

## ELECTRICAL CONDUCTIVITY AND ETHANOL RELEASE TO ASSESS RED RICE SEED VIGOR<sup>1</sup>

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**ABSTRACT** - To evaluate seed vigor, electrical conductivity and ethanol tests are fast and efficient methodologies. They have the potential to be used in several species, such as red rice. However, there are no protocols or information about their efficiency. Thus, the objective was to evaluate the efficiency, and define parameters of execution for electrical conductivity and ethanol tests, to evaluate the vigor of red rice seeds. The study was conducted using four lots of 'BRS 901' red rice, which was subjected to a germination test, as well as first count, accelerated aging, and field seedling emergence tests. The electrical conductivity test was performed with 25, 50, and 100 seeds soaked in 50 mL and 75 mL of water, at 25 °C and 30 °C, for 3, 6, 20, and 24 hours, respectively. The ethanol test was performed with 50 and 100 seeds soaked in a volume of water equivalent to 1.0, 1.5, 2.0, 2.5, and 3.0× the mass of the seed sample. To assess the vigor of red rice seeds, the electrical conductivity test is an efficient method when conducted with 50 seeds soaked in 50 mL of water at 25 °C for 20 hours. Meanwhile, the ethanol test is most effective when performed with 50 seeds, in a volume of water that is 2.5× the mass of the sample, at 40 °C for 24 hours.

**Keywords:** *Oryza sativa* L.. Breathalyzer. Fermentation. Imbibition. Leaching.

## CONDUTIVIDADE ELÉTRICA E LIBERAÇÃO DE ETANOL PARA AVALIAR O VIGOR DE SEMENTES DE ARROZ VERMELHO

**RESUMO** - Para avaliar o vigor das sementes, os testes de condutividade elétrica e etanol são metodologias rápidas e eficientes. Eles têm potencial para serem usados em diversas espécies, como o arroz-vermelho. No entanto, não existem protocolos ou informações sobre sua eficiência. Assim, objetivou-se avaliar a eficiência, e definir parâmetros de execução para testes de condutividade elétrica e etanol, para avaliar o vigor de sementes de arroz vermelho. O estudo foi realizado com quatro lotes de arroz vermelho 'BRS 901', que foi submetido a teste de germinação, primeira contagem, envelhecimento acelerado e testes de emergência de plântulas em campo. O teste de condutividade elétrica foi realizado com 25, 50 e 100 sementes embebidas em 50 mL e 75 mL de água, a 25 °C e 30 °C, por 3, 6, 20 e 24 horas, respectivamente. O teste de etanol foi realizado com 50 e 100 sementes embebidas em um volume de água equivalente a 1,0, 1,5, 2,0, 2,5 e 3,0 × a massa da amostra de sementes. Para avaliar o vigor de sementes de arroz vermelho, o teste de condutividade elétrica é um método eficiente quando realizado com 50 sementes embebidas em 50 mL de água a 25 °C por 20 horas. Já o teste do etanol é mais eficaz quando realizado com 50 sementes, em um volume de água 2,5 × a massa da amostra, a 40 °C por 24 horas.

**Palavras-chave:** *Oryza sativa* L.. Bafômetro. Fermentação. Embebição. Lixiviação.

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## INTRODUCTION

Red rice is one of the main components in the diet of the population that inhabits a large part of the Brazilian northeastern semiarid area. It is mainly cultivated as a subsistence crop (MENEZES et al., 2011). The rice is in high demand by consumers who offer high prices for it; thus, its cultivation favors the remuneration and sustainability of the family farming system (BRITO et al., 2014).

Despite its socioeconomic importance, red rice is sparsely studied in seed technology. To prioritize the use of high-quality seeds so that the stand is uniform, the plants are vigorous, and there is greater productivity potential (ORNELLAS et al., 2020), the evaluation of the physiological potential of the seeds is essential. The rice seeds with low seed vigor would result in a poor establishment of the crop and lead to great yield losses. This suggests that the seed vigor plays a vital role in increasing productivity (COSTA; KODDE; GROOT, 2014; WANG et al., 2020a).

Germination and vigor tests have been used to evaluate the physiological potential of seed lots. A germination test assesses a seed's ability to germinate under suitable environmental conditions, and determines the maximum potential under ideal and controlled conditions. It is widely used and one of its advantages is that it has a standardized methodology, although that does not always reflect performance in the field. In this case, it is necessary to use vigor tests (COSTA; KODDE; GROOT, 2014; PRADO et al., 2019). The vigor tests are related to the potential of the lots to have acceptable germination rates, to emerge and develop properly under a wide variety of environmental conditions (TEKRONY, 2003).

Limitations of vigor tests are related to execution time and assessment subjectivity (PINTO et al., 2015). Rapid tests in seed quality control programs are essential for assessing the physiological potential of seeds, and have, for this reason, warranted permanent attention from technologists, producers, and researchers (COSTA; KODDE; GROOT, 2014; MATTHEWS et al., 2018; WANG et al., 2020b). As a result, there is a need for methods that allow results to be obtained quickly and are reliable, such as electrical conductivity and ethanol production.

The electrical conductivity test meets these characteristics and has great potential, because it is a fast method and benefits both industries and farmers alike (PRADO et al., 2019). It has been useful in the evaluating the seed vigor of several species such as common bean (LEÃO et al., 2012), peanut (BARBOSA et al., 2012), graniferous sorghum (MARCOS; DUTRA, 2018), forage pea (MACHADO et al., 2019), and soybean (PRADO et al., 2019). However, there is no specific information in previous literature on the methodology of the electrical conductivity test for red rice seeds.

The ethanol test is based on the quantification of volatile ethanol produced by seeds during imbibition or at the beginning of germination. In deteriorated seeds, loss of the membrane systems integrity (mainly of the mitochondrial ones) leads to alternative ways to obtain energy to perform germination processes and thus, produce ethanol by fermentation. Depending on the amount of volatilized ethanol, information about physiological problems related to seed deterioration can be obtained (BUCKLEY; BUCKLEY, 2009). The use of the ethanol test to assess vigor was effective for canola (BUCKLEY; HUANG, 2011; BUCKLEY; HUANG; MONREAL, 2013), cabbage (KODDE et al., 2012), corn (CHAENGSAKUL et al., 2019), and melon seeds (ORNELLAS et al., 2020).

Thus, in this study it was hypothesized that the number of seeds, the water volume, and period for imbibition, can lead to variations between electrical conductivity results. For the ethanol test, the hypothesis is that the number of seeds and sample heterogeneity, in terms of seed density and size, may influence the results.

For these reasons, and considering the lack of information, this study aimed to evaluate the efficiency and set protocols for the electrical conductivity test and ethanol release test to evaluate red rice (*Oryza sativa* L.) seed vigor.

## MATERIAL AND METHODS

The research was conducted in the Plant Production Laboratory of the Department of Agricultural and Environmental Sciences, State University of Santa Cruz (UESC), using red rice seeds (*O. sativa* L.), 'BRS 901'. These were represented by four seed lots produced in 2018, from the settlement Dois Riachões, Ibirapitanga - BA, in the period from April to June 2018.

To obtain vigor differences among seed lots, they were deteriorated. In this process, the seeds were deposited on a wire mesh, coupled in plastic boxes, with 40 mL of distilled water at the bottom. The boxes were incubated at 40 °C for 24, 48, and 72 hour periods. After each period, the seeds were kept at laboratory conditions (26 °C and 70% relative humidity) for drying, and identified as lot 1 - unaged, lot 2 - aged for 24 h, lot 3 - aged for 48 h, and lot 4 - aged for 72 h. Afterwards, the physiological potential was characterized by germination and vigor tests, according to the following assessments.

Water content (WC) - determined by the oven method at 105 ± 3 °C for 24 hours (BRASIL, 2009), using two repetitions with 2 g of seeds from each lot, with results expressed as a percentage (wet basis).

Germination (GE) - performed using four repetitions of 50 seeds per lot, distributed over three leaves moistened with distilled and sterilized water, equivalent to 2.5 times the weight of the dry

substrate (paper). The rolls were kept in a germinator at 30 °C. The evaluations were performed at five and 14 days after sowing, and the percentage of normal seedlings was computed (BRASIL, 2009).

First germination count (FC) - performed together with the germination test, computes the percentage of normal seedlings obtained on the fifth day (BRASIL, 2009).

Accelerated aging (AA) - the plastic box method (11 × 11 × 3.5 cm) was used, with the seeds distributed in a single layer on the canvas (MCDONALD; PHANEENDRANATH, 1978). A measure of 40 mL of distilled water was placed at the bottom of the boxes. The boxes were kept in a germination chamber, at 40 °C for 120 h. After the aging period, four replicates of 50 seeds for each lot were subjected to a germination test, following methodology previously described, with evaluations performed on the fifth day after sowing (BRASIL, 2009).

Field seedling emergence (SE) - conducted in 10 m<sup>2</sup> (10 × 1.0 m) beds with four subsamples of 50 seeds from each lot. The seeds were sown in furrows 2 cm deep with a spacing of 0.30 m × 0.05 m. During the test, weeding and irrigation were performed. Seedling emergence was evaluated 15

days after sowing, by counting seedlings with fully developed primary leaves. The results were expressed as a percentage of emerged plants for each lot.

Electrical conductivity - carried out by testing four lots with variations in the number of seeds (25, 50, and 100), water volume (50 mL and 75 mL), and soaking period (3, 6, 20, and 24 hours). The seeds were counted, weighed (0.001 g), and placed in plastic cups containing distilled water (in the volume determined for each treatment). They were then kept in a B.O.D chamber at 25 °C and 30 °C. After each imbibition period, the electrical conductivity was determined by a conductivity meter and the results were expressed in  $\mu\text{S cm}^{-1} \text{g}^{-1}$ .

Evaluation test of ethanol - For the four lots, combinations of two quantities of seeds (50 and 100 seeds) and five volumes (mL) of water (1.0, 1.5, 2.0, 2.5, and 3.0 × the mass of the seed sample) were considered. The seeds were placed in glass jars (30 mL), plus distilled water. The flasks were sealed with a rubber stopper and metal seal, and incubated at 40 °C for 24 hours. The ethanol produced was evaluated ( $\mu\text{g.L}^{-1}$ ), with a modified breathalyzer (ethylometer) (Alcotest 6810; Dräger Safety AG & Co. KGaA; Lübeck, Germany) (Figure 1).



**Figure 1.** Equipment for assessing seed vigor based on ethanol production (breathalyzer).

A completely randomized design with four replications was used, separately for each test conducted. For the electrical conductivity test, the treatments were distributed in a 4×3×2 factorial scheme (lots × seed quantity × water volume), separately for each temperature and period. For the ethanol production test, the treatments were distributed in a 4×2×5 factorial scheme (lots × seed quantity × water volume). For both tests, means were compared using post hoc Tukey's test ( $p < 0.05$ ). Linear correlation analyses were performed with electrical conductivity and field seedling emergence tests. All analyses were performed using SISVAR

5.6 (Lavras, Brazil) software.

## RESULTS AND DISCUSSION

Values of initial water content in red rice seeds were similar for the four lots, and ranged from 11.5% to 11.9% (Table 1). This is considered adequate because there was no interference from water content variation in the vigor tests results. It was reported that the initial water content in seeds influences seed vigor evaluation (BARBOSA et al., 2012). Lot 1 showed greater physiological potential,

as first count and field seedling emergence values were found to be high (Table 1). However, germination and accelerated aging tests did not evidence this aspect, as there was no difference

among the lots. This meets the assumption of the use of the vigor test, because the germination test did not classify seed lots.

**Table 1.** Water content (WC), germination (GE), first count (FC), accelerated aging (AA), and seedling emergence in the field (SE) of red rice seed lots of the cultivar BRS 901.

Lot	WC (%)	GE (%)	FC (%)	AA (%)	SE (%)
1	11.9	92 a <sup>1</sup>	90 a	89 a	73 a
2	11.8	91 a	71 b	86 a	63 b
3	11.9	87 a	69 b	86 a	47 c
4	11.5	86 a	68 b	84 a	35 d
CV (%)	-	5.5	16.9	5.8	28.9

<sup>1</sup>Means followed by the same letter in the column do not differ, by Tukey's test ( $p < 0.05$ ).

Regarding the results of the field seedling emergency test, there was a stratification of lots at different vigor levels (Table 1). This test classified lots into four vigor levels, with lot 1 having the best vigor, lots 2 and 3 being intermediates, and lot 4 being the worst. The seedling emergence in the field test constitutes a guiding parameter for the efficiency of other tests to assess the physiological potential of seed lots (PRADO et al., 2019).

For the electrical conductivity test, considering the various combinations, there was an increase in values over the imbibition period (Tables 2 and 3). All procedures showed significant leaching after 6-hour periods of imbibition, allowing the beginning of lot stratification with regard to the physiological potential. However, lot classification became more evident after a 20-hour period of soaking.

**Table 2.** Electrical conductivity ( $\mu\text{S cm}^{-1} \text{g}^{-1}$ ) of red rice seed lots of the cultivar BRS 901, using combinations of 25, 50, and 100 seeds, per 50 mL and 75 mL of distilled water, at 25 °C in each period of soaking.

Lot	Soaking (hours)				Soaking (hours)			
	3	6	20	24	3	6	20	24
	25 seeds / 50 mL				25 seeds / 75 mL			
1	13.1 a <sup>1</sup>	20.0 a	21.2 a	26.5 a	5.4 a	10.0 a	11.2 a	13.2 a
2	13.4 a	20.7 a	25.8 ab	31.8 ab	5.2 a	10.5 a	11.7 a	13.5 a
3	13.9 a	21.2 a	28.2 b	34.8 ab	5.5 a	10.7 a	12.7 a	14.0 a
4	21.2 b	26.5 b	42.2 c	42.5 b	7.4 a	14.8 b	19.7 b	22.2 b
CV (%)	10.8	12.7	18.1	10.5	17.6	17.8	16.2	14.9
Lot	50 seeds / 50 mL				50 seeds / 75 mL			
	3	6	20	24	3	6	20	24
	50 seeds / 50 mL				50 seeds / 75 mL			
1	18.2 a	22.6 a	24.9 a	26.0 a	7.4 a	11.2 a	17.2 a	19.7 a
2	18.7 a	23.3 a	29.5 ab	31.0 b	13.4 b	14.5 ab	18.0 a	21.3 a
3	19.2 a	25.1 ab	31.5 b	33.9 b	14.7 b	15.6 ab	19.7 a	22.0 a
4	22.4 b	28.0 b	39.6 c	43.1 c	15.0 b	16.5 b	20.2 a	22.2 a
CV (%)	10.16	9.7	18.5	19.6	17.6	11.0	9.8	11.4
Lot	100 seeds / 50 mL				100 seeds / 75 mL			
	3	6	20	24	3	6	20	24
	100 seeds / 50 mL				100 seeds / 75 mL			
1	17.0 a	21.6 a	36.6 a	39.2 a	11.0 a	15.8 a	22.6 a	24.5 a
2	19.7 a	28.8 a	37.3 a	40.6 ab	12.9 ab	16.7 a	23.2 ab	27.8 a
3	23.5 a	28.7 a	43.2 ab	47.1 ab	15.5 ab	18.1 ab	25.8 ab	28.1 a
4	32.0 b	37.2 b	45.0 b	48.4 b	18.1 b	19.8 b	27.8 b	29.3 a
CV (%)	17.1	11.3	11.9	12.7	17.9	9.2	12.3	12.1

<sup>1</sup>Means followed by the same letter in the column do not differ, by Tukey's test ( $p < 0.05$ ).

**Table 3.** Electrical conductivity ( $\mu\text{S cm}^{-1} \text{g}^{-1}$ ) of red rice seed lots of the cultivar BRS 901, using combinations of 25, 50, and 100 seeds, per 50 and 75 mL of distilled water, at 30 °C in each period.

Lot	Soaking (hours)				Soaking (hours)			
	3	6	20	24	3	6	20	24
	25 seeds / 50 mL				25 seeds / 75 mL			
1	15.7 a <sup>1</sup>	24.0 a	25.5 a	31.8 a	6.5 a	12.1 a	13.4 a	15.8 a
2	16.6 a	24.9 a	30.9 b	38.8 a	6.3 a	12.6 a	14.1 a	16.2 a
3	17.1 a	25.5 a	33.8 b	41.8 ab	6.6 a	12.9 a	15.2 a	16.8 a
4	25.5 b	31.8 b	50.6 c	51.0 b	8.9 a	17.7 b	23.7 b	26.6 b
CV (%)	10.1	12.7	18.3	12.5	17.6	17.8	16.2	14.9
Lot	50 seeds / 50 mL				50 seeds / 75 mL			
	3	6	20	24	3	6	20	24
	50 seeds / 50 mL				50 seeds / 75 mL			
1	21.8 a	27.1 a	29.9 a	31.3 a	7.4 a	11.2 a	17.2 a	19.7 a
2	22.4 a	28.0 ab	35.4 b	37.3 b	13.4 b	14.5 ab	18.0 a	21.3 a
3	23.1 a	30.2 b	37.8 b	40.7 b	14.7 b	15.6 ab	19.7 a	22.0 a
4	26.9 b	33.6 c	47.6 c	51.8 c	15.0 b	16.5 b	20.2 a	22.2 a
CV (%)	10.2	9.7	18.5	19.6	17.6	11.0	9.8	11.4
Lot	100 seeds / 50 mL				100 seeds / 75 mL			
	3	6	20	24	3	6	20	24
	100 seeds / 50 mL				100 seeds / 75 mL			
1	20.4 a	25.9 a	43.9 a	47.1 a	13.2 a	18.9 a	27.1 a	29.4 a
2	23.6 ab	34.5 b	47.7 b	48.7 ab	15.5 ab	20.1 a	27.9 a	33.3 a
3	28.2 b	34.5 b	51.8 bc	56.5 ab	18.6 bc	21.7 ab	31.0 ab	33.7 a
4	38.4 c	44.6 c	54.0 c	58.1 b	21.7 c	23.8 b	33.3 b	35.1 a
CV (%)	17.1	11.4	11.9	12.7	17.9	9.2	12.3	12.1

<sup>1</sup>Means followed by the same letter in the column do not differ, by Tukey's test ( $p < 0.05$ ).

Water volume reduction was directly related to an increase in leaching for the four lots. For the water volumes for soaking, the greater the reduction in electrical conductivity values was, due to a dilution effect. Similar results were obtained with electrical conductivity tests for ryegrass (LOPES; FRANKE, 2010) and soybeans seeds (LOEFFLER; TEKRONY; EGLI, 1988). Thus, 50 mL of water for conducting the electrical conductivity test on red rice seeds proved to be more appropriate.

The association of 50 mL of water with 50 seeds indicates the best combination for carrying out the electrical conductivity test for red rice seeds (Tables 2 and 3). This combination allowed for better lot stratification and the methodology is the same as what is recommended for other crops, such as rocket salad (TORRES; PEREIRA, 2010) and ryegrass seeds (LOPES; FRANKE, 2010). The increase from 25 °C to 30 °C provided higher levels of exudate leaching, but maintained the same ranking during the imbibition periods (Tables 2 and 3). In general, high temperatures during imbibition increase the amount and speed of electrolyte leaching. Under these conditions, cell membranes became more fluid, and water and solute movement

were facilitated. A temperature of 25 °C was adequate for conducting the electrical conductivity test on red rice seeds, mainly associated with 50 mL of distilled water. This temperature is more consistent with the environmental conditions of seed analysis laboratories in tropical countries (LEÃO et al., 2012). There was a strong relationship between the electrical conductivity test and the emergence of seedlings in the field (Table 4). Both temperatures, after 20 hours of soaking, showed high values of correlation with the emergence of seedlings.

In red rice seeds, the electrical conductivity test showed a strong relationship with the emergence of seedlings in the field (Table 4) and provided early important information about its physiological potential. Some authors found that the electrical conductivity test also showed highly significant correlation with seedling emergence in the field for several species, such as peanut (BARBOSA et al., 2012), graniferous sorghum (MARCOS; DUTRA, 2018), and forage pea (MACHADO et al., 2019). Thus, coherence between the electrical conductivity results and performance in the field was established, as this is a premise for the effectiveness of a vigor test, because of limitations of the germination test.

**Table 4.** Simple correlation coefficients (r) between seedling emergence (in the field) and the electrical conductivity test of red rice seed lots of the cultivar BRS 901 conducted with 25, 50, and 100 seeds in 50 mL and 75 mL of water during 3, 6, 20, and 24 h of imbibition, at 25 °C and 30 °C.

Seeds (n)	Volume (mL)	3 h	6 h	20 h	24 h
25 °C					
25	50	-0.84 <sup>ns</sup>	-0.87 <sup>ns</sup>	-0.93 <sup>ns</sup>	-0.97*
	75	-0.81 <sup>ns</sup>	-0.85 <sup>ns</sup>	-0.87 <sup>ns</sup>	-0.82 <sup>ns</sup>
50	50	-0.89 <sup>ns</sup>	-0.97*	-0.96*	-0.97*
	75	-0.84 <sup>ns</sup>	-0.92 <sup>ns</sup>	-0.98*	-0.90*
100	50	-0.97*	-0.92*	-0.98*	-0.98*
	75	-0.87 <sup>ns</sup>	-0.99*	-0.99*	-0.99*
30 °C					
25	50	-0.84 <sup>ns</sup>	-0.87 <sup>ns</sup>	-0.93 <sup>ns</sup>	-0.97*
	75	-0.80 <sup>ns</sup>	-0.85 <sup>ns</sup>	-0.87 <sup>ns</sup>	-0.83 <sup>ns</sup>
50	50	-0.90 <sup>ns</sup>	-0.98*	-0.96*	-0.96*
	75	-0.84 <sup>ns</sup>	-0.92 <sup>ns</sup>	-0.99*	-0.91*
100	50	-0.97*	-0.92*	-0.98*	-0.98*
	75	-0.89 <sup>ns</sup>	-0.99*	-0.99*	-0.99*

<sup>ns</sup>No significant and \* Significant by t test ( $p \leq 0.05$ ).

For ethanol testing, considering lots and water volumes, regardless of the number of seeds used (50 or 100 seeds), lot 4 showed higher ethanol values, and low vigor (Table 5). The membrane disorganization and possible loss of permeability are related to initial events in the deterioration process.

Such events trigger problems in the resumption of activities by seeds after imbibition. These may include fluctuations in the respiratory rate, changes in enzyme activity, and reduced speed and germination capacity (DELOUCHE; BASKIN, 1973).

**Table 5.** Ethanol production ( $\mu\text{g.L}^{-1}$ ) by red rice seed lots of the cultivar BRS 901, using combinations of 50 and 100 seeds for every volume of distilled water at 40 °C with a 24-hour soak.

Lot	Seeds (n)	Amount of distilled water				
		1.0×	1.5×	2.0×	2.5×	3.0×
----- $\mu\text{g.L}^{-1}$ -----						
1	50	45.0 c	45.0 c	65.0 c	77.5 d	42.5 c
2		70.0 b	95.0 b	125.0 b	150.0 c	82.5 b
3		80.0 ab	110.0 ab	142.5 ab	195.0 b	95.0 b
4		95.0 a	132.5 a	180.0 a	280.0 a	145.0 a
CV (%)		12.6	11.6	14.7	11.1	13.2
1	100	62.5 b	65.0 b	90.0 d	220.0 c	50.0 b
2		117.5 a	195.0 a	230.0 c	272.5 c	115.0 b
3		130.0 a	215.0 a	317.5 b	360.0 b	157.5 b
4		137.5 a	225.0 a	437.5 a	490.0 a	215.5 a
CV (%)		14.2	19.2	9.5	9.3	17.7

<sup>1</sup>Means followed by the same letter in the column do not differ, by Tukey's test ( $p < 0.05$ ).

In order to define the ethanol test protocol, water volumes showed differences and identified variations in seed vigor. It was possible to stratify red rice seed lots from the volume of  $1.0 \times$  sample mass. For 50 seeds, a water volume of  $2.5 \times$  sample mass was used, while for 100 seeds, a water volume of  $2.0 \times$  sample mass was used. Lots were stratified into four vigor levels following the same ordering as the other tests performed, with lot 1 being more vigorous, lots 2 and 3 as intermediates and lot 4, the least vigorous.

Water is responsible for respiratory speed intensification, gas exchange benefits, and enzyme synthesis induction. The increase in respiration and/or fermentation rate is directly related to the degree of seed deterioration (MIRA et al., 2016). Thus, variation in water availability led to different metabolic responses depending on vigor level and the number of seeds in the sample (ORNELLAS et

al., 2020). However, when a water volume of  $3.0 \times$  sample mass was added, ethanol volatilization was reduced. This aspect is related to flooding, in which seeds are immersed in aqueous solution and ethanol does not volatilize. Thus, water volume of  $3.0 \times$  sample mass was not adequate for assessing seed vigor.

By correlation analysis (Table 6), the ethanol test conducted with 50 seeds and a water volume of  $2.5 \times$  sample mass, and 100 seeds using water volume of  $2.0 \times$  sample mass, were the combinations that most resembled the emergence of seedlings in the field test and seed lot classification. Considering the saving of seeds to compose the sample to be evaluated, plus the risk reduction of a particular seed being the outlier in terms of ethanol release, the amount of 50 seeds was established as the most appropriate amount.

**Table 6.** Simple correlation coefficients (r) between seedling emergence and ethanol test on red rice seed lots of the cultivar BRS 901, conducted with 50 and 100 seeds in different volumes of water.

Volume of water	50 seeds	100 seeds
1.0×	-0.8106*	-0.7363*
1.5×	-0.8647*	-0.6655*
2.0×	-0.8029*	-0.9387*
2.5×	-0.9175*	-0.9142*
3.0×	-0.8545*	-0.8652*

\*Significant by the t test ( $p \leq 0.05$ ).

The efficiency of electrical conductivity and ethanol tests for red rice seeds was evidenced by our ability to distinguish lots with differences in vigor, as well as by these tests having strong relations with the emergence of seedlings in the field. In this way, it is possible to obtain valuable information about the level of deterioration of the seeds, as well as to rank seed lots; thereby helping in decision making, whether for sowing, storage, or even disposal.

## CONCLUSION

To assess the vigor of red rice seeds, the electrical conductivity test is efficient when conducted with 50 seeds immersed in 50 mL of water at 25 °C, after 20 hours of soaking. Meanwhile, the ethanol test was most efficient when performed with 50 seeds, with a water volume of  $2.5 \times$  the mass of the sample, at 40 °C after 24 hours of soaking.

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