Ciência Rural, Santa Maria, v.54:03, e20220579, 2024 http://doi.org/10.1590/0103-8478cr20220579

ISSNe 1678-4596
CROP PRODUCTION

Sampling sufficiency, correlation and path analysis in forage pea

Alberto Cargnelutti Filho[1*] Ismael Mario Márcio Neu[2] Murilo Vieira Loro[2] Valéria Escaio Bubans[2] Vithória Morena Ortiz[2]

[1]Departamento de Fitotecnia, Programa de Pós-graduação em Agronomia, Universidade Federal de Santa Maria (UFSM), 97105-900, Santa Maria, RS, Brasil. E-mail: alberto.cargnelutti.filho@gmail.com. *Corresponding author.
[2]Programa de Pós-graduação em Agronomia, Universidade Federal de Santa Maria (UFSM), 97105-900, Santa Maria, RS, Brasil.

ABSTRACT: It is important to determine the number of plants to be evaluated to allow accurate inferences about the traits under evaluation. Investigating the linear relations among traits is important for identifying traits for indirect selection. So, the objectives of this study were to determine the sample size (number of plants) necessary to estimate the means of forage pea traits and to investigate the relations among the traits. Experiments were carried out in 2021 with three sowing dates (May 3, May 26 and July 13). Five hundred plants were randomly sampled, 100 plants in each of the five evaluation dates (June 25, August 30, July 24, September 17, September 16). In these 500 plants, the traits plant height, number of branches, number of nodes, number of leaves, number of pods, fresh matter of leaves, fresh matter of stems, fresh matter of pods, fresh matter of shoots, dry matter of leaves, dry matter of stems, dry matter of pods, and dry matter of shoots, were evaluated. The sample size was calculated to estimate the means of these traits, based on Student's t-distribution, and the relations among traits were investigated through correlation and path analysis. In an experiment, to estimate the means of these 13 traits of forage pea, with an estimation error of approximately 10% of the mean, 99 plants per treatment should be sampled. The numbers of pods and leaves have a positive linear relations with fresh and dry matter of shoots.
Key words: *Pisum sativum* subsp. *arvense* (L.) Poir, linear relations, number of plants, sample sizing.

Suficiência amostral, correlação e análise de trilha em ervilha forrageira

RESUMO: É importante dimensionar o número de plantas a serem avaliadas para possibilitar inferências precisas sobre os caracteres em avaliação. Investigar as relações lineares entre caracteres é importante para a identificação de caracteres para a seleção indireta. Assim, os objetivos deste trabalho foram determinar o tamanho de amostra (número de plantas) necessário para a estimação da média de caracteres de ervilha forrageira e investigar as relações entre os caracteres. Foram conduzidos experimentos, no ano de 2021, em três datas de semeadura (03 de maio, 26 de maio e 13 de julho). Foram amostradas, aleatoriamente, 500 plantas, sendo 100 plantas em cada uma das cinco datas de avaliação (25 de junho, 30 de agosto, 24 de julho, 17 de setembro e 16 de setembro). Nessas 500 plantas avaliaram-se os caracteres altura de planta, número de ramificações, número de nós, número de folhas, número de legumes, matéria fresca de folhas, matéria fresca de caule, matéria fresca de legumes, matéria fresca de parte aérea, matéria seca de folhas, matéria seca de caule, matéria seca de legumes e matéria seca de parte aérea. Foi calculado o tamanho de amostra para a estimação da média desses caracteres, com base na distribuição t de *Student* e investigada a relação entre os caracteres por meio de análises de correlação e de trilha. Em um experimento, para a estimação da média desses 13 caracteres de ervilha forrageira, com erro de estimação de aproximadamente 10% da média, devem ser amostradas 99 plantas por tratamento. Os números de legumes e de folhas têm relação linear positiva com as matérias fresca e seca de parte aérea.
Palavras-chave: *Pisum sativum* subsp. *arvense* (L.) Poir, dimensionamento amostral, número de plantas, relações lineares.

INTRODUCTION

Forage pea (*Pisum sativum* subsp. *arvense* (L.) Poir) is an annual winter legume crop used as a ground cover plant and with nitrogen fixation capacity. It has a high rate of shoot biomass production, with a low carbon/nitrogen ratio, favoring the decomposition and cycling of nutrients (CARVALHO et al., 2022).

Experiments with this species are conducted in the field. Limitations of time, labor and financial resources hinder the evaluation of all plants (individuals) in the usable area of the experimental unit (plot). Thus, it is common to evaluate part of the plants (sample) in the plot, and the sample should be representative of the plants in the experimental unit (STORCK et al., 2016). Thus, it is important to properly define the number of plants that must be evaluated to enable accurate inferences about the traits under evaluation.

Sample size for estimating the means of traits has been determined in species of the Fabaceae family, such as: *Cajanus cajan* (FACCO et al., 2015; FACCO et al., 2016), *Crotalaria spectabilis* (TOEBE et al., 2017), *Crotalaria juncea* (SCHABARUM et al.,

Received 10.20.22 Approved 06.02.23 Returned by the author 07.04.23
CR-2022-0579.R1
Editors: Leandro Souza da Silva Alessandro Dal'Col Lucio

2018a; SCHABARUM et al., 2018b), and *Canavalia ensiformis* (CARGNELUTTI FILHO et al., 2018b). Variation in sample size among traits and species has been reported.

Pearson's linear correlation coefficient (r) and path analysis are statistical procedures used to investigate the linear relations in a set of traits. Two traits can have perfect negative linear correlation (r = -1) or perfect positive linear correlation (r = 1), or even absence of linear relation (r = 0) (FERREIRA, 2009; BUSSAB & MORETTIN, 2017). Path analysis allows decomposing the correlation coefficients into direct and indirect effects of explanatory variables on a main variable and identifying whether there is a linear association of cause and effect, enabling the identification of traits for indirect selection (CRUZ et al., 2012; CRUZ et al., 2014). These statistical procedures have been used to study the linear association among traits of *Raphanus sativus* and *Lupinus albus* (CARGNELUTTI FILHO et al., 2014), *Crotalaria spectabilis* (TOEBE et al., 2017), *Cajanus cajan* (CARGNELUTTI FILHO et al., 2017) and *Canavalia ensiformis* (CARGNELUTTI FILHO et al., 2018a).

It is assumed that these statistical procedures, applied to a set of traits of forage pea, generate important information to support the design of experiments with better precision. Thus, the objectives of this study were to determine the sample size (number of plants) necessary for estimating the means of plant height, numbers of branches, nodes, leaves and pods, and fresh and dry matter of leaves, stems, pods and shoots of forage pea and to investigate the relations among the traits.

MATERIALS AND METHODS

Three uniformity trials (blank experiments) were conducted with the forage pea crop (*Pisum sativum* subsp. *arvense* (L.) Poir) cv. 'Iapar 83', in Santa Maria, State of Rio Grande do Sul, Brazil (29º42'S latitude, 53º49'W longitude and 95 m altitude). In this place, the climate is humid subtropical Cfa (ALVARES et al., 2013), and the soil is *Argissolo Vermelho Distrófico Arênico* (Ultisol) (SANTOS et al., 2018).

The cv. 'Iapar 83' was sown in the year 2021 on May 3 (trial 1), May 26 (trial 2) and July 13 (trial 3). In each trial, with dimensions of 8 m × 20 m (160 m²), sowing was carried out in rows, spaced 0.50 m apart, by placing 24 seeds per meter of row. Basal fertilization consisted of 35 kg ha^{-1} of N, 135 kg ha^{-1} of P_2O_5 and 135 kg ha^{-1} of K_2O. In trial 1, 100 plants were collected on June 25, that is, at 53 days after sowing (DAS), and more 100 plants were collected on August 30 (119 DAS). In trial 2, 100 plants were collected on July 24 (59 DAS), and more 100 plants were collected on September 17 (114 DAS). In trial 3, 100 plants were collected on September 16 (65 DAS).

In each of these 500 plants, randomly collected, the following traits were evaluated: plant height (PH, in cm), number of branches (NB), number of nodes (NN), number of leaves (NL), number of pods (NP), fresh matter of leaves (FML, in g plant^{-1}), fresh matter of stems (FMS, in g plant^{-1}), fresh matter of pods (FMP, in g plant^{-1}), fresh matter of shoots (FMSH = FML + FMS + FMP, in g plant^{-1}), dry matter of leaves (DML, in g plant^{-1}), dry matter of stems (DMS, in g plant^{-1}), dry matter of pods (DMP, in g plant^{-1}) and dry matter of shoots (DMSH = DML + DMS + DMP, in g plant^{-1}). For these 13 traits, measures of central tendency, dispersion, skewness and p-value of the Kolmogorov-Smirnov normality test were calculated.

For each trait, based on the number of plants sampled, i.e., 100 plants, the sample size (n) was calculated for estimation errors (semi-amplitudes of the confidence interval) fixed at 2%, 4%, 6%, 8%, 10%, 15% and 20% of the mean (m), that is, 0.02×m (higher precision), 0.04×m, 0.06×m, 0.08×m, 0.10×m, 0.15×m and 0.20×m (lower precision), with a confidence level (1-α) of 95%, through the expression $n = \left(\frac{t_{\alpha/2}\, s}{estimation\ error}\right)^2$ (FERREIRA, 2009; BUSSAB & MORETTIN, 2017), where $t_{\alpha/2}$ is the critical value of the Student's t-distribution, whose area on the right-hand side is equal to α/2, that is, the value of t, such that $P(t>t_{\alpha/2})=\alpha/2$, with α=5% significance and n-1 degrees of freedom, and s is the estimate of the standard deviation. Then, by fixing n equal to 100 plants, which was the sample size used in the sampling, the estimation error was calculated as a percentage of the mean (m) for each of the traits, using the following expression: $estimation\ error = \frac{t_{\alpha/2}\, s}{\sqrt{n}\, m} \times 100$.

To investigate the relations among the traits PH, NB, NN, NL, NP, FML, FMS, FMP, FMSH, DML, DMS, DMP and DMSH, the matrix of Pearson's linear correlation coefficients (r) was determined, and Student's t-test was used to assess the significance of r at 5%. In the matrix of correlation among the traits PH, NB, NN, NL and NP, the diagnosis of multicollinearity was made (CRUZ et al., 2014).

Afterwards, path analysis of the main variables (FMSH and DMSH) as a function of the explanatory variables (PH, NB, NN, NL and

NP) was performed according to the methodology described in Cruz et al. (2012) and Cruz et al. (2014). Statistical analyses were carried out using the applications Microsoft Office Excel® and Genes (CRUZ, 2016).

RESULTS AND DISCUSSION

Regarding the data of PH, NB, NN, NL, NP, FML, FMS, FMP, FMSH, DML, DMS, DMP and DMSH, the p-value of the Kolmogorov-Smirnov test ranged from 0.000 to 0.974, with mean of 0.229 in the five evaluations (Table 1). The higher the p-value, the greater the adherence of the data to the normal distribution curve. The proximity of the mean to the median and skewness close to zero ($-1.37 \leq$ skewness ≤ 2.11) are indicative of a slight deviation from the normal distribution curve (FERREIRA, 2009; BUSSAB & MORETTIN, 2017). Thus, this data set is considered suitable for studies of sample sizing based on Student's t-distribution and linear relations through correlation and path analyses.

Based on the dispersion measures, variation among the plants sampled in the five evaluations was observed for all traits. Such variation is important for studies on sample sizing and relations through correlation and path analyses, because it includes plants of different heights (short, medium and tall), which are common in field experiments.

In the five evaluations, it was observed that the coefficients of variation (CV) for the traits NB, NL, NP, FML, FMS, FMP, FMSH, DML, DMS, DMP and DMSH ($23.45\% \leq CV \leq 97.98\%$, mean of 51.39%) were comparatively higher than those for the traits PH and NN ($9.11\% \leq CV \leq 18.92\%$, mean of 13.88%) (Table 1). Higher CV was found for the traits NL, FML, FMS, FMP, FMSH, DML, DMS, DMP and DMSH in the two evaluations performed at 119 and 114 DAS compared to the evaluations at 53, 59 and 65 DAS. The greater permanence of plants under environmental interference may possibly explain the increased variation. Thus, for the same precision, a larger sample size is expected to estimate the mean of the traits with higher CV.

The sample sizes (number of plants) for estimating the mean, with estimation error (semi-amplitude of the 95% confidence interval) equal to 10% of the mean (m), that is, 0.10×m, ranged from 4 plants for NN to 378 plants for DMP, with mean of 99 plants (Table 2). In Microsoft Office Excel®, these sizes are obtained, respectively, by the following expressions: =ARREDONDAR.PARA.CIMA((((INVT(0.05;99)*1.0299)/(0.10*11.3000))^2);0)=4 plants and=ARREDONDAR.PARA.CIMA((((INVT(0.05;99)*1.6028)/(0.10*1.6358))^2);0) = 378 plants. For the same precision, larger sample sizes were observed for the traits NB, NL, NP, FML, FMS, FMP, FMSH, DML, DMS, DMP and DMSH in the evaluations performed at 119 and 114 DAS. Larger sample sizes of these 11 traits in comparison to PH and NN were also observed in the five evaluations. Variation in sample size between sowing dates, evaluation dates, and traits has also been found in *Cajanus cajan* (FACCO et al., 2015; FACCO et al., 2016), *Crotalaria spectabilis* (TOEBE et al., 2017), *Crotalaria juncea* (SCHABARUM et al., 2018a; SCHABARUM et al., 2018b), and *Canavalia ensiformis* (CARGNELUTTI FILHO et al., 2018b).

When planning an experiment, if the option is to allow maximum estimation error of 10%, i.e., 0.10×m, 378 plants would be sufficient to estimate the mean of these 13 traits (largest sample size). Optionally, an estimation error close to 10%, that is, a slightly below or above 10%, could be allowed with a sample of 99 plants (average of these 13 traits in these five evaluations). With 100 plants sampled, the estimation error ranged from 1.81% to 19.44%, with mean of 8.87% (Table 2). Using a sample of 99 plants, if the experiment is planned with three replicates per treatment, 33 plants would be sampled per replicate (99/3 = 33), that is, 33 plants per plot. Also, if ten treatments were evaluated in the experiment, 990 plants would be sampled (99 per treatment). For the traits PH, NB, NN, NL and NP, individual evaluations of the 33 plants of the plot are required, while for the traits FML, FMS, FMP, FMSH, DML, DMS, DMP and DMSH the weighing of the 33 plants of the plot can be performed jointly, which can facilitate the evaluation.

The fresh and dry matter of leaves, stems, pods and shoots (FML, FMS, FMP, FMSH, DML, DMS, DMP and DMSH) showed a higher degree of positive linear association (higher r values) with NP ($0.66 \leq r \leq 0.95$, mean = 0.78), NL ($0.44 \leq r \leq 0.89$, mean = 0.74) and NB ($0.18 \leq r \leq 0.83$, mean = 0.57). Conversely, there was a weak linear association or no linear association with PH ($-0.25 \leq r \leq 0.58$) and NN ($-0.16 \leq r \leq 0.25$) (Table 3). The results suggested that the numbers of pods, leaves and branches, in this order, would be more strongly associated with the fresh and dry matter of leaves, stems, pods and shoots of forage pea.

Path analysis makes it possible to decompose r into direct and indirect effects, allowing inferences on which explanatory trait(s) (PH, NB, NN, NL and NP) has(have) a cause-and-effect relationship

Table 1 - Minimum, median, mean, maximum, standard deviation, coefficient of variation (CV), skewness and p-value of the Kolmogorov-Smirnov normality test of traits[(1)] of forage pea (*Pisum sativum* subsp. *arvense* (L.) Poir) cv. 'Iapar 83', on sowing and evaluation dates in the year 2021.

Statistic	PH	NB	NN	NL	NP	FML	FMS	FMP	FMSH	DML	DMS	DMP	DMSH
----------Sowing on May 03 and evaluation on June 25 - 53 days after sowing----------													
Minimum	14.00	2.00	7.00	15.00	-	2.43	2.02	-	4.76	0.22	0.16	-	0.42
Median	22.00	4.00	11.00	38.50	-	5.47	5.52	-	11.20	0.57	0.49	-	1.06
Mean	21.77	3.59	10.71	38.46	-	5.79	5.94	-	11.73	0.59	0.52	-	1.11
Maximum	29.00	5.00	14.00	68.00	-	12.65	12.65	-	25.30	1.12	1.02	-	2.10
Standard deviation	2.98	0.84	1.34	10.53	-	2.11	2.22	-	4.28	0.21	0.18	-	0.37
CV(%)	13.71	23.45	12.54	27.37	-	36.40	37.34	-	36.51	35.59	35.73	-	33.70
Skewness	-0.10	-0.24	-0.14	0.15	-	0.76	0.76	-	0.75	0.43	0.63	-	0.41
P-value	0.359	0.000	0.008	0.825	-	0.615	0.454	-	0.611	0.709	0.167	-	0.674
----------Sowing on May 03 and evaluation on August 30 - 119 days after sowing----------													
Minimum	95.00	1.00	15.00	27.00	1.00	3.45	8.96	0.15	19.39	0.58	2.01	0.02	3.65
Median	150.00	1.00	26.00	86.50	11.00	16.29	32.12	10.19	59.76	2.48	6.47	1.33	10.23
Mean	148.91	1.89	24.79	114.00	13.99	20.34	44.99	13.89	79.22	2.90	8.39	1.93	13.23
Maximum	191.00	6.00	31.00	430.00	47.00	65.69	167.84	61.96	252.74	8.98	26.36	10.19	39.70
Standard deviation	21.75	1.29	3.47	79.71	9.12	12.75	30.00	11.51	49.82	1.78	5.18	1.78	8.06
CV(%)	14.61	68.47	14.01	69.92	65.16	62.66	66.68	82.87	62.89	61.19	61.74	92.32	60.94
Skewness	-0.17	1.52	-0.73	1.77	1.44	1.28	1.50	1.86	1.35	1.31	1.29	2.11	1.34
P-value	0.630	0.000	0.014	0.002	0.006	0.004	0.001	0.002	0.008	0.048	0.002	0.003	0.016
----------Sowing on May 26 and evaluation on July 24 - 59 days after sowing----------													
Minimum	13.00	1.00	7.00	11.00	-	1.04	1.30	-	2.40	0.14	0.17	-	0.33
Median	19.00	2.00	12.00	23.50	-	2.38	2.93	-	5.19	0.38	0.38	-	0.76
Mean	18.87	2.01	11.30	24.73	-	2.70	3.08	-	5.78	0.39	0.40	-	0.79
Maximum	25.00	4.00	13.00	50.00	-	6.14	6.66	-	12.64	0.75	0.79	-	1.53
Standard deviation	2.56	0.97	1.03	7.63	-	1.14	1.27	-	2.37	0.14	0.14	-	0.27
CV(%)	13.57	48.22	9.11	30.84	-	42.18	41.32	-	41.06	34.68	34.88	-	34.05
Skewness	0.04	0.39	-1.37	0.76	-	1.20	1.06	-	1.16	0.65	0.65	-	0.67
P-value	0.273	0.000	0.000	0.272	-	0.056	0.284	-	0.054	0.253	0.332	-	0.521
----------Sowing on May 26 and evaluation on September 17 - 114 days after sowing----------													
Minimum	106.00	2.00	17.00	20.00	0.00	3.31	15.79	0.00	20.95	0.60	3.05	0.00	3.84
Median	160.25	4.00	23.00	70.50	8.00	14.26	49.29	7.60	73.51	2.24	11.04	1.02	14.87
Mean	157.33	3.71	23.02	97.45	10.35	17.30	55.28	11.47	84.05	2.57	12.41	1.64	16.61
Maximum	210.00	7.00	30.00	369.00	45.00	56.85	145.06	46.03	230.06	8.39	32.34	6.52	43.09
Standard deviation	21.64	1.31	2.98	74.75	8.90	11.51	29.05	10.79	47.69	1.63	6.03	1.60	8.55
CV(%)	13.76	35.38	12.97	76.71	86.00	66.51	52.55	94.11	56.74	63.25	48.60	97.98	51.48
Skewness	-0.24	0.64	0.08	1.84	1.17	1.54	1.38	1.10	1.38	1.48	1.26	1.10	1.25
P-value	0.526	0.001	0.649	0.001	0.100	0.020	0.110	0.017	0.011	0.018	0.117	0.012	0.007
----------Sowing on July 13 and evaluation on September 16 - 65 days after sowing----------													
Minimum	25.00	2.00	7.00	27.00	-	2.07	3.09	-	5.16	0.31	0.39	-	0.70
Median	43.00	3.00	13.00	66.50	-	8.12	12.36	-	20.51	1.23	1.95	-	3.16
Mean	42.31	3.25	12.78	72.99	-	8.47	13.49	-	21.97	1.27	2.04	-	3.31
Maximum	62.50	6.00	18.00	170.00	-	21.61	32.44	-	52.62	2.93	4.37	-	7.30
Standard deviation	8.01	1.05	1.99	28.02	-	3.25	5.61	-	8.77	0.46	0.85	-	1.27
CV(%)	18.92	32.25	15.55	38.39	-	38.33	41.58	-	39.94	36.05	41.51	-	38.43
Skewness	0.17	0.45	-0.04	1.26	-	1.05	0.91	-	0.95	0.62	0.66	-	0.63
P-value	0.974	0.000	0.034	0.213	-	0.391	0.455	-	0.421	0.188	0.683	-	0.649

[(1)] PH - plant height, in cm; NB - number of branches; NN - number of nodes; NL - number of leaves; NP – number of pods; FML - fresh matter of leaves, in g plant^{-1}; FMS - fresh matter of stems, in g plant^{-1}; FMP - fresh matter of pods, in g plant^{-1}; FMSH - fresh matter of shoots (FMSH = FML + FMS + FMP), in g plant^{-1}; DML - dry matter of leaves, in g plant^{-1}; DMS - dry matter of stems, in g plant^{-1}; DMP - dry matter of pods, in g plant^{-1}; and DMSH - dry matter of shoots (DMSH = DML + DMS + DMP), in g plant^{-1}.

with FMSH and DMSH (CRUZ et al., 2012; CRUZ et al., 2014). The low values of condition number (CN ≤ 11.77), which is the ratio between the highest and lowest eigenvalue of Pearson's linear correlation matrix (r) between the explanatory traits, indicate weak multicollinearity (CRUZ et al., 2014; CRUZ, 2016) (Table 4).

The strong linear association between FMSH and DMSH (0.90 ≤ r ≤ 0.99, mean of 0.96) (Table 3) explains the similar results of the path

Table 2 - Sample size (number of plants) for estimating the means of traits[1] of forage pea (*Pisum sativum* subsp. *arvense* (L.) Poir) cv. 'Iapar 83', on sowing and evaluation dates in the year 2021, for estimation errors (semi-amplitudes of the confidence interval) fixed at 2%, 4%, 6%, 8%, 10%, 15% and 20% of the mean (m), i.e., 0.02×m (higher precision), 0.04×m, 0.06×m, 0.08×m, 0.10×m, 0.15×m and 0.20×m (lower precision), with a confidence level (1-α) of 95%.

Error	PH	NB	NN	NL	NP	FML	FMS	FMP	FMSH	DML	DMS	DMP	DMSH
Sowing on May 03 and evaluation on June 25 - 53 days after sowing													
2%	185	542	155	738	-	1304	1373	-	1313	1247	1257	-	1118
4%	47	136	39	185	-	326	344	-	329	312	315	-	280
6%	21	61	18	82	-	145	153	-	146	139	140	-	125
8%	12	34	10	47	-	82	86	-	83	78	79	-	70
10%	8	22	7	30	-	53	55	-	53	50	51	-	45
15%	4	10	3	14	-	24	25	-	24	23	23	-	20
20%	2	6	2	8	-	14	14	-	14	13	13	-	12
Error (%)[2]	2.72	4.65	2.49	5.43	-	7.22	7.41	-	7.25	7.06	7.09	-	6.69
Sowing on May 03 and evaluation on August 30 - 119 days after sowing													
2%	211	4615	194	4813	4180	3865	4377	6760	3894	3686	3753	8390	3656
4%	53	1154	49	1204	1045	967	1095	1690	974	922	939	2098	914
6%	24	513	22	535	465	430	487	752	433	410	417	933	407
8%	14	289	13	301	262	242	274	423	244	231	235	525	229
10%	9	185	8	193	168	155	176	271	156	148	151	336	147
15%	4	83	4	86	75	69	78	121	70	66	67	150	65
20%	3	47	2	49	42	39	44	68	39	37	38	84	37
Error (%)	2.90	13.59	2.78	13.87	12.93	12.43	13.23	16.44	12.48	12.14	12.25	18.32	12.09
Sowing on May 26 and evaluation on July 24 - 59 days after sowing													
2%	182	2289	82	937	-	1751	1681	-	1660	1185	1198	-	1142
4%	46	573	21	235	-	438	421	-	415	297	300	-	286
6%	21	255	10	105	-	195	187	-	185	132	134	-	127
8%	12	144	6	59	-	110	106	-	104	75	75	-	72
10%	8	92	4	38	-	71	68	-	67	48	48	-	46
15%	4	41	2	17	-	32	30	-	30	22	22	-	21
20%	2	23	1	10	-	18	17	-	17	12	12	-	12
Error (%)	2.69	9.57	1.81	6.12	-	8.37	8.20	-	8.15	6.88	6.92	-	6.76
Sowing on May 26 and evaluation on September 17 - 114 days after sowing													
2%	187	1233	166	5792	7279	4354	2718	8718	3169	3938	2326	9450	2609
4%	47	309	42	1448	1820	1089	680	2180	793	985	582	2363	653
6%	21	137	19	644	809	484	302	969	353	438	259	1050	290
8%	12	78	11	362	455	273	170	545	199	247	146	591	164
10%	8	50	7	232	292	175	109	349	127	158	94	378	105
15%	4	22	3	103	130	78	49	155	57	71	42	168	47
20%	2	13	2	58	73	44	28	88	32	40	24	95	27
Error (%)	2.73	7.02	2.57	15.22	17.06	13.20	10.43	18.67	11.26	12.55	9.64	19.44	10.22
Sowing on July 13 and evaluation on September 16 - 65 days after sowing													
2%	353	1024	239	1451	-	1447	1702	-	1571	1280	1697	-	1454
4%	89	256	60	363	-	362	426	-	393	320	425	-	364
6%	40	114	27	162	-	161	190	-	175	143	189	-	162
8%	23	64	15	91	-	91	107	-	99	80	107	-	91
10%	15	41	10	59	-	58	69	-	63	52	68	-	59
15%	7	19	5	26	-	26	31	-	28	23	31	-	26
20%	4	11	3	15	-	15	18	-	16	13	17	-	15
Error (%)	3.75	6.40	3.09	7.62	-	7.61	8.25	-	7.93	7.15	8.24	-	7.63

[1] PH - plant height, in cm; NB - number of branches; NN - number of nodes; NL - number of leaves; NP – number of pods; FML - fresh matter of leaves, in g plant^{-1}; FMS - fresh matter of stems, in g plant^{-1}; FMP - fresh matter of pods, in g plant^{-1}; FMSH - fresh matter of shoots (FMSH = FML + FMS + FMP), in g plant^{-1}; DML - dry matter of leaves, in g plant^{-1}; DMS - dry matter of stems, in g plant^{-1}; DMP - dry matter of pods, in g plant^{-1}; and DMSH - dry matter of shoots (DMSH = DML + DMS + DMP), in g plant^{-1}. [2] Estimation error, in % of the mean, based on the 100 plants sampled.

analyses (Table 4). A positive and high-magnitude association between fresh and dry matter of shoots has also been observed in *Raphanus sativus* (r = 0.9671), *Lupinus albus* (r = 0.9828) (CARGNELUTTI FILHO et al., 2014), *Cajanus cajan* (r = 0.994 and 0.996) (CARGNELUTTI FILHO et al., 2017), and

Table 3 - Estimates of Pearson's linear correlation coefficients among the traits[(1)] of forage pea (*Pisum sativum* subsp. *arvense* (L.) Poir) cv. 'Iapar 83', on sowing and evaluation dates in the year 2021.

-----------Sowing on May 03 and evaluation on June 25 - above the diagonal. Sowing on May 03 and evaluation on August 30 - below the diagonal------------													
	PH	NB	NN	NL	NP	FML	FMS	FMP	FMSH	DML	DMS	DMP	DMSH
PH	1	0.07	0.06	0.17	-	0.29	0.29	-	0.29	0.33	0.26	-	0.32
NB	0.10	1	-0.02	0.55	-	0.52	0.52	-	0.52	0.42	0.45	-	0.46
NN	0.62	-0.10	1	0.24	-	0.19	0.25	-	0.22	0.03	0.16	-	0.10
NL	0.07	0.63	0.10	1		0.60	0.57	-	0.59	0.44	0.54	-	0.51
NP	0.02	0.62	0.08	0.80	1	-	-	-	-	-	-	-	-
FML	0.15	0.74	0.09	0.87	0.80	1	0.96	-	0.99	0.81	0.90	-	0.90
FMS	0.23	0.83	0.04	0.75	0.69	0.93	1	-	0.99	0.75	0.93	-	0.89
FMP	-0.14	0.41	0.05	0.71	0.85	0.72	0.53	1	-	-	-	-	-
FMSH	0.15	0.78	0.06	0.84	0.81	0.98	0.96	0.74	1	0.79	0.93	-	0.90
DML	0.11	0.70	0.09	0.89	0.82	0.98	0.87	0.79	0.96	1	0.79	-	0.95
DMS	0.21	0.81	0.07	0.78	0.76	0.95	0.99	0.64	0.98	0.91	1	-	0.94
DMP	-0.18	0.34	0.04	0.68	0.79	0.65	0.45	0.99	0.66	0.74	0.56	1	-
DMSH	0.12	0.75	0.07	0.85	0.84	0.97	0.93	0.80	0.99	0.97	0.97	0.74	1
----------Sowing on May 26 and evaluation on July 24 - above the diagonal. Sowing on May 26 and evaluation on September 17 - below the diagonal----------													
	PH	NB	NN	NL	NP	FML	FMS	FMP	FMSH	DML	DMS	DMP	DMSH
PH	1	-0.02	0.52	0.19	-	0.40	0.43	-	0.42	0.42	0.44	-	0.44
NB	0.09	1	-0.12	0.68	-	0.68	0.71	-	0.71	0.67	0.70	-	0.70
NN	0.33	0.08	1	0.10	-	0.14	0.14	-	0.14	0.16	0.14	-	0.15
NL	0.08	0.40	-0.09	1		0.79	0.78	-	0.79	0.75	0.77	-	0.78
NP	-0.16	0.25	-0.11	0.63	1	-	-	-	-	-	-	-	-
FML	0.12	0.56	-0.16	0.87	0.70	1	0.94	-	0.98	0.94	0.89	-	0.93
FMS	0.16	0.61	-0.13	0.79	0.66	0.94	1	-	0.99	0.91	0.96	-	0.96
FMP	-0.20	0.21	-0.12	0.64	0.95	0.67	0.61	1	-	-	-	-	-
FMSH	0.08	0.55	-0.15	0.83	0.79	0.97	0.98	0.76	1	0.94	0.94	-	0.96
DML	0.12	0.55	-0.15	0.84	0.71	0.99	0.93	0.68	0.96	1	0.92	-	0.98
DMS	0.13	0.57	-0.15	0.78	0.67	0.90	0.97	0.63	0.95	0.89	1	-	0.98
DMP	-0.25	0.18	-0.08	0.60	0.90	0.60	0.54	0.97	0.69	0.60	0.57	1	-
DMSH	0.07	0.54	-0.15	0.82	0.78	0.94	0.96	0.75	0.98	0.93	0.98	0.70	1
---Sowing on July 13 and evaluation on September 16 - above the diagonal---													
	PH	NB	NN	NL	NP	FML	FMS	FMP	FMSH	DML	DMS	DMP	DMSH
PH	1	0.01	0.29	0.33	-	0.54	0.57	-	0.57	0.49	0.58	-	0.56
NB		1	-0.14	0.73	-	0.57	0.53	-	0.55	0.52	0.48	-	0.51
NN			1	-0.01	-	-0.01	-0.01	-	-0.01	0.12	0.06	-	0.08
NL				1	-	0.82	0.76	-	0.79	0.77	0.74	-	0.77
NP					1	-	-	-	-	-	-	-	-
FML						1	0.96	-	0.98	0.92	0.92	-	0.95
FMS							1	-	0.99	0.88	0.97	-	0.96
FMP								1	-	-	-	-	-
FMSH									1	0.90	0.96	-	0.96
DML										1	0.89	-	0.95
DMS											1	-	0.99
DMP												1	-
DMSH													1

[(1)] PH - plant height, in cm; NB - number of branches; NN - number of nodes; NL - number of leaves; NP – number of pods; FML - fresh matter of leaves, in g plant^{-1}; FMS - fresh matter of stems, in g plant^{-1}; FMP - fresh matter of pods, in g plant^{-1}; FMSH - fresh matter of shoots (FMSH = FML + FMS + FMP), in g plant^{-1}; DML - dry matter of leaves, in g plant^{-1}; DMS - dry matter of stems, in g plant^{-1}; DMP - dry matter of pods, in g plant^{-1}; and DMSH - dry matter of shoots (DMSH = DML + DMS + DMP), in g plant^{-1}. Pearson's linear correlation coefficient ≥ 0.20 or ≤ -0.20 is significant at 5% by Student's t-test, with 98 degrees of freedom.

Canavalia ensiformis (r = 0.960) (CARGNELUTTI FILHO et al., 2018a).

NP showed a positive linear correlation ($0.776 \leq r \leq 0.845$, mean of 0.805) and direct effects ($0.302 \leq$ direct effect ≤ 0.473, mean of 0.404) with the same sign and lower magnitude with FMSH and DMSH, due to the indirect effect of NP via NL ($0.254 \leq$ indirect effect ≤ 0.298, mean of 0.275). Similarly, NL showed positive linear correlation ($0.515 \leq r \leq 0.848$, mean of 0.758) and direct effects ($0.326 \leq$ direct

Table 4 - Estimates of Pearson's correlation coefficients (r) and direct and indirect effects (path analysis) of the traits plant height (PH), number of branches (NB), number of nodes (NN), number of leaves (NL) and number of pods on fresh matter of shoots (FMSH) and dry matter of shoots (DMSH) in forage pea (*Pisum sativum* subsp. *arvense* (L.) Poir) cv. 'Iapar 83', on sowing and evaluation dates in the year 2021 (D1E1, D1E2, D2E1, D2E2 and D3E1).

Effect	D1E1	D1E2	D2E1	D2E2	D3E1	D1E1	D1E2	D2E1	D2E2	D3E1
	FMSH	FMSH	FMSH	FMSH	FMSH	DMSH	DMSH	DMSH	DMSH	DMSH
Direct of PH on	0.198	0.103	0.368	0.137	0.410	0.241	0.067	0.387	0.125	0.365
Indirect of PH via NB	0.023	0.035	-0.010	0.024	0.002	0.019	0.027	-0.010	0.023	0.001
Indirect of PH via NN	0.008	-0.021	-0.024	-0.041	-0.030	0.000	-0.007	-0.021	-0.039	-0.003
Indirect of PH via NL	0.061	0.025	0.087	0.034	0.185	0.057	0.024	0.081	0.035	0.202
Indirect of PH via NP	-	0.006	-	-0.075	-	-	0.007	-	-0.072	-
Pearson's Correlation (r)	0.290*	0.146ns	0.422*	0.080ns	0.567*	0.317*	0.119ns	0.437*	0.071ns	0.564*
Direct of NB on	0.321	0.342	0.402	0.271	0.118	0.265	0.272	0.414	0.264	0.051
Indirect of NB via PH	0.014	0.010	-0.009	0.012	0.006	0.017	0.007	-0.009	0.011	0.005
Indirect of NB via NN	-0.002	0.003	0.006	-0.010	0.015	0.000	0.001	0.005	-0.010	0.001
Indirect of NB via NL	0.192	0.238	0.310	0.160	0.411	0.179	0.233	0.288	0.162	0.447
Indirect of NB via NP	-	0.188	-	0.119	-	-	0.238	-	0.115	-
Pearson's Correlation (r)	0.525*	0.782*	0.708*	0.552*	0.550*	0.461*	0.751*	0.699*	0.543*	0.505*
Direct of NN on	0.131	-0.034	-0.046	-0.124	-0.104	0.007	-0.011	-0.040	-0.121	-0.010
Indirect of NN via PH	0.012	0.064	0.193	0.045	0.119	0.015	0.042	0.203	0.041	0.106
Indirect of NN via NB	-0.005	-0.033	-0.050	0.021	-0.017	-0.004	-0.027	-0.052	0.021	-0.007
Indirect of NN via NL	0.085	0.038	0.046	-0.038	-0.006	0.079	0.037	0.043	-0.038	-0.006
Indirect of NN via NP	-	0.024	-	-0.052	-	-	0.030	-	-0.050	-
Pearson's Correlation (r)	0.224*	0.058ns	0.143ns	-0.147ns	-0.008ns	0.097ns	0.071ns	0.154ns	-0.147ns	0.082ns
Direct of NL on	0.350	0.375	0.456	0.405	0.564	0.326	0.367	0.425	0.410	0.614
Indirect of NL via PH	0.035	0.007	0.071	0.012	0.135	0.042	0.004	0.074	0.011	0.120
Indirect of NL via NB	0.176	0.217	0.273	0.107	0.086	0.145	0.173	0.281	0.104	0.037
Indirect of NL via NN	0.032	-0.003	-0.005	0.012	0.001	0.002	-0.001	-0.004	0.011	0.000
Indirect of NL via NP	-	0.240	-	0.296	-	-	0.305	-	0.288	-
Pearson's Correlation (r)	0.593*	0.835*	0.795*	0.832*	0.786*	0.515*	0.848*	0.776*	0.824*	0.772*
Direct of NP on	-	0.302	-	0.473	-	-	0.383	-	0.459	-
Indirect of NP via PH	-	0.002	-	-0.022	-	-	0.001	-	-0.020	-
Indirect of NP via NB	-	0.213	-	0.068	-	-	0.170	-	0.066	-
Indirect of NP via NN	-	-0.003	-	0.014	-	-	-0.001	-	0.013	-
Indirect of NP via NL	-	0.298	-	0.254	-	-	0.292	-	0.257	-
Pearson's Correlation (r)	-	0.812*	-	0.787*	-	-	0.845*	-	0.776*	-
Coefficient of determination	0.462	0.839	0.796	0.888	0.742	0.367	0.847	0.783	0.864	0.705
Residual variable	0.538	0.161	0.204	0.112	0.258	0.633	0.153	0.217	0.136	0.295
Condition number	4.26	11.77	6.08	6.12	8.70	4.26	11.77	6.08	6.12	8.70

D1E1 - Sowing on May 3 and evaluation on June 25; D1E2 - Sowing on May 3 and evaluation on August 30; D2E1 - Sowing on May 26 and evaluation on July 24; D2E2 - Sowing on May 26 and evaluation on September 17; and D3E1 - Sowing on July 13 and evaluation on September 16. * Significant at 5% by Student's t-test, with 98 degrees of freedom.

effect ≤ 0.614, mean of 0.429) with the same sign and lower magnitude with FMSH and DMSH, due to the indirect effect of NL via NP (0.240 ≤ indirect effect ≤ 0.305, mean of 0.282). NB showed a positive linear correlation (0.461 ≤ r ≤ 0.782, mean of 0.608) and negligible direct effects (0.051 ≤ direct effect ≤ 0.414, mean of 0.272) with FMSH and DMSH. Therefore, the association is explained by the greater indirect effects via NL (0.160 ≤ indirect effect ≤ 0.447, mean of 0.262) and NP (0.115 ≤ indirect effect ≤ 0.238, mean of 0.165).

It can be inferred that plants with more leaves and more pods have greater amounts of fresh and dry matter of shoots. The fact that it is not necessary to destroy the plants to count the number of leaves and pods is advantageous, because it allows the plants to be selected aiming at increments in fresh and dry matter, keeping them until the production of seeds. For direct selection, it would be necessary to destroy the plants for weighing FMSH and DMSH. Cause-and-effect relationships among several traits and possibility of indirect selection have also been found in *Raphanus sativus* and *Lupinus albus* (CARGNELUTTI FILHO et al., 2014), *Crotalaria spectabilis* (TOEBE et al., 2017), *Cajanus cajan* (CARGNELUTTI FILHO et al., 2017), and *Canavalia ensiformis* (CARGNELUTTI FILHO et al., 2018a).

CONCLUSION

In an experiment, for estimating the means of plant height, numbers of branches, nodes, leaves and pods, and the fresh and dry matter of leaves, stems, pods and shoots of forage pea, with an estimation error of approximately 10% of the mean, 99 plants should be sampled per treatment. The numbers of pods and leaves have a positive linear relation with fresh and dry matter of shoots.

ACKNOWLEDGEMENTS

To the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq - Processes 304652/2017-2; 304878/2022-7; 159611/2019-9), the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES), Brasil - Finance code 001, 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.

DECLARATION OF CONFLICT OF INTEREST

We have no conflict of interest to declare with respect to the research, authorship and/or publication of this article.

AUTHORS' CONTRIBUTIONS

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

REFERENCES

ALVARES, C. A. et al. Köppen's climate classification map for Brazil. **Meteorologische Zeitschrift**, v.22, n.6, p.711-728, 2013. Available from: <https://doi.org/10.1127/0941-2948/2013/0507>. Accessed: Oct. 20, 2022.

BUSSAB, W. O.; MORETTIN, P. A. **Estatística básica**. 9a ed. São Paulo: Saraiva, 2017. 568p.

CARGNELUTTI FILHO, A. et al. Linear relations among pigeon pea traits. **Comunicata Scientiae**, v.8, n.3, p.493-502, 2017. Available from: <http://doi.org/10.14295/CS.v8i3.1420>. Accessed: Oct. 20, 2022.

CARGNELUTTI FILHO, A. et al. Linear relations among traits in jack bean (*Canavalia ensiformis*). **Bioagro**, v.30, n.2, p.157-162, 2018a. Available from: <https://revistas.uclave.org/index.php/bioagro/article/view/1291>. Accessed: Oct. 20, 2022.

CARGNELUTTI FILHO, A. et al. Linear relations among characters of forage turnips and of white lupine. **Ciência Rural**, v.44, n.1, p.18-24, 2014. Available from: <http://doi.org/10.1590/S0103-84782014000100004>. Accessed: Oct. 20, 2022.

CARGNELUTTI FILHO, A. et al. Sample size to estimate the mean of traits in jack bean. **Revista Brasileira de Ciências Agrárias**, v.13, n.1, e5505, 2018b. Available from: <http://doi.org/10.5039/agraria.v13i1a5505>. Accessed: Oct. 20, 2022.

CARVALHO, M. L. et al. **Guia prático de plantas de cobertura: aspectos filotécnicos e impactos sobre a saúde do solo** [recurso eletrônico]. Piracicaba: ESALQ-USP. 2022. Available from: <http://doi.org/10.11606/9786589722151>. Accessed: Oct. 20, 2022.

CRUZ, C. D. et al. **Modelos biométricos aplicados ao melhoramento genético**: v.1. 4a ed. Viçosa: UFV, 2012. 514p.

CRUZ, C. D. et al. **Modelos biométricos aplicados ao melhoramento genético**: v.2. 3a ed. Viçosa: UFV, 2014. 668p.

CRUZ, C. D. Genes Software - extended and integrated with the R, Matlab and Selegen. **Acta Scientiarum Agronomy**, v.38, n.4, p.547-552, 2016. Available from: <http://doi.org/10.4025/actasciagron.v38i4.32629>. Accessed: Oct. 20, 2022.

FACCO, G. et al. Sample size for estimating average productive traits of pigeon pea. **Ciência Rural**, v.46, n.4, p.619-625, 2016. Available from: <http://doi.org/10.1590/0103-8478cr20150852>. Accessed: Oct. 20, 2022.

FACCO, G. et al. Sample size for morphological traits of pigeonpea. **Semina: Ciências Agrárias**, v.36, n.6, p.4151-4164, 2015. Available from: <http://doi.org/10.5433/1679-0359.2015v36n6Supl2p4151>. Accessed: Oct. 20, 2022.

FERREIRA, D. F. **Estatística básica**. 2a ed. Lavras: UFLA, 2009. 664p.

SANTOS, H. G. et al. **Sistema Brasileiro de Classificação de Solos**. 5a ed. Brasília: Embrapa, 2018. 356p. Available from: <http://www.infoteca.cnptia.embrapa.br/handle/doc/1094003>. Accessed: Oct. 20, 2022.

SCHABARUM, D. E. et al. Sample size for morphological traits of sunn hemp. **Journal of Agricultural Science**, v.10, n.1, p.152-161, 2018a. Available from: <http://doi.org/10.5539/jas.v10n1p152>. Accessed: Oct. 20, 2022.

SCHABARUM, D. E. et al. Sample sufficiency for mean estimation of productive traits of sunn hemp. **Journal of Agricultural Science**, v.10, n.9, p.209-216, 2018b. Available from: <http://doi.org/10.5539/jas.v10n9p209>. Accessed: Oct. 20, 2022.

STORCK, L. et al. **Experimentação vegetal**. 3a ed. Santa Maria: UFSM, 2016. 200p.

TOEBE, M. et al. Sample size and linear relationships between *Crotalaria spectabilis* traits. **Bragantia**, v.76, n.1, p.45-53, 2017. Available from: <http://doi.org/10.1590/1678-4499.653>. Accessed: Oct. 20, 2022.