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Soil Seed Bank Phytosociology in No-Tillage Systems in the Southwestern Amazon Region

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Abstract

Understanding the ecological dynamics of weed populations in no-tillage systems is important to establish strategies for integrated weed control capable of increasing agroecosystem sustainability. This study sought to evaluate the effect of succession systems on the seed bank in a no-tillage system. The effects of fifteen succession systems, composed of seven grasses, seven *Leguminosae*, and a fallow condition, were evaluated on the seed bank at two soil depths (0 to 10 and 10 to 20 cm). The species found in the seed bank were quantified and identified by species and family. The precision of sampling, density, dominance, the indices of diversity of Simpson and Shannon-Weiner; index of sustainability; analysis of groupings of dissimilarities; and the value of importance of each species were calculated. High weed diversity was observed; 29 species were counted, including members of 12 different families. The highest expression of weeds was observed at soil depths of 0 to 10 cm. The Simpson and Shannon-Weiner coefficients indicated high diversity in both systems of succession. The index of sustainability did not indicate significant alterations in the different systems of succession. The coefficients of confenetic correlation were 0.74% and 0.82% for the 0 - 10 and the 10 - 20 cm soil depths respectively. It is concluded that there is a high diversity of weeds in the agroecosystems of Amazonia, and different cover crops promote modifications in the community and expression of the weeds' seed bank.

Keywords

Sorghum sudanense, Phytosociology, Integrated Management, Sustainability, Brazilian Amazonia

1. Introduction

Agricultural areas are a large pool of weed seeds, which are usually a significant problem for farmers, because seeds promote weed infestation in the long term, even without introduction of new propagules in the area. This is due to the characteristic survival strategies of weeds, which produce many seeds and make use of mechanisms of dormancy in the environment [1]. These mechanisms lead to greater resistance to management strategies, and increase the competitive ability of weeds over the crop plants, which produce fewer seeds, with less dispersion capacity, and shorter seed longevity [2].

Weeds in agricultural areas cause various constraints, such as reduced agricultural efficiency, increased production costs, reduced product quality, reduced commercial value and hampered harvesting operations [3]. Among weed control practices, the chemical method is the most common, through the application of herbicides [4]. However, this isolated operation is no sufficient to control all weed interference with crops and may generate a higher latent risk of environmental contamination. Currently, the goal is to use integrated weed-control practices that are more sustainable, among which is the use of phytosociological tools such as seed banks studies [5].

The main methods used to study seed banks are seedling flotation and emergence [6]. The flotation technique infers the potential of seed diversity, viable or not, occurring in a target environment. The phytosociological study of plant emergence can simulate geo-environmental conditions to evaluate expression of the viable seed bank under specific environmental conditions. Thus, the seedling emergence method, in which soil samples are collected from the study area, has been the most widely used. The method is capable of detecting seed emergence flow when exposed to favorable environmental conditions [7]. In the seed bank survey, it detects the fraction of seeds whose emergence benefits from the established conditions. This results in more accurate results with greater correlation with field conditions [8].

In agricultural areas, seed bank estimates are important in predicting probable weed infestations; they enable a better understanding of the dynamics of species in different environmental and management situations and, consequently, the choice of more rational programs of weed control [9]. The implementation of sustainable management programs, such as no-tillage systems, may result in lower emergence of weed seeds, through minimal soil rotation, presence of live and straw cover, and crop rotation or succession [10]. According to these principles, the production of straw from cover crops can provide a physical impediment to the germination of weed seeds. In addition, the release of allelopathic substances through root exudation, volatilization, leaching, or straw decomposition, can inhibit the growth and development of invasive species. Cover plants have particular effects on ecological dynamics and the occurrence of weeds; promoting the cultural control of these invasive species can lead to reductions in herbicide use [11]. The objective of this paper was to evaluate the effect of pro-

duction systems with several cover plants on the soil seed bank under a no-tillage regime in the southwestern region of Amazonia.

2. Methods and Materials

The experiment was carried out under controlled conditions in a greenhouse at Embrapa (Brazilian Agricultural Research Company), located at Porto Velho, Rondonia. The soil used was collected in April 2015 from an experimental field area located at coordinates 08˚47'42"S latitude, 63˚50'45"W longitude, and 95 m a.s.l.

The soil collection area was previously cultivated as pasture (Urochloa brizantha cv. Marandú) for over 18 years. In 2008, the area was managed under a no-tillage regime with soybean crops. Since 2014, the area has occupied with an experiment evaluating different off-season cover crops in succession to summer maize: *Cajanus cajan*, *C*anavalia *ensiformis*, *Crotalaria juncea*, *Crotalaria ochroleuca*, *Crotalaria spectabilis*, *Mucuna aterrima*, *Mucuna cinereum*, *Pennisetum glaucum*, *Sorghum bicolor*, *Sorghum sudanense*, *Urochloa brizanha* cv. Xaraés, *Urochloa brizantha* cv. Piatã, *Urochloa ruziziensis*, and *Zea mays*, and kept fallow (without cover crop cultivation) as a control treatment [12]. The experimental field plots under the different cover land uses were 50 m^2 each (10 m × 5 m).

Prior to cover crops implantation in April 2014, sampling and analysis was performed to characterize chemical attributes of the soil (Table 1). The soil is classified as typical dystrophic Ferralsol, with a clayey texture.

Table 1. Chemical attributes of the soil of the experimental area with different soil uses. Porto Velho, Rondonia, 2014.

Depth	pH	OM	P	K	Ca	Mg	H + Al	Al	CEC	AS	SB
	H$_2$O	kg^{-1}	mg·dm^{-3}			cmol$_c$·dm^{-3}				%	
0 to 10	5.5	39.8	8.2	0.2	2.9	2.1	7.8	1.6	15.6	27.8	30.3
10 to 20	4.9	33.8	6.7	0.1	2.0	1.6	10.2	2.0	15.4	33.4	25.3

pH in water 1:2.5; organic matter (OM); P and K determined by Mehlich 1 methods: Ca, Mg and Al exchangeable extraction with KCl at 1 mol·L^{-1}.

Soil sampling for seed bank evaluation was performed after the 2015 maize harvest, after two years of cover crop effect, and one maize crop season for grain production. A systematic sampling was adopted by conglomerates, using two lines transverse to rows of maize. The sampling locations were spaced 1 m apart, excluding the border and corn rows. Collections were performed in the different treatments of soil cover, using a sample composed by four subsamples, at depths of 0 - 10 cm and of 10 - 20 cm. Each sub-sample was collected with the aid of a Dutch sampler soil (4.5 cm diameter).

The soil samples were dried, homogenized and poured into Styrofoam trays (900 cm^3) randomly distributed on a workbench in the greenhouse. The trays were previously perforated, identified and 2 cm of autoclaved sand added to each to facilitate water drainage. Eight control trays were included, each con-

taining only autoclaved sand to facilitate monitoring of external contamination.

The greenhouse used in this study has an automated irrigation sprinkler system that applied 7 mm of water over 30 minutes on a daily basis. The seedling flow measurement was performed in each tray when the seedlings received a number and a codename according to their morphotype. After six months of growth, the seedlings were collected and stored in paper bag by species and according to treatment condition. These samples were dried by oven with forced air circulation at 65˚C for further determination of dry matter mass. After two months, without new emergency flow, the soil was stirred and maintained with no irrigation for one month, after which the previous water schedule resumed.

The identification of the seedlings was carried out by comparison to the specialized literature [13] [14] [15], and by consultation with virtual herbariums [16] [17] [18] [19]. The emergence of seedlings ended at approximately 10 months since the beginning of germination. The number of viable and non-dormant seeds was obtained by summing the emerged seedlings in each sample during the whole experimental period. All of the weeds which grew from the seed bank are presented, that is, the expression of the potential of the seed bank and the wide diversity of species present in the soil in agricultural systems in the Amazon region.

Weed quantification data and their respective dry matter were processed aiming to determine phytosociological analyzes by script proposed and developed by [20] in R statistical analysis software [21].

The accuracy of the samples was determined from data on individual samples and their respective dry matter masses, obtained in each tray was evaluated by treatment, according to [22]:

$$Pr = \frac{1}{s^2 \left(\text{sample means} \right)} \tag{1}$$

where: s^2 = variance of the sample means.

The data were normalized [20] to 1, with this considered the minimum precision value for confident and high-precision sampling.

The absolute infestation was obtained by the numbers of individuals (density) and their respective dry matter mass (dominance), by sampled area, providing the results per m^2.

The cover land uses were also intra-analyzed for weed diversity by the Simpson (D) and Shannon-Weiner Indices [22], as well as by the sustainability index proposed by Shannon-Weiner (H') and coefficient of sustainability proposed by McManus and Pauly [23], as follows:

$$D = 1 - \sum n_i * \frac{n_i - 1}{N * \left(N - 1 \right)} \tag{2}$$

$$H' = \sum \left(p_i * \ln \left(p_i \right) \right) \tag{3}$$

$$SEP = \frac{Hd'}{H'} \tag{4}$$

where D = Simpson diversity index; H' = Shannon-Weiner diversity index (density); ni = number of individuals of species "i"; N = total number of individuals in the sample; pi = proportion of individuals in the sample of species "i"; SEP = Shannon-Weiner Evenness Sustainability Index; and Hd = dominant Shannon-Weiner diversity index.

Then, the succession systems were grouped by cluster analysis in each soil depth and compared by the coefficient of Jaccard binary asymmetric similarity, considering the density, frequency and dominance of weeds in treatments [24] [25]. From these data, we generated the matrix of similarity and, from this. The matrix of dissimilarity was elaborated.

$$\text{Traditional formula} : J = \frac{c}{a + b - c} \tag{5}$$

$$\text{Modified formula} : J = \frac{c}{a + b + c} \tag{6}$$

where J = Jaccard's coefficient of similarity; a = number of species in the area "a"; b = number of species in area "b"; c = number of species common to areas "a" and "b". The dissimilarities are then obtained by $Di = 1 - J$, where Di = dissimilarity (distance) of Jaccard; and J = Jaccard similarity.

3. Results and Discussion

Twenty-nine different species were observed, distributed among 12 botanical families (Table 2). The total number of individuals (736), were split fairly evenly between the two sampling depths; 377 from 0 - 10 cm, and 359 from 10 - 20 cm. The families found in the study areas were *Cyperaceae*, *Euphorbiaceae*, *Fabaceae*, *Lamiaceae*, *Loganiaceae*, *Malvaceae*, *Onagraceae*, *Poaceae*, *Pteridaceae*, *Rubiaceae*, *Scrophulariaceae*, and *Verbenaceae*. There was no external contamination of the trays, based on the lack of germination in the control trays.

The sampling accuracy for the two soil depths was verified. Based on the density data, the precision varied from 2.62 for the plots cultivated with *Urochloa brizanha* cv. Xaraés, to 18.28 for maize grown in off-season, at the 0 - 10 cm sampling depth. For the greater depth, from 10 - 20 cm, the value of 3.15 for *Crotalaria juncea* was compared to 281.06 for *Urochloa ruziziensis*. On the other hand, the amplitude of the precision, considering that the dominance was superior to the one observed for density, comprised 64.67 and 3.45 for *Mucuna cinereum* and *Mucuna aterrima*, respectively, at the depth of 0 - 10 cm. In the lower layer, the 10 - 20 cm zone, the observed extremes were 50.83 for *Canavalia ensiformis* and 2.55 for *Mucuna aterrima* (Table 3).

The number of weeds found varied with soil use systems. It was observed a greater number of individuals occurring in the depth of 0 to 10 cm, with evident predominance for the treatments *Crotalaria juncea*, *Crotalaria ochroleuca*, *Crotalaria spectabilis*, *Pennisetum glaucum*, *Mucuna aterrima* and fallow. The lowest values of density were verified in *Sorghum sudanense* (55 seedlings m^{-2}) and in *Mucuna cinereum* (56 seedlings m^{-2}), at depth of 0 to 10 cm (Figure 1),

Table 2. Families, scientific and common names of weed species found in the seed bank. Porto Velho, Rondonia.

Families	Species	
	Scientific name	Common name
Cyperaceae	*Cyperus laxus*	Tiririca-do-brejo
	Fimbristylis miliacea	Falso-cominho
	Kyllinga squamulata	*Capim*-de-uma-*só-cabeça*
	Rhynchospora nervosa	Capim-estrela
	Fimbristylis dichotoma	Falso-alecrim
	Cyperus esculentus	Tiriricão
	Kyllinga odorata	Capim-cheiroso
Euphorbiaceae	*Sebastiania corniculata*	*Guanxuma-de-chifre*
	Phyllanthus orbiculatus	Quebra-pedra-oval
	Phyllanthus urinaria	Quebra-pedra-pequeno
	Acalypha arvensis	Algodãozinho
Fabaceae	*Senna obtusifolia*	Fedegoso
	Mimosa pudica	Dormideira
Lamiaceae	*Salvia* sp.	Salvia
Loganiaceae	*Spigelia anthelmia*	Lombrigueira
Malvaceae	*Gaya pilosa*	*Guanxuma*
	Sida rhombifolia.	Vassourinha-de-relógio
Onagraceae	*Ludwigia affinis*	Cruz-de-malta
Poaceae	*Paspalum virgatum*	Capim-navalha
	Digitaria sanguinalis	Capim-amargoso
	Eleusine indica	Capim-pé-de-galinha
	Urochloa plantaginea	Capim-marmela
	Panicum maximum	Panicum
Pteridaceae	*Pityrogramma calomelanos*	Samambaia
Rubiaceae	*Spermacoce latifolia*	Erva-quente
	Spermacoce verticillata	Vassourinha-de-botão
Scrophulariaceae	*Lindernia dubia*	Agriãozinho-tapete-da-água
	Lindernia diffusa	Orelha-de-rato
Verbenaceae	*Stachytarpheta cayennensis*	Gervão-azul
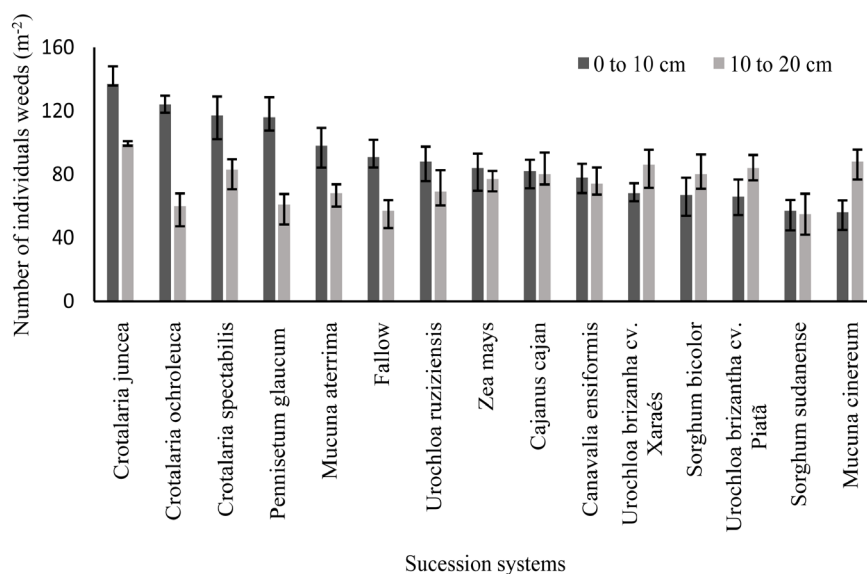
Figure 1. Total number of individuals (m^{-2}) as a function of the evaluated off-season crops use. Porto Velho, Rondonia.

Table 3. Precision of sampling by density (Pr.De) and dominance (Pr. Do.) in function of the different uses of the soil in two soil layers in Porto Velho, Rondonia.

| Treatments	Depths (cm)			
	0 - 10		10 - 20	
	Pr. De.	Pr. Do.	Pr. De.	Pr. Do.
Cajanus cajan	10.27	11.03	18.83	6.86
Canavalia ensiformis	9.56	33.78	5.41	50.83
Crotalaria juncea	17.7	5.13	3.15	30.85
Crotalaria ochroleuca	3.66	5.4	29.73	14.26
Crotalaria spectabilis	5.66	10.54	14.68	23.78
Mucuna aterrima	5.43	3.45	44.84	2.55
Mucuna cinereum	12.57	64.67	11.34	9.44
Pennisetum glaucum	7.28	6.81	19.65	5.44
Fallow	8.41	11.78	11.89	10.6
Sorghum bicolor	6.06	20.37	3.25	3.68
Sorghum sudanense	5.92	14.17	25.33	11.6
Urochloa brizanha cv. Xaraés	2.62	5.77	19.07	17.04
Urochloa brizantha cv. Piatã	7.02	7.87	5.65	3.27
Urochloa ruziziensis	13.7	21.76	281.06	6.86
Zea mays	18.28	9.2	56.89	4.27

ThPr.De.: sampling accuracy based on density data; Pr.Do.: Sampling accuracy based on dominance data.

proving to be alternatives for reducing the occurrence of weeds in no-tillage systems for grain production in the region. These results corroborate those found by [26] in the savanna in central Brazil, near Paraguay, where the genus Mucuna provided the best influence in weed suppression and the soil coverage. In turn, [27] found a high suppressive effect of *Sorghum sudanense*, generating efficient weed control in central tropical region of Brazil.

The treatments comprising the genus Crotalaria provided the highest weed density (137 seedlings m^{-2}) in the samples from 0 - 10 cm. It should be noted that these are crop species have often been used in agricultural areas in Brazil, especially areas producing soybean crops under no-tillage cultivation [28]. Crotalaria are used as an alternative for the cultural control of soil nematodes, a pest which are expensive to control and lead to negative impact on crop yields [29]. Therefore, adjustments and improvements in phytotechnical management, with increased seeding density, use of high quality physiological seeds, increasing the number of plants per row (stand), with smaller row spacing, among other practices, may represent important strategies to avoid the propensity of these species to grow as high-density weeds.

Crotalaria are plants that have a high sensitivity to photoperiod. When sown in the off-season (*i.e.* February or March) in the Brazilian savanna region, they have a faster flowering induction than when sown at the beginning of the rainy season (*i.e.* October) in the savanna or southern regions of the Amazon [30]. Thus, the plants of this type sown in the off-season have smaller size and a lower leaf-area index than plants sown in the regular season, generating lower production of biomass, of allelochemicals per unit area, and less soil coverage, which may decrease the emergence of seed bank seedlings that are photoblastic positive, or that show sensitivity to allelochemicals from the straw, or low competitiveness with straw on the ground.

The yield of dry matter mass (dry biomass) of weeds was observed to be significantly higher in the in the 0 - 10 cm layer relative to the deeper soil layer in all soil use systems (**Figure 2**).
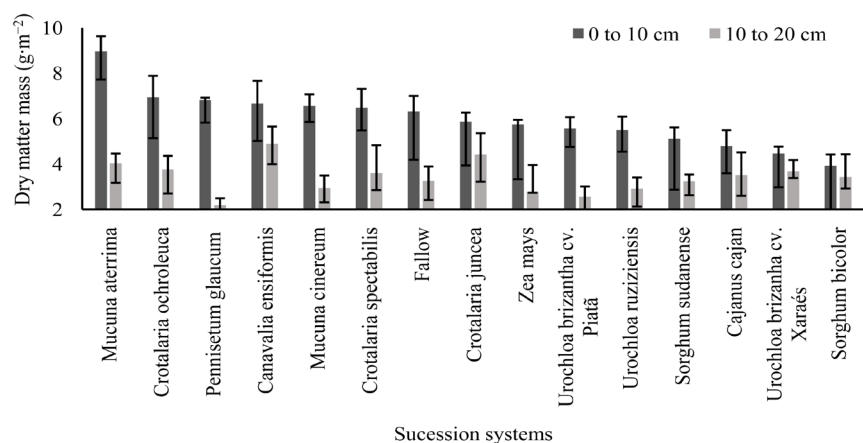
Figure 2. Dry matter biomass ($g \cdot m^{-2}$) of the weed shoot community according to the evaluated land use treatments. Porto Velho, Rondonia.

The higher biomass yield from the 0 - 10 cm layer reflects the greater potential for weed infestation in the superficial layers of areas cultivated under no-tillage systems, since the subsurface layers are not periodically revolved. According to [31], weed seeds are more concentrated in the superficial layers of 0 - 10 cm, while the population of seeds of 10 - 20 cm in perennial agricultural systems does not present as intense a dynamic of soil use as in grain production.

The highest biomass accumulation of dry matter was observed for the treatment composed by *Mucuna aterrima* at depth of 0 - 10 cm. This cover crop has a high and rapid production biomass and ground cover, and is a well-known species in allelochemical presence [32]. However, it presented one of the highest density values for *Lindernia diffusa*, an annual herbaceous species of floristic occurrence on temporarily flooded lands [14]. Regardless of the succession system evaluated, *Lindernia diffusa* was the most persistent plant under the conditions evaluated. This can be justified by the irrigation level in the greenhouse, which likely favored the emergence of weeds that develop in moist soil due to the high rainfall, a very common condition in the Amazon region. It is known that water is a limiting factor for plant development and its lack, as much as excess, affects plant growth, development, health and production [33].

The diversity coefficient *D* is a dominance index that varies from 0 to 1, and indicates the probability of two randomly selected individuals belonging to the same species. The greater the value of *D*, the lower the diversity of the population. The diversity coefficient *H* varies from 1.5 to 3.5, and higher values of H' indicate a greater diversity of species in an area; *H* is more influenced by the occurrence of rare species [34] [35]. In this study, at depths of 0 - 10 cm values of D ranged from 0.75 (*Crotalaria juncea*) to 0.90 (*Urochloa ruziziensis* and *Sorghum bicolor*), and *H* ranged from 2.49 (*Crotalaria juncea*) to 3.41 (*Urochloa ruziziensis*) (Table 4). Samples from depths of 10 - 20 cm had D values ranging from 0.77 (*Crotalaria juncea*) to 0.89 (*Crotalaria ochroleuca*, *Sorghum sudanense* and *Crotalaria spectabilis*) and *H* values from 2.41 (*Urochloa brizantha* cv. Piatã) to 3.25 (for *Crotalaria ochroleuca*, *Sorghum sudanense* and *Crotalaria spectabilis*).

Higher diversity coefficients are usually associated with a greater distribution of occurrence of spontaneous species. Thus, both coefficients indicated high diversity in all succession systems. According to [36], diversity is not directly related to higher levels of infestation. In this context, the lowest level of diversity was observed in *Crotalaria juncea*; however, it was the succession system that presented the higher density of individuals of the same species, so, greater dominance and less weed diversity.

Evaluating the sustainability index of Shannon-Weiner Evenness (SEP), no significant influence was verified between the different systems of land use. Authors have found SEP values between 0.99 and 1.33 studying succession systems of soybean cultivation with winter oilseed cover crops, and considered values below 1.50 ideal for systems considered sustainable [37]. In this work, the values obtained ranged from 0.85 (*Crotalaria juncea*) to 1.11 (*Cajanus cajan* and *Zea*

Table 4. Weed species diversity as a function of evaluated land uses. Porto Velho, Rondonia.

| Land use | Depths (cm) | | | | | |
| | 0 - 10 | | | 10 - 20 | | |
	D	H'	SEP	D	H'	SEP
Cajanus cajan	0.88	2.72	1.11	0.82	3.02	0.99
Canavalia ensiformis	0.85	3.09	1.09	0.82	3.02	0.98
Crotalaria juncea	0.75	2.49	0.85	0.77	2.55	0.98
Crotalaria ochroleuca	0.78	2.88	1.01	0.86	3.25	0.98
Crotalaria spectabilis	0.87	3.38	1.01	0.89	3.25	0.99
Mucuna aterrima	0.79	2.92	1.11	0.85	2.29	0.98
Mucuna cinereum	0.85	3.28	1.11	0.82	2.93	1.02
Pennisetum glaucum	0.81	2.99	1.05	0.79	2.66	1.12
Sorghum bicolor	0.99	3.33	1.00	0.88	2.82	0.92
Sorghum sudanense	0.85	3.15	0.98	0.86	3.25	1.02
Urochloa brizanha cv. Xaraés	0.84	2.99	0.92	0.84	3.11	1.15
Urochloa brizantha cv. Piatã	0.85	3.21	0.94	0.78	2.41	1.05
Urochloa ruziziensis	0.99	3.41	0.99	0.84	2.98	1.00
Zea mays	0.85	3.11	1.11	0.82	2.69	1.02
Fallow	0.87	3.22	1.02	0.79	2.68	0.84
Average	0.85	3.08	1.02	0.83	2.86	1.00

Simpson (*D*) and Shannon-Weiner (H') diversity indexes and Shannon-Weiner Evenness sustainability index (SEP).

mays) at depths from 0 - 10 cm, and the values for 10 - 20 cm ranged from 0.84 (fallow) to 1.15 (*Urochloa brizanha* cv. Xaraés). These results serve as indicators of the trend of the behavior of succession systems to the adopted management, and possibly indicate a correct direction of the use of cover plants as a tool to promote long-term sustainability.

The coefficient of cophenetic correlation was 0.74% for samples from 0 - 10 cm (Figure 3). When evaluating the pre-planting and post-emergence periods of soybean seedlings, [37] found cophenetic correlation values of 0.75% and 0.78%, respectively, which the authors did not consider sufficient for reliability.

A cophenetic correlation equal to 0.82% was observed for the 10 - 20 cm soil layer depth (Figure 4). The cophenetic correlation coefficient must be equal to or greater than 0.85, indicating that the grouping adequately reflects the original data [25]. However, the cut in the dendrogram at 0.46 (0 - 10 cm deep) and 0.35 (10 - 20 cm deep) suggests that the ideal number of groups formed are 2 and 3, respectively.

Spermacoce latifolia (hot herb) and *Spermacoce verticillata* (button-broom) are known weeds of agronomic importance in the region, with great potential

Cluster analysis for land use systems
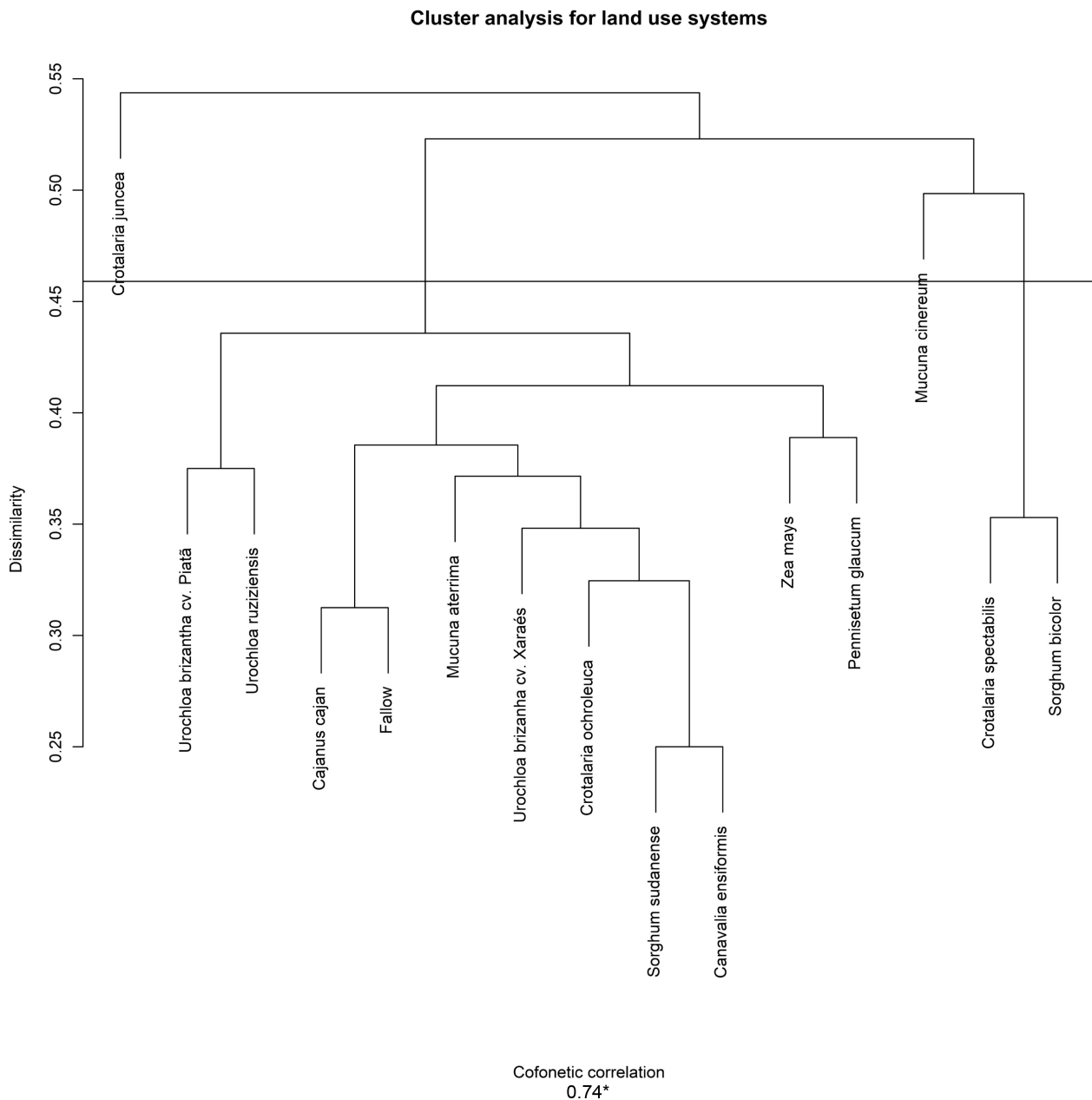
Figure 3. Cluster analysis for different land use uses, at the 0 - 10 cm sampling depth, based on the dissimilarity matrix by Jaccard's coefficient and grouping accomplished by unweighted pair group method with arithmetic mean method. Porto Velho, Rondonia.

for damaging cash crops (especially grains) through competition or negative allelopathy; both were observed in this study. Importantly, both *Spermacoce* present high levels of shade tolerance in conjunction with the annual Rubiaceae family [14], which has a high competitive capacity with annual crops, with preference for acidic soils [38]. S*permacoce verticillata* in particular is an invasive plant with high infestation capacity in arable areas, and is a difficult species to control [39].

Understanding the dynamic of the soil seed bank, the weed community and weed control strategies is key to effective weed control in a no-tillage system.

Cluster analysis for land use systems
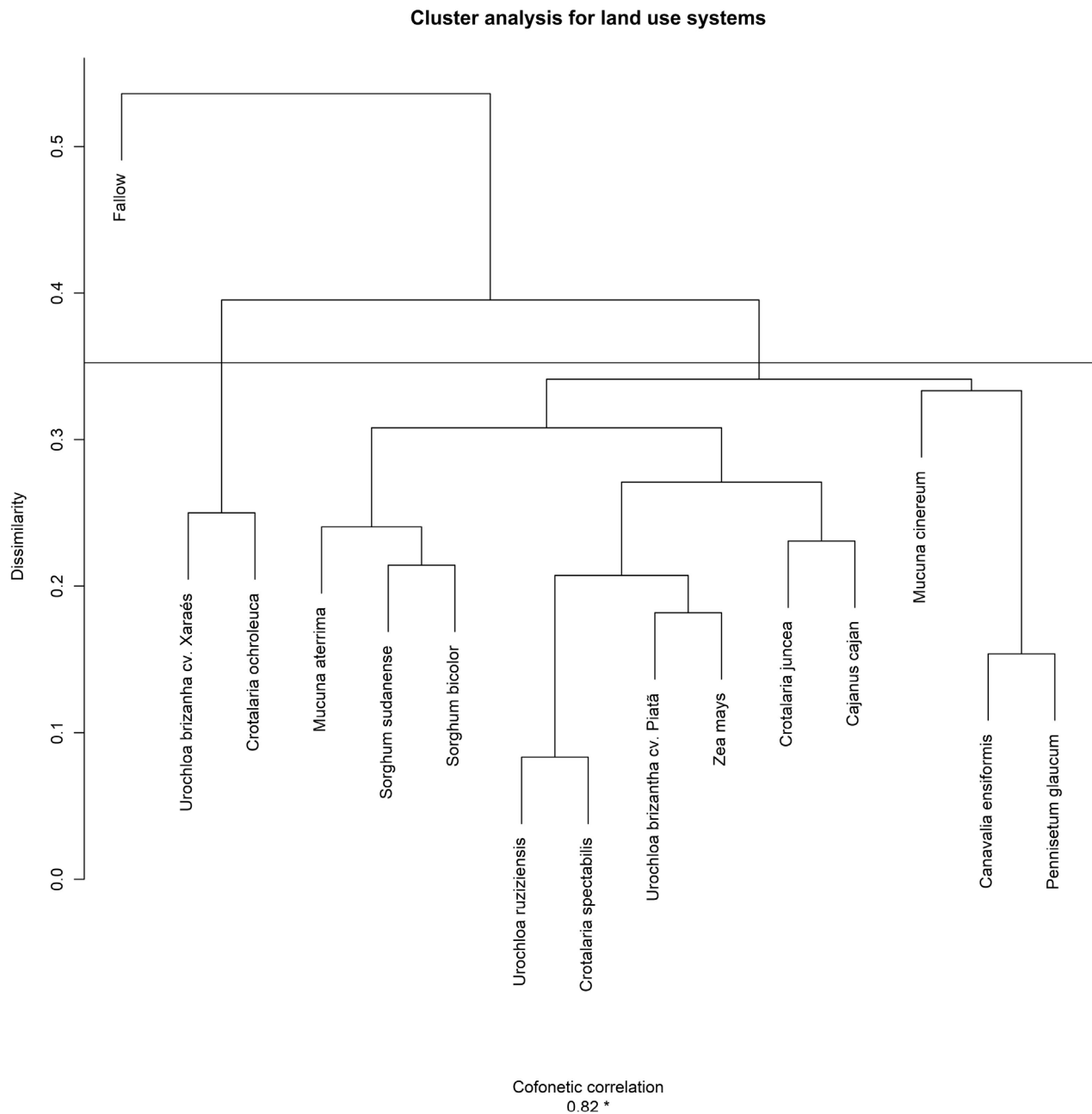
Figure 4. Cluster analysis for different land use uses, at the 10 - 20 cm sampling depth, based on the dissimilarity matrix by Jaccard's coefficient and grouping accomplished by unweighted pair group method with arithmetic mean method. Porto Velho, Rondonia.

Changes in farming practices can provide great benefits to the agroecosystem, with positive effects on weed control. In no-tillage regimes, the choice of cover crops plays a fundamental role, providing straw to protect the soil and crowd out weeds.

4. Conclusion

There is a predominance of dry biomass accumulation in the depth of 0 to 10 cm, an expression of the deleterious potential of the seed bank in soils managed

under no-tillage techniques in the southwest region of Amazonia. Crotalaria species, as cover crops, present highest weed density and a high weed diversity in the 0 to 10 cm soil depth layer compared to the other cover crops. The use of the sorghum bicolor and Urochloa spp. in the winter, as cover crops, provides suppression of weeds. These findings reinforce the need for phytosociological surveys to define the best integrated control practices for the region.

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