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**CHANGES IN GERMINATION PERFORMANCE
AND HYDROLYTIC ENZYME ACTIVITY IN
WHEAT SEEDS (Triticum aestivum L.)
CAUSED BY AGEING AND
PRE-SOWING TREATMENTS**

A thesis presented in partial
fulfilment of the requirements
for the degree of
Doctor of Philosophy
in **Seed Technology**
at **Massey University**
Palmerston North
New Zealand

SURESH NATH

1991

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ABSTRACT

Three pre-sowing hydration-dehydration treatments were evaluated for their capacity to protect or repair wheat seeds stored under two different sets of artificial ageing conditions (accelerated ageing at 100% RH, 40°C or controlled deterioration at 15% SMC, 35°C). Although similar losses in germination capacity and decreases in radicle emergence rates occurred under both ageing conditions, differences with respect to the physiology of ageing were highlighted by changes in seedling growth and seed leakage. For example, increases in seed leakage observed during storage at 15% SMC were not found at 100% RH.

Longer hydration treatments (either 24 h at 15°C in water or 20 h at 20°C in -0.37 MPa PEG solution, followed by drying) improved the vigour of unaged seeds, but treated material deteriorated rapidly in storage compared to untreated controls. In contrast short hydration treatments (2 h at 25°C followed by drying) offered some protection of germinability during subsequent storage but did not affect the vigour of unaged seeds. When seeds were treated after storage, longer hydration periods were effective in producing substantial invigoration of viable deteriorated seeds (measured by evaluating T50 or seedling growth) compared to little or no improvement by short hydration treatments. These results support earlier suggestions from work on tomato seeds that losses in seed vigour and viability are not necessarily a continuum of the same deteriorative sequence.

The mechanisms of protection of germinability by short hydration treatments were not clear. Small decreases in T50's of unaged or aged seeds as a result of these treatments were due to leakage of germination inhibitory substances. However, the rapid germination of unaged and improved responses from aged seeds caused by longer hydration treatments suggested advances in germination processes and repair activity under these conditions. This aspect was pursued in further detail by studying changes in the hydrolytic metabolism of wheat seeds using the 20 h PEG treatment.

Although the starchy endosperm of treated seeds showed some indications of protein degradation, there were no changes in proteolytic activity (determined as 'Azocoll' hydrolysing activity at pH 6.8) as a result of ageing or pre-sowing treatment after storage. However, there were some indications of loss of control over proteolytic activity in seeds subjected to treatment before storage. Severe damage to membrane permeability in these seeds appears to be a post-mortem event as this was only found in samples showing drastic losses in seed germinability.

Pre-sowing treatment caused a buildup of germinative α -amylase activity in unaged but not in aged seeds, although both showed similar radicle emergence rates. Quick resumption of α -amylase production during subsequent imbibition by treated seeds, irrespective of ageing, suggests that components involved in de novo enzyme synthesis are tolerant to desiccation in wheat seeds. Increased α -amylase activity in treated seeds or its maintenance during subsequent storage, surprisingly did not cause damage to stored starch. There was no relationship between increased α -amylase activity and early radicle emergence.

The ageing-induced delay in germinative α -amylase production appeared to be due to delayed gibberellin synthesis by the aged embryo. Pre-sowing treatment of seeds after storage effectively decreased the lag period for enzyme production in deteriorated seeds. Ageing effects on aleurone were characterised by investigating changes in the responsiveness of embryoless half seeds to gibberellic acid with respect to α -amylase production in vitro. Ageing of seeds caused a significant reduction in aleurone enzyme production. These changes were at least in part, reversed by pre-sowing treatment of aged seeds.

Abbreviations: h = hours; PEG = polyethylene glycol; RH = relative humidity; SMC = seed moisture content; T50 = time to 50% radicle emergence.

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ABBREVIATIONS

AA	accelerated ageing
ABA	abscisic acid
ATP	adenosine triphosphate
CD	controlled deterioration
cv	cultivar
d	days
DNA	deoxyribonucleic acid
dwt	dry weight
EC	electrical conductivity
EDTA	ethylenediaminetetraacetic acid
Fig	figure
g	gram
GA	gibberellic acid
GADA	glutamic acid decarboxylase activity
h	hours
HMW	high molecular weight
HPLC	high-performance liquid chromatography
ISTA	International Seed Testing Association
IUPAC-IUB	International union of pure and applied chemistry and the International union of biochemistry
kD	kilo Dalton
LSD	least significant difference
M	molar
ME	moisture equilibration
mg	milligram
mM	millimolar
MPa	megapascal
mRNA	messenger RNA
mwt	molecular weight
nm	nanometer
OD	optical density
p	probability of non occurrence of the event
PEG	polyethylene glycol

PGR	plant growth regulator
Poly(A)⁺ RNA	polyadenylated RNA
PST	pre-sowing treatment
RH	relative humidity
RNA	ribonucleic acid
SDS-PAGE	sodium dodecyl sulphate-polyacrylamide gel electrophoresis
Sec	section
SH	sulphydryl
SMC	seed moisture content
SE/SEM	standard error of mean
T50	time to 50% radicle emergence
tab	table
v/v	volume/volume
w/v	weight/volume
μS	microsiemen
α	alpha
β	beta
~	approximately equal to
<	less than
>	greater than