Optimal plot size for experiments with black oats and the common vetch

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ABSTRACT: The aim of this study was to determine the optimal plot size for evaluating the fresh weight of black oats (Avena strigosa Schreb) and the common vetch (Vicia sativa L.) in scenarios comprising combinations of the number of treatments, number of replications and levels of precision. Fifteen uniformity trials were conducted with single-crop and intercropped black oats and vetch. Fresh weight was evaluated in 540 basic experimental units (BEU), each of 1 m × 1 m (36 BEU per trial). The Smith index of soil heterogeneity (1938) was estimated. Plot size was determined using the HATHEWAY method (1961), in scenarios comprising combinations of i treatments (i = 5, 10, 15 and 20), r replications (r = 3, 4, 5, 6, 7 and 8) and d levels of precision (d = 2%, 4%, 6%, 8%, 10%, 12%, 14%, 16%, 18% and 20%). To evaluate the fresh weight of monocropped or intercropped black oats and vetch in a completely randomized or randomized complete block design, with from 5 to 20 treatments and five replications, plots of 10 m² are sufficient to identify, at a probability of 0.05, significant differences between treatments of 10% of the overall mean value of the experiment.

Key words: Avena strigosa, cover crop, experimental design, uniformity trial, Vicia sativa.
In planning such experiments, aspects related to plot size, number of treatments and the number of replications are important; these should be adequately sized, so that the results are reliable and inferences concerning the treatments under evaluation are valid. Proper sizing optimizes resources involved in the research, such as labour, time, financial resources and the experimental area. Sizing is a common question of researchers involved in this area of knowledge.

In uniformity trials or blank experiments, it is possible to divide the experimental area into basic experimental units (BEU) of the smallest possible size that is compatible with the evaluations (STORCK et al., 2016). From the data collected in these BEU, the coefficient of variation (CV) between the BEU, and the Smith index of soil heterogeneity (b) (1938) can be estimated. Estimates of CV and b can be used with the HATHEWAY methodology (1961) to calculate the optimal plot size (Xo) according to the experimental design, number of treatments, number of replications and experimental precision. After establishing the experimental design and number of treatments, the researcher can then choose the best combination of plot size, number of replications and level of experimental precision. This approach has been used with the common bean (MAYOR-DURÁN et al., 2012), and for evaluating fresh weight in potential ground cover species, such as the velvet bean (CARGNELUTTI FILHO et al., 2014b) and the forage turnip (CARNELUTTI FILHO et al., 2014c). Important results were generated in those studies, which can be applied when planning experiments with these species grown as single crops.

Although, plot size was investigated using the maximum curvature of the model of the coefficient of variation (PARANAIBA et al., 2009) in the single cultivation of black oats (CARGNELUTTI FILHO et al., 2014a) and vetch (CARGNELUTTI FILHO et al., 2017), it is assumed that intercropping, commonly used with ground cover plants, can generate distinct patterns of experimental design, and that the use of this approach can combine information for planning experiments with these two ground cover species.

Thus, the aim of this study was to determine the optimal plot size for evaluating the fresh weight of black oats (Avena strigosa Schreb) and the common vetch (Vicia sativa L.) in scenarios comprising combinations of the number of treatments, number of replications and levels of precision.

MATERIALS AND METHODS

Fifteen uniformity trials were conducted with black oats (Avena strigosa Schreb) ‘Embrapa 139’, and the common vetch (Vicia sativa L.) ‘SS Ametista’, in an experimental area located at 29º42’ S and 53º49’ W, at an altitude of 95 m. According to the Köppen classification, the climate in the area is type Cfa, humid subtropical with hot summers and no dry season (ALVARES et al., 2013); the soil is a Dystrophic Red Arenic Argisol (SANTOS et al., 2013). A physical and chemical analysis of the soil at a depth of 0-20 cm revealed: pHH 2O 1:1:5.8, Ca: 5.7 cmol-c dm-3, Mg: 2.4 cmol-c dm-3, Al: 0.0 cmol-c dm-3, H+Al: 3.5 cmol-c dm-3, SMP index: 6.2, organic matter: 2.4%, clay content: 29.0%, S: 13.3 mg dm-3, P (Mehlich): 25.7 mg dm-3, K: 0.696 cmol-c dm-3, CECpH7: 12.4 cmol-c dm-3, Cu: 1.28 mg dm-3, Zn: 0.753 mg dm-3, and B: 0.1 mg dm-3. These results were used to define the fertilization (CQFS, 2016).

The uniformity trials comprised combinations of sowing densities of black oats (Avena strigosa Schreb) ‘Embrapa 139’ (BO) and vetch (Vicia sativa L.) ‘SS Ametista’ (VE). Three trials were conducted for each of the following combinations, with the respective sowing densities shown in parentheses: 100% BO (80 kg ha-1), 75% BO (60 kg ha-1) + 25% VE (15 kg ha-1), 50% BO (40 kg ha-1) + 50% VE (30 kg ha-1), 25% BO (20 kg ha-1) + 75% VE (45 kg ha-1), and 100% VE (60 kg ha-1). On 16 June 2017, base fertilizer was applied using 20 kg ha-1 N, 80 kg ha-1 P2O5 and 80 kg ha-1 K2O (as 05-20-20 formulation NPK) with broadcast seeding.

For each uniformity trial, the central area of 6 m × 6 m (36 m²) was divided into 36 basic experimental units (BEU) of 1 m × 1 m (1 m²), to form a matrix of six rows and six columns. At flowering, the plants in each BEU were cut close to the ground and the fresh weight (FW) was determined, in g m². Weighing was carried out immediately after cutting to minimise possible variations in plant moisture.

In the period between sowing (16/06/2017) and evaluating the FW (20/09/2017), daily data were recorded for the minimum (Tmin) and maximum (Tmax) air temperature, in °C, and rainfall, in mm, at the Automatic Weather Station of the Federal University of Santa Maria, located 40 m from the experimental area. The daily mean air temperature (Tm) was calculated in °C, using the expression: Tm = (Tmin + Tmax)/2.

With the FW data of the 36 BEU in each uniformity trial, plots were marked out consisting of Xg, adjacent BEU in a row and Xc, adjacent BEU in a column. Plots of different sizes and/or shapes were marked out as (X = Xg × Xc), 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
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RESULTS AND DISCUSSION

In the 15 uniformity trials comprising combinations of the sowing densities of black oats (Avena strigosa Schreb) ‘Embrapa 139’ (BO) and vetch (Vicia sativa L.) ‘SS Ametista’ (VE), there was an increase in the mean value of the plots [M(x)] and the variance between plots [V(x)], with a decrease in the coefficient of variation [CV(x)] and the variance per BEU between plots [VU(x)], for increases in the planned size of the plot (X) (Table 1 and Figure 1). These results indicate an improvement in experimental precision (a decrease in CV and VU), with the increase in plot size. Therefore, although, it is possible to evaluate fresh weight (FW) in plots of 1 m², it is important to evaluate the precision in larger plots, i.e. it is essential to design the experiment with the optimal plot size to ensure a proper discrimination of the treatments under evaluation and the reliability of the inferences. In addition, smaller sizes may not represent plant development for either single crops or intercrops; whereas larger sizes would make it possible to evaluate the plants in the central area of the plot (working area) and disregard the borders, thereby reducing interference from plants in adjacent plots, i.e. inter-plot competition.

The mean value of FW in the three trials of each combination was 24055, 24005, 23189, 23579 and 21252 kg ha⁻¹ for the combinations of 100% BO, 75% BO + 25% VE, 50% BO + 50% VE, 25% BO + 75% VE and 100% VE respectively (Table 1). There was, therefore, adequate growth and development of the black oats and vetch, with similar values for FW between the single crops and the intercrops under the environmental conditions of the site (Figure 2).
Table 1 - Planned plot size (X = Xr × Xc) in basic experimental units (BEU), with Xr adjacent BEU in a row and Xc adjacent BEU in a column; number of plots with a size of X BEU (n = 36/X); mean value of plots with a size of X BEU [M(X); in g]; coefficient of variation (%) between plots with a size of X BEU [CV(X)]; and variance per BEU between plots with a size of X BEU [VU(X)]. Fresh weight for sowing density in black oats (BO) and vetch (VE).

| T | Xr | Xc | X | n | 100% BO | -75% BO + 25% VE | -50% BO + 50% VE | -25% BO + 75% VE | 100% VE |
|---|---|---|---|---|--------|----------------|----------------|----------------|--------|
|   |   |   |   | 1 |        |                 |                 |                 |        |
|   |   |   |   | 1 |        |                 |                 |                 |        |
|   |   |   |   | 2 |        |                 |                 |                 |        |
|   |   |   |   | 3 |        |                 |                 |                 |        |
|   |   |   |   | 4 |        |                 |                 |                 |        |
|   |   |   |   | 5 |        |                 |                 |                 |        |
|   |   |   |   | 6 |        |                 |                 |                 |        |
|   |   |   |   | 7 |        |                 |                 |                 |        |
|   |   |   |   | 8 |        |                 |                 |                 |        |
|   |   |   |   | 9 |        |                 |                 |                 |        |
|   |   |   |   | 10|        |                 |                 |                 |        |
|   |   |   |   | 11|        |                 |                 |                 |        |
|   |   |   |   | 12|        |                 |                 |                 |        |
|   |   |   |   | 13|        |                 |                 |                 |        |
|   |   |   |   | 14|        |                 |                 |                 |        |
|   |   |   |   | 15|        |                 |                 |                 |        |
|   |   |   |   | 16|        |                 |                 |                 |        |
|   |   |   |   | 17|        |                 |                 |                 |        |
|   |   |   |   | 18|        |                 |                 |                 |        |
|   |   |   |   | 19|        |                 |                 |                 |        |
|   |   |   |   | 20|        |                 |                 |                 |        |
|   |   |   |   | 21|        |                 |                 |                 |        |
|   |   |   |   | 22|        |                 |                 |                 |        |
|   |   |   |   | 23|        |                 |                 |                 |        |
|   |   |   |   | 24|        |                 |                 |                 |        |
|   |   |   |   | 25|        |                 |                 |                 |        |
|   |   |   |   | 26|        |                 |                 |                 |        |
|   |   |   |   | 27|        |                 |                 |                 |        |
|   |   |   |   | 28|        |                 |                 |                 |        |
|   |   |   |   | 29|        |                 |                 |                 |        |
|   |   |   |   | 30|        |                 |                 |                 |        |
|   |   |   |   | 31|        |                 |                 |                 |        |
|   |   |   |   | 32|        |                 |                 |                 |        |
|   |   |   |   | 33|        |                 |                 |                 |        |
|   |   |   |   | 34|        |                 |                 |                 |        |
|   |   |   |   | 35|        |                 |                 |                 |        |
|   |   |   |   | 36|        |                 |                 |                 |        |

1Each uniformity trial with a size of 6 m × 6 m (36 m²) was divided into a matrix of six rows and six columns.

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beneficial aspects of the residue from these species, when intercropped, in relation to soil protection and nitrogen supply, control and a reduction in plant density, have been highlighted (HEINRICHS et al., 2001; FORTE et al., 2018a, 2018b, 2018c), and justify the importance of evaluating the FW of these plants.

The mean value of the coefficient of variation (CV) for FW in the three trials of each combination was 11.76%, 11.58%, 13.33%, 11.53% and 18.23% for the combinations of 100% BO, 75% BO + 25% VE, 50% BO + 50% VE, 25% BO + 75% VE and 100% VE respectively. Although, in the single-crop vetch the CV was greater (CV = 18.23%) in relation to the single-crop black oats (11.76%) and to the intercropped BO and VE (11.53% ≤ CV ≤ 13.33%), at these values, all the coefficients are considered average according to the PIMENTEL-GOMES classification (2009) for agricultural crops.
in general, i.e. they are within the range of 10% and 20%. This suggested that experiments with black oats and vetch, whether as single crops or intercropped, have similar experimental precision. The variations in CV between the combinations might be associated with environmental and genotypic variability, and genotype interaction with the environment.

In the 15 uniformity trials, there were visible marked decreases in variance per BEU \(VU_{(X)}\) for plots of up to four BEU in size (4 m\(^2\)), midway between four and ten BEU, tending to stabilise for plots greater than ten BEU (Figure 1). This variance is phenotypic, and is therefore the sum of environmental and genotypic variances, and of genotype interaction with the environment. The behaviour was similar for other ground cover plants such as the velvet bean (CANGNELUTTI FILHO et al., 2014b) and the forage turnip (CANGNELUTTI FILHO et al., 2014c). As such, a plot size of up to ten BEU (10 m\(^2\)) is suggested for evaluating the fresh weight of single-crop or intercropped black oats and vetch, as the gain in experimental precision (a decrease in \(VU_{(X)}\)) with the increase in plot size starting from ten BEU was negligible. This value of 10 m\(^2\) is relatively greater than the plot size of 4.14 m\(^2\) determined to evaluate the fresh weight of black oats (CARGNELUTTI FILHO et al., 2014a) and of 4.52 m\(^2\) to assess the fresh weight of vetch (CARGNELUTTI FILHO et al., 2017). It should be considered that these authors used another methodology, i.e. the maximum curvature of the model of the coefficient of variation (PARANAÍBA et al., 2009).

To evaluate the fresh weight of single-crop or intercropped black oats and vetch in experiments conducted in completely randomized (CRD) and randomized complete block (RCB) designs, the optimal plot size \((Xo)\), estimated by the HATHEWAY method (1961) from a fixed number of treatments \((i)\) and replications \((r)\), increases with the rise in precision \((d)\) (Tables 2 and 3). For example, to evaluate the FW of black oats (100% BO) in an RCB design with five treatments \((i = 5)\) and three replications \((r = 3)\), so that in 80% of the experiments (power = 0.80) differences between treatments of \(d = 20\%\) of the overall mean of the experiment (lower precision) are detected as significant at a probability of 0.05, the plot size should be four BEU (4 m\(^2\)) (Table 3). A plot size equal to or greater than this is feasible in field experiments, which makes it possible to improve the experimental precision. For example, plots of 20 m\(^2\) would make it possible to achieve \(d = 10\%\). However, under these same conditions, for \(d = 2\%\) (greater precision), a plot of 1330 BEU (1330 m\(^2\)) would be necessary. In this situation, the experimental precision is greater, however, conducting a field experiment with a plot of 1330 m\(^2\) is impractical. As such, high experimental accuracies (low percentages of \(d\)) are difficult to achieve in practice due to the need for large plot.

Figure 2 - Daily minimum, maximum and mean air temperatures (°C) and rainfall (mm) for the period of the uniformity trials comprising combinations of sowing densities in black oats (Avena strigosa Schreb) ‘Embrapa 139’ and vetch (Vicia sativa L.) ‘SS Ametista’. Data obtained from the Automatic Weather Station of the Federal University of Santa Maria (Source: INMET Network).
Table 2 - Optimal plot size in m², for a completely randomized design, in combinations of t treatments, r replications and d levels of precision, for fresh weight at sowing densities in black oats and vetch.

| d (%) | t=5 treatments | t=10 treatments | t=15 treatments | t=20 treatments |
|-------|----------------|-----------------|-----------------|-----------------|
|       |                | 2               | 4               | 6               | 8               |
| 2     | 1236           | 770             | 548             | 420             | 337             | 279             |
| 4     | 200            | 124             | 89              | 68              | 55              | 45              |
| 6     | 33             | 20              | 15              | 11              | 9               | 8               |
| 8     | 18             | 12              | 8               | 7               | 5               | 5               |
| 10    | 12             | 7               | 5               | 4               | 3               | 3               |
| 12    | 8              | 5               | 4               | 3               | 3               | 2               |
| 14    | 6              | 4               | 3               | 3               | 2               | 2               |
| 16    | 4              | 3               | 2               | 2               | 2               | 2               |
| 18    | 4              | 3               | 2               | 2               | 2               | 1               |
| 20    | 3              | 2               | 2               | 1               | 1               | 1               |

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7% black oats + 2% vetch (index of soil heterogeneity $b=1.0292$, CV = 11.58%)

| d (%) | t=5 treatments | t=10 treatments | t=15 treatments | t=20 treatments |
|-------|----------------|-----------------|-----------------|-----------------|
| 2     | 1861           | 151             | 102             | 84              | 71              | 62              |
| 4     | 1861           | 151             | 102             | 84              | 71              | 62              |
| 6     | 1861           | 151             | 102             | 84              | 71              | 62              |
| 8     | 1861           | 151             | 102             | 84              | 71              | 62              |

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50% black oats + 50% vetch (index of soil heterogeneity $b=1.0746$, CV = 13.33%)

| d (%) | t=5 treatments | t=10 treatments | t=15 treatments | t=20 treatments |
|-------|----------------|-----------------|-----------------|-----------------|
| 2     | 193           | 159             | 109             | 91              | 78              | 68              |
| 4     | 193           | 159             | 109             | 91              | 78              | 68              |
| 6     | 193           | 159             | 109             | 91              | 78              | 68              |
| 8     | 193           | 159             | 109             | 91              | 78              | 68              |

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2% black oats + 75% vetch (index of soil heterogeneity $b=0.7215$, CV = 11.55%)

| d (%) | t=5 treatments | t=10 treatments | t=15 treatments | t=20 treatments |
|-------|----------------|-----------------|-----------------|-----------------|
| 2     | 1059           | 1002            | 722             | 545             | 652             | 634             |
| 4     | 2491           | 151             | 106             | 80              | 64              | 52              |
| 6     | 815            | 35              | 25              | 21              | 17              | 17              |
| 8     | 37             | 23              | 16              | 12              | 10              | 8               |
| 10    | 12             | 9               | 7               | 5               | 5               | 4               |
| 12    | 8              | 6               | 4               | 3               | 3               | 3               |
| 14    | 5              | 4               | 3               | 2               | 2               | 2               |
| 16    | 4              | 3               | 2               | 2               | 2               | 2               |
| 18    | 4              | 3               | 2               | 2               | 2               | 2               |

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100% vetch (index of soil heterogeneity $b=1.0968$, CV = 18.23%)

| d (%) | t=5 treatments | t=10 treatments | t=15 treatments | t=20 treatments |
|-------|----------------|-----------------|-----------------|-----------------|
| 2     | 308            | 222             | 175             | 146             | 125             | 110             |
| 4     | 87             | 63              | 50              | 42              | 36              | 31              |
| 6     | 42             | 30              | 24              | 20              | 17              | 15              |
| 8     | 25             | 18              | 14              | 12              | 10              | 9               |
| 10    | 17             | 12              | 9               | 8               | 7               | 6               |
| 12    | 12             | 9               | 7               | 6               | 5               | 5               |
| 14    | 9              | 7               | 6               | 5               | 4               | 4               |

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sizes, as noted by CARGNELUTTI FILHO et al. (2014b, 2014c). Similar behaviour was observed for the combinations of 75% BO + 25% VE, 50% BO + 50% VE, 25% BO + 75% VE and 100% VE (Table 3).

In the CRD and RCB designs, with fixed values for i and d, the value of Xo decreases for an increase in r. Furthermore, with fixed values for r and d, there is a decrease in Xo for an increase in i (Tables 2 and 3). The greater the number of treatments and number of replications, the greater the number of error degrees of freedom and, consequently, the lower the estimate of residual variance (mean square error), i.e. the greater the experimental precision.

In the HATHEWAY methodology (1961), based on a fixed value for the Smith index of soil heterogeneity (1938) (b), Xo is dependent on i, r, and d. Therefore, taking the number of treatments and the desired precision as a basis, it is possible to use the information from this study to plan the plot size and number of replications to be used. For example, if the researcher wants to evaluate the FW of ten treatments of single-crop black oats (100% BO) in an RCB design and requires a precision (d) of 10%, among the various options, he could use plots of 16 BEU (16 m²) and three replications, 11 BEU (11 m²) and four replications, eight BEU (8 m²) and five replications, six BEU (6 m²) and six replications, five BEU (5 m²) and seven replications or four BEU (4 m²) and eight replications (Table 3). For these six options, the area of the experiment would be 480, 440, 400, 360, 350 and 320 m² respectively. Therefore, for the same precision (d = 10% in this case), smaller plots and a greater number of replications are more efficient in their use of the experimental area, as discussed in CARGNELUTTI FILHO et al. (2014b, 2014c) and STORCK et al. (2016). It is important to consider that with the increase in the number of replications, a greater number of evaluations become necessary, and as the characteristic is difficult to measure and/or costly to evaluate, the use of a larger plot size and smaller number of replications might be advantageous, as long as there is sufficient area for the experiment. Therefore, depending on the available area, the number of treatments to be evaluated and the desired precision, the researcher should investigate which combination of plot size and number of replications is the most appropriate.

The information from this study makes it possible to investigate 240 scenarios comprising combinations of i treatments (i = 5, 10, 15 and 20), r replications (r = 3, 4, 5, 6, 7 and 8) and d differences between the mean values of those treatments detected as significant at 5% probability (d = 2%, 4%, 6%, 8%, 10%, 12%, 14%, 16%, 18% and 20%) for each combination and each design (Tables 2 and 3). Other scenarios can be simulated from the expression $Xo = \sqrt{\frac{(2i-1)(r-1)CV^2}{rd^2}}$ (HATHEWAY, 1961), based on the mean value of the index of soil heterogeneity (b) of the SMITH function (1938) and the mean value of the coefficient of variation (CV) for FW in the three trials of each combination. The following estimates would therefore be used for the combinations: 100% BO (b = 0.7593, CV = 11.76%), 75% BO + 25% VE (b = 1.0292, CV = 11.58%), 50% BO + 50% VE (b = 1.0746, CV = 13.33%), 25% BO + 75% VE (b = 0.7215, CV = 11.53%) and 100% VE (b = 1.0968, CV = 18.23%) (Tables 2 and 3).

In this context, as an example, to evaluate the FW of eight treatments of black oats (100% BO), with four replications and d = 10% in a randomized complete block design (RCB), we have: $b = 0.7593, DF = (8-1)(4-1) = 21, t = \text{INVT}(0.05; 21) = 2.07961383$, $t = \text{INVT}(0.40; 21) = 0.85907403, CV = 11.76\%, r = 4, d = 10\%$. Therefore, the optimal plot size (Xo) is

$$Xo = \frac{\text{INVT}(0.40; 21) \times 2.07961383}{0.85907403 \times 11.76\%} \times 10^2 = 23.49 \approx 24 \text{ BEU}.$$  

In simulating scenarios for a completely randomized design (CRD), only the expression for calculating the number of degrees of freedom will change, i.e. for this design, $DF = i(r-1)$, where i is the number of treatments and r is the number of replications. In this example, therefore, $b = 0.7593, DF = (8-1) = 24, t = \text{INVT}(0.05; 24) = 2.06389854$, $t = \text{INVT}(0.40; 24) = 0.85685545, CV = 11.76\%, r = 4$ and d = 10%. Hence $Xo = \frac{\text{INVT}(0.40; 24) \times 2.06389854}{0.85685545 \times 11.76\%} \times 10^2 = 22.02 \approx 23 \text{ BEU}$. For the same experimental values, the smaller plot size in CRD (23 BEU) compared to the RCB design (24 BEU) confirmed the greater efficiency of CRD when the experimental area is homogeneous (STORCK et al., 2016).

The results of this study make it possible to define the plot size and the number of replications in experiments to evaluate the fresh weight of black oats and the common vetch, grown as single crops or intercropped. However, it can generally be inferred that in experiments in a completely randomized or randomized complete block design, with from 5 to 20 treatments and five replications, plots of 10 m² are sufficient to identify, at a probability of 0.05, significant differences between treatments of 10% of the overall mean value of the experiment. The suggestion for a plot size of 10 m² is supported by viability in the field and the stabilized precision starting with this plot size.

**CONCLUSION**

In experiments to evaluate the fresh weight of single-crop or intercropped black oats and the common vetch, grown as single crops or intercropped.
Table 3 - Optimal plot size in m², for a randomized complete block design, in combinations of i treatments, r replications and d levels of precision, for fresh weight at sowing densities in black oats and vetch.

| d (%) | i = 5 treatments | i = 10 treatments | i = 15 treatments | i = 20 treatments |
|-------|------------------|--------------------|-------------------|-------------------|
|       |                  |                    |                   |                   |
| 2     | 1100             | 730                | 410               | 310               |
| 4     | 910              | 630                | 420               | 320               |
| 6     | 720              | 500                | 420               | 320               |
| 8     | 600              | 370                | 420               | 320               |
| 10    | 490              | 290                | 420               | 320               |
| 12    | 410              | 210                | 420               | 320               |
| 14    | 340              | 170                | 420               | 320               |
| 16    | 280              | 140                | 420               | 320               |
| 20    | 210              | 100                | 420               | 320               |
| 25    | 160              | 70                 | 420               | 320               |
| 30    | 130              | 50                 | 420               | 320               |
| 40    | 100              | 30                 | 420               | 320               |
| 50    | 90               | 20                 | 420               | 320               |
| 60    | 80               | 15                 | 420               | 320               |
| 70    | 70               | 10                 | 420               | 320               |
| 80    | 60               | 7                 | 420               | 320               |
| 90    | 50               | 5                 | 420               | 320               |
| 100   | 40               | 3                 | 420               | 320               |
| 125   | 30               | 2                 | 420               | 320               |
| 150   | 20               | 1                 | 420               | 320               |
| 200   | 15               | 0.5                | 420               | 320               |

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vetch in a completely randomized or randomized complete block design, with from 5 to 20 treatments and 5 replications, plots of 10 m² are sufficient to identify, at a probability of 0.05, significant differences between treatments of 10% of the overall mean value of the experiment.

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DECLARATION OF CONFLICT OF INTERESTS

The authors declare no conflict of interest. The founding sponsors had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, and in the decision to publish the results.

AUTHOR CONTRIBUTIONS

All authors contributed equally for the conception and writing of the manuscript. All authors critically revised the manuscript and approved of the final version.

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