

# Seed vigor in reserve mobilization and wheat seedling formation

Matheus Santin Padilha<sup>1</sup><sup>(i)</sup>, Cileide Maria Medeiros Coelho<sup>1</sup>\*<sup>(i)</sup>, Gisiane Camargo de Andrade<sup>1</sup><sup>(i)</sup>, Natalia Carolina Moraes Ehrhardt-Brocardo<sup>1</sup><sup>(i)</sup>

<sup>1</sup> Universidade do Estado de Santa Catarina, Lages, SC, Brasil. E-mail: matheus santin@hotmail.com; cileidecoelho@yahoo.com.br; gisiane@mingotti.agr.br; biosapos@gmail.com

ABSTRACT: The interactions between the mobilization of seed reserves and vigor are fundamental to research the mechanisms that contribute to the formation and growth of vigorous seedlings and their establishment in the field. The objective of this study was to evaluate the dynamics of reserves during the process of seedling formation in wheat seeds with different vigor levels. Eleven wheat cultivars were used and germination, seedling emergence in the field, speed index and average time of emergence, shoot and root length, seedling and endosperm dry mass, reduction of seed reserves, reserve reduction rate, reserve mobilization rate and reserve use efficiency were evaluated. Wheat seed lots with greater vigor demonstrate higher seed reserve reduction rate, higher rate of mobilization of reserves and greater efficiency of use of seed reserves, producing seedlings with greater length and greater dry mass. It was possible to conclude that the seed lots with greater vigor have greater capacity of reserve depletion and mobilization, and the parameter determined as reserve mobilization rate can be used to evaluate the physiological quality of seed lots.

Key words: germination; physiological quality; Triticum aestivum L.

# Vigor de sementes na mobilização de reservas e formação de plântulas de trigo

**RESUMO:** As interações entre a mobilização de reservas e o vigor é fundamental na pesquisa dos mecanismos que contribuem para formação e crescimento de plântulas vigorosas e estabelecimento destas a campo. Objetivou-se com este estudo avaliar a dinâmica das reservas durante o processo a formação de plântulas em sementes de trigo com diferentes níveis de vigor. Foram utilizadas 11 cultivares de trigo, para quais foram avaliadas: germinação, emergência de plântulas em campo, índice de velocidade e tempo médio de emergência, comprimento de parte aérea e raiz, massa seca de plântulas e endosperma, redução de reservas da semente, taxa de redução de reservas, taxa de mobilização de reservas e eficiência de uso de reservas. Os lotes de sementes de trigo com maior vigor apresentaram maior taxa de redução de reservas, maior taxa de mobilização de reservas e maior eficiência de uso das reservas da semente, produzindo plântulas com maior comprimento e maior massa seca. Foi possível concluir que os lotes de sementes com maior vigor possuem maior capacidade de depleção de reservas e mobilização e, o parâmetro determinado como taxa de mobilização de reservas pode ser utilizado para a avaliação da qualidade fisiológica de lotes de sementes.

Palavras-chave: germinação; qualidade fisiológica; Triticum aestivum L.



# Introduction

Seed germination is fundamental to the life cycle of spermatophytes. It is a process divided into three phases that have physiological and metabolic characteristics well established in the literature. Upon hydration and metabolic resumption, hydrolysis and conversion of reserves stored in tissues begins (i.e., endosperm in monocots and cotyledons in dicots) (<u>Cheng et al., 2018</u>). Reserves are transformed into simple molecules, which are mobilized toward the embryonic axis (<u>Bewley & Nonogaki, 2017</u>) to be assimilated and utilized in the growth of seedling structures (e.g., roots, leaves, coleoptile, hypocotyl).

Considering the chemical composition of the seed, wheat consists of starch, proteins and cell wall polysaccharides, which together correspond to 90% of the seed dry mass, and components in smaller quantities that include lipids, terpenes, phenolic compounds, minerals and vitamins (Shewry et al., 2013). The reserves stored in the endosperm correspond to 70% of the starch and 10% of the proteins found in the seed (He et al., 2015).

Seedling growth and establishment in the field is a critical time for grain production, and is one of the main factors that determine the success or failure of this agricultural activity (Finch-Savage & Bassel, 2015). Thus, the use of seeds with higher physiological quality, such as germination and vigor, is a determining strategy to help obtain more uniform initial plant stands.

Among the tests available for quantifying vigor, those that evaluate seedling formation and performance are widely used, since they are associated with plant establishment in the field. In general, seeds with greater vigor form seedlings with greater length and greater dry mass (Marcos-Filho, 2015). Since seed vigor is expressed during seedling formation, the association between the processes of mobilization and utilization of reserves stored in the seed needs to be better elucidated by the science of seed technology.

Thus, reserve mobilization can be investigated from the evaluation of reserve use efficiency parameters, the mobilization of stored components (<u>Soltani et al., 2006</u>) and the energy expenditure required during seedling formation (<u>Andrade et al., 2019</u>). The association between these variables can generate information about the physiological differences between seed lots and contribute to the understanding of the importance of seed vigor during seedling formation.

Seed reserve mobilization can be influenced by several factors such as genotype (Pereira et al., 2015), seed size (Steiner et al., 2019) and physiological quality (Andrade et al., 2019), in which, seeds with higher hydrolysis capacity and efficiency are able to form seedlings of greater dry mass and length.

Regarding the physiological quality of wheat seeds, a pioneering study was that of <u>Khah et al. (1989</u>), in which seeds with differences in vigor also showed differences in dry matter production in the initial period of seedling growth, contributing to the final grain yield. Similarly, in recent

In view of the above, it is assumed that the wheat seed lots with higher vigor have a greater capacity for reserve mobilization and greater efficiency in the use of stored reserves, resulting in the formation of more vigorous seedlings. Thus, this study aimed to evaluate how the difference in seed vigor influences the mobilization of reserves in wheat seeds and the formation of vigorous seedlings.

studies, the influence of vigor on initial establishment and

# **Materials and Methods**

The evaluation of reserve mobilization dynamics was performed using 11 lots of wheat (*Triticum aestivum* L.) seeds. The seed lots were obtained from different distribution sites in the states of Rio Grande do Sul and Santa Catarina, Brazil, with different production and storage conditions. The cultivars used were TBIO Toruk (L1), TBIO Sossego (L2), BRS Marcante (L3), BRS 327 (L4), ORS Madre Pérola (L5), BRS Parrudo (L6), LG Oro (L7), BRS Tarumã (L8), BRS Bela Joia (L9), BRS Reponte (L10), and BRS Umbu (L11).

The thousand seed weight (TSW) was determined as described in the Rules for Seed Analysis (Brasil, 2009). For the moisture content (MC) four repetitions of 100 seeds were and oven-dried at  $105 \pm 3$  °C for 24 hours. After this period, the capsules with the dried seeds were weighed and then the average moistures of the repetitions for each lot were obtained. With the final weight (dried seeds) the seed dry mass (SDM) was obtained by dividing by the number of seeds used in the drying.

The germination test (G) was performed on paper towel sheets in roll form, which were kept in a germinator at a temperature of  $20 \pm 2$  °C. The counts of the number of normal seedlings were performed at four and eight days after the beginning of the test (Brasil, 2009). It was run with four repetitions of 50 seeds.

The vigor index (VI) was performed as proposed by <u>Silva et</u> <u>al. (2019)</u>. Four repetitions of 20 seeds were distributed on the upper third of paper towel sheets moistened with a volume of distilled water at a rate of 2.5 mL g<sup>-1</sup> of dry paper. The rolls were kept in a germinator at  $20 \pm 2$  °C for three days. After this period, the aerial part length (APL<sub>3d</sub>), root length (RL<sub>3d</sub>) and total seedling length (TSL<sub>3d</sub>) were measured using a digital caliper. The results were divided by the number of normal seedlings in the repetition and expressed in millimeters. The VI was generated from the growth and uniformity index data (<u>Sako et</u> <u>al., 2001</u>) with the aid of R software (<u>R Core Team, 2020</u>) using the SeedCalc package as proposed by <u>Silva et al. (2019</u>).

Seedling field emergence (FE) was performed using four repetitions of 50 seeds sown in soil at a depth of four centimeters, with each plot having 0.16 m<sup>2</sup>. The count of normal seedlings emerged was done daily until the fourteenth day, in order to obtain the emergence speed index (ESI) (<u>Maguire, 1962</u>) and the average time to emergence (ATE) (<u>Labouriau, 1983</u>).

For the mobilization of seed reserves, four repetitions of 20 seeds were used and distributed on the upper third of the paper towel moistened with distilled water at a rate of 2.5 mL g<sup>-1</sup> of dry paper. The paper rolls were kept in a germinator at  $20 \pm 2$  °C for six days. From the normal seedlings obtained, the total seedling length (TSL<sub>ed</sub>) was measured with the aid of a digital caliper. The seedling (root + coleoptile + plumule) was separated from the endosperm and both were dried in an oven at 80 °C for 24 hours, obtaining the seedling dry mass (SLDM) and the dry mass remaining in the endosperm (DMRE). The seed reserve reduction (SRR) was determined according to Equation 1; the seed reserve reduction rate (SRRR) (Pereira et al., 2015) was performed as highlighted in Equation 2 and expressed as a percentage; the reserve utilization efficiency (SRUE) (Soltani et al., 2006) was expressed in mg mg<sup>-1</sup> according to Equation 3; and, the reserve mobilization rate (RMR) (Andrade et al., 2019) was performed using Equation 4 and expressed as a percentage.

$$SRR = SDM - DMRE$$
(1)

$$SRRR = \left(\frac{SRR}{SDM}\right) \times 100$$
 (2)

$$SRUE = \left(\frac{SLDM}{SRR}\right)$$
(3)

$$RMR = \left(\frac{SLDM}{SDM}\right) \times 100$$
 (4)

The data were submitted to analysis of variance (ANOVA) and the comparison of averages was performed by the Scott-Knott test at 5% probability (p < 0.05). The parameters were submitted to Pearson simple correlation analysis, and the correlation coefficients (r) were obtained at 1 and 5% probability. Principal component analysis was used to evaluate the association between reserve mobilization and the obtained vigor groups. All analyses were performed with R software (<u>R Core Team, 2020</u>).

#### **Results and Discussion**

The moisture content (MC) of the seeds was evaluated with the objective of verifying the initial uniformity among the lots, reducing possible interferences in the tests performed. The difference in MC between the lots was at most 1.5%; a fact that justifies this being the only parameter that did not show a statistically significant difference by the F test (Fcalc > Ftab $\alpha$  = 0.05). In relation to the thousand seed weight (TSW), the lots presented a variation of 27.70 g (L8) to 41.61 g (L10) (Table 1). The TSW is dependent, among other factors, on the cultivar. The influence of seed mass on seedling formation

Table 1. Moisture content (MC) and thousand se	eed weight
(TSW) of 11 lots of wheat seeds.	

Lots	MC (%)	TSW (g)
L1	12.1	32.84 d <sup>1</sup>
L2	12.7	33.36 d
L3	11.2	34.55 c
L4	11.6	41.61 a
L5	12.6	33.47 d
L6	11.4	37.57 b
L7	12.5	30.79 e
L8	12.0	27.70 f
L9	11.5	32.82 d
L10	11.4	41.37 a
L11	11.3	33.60 d
Overall average	11.9	34.52
CV (%)	5.57	1.52

<sup>1</sup>Averages followed by the same letter in the column do not differ statistically by the Scott-Knott test at 5% probability. CV (%) - Coefficient of variation.

was highlighted by <u>Steiner et al. (2019)</u>, with this, it becomes important to determine this parameter, which makes it possible to associate this attribute with the mobilization of reserves and the vigor of the seed lot.

From the results obtained in the germination test (G), the lots presented germination percentages ranging from 78% (L11) to 97% (L1), being possible to identify differences in the physiological quality among lots (Table 2). For the marketing of wheat seeds, the Ministério da Agricultura, Pecuária e Abastecimento (MAPA) establishes in Normative Instruction No. 45 of 2013, a minimum germination percentage of 80% (Brasil, 2013). Given this requirement, only lot L11 showed results outside the standard required for marketing.

Considering the variable aerial part length  $(APL_{3d})$ , seed lots L1 and L5 stood out with the highest values (23.83 and 19.50 mm, respectively); lots L2, L3, L4, L6, L7, and L10 showed intermediate values, and lots L8, L9, and L11 showed the lowest values (11.95, 11.04, and 12.85 mm, respectively). For root length (RL<sub>3d</sub>) and total seedling length after 3 days

**Table 2.** Germination percentage (G), aerial part length after 3 days (APL<sub>3d</sub>), root length after 3 days ( $RL_{3d}$ ), total seedling length after 3 days ( $TSL_{3d}$ ), and vigor index (VI) of 11 lots of wheat seeds.

Lots	G	APL <sub>3d</sub> RL <sub>3d</sub>		TSL <sub>3d</sub>	· VI	
LOTS	(%)		(mm)		VI	
L1	97 a <sup>1</sup>	23.83 a	41.75 a	65.58 a	572 a	
L2	96 a	16.97 c	39.44 a	56.41 b	554 a	
L3	91 b	17.66 c	39.71 a	57.38 b	553 a	
L4	91 b	14.52 d	36.73 b	51.25 c	531 b	
L5	91 b	19.50 b	41.91 a	61.42 a	568 a	
L6	89 b	14.48 d	34.21 c	48.70 d	517 c	
L7	88 b	15.99 c	37.73 b	53.72 c	542 b	
L8	87 b	11.95 e	28.22 d	40.18 e	477 c	
L9	82 c	11.04 f	32.72 c	43.76 e	502 c	
L10	80 c	14.83 d	36.67 b	51.51 c	527 b	
L11	78 c	12.85 e	28.83 d	41.69 e	482 c	
Overall average	88	15.78	36.17	51.96	530	
CV (%)	3.11	5.51	6.90	5.92	3.12	

<sup>1</sup> Averages followed by the same letter in the column do not differ statistically by the Scott-Knott test at 5% probability. CV (%) - Coefficient of variation.

 $(TSL_{3d})$ , the statistical segregation between seed lots was more evident. In both, the seed lots that stood out were L1, L2, L3, and L5. Thus, at three days after germination it was possible to identify differences in seedling growth between the seed lots used (Table 2).

The vigor index (VI), similarly to the results obtained in the germination test,  $RL_{3d}$  and  $TSL_{3d'}$  allowed the distinction of lots L1, L2, L3, and L5, as lots of higher physiological quality in relation to the others. The lowest VI were observed for lots L6, L8, L9, and L11. The employment of this index for seed vigor evaluation was first proposed for lettuce (Sako et al., 2001) and is currently widely used in several cultivated species and presents great ability to segregate seed lots (Medeiros et al., 2019; Silva et al., 2019).

Total seedling length was also measured after six days of germination ( $TSL_{6d}$ ), which was the same time when reserve mobilization assessments were performed (<u>Table 3</u>). Similarly to  $TSL_{3d}$  (<u>Table 2</u>), lots L1 and L5 had the longest lengths,

**Table 3.** Total seedling length after 6 days  $(TSL_{6d})$ , emergence speed index (ESI), average time to emergence (ATE), and field emergence (FE) of 11 lots of wheat seeds.

Lots	TSL <sub>6d</sub> (mm)	ESI	ATE (days)	FE (%)
L1	207.45 a	6.11 a	7.42 b	90 a
L2	189.33 b	5.51 b	8.09 a	88 a
L3	182.76 b	4.95 b	7.89 b	78 a
L4	188.38 b	4.15 c	7.76 b	64 c
L5	203.24 a	5.47 b	7.77 b	84 a
L6	187.37 b	4.50 c	8.33 a	74 b
L7	185.51 b	5.58 b	7.80 b	86 a
L8	157.41 c	4.14 c	8.34 a	69 b
L9	168.47 c	3.72 d	8.34 a	62 c
L10	192.86 b	3.14 d	8.09 a	50 d
L11	164.46 c	3.55 d	8.71 a	61 c
Overall average	184.30	4.62	8.05	73
CV (%)	6.08	8.69	3.34	7.71

<sup>&</sup>lt;sup>1</sup> Averages followed by the same letter in the column do not differ statistically by the Scott-Knott test at 5% probability. CV (%) - Coefficient of variation.

207.45 and 203.24 mm, respectively (<u>Table 3</u>). The lots L8, L9, and L11, showed the shortest total seedling length after 3 days (TSL<sub>3d</sub>) (<u>Table 2</u>) and after 6 days (TSL<sub>6d</sub>) (<u>Table 3</u>), which indicates the lower vigor of these seed lots as highlighted by Kzyzanowski et al. (2020).

The field emergence (FE), the emergence speed index (ESI), and the average time to emerge (ATE) are parameters that make it possible to characterize the vigor of seed lots, as well as to represent the behavior in the field in relation to the vigor of lots evaluated in the laboratory. Distinctly, ESI and ATE determine the speed of germination, for which, the faster the seedlings establish themselves, the sooner they begin to utilize the resources of the environment such as water, nutrients, and light to grow and develop. Considering the 11 lots evaluated, 5 showed rapid emergence (ESI) and a higher percentage (FE), being L1, L2, L3, L5, and L7 (Table 3). The field emergence percentage is fundamental to estimate the ability of the seed lot to germinate under field conditions, and is a reference tool for the other tests used in the evaluation of vigor.

According to the results already exposed considering the variables used (i.e., G,  $APL_{3d'}$ ,  $RL_{3d'}$ ,  $TSL_{3d}$ , VI,  $TSL_{6d'}$ , ESI, ATE, and FE) it was found that there is difference in vigor among the lots analyzed, being possible the separation of the lots into two groups. The seed lots L1, L2, L3, L5, and L7 were considered to be of high vigor and; the seed lots L4, L6, L8, L9, L10, and L11 as of low vigor.

For seedling dry mass (SLDM) it was observed that lots L1, L3, L4, L5, and L10 showed the highest dry mass accumulation among the lots studied (Table 4). L1, L3, and L5 belong to the high vigor group highlighted earlier. However, the L4 and L10 with lower vigor showed similar mass accumulation to the seed lots in the high vigor group. According to Pereira et al. (2015) and Steiner et al. (2019) seedling dry mass can be influenced by the initial seed dry mass (SDM) of the lot. This statement can be observed in the results obtained for SLDM of lots L4 and L10 that presented the highest SDM among the lots studied and this influenced the formation of the seedling

Table 4. Seedling dry mass (SLDM), seed dry mass (SDM), dry mass remaining in endosperm (DMRE), seed reserve reduction (SRR), seed reserve reduction rate (SRRR), reserve mobilization rate (RMR), and seed reserve utilization efficiency (SRUE) of 11 lots of wheat seeds at six days after the start of the test.

Lata	SLDM	SDM	DMRE	SRR	SRRR	RMR	SRUE
Lots	_	(m	ng)	(%)		(mg mg⁻¹)	
L1	10.93 a <sup>1</sup>	29.11 d	14.31 c	14.55 b	50.43 b	37.87 a	0.75 a
L2	9.43 b	29.12 d	16.89 b	12.22 c	41.96 b	32.41 c	0.77 a
L3	10.33 a	30.29 c	12.79 c	17.49 a	57.71 a	34.10 b	0.61 b
L4	11.14 a	36.83 a	19.21 a	17.61 a	47.81 b	30.25 c	0.63 b
L5	11.01 a	29.11 d	11.43 c	17.67 a	60.73 a	37.83 a	0.62 b
L6	10.05 b	33.29 b	16.60 b	16.68 a	50.16 b	30.23 c	0.61 b
L7	9.35 b	26.96 e	12.97 c	13.98 b	51.87 b	34.70 b	0.67 b
L8	6.79 c	24.39 f	13.19 c	11.19 c	45.91 b	27.83 c	0.61 b
L9	8.91 b	29.12 d	14.10 c	15.01 b	51.55 b	30.58 c	0.59 b
L10	10.63 a	36.46 a	20.22 a	16.24 a	44.49 b	29.19 c	0.67 b
L11	9.57 b	29.63 d	14.60 c	15.03 b	50.72 b	32.30 c	0.64 b
Overall average	9.83	30.37	15.12	15.24	50.30	32.48	0.65
CV (%)	7.61	1.38	10.49	10.70	9.98	7.62	13.29

<sup>1</sup> Averages followed by the same letter in the column do not differ statistically by the Scott-Knott test at 5% probability. CV (%) - Coefficient of variation.

with the highest mass, however, it is not associated with the greatest vigor of a seed lot.

The mobilization of stored reserves to the seedling was determined by the reserve mobilization rate (RMR). The seed lots with high vigor (L1, L3, L5, and L7) showed the highest percentages of RMR, demonstrating the greatest ability to mobilize reserves for seedling formation. Lots L4 and L10, which had the highest seed dry mass, showed the lowest mobilization rates. Thus, it is possible to state that the amount of reserves a seed has does not guarantee the efficient mobilization of these reserves, and the way these reserves are used differentiates lots of high and low vigor. This confirms the need to understand the mechanisms that are involved in the dynamics of seed reserves, which involves above all, the mobilization of the compounds that are stored to the growing points.

The dry mass remaining in the endosperm (DMRE) represents the amount of reserve that remained stored after 6 days of germination. In principle, the higher the DMRE, the less reserves have been degraded and mobilized for the growth of the embryonic axis. However, this parameter can be influenced by seed size (TSW and SDM) and reserve utilization efficiency (SRUE). When analyzing the data from lots L5 (high vigor) and L9 (low vigor), which have similar SDM and SRUE, it is observed that the low vigor seed lots made fewer reserves available for seedling formation, and the SRR of the lower vigor seed lot is lower (Table 4), indicating less capacity for hydrolysis and mobilization of stored reserves.

The seed reserve reduction (SRR) and the seed reserve reduction rate (SRRR) represent the unit mass and percentage of seed reserves has been mobilized for seedling formation. The seeds of the 11 wheat seed lots mobilized between 41 and 61% of the mass present in the endosperm during the germination process, regardless of vigor (Table 4). Lots L5 and L3, both of high vigor, showed the highest percentages of reduction of seed reserves (SRRR), showing the influence of vigor on this variable. Similar results were observed by Andrade et al. (2019) in which hybrid corn seed lots with higher vigor, showed a higher rate of reduction of stored reserves, which favored the formation of seedlings with high

vigor. The degradation of reserves is carried out by the activity of enzymes, and the released products are directed to the embryonic axis (<u>Mahender et al., 2015</u>). Thus, the greater ability to reduce seed reserves is associated with hydrolysis activity, and in seeds of corn (<u>Heberle et al., 2019</u>) and wheat (<u>Chen et al., 2017</u>) seeds with high vigor showed higher enzyme activity.

The seed reserve utilization efficiency (SRUE) is an indicator that can determine the metabolic efficiency for seedling formation during the germination process. Among the lots evaluated, the highest efficiency was found in the high vigor lots L1 and L2. The other lots showed lower use efficiency and did not differ statistically from each other (Table Whereas SRUE evaluates the seedling dry mass production per unit of reserve used (Soltani et al., 2006), in the seedling formation process not all dry mass that is degraded from the seed reserve tissue is converted into seedling. Biochemical processes, among them, respiration, need these reserves to stay active. This indicates that seed lots with high vigor have lower metabolic consumption for the formation of a seedling with high vigor (i.e., higher dry mass and length). This association between SRUE and seed lot vigor (i.e., VI) can be seen in Table 5.

The correlation coefficients showed that the vigor index (VI), field emergence (FE), emergence speed index (ESI), total seedling length (TSL<sub>6d</sub>), and seedling dry mass (SDM) showed positive association with each other, highlighting the importance of vigor for initial seedling establishment in the field (Finch-Savage & Bassel, 2015). These parameters showed significant positive correlation with the reserve mobilization rate (RMR), indicating that the reserve mobilization capacity is related to seed vigor, which favors the formation of high performance seedlings (Table 5). These associations are observed by the results obtained for the high vigor group (i.e., L1, L2, L3, L5, and L7), which showed higher values in the parameters cited (Table 2, 3, and 4). Similar results were observed in Ehrhardt-Brocardo & Coelho (2016), for which seed lots of higher vigor showed seedlings with better performance arising from the high percentage of mobilized reserve.

Var.	FE	ESI	TSL <sub>6d</sub>	SLDM	TSW	SDM	DMRE	SRR	SRRR	RMR	SRUE	VI
FE	-	0.98**	0.36*	0.04 <sup>ns</sup>	-0,41**	-0.44**	-0.43**	-0.12 <sup>ns</sup>	0.22 <sup>ns</sup>	0.48**	0.20 <sup>ns</sup>	0.60**
ESI		-	0.47**	0.16 <sup>ns</sup>	-0,35 <sup>ns</sup>	-0.38*	-0.40*	-0.08 <sup>ns</sup>	0.22 <sup>ns</sup>	0.57**	0.26 <sup>ns</sup>	0.69**
TSL <sub>6d</sub>			-	0.79**	0,32 <sup>ns</sup>	0.30 <sup>ns</sup>	0.06 <sup>ns</sup>	0.36*	0.20 <sup>ns</sup>	0.68**	0.39*	0.84**
SLDM				-	0,60*	0.60**	0.28 <sup>ns</sup>	0.53**	0.16 <sup>ns</sup>	0.64**	0.39*	0.65**
TSW					-	$0.99^{**}$	$0.71^{**}$	0.58**	-0.11 <sup>ns</sup>	-0.21 <sup>ns</sup>	-0.03 <sup>ns</sup>	0.16 <sup>ns</sup>
SDM						-	0.73**	0.58**	-0.13 <sup>ns</sup>	-0.23 <sup>ns</sup>	-0.04 <sup>ns</sup>	0.12 <sup>ns</sup>
DMRE							-	-0.14 <sup>ns</sup>	-0.77**	-0.35*	0.43**	-0.12 <sup>ns</sup>
SRR								-	0.73**	0.09 <sup>ns</sup>	-0.57**	0.31*
SRRR									-	0.32*	-0.64*	0.30*
RMR										-	0.50**	0.69**
SRUE											-	0.33*
VI												-

Table 5. Pearson simple correlation coefficients (r) among the variables related to reserve mobilization.

Legend: Not significant (<sup>ns</sup>), significant at 1% (\*\*), and significant at 5% (\*) probability by t-test. Variables (Var.); Field emergence (FE); emergence speed index (ESI); Total length of seedlings after six days (TSL<sub>6d</sub>); Seedling dry mass (SLDM); Thousand seeds weight (TSW); Seed dry matter (SDM); Dry mass remaining in the endosperm (DMRE); Seed reserve reduction (SRR); Seed reserves reduction rate (SRR); Reserve mobilization rate (RMR); Seed reserve utilization efficiency (SRUE); Vigor index (VI); n = 44 (11 lots × 4 repetitions).

The seed reserve reduction rate (SRRR) showed negative correlation with seed reserve use efficiency (SRUE) and with dry mass remaining in the endosperm (DMRE). This association between SRRR and SRUE was also verified for corn (Pereira et al., 2015) and soybean (Cheng et al., 2018), who reported that part of the seed dry mass is used by the metabolism to maintain vital functions, and this is destined for metabolic expenditure that must be considered (Table 5).

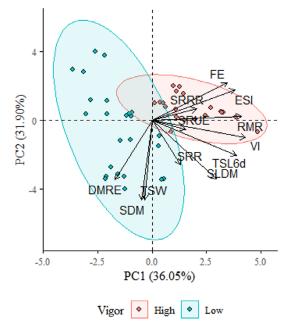
SRUE and DMRE were positively correlated with each other (r = +0.43). This result indicates that seed lots that presented higher use efficiency showed a tendency to conserve the resources available in the endosperm, that is, a smaller amount of resources was necessary for the formation of the seedling with greater mass (Table 5). This association can be observed in the results obtained from lots L1 and L9, in which, both presented DMRE without significant difference (i.e., 14.31 and 14.10 mg, respectively), however, lot L1 formed seedlings with higher dry mass (Table 4), which demonstrates the importance of efficiency for seedling formation.

The association of the high vigor (L1, L2, L3, L5, and L7) and low vigor (L4, L6, L8, L9, L10, and L11) groups obtained and the variables related to reserve mobilization, can be observed in principal component analysis (PCA). Principal components 1 (PC1) and 2 (PC2) were able to explain 36.05 and 31.90% of the data variance, respectively, for a total of 67.95% (Table 6).

In the first principal component it is possible to observe that seeds with high vigor have a positive association with SRRR and SRUE, favoring mobilization (RMR) and seedling formation (SLDM and TSL<sub>6d</sub>). SRUE and DMRE showed negative correlation between them (<u>Table 6</u>; <u>Figure 1</u>), indicating that seed lots with higher efficiency have the ability to use less reserve tissue resources for seedling formation. In contrast, the low vigor seed lots showed greater association with dry mass remaining in the endosperm (DMRE), indicating that they have a lower capacity to degrade stored reserves,

**Table 6.** Principal component analysis, eigenvalue, proportion of variance explained, and correlations between components and variables.

	PC1	PC2					
Importance of the componentes							
Eigenvalue	2.07	1.95					
Proportion of variance	36.05	31.90					
Cumulative variance	36.05	67.95					
Correlations between v	variables and comp	onents					
FE	0.72	0.45					
ESI	0.80	0.37					
TSW	-0.07	-0.96					
VI	0.90	-0.20					
TSL <sub>6d</sub>	0.81	-0.42					
SLDM	0.62	-0.71					
DMRE	-0.35	-0.72					
SDM	-0.10	-0.97					
SRR	0.27	-0.54					
SRUE	0.32	-0.10					
SRRR	0.43	0.14					
RMR	0.86	0.05					



**Figure 1.** Principal component analysis (PCA) for the reserve mobilization characters of 11 wheat seed lots grouped into high vigor (red) and low vigor (blue); n = 44.

consequently using fewer reserves and generally forming seedlings with lower dry mass.

Considering the results obtained for PC1, it was observed that seeds with high vigor have higher SRRR and higher SRUE, favoring mobilization (RMR) and seedling formation (SLDM and TSL<sub>60</sub>) (<u>Table 6</u>, <u>Figure 1</u>). One of the causes of the higher SRRR and subsequent mobilization may be associated with hydrolysis activity, according to <u>Tabatabaei et al. (2016)</u> and <u>Chen et al. (2017)</u>, the activity of the alpha-amylase enzyme is increasing during germination resulting in increased starch hydrolysis and supply of soluble sugars to the embryonic axis, being a determinant during germination and formation of wheat seedlings.

The SRRR and SRUE variables are negatively correlated with each other (Table 5, Figure 1 (PC2)), and their importance during seedling formation should be carefully considered. In general, the seeds have more dry mass than necessary for the establishment of a seedling, so it is plausible that the SRRR has great importance in the sense of rapid use of reserve components, which would favor seedling formation, even if the lot or cultivar shows less reserve utilization efficiency. Support for this approach can be seen in the results obtained for L2 (lot with higher efficiency) and L5 (lot with lower efficiency), L5 showed the highest SRRR, which provided support for forming a seedling with greater dry mass even though it showed lower efficiency compared to L2 (Table 4). Thus, the interaction between these parameters favored the formation of seedlings in the high vigor group (Table 5, Figure 1).

Considering that, the SRRR and SRUE variables are influenced by the genotype (<u>Pereira et al., 2015</u>; <u>Andrade et al., 2019</u>), it is suggested further studies with lots of the same cultivar contrasting in vigor, seeking to identify the relationship of initial seed vigor with reserve mobilization,

use efficiency and, consequently, seedling formation under different abiotic stress conditions, as well as, determine the chemical and biochemical changes during this process.

# **Conclusions**

The wheat seed lots with greater vigor have a greater capacity for reserve utilization and mobilization, forming seedlings with greater dry mass and length, carrying out this process with greater effectiveness and efficiency of reserve utilization.

Reserve mobilization rate, seedling dry mass, and total length have the greatest association with seed lot vigor, and can be used to segregate the vigor of different lots.

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#### **Compliance with Ethical Standards**

Author contributions: Conceptualization: MSP, CMMC; Data curation: MSP, GCA; Formal analysis: MSP; GCA; Funding acquisition: CMMC; Investigation: MSP, GCA, CMMC; Methodology: MSP, CMMC, NCMEB; Project administration: CMMC; Resources: CMMC; Supervision: CMMC, NCMEB; Validation: MSP; Visualization: MSP, GCA, NCMEB, CMMC; Writing – original draft: MSP, GCA; Writing – review & editin: CMMC, NCMEB.

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