Genotype x moisture interactions and wheat germinability

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Abstract

During wheat grain development, two humidity levels were compared to examine whether germinability and other aspects of ripening were altered. Six diverse wheat genotypes, with varying grain colour and putative dormancy, (Sonora 64A, Timgalen, Pembina, Gamut, Sherbati-Sonora, Karamu) were studied under two phytotron ripening environments [both at 12-18°C; one at -1.0 kPa vapour-pressure deficit (to give 28.7 % relative humidity (RH) at 12°C and 51.6 % RH at 18°C), and the other more humid at -0.4 kPa (to give 71.5 % RH at 12°C and 88.2 % RH at 18°C)], using tagged ears over entire maturation sequences. Grain moisture, mass, standard germination, dormancy-bypassed germination, and pigmentation were described, and compared via non-linear functions. Higher humidity delayed harvest-ripeness by up to two weeks, but there was much variation across genotypes. Embryonic maturity also was delayed: even failing to appear at all (in the time-frame of the experiment) for a third of the cultivars. Higher humidity greatly reduced dormancy in those lines where there was enough germinative maturity to measure. No association between graincoat redness and dormancy was apparent under humid ripening. The timing of pigmentation was relatively stable across the two moisture regimes. Grain growth was accelerated in half the genotypes, but delayed in the others. Plant breeding for resistance to sprouting-damage under humid grain-fill conditions will require special attention.

Additional key words: *dormancy; harvest ripeness, maturity, red grain coat*

Introduction

Seed maturation at some perceived level of ripeness depends on the complex interactions of many processes which are influenced by genotype and ripening environment (Bewley and Black, 1994; Gordon and Cross, 1999). While this complexity can be dissected (Flintham and Gale, 1996), the final understanding of the germinability state of mature seed requires knowledge of the interactions of several attributes in multiple environments using diverse genotypes (Wellington and Durham, 1958; Gordon and Cross, 1999). This has become very apparent when attempting to manipulate seed germinability in a consistent way, such as resistance to pre-harvest sprouting damage in wheat, where levels of "dormancy" vary across ripening environments (Stoy, 1983).

Detailed descriptions of wheat grain maturation have revealed considerable genotype x environment interaction for dessication, colouring, embryo maturity, dormancy, growth and germination (Gordon and Cross, 1999). In that work, the environmental effects were differences in ripening temperature. Many inconsistencies in germinability across genotypes and environments were revealed, and this led to the suggestion that breeding for sprouting resistance should be treated as a multiple character selection problem, perhaps by using selection indices. The present paper takes this study a step further by investigating the effects of air-humidity during pre-harvest ripening on the germinability of diverse wheat genotypes.

Materials and Methods

The gene-pool included common wheats (*Triticum aestivum* L.) of diverse origin, both white- and red-grained, and with various levels of germinability. These were: Gamut (white, highly germinable), Timgalen (white, variable sprouting sensitivity), Sherbati-Sonora (white mutant of Sonora 64, variable dormancy), Sonora 64A (*R1* gene for redness, *Rht*x gene for reduced height, moderate to low dormancy), Pembina (red, high dormancy) and Karamu (*R1* gene for redness, *Rht1* gene for reduced height/gibberellin insensitivity, high dormancy).

Both phytotron ripening environments were "cool" (18°C/12°C), but they differed in air vapour pressure deficit (vpd) throughout the ripening period, during

which the grains were attached to the plants. The "humid" environment was maintained at 0.4 kPa vpd (which was 71.5 % relative humidity (RH) at 12°C and 88.2 % RH at 18°C), while the "dry" environment was at 1.0 kPa vpd (28.7 % RH and 51.6 % RH at 12°C and 18°C respectively). Photosynthetically active radiation was at 160W/m^2 in both phytotrons. There was a 14h photoperiod with an abrupt change between light and darkness. Temperature changed over a 2h period, equally straddling the light change. Canopy-top air-flow was maintained at 0.3 - 0.5 m/s, with CO_2 concentration at 320-340 ppm. Uniform plants were placed into the phytotron at booting, having been raised previously in a glasshouse as described by Gordon and Cross (1999).

Grain development attributes were collected as described by Gordon and Cross (1999). Ears were sampled seven times during development from previously tagged spikes which had reached anthesis in the same half-day. Grains were removed by hand from the basal florets of spikelets in the lower middle of the spike. This procedure provided undamaged grains of the same age at each sampling time. The day at which dehydration reached 12.5 % grain moisture defined "readiness for harvest", i.e., harvest ripeness (HR). Two germination tests were involved: the standard procedure (measuring the net result of viability, maturity, and dormancy), and a dormancy-bypassed germination (measuring only the results of viability and maturity) (Gordon, 1979; Gordon *et al.*, 1979). Embryo maturity (EM) was defined as the day this latter test reached 95 % germination. Dormancy (DY) was estimated as the difference between the two germination tests on the same day, provided EM was above zero. This study thus distinguished between immaturity and dormancy. No inviability was encountered. Pigmentation (PG) was defined as the day colour formation first occurred in 50 % of the sample. Growth maturity (GR) was defined as the day when grain mass first reached 95 % of its asymptote.

Subsequently, asymptotic sigmoid functions were fitted on the bulked data from three replicates. The best fit was provided by the logistic for moisture, mass and colour (except for mass in cool dry Gamut, which was fitted best by a Richards function), and best-fit was a quadratic logistic for both germination tests. The genotype x moisture interactions have been described from these fits.

Results

The reliability of the individual fitted curves is of central importance, and is shown in Table 1, where the coefficients of determination are presented. Most fits are very satisfactory ($R^2 > 0.85$), with only two fits in the entire data set falling below an R^2 of 0.7. The fitted curves for the white wheats are given in Fig. 1, while those for the red wheats are in Fig. 2. Timings of major events were estimated from the fitted curves, and are shown in Table 2 as days from anthesis.

Harvest ripeness (HR) was delayed in all genotypes by the rise in humidity during grain development. There was considerable genotypic variation for this delay, ranging from 4.3 days for Timgalen to 14.7 days for Sherbati. However, embryo-maturity (EM), a more crucial measure of maturity, varied independently of dehydration. Only for Sonora did HR and EM occur together in both ripening environments. For Gamut, they occurred together in the drier environment, but embryo-maturity did not develop at all (within the experiment's time-frame) in the humid environment. This failure to develop embryo-maturity within the time-frame occurred also for Pembina, but for both environments. However, in a warm dry environment in other work both of these did attain embryo-maturity (Cross and Gordon, 1999).

Table 1. Coefficients of determination (R^2) of fitted functions for the development attributes grain moisture (GM), dry mass (DM), dormancy-bypassed germination (DG), standard germination (SG), and graincoat colour (GC) of six diverse wheat lines in moist air (-0.4 kPa vpd) and dry air (-1.0 kPa vpd) ripening environments.

Genotype	GM	DM	DG	SG	GC
Moist					
Gamut	.941	.992	--[1]	--	.799
Timgalen	.991	.943	.766	.796	.778
Sherbati-Sonora	.974	.937	.836	.830	.742
Sonora 64A	.972	.958	.993	.987	.870
Pembina	.992	.969	--	--	.884
Karamu	.970	.914	.998	.999	.865
Dry					
Gamut	.977	.973	.898	.909	.584
Timgalen	.980	.921	.900	.883	.802
Sherbati-Sonora	.968	.881	.850	.898	.815
Sonora 64A	.962	.924	.900	.886	.904
Pembina	.955	.965	--	--	.853
Karamu	.971	.954	.926	.595	.863

[1] Unable to fit function because of insufficient change within time-frame of the experiment.

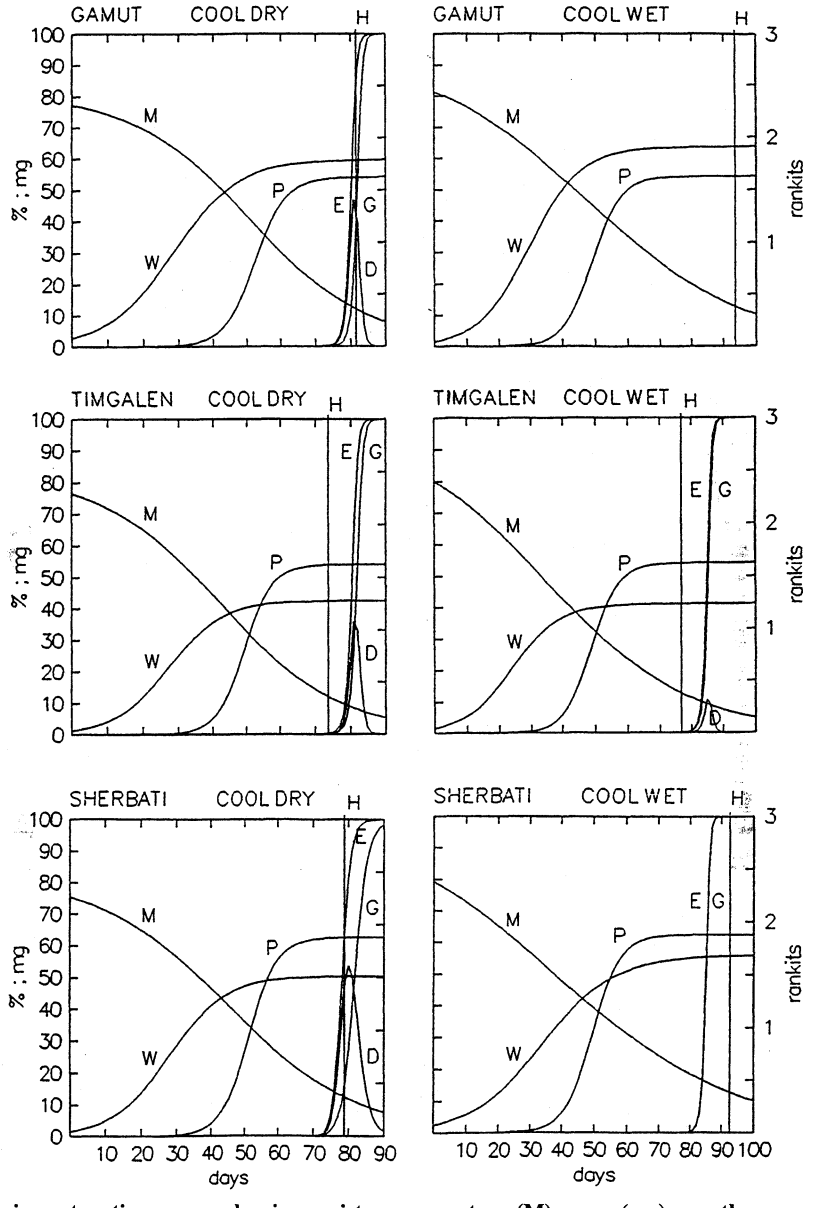

Figure 1. Grain maturation curves showing moisture percentage (M), mass (mg) growth per grain (W), dormancy-bypassed germination percentage (E), standard germination percentage (G), dormancy percentage (D), graincoat colouration (rankits) (P), and harvest ripeness day (H) for three white-grained lines. See text for further details.

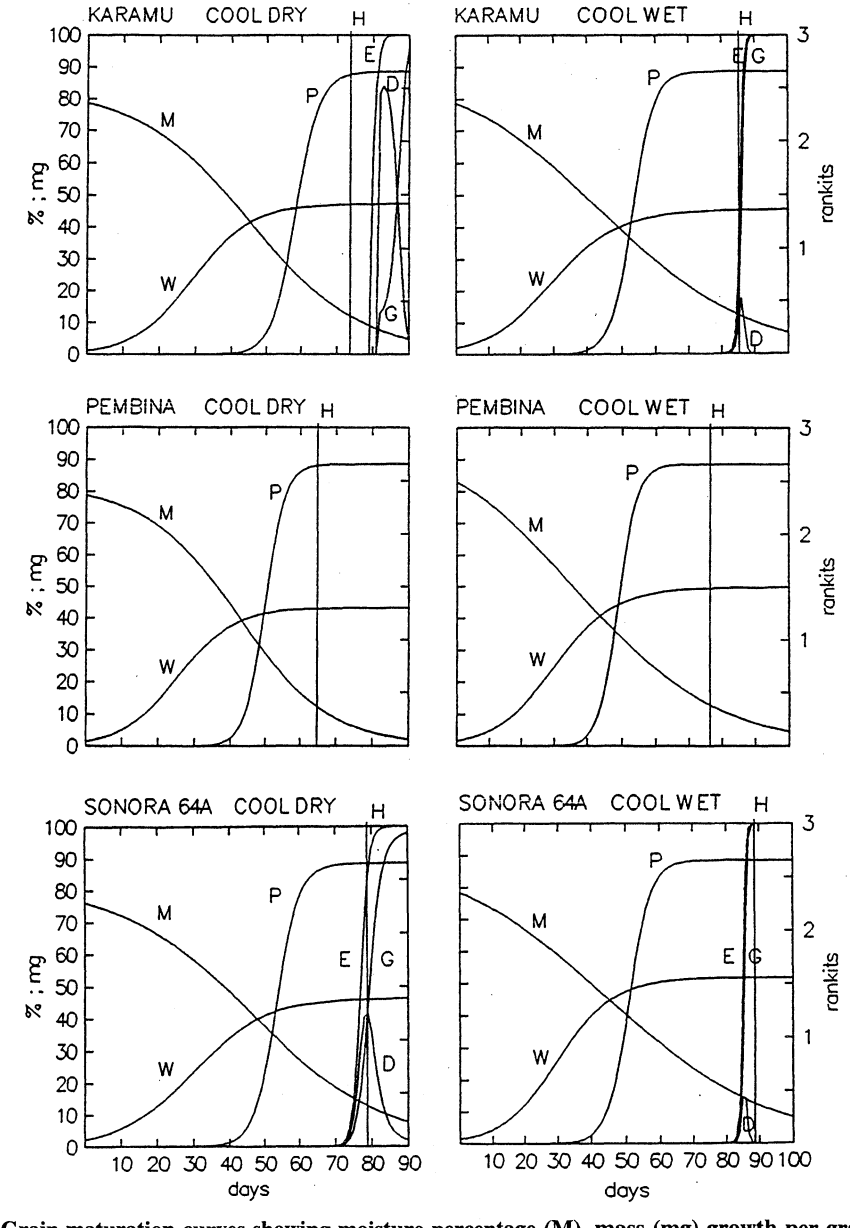

Figure 2. Grain maturation curves showing moisture percentage (M), mass (mg) growth per grain (W), dormancy-bypassed germination percentage (E), standard germination percentage (G), dormancy percentage (D), graincoat colouration (rankits) (P), and harvest ripeness day (H) for three red-grained lines. See text for further details.

For Timgalen and Karamu, embryo-maturity was later than harvest ripeness, but by varying amounts in the two environments. For Sherbati, EM was 3.4 days later than HR in the dry ripening, and 6.4 days earlier in the humid ripening.

As this study distinguished between dormancy (DY) and immaturity, it became apparent that lack of germination in these two (cool) environments was due substantially to immaturity (low embryo-maturity near HR). In cases where no EM was recorded (Pembina and Gamut in the humid environment), dormancy could not be measured. However, in the dry environment, low to moderate dormancy was found for most of the cultivars, the exception being Karamu for which dormancy was high. In the humid environment, dormancy was delayed for most cultivars. The high humidity also reduced the level of dormancy, even for Karamu. For Sherbati, the dormancy in the humid ripening disappeared altogether. Where EM or DY postdated harvest ripeness by several days, the outcome was low standard germination. Thus, Timgalen and Pembina had poor germination in both environments; while Karamu did not germinate in the dry environment but showed a hint of germination in the moist environment. Two other cultivars (Sonora and Sherbati) also had more germination in the humid ripening environment. Conversely, Gamut germinated only in the dry environment.

Table 2. Timings, in days from anthesis, of harvest ripeness (HR), growth maturity (GR), graincoat pigmentation (PG), embryo maturity (EM), and dormancy (DY) for six diverse wheat lines in moist air (-0.4 kPa vpd) and dry air (-1.0 kPa vpd) ripening environments.

Genotype	HR	GR	PG	EM	DY
Moist					
Gamut	94.2	53.2	49.1	--[1]	--
Timgalen	77.2	46.2	48.9	87.1	85.0
Sherbati-Sonora	93.3	67.0	49.6	86.9	--[2]
Sonora 64A	88.9	54.8	51.1	86.7	85.0
Pembina	76.8	56.7	49.0	--	--
Karamu	84.6	58.3	53.3	87.6	86.0
Dry					
Gamut	80.6	56.0	52.4	82.7	81.0
Timgalen	72.9	50.0	49.3	82.9	81.0
Sherbati-Sonora	78.6	51.9	51.3	82.0	80.0
Sonora 64A	79.3	59.2	53.5	80.5	79.0
Pembina	64.4	48.2	49.9	--	--
Karamu	72.9	52.8	58.3	82.1	83.0

[1] Not observed within time-frame of the experiment.
[2] No dormancy found.

Growth maturity (GR) (Table 2) was conspicuously different to the other aspects of maturity (HR and DY). In all cases, it preceded HR and EM by 20-40 days. For two genotypes (Sonora and Gamut), this gap was lengthened considerably (>10 days) by moist ripening. For Sherbati, there was little difference between the two environments. For half of the genotypes (Timgalen, Sonora and Gamut), grain growth began earlier in the moist ripening; but this led to a higher final mass only for Sonora and Gamut. After examining the general patterns amongst attributes, it is clear that growth maturity has little value as an indicator of other measures of maturity (such as germinability and harvest ripeness). Therefore, its common alternative name of "physiological" maturity implies a false generality.

The maturity attribute with most temporal stability across the two environments was colour formation (PG). Under these two "cool" ripening environments, median colouring was around 50 days from anthesis (Table 2), Karamu being about 4 days later. Half of the lines were a few days delayed in the dry ripening, but the rest were very similar in both ripenings. There was no distinction between red and white wheats in these timing patterns: even Sonora and Sherbati coloured at similar times despite the latter being the "white mutant" of the former. These results, in agreement with previous studies (Gordon, 1979; Gordon, 1983; Gordon and Cross, 1999), do not reveal a mechanism linking graincoat redness to dormancy.

Discussion

These results provide insight into the effects of air humidity on the plant-based development of wheat grain. It is clear that grain maturation consists of several separate attributes which develop at different rates in various cultivar-environment combinations. The juxtapositions of these maturity characters changed conspicuously with humidity, and these differences were inconsistent across genotypes: i.e., genotype x moisture interactions existed. Comparing results with earlier work (Gordon and Cross, 1999), it appears that the effects of humidity are less dramatic than the effects of temperature. Higher humidity caused notable delays in dehydration (harvest ripeness) and in embryo maturity (EM). The efficacy of these methods of separating immaturity from dormancy is highlighted.

There has been a long-term interest in the relationship between red graincoat and lack of germination in wheat (Gordon, 1979). In this study, the delays in EM did not appear to be associated with grain colour nor the presence of dormancy. Pigmentation timing was the attribute least affected by humid ripening, but the germinability attributes (EM and DY) were highly variable in timing. An earlier hypothesis (Gordon, 1979) suggested that hypo-oxia arising from pigment polymerisation might be linked to ABA formation and dormancy. However, because of the long and variable delays between embryo maturity and pigmentation, this hypothesis is not supported by the results presented here. The only hint of agreement was from the Karamu maturation in the humid environment, where a loss of dormancy was coincidental with the timing between PG and EM stretching to 34.3 days (Table 2).

The changes arising from humid ripening are substantially different to those due to temperature. Generally, higher temperature shortens time-spans for all attributes (Gordon and Cross, 1999), but higher humidity delays some attributes while accelerating others. This suggests that heritabilities under these conditions could be different to estimates currently extant (Gordon, 1987). It is probable, therefore, that a specifically orientated project would be required to breed cultivars resistant to sprouting under such humid ripening environments. Once again, however, it is clear that a multi-variate approach (e.g., using selection indices) would be more beneficial than emphasis on any single character.

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References

Bewley, J.D. and Black, M. 1994. Seeds: Physiology of Development and Germination, 2nd edition. Plenum Press, New York.

Flintham, J.E. and Gale, M.D. 1996. Dormancy gene maps in homoeologous cereal genomes. *In* Seventh International Symposium on pre-harvest Sprouting in Cereals (eds. K. Noda and D.J. Mares), pp 143-149. Japan Center for Academic Societies, Osaka.

Gordon, I.L. 1979. Selection against sprouting damage in wheat. III. Dormancy, germinative alpha-amylase, grain redness and flavanols. *Australian Journal of Agricultural Research* ***30***, 387-402.

Gordon, I.L. 1983. Factor analyses of characters useful in screening wheat for sprouting damage. *In* Third International Symposium on pre-harvest Sprouting in Cereals (eds. J.E. Kruger and D.E. LaBerge), pp 231-238. Westview Press, Boulder, Colorado, USA.

Gordon, I.L. 1987. Combining ability analysis of wheat grain ripening and germinability in two ripening environments. *In* Fourth International Symposium on pre-harvest Sprouting in Cereals (ed. D.J. Mares), pp 157-164. Westview Press, Boulder, Colorado, USA.

Gordon, I.L., Balaam, L.N. and Derera, N.F. 1979. Selection against sprouting damage in wheat. II. Harvest ripeness, grain maturity and germinability. *Australian Journal of Agricultural Research* ***30***, 7-17.

Gordon, I.L. and Cross, R.J. 1999. GE interaction in wheat germinability. *In* Eighth International Symposium on pre-harvest Sprouting in Cereals (ed. D. Weipart), pp 224-234. Association of Cereal Research, Detmold.

Stoy, V. 1983. Progress and prospect in Sprouting research. *In* Third International Symposium on pre-harvest Sprouting in Cereals (eds. J.E. Kruger and D.E. LaBerge), pp 3-7. Westview Press, Boulder, Colorado, USA

Wellington, P.S. and Durham, V.M. 1958. Varietal differences in the tendency of wheat to sprout in the ear. *Empire Journal of Experimental Agriculture* ***26***, 47-54.