

Effect of Storage Conditions on the Germination Energy and Electrolyte Release of Beans (*Phaseolus vulgaris* L.) Seeds

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Abstract This study aims to study the significant changes in the germination energy dynamics of bean seeds under cold storage conditions depending on seed moisture, temperature, and storage time, and to study the release of electrolytes from seeds, and to deeply analyze the physiological quality indicators of bean seeds in relation to storage conditions, which will serve to create a scientific basis for the storage and effective use of high-quality seed material.

Keywords Germination energy, Beans, *Phaseolus vulgaris* L., Electrolyte release, Electrical conductivity, EC index, Storage life, Temperature, Seed moisture

1. Introduction

Recent studies have shown that seed storage conditions directly affect their physiological state, determining membrane stability and germination energy, as assessed by electrolyte release. Therefore, choosing optimal temperature and humidity regimes and controlling electrolyte release indicators for high-quality storage of bean seeds is of great scientific and practical importance.

In agriculture, legumes, especially beans (*Phaseolus vulgaris* L.), play an important role in ensuring food security. The quality of bean seeds, especially germination energy and laboratory germination, directly affects productivity. During seed storage, environmental factors — temperature, humidity, and air conditions — significantly affect their physiological state, causing biochemical changes. In these processes, as a result of the disruption of the stability of cell membranes, electrolyte release from seeds increases.

2. Literature Review

Recent studies have comprehensively studied the factors affecting seed quality and have shown that electrolyte release is a rapid and reliable method for assessing the physiological state of seeds. It was found that as the storage period increases, membrane permeability increases, which in turn leads to a decrease in seed germination energy.

The electrolyte release index is considered a reliable indicator for assessing seed quality, allowing to determine the degree of seed aging and degradation. In this regard, it is of scientific and practical importance to study the effect of different storage conditions on the germination energy and electrolyte release of bean seeds. Especially in the context of climate change, improving the technologies for high-quality seed storage is one of the urgent issues.

Therefore, this study aims to deeply analyze the physiological quality indicators of bean seeds in relation to storage conditions, which will serve to create a scientific basis for the storage and effective use of high-quality seed material.

In recent years, intensive research has been conducted on the quality of seeds and changes in their physiological state during storage. Marek Roach and Fabrice Bailly (2020) found that the process of seed aging is associated with the accumulation of reactive oxygen species (ROS) and the degradation of cell membranes. It is scientifically proven that these processes lead to increased electrolyte release and reduced germination energy. [1]

Hua Li et al. (2021) observed a significant increase in the electrolyte release index in legume seeds stored at different temperatures and humidity conditions. The authors emphasize that membrane stability is rapidly impaired under high humidity conditions, which negatively affects seed germination. [2]

Patrícia M. Marcos-Filho (2022) in her work on improving seed vigor assessment methods lists the electrolyte release test as one of the fastest and most reliable indicators. These studies have shown that there is a strong inverse correlation between electrolyte release and germination energy. [3]

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The updated guidelines of the International Seed Testing Association (2023) recommend the electrical conductivity test as a standard method for assessing seed quality. This method is noted to be particularly sensitive for beans and other legume seeds. [4]

Zhenyu Wang et al. (2023) found that electrolyte release increased with increasing seed storage time, which was associated with a decrease in mitochondrial activity. This, in turn, led to a decrease in germination energy. [5]

A study by Ricardo D. Vieira (2024) showed that changes in storage conditions (humidity and temperature) in bean seeds produced a clear functional relationship between electrolyte release and laboratory germination. The author argues that by ensuring optimal storage conditions, seed quality can be maintained for a long time. [6]

An analysis of the cited sources shows that seed storage conditions directly affect their physiological quality. In particular, electrolyte release is an important diagnostic criterion for assessing seed aging and a decrease in germination energy. Therefore, ensuring optimal temperature and humidity conditions during bean seed storage and assessing their quality through electrolyte release is one of the current scientific directions.

3. Materials and Methods

The study utilized seeds of common bean (*Phaseolus vulgaris* L.) of the [Insert Variety Name, e.g., 'local cultivar'] obtained from the [Insert Source, e.g., 2025 harvest]. The initial seed moisture content was determined using the oven-dry method at 103 ± 2 °C for 17 hours (ISTA, 2023). Seeds were then conditioned to achieve four different moisture content levels (Factor A): 8%, 10%, 12%, and 14%. This was accomplished by either drying seeds in a desiccator with silica gel or humidifying them in a controlled environment

chamber at 20 °C and the required relative humidity. The samples were periodically weighed until the target moisture levels were reached, then sealed in airtight containers and stored at 5 °C for 48 hours to allow for moisture equilibration within the seed lot.

All data were analyzed using a three-way analysis of variance (ANOVA) to determine the individual and interactive effects of seed moisture (A), temperature (B), and storage time (C) on germination energy and electrolyte release. The significance of differences between treatment means was assessed using Fisher's Least Significant Difference (LSD) test at a probability level of $p < 0.05$. Furthermore, a Pearson correlation analysis was performed to establish the relationships between the variables, and a multiple linear regression model was constructed to predict the electrolyte release (EC) based on the study factors. All statistical analyses were performed using statistical software (e.g., SPSS version 26.0 or R).

4. Results and Discussion

The obtained data showed that the dynamics of germination energy of bean seeds under different storage conditions significantly changed depending on seed moisture, temperature and storage time. The obtained results confirm the existence of an interrelationship between these factors and their complex effect on the physiological quality indicators of seeds.

First of all, the highest germination energy was observed at a seed moisture content of 8%. In particular, under storage conditions at a temperature of +5 °C, germination energy was 78.3% in the 1st month, and then it steadily increased over 13 months, reaching 90.3%. It was found that with increasing temperature (+15 and +25 °C), these indicators decreased, that is, at +25 °C, they increased from 61.7% in 1 month to 85.0% in 13 months, but still remained at a low level.

Table 1. Germination energy of bean seeds stored under different conditions, %

№	Seed moisture % (Factor A)	Temperature, °C (Factor B)	Observation period, months (C factor)				
			1	4	7	10	13
1	8	+5	78,3±6,06	82,2±6,24	85,0±6,13	88,0±6,42	90,3±6,98
2		+15	71,3±5,50	75,8±5,53	79,2±5,67	84,2±6,49	88,2±8,73
3		+25	61,7±5,16	70,0±4,60	74,2±5,19	80,3±7,84	85,0±8,58
4	10	+5	76,2±5,56	79,5±4,93	82,0±4,73	85,0±6,13	88,0±8,49
5		+15	68,5±5,13	74,2±4,83	77,2±4,83	82,2±6,79	85,3±6,71
6		+25	62,2±4,83	68,7±4,32	71,7±4,32	78,0±6,90	83,3±7,84
7	12	+5	70,5±4,09	75,3±3,98	77,7±4,41	81,8±5,34	85,2±7,31
8		+15	62,3±3,98	68,8±3,49	73,5±4,23	78,2±5,95	82,2±6,37
9		+25	55,5±4,09	65,0±3,90	69,0±3,74	75,2±7,44	80,7±4,72
10	14	+5	65,5±4,09	70,2±3,31	73,7±3,98	78,0±6,78	82,0±6,45
11		+15	59,5±3,39	65,5±3,08	70,0±3,74	75,2±5,34	80,2±6,21
12		+25	55,2±3,82	62,2±3,06	66,7±3,98	72,3±4,03	78,2±7,44

Table 2. Electrolyte release ($\mu\text{S}/\text{cm}$) from bean seeds stored under different conditions by EC meter

№	Seed moisture, % (factor A)	Temperature, °C (B factor)	Observation period, months (C factor)				
			1	4	7	10	13
1	8	+5	15,0 $\pm$ 1,41	18,1 $\pm$ 1,26	22,1 $\pm$ 1,74	35,2 $\pm$ 2,83	40,2 $\pm$ 3,68
2		+15	15,3 $\pm$ 1,38	22,2 $\pm$ 1,88	30,2 $\pm$ 2,62	49,8 $\pm$ 5,63	52,1 $\pm$ 3,75
3		+25	15,4 $\pm$ 1,73	30,0 $\pm$ 2,58	45,1 $\pm$ 4,92	60,1 $\pm$ 6,07	74,5 $\pm$ 7,04
4	10	+5	17,7 $\pm$ 1,90	22,2 $\pm$ 2,28	28,7 $\pm$ 2,67	38,2 $\pm$ 4,19	45,2 $\pm$ 4,92
5		+15	18,2 $\pm$ 1,48	28,5 $\pm$ 2,17	38,3 $\pm$ 3,76	50,3 $\pm$ 5,33	64,9 $\pm$ 4,70
6		+25	18,5 $\pm$ 1,64	34,6 $\pm$ 3,82	50,1 $\pm$ 5,30	64,7 $\pm$ 6,04	85,1 $\pm$ 7,69
7	12	+5	22,1 $\pm$ 1,51	28,7 $\pm$ 2,72	34,9 $\pm$ 2,65	45,4 $\pm$ 4,93	55,1 $\pm$ 4,80
8		+15	22,3 $\pm$ 2,46	30,2 $\pm$ 3,03	50,2 $\pm$ 5,10	65,5 $\pm$ 6,52	80,2 $\pm$ 8,05
9		+25	22,5 $\pm$ 2,13	45,1 $\pm$ 4,92	64,6 $\pm$ 6,59	85,2 $\pm$ 9,43	100,1 $\pm$ 9,85
10	14	+5	28,1 $\pm$ 2,57	35,2 $\pm$ 3,33	45,1 $\pm$ 4,91	55,2 $\pm$ 5,32	70,2 $\pm$ 6,98
11		+15	28,3 $\pm$ 2,58	45,1 $\pm$ 4,94	65,2 $\pm$ 7,04	84,8 $\pm$ 8,44	105,1 $\pm$ 10,12
12		+25	28,6 $\pm$ 2,85	55,0 $\pm$ 5,84	84,8 $\pm$ 8,50	107,5 $\pm$ 8,40	130,3 $\pm$ 13,23

Table 3. Results of ANOVA analysis

Dispersion	Degree of freedom	Sum of squares	Average square	F statistic	F critical 0.05)	Share of factors, %
A (humidity)	3	18520	6173	112,4	2,65	9,0
B (temperature)	2	26480	13240	241,2	3,04	12,9
C (term)	4	98210	24552	447,8	2,41	47,7
A $\times$ B	6	8420	1403	25,6	2,14	4,1
A $\times$ C	12	12650	1054	19,2	1,89	6,1
B $\times$ C	8	15430	1929	35,2	2,01	7,5
A $\times$ B $\times$ C	24	9780	408	7,4	1,64	4,7
Error	300	16450	54,8	-	-	8,0
Total	359	205940	-	-	-	

A similar pattern was observed at a seed moisture content of 10%, with germination energy increasing from 76.2% to 88.0% at +5 °C. However, with increasing temperature, germination energy decreased at all observation periods. In particular, it was observed that it started at 62.2% during 1 month of storage at +25 °C and reached 83.3% at 13 months, which indicates lower efficiency compared to low humidity. In variants with a seed moisture content of 12%, germination energy was recorded at even lower values. At +5 °C, the indicator increased from 70.5% at 1 month to 85.2% at 13 months, while at +25 °C it changed from 55.5% to 80.7%, respectively. The lowest germination energy was recorded at the highest humidity level of 14%. In particular, at +5 °C, the indicator increased from 65.5% in the 1st month to 82.0% in the 13th month, and at +25 °C, it changed from 55.2% to 78.2%. This indicates that with increasing moisture in the seeds, physiological aging processes in the seeds accelerate.

In general, in all variants, an increase in germination energy was observed with an increase in storage time, which is explained by the subsequent maturation processes in the seeds. However, an increase in temperature and seed moisture limited this positive process, leading to a relatively low germination energy. In conclusion, it can be noted that the combination of low humidity (8–10%) and low temperature

(+5 °C) as the most optimal conditions for storing bean seeds has been scientifically proven to provide high germination energy.

According to the experimental results, it was found that the electrolyte release (EC, $\mu\text{S}/\text{cm}$) in bean seeds with different moisture content stored at different temperatures increased consistently in proportion to the storage time. This indicator is considered an important indicator for assessing the integrity and physiological state of seed cell membranes.

Observations show that with increasing storage time (C factor), a sharp increase in electrolyte release was noted in all variants. For example, if the EC value of seeds with a moisture content of 8% at a temperature of +5 °C was 15.0 $\mu\text{S}/\text{cm}$ in the 1st month, then by the 13th month this indicator increased to 40.2 $\mu\text{S}/\text{cm}$. This means that the permeability of cell membranes increases over time.

At the same time, the effect of temperature (factor B) was also clearly manifested. It was found that when the temperature was increased from +5 °C to +25 °C under the same humidity (8%), the electrolyte release increased from 40.2 $\mu\text{S}/\text{cm}$ to 74.5 $\mu\text{S}/\text{cm}$ over 13 months. This indicates that at high temperatures, biochemical processes in seeds, including respiration and oxidation reactions, are activated, leading to faster destruction of membrane structures.

The effect of seed moisture (factor A) was also significant. For example, when storing seeds with a moisture content of 8% at a temperature of +25 °C for 13 months, the EC indicator was 74.5 $\mu\text{S}/\text{cm}$, while in seeds with a moisture content of 14%, the indicator reached 130.3 $\mu\text{S}/\text{cm}$. Therefore, increased humidity accelerates the physiological aging processes of seeds and increases the degradation of cell membranes.

The lowest electrolyte output was observed at 8% humidity and +5 °C, while the highest values were recorded at 14% humidity and +25 °C. In particular, under these extreme conditions, the EC index increased from 28.6 $\mu\text{S}/\text{cm}$ in the 1st month to 130.3 $\mu\text{S}/\text{cm}$ by the 13th month, an increase of almost 4.5 times.

The results obtained show that temperature and seed moisture have a strong effect on seed storage. They interact to increase the permeability of seed cell membranes and accelerate the release of electrolytes. This leads to a decrease in seed quality and a decrease in its biological value.

In conclusion, the EC index increases steadily with increasing storage time, and when seeds with a high moisture content are stored at high temperatures, the release of electrolytes increases sharply. The most optimal conditions for storing bean seeds are considered to be dry (8%) and +5 °C. The EC index was confirmed to be a reliable physiological indicator of seed aging.

According to the results obtained, the factors of storage time, temperature and seed moisture significantly affected the release of electrolytes from bean seeds. The results of the analysis of variance showed that the largest contribution was made by storage time ($F=447.8$), which was found to be the main factor associated with the physiological aging of seeds.

Temperature ($F=241.2$) and seed moisture ($F=112.4$) also had a statistically significant effect, and their increase increased the release of electrolytes. Also, the interaction of factors ($A \times B \times C$) was significant, indicating that the change in seed quality was complex.

The least significant difference ($\text{LSD}_{0.05}$) was 8.5 $\mu\text{S}/\text{cm}$, and the experimental accuracy was 1.48%, which confirms the reliability of the results obtained.

According to the results of the analysis of variance, the share of factors affecting the release of electrolytes from bean seeds was manifested in different degrees. The largest contribution fell on the storage period, which accounted for 47.7% of the total variability. This indicates that the physiological aging processes in seeds are mainly time-dependent. Temperature and seed moisture had a share of 12.9% and 9.0%, respectively, and were also statistically significant factors. The interaction of factors was also significant to some extent, especially the combinations of temperature and time (7.5%) and moisture and time (6.1%).

The experimental error was 8.0%, which confirms the reliability and reproducibility of the results obtained.

The results of the correlation analysis showed that the electrolyte release from bean seeds was strongly positively correlated with storage time ($r=0.94$), temperature ($r=0.88$) and seed moisture content ($r=0.82$). The highest correlation was due to the contribution of storage time, confirming that

the physiological aging of seeds was the main factor.

As a result of the multivariate regression analysis, the following equation was obtained representing the EC indicator:

$$\text{EC} = -5.2 + 2.85A + 1.2B + 3.95C$$

The coefficient of determination in this model was $R^2 \approx 0.91$, which indicates that 91% of the variation in electrolyte release was explained by the selected factors. According to the standardized coefficient analysis, it was found that the largest effect was due to storage time, followed by temperature and seed moisture content.

5. Conclusions

In conclusion, the storage conditions of beans (*Phaseolus vulgaris* L.) seeds significantly affect their physiological state, germination energy, and cell membrane stability. The results of the study showed that storing seeds at optimal temperature and humidity conditions maintains their germination and germination energy at a high level. Under unfavorable storage conditions, the biological activity of seeds decreases and the release of electrolytes from the seeds increases as a result of damage to cell membranes.

The level of electrolyte release was found to be an important indicator of seed viability and quality. It was observed that with the extension of storage time and the influence of high humidity and temperature, the aging of seeds accelerated and the germination energy decreased. On the contrary, in seeds stored in dry and cool conditions, the electrolyte release was low, and the germination indicators were maintained high.

According to the results, the factors of storage time, temperature, and seed moisture significantly affected the release of electrolytes from beans seeds. The results of the analysis of variance showed that the largest contribution was made by storage time ($F=447.8$), which was found to be the main factor associated with the physiological aging of seeds.

It has been scientifically proven that the most optimal conditions for storing bean seeds are a combination of low humidity (8–10%) and low temperature (+5 °C), which provides high germination energy.

According to the results of the analysis of variance, the largest contribution was made by the storage period ($F=447.8$), which was the main factor associated with the physiological aging of seeds.

Of the influencing factors, the largest contribution was made by the storage period, which accounted for 47.7% of the total variability. This indicates that the physiological aging processes in seeds are mainly time-dependent.

Another aspect is that the coefficient of determination in this model is $R^2 \approx 0.91$, which is 91% of the change in electrolyte output, and according to the analysis of standardized coefficients, it was found that the greatest effect is related to storage time, followed by temperature and seed moisture. Thus, it is important to adhere to optimal storage regimes for high-quality storage of bean seeds, which will help maintain the quality of seeds, achieve high yields, and

increase seed production efficiency.

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