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# Contribution to the Optimization of Methods for Vigor Seed Evaluation of *Camelina sativa* (L.) Crantz

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**Abstract:** Camelina, a traditional oil-producing plant, has gained global interest due to the high-quality oil found in its seeds. It has numerous applications, including human dietary consumption, aviation biofuel, and biodiesel production. Seed quality testing is crucial for identifying suitable seed batches for market sale. Currently, vigor tests have been established for a limited selection of commercially cultivated plant species, including camelina. This study aims to assess seed vigor and contribute to the development and validation of methods/tests for reliable vigor assessment. The experiment used two camelina genotypes developed at the Institute of Field and Vegetables Crops in Novi Sad, Serbia. The findings revealed a noteworthy reduction in germination percentages for both genotypes across all the conducted tests, as compared to the conventional laboratory germination. Simultaneously, there was a notable increase in abnormal seedlings. However, no statistically significant differences in the values of growth parameters were observed among the applied tests. In summary, the reduced seed vigor values indicate potential issues with this trait, despite generally sound germination. Additionally, the preliminary findings and methodology developed for testing the camelina seed vigor highlighted the need for optimization when applying these tests to other species to ensure their reliability and applicability.

**Keywords:** *Camelina sativa*; germination; seed; seed ageing; vigor; seed quality; methods



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## 1. Introduction

Camelina, *Camelina sativa* (L.) Crantz, a traditional oil-producing plant, was grown in Europe until approximately the mid-20th century, at which point it gradually started to be replaced by higher-yielding species like oilseed rape [1]. As the demand for alternative and more stable sources of oil to meet human requirements continues, the cultivation of this *Brassicaceae* family plant has been resuscitated [2] and has garnered growing global attention in recent decades. This heightened interest can be attributed to the substantial quantity of high-quality oil found in its seeds, ranging from 30% to 47% of their dry weight [3]. Seeds boast nutritive value due to their rich content of essential fatty acids (FAs) and natural antioxidants, including PUFAs, phenolic compounds, tocopherols, carotenoid pigments, vitamins, phospholipids, and phytosterols. Camelina has found extensive utility in various applications, including human dietary consumption, as biofuel, in hydraulic fluids and biopolymers, as feed, and in cosmetics [4]. The high value of cold-pressed camelina seed oil comes from the presence of bioactive compounds that have a beneficial effect on human health [5]. Capitalizing on its elevated seed oil content and robust oil yield per hectare, camelina proves to be a resource-efficient candidate for the production of renewable fuels [6]. Additionally, the residual seed cake is an appreciated resource rich in proteins and omega-3 fatty acids, making it a suitable option for animal feed [7].

The presence of both winter and spring annual varieties of camelina sativa offers versatility in agricultural usage, allowing for its cultivation as a winter cover crop or a short-cycle spring crop, thereby expanding its potential applications in farming [1]. Due to its relatively limited need for agronomic inputs and high tolerance toward abiotic stresses, camelina is often suggested for cultivation in marginal soils [4]. Nevertheless, in order to achieve successful production, the genotype choice, characterized by superior seed quality, is required. In other words, the imperative for success in agricultural production lies in employing seeds with high physiological potential, which notably helps in swiftly establishing plant stands and initiating the robust development of seedlings in the field [8]. Moreover, climate change presents a big challenge to global oilseed crop production, marked by increased temperatures, heightened atmospheric $CO_2$ levels, severe climatic events, reduced water availability, and often high pressure from pests and diseases [9]. The obvious alterations in climate patterns, characterized by the increased intensity and frequency of environmental stresses [10], have the potential to impact various phases of plant growth and development [11], thereby altering the physiological quality of seeds. Germination plays a critical role in the crop life cycle, as only plants capable of making it through this stage will survive [12]. It marks the beginning of growth and development, laying the foundations for the entire crop cycle and influencing the subsequent stages of plant maturity, reproduction, and overall productivity.

Evaluating the quality and viability of seeds is crucial for identifying which seed lots are suitable for market sale. Therefore, it is essential to give careful thought to the techniques and methods employed during seed quality testing. These assessments help determine the overall quality of the seed lot, specifically by ascertaining seed viability and seed vigor. These two parameters can differentiate subtle deviations in the degree of deterioration among seed lots exhibiting comparable germination percentages [13]. This nuanced approach ensures that only the highest-quality seeds are selected for market sales, contributing to the overall success and reliability of agricultural practice. Currently, vigor tests have been established only for a limited selection of commercially cultivated plant species, with limited advancement in expanding the methodology to include a broader range of plant species, including camelina. The inclusion of seed vigor in the development of effective seed quality control strategies is justified by its substantial impact on both seedling establishment in a dynamic natural environment and the capacity for long-term storage [8]. In other words, vigor is a quantitative attribute and is influenced by various factors related to overall seed performance, encompassing the speed and consistency of seed germination, seedling growth, emergence in adverse environmental conditions, and performance following storage. At the same time, further development in this field would certainly contribute to the ongoing widespread promotion of this highly promising species. By expanding the vigor testing methodology to include more plant varieties, particularly those like camelina, the agricultural community can unlock new possibilities for enhancing seed quality, which, in turn, can positively impact the broader adoption and success of this promising crop.

To the best of the authors' knowledge, this study represents the first investigation into the vigor assessment of camelina seeds. Given the lack of established protocols and precise methodologies for evaluating seed vigor in this particular species, this research endeavors to achieve the following objectives: (i) to conduct an assessment of seed vigor to gain a deeper insight into seed quality; and (ii) to contribute to the development and validation of methods/tests that would enable a reliable assessment of vigor. These objectives are set to bridge the existing gaps in our understanding of this crop vigor assessment and to establish reliable procedures for future research and agricultural applications.

## 2. Materials and Methods

The experiment, which utilized two camelina genotypes developed at the Institute of Field and Vegetables Crops in Novi Sad, Serbia (NS Slatka and NS Zlatka), was conducted between September 2022 and September 2023.

### 2.1. Germination Assay

For the standard laboratory germination test, the seeds were placed on filter paper (120 g/m$^2$, Filtros Anoia S.A. (Barcelona, Spain) moistened with the optimal amount of distilled water (10 mL) in glass Petri dishes (90 mm × 15 mm). Subsequently, they were transferred to a germination chamber at a temperature range of 20 to 30 °C for 10 days [14]. After that period, determination of the percentage of germination, evaluation of the appearance of atypical seedlings structures, and measurement of the growth parameters (shoot and root length) were carried out. When assessing the germination percentage and abnormality of seedlings, the condition of vital structures, including the root system, shoots, and cotyledons, was examined. Seedlings that exhibited minimal damage, whether undamaged or only slightly affected, as well as those showing signs of secondary infection, constituted the percentage of germination. Seedlings were considered atypical if any of these structures were absent, significantly damaged, or deformed, or if they were decayed due to primary infection. These criteria were consistently applied across all conducted vigor tests.

### 2.2. Vigor Assessments

As part of the vigor assessment, the Accelerated Ageing test (AA test), the cold test, the Hiltner test and the Controlled Deterioration test (CD) were applied. The AA test was performed by placing the seeds in a water bath with a relative humidity of 100% using two temperature variants: temperature of 41 °C for a period of 72 h (AA1) and temperature of 43 °C for a period of 72 h (AA2). The cold test seeds were placed in a mixture of soil and sand in a 2:1 ratio and exposed to a temperature of 5–10 °C for 7 days, followed by a temperature range of 25 °C for 6 days. In the Hiltner test, moist sand served as the substrate upon which seeds were positioned and covered with a 3 cm thick layer of broken brick. The seeds underwent a cold treatment at 5–10 °C for 6 days, followed by exposure to a temperature range of 25 °C for 10 days [15] (Hampton and TeKrony, 1995, with slight modification).

The CD was conducted according to the protocol recommended for *Brassica napus* by the International Association for Seed Testing [14]. During this test, seeds are exposed to high temperatures at a predetermined and continuously increasing moisture content. This controlled environment facilitates rapid deterioration, enabling the assessment of seed vigor. Before subjecting the seeds to elevated temperatures, their moisture content is intentionally elevated, ensuring a consistent level of deterioration across all tested samples. Seeds with high vigor maintain sustained germination rates even under deteriorative conditions, while seeds with low vigor undergo a reduction in germination capability. The seeds were placed on moistened (but not wet) filter paper. Equation (1) outlines the expected seed weight at a moisture content of 20%.

$$\text{Weight of seed replicate at 20\% mc} = \text{initial seed weight} \times \frac{\text{100–initial seed mc}}{\text{100–20\%}} \quad (1)$$

where initial seed weight—the weight of each replicate measured before the seed is placed on moisture filter paper; initial seed mc—seed moisture content, determined by low-content-temperature method (seed was dried at 103 °C at 17 h).

Seed weight was consistently measured with high precision, and the obtained values were inputted into this equation. Under these conditions, approximately 1.5 h sufficed to attain the targeted moisture content of 20%. In this test, elevating and standardizing the seed moisture content to 20% serves the purpose of mitigating disparities in the water absorption rates among seeds of the same species. When the seeds reach the desired weight, the hydrated seeds are enveloped with aluminum foil, hermetically sealed, and then introduced into a refrigeration unit set at 7 °C for 24 h. Following this refrigeration phase, the seeds undergo transfer to a water bath maintained at 45 °C for an additional 24 h.

The experiments were conducted in four replicates, each comprising 100 seeds. Following the completion of all assessments of seed vigor, the same parameters were measured as those conducted in the standard laboratory test. The evaluation of seed germination was conducted using a standard laboratory assay one year after the initial seed testing.

Seedlings vigor index (SVI) was calculated according to [16]

$$SVI = (SL + RL) \times GP, \quad (2)$$

where SL—seedlings length, RL—root length and GP—germination percentage.

### *2.3. Topographical Tetrazolium Test (TTZ)*

Due to the absence of a specific protocol for camelina, the TTC test was conducted following the protocol of the International Seed Testing Association [14] designed for *Brassica napus* with slight modification. Three sets of 50 seeds each were enumerated and immersed in water for 18 h at a temperature of 20 °C. Subsequently, the seeds were rinsed to remove mucilage using a continuous water stream for 10 min, with any remaining mucilage further eliminated using a paper towel. Following careful seed extraction using a sharp implement, the seeds were immersed in a 1% 2,3,5-triphenyltetrazolium chloride solution for 5 h at 30 °C. Assessment of seed viability was based on the identification and measurement of unstained areas on the essential parts of the seed, occasionally considering the intensity of staining. While determining vitality was not the primary goal of this test, the obtained results were recalculated and presented as a percentage.

The data were analyzed using software Statistica 12.0. The analysis of variance was performed, and means were compared by Duncan's multiple range test, at 5% probability. Statistically significant differences between the control and treatments were determined at a significance level of $p < 0.05$ using the multi-range Duncan's test.

## 3. Results and Discussion

The seeds of NS Slatka and NS Zlatka, when subjected to the standard laboratory germination method, exhibited germination rates of 91% and 89%, respectively. However, the results from most of the vigor tests yielded significantly lower values (Table 1). Using the accelerated ageing test, the seed germination was 89% and 87% (NS Slatka, NS Zlatka, AA1) and 85% for both cultivars in the AA2 test. The difference between these two tests was statistically significant. Although the initial germination values varied between the two genotypes, the application of AA2 resulted in an 8% reduction in the germination in both cases. The lowest values were observed in the cold test, with a germination percentage of 84% for NS Slatka and 82% for NS Zlatka. Exposing the seeds to mechanical obstacles in the Hiltner test resulted in a 5% reduction in germination for NS Slatka, whereas NS Zlatka remained unaffected. Following the CD test, the observed percentage values for NS Slatka and NS Zlatka were determined to be 85% and 84%, respectively. Notably, a consistent trend akin to that observed for AA2 was identified.

**Table 1.** Analysis of variance for germination and growth parameters of two camelina genotypes across various vigor tests.

| Test | Genotypes | Germination (%) | Atypical Seedlings (%) | Shoot Length (mm) | Root Length (mm) | SVI |
|---|---|---|---|---|---|---|
| Standard germination test | NS Slatka | 91 $^{a}$ ± 1.26 | 2 $^{d}$ ± 0.00 | 34.75 $^{ab}$ ± 3.69 | 42.13 $^{a}$ ± 1.93 | 69.78 $^{a}$ |
| | NS Zlatka | 89 $^{b}$ ± 1.71 | 2 $^{d}$ ± 1.29 | 35.125 $^{ab}$ ± 1.03 | 40.00 $^{abc}$ ± 2.68 | 66.71 $^{ab}$ |
| Accelerate ageing test 1 | NS Slatka | 89 $^{b}$ ± 1.41 | 3 $^{cd}$ ± 1.29 | 35.75 $^{ab}$ ± 1.50 | 40.50 $^{ab}$ ± 3.06 | 67.83 $^{ab}$ |
| | NS Zlatka | 87 $^{c}$ ± 2.37 | 2 $^{d}$ ± 0.50 | 35.5 $^{ab}$ ± 1.47 | 40.88 $^{ab}$ ± 1.65 | 66.65 $^{ab}$ |
| Accelerate ageing test 2 | NS Slatka | 85 $^{cde}$ ± 1.41 | 7 $^{a}$ ± 1.29 | 33.38 $^{ab}$ ± 1.80 | 38.00 $^{bc}$ ± 3.34 | 60.66 $^{d}$ |
| | NS Zlatka | 85 $^{cde}$ ± 2.88 | 6 $^{ab}$ ± 0.50 | 34.25 $^{ab}$ ± 3.07 | 36.25 $^{c}$ ± 2.47 | 60.05 $^{d}$ |

**Table 1.** *Cont.*

| Test | Genotypes | Germination (%) | Atypical Seedlings (%) | Shoot Length (mm) | Root Length (mm) | SVI |
|---|---|---|---|---|---|---|
| Hiltner test | NS Slatka | 86 $^{cde}$ ± 2.08 | 7 $^{a}$ ± 1.89 | 32.25 $^{ab}$ ± 2.90 | 39.75 $^{abc}$ ± 3.52 | 61.50 $^{cd}$ |
| | NS Zlatka | 88 $^{bc}$ ± 2.08 | 7 $^{a}$ ± 1.89 | 31.38 $^{b}$ ± 1.43 | 38.75 $^{abc}$ ± 2.78 | 61.35 $^{cd}$ |
| Cold test | NS Slatka | 84 $^{ef}$ ± 2.22 | 8 $^{a}$ ± 2.06 | 33.38 $^{ab}$ ± 3.04 | 37.13 $^{bc}$ ± 3.07 | 59.41 $^{d}$ |
| | NS Zlatka | 82 $^{f}$ ± 1.83 | 8 $^{a}$ ± 2.16 | 33.13 $^{ab}$ ± 1.65 | 37.50 $^{bc}$ ± 1.35 | 57.90 $^{d}$ |
| Controlled deterioration test | NS Slatka | 85 $^{def}$ ± 1.26 | 7 $^{a}$ ± 1.25 | 32.25 $^{ab}$ ± 0.29 | 37.00 $^{bc}$ ± 0.41 | 58.44 $^{d}$ |
| | NS Zlatka | 84 $^{ef}$ ± 1.29 | 6 $^{ab}$ ± 0.96 | 34.00 $^{ab}$ ± 0.41 | 36.00 $^{c}$ ± 0.01 | 58.69 $^{d}$ |
| Standard germination test after 1 year | NS Slatka | 87 $^{cde}$ ± 1.73 | 3 $^{cd}$ ± 0.96 | 34.25 $^{ab}$ ± 3.30 | 40.63 $^{ab}$ ± 2.87 | 64.75 $^{bc}$ |
| | NS Zlatka | 86 $^{cde}$ ± 1.71 | 4 $^{bc}$ ± 2.16 | 33.75 $^{ab}$ ± 2.72 | 38.25 $^{abc}$ ± 0.65 | 61.72 $^{cd}$ |

The differences were statistically significant determined by Duncan's multiple range test ($p < 0.05$). The same letters indicate no significant differences between means.

The physiological potential of seeds is contingent upon two critical factors: germination viability and vigor. These factors ascertain the inherent capacity of a seed to manifest its vital biological functions within a spectrum of environmental conditions, encompassing both favorable and adverse circumstances [8]. Seed lots that demonstrate superior vigor possess the capability to endure adverse environmental stressors, consequently experiencing a more gradual rate of deterioration in comparison to those with lower vigor [17]. The assessment of the seed vigor cannot solely rely on the outcomes of a single test; instead, it necessitates the implementation of diverse methods and the incorporation of multiple tests for a comprehensive evaluation [8].

As evidenced in this investigation, a decrease in germination rates following the AA test was observed across numerous plant species: wheat [18], rise [19], oilseed rape [20], soybean [21], and safflower [22]. Elevated temperatures and high relative humidity levels to which seeds are subjected during the AA test and CD test induce heightened seed moisture content and increase the activity of hydrolytic enzymes. Simultaneously, there is an escalation of metabolic processes coupled with the ineffectiveness of enzymes responsible for neutralizing the impact of free radicals [23]. This phenomenon correlates with an elevation in lipid peroxidation levels, serving as an indicative marker of stressful environmental conditions [24,25]. Additionally, those factors can cause the carbonylation of proteins, which significantly escalates during the progression of seed decay and can eventually culminate in the death of the seed [26]. Numerous studies have demonstrated that accelerated ageing is correlated with intensified reactive oxygen species (ROS) production, resulting in diminished antioxidant activity in response to their increased accumulation [20,27,28].

This study further confirms the substantial impact of elevated temperature and relative humidity, as evidenced by a notable reduction in the germination percentage observed in both genotypes following exposure to AA2 and CD. Under these adverse conditions, the seeds experience a breakdown of hypodermic cells, manifesting anatomical changes in the hypodermic layer of the testa, which are closely linked to a decline in the seed's ability to germinate [29].

While one of the principal utilities of the AA test lies in its capacity to predict the performance of seed lots under unfavorable environmental conditions and appraise their storage potential, the results obtained in this study do not substantiate this premise completely. Notably, the correlation between the parameters examined in the standard germination after one year of storage and the AA2 test showed significance in both genotypes (r = 0.63 for NS Slatka, r = 0.76 for NS Zlatka), while, in the case of AA1, statistical significance was only observed in NS Slatka (r = 0.70). The observed correlation implies that the AA assay may possess the capacity to fulfil its fundamental objectives in the context of camelina; however, it is imperative to conduct additional optimizations of the methodology to ensure robust and accurate outcomes. Additionally, raising the temperature from 41 °C to 43 °C

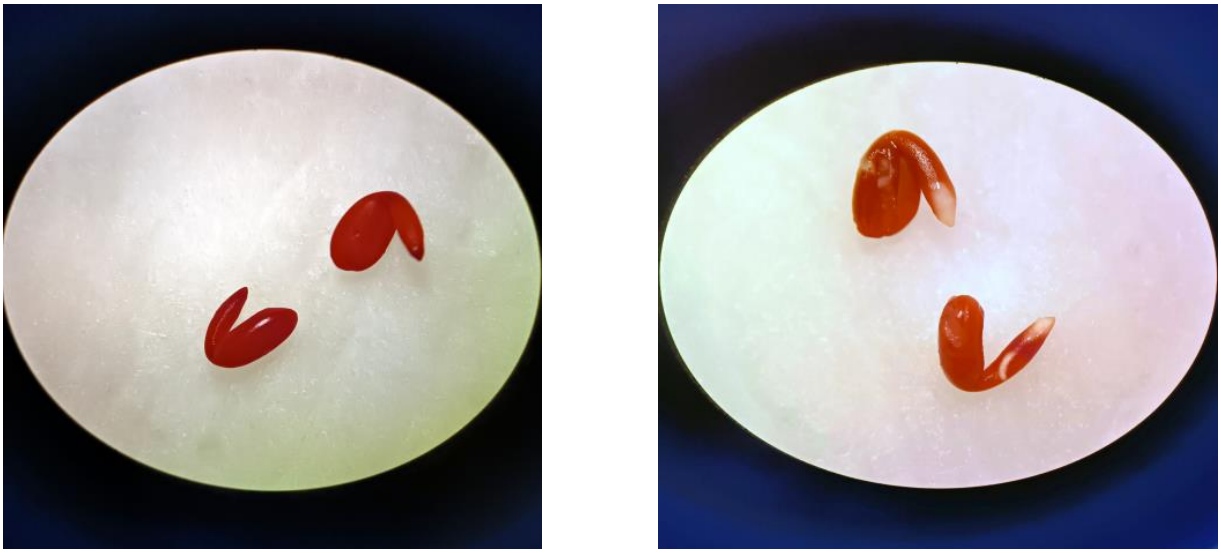
resulted in a 6% reduction in germination for NS Slatka and a 4% decrease for NS Zlatka. This indicates a moderate level of temperature sensitivity.

The controlled deterioration test is a valuable alternative for small seeds, as it enables the avoidance of the effects caused by marked differences in the rate of seed water absorption [30]. The seed moisture content is intentionally reduced to previously determined values before the stress exposure period. This characteristic is what distinguishes the CD test and positions it as an enhanced version of the AA test. In this study, the CD results showed the same trend as AA2 in both genotypes. The study by Awan et al. [31] demonstrated a direct correlation between the seed germination performance, specifically the speed of germination and tolerance to controlled deterioration, and both the mother plant environment and genotypes.

The cold test yielded the lowest values, as the seeds were exposed to low temperatures during the germination process. These results should be interpreted cautiously and evaluated against the validity of this test, due to the fact that both tested camelina genotypes belong to the spring biotype and the temperature and sowing time can vary significantly. Nevertheless, as per Baalbaki et al. [13], the implementation of a cold test enables the evaluation of seed physiological damage resulting from extended and improper storage, as well as damage caused by frost and drought conditions.

In both genotypes, there was a statistically significant increase in atypical seedlings after the AA2, CD, cold, and Hiltner tests, as compared to the standard test. It is possible that unfavorable conditions during vigor testing had a detrimental impact on the proper development of vital seed structures. In general, seeds with low physiological potential exhibit traits such as slow germination, increased susceptibility to stress during germination, and the resultant growth of plants characterized by slow, limited, and irregular growth, as well as reduced root development. These characteristics are indicative of seeds that have been adversely affected by diverse biotic or abiotic factors during their development. The consequences of such diminished seed vigor extend to reduced crop yields, particularly when seeds are produced in challenging environmental conditions [32,33].

The TTZ test is a biochemical assay that utilizes a solution of 2,3,5-triphenyltetrazolium chloride or bromide as an indicator to differentiate between viable and non-viable seeds. Seed hydration enhances the activity of the dehydrogenase enzyme, leading to the release of hydrogen ions and the formation of the stable chemical compound triphenyl formazan from colorless tetrazolium salts. This compound imparts a red color to living cells (actively involved in respiration), while the non-living cells (not actively respiring) remain uncolored. Consequently, the living seeds can be distinguished from the non-living ones [14]. In this study, the tetrazolium test was not directly used to assess the vigor and vitality of the seeds, but rather, it functions as an auxiliary tool to identify specific regions within the seed that are more susceptible to deterioration. In the images (Figure 1), non-specific and weaker staining is evident in the radicle zones. These regions are recognized as integral seed structures critical for the regular growth and developmental progression of the seedling and subsequent plant. This observation implies an increased sensitivity in these particular anatomical segments contributing to the development of atypical seedlings or seed decay. Additionally, 89% of the genotype NS Slatka seeds were viable, while 90% of the NS Zlatka seeds demonstrated viability, categorizing both of them as having very high vigor according to the criteria outlined by Rodriges et al. [34] according to Franca-Neto and Krzyzanowski (2018) [35]. Specifically, they suggest that tetrazolium viability test values exceeding 85% classify lots as possessing very high vigor. Results within the range of 84% to 75% indicate high vigor, while values below 59% correspond to lots characterized by low or very low vigor. Given that this test provides rapid and relatively reliable data, it primarily relies on the physical and physiological state of each segment of the seed embryo. Additionally, the implementation of this test excludes the influence of the external environment that could affect the germination process, as is the case with standard laboratory tests for germination [36]. Therefore, the optimization of this method for camelina should be undertaken in the shortest possible period.



(**a**) (**b**)

**Figure 1.** Illustration of camelina seeds after tetrazolium test: viable (**a**), non-viable (**b**) non-specific and weaker staining in the radicle zones.

It is recommended to analyze seedling growth parameters within a single variety rather than between varieties. This is because different varieties may have inherently distinct seedling growth rates due to their genetic differences. In this study, the seedling shoot and root length exhibited no significant variance across the various vigor tests applied to both genotypes. This observation suggests that the analysis of this parameter may not serve as a dependable indicator of vigor assessment. Nevertheless, the SVI, despite diminishing the significance of growth parameters such as seedling length and germination percentage, exhibited noteworthy variations. In both genotypes, the SVI displayed statistically lower values in the vigor tests and after-1-year storage as compared to the initial assessment. The SVI, apart from determining the germination percentage, is widely acknowledged as a highly precise and commonly employed methodology for evaluating the viability and longevity of seed lots [37,38].

The different results obtained from various tests and genotypes support the hypothesis that variability in seed survival curves depends on distinct temperatures and durations of exposure. All this implies a need for the further refinement and optimization of the experimental protocol. Several insights that potentially could have an impact on the seed vigor of this crop, as well as the further refinement of vigor assessment methodologies, encompass the following: (i) Within this species, as in the seeds of other *Brassicaceae*, there exists non-uniformity in the development and maturation of the seeds of a single plant. These seeds exhibit variations in moisture content and are subject to varying degrees of decay [39–41]; (ii) throughout the imbibition process, the seed coat of camelina generates and releases mucilage, facilitating a high degree of water absorption [42]. This mechanism potentially enhances its tolerance to drought during the initial stages of growth [43,44]; (iii) the oil content in seeds can significantly influence the mechanisms, duration, and rate of seed deterioration.

## 4. Conclusions

The considerably lower values observed in the assessment of seed vigor suggest a potential compromise in camelina's seed vitality, even though no issues with germination are evident. The preliminary findings of this experiment, which incorporated methods designed for testing other oilseeds, underscore a significant distinction for camelina. This implies that the conventional application of these tests, as recommended for other species, may not be fully effective. It is crucial to emphasize that further optimization is necessary

to enhance the reliability and applicability of these tests for camelina. It is well established that even minor deviations in the protocol can result in significant variations in the performance of seeds and seedlings. Additional research and refinement, drawing on insights from biochemistry and biotechnology, are imperative to ensure the accurate evaluation of camelina seed vigor through these testing methodologies.

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## References

1. Brock, J.R.; Ritchey, M.M.; Olsen, M.K. Molecular and archaeological evidence on the geographical origin of domestication for *Camelina sativa*. *Am. J. Bot.* **2022**, *109*, 1177–1190. [CrossRef] [PubMed]
2. Marjanović Jeromela, A.; Cvejić, S.; Mladenov, V.; Kuzmanović, B.; Adamović, B.; Stojanović, D.; Vollmann, J. Technological quality traits phenotyping of Camelina across multienvironment trials. *Acta Agric. Scand. B Soil Plant Sci.* **2021**, *71*, 667–673. [CrossRef]
3. Brock, J.R.; Scott, T.; Lee, A.Y.; Mosyakin, L.S.; Olsen, M.K. Interactions between genetics and environment shape Camelina seed oil composition. *BMC Plant Biol.* **2020**, *20*, 423. [CrossRef] [PubMed]
4. Zanetti, F.; Alberghini, B.; Marjanović Jeromela, A.; Grahovac, N.; Rajković, D.; Kiprovski, B.; Monti, A. Camelina, an ancient oilseed crop actively contributing to the rural renaissance in Europe. A review. *Agron. Sustain. Dev.* **2021**, *41*, 2. [CrossRef]
5. Ubeyitogullari, A.; Ciftci, O.N. Fabrication of bioaerogels from camelina seed mucilage for food applications. *Food Hydrocoll.* **2020**, *102*, 105597. [CrossRef]
6. Moser, B.R. Camelina (*Camelina sativa* L.) oil as a biofuels feedstock: Golden opportunity or false hope? *Lipid Technol.* **2010**, *22*, 270–273. [CrossRef]
7. Berhow, M.A.; Polat, U.; Glinski, J.A.; Glensk, M.; Vaughn, S.F.; Isbell, T.; Ayala-Diaz, I.; Marek, L.; Gardner, C. Optimized analysis and quantification of glucosinolates from *Camelina sativa* seeds by reverse-phase liquid chromatography. *Ind. Crops Prod.* **2013**, *43*, 119–125. [CrossRef]
8. Marcos-Filho, J. Seed vigor testing: An overview of the past, present and future perspective. *Sci. Agric.* **2015**, *72*, 363–374. [CrossRef]
9. Cvejić, S.; Jocić, S.; Mitrović, B.; Bekavac, G.; Mirosavljević, M.; Marjanović Jeromela, A.; Zorić, M.; Radanović, A.; Kondić-Špika, A.; Miladinović, D. Inovative approaches in breeding of climate-resilient crops. In *Climate Change and Agriculture: Perspectives, Sustainability and Resilience*; Benkeblia, N., Ed.; John Wiley & Sons Ltd.: Hoboken, NJ, USA, 2022; Chapter 6; pp. 111–156.
10. Vaughan, M.M.; Block, A.; Christensen, S.A.; Hartwell Allen, L.; Schmelz, E.A. The effects of climate change associated abiotic stresses on maize phytochemical defenses. *Phytochem. Rev.* **2018**, *17*, 37–49. [CrossRef]
11. Oliveira, G.M.D.; Silva, F.F.S.D.; Araujo, M.D.N.; Costa, D.C.C.D.; Gomes, S.E.V.; Matias, J.R.; Angelotti, F.; Cruz, C.R.P.; Seal, C.E.; Dantas, B.F. Environmental stress, future climate, and germination of *Myracrodruon urundeuva* seeds. *J. Seed Sci.* **2019**, *41*, 32–43. [CrossRef]
12. Fos, M.; Alfonso, L.; Ferrer-Gallego, P.P.; Laguna, E. Effect of salinity, temperature and hypersaline conditions on the seed germination in *Limonium mansanetianum* an endemic and threatened Mediterranean species. *Plant Biosyst.* **2020**, *155*, 165–171. [CrossRef]
13. Baalbaki, R.; Elias, S.; Marcos-Filho, J.; McDonald, M.B. *Seed Vigor Testing Handbook*; Association of Official Seed Analyst: Ithaca, NY, USA, 2009.
14. ISTA. *International Rules for Seed Testing*, 2022nd ed.; International Seed Testing Association: Wallisellen, Switzerland, 2022; Chapter 5, p. 15.
15. Hampton, J.G.; TeKrony, D.M. (Eds.) *Handbook of Vigour Test Methods*; International Seed Testing Association: Zurich, Switzerland, 1995.
16. Abdul-Baki, B.A.A.; Anderson, J.D. Relationship between decarboxylation of glutamic acid and vigour in soybean seed. *Crop Sci.* **1973**, *13*, 222–226. [CrossRef]

17. Hay, F.R.; Valdez, R.; Lee, J.S.; Sta Cruz, P.C. Seed longevity phenotyping: Recommendations on research methodology. *J. Exp. Bot.* **2019**, *70*, 425–434. [CrossRef] [PubMed]
18. Qin, P.; Kong, Z.; Liao, X.; Liu, Y. Effects of Accelerated Aging on Physiological and Biochemical Characteristics of Waxy and Non-waxy Wheat Seeds. *J. Northeast Agric. Univ.* **2011**, *18*, 7–12. [CrossRef]
19. Bijanzadeh, E.; Naderi, R.; Nosrati, K.; Egan, T.P. Effects of accelerated ageing on germination and biochemistry of eight rice cultivars. *J. Plant Nutr.* **2017**, *40*, 156–164. [CrossRef]
20. Jovičić, D.; Popović, B.M.; Jeromela, A.M.; Nikolić, Z.; Ignjatov, M.; Milošević, D. The interaction between salinity stress and seed ageing during germination of *Brassica napus* seeds. *Seed Sci. Technol.* **2019**, *47*, 47–52. [CrossRef]
21. Ebone, L.A.; Caverzan, A.; Tagliari, A.; Chiomento, J.L.T.; Silveira, D.C.; Chavarria, G. Soybean Seed Vigor: Uniformity and Growth as Key Factors to Improve Yield. *Agronomy* **2020**, *10*, 545. [CrossRef]
22. Coelho, M.V.; Lima e Silva, I.M.H.; Silva, A.A.S.; Paz, R.B.O.; Rocha, D.I.; Machado, C.G.; Silva, G.Z. Accelerated aging test in the determination of safflower seeds vigor. *Biosci. J.* **2022**, *38*, e38003. [CrossRef]
23. Tian, X.; Song, S.; Lei, Y. Cell death and reactive oxygen species metabolism during accelerated ageing of soybean axes. *Russ. J. Plant Physiol.* **2008**, *55*, 33–40. [CrossRef]
24. McDonald, M.B. Seed deterioration: Physiology, repair and assessment. *Seed Sci. Technol.* **1999**, *27*, 177–237.
25. Jovičić, D.; Štajner, D.; Popović, B.M.; Marjanović-Jeromela, A.; Nikolić, Z.; Petrović, G.; Zdero-Pavlović, R. Salt-induced changes in the antioxidant system and viability of oilseed rape. *Zemdirbyste* **2017**, *104*, 249–258. [CrossRef]
26. Zhou, W.; Chen, F.; Luo, X.; Dai, Y.; Yang, Y.; Zheng, C.; Yang, W.; Shu, K. A matter of life and death: Molecular, physiological, and environmental regulation of seed longevity. *Plant Cell Environ.* **2020**, *43*, 293–302. [CrossRef] [PubMed]
27. Yao, Z.; Liu, L.; Gao, F.; Rampitsch, C.; Reinecke, D.M.; Ozga, J.A.; Ayele, T.B. Developmental and seed aging mediated regulation of antioxidative genes and differential expression of proteins during pre- and post-germinative phases in pea. *J. Plant Physiol.* **2012**, *169*, 1477–1488. [CrossRef] [PubMed]
28. Kurek, K.; Plitta-Michalak, B.; Ratajczak, E. Reactive oxygen species as potential drivers of the seed aging process. *Plants* **2019**, *8*, 174. [CrossRef] [PubMed]
29. Silva, M.A.D.; Vieira, R.D.; Santos, J.M. Influência do envelhecimento acelerado na anatomia da testa de sementes de soja, cv. Monsoy 8400. *Rev. Bras. Sementes* **2008**, *30*, 91–99. [CrossRef]
30. Datura, A.S.; Medeiros-Filho, S. Teste de deterioração controlada na determinação do vigor em sementes de algodão. *Brazilian J. Seeds* **2008**, *30*, 19–23. Available online: http://www.scielo.br/pdf/rbs/v30n1/a03 (accessed on 20 December 2023). [CrossRef]
31. Awan, S.; Footitt, S.; Finch-Savage, W.E. Interaction of maternal environment and allelic differences in seed vigour genes determines seed performance in *Brassica oleracea*. *Plant J.* **2018**, *94*, 1098–1108. [CrossRef]
32. Zakaria, M.S.; Ashraf, H.F.; Seragm, E.Y. Direct and residual effects of nitrogen fertilization, foliar application of potassium and plant growth retardant on Egyptian cotton growth, seed yield, seed viability and seedling vigour. *Acta Ecol. Sin.* **2009**, *29*, 116–123. [CrossRef]
33. Moyo, R.; Ndlovu, E.; Moyo, N.; Kudita, S.; Maphosa, M. Physiological parameters of seed vigour in ex-situ stored sorghum germplasm. *J. Cereals Oilseeds* **2015**, *6*, 31–38. [CrossRef]
34. Rodrigues, M.; Gomes-Junior, G.F.; Marcos-Filho, J. Vigor-S: System for Automated Analysis of Soybean Seed Vigor. *J. Seed Sci.* **2020**, *42*, e202042039. [CrossRef]
35. Franca-Neto, J.B.; Krzyzanowski, F.C. *Metodologia Do Teste de Tetrazólio em Sementes de Soja*; Embrapa CNPS: Londrina, Brazil, 2018; 109p. Available online: https://ainfo.cnptia.embrapa.br/digital/bitstream/item/193315/1/Doc-406-OL.pdf (accessed on 20 December 2023).
36. Franca-Neto, J.B.; Krzyzanowski, F.C. Tetrazolium: An important test for physiological seed quality evaluation. *J. Seed Sci.* **2019**, *41*, 359–366. [CrossRef]
37. Merritt, D.J.; Martyn, A.J.; Ainsley, P.; Young, R.E.; Seed, L.U.; Thorpe, M.; Hay, F.R.; Commander, L.E.; Shackelford, N.; Offord, C.A. A continental -scale study of seed lifespan in experimental storage examining seed, plant, and environmental traits associated with longevity. *Biodivers. Conserv.* **2014**, *23*, 1081–1104. [CrossRef]
38. Zhang, T.; Fan, S.; Xiang, Y.; Zhang, S.; Wang, J.; Sun, Q. Non-destructive analysis of germination percentage, germination energy and simple vigour index on wheat seeds during storage by Vis/NIR and SWIR hyperspectral imaging. *Spectrochim. Acta A Mol. Biomol. Spectrosc.* **2020**, *239*, 118488. [CrossRef] [PubMed]
39. Still, D.W.; Bradford, K.J. Using hydrotime and ABA-time models to quantify seed quality of Brassicas during development. *J. Am. Soc. Hortic. Sci.* **1998**, *123*, 692–699. [CrossRef]
40. Bedi, S.; Kaur, R.; Sital, J.S.; Kaur, J. Artificial ageing of Brassica seeds of different maturity levels. *Seed Sci. Res.* **2006**, *34*, 287–296. [CrossRef]
41. Bewley, J.D.; Bradford, K.J.; Hilhorst, H.W.M.; Nonogaki, H. *Seeds: Physiology of Development, Germination and Dormancy*, 3rd ed.; Springer: New York, NY, USA, 2013; pp. 341–376.
42. Harper, J.L.; Benton, R.A. The behavior of seeds in soil: II. The germination of seeds on the surface of a water supplying substrate. *J. Ecol.* **1996**, *54*, 151–166. [CrossRef]

43. Cui, J.; Jiang, W.; Sun, Q.; Sun, B. Drought resistance of *Camelina sativa* (L.) Crantz seeds in germination. *Chin. Sci. Bull.* **2006**, *10*, 203–205.
44. Čanak, P.; Jeromela, A.M.; Vujošević, B.; Kiprovski, B.; Mitrović, B.; Alberghini, B.; Facciolla, E.; Monti, A.; Zanetti, F. Is Drought Stress Tolerance Affected by Biotypes and Seed Size in the Emerging Oilseed Crop Camelina? *Agronomy* **2020**, *10*, 1856. [CrossRef]