

## Germination and Vigor of Crambe Seeds Treated with Polymers under Hydric Stress

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**Abstract:** Seed treatment with polymers can be favorable for germination and the establishment of plants in deficient or hydric saturated soils. This study aimed to determine the effectiveness of the treatment with polymers under different conditions of substrate water availability on the physiological quality of crambe seeds. The seeds of 2 lots with a germination of 82 and 69% underwent the following treatments: Lot 1 with and without polymer and lot 2 with and without polymer. The polymer used was Solid Resin GV5 at a dose of 0.6 mL kg<sup>-1</sup> of seeds. In gerbox, the seeds were evaluated by germination, first counting, shoot and radicle length and seedling dry mass tests. In trays, the seeds were sown in sand substrate and humidity kept at 80, 60, 40, 20 and 10% of capacity retention. Seedlings were evaluated speed emergency index, sand emergency, shoot and radicle length and seedling dry mass tests. Researchers used a complete randomized 2×2 factorial block design (lot×polymer) for tests carried out in gerbox and a 2×2×6 factorial block design (lot×polymer×capacity retention) for tests carried out on trays. The best seed physiological performance is obtained at a capacity retention of 40%; however the germination of seedlings is not affected until the capacity retention ranges from 20-60% showing the species tolerance to water deficit conditions. Film coating is favorable to good physiological quality seeds but it also shows no change for low vigor crambe seeds.

**Key words:** Water deficit, hydric saturation, polymerization, physiological quality, Brazil

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### INTRODUCTION

Originating in the Mediterranean region, *Crambe abyssinica* (Brassicaceae) is an oilseed species which can be used in the biodiesel production, it is highly precocious, drought and frost tolerant and it has a low production cost. Machado report that crambe grows into antagonistic climatic conditions, withstanding low temperatures with frost formation in Southern Brazil and hot, dry temperatures in Midwestern Brazil. According to Ruas, it presents itself as a typical Autumn-Winter plant and it can be grown in off-season, not competing with main crops. Under these crop conditions and low water requirement, it grows atypically where most crops would not grow.

Deficient or water saturated soils may affect the germination and vigor of seeds and disadvantage the initial stand of plants, as well decrease the absorption ability of the root system, reducing the productive performance of the plant. Some practices such as seed treatment with polymers may be favorable to improve the germination performance and the establishment of plants when exposed to unfavorable conditions of abiotic

stress. According to Diniz, film coating is a technique that consists on coating or covering seeds with polymers, enabling an excellent adhesion of fungicides and insecticides to seeds, favoring the addition of agricultural inputs; moreover, it reduces contamination risks to man and to environment, allowing, thus an easy differentiation between treated and untreated seeds, without changing size or shape and protecting seeds from pathogen attacks.

The application of polymers provides a durable cover, water permeable with possible application to seeds of different shapes and sizes, maintaining the original size and shape; it regulates the germination process and allows for maximum penetration and retention of active products.

The benefits of the use of polymers in seed treatments have been reported for various species such as soybean, cotton and rice. However, researches regarding crambe crops are incipient and thus, there is no standardization of methodologies for seed coating and there are no performance data from seeds treated with these products.

Thus, the aim of this study was to determine the effectiveness of the treatment with polymers under different water availability of the substrate on the physiological quality of crambe seeds.

## MATERIALS AND METHODS

Crambe seeds, cultivar FMS Brilhante, from 2 lots (lot 1 and 2 with germination of 82 and 69%, respectively) were grown at Universidade Federal de Santa Maria (UFSM), in the agricultural year 2011/12 and stored with 10% humidity in a cold and dry room (at 15°C and relative 45% humidity). At the seed laboratory of UFSM, they underwent the following treatments: Lot 1 with polymer; lot 1 without polymer; lot 2 with polymer and lot 2 without polymer. The polymer used was Solid Resin GV5 (Laborsan, Brazil) at a dose of 0.6 mL kg<sup>-1</sup> of commercial product seeds. The seeds were not treated with fungicide and/or insecticide.

After treatment, the seeds remained at rest for 24 h and then, they were evaluated regarding physiological characteristics by the following tests:

**Germination:** It was performed by sowing 200 seeds per treatment, divided into 4 replicates of 50 seeds in gerbox plastic boxes, on germitest paper, moistened in a KNO<sub>3</sub> solution at 0.2%. The boxes were taken to the germination chamber BOD type, under constant light at 25°C. Seedling countings were performed on the 4th day for vigor (first count) and on the 7th day (germination test final counting). The results were expressed as a percentage of normal seedlings, according to Brasil.

**Radicle and shoot length (epicotyl):** This test was performed at the end of the germination test in normal seedlings (radicle and shoot) with 4 replicates of 10 seedlings each one, using a graduated scale to obtain the average value, expressed in centimeters.

**Seedling dry mass:** It was performed together with the seedling length test, where 4 replicates of 10 seedlings were placed in Kraft paper bags and taken to greenhouse at 70°C, for 48 h and then they were weighed on an analytical balance with accuracy of 0.0001 g and the results were obtained by the average of replicates and expressed in mg per seedling. The same treatments were sown in plastic trays with sand substrate previously washed, sterilized and sieved by using 50 seeds in one inch deep furrows, five inches spaced with 4 replications for each treatment. The humidity of the substrate was maintained at 100, 80, 60, 40, 20 and 10% of Capacity Retention (CR) with daily irrigation, according to the

methodology described by Brasil. Distilled water was applied at a corresponding volume to the difference of the mass of the set sand+tray at reading time and the last weighing the day before. The seedling evaluation was performed by the following tests:

**Emergency Speed Index (ESI):** This performed daily, from seedling emergency up to 1 inch ground. These seedlings were counted and the ESI was calculated using by the methodology proposed by Maguire (1962).

**Sand emergency:** The final number of seedlings after a constant value has been reached was expressed as a percentage and termed sand emergency percentage.

**Radicle and shoot length (epicotyl):** It was performed at the end of the emergency test in normal seedlings.

**Seedling dry mass:** It was performed together with the seedling length test, according to the methodology described before.

Researchers used a complete randomized block design with 4 replications in a factorial 2×2 (lot×polymer) for tests carried out in gerbox and 2×2×6 (lot×polymer×field capacity) for tests carried out on trays. Data were subjected to analysis of variance and polynomial regression analysis. Significant averages (qualitative factor) underwent the Tukey-test (p<0.05) and the models were chosen based on the significance of the degree of the equation acceptance and evaluated within the 3rd degree by adopting levels of 5% probability by using the F-test.

## RESULTS AND DISCUSSION

According to variance analysis (Table 1), for the variables: Shoot Length (SL), Primary Root Length (PRL) and Dry Mass (DM), a significant interaction between lot×polymer was observed. Thus, each lot showed a distinct performance when they underwent the treatment with polymer. Table 2 reveals that there was a significant interaction between lot×polymer×field capacity for the analyzed variables, except for the dry mass variable showing different effects for lot and polymer in different field capacities.

Table 3 presents the results of vigor tests (first counting) and germination performed in gerbox, at 25°C. The highest germination and vigor percentages were observed on lot 1 and there were no differences in these characteristics for seeds with or without polymer. These results are consistent with researches using polymers where film coated seeds did not damaged the

Table 1: The variance analysis containing Variation Sources (VS), Freedom Degrees (FD), Average Squares (AS) and the Variation Coefficient (VC) for the variables: Shoot Length (SL), Radicle Length (RL), Dry Mass (DM), first counting (V) and Germination (G)

VS	FD	AS				
		SL	RL	DM	V	G
Lot (L)	1	1.3983*	0.0612*	0.0004*	7.5013*	175.3638*
Polymer (P)	1	1.3983*	0.5814*	0.00002 <sup>NS</sup>	0.5814 <sup>NS</sup>	5.4873 <sup>NS</sup>
L×P	1	4.3160*	0.7439*	0.0006*	0.5814 <sup>NS</sup>	8.8357 <sup>NS</sup>
VC (%)		10.4600	16.9200	5.0500	7.2000	6.6900

Table 2: The variance analysis containing Variation Sources (VS), Freedom Degrees (FD), Average Squares (AS) and the Variation Coefficient (VC) for the variables: Shoot Length (SL), Radicle Length (RL), Dry Mass (DM), Emergency Speed Index (ESI) and Emergency (E)

VS	FD	AS				
		SL	RL	DM	ESI	E
Lot (L)	1	0.1920*	0.6142*	0.0064 <sup>NS</sup>	0.2832 <sup>NS</sup>	71.7257 <sup>NS</sup>
Polymer (P)	1	3.6808*	1.0881*	0.0064 <sup>NS</sup>	0.0672 <sup>NS</sup>	0.0094 <sup>NS</sup>
Field Capacity (FC)	4	44.0222*	9.2737*	0.0252 <sup>NS</sup>	320.4072*	10784.6700*
L×P	1	2.5205*	0.0074*	0.0135 <sup>NS</sup>	0.0252 <sup>NS</sup>	1.6560 <sup>NS</sup>
L×FC	4	9.0184*	1.3736*	0.0141 <sup>NS</sup>	1.1203 <sup>NS</sup>	55.3925 <sup>NS</sup>
P×FC	4	6.2325*	0.8799*	0.0126 <sup>NS</sup>	3.6476*	143.2408*
L×P×FC	4	3.8008*	1.2830*	0.0133 <sup>NS</sup>	2.0420*	121.8289*
VC (%)		11.6900	18.1000	12.6700	14.5400	14.3900

\*. <sup>NS</sup>Significant at 5% error probability by F-test and Not Significant, respectively

Table 3: Germination average (%), first counting (%), shoot length (cm), radicle length (cm) and dry mass (g) of crabe seedlings subjected to seed treatments with polymer<sup>1</sup>

Seed treatments	Lot 1	Lot 2	Average
<b>Germination</b>			
With polymer	78	70	74 <sup>a</sup>
Without polymer	82	70	76 <sup>a</sup>
Average	80 <sup>A</sup>	70 <sup>B</sup>	
<b>First counting</b>			
With polymer	52	56	54 <sup>a</sup>
Without polymer	60	50	55 <sup>a</sup>
Average	56 <sup>A</sup>	53 <sup>B</sup>	
<b>Shoot length</b>			
With polymer	4.08 <sup>Bb</sup>	5.71 <sup>Aa</sup>	4.89
Without polymer	5.71 <sup>Aa</sup>	5.25 <sup>Aa</sup>	5.48
Average	4.89	5.48	
<b>Radicle length</b>			
With polymer	0.94 <sup>Bb</sup>	1.49 <sup>Aa</sup>	1.21
Without polymer	1.74 <sup>Aa</sup>	1.44 <sup>Aa</sup>	1.59
Average	1.34	1.47	
<b>Dry mass</b>			
With polymer	0.05 <sup>Bb</sup>	0.08 <sup>Aa</sup>	0.06
Without polymer	0.07 <sup>Aa</sup>	0.07 <sup>Aa</sup>	0.07
Average	0.06	0.07	

<sup>1</sup>Values followed by the same lowercase upright and uppercase horizontally do not differ among themselves by Tukey-test, p<0.05

physiological quality of seeds in several crops. Positive results from the use of film coats are reported when seeds are sown in soil in which environmental conditions are not favorable to the germination process as moist and cold soils and also those with low hydric potential (Taylor *et al.*, 2001). According to Ni and Biddle

(2001), these hydrophilic or hydrophobic film coats delay the water entrance into seeds reducing the damage of water soaking at cold temperatures and increase the survival of the seeds. However Trentini, report that the efficiency of these polymers depends on the characteristics of each species, as well as the materials used for film coating.

**For variables:** Shoot length, radicle length and seedling dry mass, the best results were obtained in lot 1 seeds without polymer; however in lot 2 seeds, there were no differences in the seeds with or without polymer (Table 3). Trentini found that polymer can be favorable or not, depending on the initial quality of the seed lot. Thus according to the, researcher for low-vigor lots, it is not recommended to use these products because they may restrict the water entrance and oxygen, resulting in poor germination or abnormal seedlings formation. Similarly Lima, concluded that film coating was harmful to low-vigor cotton seeds; however it did not affect germination, emergency and emergency speed index of high quality seeds.

For shoot length (Fig. 1a), researchers observe that the Maximum Technical Efficiency (MTE) was obtained at capacity retention of 42% (CR42) in lot 1 by using polymer and of 53% in lot 1 without polymer showing that the polymer use has contributed positively to shoot growth under lower water availability conditions. The same trend was observed in lot 2 with polymer with MTE of CR57 and lot 2 without polymer and MTE of CR61.

This trend was also observed in polymerized seeds in lot 1 with MTE for radicle length (Fig. 1b) of CR40 and CR44 in lot 1 seeds without polymer; however in lot 2 seeds with or without polymer MET was of CR50. The MTE for ESI and seedling emergency was between capacity retention of 44-46 and 43 and 44, respectively (Fig. 2 and 3). For seedling dry mass variable (Fig. 2b) no significant effect was observed. According to Evangelista, polymer use in soybean seeds helped in the regulation of seed soaking and therefore, it provided reduction of the damage caused in the process resulting in a greater seed emergency percentage. Taylor *et al.* (2001) found that bean seeds coated showed an increased germination because polymer delayed the water entrance the first four hours, thus reducing soaking damages. Kaur and Bishnoi (2011) showed that the combination of different types and concentrations of polymers and insecticides increased the germination of canola seeds under different soil hydric stress; however according to the researchers, the efficiency of seed coating will depend on the cultivar used and environmental conditions such as soil type and the hydric potential in soil.

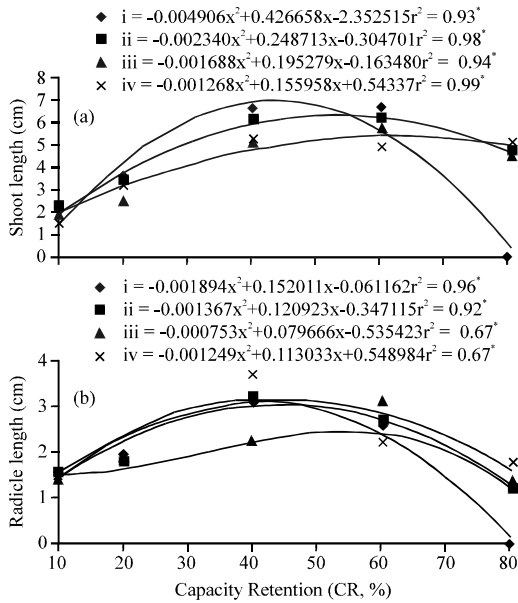


Fig. 1: Estimate of: a) Shoot length; b) Radicle length of crambe seedlings subjected to polymerized seed treatments and water availability conditions of the substrate: i) Lot 1 with polymer; ii) Lot 1 without polymer; iii) Lot 2 with polymer; iv) Lot 2 without polymer; \* $p < 0.05$

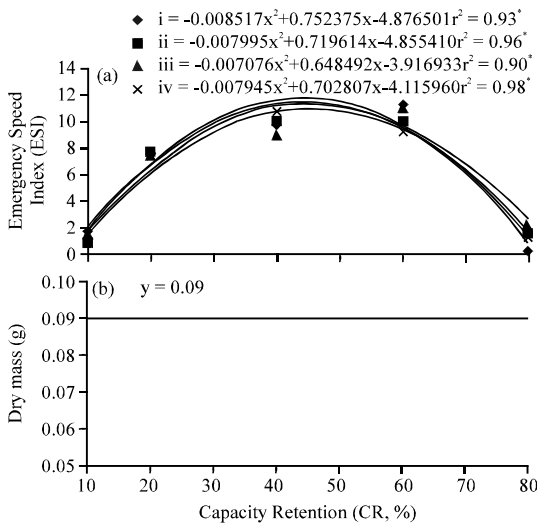


Fig. 2: Estimate of: a) Emergency Speed Index (ESI); b) Dry mass of crambe seedlings, subjected to polymerized seed treatments and water availability conditions of the substrate: i) Lot 1 with polymer; ii) Lot 1 without polymer; iii) Lot 2 with polymer; iv) Lot 2 without polymer; \* $p < 0.05$

Thus, it is clear that for these variables in general, the best results were obtained at capacity retention close to

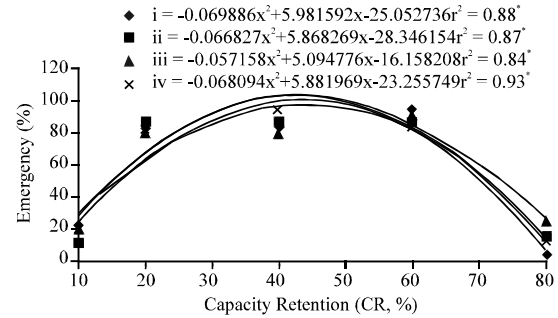


Fig. 3: Estimate of crambe seedling emergency subjected to polymerized seed treatments and water availability conditions of the substrate: i) Lot 1 with polymer; ii) Lot 1 without polymer; iii) Lot 2 with polymer; iv) Lot 2 without polymer; \* $p < 0.05$

40% (CR40) with a reduction of the values on substrates with lower (CR20 and 10) and higher water availability (CR60 and 80). Teixeira observed that more negative osmotic potential caused significant reduction of germination and of crambe seed vigor, with no normal seedling at potentials below  $-0.6$  MPa. Masetto also found that as the water availability of the substrate decreases, there is a reduction of the epicotyl and root and of the fresh mass of crambe seedlings. According to Marcos Filho, lower vigor values can be explained by the need to have an appropriate hydration level during the seed soaking stage, so that this will allow the reactivation of metabolic processes, resulting in the growth of the embryonic axis.

In CR60, 40 and 20 seedling emergency values above 80% were observed, with reduction in minor and major CRS (Fig. 3). These results show the tolerance of the species to the hydric stress conditions tested. According to Bewley and Black (1994), there is a great response variation to water stress among the species, from those very sensitive ones, to the most resistant ones so that resistant species seeds have the ecological advantage of establishing seedlings on drought-sensitive species. Roscoe reported that crambe has a high resistance to water deficit during the vegetative development stage but it does not tolerate excessive rainfall or high relative humidity and it also requires humidity in the soil to ensure an appropriate germination and the crop establishment. However Oplinger *et al.* (1991) reported that under stress conditions, crambe plants can develop long roots which confers it to be drought tolerant when compared to corn, canola, mustard or soy crops at all development stages.

In view of the roughness and the potential use of crambe crop and the diversity of growing environments in Brazil, the use of seed polymers may be an interesting tool

for the plant establishment in water stress-prone areas; however due to the wide variety and combination products, this technology requires more specific studies for its use on a larger scale.

### **CONCLUSION**

The best seed physiological performance is obtained at the capacity retention of 40%; however seedling emergency is not affected until the capacity retention ranges from 20-60%, showing the species tolerance to water deficit conditions.

Film coating is favorable to good physiological quality seeds but there is no difference for low-vigor crambe seeds.

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