

Table 3. Coefficients of Variation of Gross Morphological Parameters of the Wheat Kernel

Morphological Parameter	Coefficient of Variation
Endosperm cavity, height	314
Endosperm cavity appendices, thickness	44
Subaleurone layer, thickness	33
Crease width	27
Crease depth	18
Aleurone layer, thickness	12

It would, thus, seem that in breeding types of wheat that produce the highest yield in terms of wheat flour, changing the aleurone layer thickness is not a highly promising approach.

According to MacMasters (1962), the milling engineer is troubled by the constant minor variations in the thickness of the aleurone layer. The irregular thickness of the aleurone layer makes it difficult to scrape all of the starchy endosperm from it, in milling. That the starchy endosperm and the aleurone layer are part of one tissue, rather than separate, merely adherent

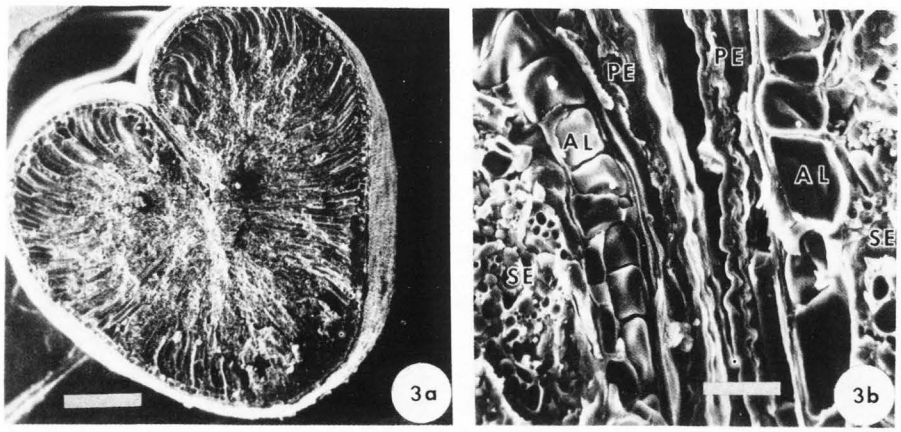

Fig. 3. SEM micrograph of a) cross section through the wheat kernel (500 μm marker) and b) through the crease area in a rye kernel, pericarp (PE), aleurone (AL), and starchy endosperm (SE) (50 μm marker).

tissues, complicates the problem (MacMasters, 1962).

Seeborg and Barmore (1952) found that differences in flour yields were attributable mainly to differences in amounts of residue remaining after separation of germ and fine bran from the shorts. This residue (irreducible endosperm) seems to correspond to the "mill finish" or reduction tails which accumulate abnormally in wheats that are difficult to mill. Seeborg and Barmore suggested that the increased yield of that material from wheats of poor milling quality corresponded to a greater amount of endosperm cell walls in such wheats.

Marked expansion in recent years of the area planted to high-yielding wheat varieties not suitable for breadmaking (i.e. Maris-Huntsman in the United Kingdom and Clement in the Netherlands) required development of a simple and quick testing procedure to distinguish between breadmaking and feed wheats (Bolling and Meyer, 1975). The authors emphasized the need to distinguish clearly between inferior milling and inferior breadmaking properties. Milling and breadmaking properties may be related, but not necessarily.

Histochemical studies of Bolling and Meyer (1975) have shown that in acceptable cultivars, a fairly strong adhesion between the aleurone-pericarp layers facilitated their separation from the starchy endosperm. In inferior wheats, the subaleurone layer is rich in protein, adheres strongly to the aleurone layer, and cannot be separated efficiently. In some inferior cultivars, the porous structure of the pericarp facilitates shattering and increases the bran content of flour (Bolling and Meyer, 1975).

Meuser and Reimer (1977) studied the possibility of influencing milling and, indirectly, baking properties. The authors investigated the effects of particle size and structure of flour particles and their composition in a wheat milled under various conditions. This was followed by determining the influence of raw material quality on the structure of flour particles from various hard wheats and the possibility of modifying processing characteristics of flours from a single wheat by altering milling parameters.

Wheat hardness is a varietal characteristic that governs particle size of milled products (Meuser and Klingler, 1979). Generally, soft wheats produce "smooth" and hard wheats "gritty" flours. The inherent capacity of hard wheat to produce semolina or farina may be important. The range of particle sizes in milling hard wheats is wider than in milling soft wheats. The former also are more granular than the latter. The strong adherence between protein and starch in hard wheats results also in greater homogeneity in composition of the milled particles than in soft wheat flours. The extensive mechanical degradation of soft wheats in milling increases their water binding capacity to such a large extent, that their breadmaking properties may be impaired. Consequently, it may be desirable to conduct the milling process in such a manner that most particles are in the 40 to 90 μm range. This modification of the milling process and resultant changes in particle size distribution could not counteract however, the inferior performance of feed wheats (Meuser and Klingler, 1979).

Yamazaki and Donelson (1972) found a high negative correlation for white layer cake volume vs. mass-median diameter of laboratory-milled cake flours obtained from pure-variety wheats. Cake volume was also associated inversely with mass median diameters of straight-grade and coarsely milled flours and directly with the quantity of sifted meal from wheats milled to obtain patent flours for cake baking. Varietal differences in cake potential for those wheats appeared to be associated largely with inherent differences in endosperm friability.

Predicting Milling Behavior

The problem of predicting the milling behavior of wheat by the use of simple chemical laboratory tests has occupied cereal chemists for many years.

The endosperm cell walls of seven Pacific Northwest (PNW) wheat cultivars ranging from excellent to poor in milling quality, were studied by Wolf et al. (1952) to determine differences in content and composition of water-insoluble hemicelluloses. In all varieties, degradation of endosperm cell walls in transections treated with 1% H_2SO_4 or 1% KOH was greatest near the aleurone and decreased toward the crease. The degradation was greatest in the first two or three cell layers just beneath the aleurone layer. The cell walls were degraded over a greater area in varieties of excellent milling quality than in those of poor milling quality, but varieties of intermediate milling quality behaved erratically (Wolf et al., 1952).

Larkin et al. (1952) found with PNW white wheats that the thickness of cell walls in the endosperm near the aleurone layer was least in the wheats of best milling quality. Differences between varieties, however, were smaller than differences between different parts of the grain. The relationship was independent of year and location (MacMasters et al., 1957). Similar results were reported for soft wheats by Popham et al. (1961) but in that case the year of growth had a significant effect on milling score. In a later study, the correlation coefficient between the pentosan value and milling score was -0.74 and -0.84 for selected winter and spring wheat, respectively. The correlation coefficients between pentosan and flour yield, bran weight, and bran cleanup were significant at the 1% level (Weswig et al., 1963).

Medcalf et al. (1968) reported that durum pentosans contained a higher proportion of arabinose, and thus more branching, than hard red spring (HRS) pentosans. Similarly, durum pentosans were somewhat higher in molecular weight than HRS pentosans. The differences were significant but relatively small. It was postulated, however, that a small difference in the degree of branching might alter markedly the degree and type of interaction of polysaccharides with proteins. Differences in molecular weight also could alter the interaction mechanism and water absorption. These differences could account, in part, for the differences in endosperm properties between durum and HRS wheats.

Mares and Stone (1973a) showed that wheat endosperm walls had an ultrastructure similar to

primary cell walls, having a microfibrillar phase embedded in an amorphous matrix. Chemical studies showed that the walls were largely composed of polysaccharide and that some protein was also present. The predominant polysaccharides are arabinoxylans of which one-third are soluble in water, the remainder requiring alkaline reagents for solubilization. The authors isolated, fractionated, and characterized wheat endosperm cell walls, free from non-endospermic cell walls, in flours from three wheats. The isolated cell walls were similar in proportions of the polymeric components and the monosaccharide composition of the walls and the wall fractions. The appearance of endosperm cell walls in situ and in wall isolates was examined by light microscopy, scanning and transmission electron microscopy. SEM showed apparent moulding of the walls on the cell contents and different fracture patterns of prismatic and central cells. The cell walls have a microfibrillar skeleton embedded in the amorphous matrix components.

In a subsequent study (Mares and Stone, 1973b), it was found that the endosperm cell walls are composed mainly of arabinoxylan and of some cellulose, glucomannan and protein. The postulated presence of cellulose is of particular interest. Stenvert and Moss (1974) applied a detergent extraction technique to quantitatively separate the outer layers of the wheat grain from the starchy endosperm. Milling studies demonstrated that the extraction technique allowed an accurate estimate of the flour yielding potential provided that the milling system was capable of fully realizing the flour milling capacity. This is much easier said than done. In certain instances the true flour yielding potential of a wheat could not be judged by the detergent method due to differences in optimum conditioning requirements, ease of separation of bran from the endosperm, dressing properties and sensitivity to moisture level. Grain hardness and test weight of hard and soft wheats were not related significantly to flour yield.

Those studies showed similarities in the chemistry of the cell walls of wheats of diverse milling quality and seem to suggest that neither variations in proportions of the component polysaccharide fractions nor in their composition are responsible for meaningful differences in milling properties. The possibility to develop a simple chemical test that relates cell wall material to varietal differences in milling cannot be excluded. It is not likely, however, that such a single test could account for differences in cell wall thickness, content, distribution, composition, and many other factors which govern milling quality. And, last but not least, we have no standardized or optimized acceptable milling test that could be used as a reference basis.

Grain Drying

Changes in texture and structure during drying of corn and rice are important in minimizing breakage during handling. Excessive cracks reduce value of corn for producing foods such as breakfast cereals. Harsh heat treatment during grain drying may reduce starch yields, impair quality of the starch, and create difficulties in corn wet-milling. If used for alcoholic beverages, overheated corn may also cause difficulties in beer brewing and in distillation. The starch granules are embedded in a proteinaceous matrix that hardens during overheating. The hardened matrix protects the starch from enzyme attack and conversion to alcohol. Broken corn is more easily attacked by insects and produces more grain dust than whole grain and creates many problems in handling, transportation, and storage. In rice milling, harsh drying and accompanying structural cracks substantially reduce yields of head rice and increase amounts of "brokens," and thus cause economic losses to the miller. The method of rice drying may also affect the texture and color of the milled rice and result in off-color, especially objectionable browning.

Nutritional Implications

The chemical composition of different cereal grains varies widely, since it is influenced by genetic, soil, cultural and climatic factors. Amounts of proteins, lipids, carbohydrates, pigments, vitamins and total ash vary; mineral elements present also vary widely. Cereals are characterized by relatively low protein and high carbohydrate contents; the carbohydrates consist essentially of starch (90% or more), pentosans, and sugars.

The various components are not uniformly distributed in the kernel. The hulls and bran are high in cellulose, pentosans and ash; the germ is high in lipid and rich in proteins, sugars, and generally, ash. The endosperm, which contains the starch, has a lower protein content than the germ and the bran (in some cereals), and is low in fat and ash.

Furthermore, the various proteins are not distributed uniformly in the kernel. Thus, the proteins fractionated from the inner endosperm of wheat consist chiefly of approximately equal amounts of prolamins (gliadins) and glutelins (glutenins). The embryo proteins consist of nucleoproteins, albumin (leucosin), globulins, and proteoses; in wheat bran prolamins predominate with smaller quantities of albumins and globulins.

Breeding efforts to improve the nutritive value of cereal grains have concentrated on increasing protein content without decreasing protein quality (mainly retaining lysine concentration in the protein). The significance of protein distribution in the endosperm depends on the type of product that is likely to be consumed. In the production of highly refined milled products, in which some of the subaleurone layer is removed, a high concentration of protein in the subaleurone layer would not be desirable. However, if the whole kernel is to be consumed, distribution of protein in the kernel is of limited nutritional consequence.

In all cereal grains, the storage protein forms a matrix which surrounds the starch granules. Protein body initiation and formation of the matrix protein were studied recently by Bechtel et al. (1982a, b). Those studies suggested a role for the Golgi apparatus in the

initiation of protein bodies. The Golgi apparatus in wheat may function as a concentration organelle to establish foci for accumulation of proteins. Protein bodies that formed in the cytoplasm were transported to the central vacuoles where the protein body membrane and tonoplast fused and deposited the protein granules into the vacuole. Protein granules in the vacuole enlarged by three mechanisms: 1. addition of membranous vesicular material of various types, 2. addition of flocculent material, and 3. fusion of granules with other newly deposited protein granules. Fig. 4 shows the relationships among kernel dry weight, water content and protein per kernel. The dry weight per kernel increased consistently during development whereas the moisture content remained relatively constant during the first 12 days and then dropped rapidly. Protein content per kernel increased consistently during caryopsis development and closely paralleled the dry weight data.

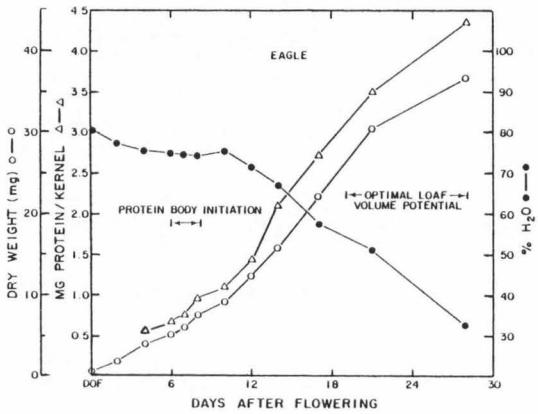

Fig. 4. Graph of dry weight, moisture content and protein content of hard red winter wheat Eagle during caryopsis development.

Fusion of protein granules to form a large protein mass is illustrated in Fig. 5 and Fig. 6. This method of protein enlargement increased noticeably during 14 to 17 days after flowering and peaked at 21 to 28 days after flowering. It is of interest that wheats harvested at about 21 to 28 days after flowering (or about 10 preripe) have maximum loaf volume potentialities. There is good indirect evidence that up to a certain level, increased protein body fusion is concomitant with increases in loaf volume potential.

The concentration of protein increases from the inner to the outer starchy endosperm. The increase may be relatively gradual, as in some soft wheats (Kent 1966), or quite steep, as in some high-protein wheat types in which some of the outer subaleurone cells contain few, if any, starch granules. Since the subaleurone region in rice is only several layers thick and lies directly below the aleurone, the subaleurone layer can be easily removed during milling. It is, therefore, desirable either to mill rice as lightly as possible for a consumer acceptable product, or to breed cultivars with an increased subaleurone layer, or cultivars with a more even distribution of protein throughout the endosperm.

Sullins and Rooney (1974) used SEM to illustrate differences in corn endosperm structure that account for differences in nutritive value of the grain. High-lysine corn has a reduced amount of protein bodies in the endosperm. SEM of soft endosperms for normal, *opaque-2* or modified *opaque-2* corn showed loosely packed, nearly round starch granules associated with thin sheets of protein and many intergranular air spaces (Robutti *et al.*, 1974). The hard endosperms had tightly packed, polygonal starch granules associated with a continuous protein matrix, and no intergranular air spaces. Normal hard endosperms had zein bodies embedded in the protein matrix; modified hard endosperms did not. Starch damage was greater in the hard endosperm than in the soft

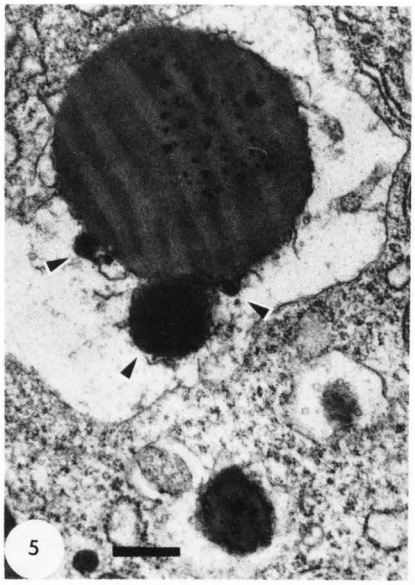

Fig. 5. Fusion of three protein granules (arrows) into a larger granule in wheat 10 days after fertilization (10.5 μm marker).

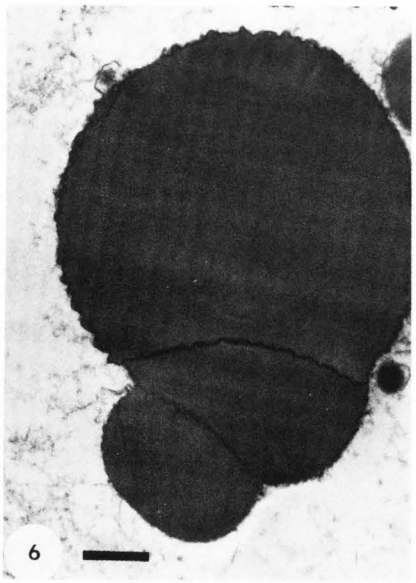

Fig. 6. Large protein granule in wheat endosperm vacuole 10 days after fertilization. Note dense line between fused granules (1 μm marker).

because of a stronger adhesion between starch and protein. The low density and opaqueness of soft endosperm were attributed to the intergranular air spaces. Interaction between protein matrix and starch granules during drying explains the shape of starch granules.

Seckinger and Wolf (1973) studied the structure of grain sorghum endosperm protein of commercial hybrids and experimental lines with the transmission electron microscope (TEM) and SEM. Vitreous endosperm showed a well developed, two-component structure consisting of concentric-ringed protein bodies (2-3 μm in diameter) embedded in an amorphous matrix protein. On the basis of solubility properties of the proteins, they suggested that the protein bodies were the site of prolamin (kafferin) deposition and that the matrix protein was the site of glutelin deposition. Distribution of protein within the sorghum grain was similar to that within other cereal grains in that the peripheral vitreous area of the kernel had the highest protein content. Interior areas had gradually decreasing amounts of protein. Protein bodies accounted for 70-80% of the sorghum protein, as determined by microscopic observations.

The structure of grain sorghum samples representing a wide genetic base was examined by SEM (Hoseney et al., 1973). The soft or opaque endosperm was characterized by relatively large intergranular air spaces. The starch granules were essentially round and covered with a thin sheet of protein. Embedded in the protein sheet were relatively large spherical protein bodies. The hard or translucent endosperm portion was characterized by a tightly packed structure with no air spaces. The starch granules were polygonal and covered with a thick protein matrix. Embedded in the protein matrix were protein bodies.

Sullins and Rooney (1973) conducted light microscopy (LM) and SEM studies of the peripheral endosperm of waxy and nonwaxy endosperm sorghum varieties. Sorghum varieties are known to differ widely in endosperm type (i.e., yellow, sugary, waxy, and nonwaxy). In feeding trials sorghum grains with waxy endosperm tended to have higher feed efficiencies than nonwaxy varieties. Sullins and Rooney found that the subaleurone endosperm area of sorghum was composed of starch granules embedded in an amorphous protein matrix that contained relatively indigestible (alcohol-soluble) protein bodies. The waxy sorghum varieties contained fewer spherical protein bodies and were, therfore, more digestible than the nonwaxy varieties. Because of its low relative proportion of protein bodies, waxy grain, apparently, is more completely broken down than is nonwaxy during processing (i.e., steam-flaking, micronizing, pulverizing, popping, exploding, and reconstituting). The protein differences also may contribute to the difference in feed efficiency between waxy and nonwaxy sorghum grains.

Germination and Malting

In malting barley for brewing, the grain is modified into a product that can yield an aqueous extract containing: (a) fermentable products, (b) available substrate for yeast nutrition, and (c) precursors for imparting desirable organoleptic qualities to beer.

The sum total of physical and chemical changes taking place during malting is termed "modification." According to MacLeod (1967), "modification" describes "a rather nebulous but nonetheless real condition which has resulted from the transformation of endospermic constituents to give the best possible material for mashing." In practice, the modification conditions of malting are selected so that yield of extractable solids is maximum, and malting losses and excessive degradation of the high-molecular-weight components of the barley are minimal. Modification transforms tough barley into friable malt. The transformation can be assessed by physical methods ranging from the simple biting test to tests involving elaborate self-recording mechanical devices. Among chemical indices, the increase in soluble proteins is probably the most important single parameter.

The pattern of endosperm modification of malted grain is critical to the science and technology of malt production. Palmer's studies (1971) indicate that the modification of the endosperm in germinated barley commences at the dorsal (nonfurrowed) surface of the grain. Palmer found that the rate of endosperm modification depended more on the effective dispersal of hydrolytic enzymes than on the total amounts of these enzymes in the grain. Microscopical analyses showed that starch granules and hemicellulosic materials of the cell walls were coated with proteinaceous materials. Proteases play a more active role than carbohydrases in the conversion of hard barley into friable malt.

Changes in the aleurone layer and in the starchy endosperm of steeped, malted, and kilned barley were examined by SEM (Pomeranz, 1972). The surfaces of aleurone cells in steeped barley were highly pitted. The walls of aleurone cells were progressively degraded during malting and kilning. Aleurone grains increased in diameter during steeping and were further distorted during kilning. Partial breakdown of cell walls in the center of the starchy endosperm of malted barley was accompanied by extensive dissolution of the protein matrix and the "freeing" of small starch granules that previously were embedded in that matrix; the effect on the appearance of the starch granules was small. In the central endosperm of kilned barley malt, the cell-wall dissolution was extensive and was accompanied by mechanical breakdown of the large starch granules. A study on modification in a kilned malt was completed recently in our laboratories (Fretzdorff et al., 1982) using a combination of histochemistry, light microscopy, and transmission and scanning electron microscopy. Hydrolysis of cell walls, proteins, and starch was most extensive in the starchy endosperm area adjacent to the scutellar epithelium (Figs. 7 to 10). Some hydrolysis occurred in areas adjacent to the aleurone layers; hydrolysis decreased as distance increased from the embryo end to the distal end and from the aleurone layer to the center of the starchy endosperm (Fig. 11). While no rigid sequence of hydrolysis was observed, generally, cell wall hydrolysis was more extensive than protein

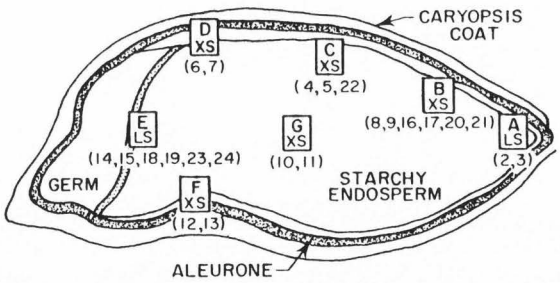

Fig. 7. Diagramatic scheme of barley or malt showing regions where various sections (X's = cross section, L's = longitudinal section) were taken.

hydrolysis, and starch hydrolysis seemed to take place gradually in the late stages of malting and kilning. Small starch granules were hydrolyzed more extensively than large granules.

The SEM was used to follow the modification in malting of a low-protein barley and a high-protein cultivar (Pomeranz, 1974). In the low-protein cultivar, the protein matrix degraded extensively, and some of the degraded protein was deposited in the kilned malt on large starch granules. In the high-protein cultivar, much of the protein matrix was intact and some protein was retained in the form of a modified but coherent and continuous thick film covering the starch granules. It was suggested that the thick film is responsible for difficulties in malting high-protein barleys, for reduction of wort extract, and for persistence of undegraded proteins, which enhance chill haze formation in beer.

Palmer (1974) suggested on the basis of SEM that during malting hydrolytic enzymes migrate into the endosperm to disrupt and solubilize mainly the cell walls, complex protein materials, and the small starch granules. Satisfactory modification in malting should result in degradation of cell-wall material throughout the endosperm and release of starch and degraded protein during mashing. However, some areas of the endosperm (especially at the distal end) may contain undegraded endosperm cell walls in which starch extract can be trapped, and the trapped starch gives rise to glucan (gum) materials during mashing.

Microbial Damage

Cereal grains are important as food because of their excellent keeping qualities. Moisture content is the major factor in determining the storage behavior of grain, which is also influenced by temperature, oxygen supply, history and condition of the grain, length of storage, and biological factors (molds and insects). The respiratory rate of dry grain is low. As the moisture content is raised above 14%, the apparent respiration increases gradually until a certain critical moisture is reached above which respiration accelerates rapidly and the grain tends to heat. This sharp increase in respiration is due to the germination and growth of certain molds

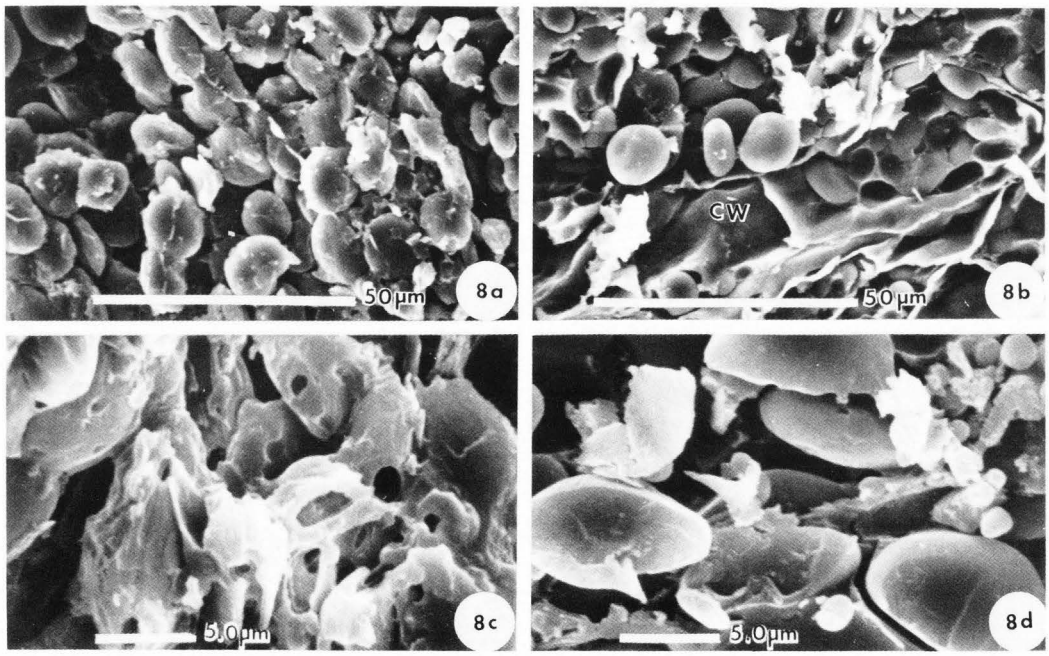

Fig. 8. SEM. a) Almost complete hydrolysis of cell wall and storage protein and extensive digestion of starch from region F of malt. b) Barley section from region F showing intact cell walls (CW), protein, and starch. c) High magnification of highly modified starch from region E of malt. d) Unhydrolyzed starch granules of barley from region E cut in half by razor blade during tissue preparation.

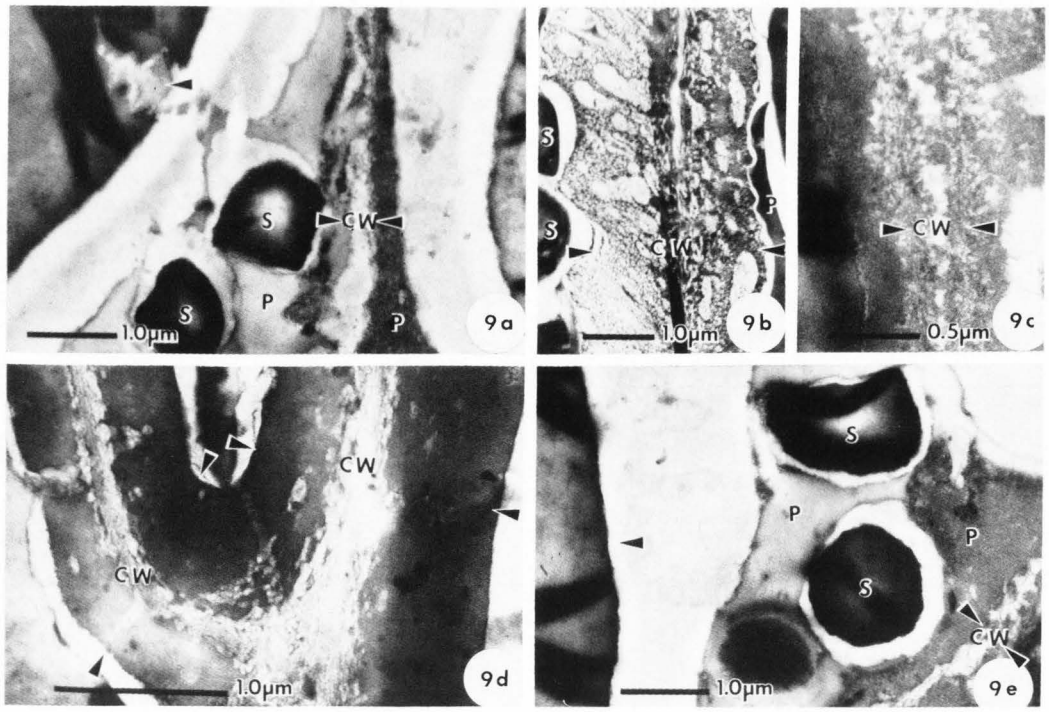

Fig. 9. TEM. a) Malt from region B showing most of cell wall (CW) digested, intact protein (P), partially digested small starch granules (S), and wedge-shaped digestion furrow in large starch granule (arrow). b) Intact cell wall (CW), starch (S), and protein (P) from barley in region B. c) Highly digested cell wall (CW) near scutellum (region E) of malt. d) Intact cell walls (CW) of barley from region E. e) Starch digestion (small starch granules (S); large starch granules, (arrow) of malt from region B was minimal; as was protein (P) degradation. Cell walls (CW) were mostly removed.

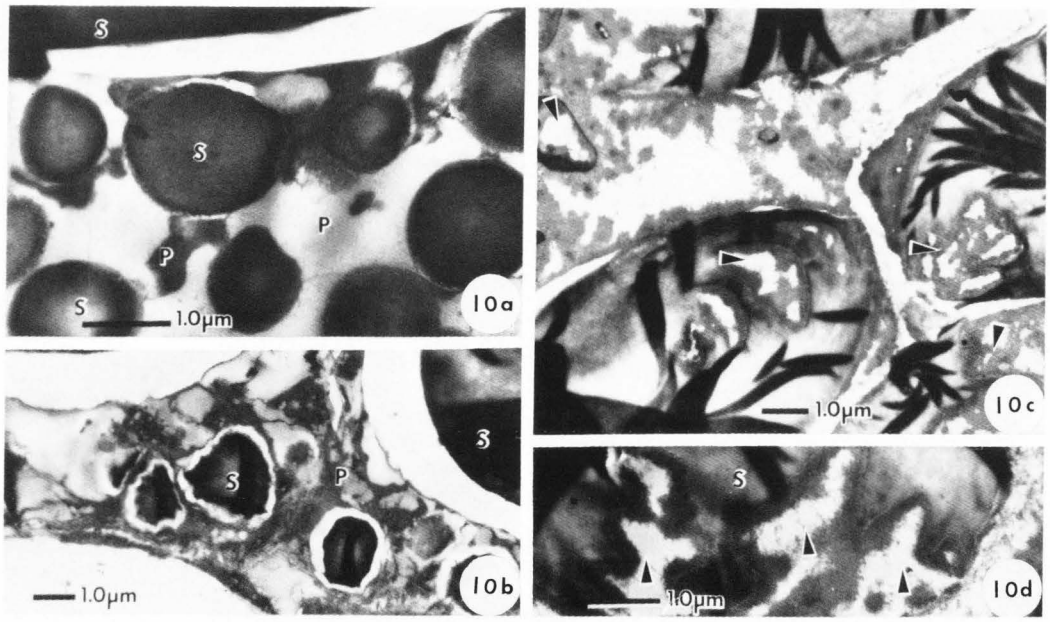

Fig. 10. TEM. a) Intact protein (P) and starch (S) of barley from region B, representative of those components in various regions. b) Malt from region C showing peripheral digestion of starch (S) and some modification of protein (P). c) Digestion of starch from region E of malt (arrows). d) High magnification of digestion furrows (arrows) of starch granule (S) of malt from region E.

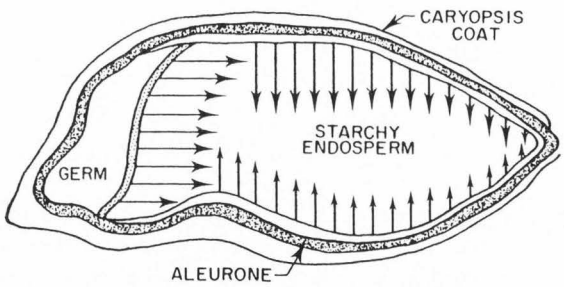

Fig. 11. Diagramatic summary of hydrolytic modification of cell walls, proteins, and starch in kilned barley malt.

(predominantly various species of *Aspergillus* and *Penicillium*) commonly found in soil and in previously used storage bins. Molds are invariably found on the grain and within the seed coats, even though the grain is harvested under ideal conditions.

Several investigations have indicated that the crease and palea are the primary site of infestation by microorganisms in oats (Pomeranz and Sachs, 1972). The area between the palea and the crease seems to be favorable for growth of microorganisms, where they are harbored in the mature and dried grain. Presence of a plaque with microbial growth (probably a slime-producing bacterial colony) beneath the palea of an oat kernel is indicated in Fig. 12a. Fungi under the hull of rice are shown in Fig. 12b.

Instrumentation and Methodology

Much of the progress on relation between structure and end-use properties of foods has been made possible through advances in instrumentation and methodology (Pomeranz, 1976).

Scientists engaged in the study of materials have a large, increasing, and sometimes bewildering number of analytical techniques at their disposal (Williams and Goldstein, 1981). Significant among these are techniques that utilize electron beams as a source of information. Some of these techniques are compared in Table 4; many are highly complimentary. New methods eliminate (or at least reduce) artifacts from specimen desiccation and damage from excessive irradiation of biological systems. This has been accomplished along with increased sensitivity. Another biological application involves labeling with heavy atoms coupled to a particular chemical group of interest within a macromolecule or to a particular macromolecule in an assembly of macromolecules (Beer et al., 1981). Chemists who study solids recognize the complicated relationships between microstructure and properties of solids. High-resolution analytical electron microscopy is especially useful in studying those relationships.

Photoacoustic spectroscopy has found for quite a number of years important applications in characterization of solids, liquids, and gases from inorganic and biological sources. Recently, there has been interest in performing photoacoustics on a microscopic scale (Rosenwaig, 1979). Tsai et al. (1979) described the application of transmission scanning acoustic microscopy in nondestructive testing and evaluation. The instrument, used in the metal industry and in production of microelectronic computers, measures material parameters such as elastic modulus, mass density, and acoustic absorption rather than the index of refraction and optical absorption as observed in an optical microscope. The instrument does not require destructive sample preparation and unlike conventional optical microscopy, can illuminate and examine details deep below the surface of an opaque sample. Williams and Goldstein (1981) emphasized that while electron microscopy continues to develop and more advanced and higher resolution instruments become available, one of the regions of greatest promise is interfacing of computers to electron optical instruments. The exciting developments in this field will change the electron microscope, now predominantly an imaging instrument, into a data-generating instrument with concurrent imaging capabilities. For example, on-line microanalysis and particle size analysis can be performed on an SEM and AEM. Concurrent diffraction pattern analysis can also be performed with minimal operator intervention. Minicomputers can be used to control operating parameters, collect data, and assist in data interpretation (Stewart, 1981).

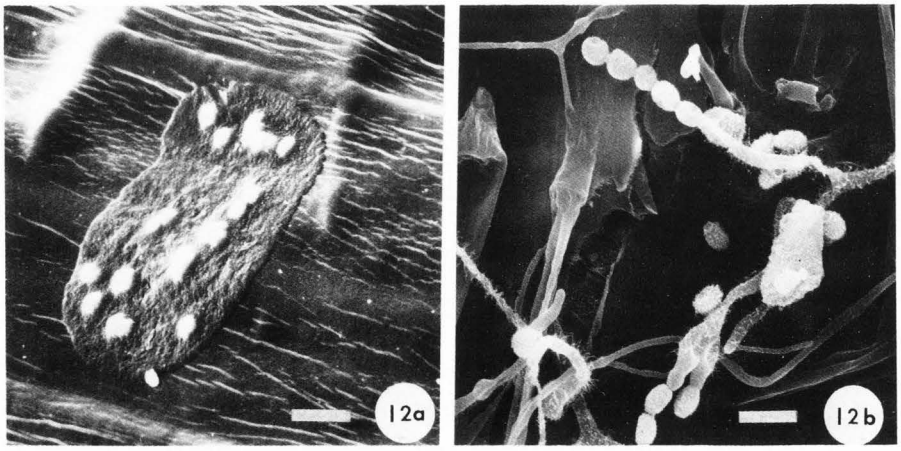

Fig. 12. a) Plaque with microbial growth on the inside of the palea of an oat kernel (20 μm marker); b) Fungi under the hull of rice (5 μm marker).

Table 4. Comparison of Techniques Utilizing Electron Beams (From Williams and Goldstein, 1981)

Characteristic	Electron Optical Equipment*			
	SEM	EPMA	TEM	AEM
1. Specimen type	Bulk	Polished bulk	Thin section	Thin section
2. Image resolution	7-10 nm	7-10 nm	~0.2 nm (line)	~0.2 nm (line) ~0.2 nm (stem)
3. Chemical analysis technique	x-ray	x-ray	-	x-ray, energy loss
a) measurement equipment	EDS	EDS, WDS	-	EDS, CBD, EELS
b) spatial resolution	~1 μm	~1 μm	-	≲50 nm
c) minimum detectability limit (WT%)	~0.1%	~0.1%	-	~0.1%
d) minimum detectable mass (g)	10^{-14}	10^{-15}	-	10^{-19}
e) accuracy	-	≥±1% (rel.)	-	±5-10% (rel.)
4. Structural analysis	-	-	Elect. diffraction ~1-5 μm	Elect. diffraction ≤10-20 nm
5. Other techniques	Magnetic contrast Electron beam induced conductivity Channeling patterns (crystal orientation) Voltage contrast Cathodoluminescence (emission of visible light)			

*AEM = analytical electron microscopy, CBD = convergent beam diffraction (microdiffraction), EDS = x-ray energy dispersive spectrometry, EELS = electron energy loss spectroscopy, EPMA = electron probe microanalyzer, SEM = scanning electron microscopy, STEM = scanning TEM, TEM = transmission electron microscopy, WDS = wavelength dispersive spectrometry (crystal spectrometer)

What are the implications of those developments for the cereal chemist and technologist? I am confident that in the not too distant future we will witness adaptation of those instruments to biological systems, in general, and cereals, in particular. I am confident that we will be able to use the stage of the microscope to see, measure, record, and interpret:

Structural features of the kernel as they relate to composition, hardness, resilience, conditioning, and milling;

Changes that occur in a dough as it is mixed, fermented, proofed, heated, and baked;

Changes in water migration, starch modification, and overall staling in a baked product as it is stored;

Modifications in a germinating barley kernel as they relate to steeping, malting, mashing, and lautering;

Extrusion of versatile products by energy-saving techniques;

And many others.

I am confident that we will be in a position to put the micro hardness tester, the mixer, the oven, the extrusion equipment, the malting chamber or some of their adaptations on the stage of a microscope and follow continuously on a microscale what is happening and learn to relate those microscopic changes to the real macro scale world.

As I have said on a previous occasion (Pomeranz, 1980), the four questions most commonly asked by workers in biological sciences are: what? how much? where? and what function? The question--*what?*--relates to the identity of components in the biological system under investigation. The answer to *how much?* provides information on the quantities of identified components in the system. The answer to the question *where?* is designed to localize the component(s) in the plant or animal tissue or in a processed food such as bread. The fourth question, has to do with function, either basic--physiological or applied--food processing. To what extent can answers to all four questions be generated by new instrumentation? I believe that recent developments show great promise in this respect.

Improving Nutritional Quality by Modifying Grain Morphology

The greatest promise, to my mind, lies not in applying instruments, but in restructuring the cereal grain. In recent years many studies have concerned the improvement of the nutritional value of cereal grains. Simple changes in grain morphology could be the basis of improvement. The embryo of cereal seeds is rich in protein (up to 38%), and the protein may contain about 7% lysine. Selection for larger embryos is particularly important if the whole seed (rather than starchy endosperm) is to be consumed. Variations in the

number of aleurone cells of the endosperm exist in corn, rice, and barley. The aleurone layer is rich in protein having a good amino acid balance. Selection for a high aleurone cell number could be useful, provided the high number is associated with improved nutritional value and the deleterious effects of high phytic acid in the aleurone layer are counteracted. Both in wheat and in rice, much of the protein is concentrated in the aleurone and outermost subaleurone. Those tissues are diverted to feed during milling and polishing of rice or during milling of highly refined wheat flour. "Restructuring" cereal grains for a more even distribution of protein throughout the whole endosperm would increase the protein content of milled products.

I am confident that by the time of the third millennium we will have as a result of a combination of classical plant breeding and modern genetic engineering a vastly improved grain. I am confident that modern genetic engineering, the precision tool of plant breeding, will be able to perform a microsurgery that will cut and paste together the genetic code of the improved cereal grain. That improved grain will be produced from high yielding cultivars, that are relatively insensitive to adverse conditions, make the best of sound cultural practices, and are uniquely structured to meet the specifications of millers, bakers, and malters. The spherical or barrel-shaped kernel with a minimum crease, will have an aleurone and germ positioned in the kernel in such a manner that it will make possible a flour extraction that approaches the starchy endosperm content. The milled product will, at least, equal in protein content the whole wheat and will be adaptable to production of a variety of cereal-based products. It will produce delectable, nutritious, and readily available foods that have excellent shelf life and meet with consumer acceptance.

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Discussion with Reviewers

R. Moss: What is meant by endosperm cavity height? The distance within the endosperm, from the dorsal to the ventral side of the grain?
Author: I assume the author (Kosina, 1979) meant the distance within the endosperm, from the dorsal to the ventral side of the grain. The author, however, did not define the term endosperm cavity height in the paper.

E. Varriano-Marston: I do not see how protein body fusion can be significant in loaf volume potential since much of the literature suggests that it is the fibrillar proteins (gluten) that are important in breadmaking potential. Please explain; maybe I am reading you wrong.
Author: I did not say "identical with", I said "concomitant with" which is what our data show. "Limited aggregation" does not contradict involvement of "fibrilar" proteins; fibrilar proteins can aggregate.

Editor: How were the grain samples prepared for Scanning Electron Microscopy?
Author: Grain and malt samples were prepared for SEM by splitting with razor blades and mounting on specimen holders which were spread with a colloidal graphite adhesive. The specimens were then coated with a thin gold film in a sputter coater. Dough or bread samples are cut with a pair of sharp scissors with minimum distortion from smooth, freshly exposed surfaces. Small pieces are quick frozen and freeze dried and the freeze-dried pieces are broken - fractured to expose interior surfaces prior to coating.