Effect of the waxy endosperm gene on germination, agronomic, and malt quality characteristics in barley (Hordeum vulgare)
by Philip Lyle Bruckner

A thesis submitted in partial fulfillment of the requirements for the degree of MASTER OF SCIENCE in Agronomy
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Abstract:
A series of four waxy-normal endosperm isogenic pairs developed in three barley (Hordeum vulgare) lines were used to evaluate the effect of the waxy endosperm gene on certain germination and agronomic traits during and after kernel maturation as well as on malting quality and speed of imbibition. Plots in two locations were sampled three times per week from shortly after anthesis to harvest maturity to evaluate seed size, alpha-amylase activity, and germination characteristics during kernel maturation. A second set of plots was used to evaluate the effect of the waxy endosperm gene on yield, malt quality, and other agronomic traits at harvest maturity. Speed of imbibition in four Compana isotypes grown in three environments was determined in a 48 hour, 20° C, unaerated steep. Isogenic analysis showed that waxy endosperm isotypes exhibited significantly less dormancy, smaller seeds, and greater alpha-amylase activity during kernel maturation than did normal endosperm isotypes. Waxy and normal endosperm isotypes did not vary significantly in germination behavior, yield, heading date, or in most malting quality characteristics at harvest maturity. Waxy endosperm isotypes were significantly lower in diastatic power, had significantly lower seed weights, and in certain genetic backgrounds were shorter than normal isotypes.

Waxy and normal endosperm isotypes did not differ significantly in speed of imbibition although preliminary information showed that waxy isotypes could reach a 45% moisture level from 9.1 to 14.0 hours sooner than normal Compana isotypes. Naked Compana isotypes imbibed water significantly faster, yielded 17% less, had a 16% smaller seed weight and exhibited significantly less dormancy and a greater germination speed during kernel maturation and at harvest maturity than did covered Compana isotypes.

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ABSTRACT

A series of four waxy-normal endosperm isogenic pairs developed in three barley (Hordeum vulgare) lines were used to evaluate the effect of the waxy endosperm gene on certain germination and agronomic traits during and after kernel maturation as well as on malting quality and speed of imbibition. Plots in two locations were sampled three times per week from shortly after anthesis to harvest maturity to evaluate seed size, alpha-amylase activity, and germination characteristics during kernel maturation. A second set of plots was used to evaluate the effect of the waxy endosperm gene on yield, malt quality, and other agronomic traits at harvest maturity. Speed of imbibition in four Compana isotypes grown in three environments was determined in a 48 hour, 20° C, unaerated steep. Isogenic analysis showed that waxy endosperm isotypes exhibited significantly less dormancy, smaller seeds, and greater alpha-amylase activity during kernel maturation than did normal endosperm isotypes. Waxy and normal endosperm isotypes did not vary significantly in germination behavior, yield, heading date, or in most malting quality characteristics at harvest maturity. Waxy endosperm isotypes were significantly lower in diastatic power, had significantly lower seed weights, and in certain genetic backgrounds were shorter than normal isotypes. Waxy and normal endosperm isotypes did not differ significantly in speed of imbibition although preliminary information showed that waxy isotypes could reach a 45% moisture level from 9.1 to 14.0 hours sooner than normal Compana isotypes. Naked Compana isotypes imbibed water significantly faster, yielded 17% less, had a 16% smaller seed weight and exhibited significantly less dormancy and a greater germination speed during kernel maturation and at harvest maturity than did covered Compana isotypes.

INTRODUCTION

Waxy barley starch is more readily modified by either chemicals or enzymes than is normal barley starch. This and other unique properties of waxy barley starch indicate that waxy barley may become increasingly important in certain industrial processes. Little information has been developed on the effect of the waxy endosperm gene on agronomic, germination, and malt quality characteristics before and after harvest maturity.

The objectives of the following studies were to determine the effect of the waxy endosperm gene on germination, dormancy, seed size, and alpha-amylase activity during kernel maturation and on imbibition and malting quality in the mature grain.

LITERATURE REVIEW

Germination Characteristics of Developing Barley Grain

Isolated barley embryos can germinate at a very early stage of development, even as early as five days after anthesis (Bishop, 1944; Gordon, 1969). The ability of the embryo to germinate at this early stage depends on a critical embryo size (Cameron-Mills and Duffus, 1980). Intact seeds, however, cannot germinate in the early developmental stages. Gordon (1969) reported that only a few intact barley seeds germinated in the first 42 days after anthesis. Wellington (1964) reported no germination in barley seeds until 3-4 weeks after anthesis, while Bilderback (1971) showed no germination up to 20 days after anthesis when the seeds were immediately placed on moist sand. Similar results are reported for wheat (Wellington, 1956a, 1956b; Mitchell et al., 1980).

The seed can, however, germinate at a very early stage if the embryo is exposed to the air by rupturing the pericarp and testa. Gordon (1969) reported a germination rate in barley of 75% ten days after anthesis under these conditions. Wheat can germinate as early as 3-4 weeks after anthesis when the embryo is exposed mechanically (Wellington, 1956b). Drying of the immature grain has a similar effect. Air dried grain germinates at an earlier stage of maturity (10 days after anthesis) and at a faster rate than undried grain

(Mitchell et al., 1980). Wellington reported germination of desiccated immature wheat three weeks after anthesis compared to five weeks after anthesis in undried grain (Wellington, 1956b).

The germination capacity of immature grain develops early after fertilization, probably within the first 10-15 days following anthesis (Gordon, 1969; LaCrox et al., 1976; Gordon et al., 1979; Mitchell et al., 1980; Takahashi, 1980). The germination capacity rises, then decreases rapidly with decreasing moisture content and the induction of dormancy (Takahashi, 1980). High temperatures during seed formation (10-20 days post anthesis) repress dormancy induction while low temperatures enhance the induction of dormancy (Takahashi, 1980). LaCroix et al. (1976) reported a decrease in germination percentage in spring wheat 34-40 days after anthesis. Wellington (1964) reported a 14% and 22% reduction in germination of Domen and Herta barleys respectively, as secondary dormancy was imposed. This dormancy breaks down with increasing maturity and storage.

The pericarp is implicated as having some control over germination in developing barley (Wellington, 1964; Gordon, 1969; Briggs, 1978; Mitchell et al., 1980). Water loss through the pericarp occurs while the embryo and remainder of the grain is still accumulating water. The capacity to germinate first occurs when 50% of the water has been lost from the outer pericarp (Mitchell et al., 1980).

The capacity to germinate at harvest time and during barley

development is a varietal character that is subject to environmental variation. A study of 4 malting and 3 nonmalting barley varieties showed initial germination capacity occurring 25-44 days after anthesis and 50% germination capacity occurring between 30 and 60 days after anthesis (LaBerge et al., 1971). Gordon et al. (1979) showed that a 50% germination potential in wheat occurred 32-38 days after anthesis. Speed of germination increases with increasing maturity in barley (Bilderback, 1971) and wheat (LaCroix et al., 1976).

Alpha-amylase Development in Developing Barley Grain

In barley and wheat at least two types of alpha-amylase exist (Daussant and Mayer, 1980; Kruger, 1980; Gordon, 1980; Sargeant, 1980). Green alpha-amylase, alpha-amylase I, is found in developing seeds. Malt alpha-amylase, alpha-amylase II, is found predominantly in germinating seeds (Daussant and Mayer, 1980; Gordon, 1980). Recent evidence indicates both types of alpha-amylase are present in germinating barley seed, although most of the alpha-amylase activity is due to alpha-amylase II (Daussant and Mayer, 1980).

Green alpha-amylase, located in the pericarp of developing seeds, is independent of the embryo or exogenous gibberellic acid for synthesis (MacGregor et al., 1972; Allison et al., 1974; Daussant and Mayer, 1980; Gordon, 1980; Sargeant, 1980). The aleurone layer and the scutellum synthesize malt alpha-amylase de novo in response to

gibberellins from the embryo or exogenous GA (Daussant and Mayer, 1980). The two types of alpha-amylase have different electrophoretic properties, pH optimums, pH and heat stabilities and starch digesting properties (Daussant and Mayer, 1980).

Alpha-amylases in immature barleys increase rapidly after anthesis, then decline to a low, stable level (Duffus, 1969; LaBerge et al., 1971; MacGregor et al., 1971; MacGregor et al., 1972; Allison et al, 1974; Riggs and Gothard, 1976; Gordon, 1980). Considerable variation in alpha-amylase developmental patterns exists due to environmental and cultivar characteristics (MacGregor et al., 1974; LaCroix et al., 1976; Riggs and Gothard, 1976). LaBerge et al. (1971) in a study of seven barley cultivars, including four malting barleys, showed that alpha-amylase levels increased immediately after anthesis, peaked in the first ten days following anthesis, then declined to a low level by 10-15 days after anthesis. Allison et al. (1974) demonstrated that pericarp anthesis in Goldfield barley peaked about fifteen days after anthesis. Riggs and Gothard (1976) reported peak alpha-amylase activity 10-16 days after anthesis in six barley cultivars although a hulless barley cultivar, Nackta, showed no definite peak. Maximum alpha-amylase activity differed markedly between cultivars although there were no cultivar differences in alpha-amylase activity from 31 days after anthesis to maturity. Conquest barley showed a peak alpha-amylase activity 11 days after ear emergence (two days after anthesis)

when grown in the field while maximum activity of Conquest and Fergus barley grown in a growth chamber occurred 9-10 days and four days after anthesis, respectively (MacGregor et al., 1971; MacGregor et al., 1972). The alpha-amylase activity in Conquest declined to 1/10 its maximum after 28 days and remained constant until maturity. Duffus (1969) reported peak alpha-amylase levels in barley grown in Britain occurred 20-30 days after anthesis and that chlorocholine chloride applied shortly after anthesis inhibited alpha-amylase synthesis. Similar enzyme development patterns and cultivar variations are reported in wheat (LaCroix et al., 1976; Gordon et al., 1979; King and Gale, 1980).

A set of alpha-amylase isozymes which have similar isoelectric points to germination alpha-amylase have been reported in developing immature wheat in addition to the green alpha-amylase normally present (Kruger, 1980). The presence of these isoenzymes indicates that some leakage may occur in the germination control mechanisms and that control over germination may not be 100% effective. Malting barleys, with characteristic strong enzyme systems and short dormancy periods, showed increased alpha-amylase activity during late ripening in response to rain (LaBerge et al., 1971). Riggs and Gothard (1976) reported a strong correlation between peak alpha-amylase activity during barley development and germination energy three weeks after harvest.

The purpose of alpha-amylase in the pericarp during grain development and the reasons for its disappearance during maturity are

unknown (MacGregor et al., 1972; Allison et al., 1974; Riggs and Gothard, 1976; Daussant and Mayer, 1980). MacGregor et al. (1972) and Allison et al. (1974) speculated that green alpha-amylase in the pericarp may hydrolyze pericarp starch early in development to provide energy for growing kernels or produce short chain primers to be used in endosperm starch synthesis. Sargeant (1980) showed that in wheat, green alpha-amylase in the pericarp is not involved in starch synthesis. Disappearance of green alpha-amylase during maturation may be due to inhibition, inactivation, insolubilization, or degradation of enzymatic protein (MacGregor et al., 1972; Riggs and Gothard, 1976; Daussant and Mayer, 1980).

The aleurone synthesizes alpha-amylase in response to gibberellic acid (Gordon, 1980). The ability to respond in this way arises early in development. Gordon (1980) stated that the ability of aleurone layers to respond to GA arises 25-30 days after anthesis, although cultivars vary considerably in this property. The stage of development at which the embryo can synthesize gibberellins remains unclear (Gordon, 1980). Cool ripening environments that slow grain dehydration also slow embryo maturation as well as the aleurone cells' ability to produce germinative alpha-amylase. Marchylo et al. (1980) harvested wheat 25, 32, and 39 days after flowering and found that the embryo scutellum could synthesize alpha-amylase in the absence or presence of GA_3 25 days after flowering. The immature endosperm-aleurone

produced alpha-amylase in response to GA_3 32 days after flowering. At successive developmental stages the tissues synthesized higher levels of alpha-amylase. Bilderback (1971) showed that aleurone layers in Himalaya barley still could not respond to exogenous GA_3 25 days after anthesis. However, dried immature seeds at least 20 days old when incubated in exogenous gibberellic acid, produced large amounts of alpha-amylase. King and Gale (1980) reported no alpha-amylase production in half seeds of wheat after GA_3 incubation until 35-40 days after anthesis. Artificially dried detached half seeds responded to GA_3 as early as 22 days after anthesis. The rate and duration of drying influenced the ability of the immature grain to respond to GA_3 and produce alpha-amylase (King and Gale, 1980).

Barley Starch in the Mature Barley Kernel

Starch, a glucan, forms the food reserve polysaccharide in plants (Greenwood, 1964). Starch, the most significant chemical component of the cereal grain constitutes two-thirds of the dry weight of barley (Harris, 1962). Starch comprises 63-65% of the dry weight of plump, two rowed barley (Harris, 1962; Briggs, 1978). The endosperm stores the majority of the starch in barley, making up 85-89% of this tissue (Briggs, 1978).

Starch is exclusive to plants, being a mixture of two types of glucose polymers, one linear and the other branched (Harris, 1962;

Bewley and Black, 1978; Briggs, 1978; Bohlinski, 1979). Amylose is a straight chain polymer containing D-glucopyranose units linked by alpha(1-4) glucosidic bonds which may be as long as 2000 glucose residues. Amylopectin is a much larger molecule with a highly branched structure in which the alpha(1-4) linked glucopyranose chains are branched with alpha(1-6) linkages every 24-26 glucose residues.

The relative proportion of amylose and amylopectin can be estimated by the iodine binding capacity or iodine affinity test, capitalizing on the fact that amylose binds iodine at constant free iodine concentrations up to a limiting value while amylopectin binds little iodine. Starch, being a mixture of the two polymers, has an intermediate binding capacity. The proportion of amylose is estimated by extrapolation of the linear portion of the amylose and starch binding curves to zero free iodine and using the following relationship (Greenwood, 1964):

$$\% \text{ Amylose} = \frac{\text{Iodine Affinity of starch}}{\text{Iodine Affinity of amylose}} \times 100$$

Enzymes degrade starch (Greenwood, 1964; Bewley and Black, 1978; Briggs, 1978; Bohinski, 1979). The two principle hydrolyzing enzymes are alpha- and beta-amylase. Both attack alpha(1-4) bonds but in different patterns. Alpha-amylase cleaves alpha(1-4) bonds at random points while beta-amylase action removes successive maltose units beginning at the non-reducing terminus of the glucopyranose chain. Neither alpha- nor beta-amylase can remove branch points, with cleavage stopping

two to three glucose units from the alpha(1-6) bond. Thus amylose can theoretically be converted entirely to glucose plus maltose and alpha and beta dextrins by the combined action of alpha- and beta-amylase. The same two enzymes will only partially degrade amylopectin. Even though the theoretical yield of maltose from amylose digested by pure beta-amylase is 100% (Harris, 1962) the actual yield is near 73%. This incomplete conversion of amylose by beta-amylase is probably due to some modification in amylose structure (Greenwood, 1964), possible anomalous alpha(1-6) linkages (Briggs, 1978). Beta-amylase converts only 58% of amylopectin to maltose (Greenwood, 1964).

Starch grains are laid down as insoluble particles. The starch content of the grain during grain development rises in a logarithmic (Harris, 1962) or sigmoidal fashion (Briggs, 1978). Ninety-five percent of the starch is deposited in the initial 11-28 days after spike emergence (MacGregor et al., 1971; Briggs, 1978). Genotype and environmental characteristics control the amount and properties of starch (Harris, 1962). Amylose and amylopectin are not synthesized simultaneously (Harris, 1962; MacGregor et al., 1971; Briggs, 1978). The amylose content of the starch increased from a value of 13.8% after 14 days to 22.5% and remained constant to maturity, indicating that during the early stages of growth, the amylopectin fraction was synthesized at a relatively faster rate than amylose (MacGregor et al., 1971). The proportion of small starch granules increases dramatically with

maturity (MacGregor et al., 1971; Briggs, 1978).

Barley starch granules are generally round and follow a bimodal size distribution pattern. The small and large size granules being 1.7-2.5 um and 22.5-47.5 um in diameter, respectively (Briggs, 1978). Two distinct starch granule sizes were found in wheat, the larger granules being 25 um in diameter and the small granules 5 um in diameter. The small starch granules accounted for 90% of the total number of granules but comprised only 10% of the total starch weight (Bewley and Black, 1978). Small starch granules have a higher gelatinization temperature and amylose content (41.3%) than large granules (26.9%) (Briggs, 1978).

Normal barley starch contains approximately 25% amylose and 75% amylopectin (Bewley and Black, 1978; Eslick, 1979). Various reports on amylose show its range to be from 11-26% of the starch (Harris, 1962; Greenwood, 1964; Briggs, 1978). Genetic mutations in barley and in other cereals have resulted in starches containing almost 100% amylopectin (Banks et al., 1970; Bewley and Black, 1978). In theory, true waxy barleys contain 100% amylopectin (Eslick, 1979) although waxy endosperm barleys do contain some amylose (0-3%) (Banks et al., 1970; Goering et al., 1973; Briggs, 1978). High amylose mutations have also been reported (Greenwood, 1964; Briggs, 1978).

Nonwaxy and waxy barley kernels and pollen grains can be distinguished by iodine staining (Eriksson, 1969). A single recessive gene

on chromosome one that expresses xenia controls the waxy endosperm character (Eslick, 1979). Ericksson (1969) reported the spontaneous mutation rate to the waxy phenotype in barley to range from 5.1×10^{-6} to 15×10^{-6}.

Many properties of waxy endosperm barleys have been studied and documented. A study of two Japanese waxy endosperm barley cultivars showed both to have less than 0.5% amylose, a beta-amylolysis limit of 54% conversion into maltose, and an average chain length of 23 glucose units (Banks, et al., 1970). The average molecular weight of this starch was 300×10^6 and showed a bimodal distribution of large and small starch granules with a range of 22 and 3 um in diameter, respectively. Waxy barley starch gelatinizes a few degrees lower than waxy corn starch.

Waxy endosperm barley starches are more susceptible to enzymatic action possibly because the pure amylopectin starch granules are less resistant to enzymolysis than the amylose/amylopectin hybrid granule found in nonwaxy barley (Goering et al., 1973; Fox and Eslick, 1979). Goering et al. (1973) has shown that waxy barley starch has a 20°C lower pasting temperature than normal barley (75°C vs. 95°C). In corn and sorghum the amylose content of the starch does not influence gelatinization temperature. Goering et al. (1973) also showed waxy Compana barley starch had a similar Brabender cooking curve to that of waxy sorghum starch but had only half the swelling power and one-third the

solubility of waxy sorghum starch, indicating that the amylopectin in waxy Compana was a unique starch. Waxy barley starch malts more rapidly than normal barley starch when measured by maltose production (Fox and Eslick, 1979). Waxy barley flour is higher than normal barley flour in beta-glucans. The starch from waxy short-awn nude Compana grown at one location retained sufficient alpha-amylase-like activity to liquify itself and four times its weight of waxy or normal corn starch (Goering and Eslick, 1976). A new process for production of ultra high maltose syrup from waxy endosperm barleys is based on some of these unique properties of waxy barleys (Goering et al., 1980). It was further suggested that the ease of liquification of waxy barley starch as well as other characteristics make waxy barleys potentially useful in the brewing industry (Goering et al., 1973; Goering and Eslick, 1976).

Sullins and Rooney (1974) demonstrated that waxy sorghum was more rapidly solubilized by alpha-amylase enzymes and by buffered rumen fluid than nonwaxy sorghum and that the waxy sorghum kernel had less peripheral endosperm. Furthermore, waxy sorghum had a peripheral endosperm less dense than nonwaxy sorghum, having large starch granules with a less dense protein matrix more susceptible to enzyme attack (Sullins and Rooney, 1975). They concluded that the properties of the peripheral endosperm were responsible for waxy sorghum starch being more susceptible to enzyme attack.

Yield trials conducted over 43 environments with waxy and normal isogenic pairs have shown no significant yield reduction in the waxy endosperm genotype (Eslick, 1979). Yield comparisons of covered and hulless isogenic pairs grown in replicated yield trials over 149 environments indicate an average yield reduction of 12% for the hulless character (Eslick, 1979). Isogenic comparisons between short awn and long awn isogenic pairs in replicated yield trials over 95 environments have shown no significant differences in yield (Eslick, 1979).

Endosperm Modification and Enzyme Development

The mobilization of cereal endosperm reserves during germination is required to replenish embryo carbohydrate reserves depleted during the first 24-48 hours following imbibition (Bewley and Black, 1978). The embryo controls the mobilization of endosperm reserves. The embryo synthesizes and releases gibberellin to the aleurone layer where production and release of hydrolytic enzymes occurs (Bewley and Black, 1978; Briggs, 1978). Mobilization commences near the scutellum andspreads along the endosperm as the aleurone layer begins enzyme release (Bewley and Black, 1978; Gibbons, 1980).

Hydrolysis of starch grains does not take place if the enclosing cell walls are intact. Wheat cell walls are composed of 15% protein and 75% carbohydrates. Beta-glucans and arabinoxylan make up most of the carbohydrate fraction. Proteinases, arabinoxylanases, and

pentosanases are synthesized in the aleurone layer and released in response to gibberellin (Bewley and Black, 1978). Acid phosphatase, ribonuclease, and endo-beta-glucanase can be synthesized in the absence of gibberellin but their activities are enhanced and their release controlled by gibberellins (Bewley and Black, 1978; Briggs, 1978). Beta-glucanase, arabinoxylanases, and proteinases degrade cell wall constituents and dissolve cell walls. Degradation of the endosperm wall occurs initially in the sub-aleurone layers then progresses to the center of the endosperm. Erosion of starch granules begins only after cell walls have been modified. Dissolution of cell walls renders the enclosed starch accessible to enzymatic attack (Bewley and Black, 1978).

Gibberellins, released by the embryo, induce the aleurone layer to undergo metabolic changes resulting in the release of many enzymes. Much of the work, however, on barley enzyme synthesis has focused on the synthesis of alpha-amylase. Levels of alpha-amylase activity increase during barley germination (Kneen, 1944; Briggs, 1964; Reynolds and MacWilliam, 1966; Briggs, 1968a; Palmer, 1969; Kruger, 1976; Bewley and Black, 1978; Briggs, 1978; MacGregor, 1978; MacGregor and Daussant, 1979; Daussant and Mayer, 1980). Barley kernels synthesize alpha-amylase de novo primarily in the aleurone layer in response to a gibberellin trigger received from the embryo. De novo alpha-amylase synthesis is documented and supported by many facts including

(a) the production of alpha-amylase in isolated barley aleurone layers in response to endogenous GA, (b) subsequent declines and increases in enzyme production as the hormone is removed and reapplied (Briggs 1964; Bewley and Black, 1978), and (c) immediate stoppage of alpha-amylase synthesis by protein synthesis inhibitors (Briggs, 1963, 1964; Bewley and Black, 1978). Definitive proof of de novo synthesis of alpha-amylase was shown by labeling amino acids with ^{18}O (Bewley and Black, 1978) or radioactivity (Briggs, 1964) and subsequently detecting the labeled amino acids in synthesized alpha-amylase.

Several studies have shown synthesized gibberellin is involved in enzyme synthesis. Experiments have shown that excised barley embryos can release gibberellin up to 60 hours in vitro (Briggs, 1978), that the endosperm of degermed grain can be modified in the presence of exogenous GA while little modification occurs without the GA (Briggs, 1968a, 1978), and that the embryo can be removed two to three days after germination begins, with little effect on enzyme level (Briggs, 1978). Up to seven isozymes of alpha-amylase including the two major groups of the enzyme appear in GA treated isolated aleurone layers and during germination (Bewley and Black, 1978; MacGregor, 1978; MacGregor and Daussant, 1979; Daussant and Mayer, 1980).

During the lag phase between gibberellin arrival at the aleurone layer and rapid alpha-amylase synthesis the hormone redirects protein synthesis in the aleurone cells, possibly by enhancing synthesis of RNA

including mRNA for alpha-amylase. Gibberellin may also enhance activation or protection of mRNA in the aleurone cells. There is little firm evidence that GA acts at the translational level of control (Bewley and Black, 1978).

The aleurone layer produces about 85% of the alpha-amylase and the scutellum 15%. Malted barley shows about 7% of the alpha-amylase is located in the embryo with the remaining 93% in the endosperm indicating that about half the alpha-amylase produced in the embryo is released to the endosperm (Briggs, 1964, 1978.). Alpha-amylase accumulates in the aleurone layer before its release (Briggs, 1978). Synthesis of alpha-amylase is probably limited not by the inability of the aleurone layer to respond to gibberellin but by the supply of gibberellin to the aleurone layer (Atanda and Miflin, 1970; Briggs, 1978). The deficiency in gibberellin supply to the aleurone layer is probably due to deficiencies in the transport of gibberellin to the aleurone rather than lack of synthesis at the embryo since endosperm areas distal to the embryo produce disproportionately small amounts of alpha-amylase (Atanda and Miflin, 1970).

High embryo sugar levels may depress levels of alpha-amylase production (Briggs, 1964) or prevent gibberellin production or release from the embryo (Briggs, 1978). Enzyme synthesis may be controlled through a feedback loop in which gibberellin production is controlled by the sugar levels in the embryo while sugar levels are controlled by

enzyme synthesis and ultimately gibberellin production (Briggs, 1978). Proteolysis, removal of starch and calcium ions, and a falling pH contribute to declining enzyme activity (Briggs, 1968a). Externally applied gibberellins can replace or augment embryo produced gibberellin to initiate or increase alpha-amylase formation (Briggs, 1963, 1964; Reynolds and MacWilliam, 1966; Briggs, 1968a; Palmer, 1969; Atanda and Miflin, 1970; Bewley and Black, 1978; Briggs, 1978; MacGregor, 1978; Smith and Briggs, 1978). The amount and rate of alpha-amylase production is also influenced by barley genotype (Atanda and Miflin, 1970; Gothard, 1974; Bewley and Black, 1978; MacGregor, 1978), temperature and water conditions during germination (Kneen, 1944; Briggs, 1968a; MacGregor and Daussant, 1979), oxygen availability (Smith and Briggs, 1978) and mechanical damage (Briggs, 1968b).

Proteinases synthesized de novo in the aleurone layer in response to gibberellin, activate beta-amylase, present in a bound insoluble form in ungerminated barley, by reducing the number of disulfide bridges between the enzyme molecule and insoluble proteins (Bewley and Black, 1978; Briggs, 1978). Beta-amylase is not synthesized de novo. Beta- and alpha-amylases work together in starch breakdown and can completely hydrolyze amylose although amylopectin digestion halts with the production of limit dextrins (Bewley and Black, 1978).

Two debranching enzymes are reported in cereals. R enzyme, found in barley malt, debranches amylopectin and beta-limit dextrins, while

limit dextrinase, reported in Proctor barley and ungerminated oats, attacks alpha- and beta-limit dextrins but does not debranch amylopectin (Bewley and Black, 1978). The aleurone layer synthesizes limit dextrinase de novo at the time of starch breakdown, its activity being enhanced 10-15 fold between the first and fourth days following imbibition (Bewley and Black, 1978).

Starch amolysis produces two major products, alpha- and beta-maltose. Alpha-glucosidase, an enzyme present in the quiescent barley embryo and aleurone layers, hydrolyzes these products (Bewley and Black, 1978; Briggs, 1978). The enzyme activity increases at both sites during the first five days following imbibition. Barley half grains release a de novo-synthesized alpha-amylase from the aleurone layer within 12 hours of the addition of GA (Bewley and Black, 1978). Glucose, the resulting product of alpha-glucosidase activity, is absorbed by the scutellum, converted to sucrose, and transported to the growing embryo (Bewley and Black, 1978).

Germination

The germination process results in growth of the grain embryo, manifested by the growth of the root and an increase in the length of the shoot, with concurrent modification of the contents of the endosperm (Schuster, 1962). Germination begins as the surface layers pick up water which penetrates through the micropyle into the kernel (Briggs,

1978). The grain swells during the first 24 hours and the embryo becomes turgid and increases in size. Germination begins with the emergence of the coleorhiza, primary, and secondary seminal roots from the embryo (Pollock, 1962; Briggs, 1978). At almost the same time the coleoptile breaks through the testa and begins elongation. The coleoptile grows up the dorsal side of the kernel, pressing a groove in the endosperm and emerging from the apex of the kernel or through the lemma along one side (Schuster, 1962; Briggs, 1978). The outside grain coverings probably restrict physically the elongation of the plumule since the elongation of the plumule in dehusked grain during germination is considerably greater than in intact barley kernels (Schuster, 1962). Endogenous reserves support initial growth of the embryo while subsequent growth makes use of products from the endosperm dissolution (Briggs, 1978).

Endosperm breakdown begins immediately below the scutellum in a layer of compressed cells and moves toward the grain apex (Schuster, 1962; Briggs, 1978). After a short period, alteration proceeds faster immediately below the aleurone layer (Briggs, 1978). The asymmetry of endosperm breakdown results from the angle of the scutellar face against the endosperm, the sequence and location in which degradative enzymes are formed, and the property of resistance to enzymatic degradation by residual walls in the nucellar sheaf cells (Briggs, 1978). Endosperm alteration begins with the hydrolytic decomposition of high

molecular weight materials by enzymes present in the barley (Schuster, 1962). The synthesis of certain enzymes follow soon after the beginning of germination (Schuster, 1962). One of these enzymes, alpha-amylase, develops rapidly in germinating grain, and with beta-amylase and certain other enzymes attacks starch granules, causing granule pitting and/or granule erosion and ultimately starch granule breakdown (Schuster, 1962; Briggs, 1978). The degradation products of the enzymes rapidly diffuse through the scutellum and are used for respiration and the energy processes of growth (Schuster, 1962).

Germination begins with water absorption and rehydration of the kernel (Brown, 1975; Bewley and Black, 1978). The kernel requires a minimum of 35% water for active germination (Essery et al., 1954; Kirsop et al., 1967; Brown, 1975; Wainwright and Buckee, 1977). Adequate kernel modification for malting barley requires a further increase in water percentage to 42-45% (Kirsop et al., 1967) or 43-46% (Brookes et al., 1976). Various researchers have concluded that good malting performance is positively correlated with the ability of barley to take up water rapidly (Brookes, et al., 1976; Ulonska and Baumer, 1976; Wainwright and Buckee, 1977).

Various factors affect water imbibition by the barley seed. Large kernels have been reported to initially imbibe water faster than small kernels although small kernels reach the appropriate level for germination more rapidly, and after prolonged steeping have a higher

moisture percentage, than large kernels (Pollock, 1962; Brookes et al., 1976; Davidson et al., 1976). Briggs (1978) reported smaller kernels take up moisture faster and to a higher final level than large kernels. A positive correlation between starch content and time required to reach a predetermined moisture level has been reported (Pollock, 1962; Brookes et al., 1976) as well as a corresponding inverse relationship between nitrogen content and imbibition rate (Brookes et al., 1976). Contradictory reports as to the influence of initial barley moisture content on water uptake exist but it is probable that barley, like other seeds, has a velocity of water uptake inversely proportional to the amount of water previously absorbed (Brookes et al., 1976). Old, dry barley seed absorbs water at a much slower rate than does fresh, moist seed, however. Increasing temperature increases the rate of imbibition (Pollock, 1962; MacLeod, 1967; Brookes et al., 1976; Briggs, 1978). All barleys probably have a similar temperature coefficient of imbibition. A linear inverse relationship exists between temperature and the log of the time required by grain to reach a particular moisture level (Briggs, 1978). Water sensitivity (Brookes et al., 1976), embryo and/or micropyle size (Davidson et al., 1976) and variety (Ulonska and Baumer, 1976) may also influence water uptake.

Upon immersion of barley in water an immediate moisture layer (2-3% fresh weight) forms around the kernel (Kirsop et al., 1967;

Brookes et al., 1976; Davidson et al., 1976; Briggs, 1978). This film of water must be removed for accurate water uptake studies (Briggs, 1978). Drying by blotting fails to remove all water on the external surface and may inflate estimates of water uptake by 1% or more (Davidson et al., 1976). Surface layers hydrate rapidly, within the first two hours in hulled barley and the first 30 seconds in hulless grains (Briggs, 1978). In naked grain the pericarp conducts water so well that the grain interior hydrates at the same rate whether the apex, the base, or the whole grain is immersed in water. During imbibition the moisture content increases rapidly, then at a progressively declining rate until, if germination does not occur, it approximates a limiting value (Briggs, 1978).

Numerous researchers have reported a triphasic pattern of water uptake in germinating seeds (Brookes et al., 1976; Bewley and Black, 1978; Takahashi, 1980). Phase I (imbibition) is purely a physical process, occurring during the first six to ten hours after immersion, characterized by rapid water uptake accounting for 60% of total water uptake. Seed colloids imbibe water previously lost during ripening regardless of dormancy or viability problems. Permeability of the seed integument controls this phase. Phase II (10-20 hours) is the lag or activation phase during which water uptake slows or ceases. Major metabolic processes begin and water uptake resumes with increasing osmotic pressure. Dormant seed does not undergo this phase as control

is exerted by embryo dormancy. Phase III (20+ hours) constitutes a steady linear increase in water uptake that can be correlated to plant metabolism. This phase, an active process, occurs only in viable seed and includes the visible onset of germination.

The surface layers of the barley kernel are the principle barriers to water entry. The testa is semi-permeable and may restrict water entry for the first few hours. The pericarp, however, is the principle barrier to water entry (Brookes et al., 1976). Gaps exist in the pericarp and testa, especially near the embryo, which may allow water entry. The embryo preferentially takes up water (Kirsop et al., 1967; MacLeod, 1967; Brookes et al., 1976). Embryo moisture content may reach 65-70% while the endosperm moisture content is 41%. Most water uptake by the kernel likely occurs in the micropylar region (Pollock, 1962; MacLeod, 1967; Brookes et al., 1976; Briggs, 1978).

A barley imbibition study conducted at 13° C indicated that the axis of the embryo and the scutellum absorb water considerably faster than all other kernel parts except the husk (Reynolds and MacWilliam, 1966). Rapid absorption of water by the embryo for six hours followed by a declining rate of absorption resulted in about 58% moisture after 24 hours in the imbibed embryo. The scutellum lagged behind the embryo in water uptake until about six hours then paralleled the embryo moisture content up to 58% moisture at 24 hours. The endosperm absorbed water more slowly, reaching 35% moisture at 24

hours. Water content of the whole grain was 38.4% after 24 hours. Another study conducted at 25° C showed water content in whole grain increasing from 17.4% to 23.8%, 26.2%, and 30.5% after 2, 4, 6, and 8 hours, respectively (Davidson et al., 1976).

Many factors affect the germination of seed, one of the most important being seed dormancy. Seed dormancy can be defined to include all instances of inhibition of germination, whether they are connected with the properties of the seeds themselves or whether they are determined by environmental conditions. In the broadest sense dormancy is the absence of germination or as a greater or lesser reduction of the germinative capacity of seeds, or finally as the maintenance of the capacity to germinate only within a small range of conditions (Nikolaeva, 1969). Barley seeds sometimes fail to germinate due to primary (intrinsic) dormancy or secondary dormancy, the latter of which is imposed by external factors (Briggs, 1978).

Dormancy varies among samples according to cultivar (Deming and Robertson, 1933; Chang, 1943; Bishop, 1944, 1945, 1946; Essery et al., 1954; Finlay, 1960a; Bell and Lupton, 1962; Carson and Horne, 1962; Pollock, 1962; Wellington, 1964; MacLeod, 1967; Belderok, 1968; Gordon, 1969; Lallukka, 1976; Briggs, 1978; Strand, 1980), weather conditions during growth and harvest (Bishop, 1945, 1946; Wellington, 1964; MacLeod, 1967; Belderok, 1968; Lallukka, 1976; Reiner and Loch, 1976; Briggs, 1978; Derera, 1980; Gordon, 1980; Strand, 1980; Takahashi,

1980), storage and handling procedures (Bishop, 1944, 1945, 1946; MacLeod, 1967; Gordon, 1969; Brookes et al., 1976; Briggs, 1978), and the methods used to test germination (Bishop, 1945, 1946; Essery et al., 1954; Essery et al., 1955; Pollock et al., 1955a, 1955b; Pollock, 1962; Wellington, 1964; Gordon, 1969, 1970; Briggs, 1978; Strand, 1980).

Dormancy expressed as a single number is usually unsatisfactory, since the germination rate, as well as final percentage may change during storage, e.g. a barley lot with 100% germination reached 50% germination within 31 hours immediately after harvest but required only 15.5 hours after three weeks of storage (Gordon, 1969). Thus a discussion of dormancy should include the rate as well as the final germination percentage under a defined set of conditions (Briggs, 1978). Numerous attempts have been made to assign single descriptive numbers to germination tests (Finlay, 1960a, 1960b; Gordon, 1969; Goodchild and Walker, 1971; Bewley and Black, 1978). The best type of presentation appears to be a graph of cumulative germination against time for each set of germination conditions (Briggs, 1978).

A mixture of factors occurring within the grain and its surface layers interacting with environmental factors probably cause barley dormancy. Embryo dormancy in barley is rare (Bishop, 1944; Pollock, 1962; Gordon, 1970; Briggs, 1978) although embryo dormancy has been reported in Hordeum spontaneum (Belderok, 1968). Briggs (1978) reported a study in which intact barley germinated 6% with germination

increasing to 12%, 46%, and 48% as 25%, 50%, and 75% of the endosperm was removed, respectively. Dehusked grain germinated at 12%, 58%, 65%, and 81% under the same conditions. Germination of isolated embryos was 100%, indicating no embryo dormancy and a certain germination inhibiting effect associated with the seed coverings.

The most promising theory on barley dormancy involves the internal level of free oxygen in the embryo (Pollock, 1962; Belderok, 1968; Briggs, 1978). Dormant and nondormant grains respire at about the same rate (Pollock, 1962; Briggs, 1978). The embryos of dormant grains, as shown by experiments with isolated embryos, have a higher free oxygen requirement or require a higher oxygen tension than nondormant embryos. This oxygen requirement of the embryo progressively declines during after-ripening (Pollock, 1962; Briggs, 1978) or the seed coats become more permeable to oxygen. Eventually enough oxygen reaches the embryo for both germination and growth (Belderok, 1968). Respiratory processes in the embryo compete for oxygen with the growth processes (Pollock, 1962; Belderok, 1968). Respiration in dormant seed may require so much oxygen that little may be left for germination (Belderok, 1968). The surface structure of the seed, surface moisture, and microbial populations on the seed limit the oxygen supply to the embryo (Briggs, 1978). The seed surface layers restrict oxygen flow, so dormancy may be broken or reduced by damaging the pericarp mechanically. The surface water film reduces oxygen uptake because of the time taken for oxygen to

dissolve into water, the time to diffuse into the grain, and the lower oxygen content in water as compared to air. Microbial populations on the seed influence seed dormancy by their high consumption of oxygen (Briggs, 1978). The production of gibberellins in plant tissues require aerobic conditions. The embryo requires oxygen in order to form gibberellins or to convert bound gibberellins to free gibberellins (Belderok, 1968).

Pronounced dormancy may be separated into three phases in barley (Belderok, 1968). The first is an immediate post-harvest phase in which practically no grains germinate in the germination test. The second is a transitional phase following the after-ripened state in which dormancy terminates in gradually increasing numbers of grains. In the third phase grains fully germinate at optimum moisture, but cannot do so if excess water is present. This latter behavior demonstrates water sensitivity. Essery et al. (1954) first detected water sensitivity when they found certain barleys would not germinate under normal steeping conditions. These barleys showed normal germinative energy under conditions of the accepted test, but if the amount of water was increased from 4ml to 8ml germination declined (Pollock et al., 1955b). Water sensitive barleys fail to germinate because the embryos fail to respond to the low internal oxygen tension resulting from the presence of an unduly thick water film (MacLeod, 1967). Low germinative energy may also reflect a deficiency of oxygen in the

vicinity of the embryo (Gordon, 1969). Finlay (1960a), however, found no water sensitivity in his study of the genetic and environmental variations in germination behavior in 800 cultivars. In fact, 8ml of water accelerated the germination rate over the use of 4ml of water in the test. Rate of water absorption (Belderok, 1968; Briggs, 1978) and nitrogen content (Bishop, 1946; Belderok, 1968) do not influence dormancy.

The following treatments reportedly break dormancy: drying with heat (Bishop, 1944; Pollock, 1962; Belderok, 1968; Briggs, 1978), gibberellin treatment (MacLeod, 1967; Belderok, 1968; Brookes et al., 1976; Briggs, 1978), removal of seed coat or mechanical damage (Bishop, 1944, 1945; Pollock et al., 1955a, 1955b; Wellington, 1964; Belderok, 1968; Gordon, 1970; Bishop, 1978), freezing, stratification or cold temperatures (Bishop, 1944; Briggs, 1978), germination in oxygen (Bishop, 1944, 1945; Pollock, et al., 1955b; Briggs, 1978) and various chemical treatments (Bishop, 1944, 1945; Pollock et al., 1955b; Pollock, 1962; Brookes et al., 1976; Briggs, 1978). One or two recessive genetic factors reportedly control dormancy in barley (Belderok, 1968).

The water conditions during a germination test also affect the final germination of the barley lot. In addition to conditions favoring water sensitivity and water imbibition as previously discussed, the position of the embryo relative to the water level, water

temperature, and dissolved ions may affect germination. Gordon (1969) reported significant differences in germination in a factorial experiment comparing tap and distilled water.

Temperature also influences germination. Kneen (1944) reported the relative growth rate of wheat and sorghum, as measured by average coleoptile length increased as germination temperature was increased from 14° C to 30° C. Gordon (1969) reported that the germination temperature influenced the expression of total germination under excessive moisture conditions but that it had little effect on the final germination percentage under optimum moisture conditions. The rate of water absorption in barley depends on temperature (Pollock, 1962). Specific temperatures have been noted such as 5° C as minimal, 29° C as optimal, and 38° C as maximal for barley germination (Briggs, 1978). Such figures are misleading since samples of grain vary widely in response to different germination conditions. Within limits mature grain germinates more rapidly at higher temperatures (Briggs, 1978). One study showed rootlet appearance in six days at 5° C, three days at 10.5° C, two days at 15.5° C, and one and three-fourth days at 17.8° C. Dormant grain, however, germinates better at lower temperatures. Most dormant barley will germinate at 2-7° C. As the grain matures, the temperature range over which germination occurs widens (Briggs, 1978). Fixed time or temperature tests may be misleading. Germination of one grain sample showed that germination in five days was best at

12° C (67%). The same seed sample germinated better (95%) in an eleven day test at 5° C. In the longer test germination at 12° C was only 86%.

Mechanical, heat, and pregermination damage reduce germination capacity of barley (Pollock, 1962). Microorganisms influence germination by increasing dormancy or by causing seed or seedling death (Briggs, 1978). Aging of seed leads to a disappearance of dormancy and a slow degeneration of the embryo leading to eventual death (Pollock, 1962). Cultivar characteristics and storage conditions affect barley germinative capacity and life (Pollock, 1962; Briggs, 1978).

Assessment of germination characteristics of grain generally falls into three groups of tests: germination capacity tests, germination energy tests, and special germination tests such as the water sensitivity test (Pollock, 1962). Germination energy tests assess the percent germination of a given seed lot at a particular time under standard conditions (Bishop, 1944; Belderok, 1968; Briggs, 1978; Brookes, 1980). Germination capacity tests determine the viability of the seed lot as the percentage of seed, both dormant and nondormant (Bishop, 1944; Belderok, 1968; Briggs, 1978; Brookes, 1980). The difference between germinative energy and germinative capacity determines the amount of dormancy in the sample (Briggs, 1978). The difference between germination with 4 ml of water and germination with 8 ml of water has been defined as water sensitivity (Essery et al.,

1955; Wainwright and Buckee, 1977; Briggs, 1978).

Many standardized germination methods exist although many have serious limitations (Bishop, 1944, 1945, 1946; Essery et al., 1954, 1955; Pollock et al., 1955a, 1955b; Pollock, 1962; MacLeod, 1967; Belderok, 1968; Gordon, 1969; Wainwright and Buckee, 1977; Briggs, 1978; Gordon et al., 1979). Tests and criteria for germination differ depending on intended use of the seed (Briggs, 1978). Maltsters require rapid germination such as 50% in one to two days and 95-100% in three days and score grains as germinated as soon as the coleorhiza appears. For seed testing purposes the seed must produce a whole, normal, vigorous seedling with an adequate root system, a coleoptile, and at least one seedling leaf (Briggs, 1978). Malting germination tests generally require only three to four days while seed testing germination require 7-14 days (Briggs, 1978). Reliable germination results require replicated subsamples under rigidly standardized conditions of temperature, amount of water, and test duration (Briggs, 1978). Inadequately controlled conditions may lead to serious discrepancies between various germination tests (Essery et al., 1954; Briggs, 1978). Micro-organism infections can cause mold growth and rotting during testing (Briggs, 1978). Most microbial infections can be controlled by seed treatment.

Germinative energy tests are usually conducted at 15-20° C and are about 72 hours in duration although some may take longer (Essery et al.,

1954). Some of the common germinative energy tests include the Schönfeld test, the Aubry method, the Cöldewe or Schönjahn method, and the Buchingor test (Bishop, 1944; Essery et al., 1954; Briggs, 1978). More satisfactory tests can be done in flat dishes, on filter papers, in graded sand or soil that is as uniform as possible to which exactly measured proportions of water are added (Briggs, 1978). No difficulties arise when determining germinative energy in fully mature barley although the precise environment during the test determines germination if some degree of dormancy exists in the barley sample (Pollock, 1962).

Germination capacity tests determine viability directly by forcing dormant seeds to germinate or indirectly by determining viability in some other way (Pollock, 1962; Briggs, 1978). Direct germination capacity tests involve pretreatment to eliminate dormancy. Common methods include steeping in dilute hydrogen peroxide (Thunaeus test) (Pollock, 1962; Wainwright and Buckee, 1977; Briggs, 1978), cutting the grains in half transversely followed by germination (Eckhardt test) (Bishop, 1944; Briggs, 1978), treatment with gibberellic acid (Pollock, 1962), removal of the husk and pericarp with sulfuric acid (Pollock et al., 1955a; Pollock, 1962; Briggs, 1978), mechanical peeling (Bishop, 1944, 1945, 1946; Pollock, 1962; Briggs, 1978), and germination in a solution of potassium nitrate (0.2%) (Briggs, 1978; Gordon et al., 1979). Indirect measurements of viability use chemical substrates to determine the viable proportion of seed. Dead seeds

take up barium chloride and resazurin or indigo carmine stains and these have been used to determine germinative capacity (Briggs, 1978). More successful indirect methods detect living tissues by the ability of their endogenous enzymes and substrates to reduce materials such as dinitrobenzene (Bishop, 1944; Briggs, 1978), salts of selenium (Bishop, 1944, 1945, 1946; Pollock, 1962; Briggs, 1978), tellurium (Briggs, 1978), or tetrazolium salts (Bishop, 1946; Pollock, 1962; MacLeod, 1967; Wainwright and Buckee, 1977; Briggs, 1978) to colored, insoluble substances which stain viable tissues. Tetrazolium tests are the most common, the chemical being reduced to highly insoluble, bright red forazans in viable tissue (Briggs, 1978).

Measurement of seed dormancy may involve determining the length of the dormant period, determining dormancy intensity (% dormant seed) or both (Strand, 1980). The length of dormancy is the number of days between the harvest stage and the time when a given percentage of grain (e.g., 50%, 85%, 98%) is capable of germinating under selected test conditions (Belderok, 1968). Strand (1980) defined the length of the dormant period as the number of storage days at 20° C required to reduce dormancy to an acceptable level. Dormancy length tests generally involve ear-germination tests which begin at harvest ripeness and are conducted at regular intervals until dormancy is gone or germination reaches a predetermined level (Belderok, 1968; Olsson and Mattsson, 1976). Olsson and Mattsson (1976) conducted a study on the

length of the dormancy period in wheat by germinating seeds at 18° C twice per week from full ripening until 50% germination. They found large cultivar differences in the length of the dormancy period. Dormancy intensity can be defined as the percentage of dormant seeds in a sample. Dormancy intensity determination may involve germination at various temperatures with the sensitivity of the test depending on germination temperature (Strand, 1980). Other dormancy intensity tests include hanging grain bundles in a closed room, keeping them moist and evaluating the extent of germination at 7, 10, or 14 days by use of a predetermined scale or alternately by terminating germination after 3-5 days and estimating the amount of germination by an alpha-amylase test (Belderok, 1968). Accurate dormancy determination requires testing at the same maturity stage (Belderok, 1968; Strand, 1980), careful threshing to avoid kernel damage (Strand, 1980), low moisture levels to eliminate water sensitivity (Strand, 1980), careful attention to germination temperatures (Strand, 1980) and standardized storage of lots to be tested (Olsson and Mattsson, 1976; Strand, 1980). Length of wheat dormancy periods increased from 12 to 15 to 50 days as storage temperature decreased from 22° C to 18° C to 2° C, respectively (Olsson and Mattsson, 1976). Noll and Czarnecki (1980) reported accurate dormancy testing could be delayed for up to 4-5 months in wheat by storage at -15° C or by increasing the germination temperature to 30° C. The same varietal ranking for dormancy was

achieved under these conditions as was found immediately after harvest. Results from spike and grain germination tests show that dormancy depends primarily on grain properties while spike type (erect or nodding), density of the head, and the presence of glumes have a minor effect on germination behavior (Belderok, 1968).

Breeding populations can be screened for dormancy in the F_2 or later generations by planting seeds in moist sand in a bench and allowing the seeds to germinate. Upon termination of the germination the seeds are sifted from the sand (Strand, 1980). The germinated seeds are discarded and the dormant seed saved and dried for subsequent planting. Selection pressure is regulated by the dormancy level in the population at the time of the test and the percentage of the population expected to survive the test. The level of dormancy of the population is a function of climatic conditions during ripening, the genetic level of dormancy, storage temperature, and the time from maturity to testing. The test is from 5-10 days in duration at temperatures from 10-20° C. The higher temperatures manifest more dormancy and are applied to low dormancy material or for a low selection pressure toward nondormancy (Strand, 1980).

Germination percentage obtained under specified conditions is the most commonly used measure of germination. It reveals nothing, however, about the rate or uniformity of germination (Bewley and Black, 1978). Another commonly used germination measure is the germination rate which