# Seed enhancements

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# Abstract

Seed enhancements may be defined as post-harvest treatments that improve germination or seedling growth, or facilitate the delivery of seeds and other materials required at the time of sowing. This definition includes three general areas of enhancements pre-sowing hydration treatments (priming), coating technologies and seed conditioning Pre-sowing hydration treatments include non-controlled water uptake systems (methods in which water is freely available and not restricted by the environment) and controlled systems (methods that regulate seed moisture content preventing the completion of germination) Three techniques are used for controlled water uptake priming with solutions or with solid particulate systems or by controlled hydration with water These priming techniques will be discussed in this paper with reference to methodology, protocol optimization, drying and storage. Coating technologies include pelleting and film coating, and coatings may serve as delivery systems. Seed conditioning equipment upgrades seed quality by physical criteria. Integration of these methods can be performed, and a system is described to upgrade seed quality in Brassica that combines hydration, coating and conditioning. Upgrading is achieved by detecting sinapine leakage from nonviable seeds in a coating material surrounding the seeds. Seedcoat permeability directly influences leakage rate, and seeds of many species have a semipermeable layer. The semipermeable layer restricts solute diffusion through the seed coat, while water movement is not impeded Opportunities for future seed enhancement research and development are highlighted

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Abbreviations PEG = polyethylene glycol, RH = relative humidity

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### Introduction

'Seed enhancement' is a term used in industry and, more recently, in the scientific literature to describe beneficial techniques performed on seeds after harvest, but prior to sowing Enhancements are 'valueadded' techniques performed on a given seedlot. The term 'seed enhancement' is often used synonymously with seed priming, but should be considered an umbrella term, which encompasses many pre-sowing techniques including priming. A functional definition of seed enhancement is post-harvest treatments that improve germination or seedling growth or facilitate the delivery of seeds and other materials required at the time of sowing. This definition includes three general methods pre-sowing hydration treatments (priming), coating technologies and seed conditioning

The objective of this paper is to provide an overview of these three general methods. These methods are not mutually exclusive, and several techniques can be combined in a particular sequence to obtain an additive effect. Integration of selected individual steps can be used to upgrade seed quality, and an example of this integration is described Review articles are cited to provide expanded literature coverage on each general method, and opportunities for future research and development are highlighted

### Pre-sowing hydration techniques

#### Regulation of water availability to seeds

Seeds require water, oxygen and a suitable temperature for germination Water uptake tollows a triphasic pattern with an initial rapid uptake phase

245

known as imbibition (phase 1), followed by a lag period (phase 2) and then a second increase in water uptake associated with seedling growth (phase 3) (Bewley and Black, 1994, Bradford, 1995) Seeds are desiccation-tolerant during phases 1 and 2, but frequently become intolerant during phase 3 Each phase of water uptake is controlled by water available to the seeds

Several methods have been described to regulate water availability to seeds both as a liquid and in vapour phase (see review by Khan, 1992) This paper deals with pre-sowing treatments using a liquid phase that allow seeds to enter phase 1 or 2 of water uptake Pre-sowing hydration techniques can be grouped into two categories depending on whether water uptake is non-controlled or controlled Non-controlled water uptake includes those methods in which water is freely available to seeds and not restricted by the environment Therefore, water uptake is governed by the affinity of the seed tissue for water Common techniques include imbibing seeds on moistened blotters or soaking seeds in water Soaking by submerging seeds in water can be performed with or without aeration (Thornton and Powell, 1992) Since water is not limited, seeds eventually germinate, assuming that seeds are viable, not dormant, oxygen is available and a suitable temperature is employed Therefore, the process must be arrested at a specific time in non-controlled water uptake systems to prevent the onset of phase 3

In controlled water uptake, water availability or water potential is regulated, thus preventing the completion of germination (seeds entering phase 3) Water potential is the algebraic sum of the osmotic, matric and pressure potentials There are three methods to limit water uptake priming with solutions, priming with solid particulate techniques and drum priming by controlled hydration Priming with solutions relies on the regulation of seed water uptake by control of the imbibing medium water potential In most liquid systems, there is an excess of solution volume to seed mass so the water potential of the environment (priming solution) remains fairly constant throughout the process The addition of inorganic salts, mannitol or glycerol to water decreases the osmotic potential of a solution, and these solutes may be used alone or in combination for priming The water potential of prepared priming solutions may be determined by thermocouple psychrometry calibrated against known NaCl solutions (Lang, 1967) Another chemical commonly used to regulate water potential is polyethylene glycol (PEG) 8000 The water potential of PEG solutions can be calculated (adapted from Michel, 1983, Hardegree and Emmerich, 1990)

$$\Psi = 0.130[PEG]^2 T - 13.7[PEG]^2$$

where [PEG] is the concentration of PEG expressed as grams per gram of  $H_2O$ , and *T* is the temperature in °C The calculated water potential is in MPa High molecular weight PEG molecules primarily regulate the water potential through matric rather than osmotic forces (Steuter *et al*, 1981) Thus PEG should be considered a matricum rather than an osmoticum

Solid particulate systems are also used to increase seed moisture in a controlled system Seeds, solid particulate and water are the three basic components Water is initially distributed throughout the seed and solid carrier, and water is largely held by the solid carrier Seeds take up water from the solid carrier until equilibrium occurs. The water potential of the solid particulate environment is determined by the physical and chemical properties of the solid carrier Specific materials can regulate total water potential by osmotic, matric or a combination of both factors (Khan, 1992)

Controlled hydration involves allowing a specific quantity of water directly to contact the seeds in order to achieve an elevated moisture content. The weight of water needed to increase a given seed mass to a desired moisture content can be calculated.

$$Wt_{H_2O} = Wt_1(MC_1 - MC_1) (100 - MC_1)^{-1}$$

where  $Wt_{H_2O}$  is the weight of water to add to the initial weight of dry seed (Wt<sub>1</sub>), MC<sub>1</sub> is the moisture content of the initial seed sample, and MC<sub>1</sub> the desired or final seed moisture content. Seed moisture content is expressed as a percentage on a fresh or wet weight basis. Water could be added to seeds at one time in a container and then seeds allowed to imbibe Alternatively, water could be added over time to allow seeds slowly to achieve the desired seed moisture content.

### Priming technology

A plethora of terms has been used in the literature to describe various pre-sowing techniques Priming with solutions has been termed liquid priming or simply 'priming' The term 'osmoconditioning' has also been used, it emphasizes the concept of using osmotic solutions to improve seed and seedling performance (Khan, 1992) This paper will use the general term 'priming' for these liquid treatments and will specify if PEG or other solutions are employed 'Solid matrix priming' and 'matriconditioning' are two terms that have been used to describe solid particulate systems (Khan, 1992) Conditioning in this context should not be confused with seed conditioning used to make physical separations of seeds (described later) A largescale method to perform the controlled hydration method is termed drum priming (Rowse, 1996)

The controlled hydration method is conducted by adding a calculated amount of water to a given seed sample of known moisture content using the formula above The senior author has used this technique for increasing the seed moisture content in laboratory studies On a commercial scale, controlled hydration by drum priming elevates seeds to the desired moisture level by applying given amounts of water with time (Rowse, 1996) The duration of the drum priming depends on the absorptive characteristics of the particular species and seedlot, and on the desired seed moisture content of the finished product (Mauromicale and Cavallaro, 1995) In European models, the priming drum sits on top of a scale, which determines seed weight increases created by a constant influx of water (Rowse, 1996) The scale must be sensitive enough to determine the incrementally small increases in weight caused by the addition of water in comparison with the weight of the apparatus itself. After the seeds have achieved the desired weight (moisture content), the hydration phase is completed Hydrated seeds can then be incubated for a desired period of time required for priming

Another drum priming system controls seed hydration by the time interval and volume of water application (Warren and Bennett, 1997) A preset volume of water is injected during each cycle as regulated by a timer attached to a solenoid Dividing the total water volume required by the preset water injection volume establishes the number of cycles. The seed absorptive pattern of the particular species and cultivar must also be known, so that the time set between injections allows no free water to be left before the next injection. After the seeds have attained the desired water content, they can be maintained in this condition for a specific period of time to complete the priming process.

Considerable research has been performed on the development and testing of various protocols for liquid, solid particulate and drum priming Two literature reviews illustrate the efficacy of liquidpriming techniques on a number of species (Bradford, 1986, Parera and Cantlitte, 1994) In solid particulate systems, the quantity of solid particulate required per given mass of seeds is dependent on the waterholding capacity of the material and the ability to remain friable throughout the process Therefore, the solid particulate acts as a means initially to hold large quantities of water, yet to give up this water freely to the imbibing seed The proportion of seed to solid particulate to water is determined on an empirical basis Two reviews provide background on the development of and results from solid particulate priming systems (Khan, 1992, Parera and Cantliffe, 1994) In the case of drum priming, once a desired seed moisture for optimal priming is determined, then large-scale applications of controlled hydration become purely the application of engineering Again, the choice of temperature and duration must still be performed on an empirical basis

The critical parameters controlled and manipulated during priming treatments are the water potential ( $\Psi$ ), temperature (T) and duration (t) A model has been used to predict the efficacy of a priming protocol to accelerate the rate of germination (Bradford and Haigh, 1994, Cheng *et al*, 1997) The model is similar to a thermal time or degree-days model, where the product of the amount by which the  $\Psi$  and T exceeds the minimum  $\Psi$  (termed  $\Psi_{min}$ ) or T (termed  $T_{mun}$ ) at which advancement will occur is multiplied by the duration of treatment ( $t_p$ )

### Hydrothermal priming time [(MPa)(°C)(h)] = $(\Psi - \Psi_{mun})(T - T_{mun})t_p$

Investigations on tomato (Lycopersicon esculentum L) seedlots tested whether the hydrothermal priming time model accounts for the effects of a range of priming treatments on subsequent germination rates (Cheng et al, 1997) Five tomato seedlots were primed at two temperatures (15 and 20°C), three water potentials (-10, -15 and -20 MPa PEG solutions)and six selected durations The effects of water potential and duration of treatment on germination rates following priming could be accounted for in all lots by a hydropriming time model For most lots, the effect of priming temperature could also be included as thermal time, but some lots showed relatively little response to temperature Overall, the accumulation of hydrothermal priming time during the treatment was linearly related to the subsequent germination rate, and a general model accounting for over halt of the variation in germination rate responses to priming was developed

### Post-treatment effects

Post-treatment processing, including drying and storage of primed seeds, has received little attention This is a significant oversight as the beneficial effects of the priming can be lost by improper post-treatment handling For example, drying certain lettuce (*Lactuca sativa* L) seedlots to below 10% moisture after priming can negate the acquired benefit of priming on germination at high temperatures (Weges and Karssen, 1990) Priming of tomato and lettuce has also been shown to accelerate the rate of ageing in comparison with unprimed seeds (Argerich *et al*, 1989, Tarquis and Bradford, 1992) Therefore, primed seeds may perform poorer than the unprimed seeds depending on the duration and conditions of storage

The vast majority of technical papers on priming deal with methodology to improve the subsequent germination and performance of the primed seeds Rarely are drying procedures even reported, except when noting that seeds were dried under 'ambient' conditions A review on dehydration after priming provides some general findings (Parera and Cantlifte, 1994)

The effect of time and extent of drying has been studied on imbibed perennial ryegrass (Lolium perenne L) seeds (Debaene-Gill et al, 1994) Seeds were imbibed on blotters at 26°C, and visible germination was first observed atter 40 h A loss in the uniformity of germination was measured if seeds were imbibed for 40 h and dried at -150 MPa (32% relative humidity (RH)) However, imbibing seeds for 40 h and then redrying at -4 MPa (97% RH) had no detrimental effect on subsequent germination rate The slower germination rate from seeds imbibed for 40 h and dried to 32% RH was negated by first drying at -4 MPa for 24 h before drying at -150 MPa Further research was conducted to evaluate the importance of drying rate on imbibed small-seeded vegetable seeds (Allen, 1997) Seeds of two cultivars each of tomato, carrot (Daucus carota L), and pepper (Capsicum annuum L) were hydrated in water and then dried at different rates over saturated salt solutions The rate and percentage of germination upon subsequent rehydration were measured Rapid dehydration of imbibed carrot and pepper seeds resulted in delayed germination upon rehydration, which often completely eliminated the germination advancement associated with hydration treatment. In contrast, tomato seeds appeared to be largely unaffected by drying rate The choice of 24 h at -4MPa was based on data for perennial ryegrass seeds and was not determined experimentally for each vegetable seedlot tested Thus, more optimum drying treatments could conceivably be developed for these seedlots While dehydration at -4 MPa removed some water, it is important to note that this drying treatment did not return seeds to the water content of air-dry seeds (see Figure 1 in Debaene-Gill et al, 1994) For situations that require drying seeds back to the original water content before sowing, exposure to lower water potentials would be required even if the first drying stage occurred at higher water potentials

After drying, primed seeds are generally stored for a period of time before sowing The storage environment can have a profound effect on primed seed efficacy and longevity A review of literature on storage of primed seeds (Parera and Cantliffe, 1994) reveals beneficial, neutral and deleterious effects of priming after storage These inconsistent results may be partially attributed the storage conditions (temperature and RH) and time of storage Storage studies were performed on primed and unprimed tomato seeds aged at 42°C and 92% RH for up to 15 days (Hacisalihoglu, 1997) Prior to ageing, primed seeds germinated faster than unprimed seeds, however, no differences in germination rate were measured between treatments aged for 6 days Primed seeds germinated more slowly and exhibited a lower final germination percentage than unprimed seeds, if aged for longer than 6 days In this study, 6 days of ageing was considered the transition point between the beneficial effects of priming and the deleterious effect of ageing

# Seed coating technologies

Seeds vary greatly in size, shape and colour. In many cases, seed size is small or irregular, making singularization and precision placement difficult. In addition, seeds should be protected from a range of pests that attack germinating seeds or seedlings. Seedcoating technologies can be employed in both situations they can facilitate mechanical sowing to achieve uniformity of plant spacing, and can act as a carrier for plant protectants, so materials can be applied in the target zone with minimal disruption to the soil ecology and environment

Two seed-coating methods will be discussed in this pelleting and tilm-coating section Working definitions have been developed in the seed industry for several seed-coating technologies (Butler, 1993) pelleting is defined as the deposition of a layer of inert materials that may obscure the original shape and size of the seed, resulting in a substantial weight increase and improved plantability, while film coating retains the shape and the general size of the raw seed with a minimal weight gain. The coating should result in a more or less continuous coating, which eliminates or minimizes product dust-off Both coating methods mav contain polymers, pesticides, biologicals, identifying colourants or dyes and other additives

## Pelleting

The pelleting operation is generally performed on a batch basis and uses an inclined drum or coating pan (Scott, 1989, N1, 1997) The speed varies with pan diameter and ranges from 10 to 35 rpm Another method used to form pellets makes use of centrifugal systems (Freund Industrial Co) Pelleting has been a labour-intensive operation that requires skilled operators and generally long working hours The pelleting process has become automated, and computer-controlled coating equipment has been described (Scott et al, 1997) In each case, seeds are coated with a combination of binder (adhesive) and tiller (bulking agent) Commercial coating tormulations usually are blends of binders and tillers, and are tormulated as drv powders. The powder is sitted on to the seeds during the coating operation, and water is misted on The water allows the

formation of the pellet and also activates the adhesive Pelleting is a wet operation, so pellets must be dried at the completion of the coating process

A number of materials have been used as binders and fillers (Scott, 1989, Ni, 1997), earlier reviews on this subject are cited by Taylor (1997) The amount of tiller used determines 'build-up' and is expressed as a ratio of filler to seed (w/w) Pellet build-up varies by crop, large or more-spherical seeds generally require less material to form a spherical end-product Pellet build-up has been reported to be 21, 4 to 91, and 17 to 351 tor sugarbeet (Beta vulgaris L), onion (Allium cepa L), and lettuce, respectively (N1, 1997, Taylor et al, 1997) Small-seeded species such as Nuotiana and Petuma require much larger build-ups, 100 to 1501 (N1, 1997) The physical nature of the filler directly attects the pellet weight or density. The density of lettuce pellets was determined from six sources (pelleting companies) and ranged from 0.8 to 1.7 g cm<sup>-3</sup> (Taylor et al, 1997) The desired pellet density is determined by sowing requirements (Hill, 1998) Low density pellets are used with vacuum-drum seeding equipment for greenhouse production, while higher density pellets are required for held sowings The highest density pellets are used in high-speed sowing operations to prevent bouncing after pellets are dropped from the metering device to the soil, while an intermediate density pellet can be used for slower tield planting operations Other considerations for choice of pelleting formulation are economics (seed value) and germination requirements or difficulty of a particular species

There have been improvements in the pelleting process and formulations made by the seed industry. The pellets should not retard the germination rate or reduce the percent stand in comparison with the non-pelleted control. The pellet may act as a barrier to oxygen diffusion, thus affecting germination (Sachs *et al*, 1981). This problem has been overcome by the addition of oxygen-liberating compounds into the pellet or modification of pellet breakdown after sowing Alkaline earth and other peroxide-containing compounds have been used to release oxygen, and CaO<sub>2</sub> has been commonly employed in pellets (Langan *et al*, 1986). Pellets have been developed that fracture in one plane during the early phases of water uptake, a commercial development termed 'split-pellet'.

The pellet can also serve as a delivery system for other materials required at the time of sowing In particular, plant protectants required at high loading rates can be applied during the coating After the first layers of coating materials have been applied to the seed, the active ingredients, formulated as dry powders, can be applied in subsequent layers Final coating applications with filler material encapsulate the plant protectants and prevent worker exposure to the seed treatments This application, termed pellet loading (Hill, 1998), avoids direct contact of the plant protectants with the seed surface. This physical separation avoids phytotoxicity caused by high concentrations of particular plant protectants. Pellet loading has been used successfully for onion seeds for application of fungicides and insect growth regulators. The pellet contains the fungicide Pro-Gro, a mixture of thiram and carboxin, at 20 g a 1 kg seed<sup>-1</sup> and the insect growth regulator, Trigard (Cyromazine) at 50 g a 1 kg seed<sup>-1</sup> (Taylor and Eckenrode, 1993)

#### Film coating

Film coating is a method adapted from the pharmaceutical and contectionery industries for uniform application of materials to seeds. The film-forming formulation consists of a mixture of polymer, plasticizer and colourants (Halmer, 1988, Robani, 1994), and formulations are commercially available that are ready-to-use liquids or prepared as dry powders (Ni, 1997). Application of the film-forming mixture results in uniform deposition of materials on each seed with little variation among seeds (Halmer, 1988). Like pelleting, the formed film may act as a physical barrier, which has been reported to reduce the leaching of inhibitors from the seed coverings and may restrict oxygen diffusion to the embryo (Duan and Burris, 1997).

Laboratory-scale tilm-coating methods and equipment have been developed. Unlike pelleting, ventilation is required to dry the aqueous tormulations sprayed on to the seeds during the coating operation A standard pelleting pan has been adapted for application of film-coating polymers, and drying is achieved by applying forced warm air into the coating pan (Taylor and Eckenrode, 1993) A smallscale, fluidized bed seed-coating apparatus has been described with controlled air velocity and temperature (Burris et al, 1994) Film coating is routinely performed in vented or perforated pans on a largescale basis either on a batch or continuous systems (Halmer, 1988, Robani, 1994) The introduction of a continuous process vented-drum coating machine by Coating Machinery Systems (Huxley, Iowa) has expanded the use of film coating. This equipment is capable of continuous application of various polymer systems and components, while providing drying capacity to prevent the seed from hydrating Capacities vary from 100 to 10 000 kg h<sup>-1</sup> depending on seed type, and target weight gain. In addition, conventional low-volume seed-treating equipment has been used for application of tilm-coating polymers, however, less material can be applied since drying capability may not be present (N1, 1997)

Film coating is versatile as a coating system or a component of a coating system Colourants provide an aesthetic appeal to the seeds, serve to colour-code

different varieties and increase the visibility of seeds after sowing Film-coated seeds have better flow characteristics in the planter (Hill, 1998) due to reduced friction between seeds Film coating and pelleting may be combined in two different processes Film coating may be performed as the final step after pelleting to provide a dust-free pellet An intermediate between film coating and pelleting is termed encrustation This hybrid technique provides some build-up, enhances plantability and is less expensive than pelleting

Film coating provides an ideal method for the application of chemical and/or biological seed treatments (Taylor and Harman, 1990, Taylor *et al*, 1994, McGee, 1995) Relatively high loading rates of plant protectants can be applied with film coating However, a spatial separation between the plant protectants and seed surface is not obtained as described for pellet loading A major impetus for using film coating is to reduce the exposure of workers to chemicals from treated seed

### Seed conditioning

The harvested seed from the field is seldom pure and therefore contains undesirable materials, including poor quality seeds that should be removed from the seedlot Conditioning has two major objectives (Copeland and McDonald, 1995) the first is the removal of contaminants, including other crop seeds, weed seeds and inert material, from the seedlots Atter this operation only seed of the same crop species should be retained. The second objective is to upgrade or eliminate the poor quality from the high quality seeds. Poor quality seeds may be in the form of immature, damaged or off-sized seeds. Upgrading can be considered as a seed enhancement as the germination and seedling growth characteristics of the seedlot are further improved.

Physical characteristics such as size, shape and weight are commonly exploited in seed conditioning (Brandenburg, 1977), while other parameters such as colour and surface texture are used as secondary techniques (Brandenburg and Park, 1977) Magnetic separators and the use of magnetic fluids have also been studied to remove contaminants (Berlage and Brandenburg, 1984) The scope of this paper does not permit a review of this equipment, seed technology books should be consulted that describe and illustrate seed-conditioning equipment used in commercial 1979, Copeland (Thompson, agriculture and McDonald, 1995, Desai et al, 1997) Seed conditioning is most commonly performed on dry seeds, and is generally conducted in a series of steps or operations using different equipment that perform specific operations For example, removal of drought-damaged soybeans was tacilitated by conditioning with three different machines used in series (Riesse *et al*, 1991) The efficiency and overall efficacy of seed conditioning have been improved by automation of existing equipment Microprocessor control of standard seedconditioning equipment has been shown to be superior to manual control and further decreases loss of high quality seed (Shvy and Misra, 1987, 1992)

Further enhancement of seed quality may be obtained if specific physical criteria that are associated with seed quality can be exploited Colour-sorting technology was described two decades ago (Brandenburg and Park, 1977), but improvements have since been made in optical systems, microprocessors and detection systems. For example, a colour sorter that senses light reflectance from 360° has been described (Lee et al, 1998) Light detectors can also quantify light reflectance in the UV and NIR regions as well as the visible, thus expanding the capability of traditional colour sorters Currently available colour sorters can be modified to sort on the basis of fluorescence, providing another tool to upgrade seed quality (Taylor et al, 1993a) Light reflectance was measured with a reflectance spectrophotometer from white-seeded beans (Phaseolus vulgaris L) (Lee et al, 1998) The greatest percent light reflectance difference between off-coloured and white seeds was measured from 275 to 450 nm with a 360 nm maximum Few differences were detected in the near intra-red (>750 nm) Off-coloured white-seeded beans had a lower percent germination than seeds with white seed coats. and colour sorting could remove the off-coloured seeds from the lot

A novel instrument has been developed at Iowa State University that combines computer imaging with ultrasound technology (Shvy and Misra, 1994) In this manner, the instrument can discriminate soybean [Glycine max (L) Merr] seeds of different size, shape, texture, colour, lustre, mass and hardness, and those with physical damage Data from the combined equipment were analysed rapidly with microprocessor control The method is not destructive and has potential for scale-up for specialized seed conditioning The ultrasound technology was also used to detect asymptomatic diseased soybean seeds The peak values of the ultrasound waves decreased, while the slope and the bandwidth values increased with increasing levels of disease infection A significant correlation was also found between the sound wave parameters and the germination percentage of the diseased seedlot (Walcott et al, 1994)

### Integration of seed enhancement technologies

Seed enhancement technologies provide unique tools that may be used directly or indirectly to facilitate the detection and removal of poor quality seeds from a seedlot Pre-sowing hydration elevates seed moisture content and, in the process, reactivates cell function Cell membrane integrity can be indirectly assessed by measuring solute leakage at the whole-seed level Seed coatings provide a system to hold compounds in close proximity to individual seeds Colour sorting can rapidly separate seeds based on differential light reflectance or fluorescence emission The following discussion will highlight the development of an integrated system that employs, in sequence, presowing hydration, seed coating and conditioning to enhance seedlot quality in *Brassica* 

Early observations reported that non-viable Brassica seeds leaked a fluorescent compound during the early phases of germination, while viable seeds did not (Taylor et al, 1988) The predominant fluorescent substance was the alkaloid sinapine, a natural reserve compound formed during seed development Sinapine is yellow at high pH ( $\geq 10$ ), and sinapine leakage can easily be visualized from single seeds An 8-h viability test was developed by measuring sinapine leakage from individual seeds (Hill et al, 1988) Sinapine leakage was found to be more accurate in predicting the success of germination than the electrical conductivity test Collectively, sinapine leakage tests can be used to estimate the ability of seeds to germinate and can be performed in a single working day

A system was developed to detect sinapine leakage on a single-seed basis to upgrade seed quality (Taylor et al., 1991) Seeds were first hydrated by soaking in water for 4 h or were primed in PEG solutions for 24 h Sinapine leakage from non-viable seeds was greater than that from viable seeds The treshly hydrated seeds were then coated with a filler that contained a finely ground cellulose The cellulose acted as an adsorbent trapping the sinapine leachate in the coating Coated seeds were dried and then sorted under UV light into two tractions fluorescent and non-fluorescent Refinements ın the coating formulation have been made (Lee et al, 1997), with a 11 build-up being optimal for sinapine detection In general, the non-fluorescent coated seeds had a higher germination percentage than the non-coated control (Taylor et al, 1991, Lee et al, 1997) Seed conditioning was performed to separate the fluorescent from the non-fluorescent coated seeds by using colour sorting equipment (Taylor et al, 1993a) A colour sorter, modified for fluorescence sorting, was able to remove a high percentage of fluorescent coated seeds with a minimal loss Overall, germination was improved in selected lots representing cabbage (Brassica oleracea L, Capitata group), broccoli and cauliflower (Biassica oleracea L, Botrytis group)

An expansion of the techniques and principles used to exploit sinapine leakage in upgrading *Brassica* 

seed quality was envisioned for other crop seeds Two major obstacles prevented the full utilization of this approach First, sinapine is found exclusively in species of the Brassicaceae (Cnuciterae) (cited by Taylor et al , 1993b) Therefore, leakage of other water-soluble compounds present in a wide range of seeds was sought Both amino acids and sugars were later studied in relation to seed ageing in cabbage, leek (Allium vorrum L), onion, tomato and pepper (Lee et al, 1995, Taylor et al, 1995) Non-germinable seeds of cabbage had the greatest leakage rate followed by leek and onion, while tomato and pepper leaked negligible amounts of compounds This finding is indicative of the second major obstacle the permeability of seed coats to solute diffusion Light and electron microscopy was used to study seed-coat permeability Semipermeability is defined as the ability of seed coverings to allow water uptake and gas exchange, while solute diffusion is restricted or prevented. It was shown that a semipermeable layer exists as the innermost layer of the seed coat, just adjacent to the endosperm, in leek, onion, tomato and pepper, while a semipermeable layer was not found in Brassica (Beresniewicz et al, 1995b) In lettuce and muskmelon (*Cucumus melo* L), a semipermeable envelope surrounds the embryo and has been shown to prevent leakage trom non-viable embryos (Hill and Taylor, 1989, Welbaum and Bradford, 1990) The chemical nature of the semipermeable layer was attributed to cutin in leek and onion, while suberin was found in tomato and pepper (Beresniewicz et al, 1995a) The perisperm envelope of muskmelon seeds has a waxy outer layer over a second callose layer ( $\beta$ -1,3-glucan) The callose layer was shown to be responsible for the semipermeability (Yim and Bradford, 1997) Leakage measured from non-germinable leek and onion seeds was related to the presence of cracks in the seed coat (Beresniewicz et al, 1995c) Thus, while amino acids, sugars and ions could be quantified in leakage studies, leakage was not a reliable measure of germinability due to permeability of the seed-covering tissues

Seed coat permeability was also shown to be important in the ability accurately to assess Brassica germinability using sinapine leakage False negatives (those seeds that did not germinate and did not leak) were the major source of errors in predicting seed germinability (Lee and Taylor, 1995) The leakage rate from intact seeds varied by seedlot, and those slow-toleak lots had a higher percentage of false negatives (Lee and Taylor, 1995) The efficacy of upgrading seed by exploiting sinapine leakage is compromised if the seed coat restricts leakage from non-viable seeds Thus, the non-viable seed would go undetected since the solute leakage would not diffuse through the seed coat into the coating Though a semi-permeable laver was not found in Brassica, chemical analysis of the seed coats has shown that cutin content was related to seed coat permeability (Lee *et al*, 1997) Cutin may have retarded the diffusion of sinapine through the seed coat Methods to enhance leakage rate were sought, and soaking seeds in a dilute solution of NaOCl was the most effective method to enhance leakage rates (Taylor *et al*, 1993b)

### A reflection on the past and a look into the future

The use of seed enhancement technologies is not new in agriculture, and earlier practices have been described for priming, coating and upgrading seed quality by differences in specific gravity Evenari (1980/81) documents practices known in antiquity that are essentially identical in principle to current priming techniques, such as the recommendation of Theophrastus (*c* 372–287 BC) to 'presoak cucumber seeds in milk or water to make them germinate quicker' The following is taken from Ohver de Serres (1600), *Le Théâtre d'Agriculture et Mesnage des Champs* 

There is another clever trick to make the planted seed more profitable than usual, it is very subtle but has been proven The grain to be used as seed, either wheat or rye or barley, will be put to soak in water tertilized with the best manure you can find The procedure is as follows fill two-thirds of a tank with the manure and then make up the filling with river water, which will stay two days, then, remove the manure which would have left its strength and power in the water so fertilized, the strength and power will be transferred to the seed soaked in the water, the seed taken out of the water must be dried in the shade, as soon as dried, sow it without waiting that the seed becomes parched, which would remove its new power By that means, the seed so prepared will yield, amazingly, eighteen or twenty for one In the following year it you wish such an increase with the same variety, you will have to repeat the same procedure as that strength of the manure does not last more than one year

Another description from the same text illustrates the use of density separation in water (de Serres, 1600)

In addition to having selected the right cultivar, and in order to improve your crop, the seed ready to be planted will be thrown into water, the soundest part, as the heaviest, will fall to the bottom, the other intertile part, the lightest one, will float on the surface, from where it will be collected with a skimmer The good seed, slightly dried but still wet, will be sown thus it will emerge quickly avoiding the danger of being eaten away by soil pests or delaved by unfavourable weather conditions

Seed coating was being practised as much as 2000 years ago (cited by Ni, 1997) The ancient Chinese coated rice seeds in mud balls to anchor the seed in a flooded rice paddy. The coating eliminated the

problem of seed drifting when seeds were sown on the surface of the flooded paddy In conclusion, many of our so called 'modern seed enhancements' have a historical basis dating back hundreds to thousands of years ago (Evenari, 1980/81) The principles and concepts have changed little with time Improvements have been gained through better technology guided by a better understanding of seed biology

Further research and development are needed on many technological aspects of all seed enhancements, as well as a better understanding of seed biology in the hydrated condition There is no single issue on which research should focus, rather, several opportunities exist to study the many components of these technologies Pre-sowing hydration methods have received the greatest attention by researchers and will continue to be of both practical and fundamental interest Solid particulate systems and controlled hydration are more recent developments than liquid priming techniques These newer systems eliminate the disposal of large quantities of solutions used in liquid priming and are generally compatible with the addition of biological control and other strategies for pest management (Taylor and Harman, 1990) The merits of these more 'environmentally safe' systems should justify commercial adoption and utilization of these technologies However, both systems are patented (Eastin, 1990, Rowse, 1991), imposing legal restrictions on their commercial usage Patenting and licensing will become even bigger issues in the future as protection will be sought by both the public and private sectors for the development of new technology that has economic value. These issues are also being tested in other areas of plant biology and agriculture In addition to the legal aspects of ownership of intellectual property, there are several other seed technology topics that need further examination

Developing a priming protocol should consider the priming step, drying and longevity Lack of efficient methods to predict seedlot performance to priming without a series of empirical treatments is a major limitation Optimization of a priming procedure requires continued effort, and seed factors may need to be considered along with environmental conditions in models or other protocols The post-treatment drying techniques have been largely overlooked, only a tew publications exist on this subject. The benefit of priming may be lost in fact, primed seeds may perform worse than nonprimed seed Simple engineering principles of seed drying are well understood, but the three parameters of drying (temperature, relative humidity and air flow) have been largely ignored with respect to drying hydrated seeds (Ptasznik and Khan, 1993) Preconditioning seeds by first drying to -4 MPa prior to drying at -150 MPa retained the rapid subsequent germination rate (cited earlier in this paper) Other methods that

employ a mild water stress after priming may improve shelf-life (Bruggink *et al*, 1997)

Hydration treatments generally reduce shelf-life, but in some cases longevity is enhanced. The faster ageing of primed seed is of commercial concern and poses many interesting questions. Is the reduced longevity due to acceleration of general ageing processes or are specific deterioration reactions being promoted? Recent research has shown that priming alters the relationship between seed moisture content and relative humidity (Sun et al, 1997) Changes in biophysical aspects of water binding may create a cascade effect on several ageing mechanisms Priming may also enhance specific ageing mechanisms such as the Maillard reactions (Wettlaufer and Leopold, 1991), since priming can initiate mobilization of reserves, creating a larger pool of reducing sugars for formation of Maillard products with proteins Alternatively, metabolism of oligosaccharides and non-reducing sugars during priming could reduce the ability of the dried seeds to form stable 'glasses' that retard deterioration reactions (Leopold *et al*, 1994) The existence of 'repair' mechanisms has been suggested for the past two decades, but concrete data are rare For example, short-term aerated soaks have been shown to improve shelf-life, but the underlying mechanisms are not well understood (Powell et al, 1997) Definitive studies are needed to examine if repair is occurring and, if so, under what conditions

In summary, developing a complete priming protocol must address the balance between germination advancement and reduced longevity Attempting to attain the most rapid germination from priming must be weighed against the enhanced liability of this pre-sowing treatment Post-treatment drying and shelf-life problems should be tackled to ensure consistent performance over a wider range of storage conditions

The challenges for technology to enhance seed performance provides an opportunity for more indepth studies on physiological and biochemical changes that occur during seed treatments Priming provides a practical basis for physiologists to study early events in germination prior to seedling growth Controlled hydration and solid particulate systems are convenient tools to hydrate seeds fairly precisely to a desired level from their initial seed moisture content to full imbibition Research is needed on cellular events associated with hydration, but should also be directed to dehydration phenomenon Physiological, biochemical and biophysical markers are needed to detect seed These markers would have practical quality application in quality control programmes, as tools to develop and monitor hydration events and possibly for upgrading seed quality

Integration of seed pre-sowing treatments with other systems will expand Seed enhancements will

become a component of the larger issues related to pest management and application of other beneficial organisms required at the time of sowing Studies on leakage and seed-coat permeability reveal that seeds of most species do not allow the diffusion of solutes through the seed coat Brassica seeds turned out to be the exception, rather than the rule, by having a relatively permeable seed coat. The uptake of many water-soluble compounds is probably severely restricted during hydration in most species, and the benefits measured after hydration are due primarily to water alone Therefore, priming, in essence, is only controlling water availability to seeds Practical methods are needed to scarify seeds to allow movement of solutes from the embryo to the environment and vice versa. In this process, the scarification method must not damage the embryo Further research is needed to exploit leakage and to find other markers that could be used to detect and upgrade seed quality

Priming and coating are commercially performed in combination on certain crop seeds, and one technique may affect the response of the other Priming overcomes thermodormancy in lettuce, however, primed seeds that were then pelleted exhibited thermodormancy when tested immediately after coating (Valdes and Bradford, 1987) Drying methods are important as they affect the uniformity of drying and subsequent seed performance Fluidized bed drying has been reported to be an effective way of drying primed seeds before tilm coating (Buljaski et al, 1992) Drying conditions determine the final seed moisture content, and quality control programmes require methods for measuring moisture content of coated seeds Water activity instrumentation has been reported as a non-destructive way of measuring the water status of pelleted or film-coated seeds (Taylor et al, 1997) Collectively, post-treatment processing, in particular the drying conditions, has a major impact on seed enhancement efficacy

Coatings will continue to be an important delivery system for materials required at the time of sowing The application of plant protectants, chemical and/or biological, for efficient and economical pest management will be a major emphasis Research will be devoted to developing specific crop seed/plant protectant combinations as the mode of actions of plant protectants become more defined and crops are engineered for chemical compatibility Film coating or the application of tilm-coating polymers will be driven forward by environmental and worker safety issues The overall impact is positive, as seed coating can reduce exposure of workers to seed treatments Seed coating should be developed as a management tool to be an integral component to enhance seedling establishment under a wide range of environmental conditions Limited research has been performed on

A G Taylor *et al* 

the addition of hydrophobic compounds for sowing into wet soils (Taylor *et al*, 1992), hydrophilic compounds in coating for sowing into dry soils and temperature-sensitive coatings to regulate the onset of imbibition at a desired soil temperature (cited by Ni, 1997) Specific formulations are needed to broaden the range of environmental conditions in which a desired stand may be established Thus, the coating improves the chances of successful germination and seedling establishment under field conditions

Research on seed conditioning has declined Presowing hydration and pelleting are generally restricted to the high-value crop seeds for which the added cost of these treatments can be justified. In contrast, seed conditioning is used on virtually all agronomic, horticultural and many other seeds Research is needed to explore various physical criteria that may be related to seed quality Future advances will be achieved by team efforts of seed technologists working with engineers Appropriate equipment could be developed or existing equipment modified to facilitate rapid sorting in order to improve seed quality The system of coating hydrated Brassica seeds to detect sinapine leakage was a novel system that included seed conditioning Continued research and development of systems are needed that are both economical and practical for a range of crop seeds

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256