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## Evaluation of Seed Vigour in White Coat French Bean (*Phaseolus vulgaris* L.) Seed Lots Under Waterlogged or Field Capacity Conditions

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**ABSTRACT:** This study was conducted to determine whether electrical conductivity and early radicle emergence count in ten white coat French bean seed lots were related to seedling emergence in waterlogged or field capacity sowing medium. Electrical conductivity (EC) measurements were performed by soaking seeds at 20 °C over 2, 4, 6, 18 and 24 h. Early radicle emergence percentages (RE) were counted after 24, 28, 32, 36, 40, 44 and 48 hours of germination. Seedling emergence percentages were tested by sowing at waterlogged or field capacity medium. Seedling emergence percentages were lower in waterlogged than in field capacity sowing in all seed lots. The EC and RE values were correlated with seedling emergence. In both waterlogged and field capacity sowings, the highest correlation values were observed in the 18 h (r=0.966; p<0.001; r=0.965, p<0.001) and 24 h of electrical conductivity (r=0.947, r=0.952, p<0.001) and in the 48 h of the RE test (r=0.905, r=0.918, p<0.001) with seedling emergence. Results showed that the seed vigour potential, in terms of emergence potential, of white coated French bean seed lots can be determined either through 18 h and 24 h EC readings or through 48 hours of early radicle emergence count during germination. Changes in seed vigour with respect to waterlogged or field capacity sowings were discussed for French bean seeds.

Keywords: Early radicle emergence, electrical conductivity, irrigation, seed germination, seedling emergence

**Abbreviations:** RE: radicle emergence, EC: electrical conductivity, SG: total and normal germination, RH: relative humidity

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#### **INTRODUCTION**

Vigour is an essential component of seed quality, which can be observed in differences in the field or glasshouse emergence potential of seed lots. Irregular and non-uniform emergence is a common phenomenon in white French bean seeds. It has been reported that seed coat affected different parameters, for example, water uptake by the seed (De Souza and Marcos-Filho, 2001), seed dormancy (Baskin et al., 2000), seed quality due to colour of seed coat (Mavi, 2010) and germination (Nerson, 2002). Seed lots with white testa showed lower emergence percentages compared to lots having dark coloured testa in French bean (Powell et al., 1986). For this reason, vigour determination in white coloured seed lots is more important to obtain an estimate on emergence potential. There are various seed vigour tests used to predict emergence potential of seed lots (Gupta, 1993; Usha and Dadlani, 2015). Common tests for legumes is the electrical conductivity of seed soaking water (ISTA, 2017). The white seeded lots had higher levels of solute leakage compared to coloured ones (Powell et al., 1986). One reason is fast imbibition, which results in higher solute leakage (higher EC values). Rapid imbibition may easily occur when free water is available in the seed bed, which is observed when irrigation is done just after sowing (water logging/excessive watering). However, the reasonable amount of water in the seed bed (i.e. the field capacity) presumably results in slow imbibition, less leakage and higher emergence. More recently, radicle emergence test (RE) during germination was considered a suitable vigour test and found to be correlated to emergence percentages (Khajeh-Hosseini et al., 2010; Ermis et al., 2015; Ozden et al., 2017). The conductivity growth due to membrane collapse, mitochondrial changes, chromosomal irregularity and free radicals in seed lots (Mavi, 2010). The aim of the present study was to investigate whether differences in the vigour of white coloured French bean seed lots can be determined by electrical conductivity and early radicle emergence tests, in relation to emergence potential, when sown under waterlogged (excessive watering) and field capacity planting conditions.

### MATERIALS AND METHODS

Ten white testae seed lots of French bean (*Phaseolus vulgaris* L. cultivar Ayşe Kadın), produced by various seed companies (all produced in 2015 and 2016), were bought in Ankara in May 2016 from retail outlets. The germination of four replicates of 25 seeds from each seed lot was assessed using the between-paper method (ISTA, 2017) at 25 °C in the dark. Rolled paper towels were placed in plastic bags to prevent water loss, and 2 mm radicle emergence counts were made after 24, 28, 32, 36, 40, 44 and 48 hours. Total and normal germination (SG) was determined after 9 days (Final germination).

Three replicates of 50 seeds in each lot were weighed (to 0.0001 g), kept at 100% relative humidity in mesh trays overnight and soaked in 200 ml distilled water at 20 °C in the dark. The conductivity of seed soak water was measured after 2, 4, 6, 18 and 24 h using a conductivity meter (Schott-Gerate GmbH, Hofheim) and expressed per gram of seeds ( $\mu$ S cm<sup>-1</sup> g<sup>-1</sup>).

For the seedling emergence tests, four replicates of 25 seeds from each lot were sown 4 cm deep in plastic seed trays (32cmx16cmx8cm / length x width x deep) containing peat moss medium (Plantaflor, Germany) and perlite mixture (2:1). Half of the trays were waterlogged and the other half was kept in field capacity. Waterlogged sowing method was obtained by excessively watering the trays (about 200 ml water in each tray), water drips (saturated) were seen under the trays. Field capacity sowing was applied by arranging seed moisture of medium about 27-30% through adding required amount of water (field capacity) by weight. Modules were placed in climatic room at 20 °C and about 70±5% RH in order to minimize evaporation.

Illumination was provided with 72  $\mu$ Mol m<sup>-2</sup> s<sup>-1</sup> light from white fluorescent lamps. Trays were irrigated, regularly by the second day of sowing to 16 days. Emergence counts were made every day for 16 days, with the appearance of leaves on the surface used as the emergence criterion. At the end of the experiment; total, normal and abnormal seedling emergence percentages were determined.

Statistical analysis was performed using SPSS to perform ANOVA and regression analysis. Comparison of means was made at 5%.

Percentages were arcsine transformed prior to analysis.

### **RESULTS AND DISCUSSION**

All seed lots had a final total germination level above 92% and a standard laboratory germination (normal seedling) level above 50% (Table 1). Despite higher germination, there were large differences in radicle emergence percentages in germination until 48 hours, seedling emergence percentages and electrical conductivity values (Table 2).

**Table 1.** Changes in radicle emergence count (RE) (24, 28 32, 36, 40, 44 and 48 h) and final (9 days) germination percentages in ten French bean seed lots. Means in the same column with the different letters are significantly different at 5% level.

Sood lota	RE Count (%)							Final Germination (%)	
Seed lots	24 h	28 h	32 h	36 h	40 h	44 h	48 h	Total	Normal
1	7 b	20 b	43 a	52 b	64 b	78 b	87 a	99 a	93 a
2	3 c	7 d	19 d	32 d	52 c	62 e	80 b	97 a	91 b
3	3 c	12 c	29 c	47 c	73 a	83 a	86 a	99 a	85 c
4	1 d	8 d	16 e	27 e	47 d	65 d	81 b	98 a	80 d
5	13 a	23 a	39 b	57 a	65 b	69 c	76 c	95 b	77 e
6	0 e	1 f	4 i	14 g	30 g	51 hi	70 d	93 c	77 e
7	0 e	1 f	11 g	25 ef	40 e	53 g	70 d	98 a	72 f
8	0 e	5 e	13 f	26 ef	47 d	59 f	69 d	97 a	66 g
9	0 e	2 f	6 h	14 g	28 h	52gh	62 e	97 a	52 h
10	0 e	4 e	10 g	24 f	36 f	50 i	62 e	92 c	50 i

**Table 2.** Electrical conductivity (EC) measurements after 2, 4, 6, 18 and 24 hours of soaking and seedling emergence percentages when sown in waterlogged and field capacity conditions in ten French bean seed lots. Means in the same column with the different letters are significantly different at 5% level.

Seed	EC (μScm <sup>-1</sup> g <sup>-1</sup> ) / h				Seedling emergence (%)		
lots	2 h	4 h	6 h	18 h	24 h	Waterlogged	Field Capacity
1	4.2 c	8.4 b	12.4 b	20.6 a	24.5 a	79 Ba	99 Aa
2	3.1 b	5.8 a	9.9 a	22.1 b	26.9 b	71 Bc	95 Ab
3	9.2 f	14.0 e	17.2 e	22.4 b	29.6 c	75 Bb	99 Aa
4	3.1 b	5.8 a	9.9 a	23.1 c	30.9 d	69 Bc	89 Ac
5	10.1 g	14.7 f	17.8 e	26.9 d	31.1 d	61 Bd	87 Ac
6	2.4 a	5.7 a	9.6 a	28.3 e	34.8 e	59 Bd	84 Ad
7	5.4 d	10.0 c	13.7 c	28.8 e	35.5 f	55 Be	81 Ae
8	8.1 e	12.2 d	15.3 d	30.7 f	37.5 g	61 Bd	83 Ade
9	8.5 e	13.5 e	19.4 f	31.8 g	39.8 h	56 Be	81 Ae
10	10.9 h	17.8 g	22.2 g	39.6 h	49.6 i	37 Bf	67 Af

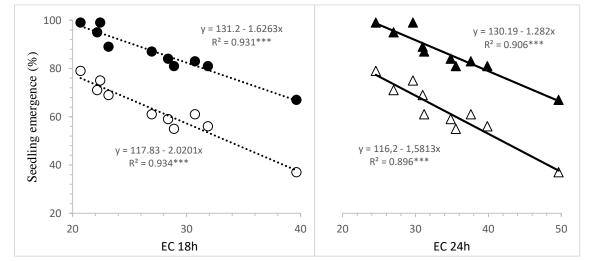
Emergence was different between seeds sown in waterlogged (excessive watering) and in field capacity conditions. In all seed lots, performing irrigation immediately after sowing led to lower emergence percentages compared to seeds sown in field capacity level. The differences were lowest in lot 1 and 4, as 20%, and the highest in lot 9 and 10, as 25 to 30% (Table 2).

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Gradual increase in EC values were observed in all seed lots as soaking time increased from 2 h to 24 h. In the final measurement of 24 h, EC varied between 24.5 (Lot 1) and 49.9 (Lot 10)  $\mu$ S cm<sup>-1</sup> g<sup>-1</sup>. Seed lots with low electrical conductivity after 18 and 24 h also showed higher seedling emergence in waterlogged and field capacity conditions. In both waterlogged and field capacity sowing emergence, the highest significant negative correlation was observed between conductivity after 18 (r=0.966; p<0.001; r=0.965, p<0.001) and 24 h (r=0.947; p<0.001; r=0.952, p<0.001) (Table 3).

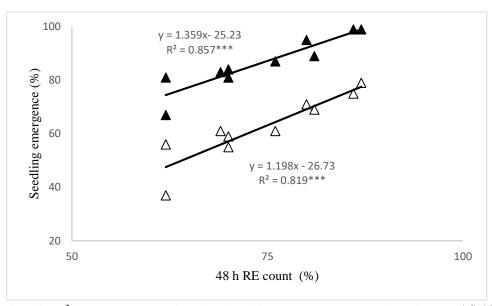
<b>Table 3.</b> Correlation coefficient (r) of between radicle emergence count (%) and EC ( $\mu$ S cm <sup>-1</sup> g <sup>-1</sup> ) values in different hours
of French bean seed lots with seedling emergence at waterlogged and field capacity sowings.

Crittania	Seedling Emergence Percentages			
Criteria	Waterlogged	Field capacity		
Total germination	0.765**	0.726*		
Normal germination	0.877***	0.897***		
Radicle Emergence Test (RE)				
24 h	0.369	0.427		
28 h	0.522	0.553		
32 h	0.615*	0.653*		
36 h	0.542	0.600		
40 h	0.684*	0.736*		
44 h	0.809**	0.838**		
48 h	0.905***	0.918***		
Electrical Conductivity (EC)				
EC 2 h	0.482	0.421		
EC 4 h	0.545	0.483		
EC 6 h	0.592	0.537		
EC 18 h	0.966***	0.965***		
EC 24 h	0.947***	0.952***		



**Figure 1.** Relationship between electrical conductivity (EC,  $\mu$ S cm<sup>-1</sup> g<sup>-1</sup>) values of 18 h and 24 h and seedling emergence at waterlogged ( $\circ, \Delta$ ) and field capacity ( $\bullet, \blacktriangle$ ) sowings in ten French bean seed lots.

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**Figure 2.** Relationship ( $R^2$ ) between RE test of 48 h and seedling emergence at waterlogged ( $\triangle$ ) and field capacity ( $\blacktriangle$ ) conditions in ten French bean seed lots.

The highest significant positive correlation between early radicle emergence after 48 h was correlated with waterlogged and field capacity emergence percentages (r=0.905, p<0.001, r=0.918, p<0.001). Results in Figure 1 and 2 showed that EC of 18 h and 24 h and RE of 48 h were highly related (p<0.001) to seedling emergence at waterlogged and field capacity sowings.

The significant negative correlation of leachate conductivity with seedling emergence suggests that a conductivity test for seed vigour could be applied to French bean to identify seed lots with poor emergence potential, which is already been introduced into the ISTA Rules (ISTA, 2017) as a validated vigour test. Radicle emergence test (RE) is found to be related to emergence percentages and seedling size in different crop seeds (Khajeh-Hosseini et al., 2010; Ermis et al., 2015). Our results indicated RE can possibly be used as vigour test in French bean seed lots. Further studies with larger numbers of seed lots are needed.

Differences in seed vigour, reflected in a range in emergence performance, can be attributed to imbibition damage (Legesse and Powell, 1996; Usha and Dadlani, 2015), which is related to testa colour. White coated beans adhere loosely to the seed coat, which allows the rapid movement of water through the gap between the seed coat and cotyledons, resulting in imbibition damage and poor emergence. On the other hand, the dark seed coat adheres closely to the cotyledons, thereby limiting water movement into the seed and reducing imbibition damage (Powell et al., 1986; De Souza and Marcos-Filho, 2001). In our study, we compared the vigour of white coated seed lots. Excessive watering immediately after sowing reduced emergence percentages compared to sowing in field capacity. It is obvious that excessive watering after sowing induced imbibition damage, which resulted in lower emergence. Additionally, vigour differences reflected in emergence percentages in the same planting method can be due to physiological ageing, such as storage, pre-harvest factors (Khajeh-Hosseini et al., 2010). Seed germination and emergence rate may not be successful in unfavorable environmental and soil conditions. Especially if the soil structure is degraded, there is no quality emergence. Because the water can not keep the soil and the condition will not be suitable for plant development.

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For bean lots sown in both waterlogged and field capacity conditions, electrical conductivity readings at 18 and 24 h in field capacity waterlogged conditions and at 48 h of RE were highly correlated with seedling emergence. Relationship between earlier EC readings and at 48 h of RE were not as high as these aforementioned correlations. Similar between relationships EC, early radicle emergence count and seedling emergence were also reported in previous studies (Khajeh-Hosseini et al., 2010; Matthews et al., 2009).

# CONCLUSION

Both methods can potentially be used as quick, cheap and simple methods for seed vigour testing prior to sowing. Routine testing EC readings can be done overnight (18 hours) or a daily routine (24 hours). RE test of 48 hours is slightly longer but it also gives results in two days. The tests would be particularly useful for distinguishing seed vigour ranking in bean seed lots when sowing in adverse conditions, such as excessive watering after sowing.

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