WEED INTERFERENCE IN THE MORPHOPHYSIOLOGICAL AND NUTRITIONAL CHARACTERISTICS OF ERVA-MATE (*Ilex paraguariensis*)

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Abstract

The erva-mate (*Ilex paraguariensis*) represents an alternative crop for agricultural exploitation in South Brazilian farms with potential to export the harvested product. However, there is scarce information about the interference caused by weeds on this crop. The objective of this work was to evaluate the interference of weed species on the morphophysiological and nutritional characteristics of erva-mate. The experiment was installed in greenhouse at the Federal University of Fronteira Sul (UFFS), *Campus* Erechim/RS, Brazil, in randomized blocks design, arranged in a 4 x 6 factorial scheme, with four replications. In factor A, weed species (*Urochloa plantaginea* – Alexandergrass, *Bidens pilosa* – Hairy beggarticks, *Ipomoea indivisa* – Morning glory and *Conyza bonariensis* – Hairy fleabane) were allocated, and in B the populations of these species competing with the erva-mate (0, 1, 2, 3, 4 and 5 plants per pot) were considered. The variables evaluated in erva-mate were sub-stomatal CO_2 concentration, photosynthetic rate, CO_2 consumed, stomatal conductance, transpiration rate, water use efficiency, height, stem diameter, leaf area and shoot dry mass. Concentrations of nitrogen (N), phosphorus (P), calcium (Ca) and magnesium (Mg) were also evaluated. Overall, all weed species and densities among the tested ones harmed the development of erva-mate plants, which was considered as low competitive with weeds.

Keywords

Urochloa plantaginea; Bidens pilosa; Ipomoea indivisa; Conyza bonariensis.

1. Introduction

Erva-mate (*Ilex paraguariensis* St. Hil.) is an arboreal species found in South America. It has 80% of its natural occurrence in Brazil [1], playing an important economic, social and even cultural role, mainly for smallholders.

Despite the importance of this herb for the economy, its average productivity of about 7.65 t ha^{-1} (510 @ ha^{-1}) is far below what could be effectively produced [2], an appropriate management system is applied.

Productivity losses caused by weeds in erva-mate are associated with competition [3], allelopathy, and indirectly to a reduction in product quality [4]. As naturally occurring species, weeds have genetic variability that guarantee them a high competitive ability [5], whether annual or perennial.

Martins [6] and Medeiros [7] verified that weeds significantly competed for nutrients with olive cultivars and eucalyptus clones. For the erva-mate, however, there are only a few studies about weed interference and their effects on the morphophysiological characteristics and nutrient accumulation by this crop.

Therefore, the hypothesis of this work is that weeds cause negative effects in morphophysiological characteristics and nutrient accumulation in erva-mate. To test the hypothesis, we aimed to evaluate weed interference on the morphophysiological characteristics and nutritional status of erva-mate.

2. Material and Methods

Experimental data most directly related to management practices recommendations are preferably obtained in additive experiments [8], where a single crop plant is maintained in a constant plant arrangement, while the weed is planted around in variable density [9].

The study was established in greenhouse, at the Federal University of Fronteira Sul (UFFS), *Campus* Erechim, from May to December 2015, in randomized blocks design, arranged in a 4 x 6 scheme, with four replications. In factor A, the weed species were allocated (*Urochloa plantaginea* - alexandergrass, *Bidens pilosa* – hairy beggarticks, *Ipomoea indivisa* – morning glory and *Conyza bonariensis* – hairy fleabane) and in B the populations of these species (0, 1, 2, 3, 4, 5 plants per pot) were allocated.

The erva-mate native genotype seedlings were selected from the same planting pot, with uniform size of 0.2m. The experimental unit consisted of a pot containing eight liters of Red aluminoferric Latosol (Hapludox), to where an erva-mate seedling was transplanted. The soil had the following chemical characteristics: $pH_{(water)}$ 5.2 (SMP index); 2% organic matter content; 6.4 mg dm^{-3} of P; and Ca, Mg and $CTC_{(pH\ 7,0)}$ of 12; 3.8 and 21.8 $cmol_c\ dm^{-3}$, respectively.

Transplanting was carried out in the first half of May 2015, and soil fertilization was accomplished according to the technical recommendations for the erva-mate [10], 15 days after transplanting (DATp), based on soil analysis. Seedlings were protected with 50% shading screen in the first 30 DATp.

Weeds were transplanted to plots, according to the treatments, 90 DATp of the erva-mate. This methodology aimed to approach field conditions, where the erva-mate is transplanted to weed-free areas, where weeds start to compete with the crop only about 90 days after crop planting. Irrigation was performed on demand to keep plots moist and avoid water stress.

At 160 DATp, when weeds entered the reproductive stage, the following variables were evaluated for erva-mate: sub-stomatal CO_2 concentration (*Ci*-μmol mol-1), photosynthetic rate (*A* - μmol $m^{-2}\ s^{-1}$), CO_2 consumed (*ΔC* - μmol mol^{-1}), stomatal conductance (*Gs* - mol $m^{-1}\ s^{-1}$), transpiration rate (*E* - mol $H_2O\ m^{-2}\ s^{-1}$) and water use efficiency (*EUA* - mol CO_2 mol H_2O^{-1}). These variables were determined in the middle third of the first fully expanded leaf of erva-mate. To evaluate the physiological variables, it was used an infrared gas analyzer (IRGA), ADC LCA Pro-SD (Analytical Development Co. Ltd, Hoddesdon, UK), being two blocks evaluated per day, between eight and ten o'clock in the morning, in order to maintain homogeneous environmental conditions during the analysis of each block.

At 180 DATp, the plant height (cm) of erva-mate was assessed with graduate ruler from the soil to the plant apical meristem. Stem diameter (mm) was determined with digital caliper 5 cm above soil surface. Leaf area (cm^2 $plant^{-1}$) was determined with portable leaf area meter model CI-203, BioScience Industries.

At 190 DATp, erva-mate plants were cut to soil level, packed in kraft paper bags and set for drying into forced air circulation oven at 65 ± 5 ºC for four days, to determine its dry mass (g $plant^{-1}$). Afterwards, only the mate leaves were ground and passed through a process of digestion with H_2O_2, H_2SO_4 for determination of nitrogen (N), phosphorus (P), calcium (Ca) and magnesium (Mg), according to procedures described by Tedesco [11].

The data were submitted to analysis of variance by the F-test, and when significant, data was explored either by regression analysis of by Tukey's test. All tests were performed at 5% probability ($p < 0.05$), and results in Tables and graphs were presented according to the respective significances in the ANOVA analysis.

3. Results and Discussion

Physiological parameters

The population of Alexandergrass and morning glory competing with erva-mate provided significant increase in *Ci* of this crop compared to the control (Figure 1A). The *Ci* increase in erva-mate when competing with weeds is related to depletion in environmental resources. The increase in *Ci* can indicate an attempt of the plant to reduce stress generated by the competition, since this is a physiological variable influenced by environmental conditions like water and light availability, among others [12], which partly command the stomatal opening/closure mechanism.

It was observed that erva-mate photosynthetic rate (*A*) decreased with increase in population of either hairy beggarticks or morning glory (Figure 1B). With three plants of hairy beggarticks per plot, a decrease of 54.6% in *A* of erva-mate was reported. When competing with morning glory, there was a linear reduction in *A* of the crop as the weed density increased, reaching about 61% reduction when plant proportion was 1:5 (1 crop plant, 5 morning glory plants) (Figure 1B). For the other weeds, there was effect of the competition in the rate of *A* in erva-mate, but there was no effect of weed density on this parameter; in other words, the presence of the weeds affected the rate of *A*, but not its density of occurrence (Figure 1B).

Concenço [13] highlighted that *A* (Figure 1B) is related to CO_2 consumed (Figure 1C) and increase in plant biomass. As the weed population increased, the level of competition also increased with reduction in the consumed CO_2 (ΔC) by erva-mate (Figure 1C), resulting also in lower *A* (Figure 1B). Similar results were observed by Matos [12], who reported reduction of *A* in coffee trees when under competition with *Mucuna aterrima* and *Urochloa decumbens* in densities of 28 and 84 plants m^{-2}, respectively.

The ΔC of erva-mate plants decreased drastically when this crop was infested with alexandergrass, hairy beggarticks or morning glory, with respective decreases in ΔC of about 80, 49 and 59%, but there was no impact of hairy fleabane (Figure 1C).

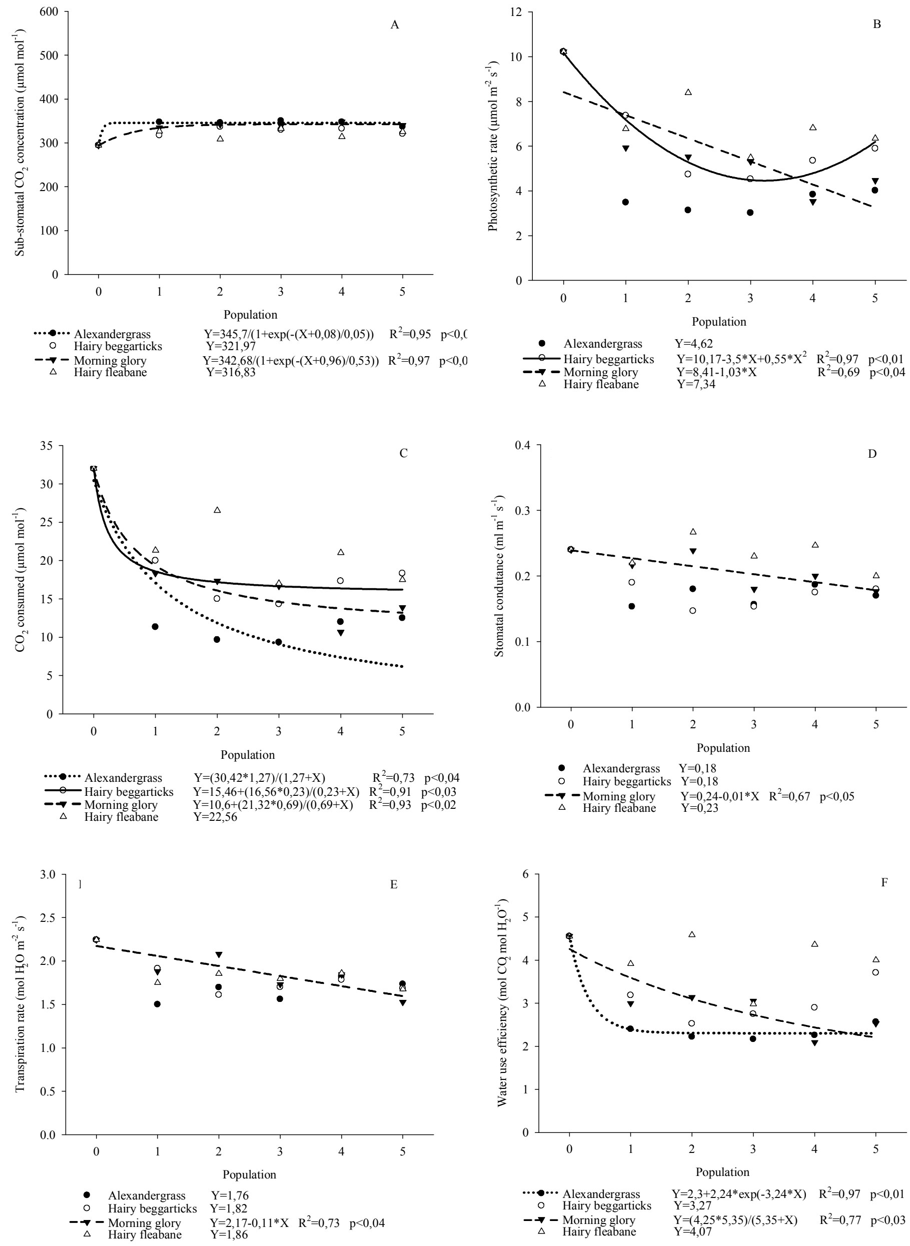


Figure 1. Sub-stomatic CO_2 concentration (*Ci* - µmol mol^{-1}) (A), photosynthetic rate (*A* - µmol m^{-2} s^{-1}) (B), CO_2 consumed (*ΔC* - µmol mol^{-1}) (C), stomatal conductance of water vapors (*Gs* - mol m^{-1} s^{-1}) (D), transpiration rate (*E* - mol H_2O m^{-2} s^{-1}) (E), water use efficiency (*EUA* - mol CO_2 mol H_2O^{-1}) (F), of mate in function of species and weed populations. UFFS-Erechim, 2016. ANOVA: For all variables, the interaction A x B is significant ($p < 0.05$).

For stomatal conductance (*Gs*) of erva-mate, a decrease of approximately 21% was observed when comparing the maximum infestation level of morning glory with the control free from infestation (Figure 1D); the reduction in *Gs* was linear to the increase in number of plants of this weed. The other weed species did not affect the erva-mate in a way that could be described by a regression. The water is only lost by transpiration while the stomata are open. Stomatal opening and closing depends on factors such as solar radiation, CO_2 level in the mesophyll, relative humidity, water potential, wind and growth substances of each species [12].

Similar results were found by [14], who reported decrease in *Gs* of coffee trees, when competing with *M. aterrima* or *U. plantaginea*. In this way, the reduction in photosynthetic rate of erva-mate (Figure 1B) caused by the coexistence with morning glory also affected directly the *Gs.*

Analyzing the transpiration rate (*E*) of erva-mate (Figure 1E), it was verified adjustment to the linear model for morning glory, similarly to what was observed for *Gs* (Figure 1D), where the decrease in *E* was of about 25% in the largest morning glory population (Figure 1E). Galon [12] observed a gradual reduction of *E* for glyphosate resistant and susceptible biotypes of *C. bonariensis,* similarly to what was observed in the present study.

The water use efficiency (*WUE*) of erva-mate (Figure 1F), presented exponential and hiperbolic models, respectively for alexandergrass and morning glory, as function of the increase in weed density. It was observed, compared to the control (weed free plots), decrease of approximately 49% in erva-mate *WUE* when under competition with alexandergass, independent of population.

The *WUE* reduction of erva-mate as function of its competition with weeds is related directly to a decrease in the CO_2 consumed (Figure 1C), transpiration (Figure 1E), stomatal conductance (Figure 1D) and photosynthetic rates (Figure 1B). The results corroborate with those found by Ferreira [15] when verifying that soybean in competition with *Bidens pilosa* and *Urochloa brizantha* affected considerably the crop *WUE.*

When analyzing the physiological data of erva-mate as a whole in function of the competition, it is observed that hairy fleabane and hairy beggarticks were the less competitive weed species, as they caused the smaller decreases in erva-mate physiological parameters (Figure 1).

Alexandergrass was more aggressive when competing with the erva-mate, because there was a significant decrease in most physiological parameters of the crop when under competition with this weed. From the population of four plants per pot onwards, it was noticed that morning-glory also started to interfere in erva-mate physiological traits (Figure 1).

Biometrical variables

Regarding plant height, there was no model adjustment to the data (data not shown). Plant height of erva-mate may have been affected not only by weed species and / or population density, but also by the natural variability in genetic traits inherent to the erva-mate native genotypes.

In plant densities of 4 : 1 alexandergrass / erva-mate plant proportion, there was a big decrease in erva-mate height. The other weed species caused also reduction in crop height (Table 1). In the largest population tested, hairy beggarticks, morning glory and hairy fleabane significantly affected the erva-mate height as function of the interspecific competition (Table 1).

Table 1. Height, steam diameter, leaf area and dry mass of the native mate genotype in competition with alexandergrass, hairy beggarticks, morning glory and hairy fleabane. UFFS-Erechim, 2016.

Specie	Height (cm)	Steam diameter (mm)	Leaf area (cm^2 pot^{-1})	Dry mass (g pot^{-1})	Height (cm)	Steam diameter (mm)	Leaf area (cm^2 pot^{-1})	Dry mass (g pot^{-1})
	Population 0				**Population 1**			
Alexandergrass	44.67 a[1]	7.24 a	692.96 a	14.93 a	46.50 a	7.36 a	781.22 a	15.45 a
Hairy beggarticks	44.67 a	7.24 a	692.96 a	14.93 a	46.25 a	6.79 a	553.97 b	10.41 b
Morning glory	44.67 a	7.24 a	692.96 a	14.93 a	46.67 a	6.70 a	725.78 a	15.48 a
Hairy fleabane	44.67 a	7.24 a	692.96 a	14.93 a	49.83 a	7.30 a	710.83 a	16.13 a
	Population 2				**Population 3**			
Alexandergrass	46.89 c	7.63 ab	769.77 b	16.97 a	45.67 b	7.16 ab	602.53 b	11.28 b
Hairy beggarticks	51.67 b	6.90 b	622.53 c	13.42 b	54.50 a	6.32 b	670.10 ab	11.64 b
Morning glory	56.78 a	7.53 ab	879.98 a	16.12 a	46.44 b	7.39 ab	632.95 b	12.35 ab
Hairy fleabane	54.00 b	7.77 a	788.46 ab	17.45 a	46.78 b	7.50 a	741.63 a	14.11 a
	Population 4				**Population 5**			
Alexandergrass	30.50 b	5.93 b	407.88 c	9.55 b	45.89 a	7.16 a	604.95 c	10.04 c
Hairy beggarticks	49.22 a	7.88 a	441.08 c	9.71 b	42.50 b	4.52 c	728.71 b	14.89 a
Morning glory	42.33 a	7.54 a	783.62 a	15.86 a	40.67 b	6.87 a	853.19 a	14.48 a
Hairy fleabane	48.00 a	7.21 a	529.09 b	13.85 a	40.67 b	6.18 b	416.39 d	11.54 b
C.V. (%)	**4.98**	**8.39**	**4.91**	**7.45**	**4.98**	**8.39**	**4.91**	**7.45**

[1] Means followed by the same lowercase letter in the column within each population do not differ by Tukey test ($p \leq 0.05$). ANOVA: For all variables, tfactors A and B were significant ($p < 0.05$). Comparison within factor B is not presented.

Similar results were observed by Martins [16] who verified that seven plants m^{-2} of *Amaranthus retroflexus, U. brizantha, B. pilosa or C. echinatus,* interfered differentially in seedling height of two olive cultivars (Arbequina and Ascolano) compared to the weed free control. Arbequina competing with *B. pilosa* and *A. retroflexus* presented lower seedling height, attributed to genetic differences between cultivars in terms of competitive ability [12].

The stem diameter of erva-mate in competition with hairy fleabane, was not affected up to three plants of the competitor. There was reduction of 2.8 and 17.4% in stem diameter when the erva-mate competed with four and five hairy fleabane plants, respectively (Table 1). This behavior is directly associated with the reduction in the physiological characteristics of erva-mate, as consequence of the competition. Toledo [17] also verified reduction in diameter of eucalyptus in competition with *U. decumbens.*

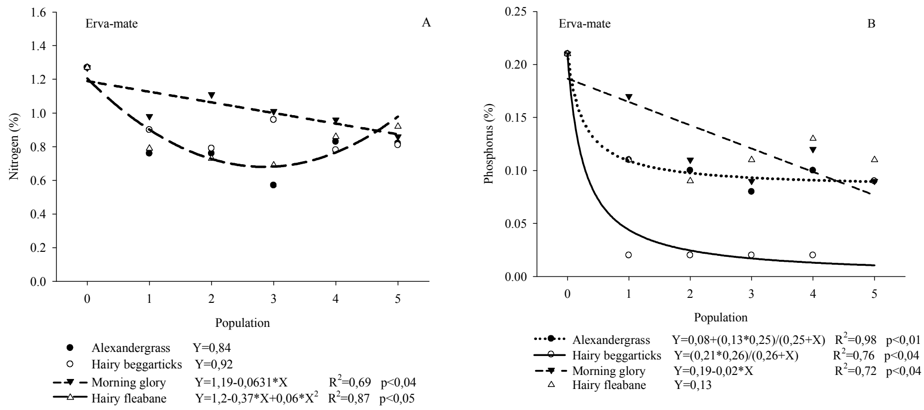
Regarding the erva-mate leaf area, the competition of two hairy fleabane plants with the crop caused increase of approximately 12% compared to the control plot with further reduction in this variable in higher competition densities, reaching about 41% reduction with five plants, compared to the control (Table 1). This instantaneous increase in leaf area at low competition levels is a natural mechanism that plants use to try to escape competition in its initial stages, before it becomes severe; the plant tries to increase its interception of light energy and overcome shading [18]. With increase in the population of hairy beggarticks, there was a proportional reduction in leaf area, except for population 5, where it tended to stabilize. Costa [19] found a reduction of 42% in eucalyptus leaf area after 60 days of competition with *Commelina benghalensis*.

When analyzing the erva-mate stem diameter, leaf area and dry mass in competition with the studied weeds, in general, there was a considerable reduction in dry mass 4 : 1 crop : hairy beggarticks proportion, and with three or more alexandergrass plants competing with the crop. Hairy fleabane affected the crop only at the 5 : 1 proportion, and the morning-glory was the least aggressive weed (Table 1).

Weed species also differ in the mechanism of competition [20]. Alexandergrass is highly competitive because of its efficiency in carbon fixation and water utilization. Hairy beggarticks is also very resistant to water stress [12], but its high competitive ability lies on the high number of individuals, and not in the individual competitive ability [12]. Morning-glory, besides water and nutrient competition, is highly competitive for light, since it has the ability to rapidly establish its vegetative structure and reproduce even under competition. Hairy fleabane, which has numerous offspring [12], similarly to hairy beggarticks, has a higher competitive aggressiveness when its population increases.

Nutrient content

Regarding the nitrogen content (N) in erva-mate plants, morning-glory provided a linear decrease of 5.3; 10.6; 15.9; 21.2 and 26.5% in leaf N content, respectively in the populations of 1, 2, 3, 4 and 5 plants, compared to the control treatment (Figure 2A); that is, as the weed population increased, there was a proportional decrease in N accumulation in leaves. Martins [12] found that in olive cultivars, there were smaller N accumulations in leaves under competition with *B. pilosa*, *C echinatus*, *A. retroflexus* and *U. brizantha*, compared to the control plot.



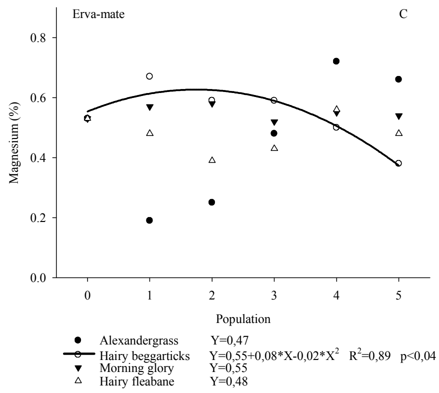


Figure 2. Nitrogen (%) (A), phosphorus (%) (B) e magnesium (%) (C) contents in leaf tissues in native mate genotype in function of species and population of weeds. UFFS-Erechim, 2016.
ANOVA: For all variables, the interaction A x B is significant ($p < 0.05$).

Comparing the N content within each plant population, it was possible to verify that morning glory was less competitive than the other species for N, as the erva-mate absorbed more N under competition with this weed compared to the other ones (Table 2). ANOVA: For all variables, the interaction A x B is significant ($p < 0.05$).

Table 2. Nitrogen (N), phosphorus (P), calcium (Ca) and magnesium (Mg) contents in foliar tissues in native mate genotype in competition with alexandergrass, hairy beggarticks, morning glory and hairy fleabane UFFS-Erechim, 2016.

Specie	%											
	N	P	Ca	Mg	N	P	Ca	Mg	N	P	Ca	Mg
	Population 0				**Population 1**				**Population 2**			
Alexandergrass	1.27 a[1]	0.21 a	0.17 a	0.53 a	0.76 c	0.11 a	0.32 a	0.19 b	0.76 b	0.10 a	0.34 a	0.25 c
Hairy beggarticks	1.27 a	0.21 a	0.17 a	0.53 a	0.90 ab	0.02 b	0.29 a	0.67 a	0.79 b	0.02 b	0.27 ab	0.59 a
Morning glory	1.27 a	0.21 a	0.17 a	0.53 a	0.98 a	0.17 a	0.30 a	0.57 a	1.11 a	0.11 a	0.33 a	0.58 a
Hairy fleabane	1.27 a	0.21 a	0.17 a	0.53 a	0.79 bc	0.11 a	0.25 a	0.48 a	0.73 b	0.09 a	0.21 b	0.39 b
	Population 3				**Population 4**				**Population 5**			
Alexandergrass	0.57 b	0.08 b	0.21 a	0.48 ab	0.83 ab	0.10 b	0.42 a	0.72 a	0.82 a	0.09 a	0.32 a	0.66 a
Hairy beggarticks	0.96 a	0.02 c	0.29 a	0.59 a	0.78 b	0.02 c	0.29 b	0.50 b	0.81 a	0.09 a	0.21 b	0.38 b
Morning glory	1.01 a	0.09 ab	0.22 a	0.52 ab	0.96 a	0.12 ab	0.27 b	0.55 b	0.86 a	0.09 a	0.24 b	0.54 ab
Hairy fleabane	0.69 b	0.11 a	0.21 a	0.43 b	0.86 ab	0.13 a	0.24 b	0.56 b	0.92 a	0.11 a	0.19 b	0.48 b
C.V. (%)	**8.98**	**15.75**	**20.07**	**15.02**	**8.98**	**15.75**	**20.07**	**15.02**	**8.98**	**15.75**	**20.07**	**15.02**

[1] Means followed by the same lowercase letter in the column within each population do not differ by Tukey test ($p \leq 0.05$).

For phosphorus (P) content, overall, alexandergrass and hairy beggarticks caused reduction of approximately 57 and 92% in P content compared to the control (Figure 2B). Comparing weed species within each population, it was observed in general that hairy beggarticks was the most competitive one by the nutrient, and hairy fleabane and morning glory were the least competitive ones by P (Table 2). The reduction in P content in erva-mate as function of the weed populations can be explained by the fact that P has low mobility in soil. Weeds develop faster compared to erva-mate, and their roots are more aggressively distributed in soil, providing greater absorption of less mobile elements as P.

For Mg content in erva-mate, it was generally observed that the hairy beggarticks, morning glory and hairy fleabane were more competitive by Ca and Mg in the larger populations (Table 2), as the erva-mate content for these nutrients was reduced under competition with these weeds. These results corroborate with those found by [21], who verified that coffee leaves collected from plants free of competition had P concentration 19.5% higher than leaves coming from plants under competition; in the same study, Mg contents were reduced as the weed population increased.

In general, the competition between the erva-mate and the weeds early in the crop development may compromise the entire crop cycle. It was verified that weeds caused reduction on erva-mate metabolism.

4. Conclusion

Morning glory and alexandergrass were the weed species which most consistently affected the physiology of the primary metabolism of erva-mate plants;

Overall, all weed species and densities among the tested ones harmed the development of erva-mate plants, which was considered as low competitive with weeds. Thus, under field conditions, erva-mate plants should have a ring around it where all weeds need to be controlled. The minimum ringing size allowed for weed elimination should be studied in future works.

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