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Temporal Changes in Abscisic Acid Concentration in Dormant and Non-Dormant Seeds of Wheat (*Triticum spp.*) Genotypes

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ABSTRACT

Little is known about the levels and physiological role of endogenous abscisic acid (ABA) during after-ripening and germination. Genetic variants for ABA content were investigated to account for the role of ABA in the persistence of seed dormancy. In this study, genotypic variation in ABA contents at Zadok's Growth Stage 92 (ZGS 92), temporal changes in ABA during two physiological stages (after-ripening and water uptake), and responsiveness of wheat seed to applied ABA at different concentrations, of two tetraploid and two hexaploid wheat genotypes were examined during 1996 and 97. A combined analysis of variance indicated no significant differences in ABA among genotypes at ZGS 92. During the early stages of germination, endogenous ABA in the caryopses of the four wheat genotypes was analyzed. The results showed a transient increase in ABA content (up to 4 hours) occurring first during imbibition, followed by a decline up to 12 hours and an increase thereafter. ABA declined in all genotypes during seven weeks of after-ripening (dry storage). The four genotypes had reductions in ABA up to 4 weeks of after-ripening. An increase in ABA was observed during the fourth and fifth weeks of after-ripening with a decline after seven weeks. ABA had a little effect on germination index at the lower temperature (10 °C). Our results suggested that wheat grains are able to synthesize ABA during imbibition. However, no significant differences between dormant and non-dormant genotypes were detected. A decrease in ABA during after-ripening could have a role in loss of seed dormancy.

Keywords: Abscisic acid, After-ripening, Dormancy, Germination, Imbibition, Seed.

INTRODUCTION

Seed dormancy in wheat (*Triticum* spp.) is an important agronomic trait. Lack of seed dormancy can result in pre-harvest sprouting under wet climatic conditions, which adversely affects end-use quality. In contrast, excessive dormancy results in poor stand establishment (Derera, 1989). The intensity of seed dormancy varies greatly and this variation may be genetic in origin but influenced by the environmental conditions occurring during seed development (Simpson, 1990). For several decades, the study of the internal factors that regulate seed dormancy was principally conducted by the comparison of the biochemical and physiological behaviors of dormant and non-dormant seeds. One of the most studied research fields has been the involvement of plant growth regulators in seed dormancy. Abscisic acid (ABA) is known to promote normal embryogenesis and prevent precocious germination in immature wheat embryos (Williamson and Quatrano, 1988). Investigations on the role of ABA in grain dormancy

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of cereals have focused mainly on the developmental stages of seed. However, there is no general agreement on the role of ABA content in seed dormancy at physiological maturity and during after-ripening (Black, 1991).

The objectives of this study were to determine ABA levels at harvest maturity (ZGS92), differential changes in ABA during after-ripening and early stages of germination, and responsiveness of wheat seed to ABA in dormant and non-dormant wheat genotypes.

MATERIALS AND METHODS

Plant Material

In this study, spring wheat genotypes, two hexaploid, RL4137 and AUS 1408, (red and white, highly dormant), and two tetraploid, 93-282 and Sceptre (highly dormant and non-dormant) were used. Seeds from plants grown under field conditions at the University of Saskatchewan's Seed Farm (Saskatoon) in 1996 and 1997 were used for this experiment. A randomized complete block design (RCBD) with four replications was used in field experiments. Each plot consisted of four rows (2.4 m long) spaced 30 cm apart and was sown at approximately 250 seeds/m^2. Fertilizer (11-51-0) was drilled with the seed at approximately 50 Kg ha^{-1} and all experiments were conducted on fallow land. The spikes were harvested at ZGS 92, dried for one week at room temperature and kept in a freezer at –20 °C. Ten spikes from each replication were threshed by hand.

ELISA and GC/MS

For ABA extraction and quantitative analysis of ABA in intact wheat seeds, a modified form of an indirect enzyme-linked immunosorbent assay (ELISA) utilizing a monoclonal antibody (Walker-Simmons, 1987), developed by Dunstan and Bock (1997) was used. Analysis of ABA content by gas chromatography/mass spectrometer (GC/MS) (Walker-Simmons *et al*., 1998) were performed to test the efficiency of extraction procedure and to verify the ELISA technique.

Genetic Variation in ABA Content at ZGS 92 (Maturity)

Twenty-five intact seeds of three field replicates were taken from the freezer. Ten mg of powdered samples were used to measure ABA content. The ELISA experiment was conducted in a randomized block design and it was conducted on all genotypes five times in each replication (each extraction solution).

Genetic Variation and Differential Changes in ABA Levels During Early Periods of Germination

Twenty-five seeds from each of three field replicates were placed on two layers of Whatman No. 1 filter paper containing 3 ml distilled water. Wheat grains were allowed to imbibe for 4, 8, 12, 24, and 36 hours at 20 °C in the dark. Grains with no imbibition were used as controls (0 time). At each interval, samples were taken from Petri plates, frozen immediately in liquid nitrogen, lyophilized, and ground. Extraction was conducted for each sample. A randomized complete block design with three replications was used. ELISA was conducted on all genotypes three times in each replication (extraction solution).

Genotypic Variation in ABA Level During Seed After-Ripening

To assess ABA levels, twenty five seeds stored at - 20 °C were taken from the freezer at one-week intervals up to seven weeks and kept at room temperature (23±2 °C). The seeds were frozen in liquid nitrogen, lyophi-

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lized, and powdered. Ten mg samples were used for the extraction. ELISA experiments were conducted in a RCBD with three replications.

Response of Wheat Seed to ABA Concentration at Two Temperatures

Two replicates each containing 50 seeds from four wheat cultivars were placed on two layers of Whatman no. 1 filter paper in 15×100 mm Petri plates. The plates were irrigated with 5 ml of test solution containing 0, 5, 10, and 20 M ABA and incubated at 10 °C and 20 °C in the dark. The Petri plates were kept under sealed conditions to prevent moisture loss. Germination counts were performed for seven days and seeds with radicle growth in excess of 3-4 mm were considered germinated and removed from the plate. A factorial experiment design with a split plot arrangement along with three replications was used. A weighted germination index (GI) for seeds (Walker-Simmons and Sesing, 1990) was used in the data analysis as follows:

$$GI=\frac{7\times n_1+6\times n_2+5\times n_3+..+1\times n_7}{7\times \text{total seeds}}$$

where n is the number of germinated seeds at day 1, 2, and etc.

The germination index is useful because the plot of the cumulative germination percentage versus time can be reduced to a single germination index point.

Data Analysis

Analysis of variance (ANOVA) was performed for individual experiments separately. Subsequently, combined analyses were used over years. For all experiments, years and replications were considered random. To test the homogeneity of variances, the Bartlett's test was applied. Least significant differences (LSD) were calculated to measure the differences between cultivars and germination indices.

RESULTS

Genetic Variation of ABA Content at ZGS 92

In this experiment, ABA content of two hexaploid wheat genotypes with high levels of seed dormancy and two tetraploid wheat genotypes, one highly dormant and the other non-dormant, were evaluated.

Although in 1996 and 1997, hexaploid wheat cultivar RL4137 exhibited higher ABA at ZGS 92 than did Sceptre (non-dormant), the analysis of variance detected no statistically significant differences among genotypes. On average, RL4137 (highly dormant) exhibited the highest ABA (837 pg/mg DW) while Sceptre the non-dormant cultivar had the lowest ABA (673 pg/mg DW) at ZGS 92 (Table 1 and 2).

Averaged over years, no significant differences were detected among genotypes. ABA for wheat genotypes significantly varied from year to year. On average, the ABA was 49% higher in 1996 than in 1997.

Table 1. ABA contents in intact seeds of wheat cultivars at ZGS92 in 1996 and 97.

Genotype	ABA (pg/mg dry weight)		
	1996	1997	Average
RL4137	970	705	837
Sceptre	869	478	673
AUS1408	1143	472	808
93-282	1134	426	780
LSD (0.05)	328	187	

Table 2. Analysis of variance for ABA content in seeds of wheat genotypes at ZGS92 in 1996 and 1997.

Source	DF	Mean Squares	
		1996	1997
Genotype (G)	3	265966	235246
Replication (R)	2	44773	268000
ELISA (Replication)	12	557125**	121168
G × R	6	361753**	192593*
Error	36	107040	65737

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Differential Changes in ABA During the Early Stages of Germination

The aim of this study was to investigate the endogenous ABA during imbibition in intact grains from both dormant and non-dormant genotypes. The ABA extraction solution from each replication in all four genotypes was tested by ELISA three times. There were no statistically significant differences in ABA among cultivars in 1996 and 1997 (table 3).

Differences in ABA varied significantly with length of imbibition in both years. The interaction between cultivar and imbibition time was not significant in either year. On average, AUS1408 (dormant genotype) had the highest ABA in 1996 and 1997 (826 and 559 pg/mg DW, respectively), while Sceptre (non-dormant) had the lowest (data not shown).

With respect to the ABA levels following various imbibition times, a transient increase in ABA (up to four hours of imbibition) was observed followed by a decline (up to 12 hours) and an increase in the post-12 hour period (table 4). The ABA levels of all cultivars at 12 hours of imbibition were the lowest and an increase in ABA after 24 hours of imbibition was observed for all cultivars.

Table 3. Analysis of variance for ABA contents during the early stages of germination (various imbibition intervals) in 1996 and 1997.

Source	DF	Mean Squares	
		1996	1997
Cultivar (C)	3	378327	27734
Imbibition time(I)	5	885440**	1791564**
Replication (R)	2	303483	1355777*
ELISA within (R)	6	94032*	38101
C × I	15	64354	55240
C × R	6	149188	120676*
I × R	10	96664	86079
C × I × R	30	80106**	40024**
Error	138	33225	21747

* (p=0.05), ** (p=0.01)

In both years, a highly significant three way interaction between cultivar, imbibition, and replication was detected (table 3). In 1996 (data not shown), it appears that the nature of the interaction exists in the second replication. Except for AUS1408, the genotypes did not show an increase in ABA between 12 h and 24 h. The ABA increased in AUS1408 after 12 h of imbibition. The patterns of ABA response to imbibition in the first and third replications were similar for

Table 4. Temporal changes in ABA contents (pg/mgDW^{-1}) of wheat genotypes during imbibition over 1996 and 97.

Genotype	Imbibition periods (hours)						
	0	4	8	12	24	36	Average
RL4137	593.12	693.88	492.32	368.29	569.64	814.96	588.70
AUS1408	628.65	728.70	658.44	460.72	768.90	909.83	692.54
93-282	565.15	718.50	579.60	317.39	519.43	949.91	608.33
Sceptre	501.57	685.93	549.51	387.33	548.48	776.26	574.84

LSD (0.05) between treatments=132.17

Table 5. Comparison of ABA levels measured by ELISA or GC-MS in sample extracts of RL4137 (highly dormant genotype).

Imbibition (hours)	(+) ABA (pg/mg DW)			
	GC-MS (#1)	GC-MS (#2)	Average (adj.)[a]	ELISA
0	564	555	528	622
4	847	840	826	866
8	693	577	583	541
12	614	573	555	532
24	734	680	655	684
36	847	893	806	790

[a] Adjusted value based on (+) ABA level in one mg of seed sample.

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all genotypes. In 1997, the interaction appears to be caused by the third replication (data not shown). The level of ABA decreased in all genotypes at 4 h of imbibition, except for accession 93-282 which showed an increase. Moreover, unlike other genotypes, the level of ABA in RL4137 increased at 12 h. The patterns of ABA level among genotypes in the first and second replication were similar.

ELISA Verification by GC/MS

ABA immunoassay results of the highly dormant cultivar, RL4137, from the previous study utilizing the ABA-4-BSA conjugate in an indirect ELISA assay were verified by GC-MS (table 5). For verification by GC-MS, the seed samples from the second replication of the 1996 field trial were used. This replication was selected at random. The ABA extraction from the one replication was repeated twice and compared with the ELISA results.

The results of GC-MS verified the pattern of ABA inside the intact seed during various imbibition periods as measured by ELISA. That is, a transient increase in ABA after four hours, followed by a decline up to 12 hours and an increase thereafter. Correlation between the results of ELISA and GC-MS in this experiment was highly significant (r=0.89, p=0.05), verifying the validity of the ELISA analysis.

Differential Changes in ABA Level with Length of After-Ripening

In 1996, no significant differences were detected among genotypes. However, in 1997 these differences were statistically significant (table 6). In both years, the genotype by after-ripening interaction was not statistically significant. The results indicated that seven weeks of after-ripening resulted in a large reduction in ABA in the seed in all four cultivars for both years. A greater reduction in ABA levels was detected in 1997 than in 1996. A combined ANOVA did not detect significant differences among genotypes and year (data not shown).

A similar response pattern for ABA to after-ripening was observed among genotypes in 1996 and 1997. Averaged over years, all genotypes exhibited a reduction in ABA up to four weeks of after-ripening followed by

Table 6. Analysis of variance for differential changes in ABA levels during after-ripening in 1996 and 1997.

Source	DF	Mean Square	
		1996	1997
Replication	2	91030**	339388**
Genotype (G)	3	30816	133359*
Length of After-ripening (L)	7	115446**	57682**
G × L	21	7023	22937
Error	62	12311	21505

* (p=0.05), ** (p=0.01)

Table 7. Differential changes in ABA levels (pg/mg DW) during after-ripening averaged over two year (1996 and 1997).

	After-ripening period (week)							
Genotype	0	1	2	3	4	5	6	7
RL4137	533	442	313	184	310	503	375	216
AUS1408	533	488	435	234	489	656	482	272
93-282	570	563	363	149	356	675	354	200
Sceptre	521	589	296	148	307	548	451	211

LSD (0.05) between two genotypes at one level of after-ripening=469

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an increase in ABA at four and five weeks and a further decline up to the seventh week (table 7).

Response of Wheat Genotypes to Exogenous Application of ABA at Two Imbibition Temperatures

Analysis of variance of germination indices indicated significant differences among genotypes (table 8). No significant differences were detected among ABA concentrations or imbibition temperatures. However, the genotype x temperature interaction was statistically significant. On average, RL4137 had the lowest germination index (0.26).

Table 8. Analysis of variance for germination indices of wheat genotypes at four different concentrations of ABA and two temperatures in 1997.

Source	DF	Mean Squares
Replication (R)	1	0.0153
Temperature (T)	1	0.0791
R × T	1	0.0006
Genotype (G)	3	0.0948**
ABA Concentrations (A)	3	0.0052
T × G	3	0.0457**
T × A	3	0.0013
G × A	9	0.0008
T × G × A	9	0.0012
Error	30	0.0027

** (p=0.01)

Increasing the germination temperature resulted in lower germination indices for the dormant genotypes (table 9).

At 20 °C, except Sceptre, application of 20 M of ABA reduced the germination indices of RL4137, AUS1408, and 93-282 by approximately 30%, 16%, and 21%, respectively as compared to the distilled water (table 9). The germination characteristics of dormant genotypes were temperature sensitive at elevated exogenous ABA concentrations, while seeds of the cultivar Sceptre exhibited little such sensitivity.

ABA concentration had a slight effect on germination index at the lower temperature (10°C) (table 9). Germination indices of all genotypes remained similar over ABA concentrations at the low temperature.

DISCUSSION

Abscisic acid plays an important role in seed development, being involved in storage protein synthesis and prevention of precocious germination (Black, 1991). Likewise, research on the mechanism of seed dormancy suggests a strong involvement of ABA (Kermode, 1990). Although much information exists concerning ABA content and the sensitivity of seed to ABA during its development, little is known about endogenous ABA content during germination and after-ripening. In the present study, potential genetic variants for ABA after varying lengths of after-ripening and early germination were evaluated.

At ZGS 92 (harvest maturity), there were small differences in the endogenous ABA of four wheat genotypes. Although the ABA in the non-dormant tetraploid cultivar Sceptre was the lowest in both years, the differences among genotypes were not statistically significant. There is not a general agreement on the role of ABA in seed dormancy at maturity. Walker-Simmons (1987) demonstrated

Table 9. Effect of different ABA concentrations and two temperatures on germination indices of wheat genotypes averaged over replications in 1997.

Genotype	10^0C				20^0C			
	0	5 µM	10 µM	20 µM	0	5 µM	10 µM	20 µM
RL4137	0.32	0.28	0.31	0.29	0.23	0.26	0.17	0.16
AUS1408	0.41	0.39	0.39	0.38	0.19	0.19	0.20	0.16
93-282	0.44	0.41	0.41	0.39	0.48	0.43	0.39	0.38
Sceptre	0.41	0.37	0.38	0.36	0.40	0.39	0.35	0.40

LSD (0.05) (between all numbers in the table)=0.11

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that ABA was similar in the embryos of sprouting-resistant and sprouting-susceptible wheat cultivars. In contrast, Wang (1996) showed that in barley grains, a very high ABA in the intact dormant seed was correlated with a lack of germination.

One of the first aspects to be tested in this study was the endogenous ABA during imbibition in the intact grains from both dormant and non-dormant wheat genotypes. A similar pattern was observed among genotypes, that is, an increase in ABA content (up to 4 hours) followed by a decline up to 12 h and an increase in the post-12 h period. The phenomenon of ABA resynthesis during early periods of germination was established in this study. On average, the ABA was increased by 7% and 23% at 24 and 36 hours of imbibition, respectively. In a similar study with barley (Wang, 1996), an increase in ABA during the first ten hours of imbibition was observed followed by a decline. The ABA; however, still remained higher after 48 h than at the start of imbibition. Wang (1996) concluded that the differences in ABA content among imbibition intervals could indicate a faster diffusion or turn-over of endogenous ABA in the grain.

The results of this study suggest a transient increase in ABA during the first four hours of seed imbibition. The physiological reason for such an increase is not clear yet. Bianco *et al.* (1997) and Le Page-Degivry *et al.* (1997) suggested that this increase results from the hydrolysis of ABA conjugates stored in dry seeds. According to Le Page-Degivry *et al.* (1997) in a tree species, *Fagus sylvatica* L., hydrolysis of conjugates present in cotyledons could be a potential source of free ABA upon imbibition. This pool of conjugates appears to be relatively large. However, in other species such as Douglas fir (*Pseudotsuga menziesi*), dry seeds contained a low ABA esters, and therefore their hydrolysis could not be responsible for the increased ABA in dormant embryos (Bianco *et al*., 1997).

The decrease in ABA between 4 and 12 h of imbibition could be a result of outward diffusion of endogenous ABA from the seed. In a study by Wang *et al.* (1995), when the embryos were incubated in water, the amount of ABA in the embryo decreased exponentially with time. In accordance with the decrease in embryo ABA, the ABA in the medium increased with time. The authors also indicated that the ABA in the medium of dormant embryos was higher for a longer period. Visser *et al.* (1996) hypothesized that an increased ability to degrade extracellular ABA could play a role in germination of non-dormant seeds. The authors revealed that the ABA in the incubation medium of embryos from an ABA-insensitive mutant of barley decreased much more rapidly than those of the wild-type incubation medium. Visser *et al.* (1996) suggested that the inhibitory action of ABA occurs outside of the embryo. Thus, an ABA-perception site might be located at the surface of the embryo.

Results from this study provide evidence that all wheat genotypes were able to resynthesize ABA. According to Wang *et al.* (1995), *de-novo* synthesis of ABA could play a role in the control of germination. The authors reported that embryos isolated from dormant barley seeds and incubated are able to resynthesize ABA. However, in contrast to our finding, they indicated that embryos from non-dormant barley seeds are unable to resynthesize ABA. At present, the role of ABA resynthesis in seed dormancy awaits evaluation.

In some species, mature, dehydrated seeds are dormant and additional dry storage (after-ripening) is necessary and sufficient to overcome primary dormancy. Our results demonstrated that after-ripening was associated with a decline in ABA. Such a decrease was noticed by King (1976) who showed that forced drying of wheat seeds induced both a rapid drop in seed ABA and an increase in germinability. In soybean, drying of the embryos slowly within the pod caused a gradual decline in ABA and, in parallel, an increase in the ability of immature embryos to germinate (Ackerson, 1984). Similarly, Bianco *et al*. (1994) reported that slow drying of dormant embryos in sunflower (*Heli-*

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anthus annuus) caused a decline in ABA of whole embryos as well as of isolated axes. However, dry storage for a period of six weeks did not induce any additional significant modification in ABA level of either whole embryos or of isolated axes.

The results of this study indicated that, with the exception of weeks four and five, seven week after-ripening resulted in a lower ABA content of the seed. With respect to the role of after-ripening, one point is worth noting and this concerns the effect of drying. Generally, after the seeds reach maximum dry or fresh weight, the level of ABA declines sharply. Artificial drying confers germinability upon very young wheat grains, an effect that has been attributed to the lowering of ABA brought about by dehydration (King, 1976). There is evidence to support the possibility that some of the free ABA is first transformed into bound ABA. In wheat cultivars studied by King (1976), the level of bound ABA appeared to be greater than the amount of free ABA that has been lost. The drop in ABA content, as we have seen, generally occurs after the seed has accumulated its food reserves and when maturation begins. Loss of water takes place during after-ripening, and it may be argued that this fall in water content precipitates the depletion in ABA. The coincidence between the reduced water content of wheat grains and the decrease in ABA has been found in other investigations on wheat (King 1976).

Dormancy in intact seeds of wheat is closely associated with responsiveness of the embryo to ABA (Morris *et al*. 1989). Likewise, embryonic responsiveness to ABA in wheat has been found to be developmentally regulated and to be retained longer in cultivars resistant to pre-harvest sprouting (Walker-Simmons, 1987). In our study, no genotypic variation existed for seed germination response to exogenous application of ABA. At a lower temperature (10 °C), seeds of all the genotypes showed 100% germination. At a higher (20°C) temperature, increased responsiveness to ABA (20 M) was observed only in the dormant genotype RL4137. Seeds of the non-dormant cultivar, Sceptre, lacked high-temperature dormancy and were not responsive to ABA. Similarly, Walker-Simmons (1988) has reported enhancement of ABA responsiveness of dormant seeds and embryos at higher temperatures. According to Walker-Simmons (1988), differences in the capacity of ABA to block germination may be due to variation in the action of hormone in binding to the hormone receptor or in the reactivity of the ABA-receptor complex.

In conclusion, the data presented here suggest that wheat grains are able to resynthesize ABA upon imbibition. The role of ABA resynthesis in seed dormancy was not investigated in this study. Therefore, further research will be required to examine possible mechanisms involved in resynthesis and reduction of ABA in wheat seeds. Moreover, further research will be required to investigate the possible roles of resynthesized ABA in dormancy induction.

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تغییرات زمانی غلظت اسید ابسایزیک در بذر ژنوتیپ های دارای خواب و بدون خواب گندم (*Triticum spp.*)

چکیده

هورمون اسید ابسایزیک به عنوان یکی از فاکتورهای مهم فیزیولوژیکی در ایجاد و استمرار خواب بذر در نظر گرفته می‌شود ولی یافته‌های علمی مرتبط با غلظت این هورمون و نقش آن در مراحل پس از رسیدگی و جوانه زنی بذر بسیار محدود است. در این آزمایش چهار ژنوتیپ گندم (هگزاپلوئید و تتراپلوئید) مورد استفاده قرارگرفت. مطالعه فوق در سالهای ۱۹۹۶ و ۱۹۹۷ با اهداف ذیل انجام گردید:

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الف) ارزیابی همبستگی فیزیولوژیکی بین غلظت اسید ابسایزیک و تنوع ژنتیکی خواب بذر در مرحله رسیدگی، ب) اندازه‌گیری غلظت ابسایزیک اسید در مراحل فیزیولوژیکی پس از رسیدگی ومراحل اولیه جوانه زنی و ج) واکنش بذر نسبت به غلظتهای متفاوت از اسید ابسایزیک در درجـــه حرارتهای ۱۰ °C و ۲۰ °C . نتایج این تحقیق نشان داد که در مرحله رسیدگی تفاوت معنی‌داری بین ژنوتیپ‌های مختلف گندم از نظر غلظت اسید ابسایزیک وجود ندارد. اندازه‌گیری غلظت هورمون در مدت ۳٦ ساعت جذب آب نشان داد که در ٤ ساعت اولیه میزان اسید ابسایزیک افزایش یافت. در حدفاصل ٤ تا ۱۲ ساعت کاهشی تدریجی در غلظت اسید ابسایزیک مشاهده گردید. مجدداً افزایش میزان این هورمون پس از ۱۲ ساعت در بذر مشاهده شد و این افزایش تا انتهای زمان آزمایش (۳٦ ساعت) ادامه داشت. با گذشت ۷ هفته از دوره پس از رسیدگی میزان اسید ابسایزیک به مقدار قابل ملاحظه‌ای کاهش پیدا کرد. غلظتهای متفاوت این هورمون فاقد اثر معنی‌دار بر شاخص جوانه‌زنی در درجه حرارت پائین بود. نتایج تحقیق فوق نشان داد که در مراحل اولیه جوانـــه‌زنی، بذرهای هر چهار ژنوتیپ قادر به سنتز اسید ابسایزیک بوده و همچنین کاهش میزان این هورمون در طی فرایند پس از رسیدگی می‌تواند در کاهش خواب بذر نقش داشته باشد.

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