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The Role of Seed Coats in Seed Viability

YASSEEN MOHAMED-YASSEEN[1], SHERYL A. BARRINGER[2], WALTER E. SPLITTSTOESSER[1], AND SUZANNE COSTANZA[3]

[1]*University of Illinois*
Department of Horticulture
S–406 Turner Hall
1102 South Goodwin Avenue
Urbana, Illinois 61801, U.S.A.

[2]*Ohio State University*
Department of Food Sciences & Technology
122 Vivian Hall
2121 Fyffe Road
Columbus, Ohio 43210–1097, U.S.A.

[3]*St. John's University*
Department of Natural Science
300 Howard Avenue
Staten Island, New York 10301, U.S.A.

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I. Abstract

The seed coat is the seed's primary defense against adverse environmental conditions. A hard seed coat protects the seed not only from mechanical stress but also from microorganism invasion and from temperature and humidity fluctuations during storage. Phenolic compounds in the seed coat contribute to seed hardness and inhibition of microorganism growth. During germination, the seed coat protects the seed from hydration stress and electrolyte leakage.

Resumen

La cubierta de la semilla es la defensa primaria contra el medio ambiente adversa. Una cubierta dura proteja la semilla no solo de tensión mecanica sino también de ana invasion de microorganismos y de cambios en temperatura y humedad durante almacenaje. Los phenolics en la cubierta de la semilla contribuyan a la dureza de la semilla y la inhibición de crecimiento de microorganismos. Durante la germinación, la cubierta proteja la semilla de la tension de hidratación y del escape de electrolytes.

II. Introduction

Seeds are fundamentally important to people, not only because they constitute the chief method of plant propagation, but also because they provide an important food (Duffus & Slaughter, 1980). Seed storage, critical for germplasm preservation, is important to farmers, breeders, and industries interested in seed processing and commercial trade. Seeds of many species, however, lose viability after short periods of storage, making their species prone to extinction and causing extensive losses. Strategies for preservation of seeds are often used, including in situ conservation in natural preserves and ex situ preservation usually in gene banks (Roos, 1988). In situ maintenance allows continued evolution of a species, but large areas of land are needed and species are still in danger of extinction as a result of natural disasters. Preservation ex situ is thus a preferred method for genetic conservation.

The seed coat is the seed's primary defense against adverse environmental conditions: It provides the embryo and other seed components with physical and chemical barriers to unfavorable conditions and protects the seed against infection and deterioration caused by microorganisms. The importance of the seed coat in seed longevity has long been noted (Becquerel, 1906). Seeds with hard seed coats are generally long-lived (Bass, 1980; Priestley, 1986)—hard seeds such as *Canna, Lotus,* and *Lupinus* have survived for over 500 years (Bass, 1980). Weak structure and cracks in the seed coat permit fungal infection, causing seed deterioration (Christiansen et al., 1960; Christiansen & Justus, 1963; Mayne et al., 1969; Halloin, 1986b; Mohamed-Yasseen et al., 1993).

III. Definition of Seed and Seed Coat

A seed is a structure that contains at least an embryo and usually a supply of stored nutrients (Mauseth, 1988). The seed coat is the outer coat of seed derived from the integument. It is also called the "testa." The term "seed" used in this context is meant to include dry fruit, in which the ovary wall has become a part of the dispersal unit. Only seeds which can be stored in a dry state, known as orthodox seeds (King & Roberts, 1979), will be discussed in this paper.

IV. Definition of Seed Aging

Seed aging can be defined as the progressive deterioration of the structures and functions of the seed over time (Mohamed-Yasseen, 1991). This ultimately leads to the death of the organism. Roberts (1972) defined seed aging as an irreversible degenerative change which is generally considered to represent the death of the seed. The term "senescence" is frequently used for deteriorative changes in whole plants or plant organs that tend to occur at well-defined points in their life cycle and normally result in death (Priestley, 1986). Whereas senescence represents endogenously controlled degenerative processes leading to death, aging includes a wide array of passive or nonregulated, degenerative processes driven primarily by exogenous factors (Leopold, 1975). This passive degeneration is a consequence of lesions ("wear and tear") that accumulate over time (Leopold, 1975; Nooden & Leopold, 1978). Aging does not in itself necessarily cause death, but it may decrease resistance to a variety of stresses and otherwise increase the probability of death. Because the biochemical nature of senescence and aging is not precisely known, it is premature to attempt to define these processes more exactly or to draw a fine line between them (Nooden, 1988).

Longevity of a seed is the period from seed maturation until seed death (Ellis & Roberts, 1981). It has long been known that the greater the moisture content and storage temperature of orthodox seeds, either singly or in combination, the shorter the longevity (Roberts, 1973).

V. Seed Coat as Physical Defense

Hard seed coats lessen or alleviate habitual stress during and after harvest. Susceptibility to mechanical injury (injury that occurs during harvest and/or transport) is a significant factor in seed storability (Bass, 1980). Corner (1976) reported that larger seeds live longer than smaller seeds and suggested that longer viability was due to a hard seed coat. Mechanical injury reduces the storability of seeds (Burns et al., 1958; Metzer, 1961; Mamicipic & Caldwell, 1963; Arnold, 1963; Moore, 1972; Almedia & Falivene, 1982). Removing the seed hull from sorghum (Haferkamp et al., 1953; Kalashink & Naumenko, 1979; Esbo, 1954, 1960), delinting of cotton seeds (Flores, 1938; Simpson, 1946), dewinging of pine seeds (Barner & Dalskov, 1954; Huss, 1956; Kamara, 1967), and scarification of alfalfa and *Pelargonium* seeds (Graber, 1922; Battle, 1948; Bachthaler, 1983) all reduced viability. Scarification of onion (*Allium cepa* L.) seeds increased electrolyte leakage, caused fungal infection, and reduced germination rate (Splittstoesser & Mohamed-Yasseen, 1991; Splittstoesser et al., 1994).

An impermeable or hard seed coat provides the surest protection a seed can have against fluctuations in humidity and temperature which could damage the embryo or encourage growth of microorganisms (Christiansen et al., 1960; Christiansen & Justus, 1963; Mayne et al., 1969; Halloin, 1986b). A negative correlation was found between cracked seed coats and the thickness of seed coats of soybean (Yasue & Kinomura, 1984). Soybean with ruptured or shrunken seed coats (Singh & Setia, 1974), shrunken sweet corn (Yarchuk, 1966; Yarchuk & Leizerson, 1972; Styer et al., 1980; Schmidt & Tracy, 1988), and wrinkled peas (Adamova, 1964) deteriorated faster than smooth ones. Wrinkled seeds (Fig. 1) were found in all ten cultivars of onion, a short-lived seed, obtained from different sources (Mohamed-Yasseen & Splittstoesser, 1990a; Mohamed-Yasseen et al., 1991).

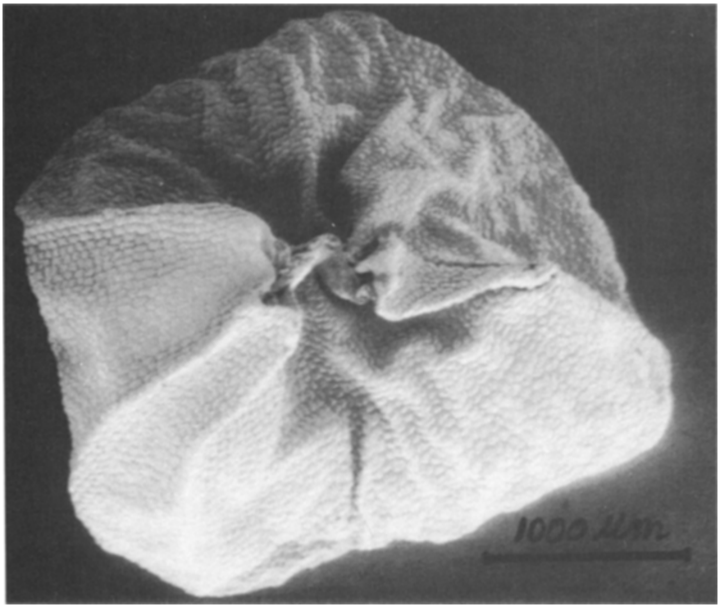

Fig. 1. Typical seed coat shrinkage in onion seed; bar = 1000 µm.

The position of the embryo in relation to seed coats and other seed components may also affect seed storability. Peanut seeds lose viability due to damage to the radicle tip, which is located under the seed coat and protrudes outside the cotyledons (Roberts, 1972). Onion seed, a short-lived seed, has its root tip located just under the protruding part of the seed coat (Fig. 2). This fragile position makes the root tip vulnerable to mechanical damage and an easy target for microflora found growing abundantly over the hilum arena (Splittstoesser & Mohamed-Yasseen, 1991). Similar observations were noted by MacKay et al. (1970) on a short-lived rye seed.

Studies on the genetics of longevity in soybean (Kueneman, 1983) and corn (Cal & Obendorf, 1972; Scott, 1981) suggest that the trait for longevity is inherited maternally. Potts (1978), Potts et al. (1978), and Minor and Paschal (1982) showed that hardseededness in soybeans greatly reduces field deterioration under simulated tropical conditions. Unfortunately, a hard seed coat is considered an undesirable characteristic, as this high level of resistance to deterioration is accompanied by resistance to germination. This trait is also undesirable if seeds are to be consumed.

VI. Nutrient and Electrolyte Leakage

In addition to forming a direct physical barrier to microorganism infection, the seed coat indirectly protects the seed from infection by restricting the diffusion of nutrients from the seed into the soil. Nutrients, such as carbohydrates and amino acids, diffuse into the soil when seeds imbibe, and they contribute to the proliferation of pathogenic

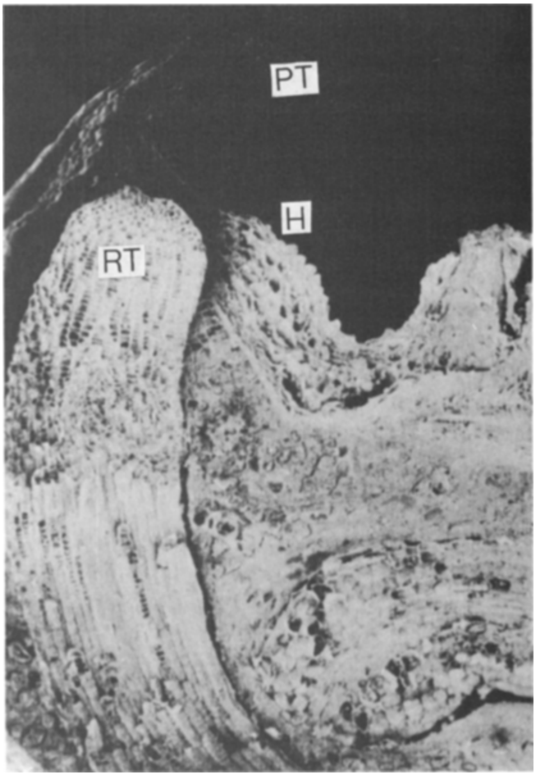

Fig. 2. SEM micrograph of bisected onion seed showing the location of root tip (RT) adjacent to the hilum (H) and the protruding tissue (PT) of seed coats.

fungi immediately around seeds (Mayne et al., 1969; Halloin, 1986b; Mohamed-Yasseen, 1991; Splittstoesser & Mohamed-Yasseen, 1991). The leakage of nutrients and elements during seed germination has been demonstrated in such diverse crops as barley and wheat (Abdul-Baki & Anderson, 1970; Abdul-Baki, 1980; Petruzzelli et al., 1982). The increase in electrolyte leakage in deteriorated seeds is an indication of membrane deterioration, which Parrish et al. (1982) suggested is the primary factor in the aging process. However, the relationship between electrolyte leakage and aging may not be assumed to occur in all stages of seed aging—Abdul-Baki and Anderson (1970) reported that leakage of sugar from aged barley seeds was unrelated to early stages of deterioration. Scarified onion seeds, which gave higher germination rates than aged seeds, had greater electrolyte leakage (Splittstoesser et al., 1994).

VII. Seed Coat and Microflora

The involvement of fungal infection in seed aging was demonstrated by Christensen and Kaufman (1969), Christensen (1967, 1972, 1973), and Neergard (1977), showing that seeds infected with fungi deteriorated faster during storage than uninfected seeds.

Bacterial infection plays a secondary role in seed deterioration (Angelo & Ory, 1983; Cherry, 1983). The interaction of seed microflora with seed components has been extensively studied in wheat and cotton (Anderson et al., 1970; Anderson & Baker, 1983; Halloin, 1983; McGee, 1983; Mills, 1983, 1986).

A major way in which microorganisms damage seeds is the production of exocellular enzymes and toxins. Among the enzymes produced, cellulases, pectinases, amylases, lipases, proteases, and nucleases are likely to be of major importance. Aflatoxins and mycotoxins, produced by fungi in the *Aspergillus* group, reduce seedling elongation, inhibit chlorophyll synthesis, inhibit various enzymes, and degenerate the endoplasmic reticulum (Halloin, 1986a). Microorganism infection may also cause an increase in electrolyte leakage, which is apparently due to the damage of the cell membrane and seed integuments.

Microorganisms that infect seeds can enter through natural openings, such as the micropyle, or through wounds or cracks. Some fungi penetrate directly through thin seed coats (Neergard, 1977; Herman, 1983). Scarification or punctures of the pericarp of corn seed allow rapid invasion by storage fungi, leading to reduced germination (Christensen & Lopez, 1963).

Scanning electron microscopy (SEM) is a powerful tool for detecting seed coat cracks and microflora in asymptomatic seed, and could be useful in determining seed storability (Mohamed-Yasseen & Splittstoesser, 1990a, 1990b). SEM observation of asymptomatic and symptomatic seed coats of onion revealed the presence of fungal infection (Mohamed-Yasseen & Splittstoesser, 1990b), which was more pronounced in seed cracks (Fig. 3) and natural openings of seeds (Fig. 4). When onion seeds were surface sterilized and plated on potato agar medium, SEM observation showed the presence of *Aspergillus* (Fig. 5), *Penicillium, Rizopus,* and other unidentified fungi. The control of seed-borne disease using such fungicides as benomyl, captan, and thiram improved seed viability of several species (Maude, 1972; Moreno-Martinez & Mandugano, 1985).

VIII. Seed Coat and Hydration Stress

Seeds with damaged seed coats imbibe water rapidly. Their embryos are, as a result, susceptible to imbibitional injury (Powell & Matthews, 1978; Duke & Kakefuda, 1981; Tully et al., 1981; Duke et al., 1983; Oliveria et al., 1984; Duke et al., 1986; Powell et al., 1986), which can be exacerbated by cold soil. Cracking or removal of lima bean seed coats increases the rate of water uptake and solute leakage at low temperatures (Pollock & Toole, 1966). This imbibitional chilling injury can be diminished by either retarding water uptake with polyethylene-glycol or applying a thin coat of lanolin to the seed coat (Priestley & Leopold, 1986). Soybean seeds that were insensitive to chilling injury could be rendered susceptible through scarification of the seed coats (Tully et al., 1981).

IX. Phenolic Compounds in the Seed Coat

Seeds with pigmented seed coats, in a number of crops, have a longer storage life. Beans (*Phaseolus vulgaris* L.) with pigmented seed coats perform better in cold soils than white seeds (Dikson, 1971). Soybeans with black seed coats imbibe water more slowly than unpigmented soybeans and show less imbibitional chilling injury (Tully et al., 1981). Chickpeas with dark seed coats store better than lighter-colored varieties

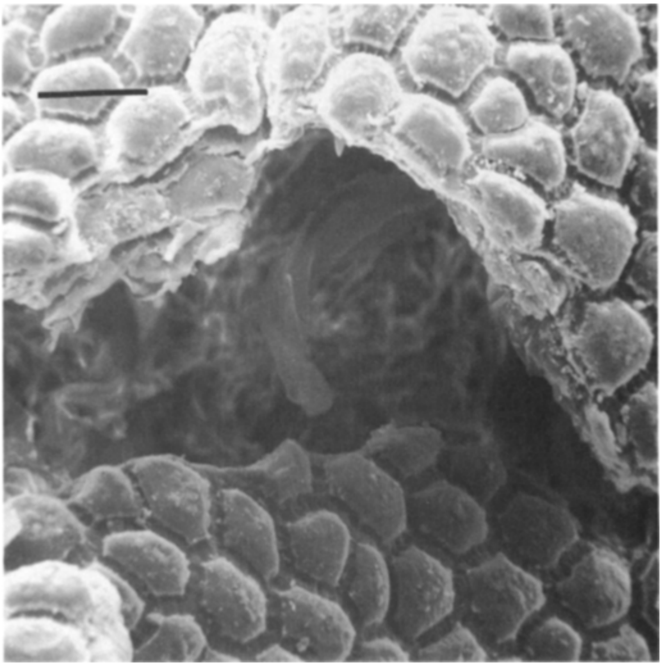

Fig. 3. Typical seed coat cracks in onion seed; fungal infection can be seen inside it. Bar = 100 μm.

(Gvozdeva & Zhukova, 1971). Van der Maesen (1984) has also noted that pale-seeded chickpeas are shorter lived than those with thicker, harder coats. In a study of inheritance of water permeability in soybean seed, Shahi and Pandey (1982) found a linkage between seed coat color and seed impermeability. Yellow seeds are more permeable than black seeds. Black-seeded soybeans were more resistant to deterioration in high-humidity conditions than were pale varieties, a trait that was associated with a decrease in fungal growth (Starzinger et al., 1982). Roos (1984) similarly reported that white-seeded lines of snap bean (*Phaseolus vulgaris* L.) deteriorated faster than colored ones under unfavorable storage conditions. Seed coat cracking and leakage was greater in white-seeded than black-seeded snap bean, making them more susceptible to attack by soil pathogens (Prasad & Weigle, 1976). Powell et al. (1986) found that differences in seed vigor in dwarf French were associated with the color of the testa. French beans with white seed coats were more sensitive to imbibitional damage than colored seeds. Colored snap beans had greater seed coat dry weight and thickness and less permeability to water than did white seeds (Kannenberg & Allard, 1964). Lignin comprised about 15% of the total weight of colored lima bean seeds but only 1% of white seeds (Wyatt, 1977). Red wheat is also found to be longer lived than white wheat (Khoroshailov & Zhukova, 1973). Drying seeds in the absence of oxygen resulted in seed coats without pigmentation and high permeability to water, while drying in the air or in oxygen resulted in colored seed coats and less permeability (Mayer & Poljakoff-Mayber, 1989). It was suggested that the resistance of colored seed coats to deterioration was a result of impermeably thick seed coats created by the

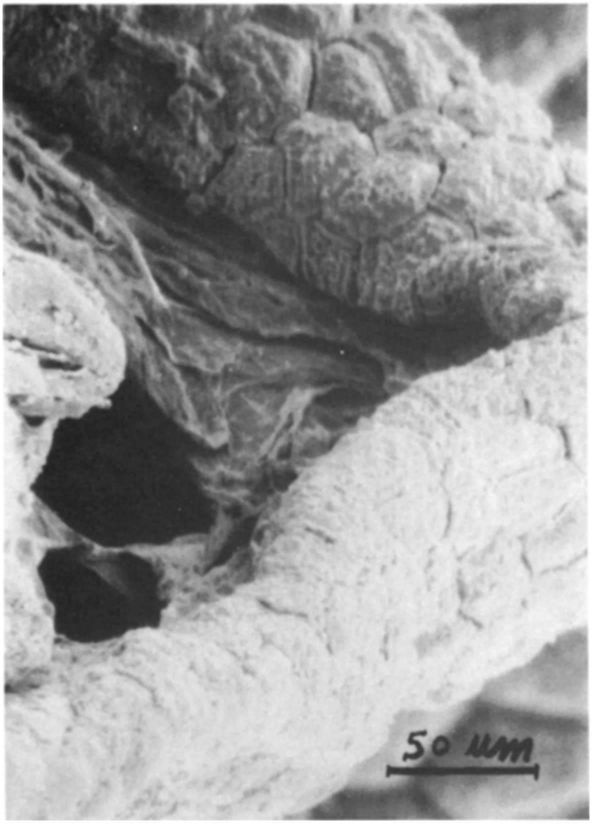

Fig. 4. Typical fungal infection in natural opening, shown with the micropyle.

oxidation of phenolic compounds by polyphenoloxidase or peroxidase. It was found recently that the majority of peroxidase activity in soybean seed was localized in the seed coat, suggesting that it may play a role in the hardening of the seed coat (Gillikin & Graham, 1991). Colored snap bean seeds are often more resistant to mechanical damage (Dikson & Boettger, 1976). Such resistance means less nutrient leakage from the seed during germination and, as a consequence, less attack by soil-borne fungi (York et al., 1977).

Phenolic compounds in seed coats play another role in seed longevity and act as a chemical defense against microorganisms. Monomeric phenols, which are more soluble than polymerized forms, may act as inhibitors to fungal growth and seed germination under humid storage conditions (Halloin, 1986b). Peas with dark seed coats were less susceptible to root rots than peas with light-colored coats; this resistance was due to anthocyanins (Clauss, 1961). Electrolyte leakage, however, may antagonize the inhibitory effect of phenols. Kraft (1977) observed that the anthocyanin delphinindin was inhibitory to *Fusarium*, but this inhibition was overcome by the

Fig. 5. Fungal infection with *Asperigillus* over onion seed coats.

presence of glucose. Wrinkle-seeded peas were more susceptible to preemergence rotting than were smooth-seeded types, but the resistance did not persist once the seed coats were broken, resulting in sugar leakage (Clauss, 1961).

X. Selection for Hard Seed Coat for Longevity

Few attempts have been made to select for long-storage traits in seeds (Halloin, 1986b). Bird (1982) developed the multi-adversity resistance system to select for resistant cottonseed. The selected cottonseed showed reduced leakage (Halloin, 1986b) and resistance to field weathering. Peanut seed has developed resistance to *Aspergillus*, due to better seed coat integrity (Mixon & Rogers, 1973; Taber et al., 1973). The water-soak method to select for impermeable seed coats was held to be effective in increasing storability of crimson clover and cottonseed (Bennett, 1958; Christiansen & Justus, 1963). Storability of a population of corn seeds was improved by successive selection through accelerated aging (Scott, 1981). The hot-water

(>50°C) technique was initially developed and applied as an alternative to accelerated aging for predicting storability (Bourland & Welch, 1985; Furbeck et al., 1989). The hot-water procedure (70°C, 4 min.) (Splittstoesser et al., 1994) is more rapid than either the water-soak or accelerated-aging method and can be used to screen a high volume of seeds within a few minutes. Seeds treated with hot water are nearly free from microorganism contamination, a problem always found with the accelerated-aging method, even when a fungicide and bactericide were used (Bahattacharyya et al., 1985; Halloin, 1986a). The hot-water treatment is widely used to control both external and internal seed-borne pathogens including fungi, bacteria, and nematodes (Ventura & Garrity, 1987). The treatment, however, creates mechanical stress to seed coats (Brant et al., 1971). For example, with onion seeds (Splittstoesser et al., 1994), only 10% of the seeds remained viable after hot-water treatment; but these seeds may contain hard seed coats and be free from cracks and an occluded hilum. The surviving plants are presently being evaluated to determine if second-generation plants inherited hard seed coats.

XI. Conclusion

The seed coat plays an important role in seed longevity since it provides the primary defense against harmful microorganisms and unfavorable environmental conditions. Cracked seed coats permit electrolyte leakage, which encourages the growth of microorganisms. Weak seed coat structures also permit rapid water uptake, leading to imbibitional injury. Seed coat color helps increase mechanical resistance of seeds through polymerized phenol, and acts as a chemical defense against microorganisms through soluble phenolic compounds.

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