

## Experimental design and precision in trials with black oat, common vetch, and forage turnip intercropping

Alberto Cargnelutti Filho<sup>1\*</sup>, Daniela Lixinski Silveira<sup>1</sup>, Marcos Vinícius Loregian<sup>1</sup>,  
Lucas Fillipin Osmari<sup>1</sup>, Vithória Morena Ortiz<sup>1</sup>

<sup>1</sup> Universidade Federal de Santa Maria, Santa Maria, RS, Brasil. E-mail: [alberto.cargnelutti.filho@gmail.com](mailto:alberto.cargnelutti.filho@gmail.com); [danielisil@gmail.com](mailto:danielisil@gmail.com); [vinicius.loregian@hotmail.com](mailto:vinicius.loregian@hotmail.com); [lucasfosmari@gmail.com](mailto:lucasfosmari@gmail.com); [vithoria.ortiz159@gmail.com](mailto:vithoria.ortiz159@gmail.com)

**ABSTRACT:** The objective of this study was to determine the optimal plot size for evaluating the fresh matter of black oat (*Avena strigosa* Schreb), common vetch (*Vicia sativa* L.), and forage turnip (*Raphanus sativus* L.), grown in intercrop, in scenarios formed by combinations of treatments numbers, repetitions numbers, and experimental precision levels. Six uniformity trials were conducted with the consortium of black oat, common vetch, and forage turnip, with three trials evaluated at 84 days after sowing (DAS) and the other three trials at 119 DAS. Fresh matter was assessed in 216 basic experimental units (BEU) of 1 × 1 m (6 trials × 36 BEU per trial). The heterogeneity index of Smith (1938) was estimated. Plot size was determined by the method Hatheway (1961) in scenarios formed by combinations of *i* treatments (*i* = 5, 10, 15, 20, and 25), *r* repetitions (*r* = 3, 4, 5, 6, 7, 8, 9, and 10), and *d* precision levels (*d* = 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, and 20%). To evaluate the fresh matter of black oat, common vetch, and forage turnip, in intercropping, with 5 to 25 treatments and with six repetitions, plots of 15 m<sup>2</sup> of usable area are sufficient for differences between treatments of 12% of the overall average of the experiment to show differences at the 0.05 level of significance.

**Key words:** *Avena strigosa* Schreb; optimal plot dimension; *Raphanus sativus* L.; repetitions numbers; *Vicia sativa* L.

## Dimensionamentos experimentais e precisão em ensaios com consórcio de aveia preta, ervilhaca e nabo forrageiro

**RESUMO:** O objetivo deste trabalho foi determinar a dimensão ótima de parcela para avaliar a matéria fresca de aveia preta (*Avena strigosa* Schreb), ervilhaca (*Vicia sativa* L.) e nabo forrageiro (*Raphanus sativus* L.), cultivadas em consórcio, em cenários formados por combinações de números de tratamentos, números de repetições e níveis de precisão experimental. Foram conduzidos seis ensaios de uniformidade com o consórcio de aveia preta, ervilhaca e nabo forrageiro, sendo três ensaios avaliados aos 84 dias após a semeadura (DAS) e os outros três ensaios aos 119 DAS. Foi avaliada a matéria fresca em 216 unidades experimentais básicas (UEB) de 1 × 1 m (6 ensaios × 36 UEB por ensaio). Foi estimado o índice de heterogeneidade de Smith (1938). Foi determinada a dimensão de parcela por meio do método de Hatheway (1961) em cenários formados pelas combinações de *i* tratamentos (*i* = 5, 10, 15, 20 e 25), *r* repetições (*r* = 3, 4, 5, 6, 7, 8, 9 e 10) e *d* níveis de precisão (*d* = 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19 e 20%). Para avaliar a matéria fresca de aveia preta, ervilhaca e nabo forrageiro, em cultivo consorciado, com 5 a 25 tratamentos e com seis repetições, parcelas de 15 m<sup>2</sup> de área útil são suficientes para que diferenças entre tratamentos de 12% da média geral do experimento apresentem diferenças ao nível de 0,05 de significância.

**Palavras-chave:** *Avena strigosa* Schreb; dimensão ótima de parcela; *Raphanus sativus* L.; número de repetições; *Vicia sativa* L.



## Introduction

Important contributions of the consortium of black oat (*Avena strigosa* Schreb), common vetch (*Vicia sativa* L.), and forage turnip (*Raphanus sativus* L.) (O+V+T) have been demonstrated (Rigon et al., 2011; Ziech et al., 2015; Wolschick et al., 2016; Michelon et al., 2019; Haskel et al., 2020). Increases in bean yields were obtained after black oat, common vetch, and forage turnip were grown alone and in consortia when compared to ryegrass and linseed alone (Rigon et al., 2011). The O+V+T consortium shows balanced C/N ratio and intermediate decomposition compared to single cropping (Ziech et al., 2015). At the beginning of the cycle, soil cover by the plant canopy is higher in the fodder turnip and O+V+T consortium crops while at the end it is higher in the common vetch and O+V+T consortium (Wolschick et al., 2016).

Evaluating winter cover crops grown alone and in a consortium over three agricultural seasons, Michelon et al. (2019) observed that the O+V+T consortium stood out with more dry matter and higher corn grain yields, grown in succession. They also concluded that the cultivation of cover crops for three consecutive years increased the organic matter content and availability of phosphorus and potassium in the soil. Haskel et al. (2020) concluded that black oat and the O+V+T consortium have faster development and higher initial soil cover capacity compared to common vetch and forage turnip grown alone.

In these experiments winter cover plants, in stand-alone and intercropping, were evaluated in three repetitions and with plots of 25 m<sup>2</sup> (Ziech et al., 2015), 40 m<sup>2</sup> (Haskel et al., 2020), and 48 m<sup>2</sup> (Michelon et al., 2019) or with four repetitions and plots of 18 m<sup>2</sup> (Rigon et al., 2011) and 30 m<sup>2</sup> (Wolschick et al., 2016). In these surveys, a randomized block design was used and the number of treatments ranged from 5 to 20. However, the criteria used to define the plot size and the number of repetitions were not mentioned.

The definition of experimental designs in trials with intercropping of black oat, common vetch, and forage turnip is important to obtain accurate results and reliable inferences about the treatments under evaluation. Proper sizing optimizes the resources involved in the research, such as manpower, time, financial resources, and experimental area. These dimensions are common questions from researchers involved in this area of knowledge.

The methodologies of Smith (1938) and Hatheway (1961) can be applied to uniformity trials data (trials without treatments) in order to calculate the optimal plot size according to the experimental design, treatment numbers, repetition numbers, and experimental precision levels. These methodologies have been used in sunflower (Sousa et al., 2016), in banana (Donato et al., 2018), in forage palm (Guimarães et al., 2020), and in species with potential for ground cover, such as: velvet bean (Cargnelutti Filho et al., 2014b); forage turnip (Cargnelutti Filho et al., 2014c); flax (Cargnelutti Filho et al., 2018); and black oat with common vetch (Cargnelutti Filho et al., 2020).

Plot size has been investigated in single cropping of black oat (Cargnelutti Filho et al., 2014a), common vetch (Cargnelutti Filho et al., 2017), and forage turnip (Cargnelutti Filho et al., 2011, 2016) by averages of the maximum curvature method of the coefficient of variation model (Paranaíba et al., 2009). It is assumed that the O+V+T consortium, commonly used with ground cover plants, may generate distinct experimental planning patterns. Thus, the objective of this study was to determine the optimal plot size for evaluating the fresh matter of black oat (*Avena strigosa* Schreb), common vetch (*Vicia sativa* L.), and forage turnip (*Raphanus sativus* L.), grown in intercrop, in scenarios formed by combinations of treatments numbers, repetitions numbers, and experimental precision levels.

## Materials and Methods

Six uniformity trials (blank experiments) with the intercrop of black oat (*Avena strigosa* Schreb), cultivar Embrapa 139, common vetch (*Vicia sativa* L.), and forage turnip (*Raphanus sativus* L.), cultivar IPR-116 (O+V+T), were conducted in an experimental area located at 29° 42' S, 53° 49' W and 95 m altitude. At this site, the climate is Cfa humid subtropical, according to Köppen classification, with hot summers and no dry season (Alvares et al., 2013) and the soil is Arenic Dystrophic Red Argissolo (Santos et al., 2018).

On June 2, 2020, the base fertilization was performed, with 20 kg ha<sup>-1</sup> of N, 80 kg ha<sup>-1</sup> of P<sub>2</sub>O<sub>5</sub> and 80 kg ha<sup>-1</sup> of K<sub>2</sub>O (only N-P-K, of the 05-20-20 formulation) and the sowing of the consortium O+V+T, by broadcast, with the following sowing densities: black oat (60 kg ha<sup>-1</sup>) + common vetch (30 kg ha<sup>-1</sup>) + forage turnip (15 kg ha<sup>-1</sup>).

Three trials were evaluated at 84 days after sowing (August 25, 2020) and the other three trials at 119 days after sowing (September 29, 2020). In each uniformity trial, the central area of dimension 6 × 6 m (36 m<sup>2</sup>) was divided into 36 basic experimental units (BEU) of 1 × 1 m (1 m<sup>2</sup>), forming a matrix of six rows and six columns. In each BEU, the plants were cut, close to the soil surface, and the fresh matter (FM) was weighed, in g m<sup>-2</sup>. Weighing was performed immediately after cutting, in order to minimize possible variations in plant moisture.

For each uniformity trial, with the FM data from the 36 BEU, plots with X<sub>R</sub> BEU adjacent in the row and X<sub>C</sub> BEU adjacent in the column were planned. The plots with different dimensions and/or shapes were planned as (X = X<sub>R</sub> × X<sub>C</sub>), i.e., (1 × 1), (1 × 2), (1 × 3), (1 × 6), (2 × 1), (2 × 2), (2 × 3), (2 × 6), (3 × 1), (3 × 2), (3 × 3), (3 × 6), (6 × 1), (6 × 2) and (6 × 3). The abbreviations X<sub>R</sub>, X<sub>C</sub> and X, stand for number of adjacent BEU in the row, number of adjacent BEU in the column and plot size in number of BEU, respectively.

For each plot size (X) were determined: n - number of plots with X BEU size (n=36/X); M<sub>(X)</sub> - average of the plots with X BEU of size; V<sub>(X)</sub> - variance among plots of X BEU of size; CV<sub>(X)</sub> - coefficient of variation (in %) between plots of X BEU size; and VU<sub>(X)</sub> - variance per BEU among the X BEU plots of size [VU<sub>(X)</sub>=V<sub>(X)</sub>/X<sup>2</sup>].

The parameters V1 (variance per BEU among the plots in a BEU of size) and b (index of heterogeneity) and the coefficient of determination ( $r^2$ ) of the function  $VU_{(x)} = V1/X^b$  of Smith (1938) were estimated. These parameters were estimated by logarithmic transformation and linearization of the function  $VU_{(x)} = V1/X^b$ , i.e.,  $\log VU_{(x)} = \log V1 - b \log X$ , whose estimation was weighted by the degrees of freedom (DF=n-1), associated with each plot dimension, as applied by Sousa et al. (2016). The observed values of the dependent [ $VU_{(x)}$ ] and independent (X) variables and the function  $VU_{(x)} = V1/X^b$  (Smith, 1938) were plotted graphically.

Experimental plans were simulated for the scenarios formed by combinations of i treatments (i = 5, 10, 15, 20, and 25), r repetitions (r = 3, 4, 5, 6, 7, 8, 9, and 10) and d differences between treatment averages to be detected at the 0.05 level of significance, expressed as a percentage of the overall average of the experiment, that is, in levels of precision [d = 4% (highest precision), 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, and 20% (lowest precision)]. For each experimental plan, optimal plot size ( $X_o$ ) was calculated, in number of BEU, using the expression

$$X_o = \sqrt[3]{2(t_1 + t_2)^2 CV^2 / rd^2} \quad (\text{Hatheway, 1961}).$$

In this expression b is the estimate of the heterogeneity index (in this study, the average of b from the six uniformity trials was taken);  $t_1$  is the critical value of Student t distribution for the significance level of the test (type I error) of  $\alpha = 5\%$  (two-sided test at 5%), with DF degrees of freedom;  $t_2$  is the critical value of Student t distribution, corresponding to 2(1-P) (two-sided test), where P is the probability of obtaining a significant result, that is, the power of the test (P = 0.80, in this study), with DF degrees of freedom; CV is the estimate of the coefficient of variation among plots of a BEU size (in this study, the average CV of the six uniformity trials was taken), in percent; r is the number of repetitions; and d is the difference between treatment averages to be detected at the 0.05 level of significance, expressed as a percentage of the overall average of the experiment (precision). The

degrees of freedom (DF) for obtaining the critical values (tabulated) of Student t-distribution were obtained by the expression  $DF = (i)(r-1)$ , where i is the number of treatments and r is the number of repetitions. The values of  $t_1$  and  $t_2$ , in this study, were obtained with the Microsoft Office Excel® application, by averages of the functions  $t_1 = \text{INVNT}(0.05; DF)$  and  $t_2 = \text{INVNT}(0.40; DF)$ , respectively.

Average comparisons of FM, CV and b estimates between the two evaluation seasons (n=3 uniformity trials per season) were performed using Student t-test (two-sided) for independent samples at 5% significance level. The remaining statistical analyses were performed with the aid of the applications SISVAR (Ferreira, 2019) and Microsoft Office Excel®.

## Results and Discussion

In the six uniformity trials with the intercrop of black oat (*Avena strigosa* Schreb), cultivar Embrapa 139, common vetch (*Vicia sativa* L.), and forage turnip (*Raphanus sativus* L.), cultivar IPR-116 (O+V+T), the fresh matter (FM) ranged between 2656 and 3367 g m<sup>-2</sup>, or 26.56 and 33.67 Mg ha<sup>-1</sup>, respectively (Table 1). The average FM, of the trials evaluated at 84 and 119 days after sowing (DAS) were 2992 and 3087 g m<sup>-2</sup>, respectively, and did not differ (t = -0.39707; p-value = 0.7116, with 4 degrees of freedom) and the overall average of the six trials was 3040 g m<sup>-2</sup>.

The coefficient of variation (CV) of FM, obtained among the 36 BEU in each of the six uniformity trials, ranged from 25.22 to 30.68%, with an average of 28.67% (Table 1). The average CVs of the three trials for each evaluation season were 27.74 and 29.60% for the evaluations at 84 and 119 DAS, respectively, and by Student t-test (two-sided), for independent samples, at 5% significance, they did not differ (t = -1.18258; p-value = 0.3025). This suggests that experiments with the consortium of black oat, common vetch, and forage turnip have similar experimental accuracy. Additionally, it can be inferred that using the average CV of the six trials (CV = 28.67%), in Hatheway (1961) methodology, is adequate to represent the evaluation seasons.

**Table 1.** Fresh matter (FM), coefficient of variation (CV) and Smith (1938) index of heterogeneity (b) in three uniformity trials (repetitions) of each evaluation season s of the consortium of black oat, common vetch and forage turnip. Student t-test value and respective p-value for FM, CV and b.

Evaluation seasons	Trials <sup>(1)</sup>	FM (g m <sup>-2</sup> ) <sup>(2)</sup>	CV (%) <sup>(2)</sup>	b <sup>(2)</sup>
84 days after sowing	1	3367	28.77	1.0985
	2	2953	29.24	0.9402
	3	2656	25.22	1.0046
	Average	2992	27.74	1.0144
119 days after sowing	1	2860	30.68	0.6135
	2	3134	27.74	1.3530
	3	3267	30.39	1.0666
	Average	3087	29.60	1.0111
	Overall average	3040	28.67	1.0127
Student t-test value		-0.39707	-1.18258	0.01524
p-value		0.7116	0.3025	0.9886

<sup>(1)</sup> Each uniformity trial of size 6 × 6 m (36 m<sup>2</sup>) was divided into 36 BEU of 1 × 1 m (1 m<sup>2</sup>), forming a matrix of six rows and six columns.

<sup>(2)</sup> The averages of the evaluation seasons do not differ by Student t-test (two-sided), with 4 degrees of freedom (p > 0.05).

Smith (1938) heterogeneity index (b), among the six uniformity trials, ranged from 0.6135 to 1.3530, with an average of 1.0127 (Table 1). The averages of b, of the three trials of each evaluation season, were 1.0144 and 1.0111, for the evaluations at 84 and 119 DAS, respectively, and by Student t-test (two-sided), for independent samples, at 5% significance, they did not differ ( $t = 0.01524$ ;  $p\text{-value} = 0.9886$ ). Then, one can use the average of b from the six trials ( $b = 1.0127$ ) in Hatheway (1961) methodology, to represent the two evaluation seasons. Values of b close to unity indicate high heterogeneity or low correlation between adjacent plots. According to Lin & Binns (1986), when  $b > 0.7$ , it is recommended to increase the plot size, when  $b < 0.2$ , one should increase the number of repetitions, and in cases of  $0.2 \leq b \leq 0.7$  it is appropriate to investigate the best combination of plot size and number of repetitions. So, it can be inferred that in experiments with the

intercrop of black oat, common vetch, and forage turnip, one should prioritize the use of larger plots.

In all six uniformity trials, there was a decrease in variance per BEU among plots  $[VU_{(x)}]$ , which indicates improvement in experimental precision with increasing planned plot size (X) (Figure 1). The decreases were steep up to plots four BEU in size ( $4 \text{ m}^2$ ), intermediate between four and 15 BEU, and a stabilizing trend with plots larger than 15 BEU ( $15 \text{ m}^2$ ). Similar pattern to this was observed in velvet bean (Cargnelutti Filho et al., 2014b); forage turnip (Cargnelutti Filho et al., 2014c); flax (Cargnelutti Filho et al., 2018); and black oat with common vetch (Cargnelutti Filho et al., 2020). So, to evaluate the fresh matter of the consortium of black oat, common vetch, and forage turnip, a plot size of up to  $15 \text{ m}^2$  is indicated.

In Hatheway (1961) methodology, from fixed values of the coefficient of variation ( $CV = 28.67\%$ ) and Smith (1938)

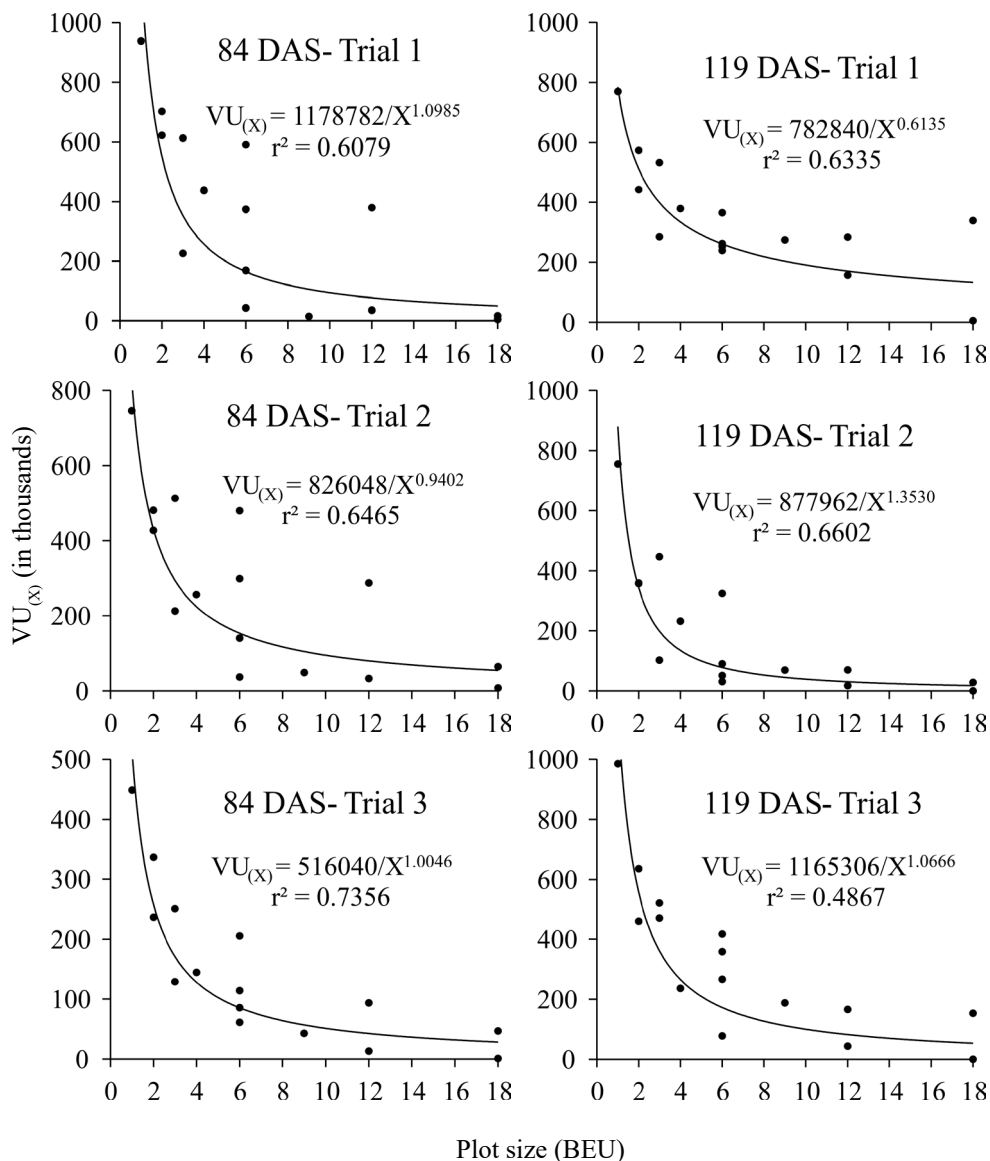


Figure 1. Relationship between the variance per basic experimental unit (BEU) between X BEU plot sizes  $[VU_{(x)}=V_{(x)}/X^2]$  and the planned plot size (X), in BEU, and the parameter estimates of the function  $VU_{(x)}=V1/X^b$  of Smith (1938). Fresh matter data obtained in uniformity trials with the consortium of black oat, common vetch, and forage turnip, evaluated at 84 and 119 days after sowing (DAS), with 36 BEU of  $1 \text{ m}^2$ .

index of heterogeneity ( $b = 1.0127$ ), it is possible to determine distinct optimal plot sizes ( $X_0$ ), as a function of the number of treatments ( $i$ ), the number of repetitions ( $r$ ), and precision ( $d$ ) (Table 2). Therefore, besides the indicated size of 15 m<sup>2</sup>, the researcher can investigate within his availability of experimental area, number of treatments to be evaluated and desired precision, which combination of plot size and number of repetitions is the most appropriate. In crops, such as: sunflower (Sousa et al., 2016); banana (Donato et al., 2018); forage palm (Guimarães et al., 2020); velvet bean (Cargnelutti Filho et al., 2014b); forage turnip (Cargnelutti Filho et al., 2014c); flax (Cargnelutti Filho et al., 2018); and black oat with common vetch (Cargnelutti Filho et al., 2020), the application of Smith (1938) and Hatheway (1961) methodologies has generated important subsidies for planning the experiments.

For fixed values of  $i$  and  $r$ ,  $X_0$  increased with increasing precision ( $d$ ) (Table 2). For example, to evaluate FM in an experiment conducted in completely randomized design (CRD), with five treatments and three repetitions, aiming that in 80% of the experiments (power = 0.80) differences between treatments of  $d = 20\%$  of the overall average of the experiment (lower precision) will be detected at the 5% significance level, the plot size should be 12.8 BEU (12.8 m<sup>2</sup>) (Table 2). At the other extreme, i.e. plots of 307.5 m<sup>2</sup> would make it possible to improve accuracy and obtain  $d = 4\%$ . However, conducting an experiment with a 307.5 m<sup>2</sup> plot requires a larger experimental area and can make the experiment difficult to run. Therefore, in practice, high experimental accuracies (low percentages of  $d$ ) are difficult to achieve, due to the need for high plot size, as already pointed out by Cargnelutti Filho et al. (2014b);

**Table 2.** Optimal plot size, in m<sup>2</sup>, in combinations of  $i$  treatments,  $r$  repetitions, and  $d$  precision levels (%), for fresh matter of the consortium of black oat, common vetch, and forage turnip (CV = 28.67%; heterogeneity index  $b = 1.0127$ ).

i	r	d (%)																	
		4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	
5	3	307.5	197.9	138.0	101.8	78.2	62.0	50.3	41.7	35.1	30.0	25.9	22.6	19.9	17.7	15.8	14.2	12.8	
	4	215.6	138.8	96.8	71.4	54.8	43.5	35.3	29.2	24.6	21.0	18.2	15.8	14.0	12.4	11.1	9.9	9.0	
	5	167.1	107.6	75.0	55.3	42.5	33.7	27.4	22.7	19.1	16.3	14.1	12.3	10.8	9.6	8.6	7.7	7.0	
	6	136.8	88.0	61.4	45.3	34.8	27.6	22.4	18.6	15.6	13.3	11.5	10.1	8.9	7.9	7.0	6.3	5.7	
	7	115.9	74.6	52.0	38.4	29.5	23.4	19.0	15.7	13.2	11.3	9.8	8.5	7.5	6.7	5.9	5.3	4.8	
	8	100.6	64.8	45.2	33.3	25.6	20.3	16.5	13.6	11.5	9.8	8.5	7.4	6.5	5.8	5.2	4.6	4.2	
	9	88.9	57.2	39.9	29.5	22.6	17.9	14.6	12.1	10.2	8.7	7.5	6.5	5.8	5.1	4.6	4.1	3.7	
	10	79.7	51.3	35.8	26.4	20.3	16.1	13.0	10.8	9.1	7.8	6.7	5.9	5.2	4.6	4.1	3.7	3.3	
	10	3	276.7	178.1	124.3	91.6	70.4	55.8	45.3	37.5	31.6	27.0	23.3	20.3	17.9	15.9	14.2	12.8	11.5
		4	201.4	129.6	90.4	66.7	51.2	40.6	33.0	27.3	23.0	19.6	17.0	14.8	13.0	11.6	10.3	9.3	8.4
5		158.9	102.3	71.3	52.6	40.4	32.0	26.0	21.6	18.1	15.5	13.4	11.7	10.3	9.1	8.1	7.3	6.6	
6		131.4	84.6	59.0	43.5	33.4	26.5	21.5	17.8	15.0	12.8	11.1	9.7	8.5	7.5	6.7	6.1	5.5	
7		112.1	72.2	50.3	37.1	28.5	22.6	18.4	15.2	12.8	10.9	9.4	8.2	7.3	6.4	5.7	5.2	4.7	
8		97.8	63.0	43.9	32.4	24.9	19.7	16.0	13.3	11.2	9.5	8.2	7.2	6.3	5.6	5.0	4.5	4.1	
9		86.8	55.8	39.0	28.7	22.1	17.5	14.2	11.8	9.9	8.5	7.3	6.4	5.6	5.0	4.4	4.0	3.6	
10		78.0	50.2	35.0	25.8	19.8	15.7	12.8	10.6	8.9	7.6	6.6	5.7	5.0	4.5	4.0	3.6	3.2	
15		3	267.6	172.2	120.1	88.6	68.1	53.9	43.8	36.3	30.6	26.1	22.5	19.7	17.3	15.4	13.7	12.3	11.1
		4	197.0	126.8	88.4	65.2	50.1	39.7	32.3	26.7	22.5	19.2	16.6	14.5	12.7	11.3	10.1	9.1	8.2
	5	156.3	100.6	70.2	51.8	39.8	31.5	25.6	21.2	17.9	15.2	13.2	11.5	10.1	9.0	8.0	7.2	6.5	
	6	129.7	83.5	58.2	43.0	33.0	26.1	21.2	17.6	14.8	12.6	10.9	9.5	8.4	7.4	6.7	6.0	5.4	
	7	110.9	71.4	49.8	36.7	28.2	22.4	18.2	15.0	12.7	10.8	9.3	8.2	7.2	6.4	5.7	5.1	4.6	
	8	96.9	62.4	43.5	32.1	24.7	19.5	15.9	13.1	11.1	9.5	8.2	7.1	6.3	5.6	5.0	4.5	4.0	
	9	86.1	55.4	38.6	28.5	21.9	17.4	14.1	11.7	9.8	8.4	7.3	6.3	5.6	4.9	4.4	4.0	3.6	
	10	77.4	49.8	34.8	25.6	19.7	15.6	12.7	10.5	8.8	7.6	6.5	5.7	5.0	4.4	4.0	3.6	3.2	
	20	3	263.2	169.4	118.2	87.1	66.9	53.0	43.1	35.7	30.1	25.7	22.2	19.3	17.0	15.1	13.5	12.1	11.0
		4	194.8	125.4	87.5	64.5	49.6	39.3	31.9	26.4	22.3	19.0	16.4	14.3	12.6	11.2	10.0	9.0	8.1
5		155.0	99.8	69.6	51.3	39.4	31.3	25.4	21.0	17.7	15.1	13.1	11.4	10.0	8.9	8.0	7.1	6.5	
6		128.9	82.9	57.9	42.7	32.8	26.0	21.1	17.5	14.7	12.6	10.9	9.5	8.3	7.4	6.6	5.9	5.4	
7		110.3	71.0	49.5	36.5	28.1	22.2	18.1	15.0	12.6	10.8	9.3	8.1	7.1	6.3	5.7	5.1	4.6	
8		96.5	62.1	43.3	31.9	24.5	19.4	15.8	13.1	11.0	9.4	8.1	7.1	6.2	5.5	4.9	4.4	4.0	
9		85.7	55.2	38.5	28.4	21.8	17.3	14.0	11.6	9.8	8.4	7.2	6.3	5.5	4.9	4.4	4.0	3.6	
10		77.1	49.7	34.6	25.5	19.6	15.6	12.6	10.5	8.8	7.5	6.5	5.7	5.0	4.4	4.0	3.6	3.2	
25		3	260.6	167.7	117.0	86.3	66.3	52.5	42.7	35.3	29.8	25.4	21.9	19.2	16.9	15.0	13.4	12.0	10.9
		4	193.6	124.6	86.9	64.1	49.2	39.0	31.7	26.3	22.1	18.9	16.3	14.2	12.5	11.1	9.9	8.9	8.1
	5	154.3	99.3	69.3	51.1	39.2	31.1	25.3	20.9	17.6	15.0	13.0	11.3	10.0	8.9	7.9	7.1	6.4	
	6	128.4	82.6	57.6	42.5	32.7	25.9	21.0	17.4	14.7	12.5	10.8	9.4	8.3	7.4	6.6	5.9	5.3	
	7	110.0	70.8	49.4	36.4	28.0	22.2	18.0	14.9	12.6	10.7	9.3	8.1	7.1	6.3	5.6	5.1	4.6	
	8	96.2	61.9	43.2	31.9	24.5	19.4	15.7	13.0	11.0	9.4	8.1	7.1	6.2	5.5	4.9	4.4	4.0	
	9	85.5	55.0	38.4	28.3	21.8	17.2	14.0	11.6	9.8	8.3	7.2	6.3	5.5	4.9	4.4	3.9	3.6	
	10	77.0	49.5	34.6	25.5	19.6	15.5	12.6	10.4	8.8	7.5	6.5	5.7	5.0	4.4	3.9	3.5	3.2	

2014c; 2018; 2020). Additionally, for fixed values of *i* and *d*,  $X_o$  decreased with the increase of *r*, and for fixed values of *r* and *d*,  $X_o$  decreased with the increase of *i*. Similar pattern has been found by Cargnelutti Filho et al. (2014b; 2014c; 2018; 2020).

The information from this study enables investigations into 680 scenarios formed by combinations of *i* treatments (*i* = 5, 10, 15, 20, and 25), *r* repetitions (*r* = 3, 4, 5, 6, 7, 8, 9, and 10) and *d* differences between treatment averages to be detected at the 5% significance level (*d* = 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, and 20%). For example, if the researcher wants to evaluate the FM of five treatments, in CRD, and wants precision (*d*) of 10%, among the various options, he could use plots of 50.3 BEU (50.3 m<sup>2</sup>) and three repetitions, 35.3 BEU (35.3 m<sup>2</sup>) and four repetitions, 27.4 BEU (27.4 m<sup>2</sup>) and five repetitions, 22.4 BEU (22.4 m<sup>2</sup>) and six

repetitions, 19.0 BEU (19.0 m<sup>2</sup>) and seven repetitions, 16.5 BEU (16.5 m<sup>2</sup>) and eight repetitions, 14.6 BEU (14.6 m<sup>2</sup>) and nine repetitions and 13.0 BEU (13.0 m<sup>2</sup>) and ten repetitions (Table 2). In this situation, the required experimental area is 755, 706, 684, 672, 664, 659, 655, and 652 m<sup>2</sup>, respectively (Table 3).

Other scenarios can be simulated using the expression

$$X_o = \sqrt[3]{2(t_1 + t_2)^2 CV^2 / rd^2} \text{ (Hatheway, 1961).}$$

For example, to evaluate the FM of 13 treatments, with six repetitions and with *d*=12%, in CRD, one has:  $b=1.0127$ ;  $DF=(13)(6-1)=65$ ;  $t_1=INV(0.05;65)=1.997137887$ ;  $t_2=INV(0.40;65)=0.847186101$ ;  $CV=28.67\%$ ;  $r=6$ ;  $d=12\%$ . Then,

**Table 3.** Experiment size, in m<sup>2</sup>, in combinations of *i* treatments, *r* repetitions, and *d* precision levels (%), for fresh matter of the consortium of black oat, common vetch, and forage turnip (CV = 28.67%; heterogeneity index *b* = 1.0127).

i	r	d (%)																	
		4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	
5	3	4612	2968	2071	1527	1173	930	755	626	527	450	388	339	298	265	237	213	192	
	4	4312	2775	1936	1428	1097	869	706	585	492	420	363	317	279	248	221	199	180	
	5	4178	2689	1876	1383	1063	842	684	567	477	407	352	307	270	240	214	193	174	
	6	4103	2641	1842	1359	1044	827	672	557	469	400	346	302	266	236	210	189	171	
	7	4056	2611	1821	1343	1032	818	664	550	463	396	342	298	262	233	208	187	169	
	8	4025	2590	1807	1333	1024	811	659	546	460	392	339	296	260	231	206	185	168	
	9	4002	2576	1797	1325	1018	807	655	543	457	390	337	294	259	230	205	184	167	
	10	3985	2565	1789	1320	1014	803	652	541	455	389	336	293	258	229	204	184	166	
	10	3	8302	5343	3728	2749	2112	1674	1359	1126	948	810	699	610	537	477	426	383	346
		4	8056	5185	3617	2668	2049	1624	1319	1093	920	786	679	592	521	462	413	371	336
5		7945	5114	3567	2631	2021	1602	1301	1078	907	775	669	584	514	456	407	366	331	
6		7885	5075	3540	2611	2006	1590	1291	1069	901	769	664	580	510	453	404	363	328	
7		7849	5051	3524	2599	1997	1582	1285	1065	896	765	661	577	508	451	402	362	327	
8		7825	5036	3513	2591	1991	1577	1281	1061	894	763	659	575	506	449	401	361	326	
9		7809	5026	3506	2586	1987	1574	1279	1059	892	762	658	574	505	448	400	360	325	
10		7799	5019	3501	2582	1984	1572	1277	1058	891	760	657	573	505	448	400	359	325	
15		3	12040	7749	5406	3987	3063	2427	1971	1633	1375	1174	1014	885	779	691	617	555	501
		4	11819	7607	5307	3914	3007	2383	1935	1603	1350	1153	996	869	765	679	606	545	492
	5	11724	7545	5264	3882	2982	2363	1919	1590	1339	1143	988	862	759	673	601	540	488	
	6	11674	7513	5241	3866	2970	2353	1911	1583	1333	1138	983	858	755	670	599	538	486	
	7	11646	7495	5229	3856	2962	2348	1907	1580	1330	1136	981	856	754	669	597	537	485	
	8	11629	7484	5221	3851	2958	2344	1904	1577	1328	1134	980	855	753	668	596	536	484	
	9	11619	7478	5217	3848	2956	2342	1902	1576	1327	1133	979	854	752	667	596	536	484	
	10	11614	7475	5214	3846	2954	2341	1901	1575	1326	1133	978	854	752	667	596	535	484	
	20	3	15789	10162	7089	5229	4017	3183	2585	2141	1803	1540	1330	1161	1022	906	810	728	658
		4	15588	10032	6999	5162	3965	3142	2552	2114	1780	1520	1313	1146	1009	895	799	718	649
5		15504	9978	6961	5134	3944	3125	2538	2103	1771	1512	1306	1140	1003	890	795	715	646	
6		15464	9953	6943	5121	3934	3117	2532	2097	1766	1508	1303	1137	1001	888	793	713	644	
7		15444	9940	6934	5114	3929	3113	2528	2095	1764	1506	1301	1135	999	887	792	712	643	
8		15434	9933	6930	5111	3926	3111	2527	2093	1763	1505	1300	1135	999	886	791	711	643	
9		15430	9931	6928	5110	3925	3111	2526	2093	1762	1505	1300	1134	998	886	791	711	643	
10		15430	9930	6928	5109	3925	3110	2526	2093	1762	1505	1300	1134	998	886	791	711	643	
25		3	19542	12577	8774	6471	4971	3940	3199	2650	2232	1906	1646	1437	1265	1122	1002	901	814
		4	19358	12458	8691	6410	4924	3902	3169	2625	2211	1888	1631	1423	1253	1111	993	892	806
	5	19286	12412	8659	6386	4906	3888	3157	2616	2203	1881	1625	1418	1248	1107	989	889	803	
	6	19255	12392	8645	6376	4898	3882	3152	2612	2199	1878	1622	1415	1246	1105	987	887	802	
	7	19242	12384	8640	6372	4895	3879	3150	2610	2198	1876	1621	1414	1245	1105	987	887	801	
	8	19239	12382	8638	6371	4894	3878	3150	2609	2197	1876	1621	1414	1245	1105	987	887	801	
	9	19241	12383	8639	6372	4895	3879	3150	2610	2198	1876	1621	1414	1245	1105	987	887	801	
	10	19246	12386	8641	6373	4896	3880	3151	2610	2198	1877	1621	1415	1245	1105	987	887	802	

$$X_o = 1.0127 \sqrt{2(1.997137887 + 0.847186101)^2 28.67^2 / 6 \times 12^2} = 14.87 \text{ BEU.}$$

If the researcher wants to conduct the experiment in a randomized complete block design, he has:  $b=1.0127$ ;  $DF=(13-1)(6-1)=60$ ;  $t_1=INVT(0.05;60)=2.000297804$ ;  $t_2=INVT(0.40;60)=0.847653006$ ;  $CV=28.67\%$ ;  $r=6$ ;  $d=12\%$ . Then,

$$X_o = 1.0127 \sqrt{2(2.000297804 + 0.847653006)^2 28.67^2 / 6 \times 12^2} = 14.91 \text{ BEU.}$$

Therefore, using the criterion of rounding up to the nearest whole number to ensure the desired accuracy, for these examples, the plot size would be 15 m<sup>2</sup> and the experimental area 1170 m<sup>2</sup>.

The results of this study serve as a reference for defining plot size and number of repetitions in experiments to evaluate the fresh matter of black oat, common vetch, and forage turnip, grown as intercrops. It is recommended to use 15 m<sup>2</sup> plots, due to practical feasibility in the field and the stabilization of accuracy at this size.

This size of 15 m<sup>2</sup> is relatively larger than those established to evaluate the fresh matter of the single crops of black oat (Cargnelutti Filho et al., 2014a), common vetch (Cargnelutti Filho et al., 2017), and forage turnip (Cargnelutti Filho et al., 2011, 2016) and the consortium of black oat and common vetch (Cargnelutti Filho et al., 2020) which were 4.14, 4.52, 1.20, and 10 m<sup>2</sup>, respectively. Additionally this size of 15 m<sup>2</sup> is relatively smaller than used in experiments with the O+V+T consortium along with other ground cover species by Rigon et al. (2011), Ziech et al. (2015), Wolschick et al. (2016), Michelon et al. (2019), and Haskel et al. (2020), which ranged between 18 and 48 m<sup>2</sup>.

## Conclusions

From Smith (1938) index of heterogeneity and Hatheways (1961) method, it was concluded that in experiments to evaluate the fresh matter of the consortium of black oat, common vetch, and forage turnip, with 5 to 25 treatments and with six repetitions, plots of 15 m<sup>2</sup> of useful area are sufficient for differences between treatments of 12% of the overall average of the experiment to show differences at the 0.05 level of significance.

## Acknowledgments

To the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq - Processes 146258/2019-3, 158165/2018-7, and 304652/2017-2), the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES), and the Fundação de Amparo à Pesquisa do Estado do Rio Grande do Sul (FAPERGS) for granting scholarships to the authors. To the scholarship students and volunteers for helping in data collection.

## Compliance with Ethical Standards

**Author contributions:** Conceptualization: ACF; Data curation: ACF, DLS, MVL, LFO, VMO; Formal analysis: ACF; Funding acquisition: ACF; Investigation: ACF, DLS, MVL, LFO, VMO; Methodology: ACF; Project administration: ACF; Resources: ACF; Software: ACF; Supervision: ACF; Validation: ACF; Visualization: ACF; Writing – original draft: ACF; Writing – review & editing: ACF, DLS, MVL, LFO, VMO.

**Conflict of interest:** The authors declare no conflict of interest.

**Financing source:** To the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq - Processes 146258/2019-3, 158165/2018-7, and 304652/2017-2), the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES – Finance Code 001), and the Fundação de Amparo à Pesquisa do Estado do Rio Grande do Sul (FAPERGS).

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