INFLUENCE OF MAIZE HERBICIDES ON WEED SEED BANK DIVERSITY IN A HUMID FOREST AGRO-ECOLOGY OF SOUTH-EASTERN NIGERIA

[1]Udensi Ekea Udensi, [2]Ikechukwu Asogwa and [2]Ochekwu Benard

[1]Department of Crop and Soil Science, University of Port Harcourt, P. M. B 5323, Port Harcourt, Nigeria

[2]Department of Plant Science and Biotechnology, University of Port Harcourt, P.M. B 5323, Port Harcourt, Nigeria

Corresponding author's email: ueudensi@yahoo.co.uk; udensi.udensi@uniport.edu.ng

ABSTRACT

A Screen house study was carried out to determine the weed seed bank diversity following pre-emergence weed control in maize (*Zea mays* L.) farm. The Soil used for the study was collected from Faculty of Agriculture Research and Teaching farm (04^{o} 90 889`N and 006$^{o}$ 92 240` E a.s.L 2,7 m) of the University of Port Harcourt, Rivers State, Nigeria (04^{o} 54 538'N and 006^{o} 55 329'; a.s.l. 17m) . The weed seed bank study was conducted at the Green house, Centre of Ecological study University of Port Harcourt. Soil for this study was collected at three depths-0-5cm, 5-10cm and 10-15cm respectively. Nine core samples were collected in a diagonal transect from each maize plot in a manner that depicts a "Z" and bulk for each soil depth. The soil for the study was collected 40 weeks after the pre-emergence maize herbicides treatment from previously treated maize weed control trial. The herbicide treatments were as follows: Primextra Gold at 1.5kg ai ha^{-1} ; Primextra Gold at 3.0kg ai ha^{-1} ; Pendimethalin + Atrazine at 1.0 + 1.0kg ai ha^{-1} ; Pendimethalin + Atrazine at 2.0 + 2.0 kg ai ha^{-1} ; Gardoprim A (Atrazine + Terbutylazine) at 1.0kg ai ha^{-1} ; Gardoprim A (Atrazine + Terbutylazine) at 2.0kg ai ha^{-1}. Hand Weeded x 2 (3+6 WAP), Unweeded and Weed Free treatments were the controls. The size of the pots used was 0.029m$^{2.}$ Result shows that hand weeded treatments: weedfree (0.996), weed x 2 (1.069) had lower diversity indices compared to the herbicides with a mean index of 1.34 and the unweeded with an index of 1.187. It was also observed that the pre-emergence herbicides used may have controlled grasses more than broad-leaved weeds. The result of this study showed that the herbicide (Pendimethalin + Atrazine) at 2.0 +2.0kg ai ha-^{1} was more effective in reducing weed seeds (seed bank) than the other five herbicide treated plots, with a mean diversity index value of 1.083, while the highest weed diversity was recorded in plot treated with Gardoprim A herbicide (Atrazine +Terbutylazine) at 2.0 +2.0kg ai ha^{-1}, with a mean diversity index of 1.260. . The mean average weed diversity was highest (1.214) in 0-5cm soil depth followed by 1.149 in 5-10cm soil depth and then 1.068 in 10-15cm depth. This shows that as soil depth increases the number of weed species decreases. The result also implied that Gardoprim A may not provide some residual control of late emerging weed seeds or seedlings that germinate several months after the initial treatment. This calls for a careful selection of appropriate pre-emergence weed control program in diverse arable cropping systems that will provide lasting weed control for late emerging weeds in the system.

Keywords: Index of diversity, seedling emergence, soil depth and pre-emergence

INTRODUCTION

Soil seed bank is a reservoir of viable seeds in the soil. However, seed banks could also be referred to as a place where seeds are collected or gathered until germination. It consists of both recent and older seeds shed in, and dispersed into a locality. This reserve of propagules is the source of local weed diversity, and is essential for the continuity of any local weed flora. Large seeds bank is a common attribute of the wide range of plant communities (Dekker, 1999). The species composition of seed bank in any plant community depends on seed inputs from present and previous vegetation and the longevity of viable seed under local condition (Robert, 1981). The abundance and composition of species in arable weed seed bank and established or aboveground weed communities is a reflection past and current management of soils, crops and weeds (Lègeré and Samson, 1999; Mendalled *et al.*, 2001; Tørresen and Ŝkuterud, 2002). The survival and fate of seeds in the soil represent a crucial step in weed flora dynamics (Baldoni *et al*., 2000). The impact of chemical control on seed bank is either selective or non-selective. Its influence can be direct, when soil applied herbicides become selective (Wright *et al*., 1993). The management of weed seed bank is based on knowledge and modification of the behaviour of seeds within the soil seed bank matrix (Dekker, 1999). Weed seeds are important component of weed and crop life cycle as they are the origin of future populations. The success of any crop production activity depends to some extent on the ability to modify the seed bank in favour of crop growth. Weed management outcomes have often been linked to infestation size, as larger resident weed seed banks often require more weed control to adequately manage the population (Hartzler & Roth, 1993). The ability of a succeeding crop to survive depends on the size of the previous seed bank. According to Nobel and Slatyer (1980), knowledge of this seed bank is essential to achieve a more complete understanding of local vegetation dynamics, since composition of the soil seed bank may predict what species dominate a plant community after disturbance. The studies of samples of weed seed bank will also enable the identification, classification of the flora and subsequent plant community according to Voll *et. al.*(1995). Similarly, the study of weed seed band will help us in judging the efficiency of previous management strategies as well as predicting the need for subsequent control and rational herbicide use in relation to economic considerations in production. Conventional and traditional agriculture alike dwells on trying to reduce seed bank size. The choice of weed management strategies must reflect their efficiency in reducing the local seed bank size.. Herbicides are the most effective weed management tool of the 20th century. Herbicides are very effective at reducing weed populations and at the same time the number of seed added to the soil seed bank. Changes in crop rotation and herbicides use can result in changes in weed seed banks in arable soil (Squire *et al.,* 2000). Numbers of species can increase if herbicide use is reduced. The estimation of viable seeds in seed banks is crucial to understanding seed bank dynamics (Forcella, 1992), will enable planning for subsequent vegetation management. Thus the present study is aimed at determining the number of viable seeds (seed bank) and their diversity and distribution within 0-15 soil depth profile following previous maize pre-emergence herbicides.

MATERIALS AND METHODS

Land Preparation and Plot Layout

A 22m by 35m portion of land which was previously treated with pre-emergence herbicides for weed control in maize was cleared with cutlass and all the trashes gathered and removed from the plot. The plot was re-plotted with wooden pegs and twines according to the previously treated pre-emergence herbicides 40 WAT as follows:

1. PrimextraGold at 1.5 kg ai/ha
2. PrimetraGold at 3.0kg ai/ha
3. Pendimethalin + Atrazine at 1.0+1.0 kgai/ha
4. Pendimethalin + Atrazine at 2.0+2.0 kgai/ha
5. Gardoprim A at 1.0kg ai/ha
6. Gardoprim A at 2.0 kg ai/ha
7. Hand weeded twice (3 and 6 WAP)
8. Weedfree
9. Unweeded check

Weed Seed Bank Procedures:

Soil sample collection and preparation: Soil samples were collected from this previously treated maize herbicide trial from the Research and Teaching Farm of the Department of Crop and Soil Science Faculty of Agriculture, University of Port Harcourt Rivers State, Nigeria (04 54 538'N, 006 55 329' E, 17m. a.s.l). Soil samples were collected at three different depths: 0-5cm, 5-10cm and10-15cm in each plot replicate using Soil Auger along each segment transect in a manner that depicts "**W**" and then bulked into a well-labelled nylon or polyethylene bag. Thus a total of 27 subsamples of each soil depth were collected from the three field sample replicates of the previous nine treatments listed above giving a total of 81 samples. Thereafter, samples were taken to the greenhouse at the Center for Ecological Study of the Department of Plant Science and Biotechnology, University of Port Harcourt, to initiate the weed seed bank study. Three samles of 100g of each were measured out to represent three replicates of the previous nine weed control treatments. A total of 27 treatment samples were measured out from each soil depth. Each sample was put into a small white plastic container of diameter 9cm and height 4.5cm, containing Whatman no.1 filter paper. The containers were perforated underneath to allow water absorption by sub-irrigation. These samples were arranged in randomized complete block design at the greenhouse with 54 pots per block in three location of the greenhouse as replicates (9 treatments x 3 soil depth x 3 replications x 2 repeats) and placed upon each of the container's cover. The samples were sub-irrigated with tap water and the absorption was through the perforated holes, to avoid splashing that will lead to weed seed lost. The first seedling count started one week after watering (WAW)

Weed seedling identification and count

The emerged seedlings were counted, identified (at the level of broad-leaved and grasses and not at species taxa level) and uprooted or removed weekly. The reason for removing seedlings was to avoid mistake of double counting in the next counting day. After the first three weeks of count, samples were allowed to dry for two days inside the green house and then upturned to prepare for the next counting. The soil samples were squeezed lightly with hand to make them homogenous. However, the old filter papers were removed and then replaced with new ones. Then the soil samples were upturned into the individual container and moistened with tap water as usual. This was done to give all the seeds equal chances to germinate, the seedling emergence, identification and count continued for seven weeks. The seed bank study process was repeated and the data from the repeat trials were combined to represent the information sought from the experiment.

Data collection and analysis

Shannon-Wiener index was used to calculate the diversity of the seedling broad-leaved and grass species as affect by the previous management of *Panicum maximum* weed community in maize with selected pre-emergence herbicides. This index is based on proportional abundance of each species. The Shannon-Wiener diversity index (**H^1**) was calculated using the following formula

$H^1 = -\Sigma\ [Pi\ (InPi)]$

Where pi= proportional abundance of a species (pi=ni/N), ni= the number of ith species and N= total numbers of individuals of all species in the community (Booth *et al.,* 2003). In addition to the diversity index, data were recorded on composition, density and percentage contribution, to the index by the broad category of the weeds. The germinable seeds were converted to the number of seeds per square meter. The density values were transformed prior to analysis to improve normality and homogeneity of variance. The data obtained were subjected to Analysis of Variance and Least Significance Difference (LSD) was used in comparing the means.

RESULTS AND DISCUSSION

Weed seed bank diversity

The result obtained showed that weed seedling diversity was affected by depth. This means that majority of the seeds were domiciled within 0-10 cm soil depth, probably because of the field was fallow for over 40 weeks after the previous treatment or disturbance of the site. This result is in agreement with the observations of Pereira *et al.*, 2013, that majority of viable weed seeds were found at 0-10 cm soil depth, probably because of previous management. Mean species diversity indices per soil depth were as follows 0-5 cm (1.214)> 5-10 cm (1.149)> 10-15 cm (1.068) (Table 1). Across soil depth weed-free treatment had the least weed seedling diversity (compared to the other treatments, while the unweeded check had the highest. The herbicide treatment did not differ in their effect on weed seedling diversity across soil depth ($P<0.05$). However, Pendimethalin + Atrazine at 2.0 kg ai/ha, had the lowest diversity index, compared to the other herbicides; meaning that it was more effective in controlling weed seeds in the soil while Gardopim A (Atrazine + Terbutylazine) had the highest index (Table 1).

Table 1: Effect of treatment on weed seed bank diversity at three soil depth 40 WAT

Treatment	Shannon-Weiner's diversity index per soil depth		
	0 - 5cm	5 – 10cm	10 – 15cm
Prime extra Gold at 1.5kg ai ha^{-1}	1.281	1.183	1.110
Prime extra Gold at 3.0kg ai ha^{-1}	1.328	1.059	1.127
Pendimethalin + Atrazine at 1.0 + 1.0kg ai ha^{-1}	1.277	1.226	0.938
Pendimethalin + Atrazine at 2.0 + 2.0kg ai ha^{-1}	1.148	0.995	1.105
Gardoprim A (Atrazine + Terbutylozine) at 1.0kg ai ha^{-1}	1.376	1.219	0.964
Gardoprime A (Atrazine + Terbutylazine) at 2.0kg ai ha^{-1}	1.282	1.136	1.373
Hand weeded x 2 (3 + 6 WAP)	1.011	1.214	0.986
Unweeded	1.362	1.141	1.058
Weed free	0.864	1.170	0.953
LSD (P=0.05)	0.253	0.214	0.276

This is an indication that the herbicide Gardoprim A was less effective in control a wide range of germinable seed bank from the treated site, hence may not offer lasting or acceptable control of germinable seed bank overtime. Herbicide combinations involving Pendimethalin have been reported to be one of the most efficient in controlling emerging weed flora (Pereira *et al.,* 2013). Our result seems to be consistent with this report as Pendimethalin + atrazine in the present study was the most efficient in reducing germinable weed seed bank. The result also shows that hand weeded treatments (weed twice and Weedfree) had low seedling weed diversity compared to the unweeded check, although the differences were not significant. , the unweeded check recorded the highest weed seedling diversity (Table 2).

Averaged over soil depth (0-15 cm), the result showed that weed seedling were more diverse in plots that were treated with herbicides and unweeded check (No herbicide), than in plots that were weeded weekly (weedfree) and weeded twice (at 3 and 6 WAT), this might be probably due to lack of further soil disturbance after the treatments were imposed for the herbicide treatments and the unweeded check. These results seem to agree with the reports of Carter and Ivany (2006), Sosnoskie *et.al*.(2006), and Feldman *et.al*. (1997), who reported that species diversity higher were tillage intensity was reduced. For the herbicide treatments germinable weed seed were more diverse in plots treated with

Gardoprim A than the rest of the herbicide treatments with an average index of 1.225, but the differences were not significant when compared with the other herbicide treatments.

Table 2: Effect of treatments on weed seedbank diversity 40 WAT

Treatment	Diversity index
Prime extra Gold at 1.5kg ai ha^{-1}	1.192
Prime extra Gold at 3.0kg ai ha^{-1}	1.171
Pendimethalin + Atrazine at 1.0 + 1.0kg ai ha^{-1}	1.147
Pendimethalin + Atrazine at 2.0 + 2.0kg ai ha^{-1}	1.083
Gardoprim A (Atrazine + Terbutylazine) at 1.0kg ai ha^{-1}	1.186
Gardoprime A (Atrazine + Terbutylazine) at 2.0kg ai ha^{-1}	1.26
Hand weed x 2 (3 + 6 WAP)	1.069
Unweeded	1.187
Weed free	0.996
LSD (P=0.05)	0.237

However, Gardoprim A diversity index was significantly higher than was recorded with the weed-free treatment (0.966). All the herbicide rates except Gardoprim A at 1.0 kg ai/ha did not differ significantly in their effect in reducing weed seedling diversity than the hand weeded treatments (Table 2). Diversity indices measured in this study were within the range of those reported for various cropping systems in diverse geographical areas (Clements *et al*., 1994; Derksen *et al.* 1995). Values reported in the literature for Shannon's H' for weed communities generally are <2.0.

Percentage weed type contribution to weed seedling diversity

The result obtained shows that broadleaved weed seeds were higher in 5-10 cm soil depth than the grasses contributing about 65.48 % of the germinable seed bank. When compared to grasses (5.15%), and this represents 93% of the total germinable weed seeds at this soil depth (Table 3). The mean percentage value of grasses in 0-5cm soil depth is higher (8.52) than that of 5-10cm (5.15) and that of 10-15cm (3.17) soil depth (Table 3). The result also showed that germinable grass weed seeds concentrated more at of 0-5cm depth than at 5-10cm and 10-15cm soil depths respectively. The values in parenthesis are the transformed values. Our results also indicated that all the treatments used controlled grass weed seeds more than the broadleaved weed seeds. The mean values of broadleaves at the soil depth 0-5, 5-10 and 10-15cm are 43.5, 65.5 and 28.1% respectively and the mean values of grasses were 8.5, 5.2 and 3.6 % respectively at the same soil depths (Table 3).

Table 3: Percentage weed type contribution to weed seedling diversity at three soil depth 40 wat

	Broadleaves			Grasses			Total		
TREATMENT	**0-5 cm**	**5-10cm**	**10-15cm**	**0-5 cm**	**5-10 cm**	**10-15 cm**	**0-5 cm**	**5-10 cm**	**10-15 cm**
PrimextraGold 1.5Kg	38.67(6.09)	66.33(8.01)	33.17(5.57)	13.50(3.68)	7.50(2.82)	5.33(2.41)	52.17(7.09)	73.83(8.47)	38.50(6.08)
PrimextraGold 3.0Kg	49.67(6.89)	68.17(8.26)	29.17(5.41)	10.83(3.35)	5.83(2.49)	4.00(2.11)	60.50(7.66)	74.00(8.60)	33.17(5.77)
Pendi + Atraz1. 0+1.0Kg	41.67(6.25)	63.33(7.77)	30.67(5.26)	8.67(2.85)	4.83(2.26)	3.50(1.85)	50.33(7.05)	68.17(8.06)	34.17(5.55)
Pendi + Atraz2. 0+2.0Kg	38.00(6.13)	90.00(8.98)	46.67(6.14)	8.33(2.89)	5.33(2.13)	4.83(2.22)	46.33(6.75)	95.33(9.22)	51.50(6.51)
Gardoprim A 1.0Kg	46.50(6.70)	86.83(8.13)	39.50(5.72)	9.00(3.01)	6.00(2.45)	3.50(1.93)	55.50(7.41)	92.83(8.48)	43.00(5.99)
Gardoprim A 2.OKg	42.17(5.85)	72.67(8.35)	19.33(4.18)	5.33(2.31)	4.00(2.03)	2.17(1.61)	47.50(6.33)	76.67(8.60)	21.50(4.44)
Hand weeded x 2	66.33(7.96)	58.83(7.70)	19.50(4.28)	5.83(2.44)	3.00(1.78)	2.17(1.57)	72.17(8.29)	61.83(7.89)	21.67(4.50)
Unweeded	30.33(5.34)	41.17(6.02)	16.00(3.99)	9.00(2.91)	5.83(2.33)	5.17(2.32)	39.33(6.23)	47.00(6.55)	21.17(4.59)
Weedfree	37.83(6.14)	42.00(6.46)	18.67(4.36)	6.17(2.54)	4.00(2.11)	1.50(1.41)	44.00(6.63)	46.00(6.76)	20.17(4.53)
LSD (P=0.05)	33.15(2.57)	69.02(3.76)	34.09(2.57)	8.97(1.53)	6.16(1.34)	4.05(1.93)	35.23(2.53)	72.61(3.77)	36.19(2.64)

Of all the treatments, unweeded check contributed less (39%) to the total germinable weed seed bank diversity at 0-5 cm soil depth while the hand weeded twice contributed about 72% to the total weed seed diversity at the same soil depth. The Weedfree treatment (weekly weeding) contributed about 44% (Figure 1). The herbicide treatments irrespective of rates contributed to the total germinable seed diversity at 0-5 cm soil depth as follows: PrimextraGold (56.1%) > Gardoprim A (51 %) > Pendimethalin + Atrazine (48%) (Figure 1).

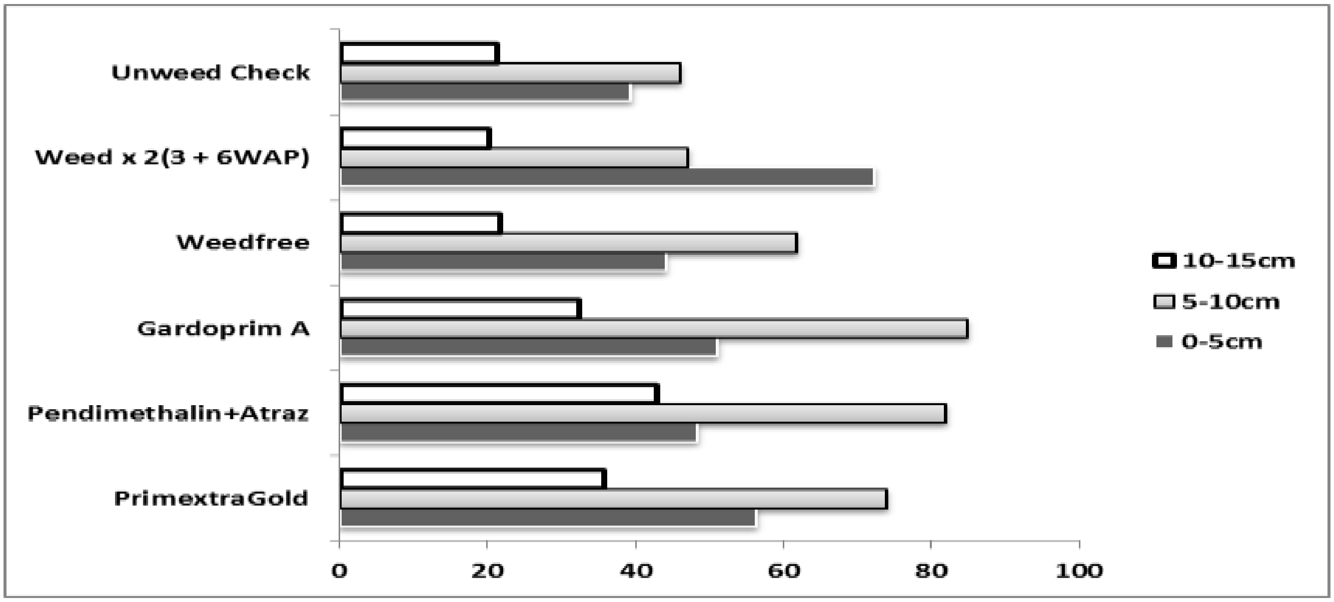

Figure 1: Percent (%) contribution by treatment to the total germinable seed bank at three soil depth 40 WAT

At 5-10 cm soil depth there was a trend observed from the treatments that was different from the previous depth; which was as follows: Gardoprim A and Pendimethalin + Atrazine contributed ± 82 % each to the total weed seed diversity, hand weeded twice contributed 61.8%, Weedfree and unweeded check contributed 46 % and 47 % respectively (Figure 1). The trend was also variable at 10-15 cm soil depth with the weeded and unweeded check contributing less (≥ 20 %) than the herbicide treatments (≥ 30 % and ≤ 43 %) to the total weed seed diversity (Figure 1). Though soil depth and treatment contribution to the total weed seed diversity was variable but the differences were not significant (P > 0.05). This variable in soil depth and treatment effect on weed seed diversity may be attributed to previous vegetation management and seeds recruited into seed bank previously and 40 weeks before the seed bank study, and length of their viability, as influenced by the soil micro-environment. This result may partially align with the report of Roberts (1981) that the composition and size of soil seed bank in a plant community depends on seed inputs from present and previous vegetation and the longevity of viable seed under the local soil environment. The overall effect of treatments interms of contributing to the total weed seed bank diversity were as follows: unweeded check (No herbicide) 35.5 %, weedfree 42.5%, weed x 2 (3 + 6 WAP) 46.5%, PrimextraGold 55.3%, Gardoprim A 56 % and Pendimethalin + atrazine 57.7 %. Pereira et al. (2013), in their work on with herbicide combination for weed seed bank control concluded that treatments Metolachlor and Pendimethalin mixture, no herbicide treatment and weekly weeding were the most effective in reducing the weed seed bank in the study area. Our result is consistent with this report with respect to unweeded, weedfree and hand weeded twice but variable with the herbicides, which were similar in their effect on weed seed bank irrespective of rates

Weed Seedling Composition and Density

In terms of weed seedling composition broad-leaf weed seedlings were more dominant across soil depth compared to grasses. The germinable mean broadleaved soil seed bank varied from 542 m^{-2} at soil depth of 10- 15 cm to 2339 m^{-2} at soil depth of 5-10 cm (Table 4).

Table 4: Weed seedling density (no.m^2)

Treatments	Broadleaves			Grasses			Total		
	0-5	**5-10**	**10-15**	**0-5**	**5-10**	**10-15**	**0-5**	**5-10**	**10-15**
Primextra1.5kg	1311ba	2249a	1124a	458a	254a	181a	1768a	2503a	1305a
Primextra3.0kg	1684ba	2311a	989a	367a	198a	136a	2051a	2508a	1124a
Pendi+Atraz1.0kg	1412ba	2147a	1039a	394a	164a	119a	1706a	2311a	1158a
Pendi+Atraz2.0kg	1288ba	3051a	1582a	283a	181a	164a	1571a	3232a	1745a
GardoprimA1.0kg	1576ba	2944a	1339a	305a	203a	119a	1881a	3147a	1458a
GardoprimA2.0kg	1429ba	2463a	655a	181a	136a	73a	1610a	2599a	729a
Handweededx2	1249a	1994a	661a	198a	102a	73a	2446a	2096a	735a
Unweeded	1028b	1395a	542a	305a	198a	175a	1333a	1593a	718a
Weed Free	1283ba	1424a	633a	209a	136a	51a	1492a	1559a	684a
LSD(p=0.05)	1123.8	2339	1155.5	303.9	208.8	137.3	1194	24.61	1226.8

The broadleaved weeds were distributed in this order 5-10 cm > 0-5 cm > 10-15 cm across the soil depth while the grasses followed according to the depth profile as follows: 0-5 cm >5-10 cm > 10-15 cm soil depth (Table 4). The reason for this type of distribution for the grasses may be that at depth more than 0-10 cm they may not survive as they are small seeded species compared to the broadleaved weeds which are somehow large seeded weeds. Broadleaved weeds dominated the composition of weed seedling emerging at any sampling depth (0-5 cm 16.4 % grasses and 83.6% broadleaves, 5-10 cm 7.3 % of grasses and 92.7 % broadleaves; 10-15 cm 11.3 % of grasses and 88.7% of broadleaves. Across soil depth and treatment broadleaves accounted for about 86.4% and grasses about 13.6% of the total weed composition (Table 4). Across soil depth treatment did not differ significantly in their effect on germinable weed seedling composition. With respect to soil depth, total weed seedling density composition was as follows 0-5 cm soil depth 33.7%, 5-10 cm depth 45.8% and 10-15 cm depth 20.5%. This result shows that weed seedlings were greater or more abundant within 0-10 cm soil depth and less as the depth goes beyond 10 cm. This may have been due to previous disturbance and the length of fallow after the herbicides application (40 weeks), as more weed seeds may have been recruited into the seed bank. This result is in agreement with the observations of Pereira et al., 2013, that majority of viable weed seeds were more at 0-10 cm soil depth, probably due to previous management impact. Irrespective of soil depth, total weed seedling density was variable with the herbicide treatments.The least weed seedling density was recorded in the plots treated with Gardoprim A at 2.0kgha while the highest was recorded in the plots treated with Pendimethalin + Atrazine @ 2.0kg/ha. Considering the treatment effect in reducing total weed seedling density the effect was as follows: Weedfree (1245/m^2) > unweeded (1256 seeds /m^2) >Gardoprim (1646 seeds/m^2) > Pendi + Atraz. at 1.0kg/ha (1725 seeds /m^2) > Hand weed x 2 (1759 seeds/m^2) > Primextra at 1.5kg/ha (1860 seeds/m^2) > Primextra at 3.0 kg/ha (1894 seeds/m^2)> Gardoprim at 1.0kg/ha (2162 seeds/m^2) > Pendi + Atraz. 2.0kg/ha (2183 seeds/m^2) Table 4. Seedling density did not differ with depth in both plots hand weeded and those treated with herbicides.

CONCLUSION

Findings from our study showed that seed bank from the study area is characterized by an abundance of viable broadleaved weeds that are resident within 0-10 cm soil depth and grasses that are more abundant at the 0-5 cm soil depth. This study has also shown that the weed spectrum in the humid forest zone may shift in favour of broadleaved weeds after a pre-emergence weed control program. Therefore, emphasis should be directed towards solving the problem of late emerging weeds that may be dominated by the broadleaved weeds. Nevertheless more research is needed to develop management strategies that will minimize this shift and dominance in flora by specific weed type.

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