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Conformity of vigor tests to determine the seed quality of safflower (*Carthamus tinctorius* L.) cultivars

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Abstract

Safflower seeds vary in size, shape and oil content; therefore, a suitable method for testing seed vigor has not been properly developed. This study aimed to create a reliable procedure for combining temperature and time for accelerated aging to be potentially used to perform deterioration and electrical conductivity tests for predicting safflower seed vigor of safflower. Seeds from Balci, Dincer and Remzibey were evaluated in the laboratory for germination, emergence and accelerated aging (AA) at 41, 43 and 45 °C for 24, 48, 72 and 96 h. Controlled deterioration (CD) was assessed at a 20% moisture content at 45 °C for 48 h, and the electrical conductivity (EC) test was performed in artificially aged seeds. The initial germination rates of Balci, Dincer and Remzibey seeds were determined to be 95.0, 96.5 and 97.5%, respectively, and the emergence percentages were 81.8, 86.5 and 86.0%, respectively, under laboratory conditions. The results demonstrated that increased aging, temperature and time remarkably decreased the germination percentage. For the AA test, 45 °C for 96 h was the most effective temperature and time combination for distinguishing the seed vigor of safflower cultivars. We demonstrated that the conductivity test can be used to evaluate safflower seed vigor because it also negatively correlated with germination and the accelerated aging test, whereas the CD test was not efficient for evaluating seed vigor.

Keywords: *Carthamus tinctorius* L., accelerated aging, controlled deterioration, electrical conductivity, germination.
Abbreviations: AA_Accelerated aging, CD_Controlled deterioration, EC_Electrical conductivity, MGT_Mean germination time, LEP_Laboratory emergence percentage, FEP_Field emergence percentage.

Introduction

Safflower (*Carthamus tinctorius* L.) is an important oilseed crop that harbors a 30-45% oil content and that has been widely cultivated as a source of edible oil and dyes in the world. The cultivation of safflower is especially widespread in dry areas where rainfall is insufficient to promote the survival of the other oilseed crops, as it is tolerant to drought and salinity and requires low labor costs for mechanization (Weiss, 2000). It is a highly branched plant and exhibits a regular flowering order that begins on the main stem head and continues to the primary, secondary and tertiary heads (Baydar and Ulger, 1998). This flowering pattern, which can last 10-40 days, results in different sized seeds in the plant, which yield a variable seed quality (Weiss, 2000). A germination test demonstrates the viability of a seed lot under optimum conditions and can predict seed quality if the emergence conditions are favorable (Kolasinska et al., 2000; Rodo and Marcos-Filho, 2003). Seed vigor, an important component of seed quality, depends on genetic and environmental factors, such as maternal plant nutrition, seed maturity, reserve and seed moisture content (Ramamoorthy and Natarajan, 1997; Taylor, 2003). It is also influenced by mechanical damage, drying, seed age, storage conditions and pathogens (Perry, 1981). The seed vigor tests increase the ability to predict seed quality and stand establishment, although various vigor tests can provide conflicting results for several crop species (Powell et al., 1997; Jotai et al., 2001; Hampton et al., 2004). One of the most commonly used seed vigor tests is the accelerated aging (AA) test, which maintains seeds under a high temperature and high humidity for a specified time (Kulik and Yaklish, 1982). This method has been useful for classifying and predicting the field emergence performance of various crops, such as corn (Woltz and Tekrony, 2001), onions (Rodo and Marcos-Filho, 2003), soybeans (Torres et al., 2004), melons (Torres et al., 2009) and sesame (Thant et al., 2010). More recently, Godakahriz et al. (2012) reported that the accelerated aging of safflower seeds at 40 °C for 6 d was helpful for determining seed vigor. Controlled deterioration (CD) is another seed vigor test, which allows seeds to increase their initial moisture content followed by exposure to high temperature (45 °C) for 24 or 48 h. A positive relationship was observed between the CD result and the field emergence capacity by Rodo and Marcos Filho (2003) in onions, Kavak et al. (2007) in peppers, Modarresi and Van Damme (2003) in wheat, Hamidi et al. (2009) in rape and Wang et al. (2004) in forage crops. Braz et al. (2008) reported that the most efficient combination for the controlled deterioration of sunflower seeds was a 20% moisture content for 72 h or a 25% moisture content for 48 and 72 h at 42 °C. The electrical conductivity (EC) test can also effectively select highly vigorous seed lots under unfavorable stress conditions (Matthews and Powell, 1981). It has been successfully used as vigor test to evaluate the relationship between EC measurements and the field emergence of pea (Siddique et al., 2002), bean (Kolasinska et al., 2000), *Brassica* (Hampton et al., 2009) and wheat (Khan et al., 2010) seeds. Coimbra et al. (2009) suggested that the EC test most efficiently distinguished the vigor of sweet corn seeds lots. Although the seed lots of several crops are classified as high or low vigor by using different vigor tests, published detailed information on vigor tests used on safflower

Table 1. Seed characteristics, germination and emergence of the investigated safflower cultivars used in the vigor tests.

Cultivar	1,000 seeds weight	Seed moisture content	Hull percentage	Oil content	GP	LEP	FEP
	g	%					
Balci	38.6[b]	6.2	41.0[b]	38.0[a]	95.0	81.8	69.0[b]
Dincer	45.4[a]	6.3	47.5[a]	29.2[b]	96.5	86.5	84.0[a]
Remzibey	39.2[b]	5.7	47.0[a]	31.4[b]	97.5	86.0	74.0[b]
LSD $_{0.05}$	1.71	ns	1.17	2.78	ns	ns	6.92

ns: not significant. GP- germination percentage, LEP- laboratory emergence percentage, FEP- field emergence percentage. *: Means followed by the same letter (s) are not significantly different at $P \leq 0.05$.

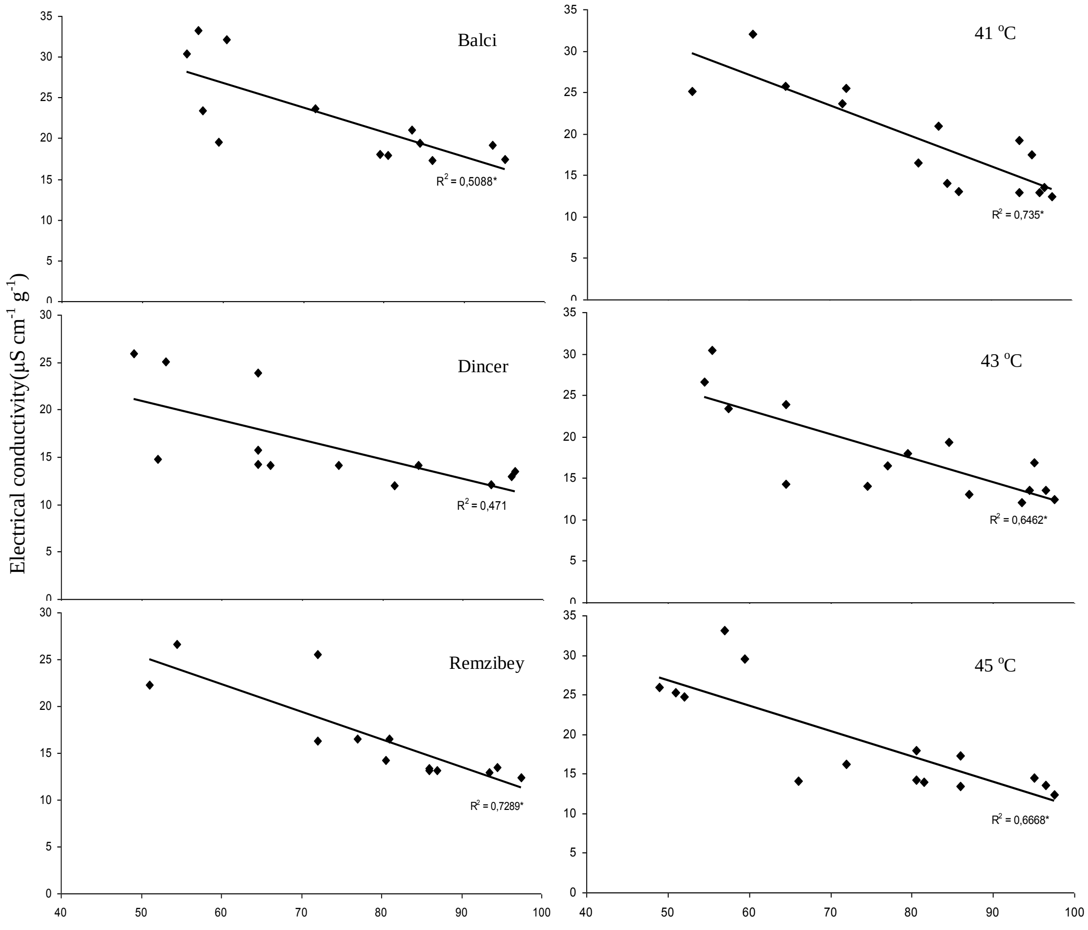

Fig. 1. Relationship between the electrical conductivity (EC) and germination percentage of safflower cultivars after accelerated aging. The determination coefficient (R^2) was based on the regression equations. *: Significant at $P \leq 0.05$.

seeds is still lacking. The objective of the present study was to determine the appropriate combination of temperature and time for accelerated aging and to identify the conformity of the AA, CD and EC tests in safflower.

Results

Seed characteristics

One thousand seed weight, hull percentage, oil content and field emergence percentage of the safflower cultivars were significantly different ($P \leq 0.05$; Table 1). The highest thousand seed weights and hull percentages were observed in Dincer, whereas the lowest were in Balci. A heavier seed weight significantly resulted in a higher hull content. The genotypes with a low hull percentage correlated with a higher oil content. Dincer seeds possessed high laboratory emergence percentage (LEP) and field emergence percentage (FEP) values of 86.5 and 84.0%, respectively.

Seed vigor tests

A significant three-way interaction (cultivar × aging temperature × time) was observed ($P \leq 0.05$) for the germination percentage, mean germination time and electrical conductivity. The germination percentage for controls and seeds aged at 41 °C for 24 h were similar but it declined with time after 48 h and greater. The highest germination percentage decrease was detected in Balci at 41 °C for 96 h and for aging at 45 °C for 96 h in Dincer and Remzibey. The highest germination percentage (72.5%) with an aging time

Table 2. Germination percentage (GP, %), mean germination time (MGT, d) and electrical conductivity (EC, µS cm^{-1} g^{-1}) values after accelerated aging (AA), controlled deterioration (CD) and standard germination (SG) of safflower cultivars.

Temp.	Time (h)	Balci			Dincer			Remzibey		
		GP	MGT	EC	GP	MGT	EC	GP	MGT	EC
41 °C	24	93.5^{abc}	1.88^{o-t}	19.2^{gh}	96.0^{a}	1.81^{g-u}	13.0^{mn}	93.5^{abc}	1.63^{uvw}	12.9^{mn*}
	48	83.5^{de}	1.93^{n-s}	21.0^{fg}	84.5^{de}	1.80^{g-u}	14.1^{k-n}	86.0^{cd}	1.76^{r-u}	13.1^{mn}
	72	71.5^{h-k}	2.10^{j-n}	23.7^{de}	64.5^{kl}	2.02^{l-p}	15.8^{ijk}	81.0^{def}	2.04^{l-p}	16.5^{ijk}
	96	60.5^{lm}	2.41^{gh}	32.1^{ab}	53.0^{mn}	2.39^{gh}	25.1^{cd}	72.0^{g-k}	2.49^{fg}	25.5^{cd}
43 °C	24	84.5^{de}	1.96^{n-r}	19.4^{gh}	93.5^{abc}	1.69^{tuv}	12.1^{n}	94.5^{ab}	1.73^{s-v}	13.5^{lmn}
	48	79.5^{d-g}	2.11^{j-n}	18.0^{hi}	74.5^{f-j}	1.74^{s-v}	14.1^{k-n}	87.0^{bcd}	1.73^{s-v}	13.1^{mn}
	72	57.5^{lm}	2.26^{hij}	23.4^{def}	64.5^{kl}	2.12^{j-n}	14.3^{k-n}	77.0^{e-h}	2.22^{h-l}	16.5^{ijk}
	96	55.5^{mn}	2.26^{hij}	30.4^{b}	64.5^{kl}	2.69^{f}	23.9^{de}	54.5^{mn}	2.25^{h-k}	26.6^{c}
45 °C	24	86.0^{cd}	1.95^{n-r}	17.3^{hij}	81.5^{def}	1.85^{p-t}	12.0^{n}	86.0^{cd}	1.97^{n-q}	13.4^{lmn}
	48	80.5^{def}	2.06^{k-o}	17.9^{hi}	66.0^{kl}	2.00^{m-q}	14.1^{k-n}	80.5^{def}	2.18^{i-m}	14.2^{k-n}
	72	59.5^{lm}	3.23^{bcd}	19.5^{gh}	52.0^{n}	3.14^{cde}	14.8^{j-m}	72.0^{g-k}	2.33^{gh}	16.3^{ijk}
	96	57.0^{lm}	3.03^{de}	33.2^{a}	49.0^{n}	3.38^{ab}	25.9^{cd}	51.0^{n}	3.24^{bc}	22.3^{ef}
CD		70.0^{jk}	1.78^{m-u}	16.9^{ijk}	68.0^{kl}	1.33^{y}	13.8^{lmn}	72.0^{g-k}	1.31^{y}	14.5^{j-m}
SG		95.0^{a}	1.87^{o-t}	17.5^{hi}	96.5^{a}	1.46^{w}	13.6^{lmn}	97.5^{a}	1.54^{vw}	12.4^{mn}
$LSD_{int.}$ ($P \leq 0.05$)		7.95	0.20	2.56	7.95	0.20	2.56	7.95	0.20	2.56

*: Means followed by the same letter(s) are not significantly different at $P \leq 0.05$.

of 96 h was detected in Balci (75.5%) at 43 °C. An increased temperature considerably decreased the cultivar germination. A lower germination was obtained at higher temperatures and a longer aging time. All aging treatments demonstrated a delayed in MGT, where the maximum was at 45 °C for 96 h. An increased aging temperature and time delayed the MGT; however, treatment at 45 °C for 96 h extended the MGT more than the other treatments (Table 2). The MGT was significantly longer at all temperatures for 96 h than during the other treatments. The longest time to germination was obtained at 45 °C. When the seed germination decreased, the EC values increased accordingly. The highest EC values were recorded at all temperatures for 96 h. The cultivar germination percentages after the CD test were similar, and significant differences were not observed. The germination percentages of Remzibey, Balci and Dincer were 72, 70 and 68%, respectively. The smallest MGT was in Remzibey at 1.31 d, followed by Dincer (1.33 d) and Balci (1.78 d). The EC values after CD were 16.9, 13.8 and 14.5 µS cm^{-1} g^{-1} for Balci, Dincer and Remzibey, respectively. A significant relationship between the EC values and germination percentages after accelerated aging was observed (Fig. 1). Reduced germination percentages correlated with increased EC values. The highest and most significant EC value (R^2=0.7289; $P \leq 0.05$) was obtained from Remzibey; however, a strong relationship (R^2= 0.735, $P \leq 0.05$) was detected at 41 °C for the cultivars.

Discussion

There was no difference in the initial seed viability of safflower cultivars, of which the seed characteristics and emergence percentages were different. A lower seed weight and hull percentage resulted in a lower germination and emergence percentage in Balci. Significant differences in the seed vigor were observed. An increased time and temperature in the AA test reduced germination but increased EC values and the MGT. The detrimental effect of AA was more prominent in Balci compared to the other cultivars. These results agreed with those of Santipracha et al. (1997) in corn and Cisse and Ejeta (2003) in sorghum, as they confirmed that genetic differences may affect seed vigor. AA adversely influenced the MGT of safflower cultivars, but exhibited inhibitory effects at 72 or 96 h. An increased time and temperature attenuated the germination time. Additionally, aged seeds exhibited higher EC values compared to the unaged seeds of each cultivar, which is consistent with earlier observations with *Brassica* species presented by Hampton et al. (2009). Vieira et al. (1999) detected a significant correlation between germination, field emergence and EC in soybeans. Conservely, Torres et al. (2009) reported that the EC test was not efficient for evaluating the seed vigor of melon seeds. The results of this study are consistent with the observations of Atak et al. (2002), who observed that the EC value was a valuable indicator of seed vigor in pea seeds. A highly significant relationship between the EC values and germination percentages suggested that the EC value is an efficient indicator of safflower seed vigor. Coimbra et al. (2009) observed that the EC test was the most efficient method to determine sweet corn seed vigor but that it did not correlate with field emergence or storage capacity. The CD test could not clearly distinguish the vigor of different safflower cultivars. All of the cultivars presented similar germination percentages and EC values after the CD test. However, useful CD test results were obtained by Braz et al. (2008) in sunflowers and by Rodo and Marcos-Filho (2003) in onions.

Materials and Methods

This study was performed at the Seed Science Laboratory in the Field Crops Department, Faculty of Agriculture, Eskisehir Osmangazi University, Turkey. Safflower cultivar seeds from Balci, Dincer and Remzibey were obtained from the Transitional Zone Agricultural Research Institute in 2011. All of the seeds were stored at 4 °C to maintain same moisture content until the start of the experiment.

Germination and emergence tests

Four replicates of 50 seed samples from each cultivar to be assessed for seed vigor were germinated in three rolled filter papers with 10 mL of distilled water. Each rolled paper was placed into a sealed plastic bag to prevent moisture loss. The seeds were allowed to germinate at 25 ± 1 °C in the dark for 10 days. The seeds were considered germinated when the emerging radicle was at least 2 mm long. The germination percentage (GP) was recorded every 24 h for 10 days. The mean germination time (MGT) was calculated to assess the speed of germination (ISTA, 2003). Four replicates of 50 seeds from each cultivar were sown at 2 cm depths in sand in a seedling tray (30 cm × 20 cm × 7 cm) to determine the laboratory emergence percentage (LEP). The seedlings were grown in an incubator at 25 ± 1 °C for 10 days. The emerged seedlings (appearance of hypocotyls at the surface) were counted at 10 days after sowing. Four sets of 50 seeds for each cultivar were sown in 2-m-long plots at a depth of 2.5-3.5 cm at the experimental field at the Field Crop Department. The emerged seedlings with cotyledon leaves were counted 21 days after sowing and represented the field emergence percentage (FEP).

Seed vigor tests

Accelerated aging (AA) test: Two hundred seeds were sampled from the each genotype. The AA test was performed with an aging temperature and time combination of 41, 43 and 45 °C for 24, 48, 72 and 96 h in plastic boxes (11 × 11 × 4 cm) with 40 mL of distilled water (Hampton and Tekrony, 1995). The seeds were placed on a 10 × 10 × 3 cm wire mesh tray and placed in a box after they were uniformly distributed. Each box was used for only one temperature and time combination in each cultivar. After aging, fifty seeds per replicate were germinated using filter paper at 25 ± 1 °C in a dark growth chamber for 10 days. Electrical conductivity (EC) test: The electrolyte leakage was measured with four replicates of 50 weighed seeds for each aging combination. The seeds were immersed in 200 mL of deionized water at 20 ± 1 °C for 24 h (ISTA, 2003). The electrical conductivity of the soaked water was measured using a conductivity meter (Model WTW Cond 314i, Germany). The results were expressed in $\mu S\ cm^{-1}\ g^{-1}$ to account for the variability in the seed weight among the seed lots. Controlled deterioration (CD) test: The moisture content of each cultivar was first determined at 105 ± 2 °C for 3 h. The seeds were placed into plastic tubes, and the required amount of water was added to increase the seed moisture content up to 20% by using the following equation (Rutzke et al., 2008):

X mL water = (g seed) × (MC_f - MC_i / 1 + MC_i)

where MC_f = desired final moisture concentration and MC_i= initial moisture concentration.

The tubes were sealed with parafilm to prevent moisture evaporation and incubated at 5 °C for 48 h to allow the moisture to equilibrate among the seeds. The CD test was performed at 45 °C for 48 h.

Statistical analysis

The experiment design consisted of a three factor factorial arranged in a completely randomized design with four replicates and 50 seeds per replicate. The germination percentage data were subjected to an arcsine transformation before an analysis of variance was performed using the MSTAT-C program (Michigan State University). The differences among the means were compared using the LSD values ($P \leq 0.05$).

Conclusion

Standard laboratory germination and the AA, EC and CD tests were compared in their ability to assess safflower seed quality. We concluded that the controlled deterioration test should not be used to clarify seed vigor. However, the electrical conductivity test was a reliable indicator of safflower seed vigor. A temperature of 45 °C for 96 hours was the optimum combination for the accelerated aging test.

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