SURVIVAL ANALYSIS OF THE WHEAT GERMINATION DATA

Jerko Gunjača, Hrvoje Šarčević
Faculty of Agriculture University of Zagreb
Svetošimunska 25, HR-10 000 Zagreb, Croatia
jgunjaca@agr.hr

Abstract: *Pre-harvest sprouting can seriously reduce wheat grain yield. To investigate one of possible natural mechanisms of protection, bract inhibition, two wheat varieties' seeds have been germinated in four different D-mannitol solutions. Survival analysis of germination data provided evidence that increase of the germination media concentration, resulting in its higher osmotic pressure, could magnify germination inhibition.*

Key words: wheat germination, bract inhibition, survival analysis, wheat (*Triticum aestivum* L.)

1. Introduction

In the seasons with frequent rainfalls during harvest, pre-harvest sprouting of wheat grain can cause serious yield reduction. There are two natural mechanisms of protection: dormancy and bract inhibition. Investigation on these mechanisms in an experiment with 50 wheat genotypes (Šarčević et al., 1998), showed that they have different levels of inhibition. The explanation of these differences without further knowledge of the nature of inhibition is difficult. The cause of inhibition is still largely unknown; it can be argued that it is the presence of chemical inhibitors in bracts, or solely the change in osmotic potential caused by dissolved bract organic compounds (Kaufmann and Ross, 1970). This investigation is focused on the later assumption. Thus, an experiment has been carried out to investigate the effect of solutions with different concentrations (hence different osmotic pressure) on seed germination rates. Results of the experiment are presented in this paper.

2. Materials and methods

D-mannitol is chosen as a germination medium, assuming it could not be involved in chemical reactions causing inhibition. Non-dormant seeds of two winter wheat varieties, Goranka and Dukat were germinated in 5 ml of of 0, 2, 4 or 5% D-mannitol solutions. The germination test has been performed in Petri dishes (containing 50 seeds) at 20^0C in the dark. Each variety/concentration combination has been set in three Petri dishes (150 seeds). After 41 hours grains with three developed roots longer than 3 mm were counted and removed. This procedure was repeated after 48, 65, 72 and 89 hours, until most of the seeds have germinated.

Survival analysis has been rarely used in seed germination studies (Scott, 1984), so there were few references for the choice of the appropriate model. In the beginning there had to be decided whether to use the Kaplan-Meier or the actuarial life-table estimates of the survival distribution function. In this experiment, each collected

datum represented an interval of time in which certain seed germinated, rather than the actual time of germination. For this type of the data, the actuarial life-table estimates are more appropriate.

The objective of this study was to explore the association between the concentration of the D-mannitol solution and the germination rate. With no previous knowledge about the association of the germination media thickness with the germination rate, the analysis has been carried out in two stages. First stage included separate analysis for each variety, with the D-mannitol concentration as a stratification variable, and comparison of resulting survival curves and log rank statistics. If trend of decreasing germination rate with increasing of the D-mannitol concentration can be observed, then in the second stage data are pooled. At this stage model includes variety as a stratification variable and D-mannitol concentration as a covariate, whose association with the germination rate could be tested using log rank test (Cantor, 1997).

3. Results

After 89 hours 948 out of 1200 seeds have germinated. At this point, amount of the solution left in the Petri dishes was decreased to the level that could cause unequal conditions for among seeds, so the experiment has been stopped. Thus, 21% of the observations are censored.

Development of germination rates is presented in Fig. 1 and 2, where percent of non-germinated seeds is plotted against time, showing survival distribution functions for different D-mannitol concentrations. Germination in the solutions with higher D-mannitol concentration was always much slower, with the only one exception.

Figure1. Survival distribution function estimates for the germination of variety Goranka

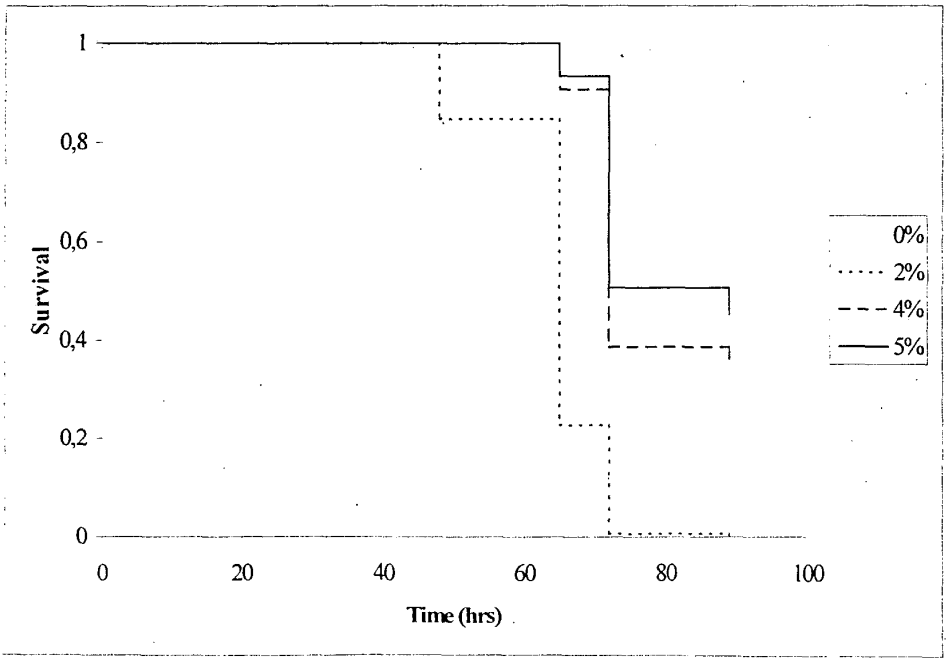
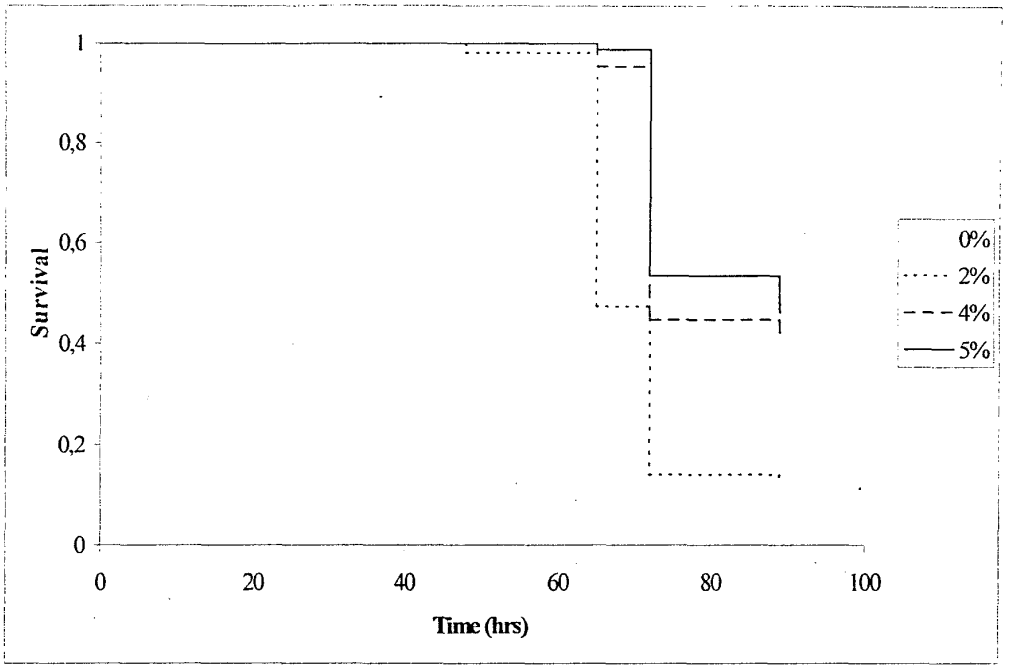


Figure 2. Survival distribution function estimates for the germination of variety Dukat



For both varieties it is obvious that the thickening of the D-mannitol solution increases the level of germination inhibition. Log rank tests for varieties Goranka (χ^2=398.74, p=0.0001) and Dukat (χ^2=286.11, p=0.0001) both proved statistical evidence that different D-mannitol concentrations in germination media caused different germination rates. Furthermore, the value of log rank statistics (Tab. 1), showing deviations from the median number of germinated seeds, decreased as D-mannitol concentration increased.

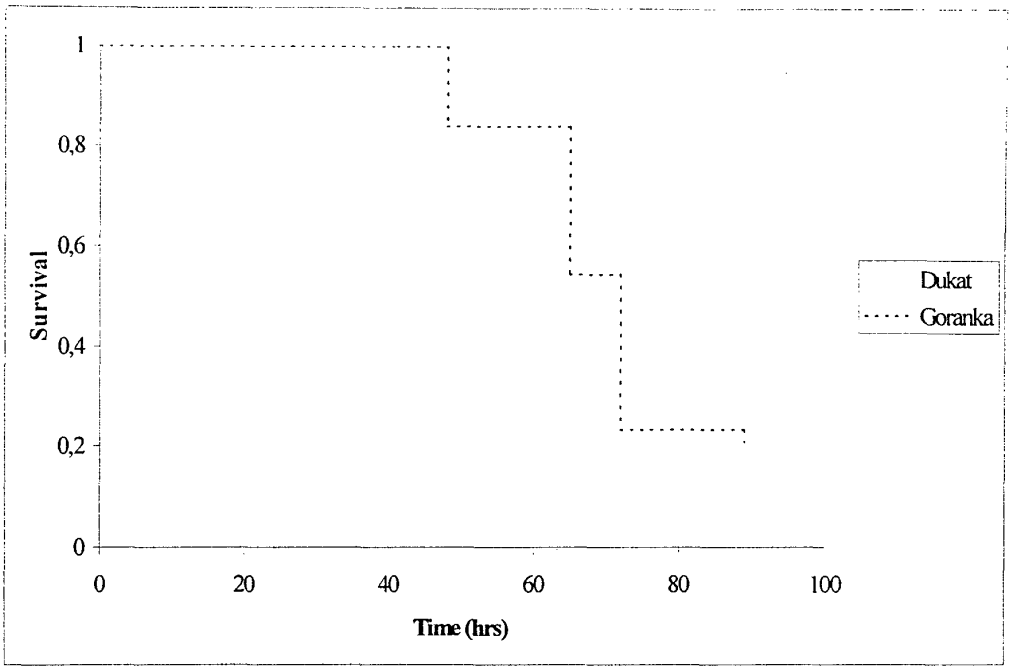
Table 1. Log rank statistics for D-mannitol concentrations

D-mannitol	Goranka	Dukat
0%	86.98	80.98
2%	61.55	41.23
4%	-69.32	-54.76
5%	-79.21	-67.46

There are similar patterns of germination rate variability in time and at different levels of D-mannitol concentration, therefore the test for overall dependence of germination inhibition on the germination media density could be carried out.

Two varieties showed different germination progress (Fig 3.), statistical significance of this difference tested in a log rank test (χ^2=15.83, p=0.0001). However, log rank test for the association of time with D-mannitol concentration over pooled strata was significant (χ^2=418.9, p=0.0001).

Figure 3. Survival distribution function estimates for the germination of varieties Goranka and Dukat



These results have given evidence that germination inhibition could be associated with osmotic pressure of the germination media. Differences between germination curves for two varieties are either a product of their different germination potential or due to the presence of other sources of inhibition. Further research should be done to investigate other possible mechanisms of inhibition, as well as the amount of inhibition due to osmotic pressure.

4. References

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