

PREDICTION OF VIABILITY AND EMERGENCE CAPACITY OF SAFFLOWER SEED LOTS

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ABSTRACT

Predicting seedling emergence potential of safflower under field conditions is very difficult because of a wide variation of seed size, weight, shape and oil content. This study aimed to determine the most useful seed testing method predicting the field emergence of safflower seed lots. Thirty-four seed lots of 16 safflower genotypes were evaluated for field emergence, germination, seedling growth, accelerated ageing (AA) at 41, 43 and 45 °C for 48, 72, 96, 120 and 144 h, cool germination (18°C) and cold tests (10°C for 7 days), pH and electrical conductivity (EC) values of soak water for 4, 8 and 24 h. The seed lots showed a high variability for germination from 77.0% to 98.5% in laboratory conditions, and the field emergence ranged between 30.5% and 84.5%. The highest correlation with field emergence was detected in the second day germination in cool test ($r=0.586^{**}$), followed by the cold ($r=0.556^{**}$) and cool germination ($r=0.527^{**}$). No significant correlations were determined between the field emergence and seed weight, oil rate, seedling growth parameters, EC, pH and mean germination time. The AA test conducted at 41°C for 48 h significantly related to the field ($r=0.492^{**}$) and laboratory emergence ($r=0.607^{**}$); consequently, this combination is advised as the optimum ageing condition. The only one seed testing method does not reflect the emergence potential of safflower seed lots so, the tests should be combined by a linear multiple regression analysis. It was concluded that second day germination in cool test (SDC), laboratory emergence (LE) and cold test (CT) were found the promising methods in order to determine seed vigor of safflower. The best model was discovered as second day germination in cool test, laboratory emergence and cold test with the model of $y=-37.8+0.64*SDC+0.17*LE+0.32*CT$.

Key words: *Carthamus tinctorius* L., germination, emergence, accelerated ageing, cold test.

INTRODUCTION

Safflower is one of the most promising oilseed crops to be cultivated under rain-fed conditions in Turkey. It has been extensively grown as a spring crop in cold climatic conditions with the rotation after winter wheat or barley (Coşge and Kaya, 2008). Its cultivation has been supported by the government to increase oilseed production, especially in fallow lands and decrease the import of edible oil (Anonymous, 2017). By the reasons of its tolerance to drought and salinity, low labor and mechanization requirements, safflower has been potentially considered as a plant to replace the fallow that is obligatory due to insufficient and irregular precipitation refusing the survival of the other crop plants (Kaya *et al.*, 2015). It is especially preferred in fallow areas suffering from low soil fertility and a limited rainfall. Vigorous seed utilization to produce the healthy seedlings under these adverse conditions is crucial to provide the optimum seed density and plant population.

Seed vigor depends primarily on plant life cycle from pre-harvest, namely, maternal plant nutrition, seed reserve and maturity which can be largely controlled by soil and climatic conditions, secondarily mechanical damage during harvest and lastly post-harvest processes such as drying and storage conditions. Because seed

viability and emergence potential is ordinarily determined by standard germination test conditioning under optimum moisture and temperature, it fails to describe adequately the emergence performance under field conditions. So, a number of seed testing methods such as electrical conductivity, tetrazolium, accelerated ageing, cold and cool tests have been developed to assess the field emergence capacity more accurately than the standard germination (Milošević *et al.*, 2010). The previous researches showed that seed vigor in safflower has been successfully determined by the electrical conductivity (EC) test (Khavari *et al.*, 2009; Kaya, 2014) and accelerated ageing (Zamani *et al.*, 2010; Godakahriz *et al.*, 2012) in consequence of using limited seed lots. In the present study, the objective was to find a suitable, simple and effective seed test method predicting field emergence capacity, to combine two and more methods to estimate it better than the standard procedures, and to identify the most suitable condition of time and temperature for accelerated ageing test in safflower.

MATERIALS AND METHODS

The experiment was conducted at the Seed Science Laboratory in Department of Field Crops, Eskişehir Osmangazi University-Turkey in 2016. Thirty-

four seed lots of 11 registered safflower varieties and 5 inbred lines produced by seed companies, breeders and farmers were used after their seeds were reproduced at the experimental fields of the university during growing season in 2015 (March to August).

A four-replicated experiment carried out using 200 seeds from each seed lot to assess the seed viability between three layers of filter papers wetted with 21 mL of deionized water, treated with 1 g L⁻¹ Thiram fungicide. After the papers had been rolled, they were sealed in plastic bags to prevent moisture loss. The seeds were allowed to germinate for 10 days at 25±1°C in the dark incubator and were counted as germinated when the radicle elongated to 2 mm long. The germinated seeds were recorded every 24 h for 10 days to calculate the speed of germination (mean germination time, MGT) and the germination percentage (GP) was calculated according to ISTA (2003) rules. Seedling length, seedling fresh and dry weights were also measured at 10th day. The laboratory emergence test was conducted with 200 seeds from each seed lot in a seedling tray filled with peat. They were transferred into a growth chamber at a constant temperature of 25±1°C and 70% relative humidity, and the emerged seedlings (appearance of hypocotyls at the surface) were counted at 10th day. The field emergence test was performed at experimental areas of the university and determined at four replicates of 50 seeds which were planted on March 15th into seedbed spaced at 45 cm and 2-3 cm depth in a 3 m long plot. The soil of the experimental area was clay loam, alkaline (pH=8.1) and low in organic matter (1.1%). No irrigation was implemented after sowing because natural rainfall occurred. The seedlings were counted at 30th day and calculated as a percentage.

The cold germination test was applied to 4×50 (200 seeds per lot) seeds as mentioned in germination test. The test was performed at 10°C for seven days in a cold chamber. The rolled packages were transferred after 7 days to an incubator at 25°C (Vieira *et al.*, 2010). The germinated seeds were counted every day to calculate the mean germination time under cold stress.

The cool germination test was conducted between papers at 18°C (Hampton and TeKrony, 1995) and it was continued up to 10 days in the dark.

After four replications of 50 healthy seeds were weighted, they were immersed for 24 h in 200 mL of distilled water at 20±1°C in the dark conditions. The electrical conductivity (EC) and pH of soak water were read by an EC meter (WTW Cond 7310i) and pH meter (WTW pH 7110) after 4, 8 and 24 h incubation. The readings were converted to µS cm⁻¹ g⁻¹ to express per gram seeds.

The accelerated ageing (AA) test was performed with 200 seeds from each lot. The test was conducted with various combinations of temperatures (41, 43 and 45°C) and time (48, 72, 96, 120 and 144 h) at

approximately 100% air relative humidity in ageing boxes (11×11×4 cm) containing 40 mL of distilled water (Hampton and Tekrony, 1995). The seeds were uniformly distributed on a wire mesh tray with dimension 10×10×3 cm and covered plastic boxes. After ageing treatments, standard germination test was performed at 25±1°C in dark condition for 10 days.

The experimental data were analyzed using a completely randomized design with four replicates. After an arcsine transformation of percentages, ANOVA was performed using the statistic program of MSTAT-C (Michigan State University, v. 2.10). All correlations between the investigated parameters were calculated at 5% and 1% level of significance. A stepwise multiple regression analysis was conducted to determine if any combination of vigor tests could more precisely forecast the field emergence using SPSS.

RESULTS

A thousand seed weight and oil content of safflower seed lots showed a considerable variation. The oil content changed between 21.4 and 37.6%, and a thousand seed weight was determined from 33.3 to 52.6 g. The initial seed viabilities of the lots ranged from 77.0 to 98.5%, but field emergence was ranged from 30.5 to 84.5%. The higher EC readings were recorded at 24 h and extending duration led to increasing EC values (Table 1).

There were several significant correlations among the investigated parameters of seed quality in 34 seed lots while neither oil content nor a thousand seed weight was significantly associated with germination and emergence performance of safflower (Table 2). Also, the seedling growth parameters were not related to both germination and emergence. None of EC and pH measurements appeared to be correlated with seed performance, such as germination percentage, laboratory and field emergence percentages. MGT calculated in germination test, cool germination and cold tests failed to estimate germination and emergence performance of seed lots due to the absence of significant correlations. The highest significant correlation coefficient for standard germination was calculated in second day germination percentage of standard germination test ($r=0.936^{**}$), followed by the cold ($r=0.838^{**}$) and the cool ($r=0.792^{**}$) tests. Second day germination percentage in cool test showed a significant coefficient with laboratory emergence; however, it had the highest correlation coefficient with field emergence ($r=0.586^{**}$). The best relationship and higher significant positive correlation for germination and emergence was sequentially obtained in cold, cool and laboratory emergence. Regression analysis showed that a significant relationship was detected between field emergence and second day germination in cool test ($R^2=0.34^{**}$), cold test ($R^2=0.31^{**}$) and laboratory emergence ($R^2=0.19^{**}$) (Figure 1a, b and c).

Various time and temperature combinations were also tested for the accelerated ageing. The significant correlations with germination percentage, laboratory and field emergence percentage were detected, whereas the cooler temperature and the shorter exposure time exhibited the better correlations compared to the increased temperature and time (Table 3). Due to no germination at 45°C for 144 h, the results could not be illustrated. All of the correlations were positive except for a combination of 45°C for 120 h in field emergence. For germination percentage, the highest correlation coefficient was recorded at 41°C for 72 h ($r=0.674^{**}$), although the condition of 41°C for 48 h was the best for laboratory emergence ($r=0.607^{**}$) and field emergence ($r=0.492^{**}$). Extension of duration and warmer temperature in AA conditions led to reducing coefficients and significance level. The correlation coefficient for

field emergence was lower than that of germination and laboratory emergence percentage.

A multiple regression analysis was performed on 34 seed lots to search for better combination of vigor tests than a single test. The best single method for predicting field performance of safflower was second day germination percentage in cool germination test (SDC) (Table 4) and the highest significant determination coefficient was calculated as $R^2=0.34$ (Figure 1). Furthermore, a combination of SDC and laboratory emergence (LE) gave a better prediction than this test. When the LE was included to the model, the R^2 was enhanced from 0.34 to 0.39. However, the best model predicting the field emergence included SDC, LE and cold test (CT) with the formula $y = -37.8 + 0.64^*SDC + 0.17^*LE + 0.32^*CT$ ($R^2=0.40^{**}$).

Table 1. Minimum, maximum and mean values of the investigated parameters for 34 seed lots of safflower.

	Minimum	Maximum	Mean	Standard Error	Probability
Oil content (%)	21.4	37.6	29.1	0.76	<0.01
A thousand seed weight (g)	33.3	52.6	40.5	0.82	<0.01
Germination percentage (%)	77.0	98.5	90.9	0.89	<0.01
2 nd day germination (%)	77.0	97.5	89.5	0.89	<0.01
MGT (d)	1.02	1.77	1.22	0.03	<0.01
Seedling length (cm)	3.38	12.43	5.98	0.31	<0.01
Seedling fresh weight (mg plant ⁻¹)	127.9	268.2	189.3	5.34	<0.01
Seedling dry weight (mg plant ⁻¹)	16.2	31.5	20.9	0.61	<0.01
Laboratory emergence (%)	27.5	95.5	70.3	2.55	<0.01
Field emergence (%)	30.5	84.5	60.1	2.04	<0.01
Cool test (%)	80.0	99.5	92.9	0.85	<0.01
2 nd day germination (%)	64.5	99.0	89.0	1.33	<0.01
MGT (d)	1.29	2.29	1.91	0.03	<0.01
Cold test (%)	83.0	100.0	92.4	0.71	<0.01
2 nd day germination (%)	13.5	97.0	76.0	3.23	<0.01
MGT (d)	2.11	3.88	3.04	0.06	<0.01
EC 4h	8.5	28.5	14.8	0.77	<0.01
EC 8h	11.4	31.7	17.7	0.82	<0.01
EC 24h	16.6	37.9	21.8	0.91	<0.01
pH 4h	5.67	6.14	5.91	0.02	<0.01
pH 8h	5.67	6.02	5.87	0.02	<0.01
pH 24h	5.65	6.17	5.82	0.02	<0.01

Table 2. Simple correlation coefficients among standard germination, laboratory emergence, field emergence and seed vigor tests in 34 seed lots of safflower.

	Standard germination (%)	Laboratory emergence (%)	Field emergence (%)
Laboratory emergence (%)	0.546 ^{**}	-	0.435 ^{**}
Field emergence (%)	0.468 ^{**}	-	-
MGT (d)	-0.242 ^{ns}	-0.243 ^{ns}	-0.324 ^{ns}
Oil content (%)	-0.302 ^{ns}	-0.291 ^{ns}	-0.011 ^{ns}
1000 seeds weight (g)	-0.252 ^{ns}	0.108 ^{ns}	-0.141 ^{ns}
2 nd day germination (%)	0.936 ^{**}	0.541 ^{**}	0.497 ^{**}
Seedling length (cm)	0.144 ^{ns}	0.341 [*]	0.007 ^{ns}

Seedling fresh weight (mg plant ⁻¹)	-0.066 ^{ns}	0.250 ^{ns}	-0.018 ^{ns}
Seedling dry weight (mg plant ⁻¹)	-0.304 ^{ns}	0.012 ^{ns}	-0.149 ^{ns}
Cool test (%)	0.792 ^{**}	0.587 ^{**}	0.527 ^{**}
2 nd day germination (%)	0.637 ^{**}	0.380 [*]	0.586 ^{**}
MGT (d)	-0.391 ^{ns}	-0.268 ^{ns}	-0.360 ^{ns}
Cold test (%)	0.838 ^{**}	0.553 ^{**}	0.556 ^{**}
2 nd day germination (%)	0.492 ^{**}	0.367 [*]	0.495 ^{**}
MGT (d)	-0.330 ^{ns}	-0.268 ^{ns}	-0.317 ^{ns}
EC 4h	-0.189 ^{ns}	-0.218 ^{ns}	-0.162 ^{ns}
EC 8h	-0.176 ^{ns}	-0.155 ^{ns}	-0.145 ^{ns}
EC 24h	-0.216 ^{ns}	-0.107 ^{ns}	-0.169 ^{ns}
pH 4h	0.086 ^{ns}	-0.150 ^{ns}	-0.042 ^{ns}
pH 8h	0.112 ^{ns}	-0.180 ^{ns}	-0.107 ^{ns}
pH 24h	-0.064 ^{ns}	-0.192 ^{ns}	-0.165 ^{ns}

*, ** significant at %5 and %1 levels of probability, respectively; ns: not significant

Table 3. Simple correlation coefficients among germination percentage after AA test and germination, laboratory and field emergence.

Temp. (°C)	Hours (h)	Germination percentage (%)	Laboratory emergence (%)	Field emergence (%)
41	48	0.634 ^{**}	0.607 ^{**}	0.492 ^{**}
	72	0.674 ^{**}	0.488 ^{**}	0.332 [*]
	96	0.502 ^{**}	0.390 [*]	0.247 ^{ns}
	120	0.527 ^{**}	0.395 [*]	0.205 ^{ns}
	144	0.521 ^{**}	0.389 [*]	0.229 ^{ns}
43	48	0.538 ^{**}	0.507 ^{**}	0.332 [*]
	72	0.520 ^{**}	0.431 [*]	0.443 ^{**}
	96	0.525 ^{**}	0.426 [*]	0.335 [*]
	120	0.321 [*]	0.396 [*]	0.346 [*]
	144	0.438 [*]	0.421 [*]	0.252 ^{ns}
45	48	0.518 ^{**}	0.458 ^{**}	0.249 ^{ns}
	72	0.312 ^{ns}	0.140 ^{ns}	0.223 ^{ns}
	96	0.407 [*]	0.303 ^{ns}	0.201 ^{ns}
	120	0.232 ^{ns}	0.121 ^{ns}	-0.066 ^{ns}
	144	-	-	-

*, ** significant at %5 and %1 levels of probability, respectively. ns: not significant. Bracket (-) shows no seed germination and emergence.

Table 4. Summary of multiple regressions predicting the field emergence with vigour tests for 34 lots of safflower.

Step	Variable	R	R ²	Regression equation
1	2 nd day germination in cool test (SDC)	0.586	0.34	$y = -19.4 + 0.89 \cdot \text{SDC}$
2	Laboratory emergence (LE)	0.629	0.39	$y = -20.6 + 0.75 \cdot \text{SDC} + 0.20 \cdot \text{LE}$
3	Cold test (CT)	0.632	0.40	$y = -37.8 + 0.64 \cdot \text{SDC} + 0.17 \cdot \text{LE} + 0.32 \cdot \text{CT}$

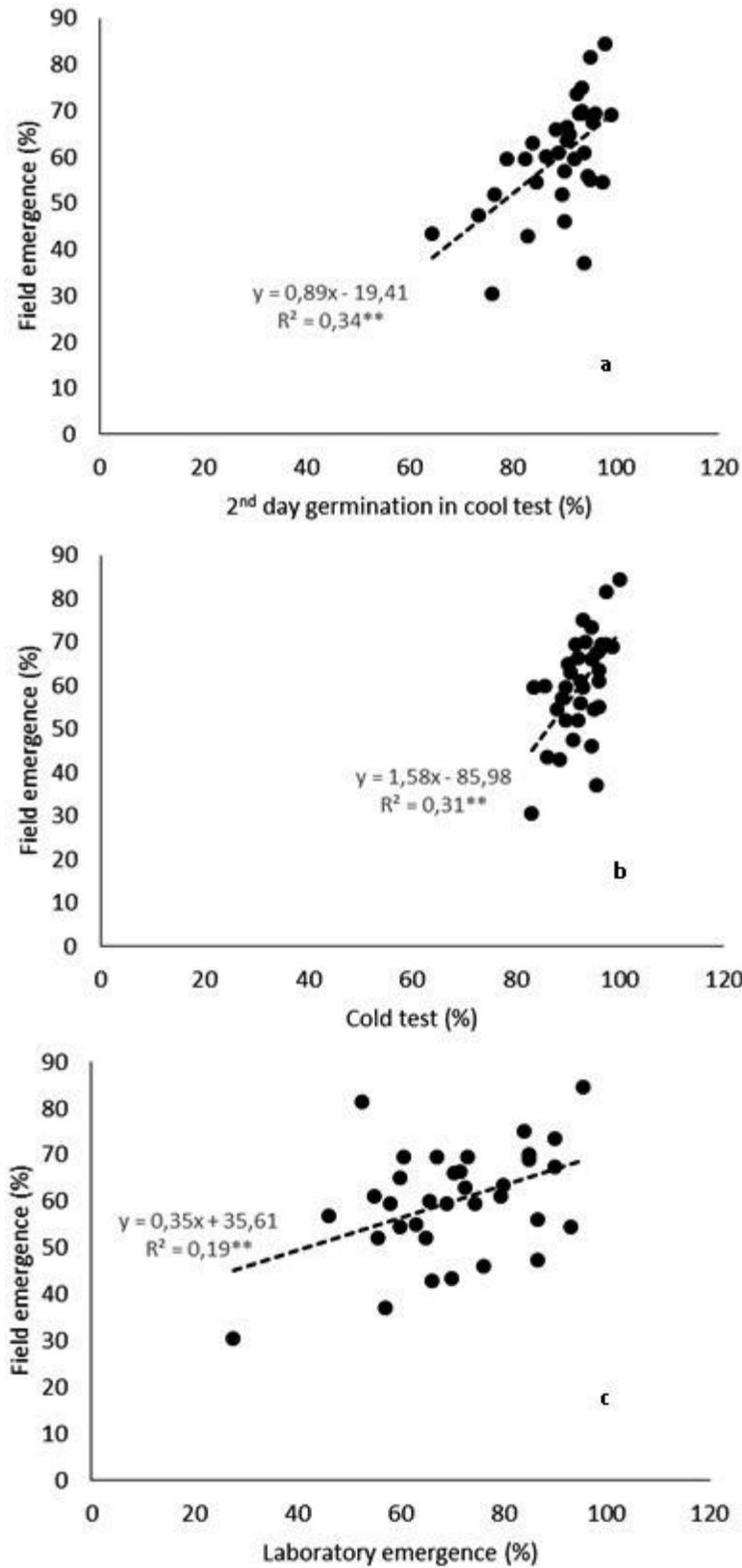


Figure 1. Regression analysis between field emergence and second day germination in cool germination (a), cold test (b) and laboratory emergence (c).

DISCUSSION

Although initial seed properties such as thousand seed weight and oil content showed a wide difference, no significant correlation was determined with germination and emergence performance of safflower seeds. In contrast to our findings, a considerable positive relationship between the seed weight and the field emergence were observed in cotton seed (Pahlavani *et al.*, 2008; Snider *et al.*, 2016). The seedling length, seedling fresh and dry weights showed insignificant correlations with the field emergence. The MGTs calculated from standard germination, cool and cold tests were also failed to predict the field emergence because of the lack of significant correlations. This finding is in contrast to those of Matthews and Khajeh-Hosseini (2006), who indicated that the MGT was a valuable evidence of seedling emergence in maize. Moreover, none of durations for EC and pH gave significant correlations. Khavari *et al.* (2009) and Kaya (2014) found a significant relationship between EC and germination percentage of safflower but they used small-scale seed lots to produce practical uses. Our results disagreed with those of other crops such as lentil (Makkawi *et al.*, 1999), soybean (Vieira *et al.*, 2004), chickpea (Khajeh-Hosseini and Rezazadeh, 2011), common bean (Silva *et al.*, 2013), sorghum (Kaya and Ileri, 2015), who found a considerable relationship between EC and field emergence. They reported that the EC readings were negatively linked to germination and seedling emergence, and the higher emergence percentage was obtained in seeds having lower EC. Unlike the other crop plants, the seed tests mentioned above were not found to be an indicative of the potential of field emergence of safflower.

The second day germination percentages from standard germination, cool and cold tests were evaluated for a valuable indicator of seed vigor because all of them were strongly correlated with the field emergence. Among them, the highest correlation coefficients with the field emergence and germination were observed in the second day germination in cool test, except for the laboratory emergence. Our results were similar to the findings of Demir *et al.* (2011); indicating that second day germination percentage was related to seedling emergence in field and germination after storage in *Viola* seeds. The field emergence was significantly correlated with the germination percentage of the cold and cool test and their coefficients displayed similarity to each other. Although both the cold and cool tests have been developed and performed for tropic or warm season crops (Milošević *et al.*, 2010), they could be used for ranking the seed lots of safflower. These results are confirmed by the findings of Lovato *et al.* (2005) in maize and Vieira *et al.* (2010) in soybean. They proved that the cold

germination showed a significant correlation with the field emergence.

AA is generally used for distinguishing vigor of seed lots and is usually found suitable for several crop plants. In this study, increasing temperature and time of ageing conditions resulted in decreasing both germination percentage and correlation coefficient with field emergence. Our findings revealed that the optimum combination of ageing conditions for safflower was determined at 41°C for 48 h because of the highest correlation with field emergence. In earlier researches, Kaya (2014) reported that the ageing condition at 45°C for 96 h was the most successful for safflower, while Godakahriz *et al.* (2012) found at 40°C for 6 days. Also, Khavari *et al.* (2009) stated that germination after AA test provided a significant relationship with field emergence of six safflower lots.

Despite the low and significant correlation coefficient, the second day germination, cold and cool tests were the most appropriate methods for estimating field emergence performance among the investigated tests. Furthermore, a multiple regression analysis provided a better prediction and higher the coefficient of multiple determination. The second day germination in cool test was the best single test, but the determination coefficient (R^2) was improved from 0.34 to 0.39 when the laboratory emergence was included to the model. The equation was fitted by adding the cold germination as the third component. In conclusion, two or more methods should be suggested to forecast the field emergence percentage instead of relying on a single test, so the second day germination in cool test, laboratory emergence and cold germination should be considered as the most proper indicators for seed vigor in safflower.

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