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Selecting maize for rapid kernel drydown: timing of moisture measurement

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Abstract

Previous studies have shown that maize ear moisture measured using a modified Electrophysics Moisture Meter model MT808 was highly correlated to kernel moisture and could be used as a selection tool in breeding maize genotypes with faster rates of kernel drydown. Such a tool would need to be standardized for practical and routine use in a breeding program with large numbers of plants. The objective of this study was to determine the optimum time for measuring ear moisture using this meter. In a split-plot design with three replicates in 2007, 2008 and 2009, ear moisture of six inbred lines and eight F₁ hybrids were measured weekly from one to eight weeks post-silking using a modified MT808 moisture meter. To determine if multiple ear moisture readings (EMRs) could be made on the same ear, an additional treatment was added so that all eight readings were made on the same ear. There was a positive correlation between weekly EMRs readings done on separate ears and those done on the same ear, indicating that repeated readings, if desired, could be made on the same ear. Significant genotypic differences in EMRs were found five to eight weeks post-silking. The EMRs at week one, five, and eight could be used to calculate a daily drydown rate (DDR). Maize genotypes (hybrids and inbreds) could be divided into four groups based on their DDRs during development as: high-high, high-low, low-high, and low-low DDRs from weeks one to five and five to eight, respectively. Genotypes with higher DDRs from weeks one to five tended to have overall higher DDRs by eight weeks post-silking. Inbred lines with higher DDRs at either stage expressed this trait in their hybrid crosses. This non-destructive method will improve selection for fast kernel drydown in maize breeding programs, especially in short-season areas.

Keywords: corn, selection, kernel moisture, breeding

Introduction

The drydown rate of maize or corn (*Zea mays* L) kernels after grain-filling plays an important role in a successful harvest, especially in cool, short-season environments (Eckert, 1978; Cross, 1985). The lower the kernel moisture is at harvest, the less the drying and storage costs will be (Hellevang and Reff, 1987; Hellevang, 2004). As kernels develop from the blister stage to the formation of the black layer, which indicates physiological maturity, kernel moisture decreases from approximately 85% to 30% (Schmidt and Hallauer, 1966; Baute et al, 2002). After black layer formation, further moisture loss primarily occurs via physical evaporation through the kernel surface (Kiesselbach and Walker, 1952). Maize hybrids with lower kernel moisture content at black layer are more desirable than those with higher moisture content. Continued loss of moisture from the kernel is related to many factors including: air temperature and humidity (Schmidt and Hallauer, 1966), premature plant death (Troyer and Ambrose, 1971), husk tightness (Troyer and Ambrose, 1971; Hicks et al, 1976), kernel test weight (Troyer and Ambrose, 1971; Cross,

1985), pericarp thickness and permeability (Crane et al, 1959), kernel and ear size and shape (Cross, 1985; Hunter et al, 1979), rate of grain fill (Kang et al, 1986), and plant defoliation (Tollenaar and Daynard, 1978).

In general, the more time a plant has for kernel filling, the higher the yields will be (Daynard and Kannenberg, 1976; Tollenaar, 1977; Cavalieri and Smith, 1985). While earlier flowering hybrids can be selected to facilitate kernel drydown before harvest, this usually results in plants with smaller ears and lower yields (Corke and Kannenberg, 1989; Troyer, 1990). Growers in short-season areas, preferring hybrids with high yield potential, will often select later maturing hybrids than what is recommended for their region. If these intermediate to late maturing hybrids do not flower until August, there is a risk of insufficient time for kernel filling and drydown before cooler fall weather slows development or an early frost occurs (Baute et al, 2002; Lauer, 2004). A desirable hybrid would be one with intermediate to late maturity and a fast kernel drydown rate. Several studies have been conducted on selecting maize genotypes with fast kernel drydown traits (Cross, 1995; Magari et al, 1997; Poneleit and Egli, 1979). Genetic variation

for kernel drydown exists and quantitative trait loci have been identified (Kang and Zuber, 1989; Sala et al, 2006; Wang et al, 2012; Zhang et al, 1996). Selection based on low ear moisture content at a given date after pollination has been reported to be an effective way to predict kernel moisture content at harvest (Cross, 1985; Cross et al, 1987). Cross and Kabir (1989), studying the rate of moisture loss in detached ears in the laboratory, suggested that simultaneous selection for low moisture at 45 days after pollination and for fast relative moisture loss was more effective in producing hybrids with low harvest moisture than using either method alone. These studies used a traditional oven drying method to measure kernel moisture. This destructive technique is not efficient for breeding purposes. To find a simple method of measuring kernel moisture, Kang et al (1978) designed an electronic probe (DC-10 moisture meter) which non-destructively estimates kernel moisture content in field grown plants; this meter was patented by Spry (1990). The DC-10 moisture meter was used to measure the kernel moisture three times at 10-day intervals (between 30 – 60 days after mid-silk) and moisture loss across the three dates was then used to determine the kernel drydown rate. Freppon et al (1992) studied the use of a hand-held moisture meter (Delmhorst JCS-1) to select for low ear-moisture content in a set of S1 plants that had similar pollination dates in the field at 30 days after pollination. More recently, two non-destructive tools for measuring kernel moisture in the field were reported to potentially be very useful for breeding for fast kernel drydown in maize (Reid et al, 2010; Yang et al, 2010). In one study, the area under the drydown curve calculated from four measurements of moisture content using a digital moisture meter (manufacturer model BLD5601) was used to evaluate the rate of drydown (Yang et al, 2010). In a previous study we reported

how a modified timber moisture meter (Electrophics model MT808) was used to measure ear moisture, which was then multiplied by 1.11 to calculate kernel moisture when the ear moisture reading (EMR) was under 40 (Reid et al, 2010). The modified MT808 meter can be used in the field to screen a large number of potential lines per hour (200-300 ears per hour). All of these studies concluded that these light weight, non-destructive tools for measuring kernel moisture in the field will be very useful for breeding for fast kernel drydown in maize.

The objective of this study was to determine the optimum time for measuring ear moisture content using a modified moisture meter, as proposed by Reid et al (2010). The use of the moisture meter was verified with a series of inbred lines and hybrids made from them.

Materials and Methods

A field experiment in a split-plot design, with three replicates, was conducted in 2007, 2008 and 2009 at the Eastern Cereal and Oilseed Research Centre, Central Experimental Farm, Agriculture and Agri-Food Canada (AAFC), Ottawa, Ontario, Canada (45°22'N, 75°43'W). The planting dates were May 9, 2007, May 14, 2008 and May 18, 2009. Fourteen maize genotypes: six inbred lines (CL30, CO388, CO441, CO442, CO444, and MBS1236) and eight F₁ hybrids (CO388xCL30, CO441xCL30, CO442xCL30, CO444xCL30, CO388xMBS1236, CO441xMBS1236, CO442xMBS1236, and CO444xMBS1236) were the main plot units while nine sampling dates within each main plot were the split-plot units. The inbreds were selected to represent a range in maturities from early (CL30) to late (CO388 and MBS1236) for the Ottawa region. All inbreds were from the AAFC maize breeding program except MBS1236, which was a success-

Table 1 - Mean square results from a mixed model ANOVA by year and combined across years.

	df	Year		
		2007	2008	2009
<i>by year</i>				
Genotype	13	104.4 ***§	113.6 ***	78.8 ***
Sampling time	7	5732.9 ***	480.7 ***	5049.8 ***
Genotype x sampling time	91	17.7 ***	22.5 ***	13.2 **
Residual	222	4.8	9.2	9.5
Total	333			
<i>combined data across years</i>				
Year	2	429.6 ns		
Genotype	13	189.3 **		
Sampling time	7	15474.0 ***		
Genotype x sampling time	14	22.2 ***		
Year x sampling time	26	56.7 ***		
Year x genotype	91	54.2 ***		
Year x genotype x sampling time	182	17.7 ***		
Residual	666	7.8		
Total	1001			

§ns, **, *** stands for not significant, significant at the P= 0.01 or P = 0.001 level of probability.

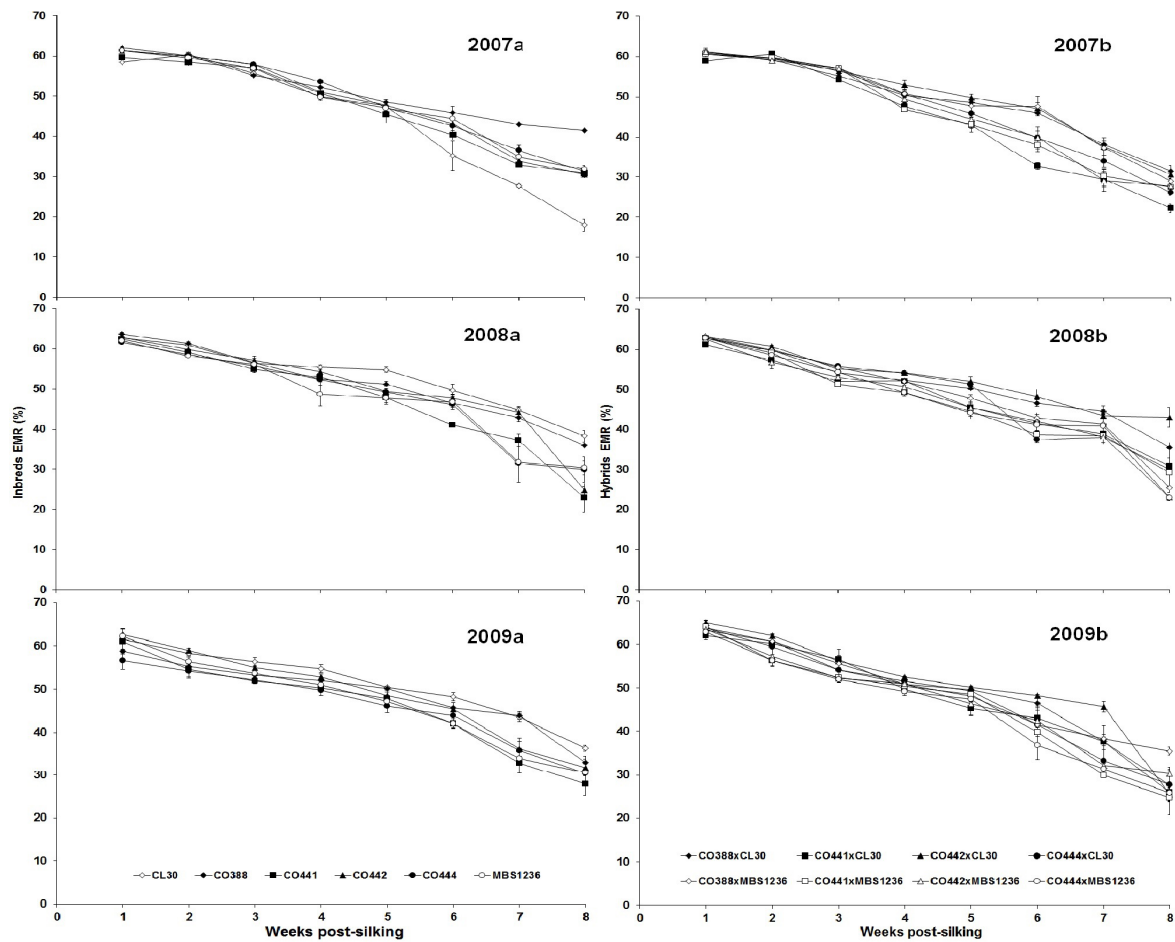


Figure 1 - Ear moisture readings (EMRs) from one to eight weeks after silking for six maize inbred lines and eight hybrids measured using a modified moisture meter (Electrophysics Model MT808): 2007a, 2008a and 2009a, inbreds; 2007b, 2008b, 2009b, F₁ hybrids crossed with the earliest maturing inbred CL30 and the later maturing inbred MBS1236.

ful tester line for AAFC genotypes from MBS Genetics LLC. The hybrids were a cross of the four CO inbreds as females with CL30 and with MBS1236 as males (females are listed first in a cross, e.g. female x male). Each main plot consisted of 11 rows, each 3.5 m long, with 15 plants and 0.76 m between rows. The first and the last row of each main plot were used as border rows to minimize the effect of having shorter stature inbreds in the same experiment as taller hybrids. The inner nine rows were randomly assigned to eight weekly sampling dates starting post-silking and one row where the ears were sampled each week. This represented the time span for moisture loss from silking, when the ears had their highest moisture, to late September depending on genotype maturity. The ninth treatment consisted of taking all eight readings on the same row such that each ear was punctured eight times to determine if multiple moisture measurements could be taken on the same ear. Ear moisture readings were recorded using a non-destructive method with a modified Electrophysics Moisture Meter, model MT808 (Reid et al, 2010) which can measure moisture from 4% to 100%. Two pins of the MT808

were inserted into the middle of each ear through the husks and into the kernels and cob. The center 12 plants of each row were used for moisture measurements and a mean of the 12 plants was calculated. The silking date for each genotype was recorded when 50% of the plants had silk protruding from the primary ears. Daily maximum and minimum air temperatures, rainfall and solar radiation data were recorded at the Central Experimental Farm weather station approximately 100 m from the experimental field site. Daily drydown rate (DDR) of the ears was calculated using the following formula:

$$DDR = (EMR_t - EMR_{(t+\Delta t)})/\Delta t$$

where: EMR_t and $EMR_{(t+\Delta t)}$ are, respectively, the EMRs at time t and $t+\Delta t$ and Δt is the period of days between the first and second EMR in days.

A mixed-model ANOVA (Proc mixed, SAS version 9.1, 2003) was used to determine significant differences among EMRs for each individual year prior to combining the data to determine if there was significant year, or year interactions. In each analysis, genotype and sampling time were fixed effects while year was considered random.

Results and Discussion

Initially each year was analyzed separately (Table 1), and there were significant differences in EMRs among genotypes, sampling times and the interaction of genotype by sampling time. A test of homogeneity of error variance of the years analyzed independently indicated that the three years could be combined into one mixed model ANOVA. There were no significant differences among years for EMR but there were significant differences among sampling times, genotypes and the interactions of year by genotype, year by sampling time, and year by sampling time by genotype. Ear moisture readings decreased weekly from silking for all genotypes (Figure 1). Average EMRs decreased from 61.9% one week post-silking to 47.8% by the fifth week and 29.5% by the eighth week. This resulted in a 14.1% decrease during the first four weeks and an additional 18.3% during last three weeks post-silking.

Significant genotypic differences in EMRs were found each year (Table 1, Figure 1). The decrease in EMRs from the first to the fifth week was almost linear after which the EMR dropped rapidly. Genotypic differences from the first to the fifth week were smaller than differences from the fifth to eighth week among all 14 genotypes. By the fifth week, significant differences between genotypes could be observed (Figure 1), with the exception of the inbreds in 2007 where differences could only be observed by the sixth week. Seed from the earliest maturing inbred, CL30, had the lowest EMRs in 2007, but the highest in 2008 and 2009 (Table 2). The other five inbreds had intermediate EMRs in all three years although there were clear-cut differences by 8 weeks post-silking; CO441 had the lowest EMRs in most years and CO388 the highest. Among the four CL30 hybrids, CO441xCL30 and CO444xCL30 had consistently lower EMRs, whereas CO388xCL30 and CO442xCL30 had con-

sistently higher EMRs. Similar results were obtained with the four MBS1236 hybrids; CO441xMBS1236 and CO444xMBS1236 had consistently lower EMRs overall, whereas CO388xMBS1236 and CO442xMBS1236 had higher EMRs (Figure 1, Table 2).

The weekly EMRs, taken from one to eight weeks post-silking, when individual ears were punctured only once, were compared to row 9 where the EMRs were taken weekly on the same ear. There was a high correlation between these two sets of readings ($r=0.97$, $p<0.01$, data not shown) indicating that EMRs can be done multiple times on the same ear without affecting results; however, it should be noted that this may increase the chance of fungal infections entering these ears through puncture wounds. For example, *Fusarium* species often enter the ear through puncture wounds created by insects, birds or hail (Sutton, 1982).

The mean of the EMRs from the three years at 1, 5, and 8 weeks post-silking was used to calculate the DDR (Table 2). The mean DDR for all of the genotypes for the period from week 1 to week 5 was 0.5 % per day and from week 5 to week 8 was 0.87 % per day. There appeared to be slightly more variation in DDR between years later rather than earlier in the season; DDRs were 0.83%, 0.88%, and 0.89% per day from five to eight weeks in 2007, 2008, and 2009, respectively. It was clear from the data that DDR was influenced by genotype. Of the six inbred lines, CL30 was the earliest to silk and CO441 silked five days after it (Table 2). While CO441 had the lowest EMRs at both the fifth and eighth week, CL30, which was faster to mature, had the highest EMRs on the fifth week and the second highest on the eighth week. Similarly, CO388 and MBS1236 were the latest maturing inbreds, both silking at approximately 80 days after planting, but MBS1236 had lower EMRs than CO388 on both the fifth and eighth week. Similar complexi-

Table 2 - Average silking date, ear moisture reading (EMR) (one, five and eight weeks post-silking) taken with a modified MT808 meter and daily drydown rate (DDR) from weeks one to five, five to eight and one to eight for six inbreds and eight hybrids, 2007 to 2009, at Ottawa, ON, Canada.

Genotype	Number	Silking Date ^a	EMR (%)			DDR (%) ^b		
			Week 1	Week 5	Week 8	Weeks 1-5	Weeks 5-8	Weeks 1-8
CL30	1	67e	60.9ab	50.9a	30.8ab	0.36d	0.95a	0.61ab
CO388	2	81a	61.5ab	50.0ab	36.8a	0.41cd	0.63a	0.50b
CO441	3	72cd	61.0ab	47.1bc	27.2b	0.50bc	0.95a	0.69a
CO442	4	76bc	62.3a	48.5b	29.1ab	0.49bc	0.92a	0.68a
CO444	5	78ab	59.8b	47.5bc	30.6ab	0.44bcd	0.81a	0.60ab
MBS1236	6	81a	61.9ab	47.3bc	30.7ab	0.52b	0.79a	0.64ab
CO388xCL30	7	69de	62.4a	49.5ab	31.5ab	0.46bc	0.86a	0.63ab
CO441xCL30	8	69de	61.3ab	44.6d	26.2b	0.60ab	0.88a	0.72ab
CO442xCL30	9	69de	63.1a	50.7ab	33.1ab	0.44bcd	0.84a	0.61ab
CO444xCL30	10	69de	62.5a	48.9ab	28.0b	0.49bc	1.00a	0.70a
CO388xMBS1236	11	75bc	62.6a	47.9bc	30.0ab	0.53ab	0.85a	0.66ab
CO441xMBS1236	12	72cd	62.5a	45.4cd	27.2b	0.61a	0.87a	0.72a
CO442xMBS1236	13	74bc	62.4a	45.9c	27.3b	0.59ab	0.88a	0.72a
CO444xMBS1236	14	76bc	62.3a	45.4cd	25.0b	0.61a	0.97a	0.76a
Mean	73	61.9	47.8	29.5	0.50	0.87	0.66	
LSD (0.05)		4.1	2.35	2.34	8.52	0.09	0.38	0.18

Means followed by the same letter within a column are not significantly different (0.05).

^anumber of days from planting until 50% of the plants of a given genotype have silk protruding from the ears < ^bDDR (daily drydown rate) = $(EMR_t - EMR_{t+\Delta t}) / \Delta t$, where EMR_t and $EMR_{t+\Delta t}$ were the ear moisture readings at time t and $t+\Delta t$ and Δt is the period of days between the first and second moisture reading.

ties were observed with the hybrids. The CL30 hybrids silked earlier than the MBS1236 hybrids, but the MBS1236 hybrids had lower EMRs than all of the CL30 hybrids, with the exception of the hybrid between the two earliest inbreds, CO441xCL30. These results indicate that genotypes with the potential for silking early may still dry slower than those that silk later, highlighting that rapid kernel drydown is not always correlated with early silking.

When the DDR for the genotypes from week 1 to 5 and week 5 to 8 were plotted (Figure 2), the genotypes used in this study could be classified into four groups. There were three genotypes with a lower DDR than the mean for week 1 to 5 and for week 5 to 8 (CO388, CO444 and CO442 x CL30). The CO388xCL30 hybrid had a lower DDR than the mean for the first duration but a similar DDR to the mean for the second duration. Three genotypes had a low:high DDR combination (CL30, CO444xCL30 and CO442) while CO441 had a similar DDR to the mean for the first duration and a higher DDR for weeks 5 to 8. There were two genotypes with a high:low combination (MBSS1236 and CO388xMBS1236) and one with a higher first duration but a similar second duration (CO441xMBS1236). Finally, CO444xMBS1236 had higher DDR in the first and second duration compared to the mean while two other genotypes (CO442xMBS1236 and CO441xCL30) had higher first durations, but a similar week 5 to 8 compared to the mean. There were two inbreds (CO441 and CO442) and six hybrids (all the MBS1236 crosses and CO441xCL30) with overall DDRs from the first to eighth week equal to or higher than the mean overall DDR. Of these genotypes, six had higher DDRs by the fifth week and the remaining two had higher DDRs by the eighth week. This suggests that to obtain the fast drydown characteristic, the genotype should have fast drydown in the first five weeks after silking or have a significant drydown between five and eight weeks after silking. All four MBS1236 hybrids had slightly higher DDRs from week 1 to week 5 than the corresponding CL30 hybrid with the same female inbred parent. Similarly, three CL30 hybrids had slightly higher, but not statistically significant, DDRs from week 5 to week 8 than their corresponding MBS1236 hybrids, with the exception of the hybrid with CO442. The overall DDRs were slightly higher with the MBS1236 hybrids than with the CL30 hybrids. These data may indicate that early stage DDRs, may play a more important role than later stage DDRs.

The above results indicate that rather than repeatedly measuring kernel moisture, EMRs taken at silking, at the late grain filling stage (five weeks post-silking), and just before harvest (eight weeks post-silking), are sufficient to calculate DDR for a given genotype. This could be further reduced to two measurements if it could be shown that the kernel moisture at silking was a constant. This would mean that only the two remaining EMRs need be done, the first as early

as five weeks (35 days) post-silking and the second at eight weeks (56 days) post-silking at Ottawa. The first measurement is later than that recommended by Freppon et al (1992) (30 days after pollination) and earlier than Cross et al (1987) (45 days after pollination). Other researchers have recommended that two or more moisture measurements be taken (Kang et al, 1978; Yang et al, 2010) because kernel drydown in maize is a trait influenced by many factors with low heritability.

For some genotypes, there was significant genotype by sampling time, or genotype by sampling time by year interactions. These interactions may have been caused by the environment in which the EMR was taken; other researchers have reported significant genotype by environment interactions for moisture loss in corn (Magari et al, 1997). For example, EMRs are taken on living plants in the field, but the MT808 Electrophysics Moisture Meter is sensitive to water that may be present on the husks. Another important factor was that overall, the air temperature and amount of sunshine became lower as the season progressed from late July to August and into the fall. Thus, earlier silking genotypes like CL30 often have grain-filling periods with higher temperatures and more sunshine than later silking genotypes such as CO388.

The results of this study indicated that when moisture readings were made using the modified MT808, the rate of moisture loss from the first to the fifth week post-silking was almost linear, but then dropped from the sixth to eighth week. Ear moisture readings between genotypes can be significantly different by the fifth week (35 days post-silking). Three EMRs can

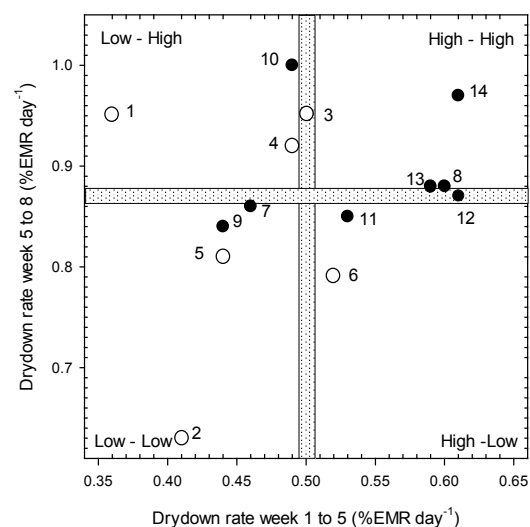


Figure 2 - Drydown rates from week one to five plotted against the drydown rate from week five to eight. The shaded line represents the mean and standard deviation of 14 genotypes, while clear and filled circles represent inbred lines and hybrids, respectively, numbered according to Table 2.

be used to calculate DDRs for evaluating the kernel drydown traits. Genotypes with DDRs ≥ 0.5 from the first to fifth week, ≥ 0.87 from the fifth to eighth week, or overall ≥ 0.66 from the first to eighth week can be classified as possessing a fast drydown trait at early, late, and entire growth stages, respectively. DDRs from the first to fifth week played a more important role in determining the final DDRs than DDRs from the fifth to eighth week.

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