



# Seed Vigour Tests to Predict Field Emergence of White-seeded Cowpea [*Vigna unguiculata* (L.) Walp] Seed Lots

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## ABSTRACT

**Background:** Seed vigour is considered one of the most important seed quality criteria for achieving rapid and uniform emergence and good stand establishment in plant production. The present study was conducted to determine which seed vigour tests best predict field emergence of cowpea seed lots.

**Methods:** Standard germination (SG), cool germination (CG, 18°C), single seed conductivity (SSC) and bulk seed conductivity (BSC) tests were conducted to estimate field emergence (FE) of eleven white-seeded cowpea lots with germination percentages  $\geq 85\%$ . Field emergence tests (FE-1 and FE-2) were conducted at two different seed sowing dates in early spring.

**Result:** The BSC of seeds from 11 seed lots varied from 17.3 to 41.6  $\mu\text{S cm}^{-1} \text{g}^{-1}$ , while SSC varied from 63.7 to 130.5  $\mu\text{S cm}^{-1} \text{g}^{-1}$ . Field emergence of 11 seed lots ranged from 53 to 79% for FE-1, from 61 to 93% for FE-2 and statistically significant differences were found between seed lots in both FE tests. Both SSC and BSC methods showed significantly higher negative correlations than standard germination and cool germination tests with FE-1 ( $r=-0.794$  and  $r=-0.891$ ,  $p<0.01$ ) and FE-2 ( $r=-0.899$  and  $r=-0.954$ ,  $p<0.01$ ), respectively. Therefore, both the SSC and BSC methods can be used to predict field emergence of white-seeded cowpeas.

**Key words:** Cool germination, Cowpea, Electrical conductivity, Field emergence, Seed vigour.

## INTRODUCTION

Cowpea, also known as black-eyed peas, is an important food legume and a valuable component of the traditional cropping systems (Rathore *et al.*, 2015) and is grown as a fodder crop, green manure and cover crop (Backiyarani and Nadarajan, 1996; Arora, 2004; Meena *et al.*, 2015). It is a valuable source of protein in human diets; its fresh or dried seeds and fresh pods are eaten in salads and soups (Nalawade *et al.*, 2021; Özel *et al.*, 2023). A total of 11.425 tons of fresh cowpea and 1.298 tons of grain cowpea (Anonymous, 2024) were produced in Türkiye. In this production, certified seeds and producers' own seeds are still used and cowpea farmers continue to obtain seeds from their cultivation or from the local market. The most common problem encountered by farmers who use their own cowpea seeds is low seed quality due to a lack of expertise throughout production, harvesting, conditioning and cowpea seeds are more sensitive to storage under ambient conditions, thereby increasing the loss of physiological quality before sowing (Smiderle *et al.*, 2017; Arun *et al.*, 2021). As a result, using those seeds may lead to poor germination and stand establishment, resulting in a significant reduction in crop (Raj *et al.*, 2020).

The most important factors affecting plant production are the viability and vigour of the seed lot, which have direct or indirect effects on field emergence, performance, plant vigor and yield and quality of most crops. Seed viability is assessed using the standard germination test, an acceptable seed quality test in quality control programs and it can be used to predict emergence under optimum field conditions (ISTA, 1995). However, the standard germination test cannot detect quality differences between

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seed lots in estimating field emergence under suboptimal field conditions. Therefore, there is a need for tests to determine the field emergence and storage potential of seed lots with acceptable germination (ISTA, 1995; Marcos-Filho, 2015). Seed vigour tests provide more detailed information to seed producers and users about the physiological seed quality of seed lots, as well as the identification of seed lots with a higher probability to perform well after sowing and/or during storage (Marcos-Filho, 2015). Today, several standardized seed vigour tests have been validated or recommended by ISTA and are still under research to better understand seed quality and detect vigour differences within seed lots; however, there is no obligation in seed quality control programs/systems.

Electrical conductivity (EC) is a validated vigour test for garden pea, chickpea, soybean, bean and radish (ISTA, 2022).

This test can also be applied to other non-endospermic seeds with a high number of active embryo tissues, as well as to small-seeded dicotyledonous species (Mavi *et al.*, 2014; Powell, 2022). EC testing is performed in two ways: the bulk conductivity method, which uses a specified number of seeds (ISTA, 1995) and the single-seed technique, which uses a single seed (Siddique and Goodwin, 1985; Hamman *et al.*, 2001). Several studies reported on the determination of seed vigour differences, changes in viability during storage and the prediction of field emergence in cowpea seed lots using the EC test. Pekşen *et al.* (2004) determined a significant negative correlation between the EC test and field emergence in white-seeded cowpea seed lots, but not in colored seed lots. Similarly, Sangwan *et al.* (2005) found a negative, significant correlation between seedling establishment and EC in cowpea genotypes. According to Batista *et al.* (2012), the EC test can be used to evaluate and rank cowpea seed lots based on seed vigor. EC was found to be effective at detecting reductions in cowpea seed viability due to storage (Araméndiz-Tatis *et al.*, 2022). Tyagi *et al.* (2024) stated that field emergence cannot be predicted with the standard germination test; however, the EC test can be used. EC tests have also been used to predict viability and as a good uniformity indicator of the seed quality between the seed lots, seedling emergence in the field or estimate storage potential in grain legumes such as bean (Siddique and Goodwin, 1985; Barros *et al.*, 1999; Kolasinska *et al.*, 2000; Ermis, 2022), soybean (Vieira *et al.*, 2004; Colete *et al.*, 2004; Khaliliaqdam *et al.*, 2012), chick pea (Khajeh-Hosseini and Rezzadeh, 2011), in deed some other species such as yellow mustard (Verma *et al.*, 2003), sweet corn (Dungjunchot and Chanprasert, 2008), radish (Mavi *et al.*, 2014) and tagetes (Gülöksüz *et al.*, 2025).

The cool germination test (CG) is a commonly used measure of seed vigor in cotton seed lots, determining which seed lots are appropriate for planting in cold soils or less than ideal conditions and recommended as a seed vigour test for cotton (ISTA, 1995; AOSA, 2009) and has also been reported by researchers as a good indicator of field emergence and vigour difference in among seed lots: cotton (Savoy, 2005), sweet corn (Dungjunchot and Chanprasert, 2008) and maize (Ilbi *et al.*, 2009; Noli *et al.*, 2010).

The present study was conducted to predict field emergence of 11 cowpea seed lots using standard germination, cool germination and single and bulk-seed conductivity tests.

## MATERIALS AND METHODS

This study was conducted in the Seed Science and Technology Lab and the Research and Application Farm of the Department of Horticulture, Faculty of Agriculture, Isparta University of Applied Sciences, Isparta, from February to July 2022.

### Seed lots

In the study, 11 white-seeded cowpea seed lots of “Karnikara” genotype obtained from different production areas of the Western Anatolian province of Türkiye (4 seed lots from Muğla, 3 seed lots from Uşak and 4 seed lots from Isparta) were used. The germination of all seed lots was  $\geq 85\%$ . After determining the moisture content of the seed lots, all lots were calibrated to 12% moisture to minimize seed moisture-related error in subsequent analyses (ISTA, 2022). Moisture-calibrated seed lots were placed in hermetic jars and stored in a refrigerator at  $+4^{\circ}\text{C}$  throughout the study.

### Standard germination test (SG)

Four replicates of 25 seeds were germinated between moistened paper towels for 8 days at  $25^{\circ}\text{C}$  (ISTA, 2022) and the number of normal seedlings was evaluated after 8 days (ISTA, 2018). To prevent etiolation in seedlings and ensure healthy seedling development during the germination test, an 8 h light/16 h dark photoperiod is used.

### Cool germination test (CG)

The cool germination test was conducted at  $18^{\circ}\text{C}$  for 12 days, as described in the SG test above, with four replicates of 25 seeds each and the number of normal seedlings was determined at the end of the test (Ilbi *et al.*, 2009).

### Single seed conductivity (SSC)

Four replicates of 10 individual seeds from each seed lot were weighed on an analytical balance with a precision of 0.0001 and each individual seed for each replicate is placed in a 100 ml volume beaker containing 80 ml of distilled water ( $\text{EC} < 5 \mu\text{S}/\text{cm}$ ) and the beakers were covered with stretch film and kept in the incubator at  $20^{\circ}\text{C}$  for 24 hours  $\pm$  15 minutes (Siddique and Goodwin, 1985). The electrical conductivity of each seed was measured using a conductivity meter (WTW inoLab Cond 720, Germany). The distilled water EC value was subtracted from each reading, the result divided by the individual seed weight and the SSC determined as  $\mu\text{S cm}^{-1} \text{g}^{-1}$  (ISTA, 2022).

### Bulk seed conductivity (BSC)

Four replicates of 50 seeds from each seed lot were weighed on an analytical balance with a precision of 0.0001 g and placed in conical 500 ml flasks (with a bottom diameter of  $80 \pm 5 \text{ mm}$ ) containing 250 ml of distilled water ( $\text{EC} < 5 \mu\text{S}/\text{cm}$ ), covered with stretch film and placed in the incubator at  $20^{\circ}\text{C}$  for 24 hours  $\pm$  15 minutes. The conductivity of each replicate was measured with a conductivity meter (WTW inoLab Cond 720, Germany). The EC value of distilled water was subtracted from each reading, the result was divided by the bulk seed weight and the BSC was determined as  $\mu\text{S cm}^{-1} \text{g}^{-1}$  (ISTA, 2022).

### Field emergence tests (FE)

Two field emergence tests (FE-1 and FE-2) were carried out in spring and four replicates of 25 seeds were sown by

hand at a soil depth of 4 cm, with a row spacing of  $15 \times 4$  cm, on the 3<sup>rd</sup> of May for FE-1 and on the 24<sup>th</sup> of May for FE-2, 2022, using a randomized plot design. The experimental plot was irrigated every few days during field emergence tests. Seedlings with cotyledons parallel to the soil surface were counted daily and seedling emergence was counted until 21 days after sowing in both FE tests. The soil was a sandy clay loam and the average air and soil temperatures were 14.4 and 17.0°C in FE-1 and 17.6 and 21.1°C in FE-2, respectively.

### Statistical analysis

All data obtained from the study were subjected to analysis of variance and Pearson's correlation analysis using the package for Social Sciences (IBM SPSS 21 package program) statistical program. The Duncan multiple comparison test was used to determine differences between the means. The relationships between field emergences (FE-1 and FE-2) and SG, CG, SSC and BSC were assessed using coefficients of determination ( $R^2$ ).

## RESULTS AND DISCUSSION

Changes in normal seedling percentages in the SG and CG tests, EC values for both SSC and BSC and field emergences for FE-1 and FE-2 were given in Table 1. Normal seedling percentages in the SG test ranged from 85% to 98% and all seed lots met the acceptable germination requirements of the Seed Certification Standards of Türkiye. However, the field emergence of 11 cowpea seed lots ranged from 53% to 79% in FE-1 and from 61% to 93% in FE-2 (Table 1). These results showed that variable stress factors at field conditions had a significant impact on field emergence. Pekşen *et al.* (2004) reported that poor field emergence in cowpea is related to adverse soil conditions. The SG test was correlated only with FE-2 ( $r=0.621^*$ , Table 2). The correlation between SG and FE-2 is thought to be due to the fact that the average air and soil temperatures during FE-2 were 3-4°C higher than

during FE-1. The SG test is performed at optimum germination temperatures and therefore provides reliable results in favorable field conditions; however, its effectiveness may decrease under adverse conditions (ISTA, 1995). Sangwan *et al.* (2005) determined a significant and positive correlation between the standard germination test and seedling emergence in cowpea. Similarly, Perissé *et al.* (2005) stated that the standard germination and EC tests showed high correlation with field emergence in white lupin. However, Tyagi *et al.* (2024) stated that there was no significant correlation between field emergence and the standard germination test in cowpea and therefore, it could not be used to predict field emergence. Similarly, Sridhar and Nagaraja (2004) reported no correlation between the standard germination test and field emergence in field-crop species such as maize, sorghum, cotton and pigeon pea.

The cool germination vigor test, performed slightly below the optimum germination temperature (18°C), can be used to determine differences in seed vigor among cotton seed lots (ISTA, 1995; AOSA, 2009). In our study, cowpea seed lots were classified into 6 different statistical groups in the CG test. Especially, the decreases in normal seedling percentage in the CG test were greater in seed lots 5, 7 and 9 and these lots showed lower field emergence for both FE-1 and FE-2 compared with other lots (Table 1). High and significant correlations were determined between the CG test and both FE tests,  $r=0.680^*$  with FE-1 and  $r=0.763^{**}$  with FE-2 (Table 2). Several researches showed that the CG test (18°C) can be used as effectively in prediction of the quality and field emergence of maize seed lots (Savoy, 2005) and sweet corn (Dungjunchot and Chanprasert, 2008) and also can be used as an alternative test to the cold test to evaluate vigour differences among maize seed lots (Ilbi *et al.*, 2009). In another study, the final count (144 hours) and the number of normal seedlings in the cool-germination test at 13°C showed significant positive correlations with field emergences (Noli *et al.*, 2010).

**Table 1:** Results in the standard germination test at 25°C (SG) and cool germination test carried out at 18°C (CG), single seed (SSC) and bulk seed conductivity (BSC) and field emergence tests at different sowing times (FE-1 and FE-2) of 11 cowpea seed lots.

| Lots | SG (%) | CG (%) | SSC ( $\mu\text{S}^{-1} \text{cm}^{-1} \text{g}$ ) | BSC ( $\mu\text{S}^{-1} \text{cm}^{-1} \text{g}$ ) | FE-1 (%) | FE-2 (%) |
|------|--------|--------|----------------------------------------------------|----------------------------------------------------|----------|----------|
| 1    | 97a    | 86abcd | 94.2b                                              | 24.1e                                              | 78a      | 92a      |
| 2    | 96a    | 89ab   | 66.1d                                              | 17.3g                                              | 79a      | 93a      |
| 3    | 95a    | 85bcd  | 92.3b                                              | 27.7d                                              | 71abc    | 81b      |
| 4    | 94a    | 93a    | 95.7b                                              | 27.6d                                              | 68bc     | 81b      |
| 5    | 85b    | 79de   | 120.9a                                             | 35.6b                                              | 67bc     | 71c      |
| 6    | 95a    | 91ab   | 94.0b                                              | 26.7d                                              | 67bc     | 79bc     |
| 7    | 92a    | 73ef   | 129.7a                                             | 33.6c                                              | 65c      | 70c      |
| 8    | 94a    | 89ab   | 82.2b                                              | 22.1f                                              | 74ab     | 88ab     |
| 9    | 93a    | 68f    | 130.5a                                             | 41.6a                                              | 53d      | 61d      |
| 10   | 98a    | 81cd   | 77.4c                                              | 21.0f                                              | 74ab     | 87ab     |
| 11   | 96a    | 88abc  | 63.7d                                              | 18.2g                                              | 73abc    | 88ab     |
| Mean | 94.0** | 83.8** | 95.1**                                             | 26.9**                                             | 69.9**   | 81.0**   |

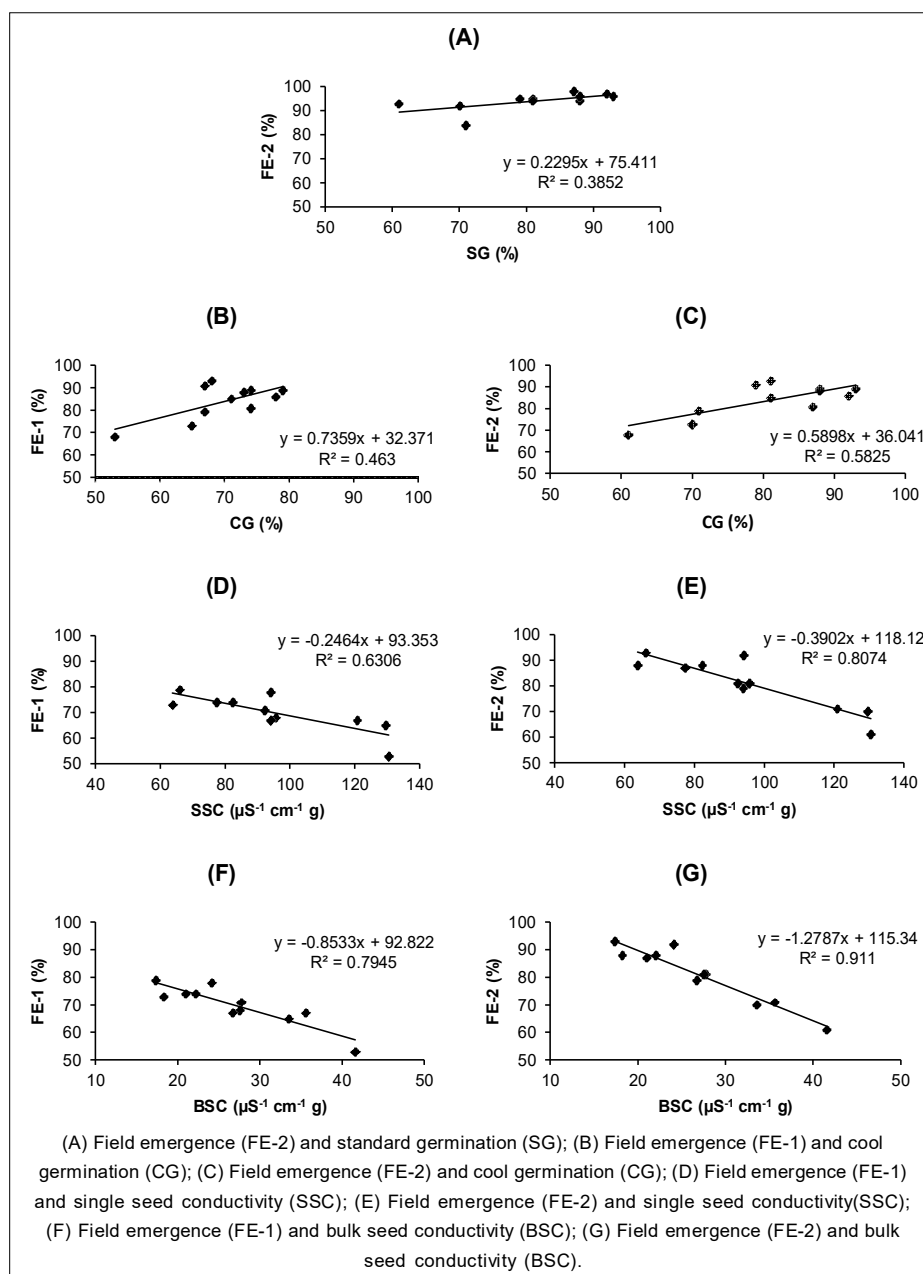
Significance levels: \*\* $p < 0.01$ .

**Table 2:** Correlation coefficients between the various quality tests and the field emergence tests carried out on 11 cowpea seed lots.

| Tests  | All seed lots            |                          |
|--------|--------------------------|--------------------------|
|        | Correlation of FE-1 with | Correlation of FE-2 with |
| SG (%) | 0.440                    | 0.621**                  |
| CG (%) | 0.680*                   | 0.763**                  |
| SSC    | -0.794**                 | -0.899**                 |
| BSC    | -0.891**                 | -0.954**                 |

Significance levels: \* $p < 0.05$ , \*\* $p < 0.01$ .

Powell (2022) reported that the conductivity test has the greatest potential for determining seed vigour in species with large living cotyledons. The changes in the seed lots' conductivity values were statistically significant in both EC tests. The seed lots were divided into four statistical groups in SSC and into seven groups in BSC. SSC of different seed lots ranged from 63.7 to 130.5  $\mu\text{S cm}^{-1} \text{g}^{-1}$  and BSC ranged from 17.3 to 41.6  $\mu\text{S cm}^{-1} \text{g}^{-1}$ . Seed lots 2 and 11 had the lowest conductivity values in both EC tests among seed lots and showed higher emergence in both field emergence tests. It was determined that seed

**Fig 1:** Regression analysis of various tests for 11 cowpea seed lots.

lots 5, 7 and 9, which had the highest conductivities in both SSC and BSC tests, gave the lowest emergence rates in both field emergence tests (Table 1). Seeds with high EC values often have poor physical quality due to damage to cell membranes and dead tissues resulting from ageing (Powell, 2022). Higher leakage levels in seed lots of chickpea (Khajeh-Hosseini *et al.*, 2011) and radish (Mavi *et al.*, 2014) indicate slower and lower emergence, as well as differences in seed vigour. Similarly, Khaliliaqdam *et al.* (2012) reported that low seed vigour resulted in significant delays and reductions in field emergence in soybean. Both SSC ( $r=-0.794^{**}$  with FE-1 and  $r=-0.899^{**}$  with FE-2) and BSC ( $r=-0.891^{**}$  with FE-1 and  $r=-0.954^{**}$  with FE-2) showed highly negative correlations with both field emergence tests than SG and CG tests (Table 2). Our results align with the prediction of field emergence by conductivity in cowpea seeds conducted by Pekşen *et al.* (2004), Sangwan *et al.* (2005) and Tyagi *et al.* (2024). Moreover, previous studies have shown that the electrical conductivity test can be used to predict seedling and field emergence, such as bean (Kolasinska *et al.*, 2000), yellow mustard (Verma *et al.*, 2003), soybean (Vieira *et al.*, 2004), purple vetch and alfalfa (Wang *et al.*, 2004), white lupin (Perissé *et al.*, 2005), chick pea (Khajeh-Hosseini *et al.*, 2011), soybean (Khaliliaqdam *et al.*, 2012) and tagetes (Gülöksüz *et al.*, 2025).

Based on both EC tests and FE tests results, seed lots 5, 7 and 9 were classified as low-vigor, seed lots 3, 4 and 6 as medium-vigor and seed lots 1, 2, 8, 10 and 11 as high-vigor. However, the BSC was found to be more effective than the SSC because it exhibits a higher correlation with field emergence tests and classifies seed lots into more statistically distinct groups (Table 1). Previous studies have indicated that single seed conductivity (Siddique and Goodwin, 1985) and bulk seed conductivity in beans (Barros *et al.*, 1999), in maize (Ribeiro *et al.*, 2009) and in cowpeas (Batista *et al.*, 2012), in purple vetch and alfalfa (Wang *et al.*, 2004), in soybean (Khaliliaqdam *et al.*, 2012) can be used to differentiate and rank seed lots according to their seed vigor levels. Other studies have shown that the EC test can predict bean germination (Ermis, 2022) and identify reductions in seed viability due to storage effects in cowpea (Araméndiz-Tatis *et al.*, 2022).

In this study, although the standard germination test was ineffective at explaining variation in FE-1, it accounted for 38.5% of the variation in FE-2 (Fig 1A). The CG test accounted for 46.3% of the variation in FE-1 and 58.3% of the variation in FE-2 (Fig 1B and 1C). However, it was determined that 63.1% of the variation in FE-1 and 80.7% of the variation in FE-2 could be predicted with SSC (Fig 1D and 1E) and 79.5% of the variation in FE-1 and 91.1% of the variation in FE-2 could be predicted with BSC (Fig 1F and 1G). These results showed that both conductivity test methods can be used more effectively to predict the field emergence of cowpea seed lots than standard germination and cool germination tests. Kolasinska *et al.*

(2000) stated that electrical conductivity testing can be used to predict field emergence of bean seed lots regardless of soil temperature. Hamman *et al.* (2001) reported that single-seed conductivity was not effective in predicting field emergence performance in soybeans, whereas Colete *et al.* (2004) stated that bulk seed conductivity could be used effectively to predict the vigour and field performance of soybean seeds.

## CONCLUSION

In this study, both electrical conductivity tests showed higher correlations with field emergence at different sowing dates than other laboratory tests, suggesting that they can be successfully used to predict field emergence of white-seeded cowpea seed lots. Furthermore, both conductivity tests are more reliable than standard germination and cool germination tests because they are cheaper, faster, more measurable and simpler to conduct.

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## Disclaimers

The views and conclusions expressed in this article are solely those of the authors and do not necessarily represent the views of their affiliated institutions. The authors are responsible for the accuracy and completeness of the information provided, but do not accept any liability for any direct or indirect losses resulting from the use of this content.

## Conflict of interest

The authors declare that there are no conflicts of interest regarding the publication of this article. No funding or sponsorship influenced the design of the study, data collection, analysis, decision to publish, or preparation of the manuscript.

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