Considerations regarding barley germination

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Abstract. Naturally, germination takes place under the soil surface hidden from the eye of the observer. Therefore, the main goal of the experiment is the understanding of the germination process in correlation with the growing degree days. The experiment is performed on samples of 100 untreated barley seeds placed in Petri dishes, subjected to four fixed temperatures, 5°C, 10°C, 15°C and 20°C, in four incubators, continuously over a period of 6 days. From the 3rd to the 6th day, a sample is taken from each incubator and the following data is collected: the number of germinated seeds, the number of stems, the length of the longest radicle, and the length of the stem. The data analysis shows that the barley seed germination rate reaches at least 50% after reaching 30 degree-days.

1 Introduction

Germination is a continuous topic of research since it can be approached from different points of view. For example, from the perspective of the metabolites' localization in the first seven days of the barley germination [1]. As is known, barley (Hordeum vulgare L) is considered a model organism for the investigation of the germination process of cereals, being mentioned the use of germination and growth of barley as a test for the evaluation of fresh compost substrates or potential toxic effects of soil samples [2]. Barley is a species of cereal grain grown for its seed since the early agricultural human communities; nowadays, the world production of barley is the fourth among cereals, around 155 million tons [3]. Barley prefers relatively low temperatures to grow, and well-drained soil, because successful germination requires both water and oxygen; it is also relatively tolerant of drought and soil salinity. According to GAEZ Data Portal, the optimal temperatures for barley germination are between 16°C to 20°C, while absolute temperatures are 5°C and 30°C; other sources state that germination may occur at a larger interval between 3°C to 30°C [4], or 4°C to 37°C [5]; moreover, the minimum temperature for germination is 1°C - 2°C [5]. The speed of seed germination depends on the accumulated temperature, or growing degree-days, GDD. GDD is a very important tool in agriculture because it can be used to estimate the optimal time to plant, grow or harvest crops. In other words, GDD can predict the phenological stage of a plant [6, 7]. For this to be possible, two main requirements are necessary: i) there must be a standardization of the phenological phases of the plants, and ii) there must be a correlation

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between the phenological phase and the accumulated temperature. The first requirement is fulfilled by the development of the Cereal growth staging scales from early research in the mid-XX century [8] to more recent Zadoks [9] and BBCH scale [10]. The BBCH scale has become an international standard in agriculture after its recognition by the European and Mediterranean Plant Protection Organization (EPPO). This scale includes ten main stages of growth, and each of them has ten secondary stages. The stages are coded with a two-digit number, the first of which represents the principal growth stage. In the context of cereals, the principal growth stage 0, Germination, has only 7 secondary stages which characterize the development of the seed: 00: dry seed (caryopsis); 01: beginning of seed imbibition; 03: seed imbibition complete; 05: radicle emerged from caryopsis; 06: radicle elongated, root hairs and/or side roots visible; 07: coleoptile emerged from caryopsis; 09: emergence: coleoptile penetrates soil surface (cracking stage). Regarding the second requirement, here too a standardized element is needed - that of calculating the temperature accumulation. Traditionally, GDD is the arithmetic mean between the daily maximum, T_{MAX} and minimum, T_{MIN} temperatures from which the biological threshold temperature of the plant, T_{BASE} is subtracted (1), calculated over consecutive days [11].

$$GDD = [0.5\,(T_{MAX}+T_{MIN})] - T_{BASE} \quad (1)$$

Correlations between GDD and BBCH are reported in scientific papers [12, 13]; few of them refers to the germination stage. However, 109-145°C-days and 125-160°C-days are mentioned for the last stage of the Germination: Emergence: Leaf tip just emerging from above-ground coleoptyle for barley and wheat [6]. This paper refers to a study of the germination of barley seeds in correlation with the GDD. The purpose of this experiment is to determine the thermal time required for barley seeds to undergo the physiological process of germination. The results are expected to complete the information needed for a decision support system for precision agriculture. The study approaches a macro level, with unselected material, as it results from a harvest.

2 Materials and methods

100 seeds of barley are placed in a Petri dish (Fig. 1). Cardinal FD barley harvested by farmers in 2023, a Romanian. registered variety with six rows of grain on the barley spike which is supposed to have a reduced growing period [14], is used for the experiment.

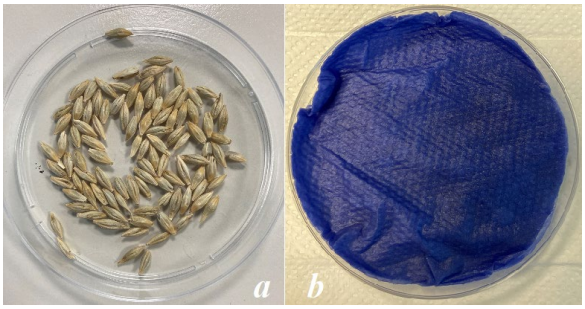

Fig. 1. The samples: a) dry seeds; b) after adding the water.

According to the product sheet from the Fundulea National Institute of Agricultural Research and Development, the creator of the Cardinal FD, the weight of 1000 grains varies between 46-50g, under optimal conditions of culture, respectively irrigated regime [15]. To evaluate the amount of water requested for germination the weight of the seeds is measured with a Digital Weighing scale EMB 500-1 (Kern), and the seeds humidity is determined with a Moisture Analyzer MAC 50/NH (Partner, Fig. 2).

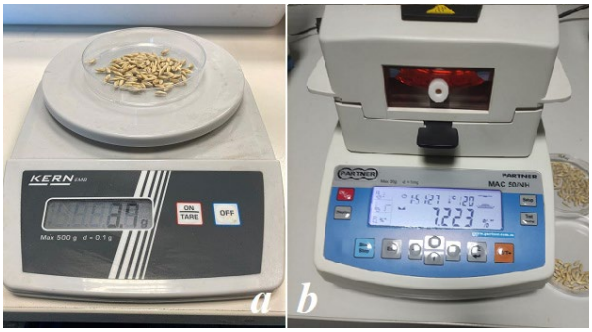

Fig. 2. Measuring instruments: a) digital weighing; b) moisture analyzer.

The average weight of the seeds is around 3.7g, while the seed moisture is 7.22%. Hence, 5ml of water is added to the Petri dish two times: over the seeds and after applying three rounded foils of cellulose paper over them. With this amount of water and by covering the seeds, we aim to ensure 100% humidity for the first 6 days of the experiment. Generally, barley seed germination takes about 2-4 days depending on several factors, especially type of the barley, temperature, humidity, quality of the seeds and aeration. 16 Petri dishes are prepared in this way. Four of them are inserted into four incubators programmed to maintain a constant temperature of 5°C, 10°C, 15°C and 20°C. The equipment used consists of a Cooled Incubator type FOC 120E Connect (Velp Scientifica) for 5°C, and three Digital Thermoelectric Incubators (Vevor) for the other three temperatures. The length of the radicles is measured manually using a Digital caliper (Burg Wächter) with a resolution of 0.01mm. After three days, daily until the 6th day, one Petri dish is removed from each incubator, the germinated seeds are counted and the lengths of their outgrowths are measured. The experiment took place for 7 days, twice. The seeds were not treated before the experiment.

3 Results and discussion

The temperature inside each incubator is continuously monitored using a wireless temperature sensors network. This highlighted a small difference between the temperature values set at the incubators and those actually measured inside them, but allowed them to be adjusted so that the average values of the measured temperatures during the two experimental periods are very close to the targeted ones: 5.24°C, 9.52°C, 14.83°C, 19.83°C, and 5.00°C, 10.08°C, 14.78°C, 20.163°C, respectively.

3.1 Germination rate

It was observed in the period from the 3rd to the 6th day that a significant number of seeds developed radicles, and another smaller number developed stems. As seen in Fig. 3a, for a

sample Petri dish exposed at 10°C temperature after the 6th day, only 4 seeds developed stems.

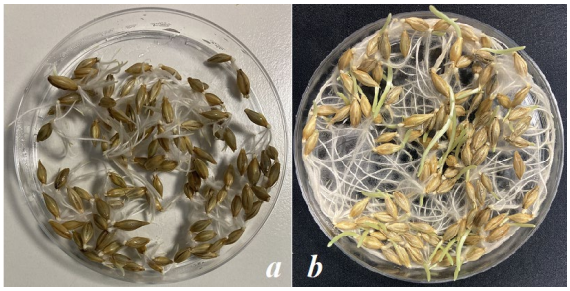

Fig. 3. Sample seeds after the 6th day exposed at: a) 10°C, b) 15°C.

On the other hand, since it is found that there are seeds exposed to the 5°C temperature that germinated progressively over time, this means temperature accumulation. According to (1) this would not be theoretically possible if the biological threshold temperature of the barley of 5°C is taken into account in the GDD calculation. As a result, considering some reports that barley germination can also take place at 1°C, the value of 0°C is taken as T_{BASE} in the GDD calculation. The germination rates for all samples are presented in Table 1, while Fig. 4 presents the evolution of barley seed germination in time from the 3rd to the 6th day for all four temperatures.

Table 1. The percentage of germinated seeds.

Day	Germination rate [%]			
	5°C	10°C	15°C	20°C
3rd	5	53.5	68.5	66.5
4th	30.5	71	72	81
5th	39.5	70	74.5	75
6th	50	67.5	74	77.5

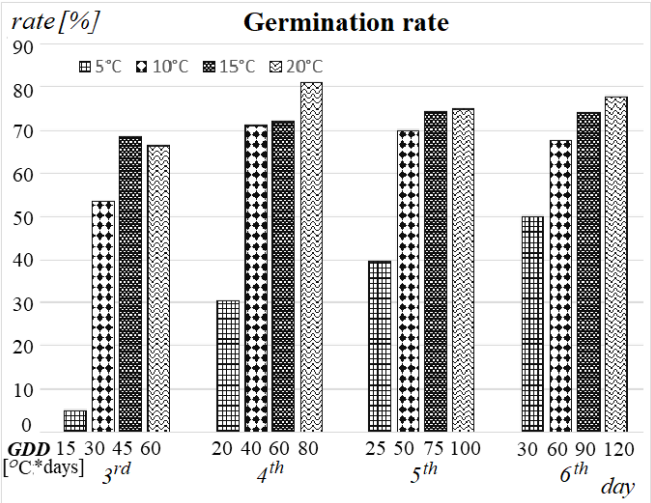

Fig. 4. The graphical representation of the germination rate.

Under each column of germination rate it is noted the growing degree days. It can be seen that the lowest germination rate occurs at the samples kept at 5°C temperature after 3 days (GDD = 15°C-days), but it increases day by day. It can be said that the secondary growth stage 05: Radicle emerged from caryopsis, according to BBCH scale, begins between 15-20°C-days. This is due to the fact that this temperature is well below the optimum temperature range for barley. However, it can be noted that this sample, after accumulating 30°C-days presents a germination rate (50%) very close to the sample kept at 10°C after 3 days, when GDD is also 30 (53.5%). The same observation can be made for GDD = 60°C-days: the sample exposed at 20°C after 3 days presents a germination rate (66.5%) very closed to the sample exposed at 10°C during 6 days (67.5%), and not far from the sample exposed at 15°C after 4 days (72%). Highest germination rates are obtained for the samples exposed at 15°C and 20°C; after four days all present rates greater than 70%, higher for the latter sample, which is in accordance with the optimum temperature range. The variation of the germination rate with GDD is not linear but rather polynomial of the 4th degree, which presents the best coefficient of determination, R2 = 0.9735 (Fig. 5).

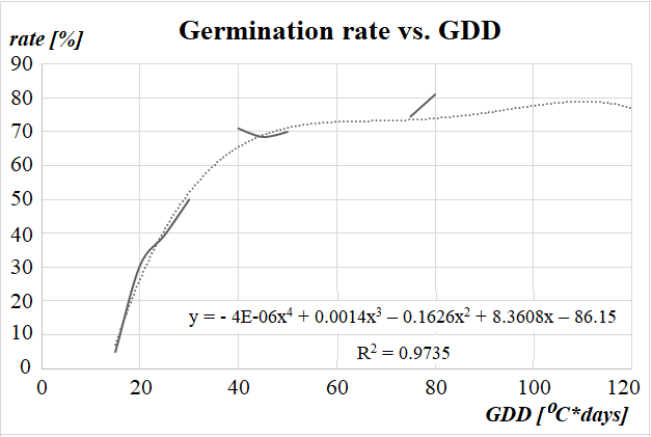

Fig. 5. Germination rate evolution with GDD.

Although the results of barley samples permanently exposed to a temperature of 5°C are not favorable from the above criterion, this temperature is reported as beneficial to wheat for the removing of embryos dormancy, thus allowing a high germination percentage [16].

3.2 Stem rate

After the fourth day some barley seeds presented stem growing. The rate of stem development is presented in Table 2, while Fig. 6 presents a chart of the data where the stem rate is represented as a function of the temperature.

Table 2. The percentage of stem development.

Day	Stem rate [%]			
	5°C	10°C	15°C	20°C
3rd	0	1.5	4	27
4th	0	0.5	14	24.5
5th	0	1	35	36.5
6th	0	2.5	46.5	47

It is observed that no stem is growing at 5°C temperature, at least within 6 days period. On the other side, the stem rate is increasing after each day, the greater incubator temperature, the higher stem rate. Here, no correlation can be made from the GDD point of view since for 60°C-days the stem rate has too different values: 2.5%, 14% and 27%.

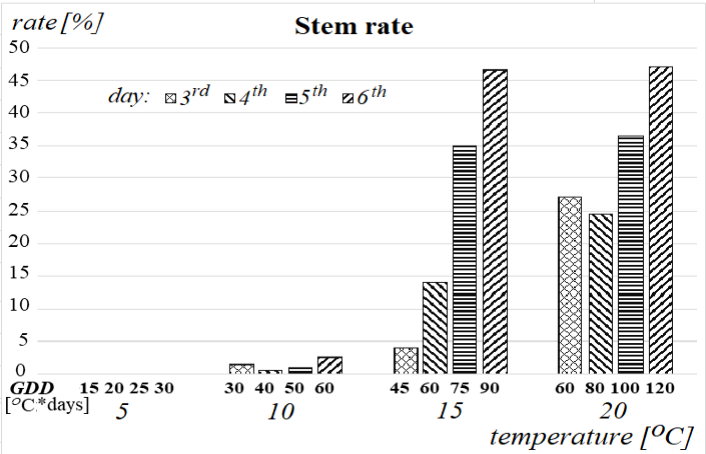

Fig. 6. The graphical representation of the stem rate.

3.3 The length of radicles

Table 3 and Fig. 7 presents the average radicle lengths for all germinated seeds and the graphical representation of its variation with the temperature.

Table 3. The average length of the radicles.

Day	Length [mm]			
	5°C	10°C	15°C	20°C
3rd	0.55	3.96	11.58	20.12
4th	0.82	6.04	16.99	19.04
5th	1.70	10.40	23.25	25.86
6th	2.32	12.30	28.44	27.51

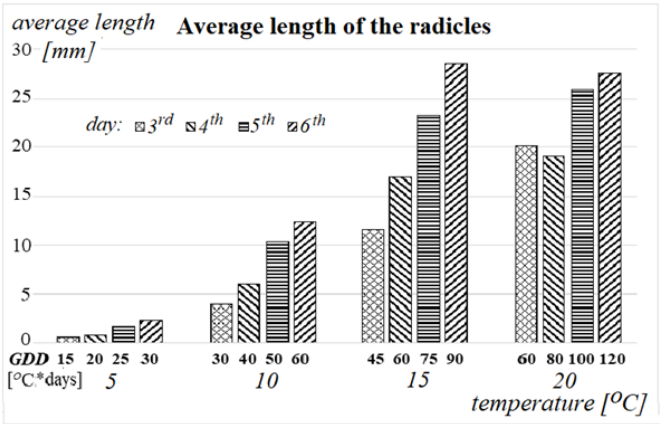

Fig. 7. The graphical representation of the stem rate.

Depending on the temperature in the incubator and the number of days of germination, the seeds developed radicles. The number of radicles may be from one to several in the same Petri dish. The length of the radicles also very different in the same sample (Fig. 8).

Fig. 8. Samples of seeds with: a) two radicles, b) four radicles.

Also, even after 6 days, even at a temperature of 20°C, there are seeds that have not germinated. This could be caused by the presence of dead seeds or diseased seeds that have infested the entire sample disrupting their development. A singularity is the identification of a single seed in the sample subjected to 10°C after 4 days of germination which, in addition to the fact that it has a very long root compared to the other seeds, has also developed an unexpectedly large stem. Therefore, the analysis according to the maximum root length does not seem to be relevant, but rather according to the average root length of all the seeds germinated at the same temperature. As expected, the average length of the radicles increases with the number of days of germination and with the temperature. But a fair correlation between GDD and average length of the radicles cannot be made. For the same GDD (e.g.: 60), there are three different values (12.30 mm, 16.99 mm, and 20.12 mm). Moreover, for some smaller GDDs correspond to longer average lengths than for larger GDDs (e.g: 75 versus 80°C-days, or 50 versus 45°C-days), even if within a reasonable range. However, this could also be a shortcoming of the experiment, because if there are several roots, only the longest of them was measured. In order to more correctly evaluate the germination power of the seed, all the roots should have been measured.

3.4 The length of the stems

Since the seeds generated only one stem, it is expected that there is a clearer correlation here. The results of the measurement and calculations are presented in Table 4 and Fig. 9.

Table 4. The average length of the stems.

Day	Length [mm]			
	5°C	10°C	15°C	20°C
3rd	0	2.87	2.32	7.69
4th	0	1.87	6.64	7.43
5th	0	3.80	10.35	17.58
6th	0	7.04	11.81	11.95

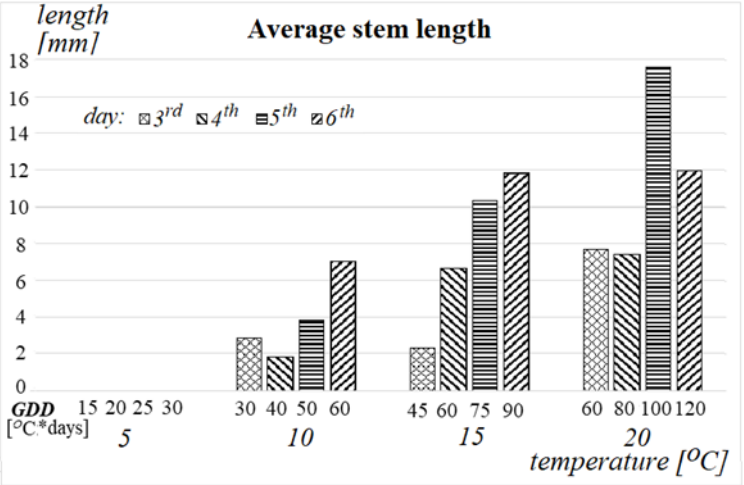

Fig. 9. The graphical representation of the average length of stems.

The seeds exposed to 5°C temperature do not present stem growing within the experimental period, even after 30°C-days, when, apparently, the first stems are developed (10°C sample, after three days). An explanation can be that biochemical reactions do not vary linearly with temperature.

4 Conclusions

An experiment on the germination of barley seeds depending on the temperature was carried out. The material used for experimentation came directly from the producer's harvest and the samples were not selected or treated. As a result, the ideal conditions for experimentation were not created, but real ones, so that in some Petri dishes, diseases were triggered that may have affected the germination. For barley seeds phenological stage 02 occurs after GDD reaches 15-20°C-days. It was found that the germination rate of the seed sample reaches 50% after the accumulation of 30°C-days. The variation of the germination rate with GDD is not linear but polynomial of the 4th degree. In the context of very fast temperature variations and with extreme values due to climate changes, it is rather useful to take into account the accumulated degrees (°C-days) instead of the number of days when assessing the phenological state.

This work was supported by a grant of the Ministry of Research, Innovation and Digitalization, CCCDI-UEFISCDI, project number PN-III-P2-2.1-PTE-2021-0363, within PNCDI III.

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