Original Research Article

Effects of selected crops on germination and growth of *Bidens pilosa* and other weeds

ABSRACT

A screen house pot study was done at Uganda National Crops Resources Research Institute, Namulonge in Uganda during September-December 2021. Profiled metabolites from 5-donor plants; *Mucuna pruriens,* L.(Mc), *Cymbopogon nardus* L. (C), *Desmodium uncinatum* Jacq. DC.(D), (*Zea mays,* L. (Mz) and New Rice for Africa 1 (NERICA 1) were Terpenoids and Phenols. NERICA 1 (R) is a cross of *Oryza sativa* and *Oryzaglaberrima*. C produced 10 terpenoids + 1 ester, R and D each released 6 Terpenoids + 3 Phenols. Mc produced 2 Terpenoids + 4 Phenols and 5 Terpenoids + 4 Phenols were profiled from Mz. We adopted a completely randomised block design with 3 replicates and eight combinations of 3 leaf+stem+root powders namely; Mc+D+C, Mz+Mc+C, RDC, Mc+D+C, Mz+R+Mc, Mz+D+Mc, R+Mc+C and Mz+R+C, mixed with 1.5 kg soil (15, 30 and 45 %, w/w). Controls had no powders. Mixtures were placed in plastic pots and 20 seeds of *Biden pilosa* L. were sown in each pot. Every 2 days, 200 ml of water was applied to all treatments for 60 days. Mz+Mc controlled *B. pilosa* and volunteer weeds relative to R+C and C+D. Weed germination and biomass reduced with increased powder doses. Volunteer monocot weeds were more inhibited than dicots. High potential exists for functional allelopathy using allelochemicals in the donor plants to develop bio-herbicides.

Key words: *Allelopathy, Bidens pilosa, Cymbopogon nardus, Desmodium uncinatum, Mucuna pruriens, Oryza sativa, Volunteer weeds, Zea mays.*

1.INTRODUCTION

Weeds are a major constraint to directly sown crops like rice (1). Development of resistance in previously susceptible weed species and environmental pollution in soils are the major drawbacks of chemical herbicide use (2). Reports indicate potential plant species that control weeds, which are both cost effective and environmentally friendly when used as allelochemicals (3, 4, 5, 6). Allelopathy is described as any direct or indirect harmful or beneficial effects of one organism on another through the production of chemical compounds released into the environment (7). Allelochemicals are a subset of secondary metabolites, which are not required for metabolism of an allelopathic organism and are released via leaf leachates, volatiles or root exudates (8), from seeds and flowers (9) and from plant mulches in both natural and agricultural systems (10). Terpenoids and Phenolic acids are the most common plant allelochemicals serving a range of important physiological and societal functions. Terpenoids are classified by the number of carbons in the skeletal structure, typically in units of five carbons. Phenolic

acid compounds are universally distributed in plants. The inhibitory effects of terpenoids and phenolic compounds on seed germination and crop growth have been widely reported. (11) observed some phenolic compounds that inhibited amino acid transport and protein synthesis. (12) attributed the allelopathic contribution made by phenolic compounds to more than one substance. (13) noted reduced growth of *Echinochloa crus-galli* in paddy fields due to allelochemicals released by the roots of *Oryza sativa*. (14) recognized significant correlation between phenolic contents and percent inhibition of root growth in barnyard grass. Weed control using the donor crops like NERICA 1, *C. nardus*, *D. uncinatum*, *M. pruriens* and *Z. mays* stover water extracts (4), crop mulches (5), intercrops (6) have been documented.

Rice and maize are the leading food crops in the world and together with wheat supply more than 50 % of the calories. Considering their significance and economic importance, cultivation of these crops is an option for improved livelihoods and basic service support in dry lands. *M. pruriens* and *D. uncinatum* are major livestock feeds, medicinal and used in environment management. *C. nardus* is a medicinal plant and used in scenting of soaps, detergents and insect repellent preparations, antibacterial and an antifungal agent. *B. pilosa* is a notorious weed but also used as food and medicine in sub-Saharan Africa. *B. pilosa* has been identified as a fast-growing weed with competitive abilities and allelopathic properties in many cropping systems. The weed is also a host and vector to *Meloidogyne* nematodes (15).(16) noted that allelopathic inhibition is complex and can involve the interaction of the different classes of chemicals with mixtures having a greater allelopathic effect than individual compounds. In the laboratory and 2 screen house studies we profiled the bio-active compounds vegetative parts of the donor plants and determined the herbicidal effects of secondary metabolites in mixed leaf+stem+root powders of *M. pruriens*, *C. nardus*, *D. uncinatum*, *Z. mays* and NERICA 1 crops on the germination of *B. pilosa*, volunteer monot and dicot weeds.

2. MATERIALS AND METHODS

2.1 Root Exudates

2.1.1 Pot culture in screen house

A screen house pot study was done at Uganda National Crops Resources Research Institute, Namulonge in Uganda during September-December 2021. Plastic pots (radius, 7 cm; height; 26 cm) were filled with 3 kg of forest loam soil. Rainfall of 638 mm was received during the cropping season with minimum and maximum temperatures of 19 and 30^{o}C respectively. Five 4-day old pre-germinated seeds ofNERICA 1, *D. uncinatum*, *Z. mays* and *M. pruriens* seeds were sown in the pots. Since *C. nardus* ispropagated by vegetative means, five plantlets were uprooted, cleaned and immediately potted and placed in a screen house. One pot without any plant was maintained as control. About 200 ml tap water was applied to each pot every two days till harvest (60 days after planting), when the crops were at peak vegetative growth (17). Five plants were taken out of each plastic pots at harvest and the plant roots were shaken gently to remove the rhizosphere soil. The samples were oven dried at 65 ^{o}C for 48 h to constant weight for compound analysis.

2.1.2 Extraction and analysis of potential organic compounds from plants

Organic compounds in plants were extracted using solid phase micro-extraction (SPME) and by solvent extraction. Prior to the extraction, the SPME fibre was pre-conditioned for 1.0 hr at 250 ^{o}C under a stream of helium inside the gas chromatograph (GC) injection port liner. The SPME fibre used was gauge 24, 1 cm long, coated with divinyl benzene/ polydimethylsiloxane and with 65 µM film thickness. In a single manual injection, one g of each plant sample was weighed into a 10 ml air tight glass vial. The sample and control extractions were placed into a thermostat heated block at 60 ^{o}C for 1.0 h with the fibre exposed to the headspace for the entire duration. The fibre was retracted and introduced into the injection port of the GC in splitless mode. One g of plant sample was accurately weighed into a 50 ml extraction tube and extracted with 10 ml of hexane by shaking at 250 revolutions per minute (rpm) in an orbital shaker for 1.0 h. Two millilitre extract was aliquoted into an Eppendorf tube and centrifuged at 5000 rpm for 10 min and 1 ml extract was aliquoted into a GC vial for injection. One hundred mg of freeze dried samples were weighed into 2 ml Eppendorf and 1800 µL of hexane was added with two mini steel balls placed inside the Eppendorf. The samples were vigorously ground in a Geno grinder for 10 min. The extract was centrifuged at 5000 rpm for 5 min and 200 µL of extract was diluted with 800 µL of hexane in a GC vial for injection.

2.1.3 Gas Chromatography and Mass spectrometry instrumental analysis conditions

Samples were subjected to analysis using a 7890A GC system (Agilent Technologies, USA) coupled to a 240-ion trap mass spectrometer detector (Agilent Technologies) using the Agilent 7693A automatic liquid sampler for solvent extracted samples. A VF5-MS (5 % phenyl methylpolysiloxane), 30 m × 0.25 mm id, 0.25 µm film capillary column was used with the injector port set at 280 °C. Helium was used as carrier gas at a flow rate of 1 ml/min. The oven temperature was programmed to rise from 50 °C to 180 °C at 4 °C/min followed by an increase to 250 °C at 3 °C/min. The ion trap mass spectrometer parameters were as follow: scan range 50-540 (m/z), ionization mode EI and transfer line temperature, manifold temperature and trap temperature of 250 °C, 100 °C and 150 °C, respectively. Chromatograms and spectra representing individual samples were analysed using the automated mass spectral deconvolution and identification system software (AMDIS, US). The identification of the individual compounds was performed by comparing each of the mass spectra with the database of NIST 11 (Gaithersburg, MD, USA) and Wiley 7N (John Wiley, NY, USA) and also by comparing the calculated Kovats linear retention indices using retention times of n-alkane series against the values obtained in the NIST web book for the same capillary column stationary phase.

2.2 WEED CONTROL BY LEAF/STEM/ROOT POWDER

The Volunteer weeds were identified as the non- sown weeds that comprised Dicots and Monocot weeds. The sown weeds (Bidens pilosa) and volunteer weeds constituted total weeds in the study.

2.2.1 Experimental design and treatments

A screen house experiment was done during September-December 2022 at Namulonge Research Station in Uganda and herbicidal efficacy of powdered formulations from dry R+C+D+Mc+Mz; Leaf+stem+root powders on germination of *B. pilosa* were determined. The inhibitory effects of secondary

metabolites in the donor crops on volunteer weeds (seed bank of monocots + dicots other than *B. pilosa* in the soil) were also assessed. During the cropping season the area received a total of 638 mm rainfall with minimum and maximum temp of 19 and 30^{o}C respectively. Two months old whole plants each of NERICA 1, *D. uncinatum*, *M. pruriens*, *Zea mays* and *C. nardus* plants were harvested, dried at 65 ^{o}C for 48 h to constant weight and separately crushed to powders. LSR powders were combined in equal proportions of three powders to make eight powdered mixtures namely; Mz+D+C, Mz+Mc+C, R+D+C, Mc+D+C, Mz+R+Mc, Mz+D+Mc, R+Mc+C and Mz+R+C. The powders were mixed with sandy loam soil excavated from 10-20 cm depth of a fallowed field to make ratios of 15 %, 30 % and 45 % w/w (Powders to soil mixtures). 1 kg air dried soil was filled into seventy-two plastic pots (radius, 7 cm; height; 26 cm). Treatments were arranged in a completely randomised block design with three replicates. Control pots were not supplied with LSR powders. Twenty seeds of *B. pilosa* weedwere sown in each pot and watered initially with 250 ml and later 100 ml of tap water every two days for 4 weeks.

2.3 DATA COLLECTION

Germinated *B. pilosa* and volunteer weeds were populationed on a weekly basis. All the weeds in each pot were harvested at 30 days after application of powders, populationed and classified into monots (narrow leaves) and dicots (broad leaves) and percentages ratios were calculated.

3.RESULTS

3.1 Bio-active compounds in donor plants

3.1.1 Cymbopogon nardus

Retention time, relative match factors and compounds identified in *Cymbopogon nardus* stalk are indicated in Table 1. *C. nardus* vegetative material released 10 terpenoids namely Citronellal, β-Citral, cis-Geraniol, trans-Carane, Eugenol, Geraniol acetate, β-Elemen, Caryophyllene, α-Gurjunene and γ-Cadinene and one ester named Citronellyl butyrate.

Table 1. Retention time, relative match factors and compounds in *Cymbopogon nardus* stover.

S/No	Retention time (Min)	Relative match factor	Identified compound	Class of compound
1	13.59	854	Citronellal	Terpenoid
2	16.20	859	Citronellyl butyrate	Ester
3	16.57	847	β-Citral	Terpenoid
4	16.99	870	cis-Geraniol	Terpenoid
5	20.24	874	trans-Carane	Terpenoid
6	20.42	888	Eugenol	Terpenoid
7	21.17	875	Geraniol acetate	Terpenoid
8	21.60	862	β-Elemen	Terpenoid

9	22.57	892	Caryophyllene	Terpenoid
10	24.51	854	α-Gurjunene	Terpenoid
11	25.48	895	γ-Cadinene	Terpenoid

3.1.2 *Desmodium uncinatum*

Retention time, relative match factors and compounds identified in *Cymbopogon nardus* stalk are indicated in Table 2. The 9 organic compounds profiled from *D. uncinatum* stover (Table 2) included six terpenoids namely Butylated Hydroxytoluene, 1,2,3-Trimethyl-4-[(1E)-1-propenyl]naphthalene, 1,4-Eicosadiene, 1-Ethyl-2-(1-phenylethyl)benzene, 3,4-Diethyl-1,1'-biphenyl and 2, 2'-Diethylbiphenyl plus three phenols namely 2,5-Di-tert-butylphenol; 3, 7, 11, 15-Tetramethyl-2-hexadecen-1-ol and 3,7,11,15-Tetramethyl-2-hexadecen-1-ol.

Table 2. Retention time, relative match factors and compounds in *Desmodium uncinatum* stover

S/No	Retention time (Min)	Relative match factor	Identified compound	Class of compound
1	25.12	849	Butylated Hydroxytoluene	Terpenoid
2	25.33	867	2,5-Di-tert-butylphenol	Phenol
3	29.57	845	3,4-Diethyl-1,1'-biphenyl	Terpenoid
4	30.15	863	1,2,3-Trimethyl-4-[(1E)-1-propenyl]naphthalene	Terpenoid
5	30.45	842	2,2'-Diethylbiphenyl	Terpenoid
6	31.02	785	1-Ethyl-2-(1-phenylethyl)benzene	Terpenoid
7	34.42	860	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	Phenol
8	35.09	759	(9Z)-9-Icosen-1-ol	Phenol
9	35.59	778	1,4-Eicosadiene	Terpenoid

3.1.3 Nerica 1

Retention time, relative match factors and compounds identified in *Cymbopogon nardus* stalk are indicated in Table3. Butylated Hydroxytoluene; 3,4-Diethyl-1,1'-biphenyl, 1,2,3-Trimethyl-4-[(1E)-1-ropenyl]naphthalene;2,2'-Diethylbiphenyl; 1-Ethyl-2-(1-phenylethyl) benzene and 1,4-Eicosadiene terpenoids were produced by NERICA 1 stover (Table 1). 2, 5-di-tert-butyl- Phenol, 3, 7, 11, 15-Tetramethyl-2-hexadecen-1-ol and (9Z)-9-Icosen-1-ol and an ester called Hexadecanoic acid were the phenols identified in the NERICA 1 stover.

Table 3. Retention time, relative match factors and compounds in *Oryza sativa* stover

S/no	Retention time (Min)	Relative match factor	Identified compound	Class of compound
1	25.13	849	Butylated Hydroxytoluene	Terpenoid
2	25.32	845	2,5-di-tert-butyl- Phenol	Phenol
3	29.56	862	3,4-Diethyl-1,1'-biphenyl	Terpenoid
4	30.15	832	1,2,3-Trimethyl-4-[(1E)-1-propenyl]naphthalene	Terpenoid
5	30.45	865	2,2'-Diethylbiphenyl	Terpenoid
6	31.01	792	1-Ethyl-2-(1-phenylethyl)benzene	Terpenoid
7	34.40	874	3,7,11,15-Tetramethyl-2-hexadecen-l-ol	Phenol
8	35.07	803	(9Z)-9-Icosen-1-ol	Phenol
9	35.60	782	1,4-Eicosadiene	Terpenoid
10	36.91	845	Hexadecanoic acid	Ester

3.1.4 *Mucuna pruriens*:

Retention time, relative match factors and compounds identified in *Cymbopogon nardus* stalk are indicated in Table 4. Compounds identified in the *M. pruriens* stover included two terpenoids named Butylated Hydroxytoluene and 1,4-Eicosadiene besides four Phenols named 2,5-di-tert-butyl-Phenol, Hexa-hydro-farnesol, 3,7,11,15-Tetramethyl-2-hexadecen-1-ol, (9Z)-9-Icosen-1-ol and (9Z)-9-Icosen-1-ol.

Table 4. Retention time, relative match factors and compounds in *Mucuna pruriens* stover

S/No	Retention time (min)	Relative match factor	Identified compound	Class of compound
1	25.13	856	Butylated Hydroxytoluene	Terpenoid
2	25.32	841	2,5-di-tert-butyl- Phenol	Phenol
3	26.24	786	Hexa-hydro-farnesol	Phenol
4	34.41	856	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	Phenol
5	35.10	796	(9Z)-9-Icosen-1-ol	Phenol
6	35.60	814	1,4-Eicosadiene	Terpenoid

3.1.5 Zea mays (Longe 6h)

Retention time, relative match factors and compounds identified in *Cymbopogon nardus* stalk are indicated in Table 5. Five terpenoids (Table 1) namely Ionene, Butylated Hydroxytoluene, 3,4-Diethyl-1,1'-biphenyl, 1,2,3-Trimethyl-4-[(1E)-1-propenyl]naphthalene, 1,4-Eicosadiene and four phenols called Falcarinol, 2,5-Di-tert-butylphenol, 3,7,11,15-Tetramethyl-2-hexadecen-1-ol and (9Z)-9-Icosen-1-ol were profiled from *Z. mays* stover.

Table 5. Retention time, relative match factors and compounds in *Zea mays* (LONGE 6H)

S/No	Retention time (Min)	Relative match factor	Identified compound	Class of compound
1	20.51	876	Falcarinol	Phenol
2	20.54	904	Ionene	Terpenoid
3	25.13	903	Butylated Hydroxytoluene	Terpenoid
4	25.32	896	2,5-Di-tert-butylphenol	Phenol
5	29.56	842	3,4-Diethyl-1,1'-biphenyl	Terpenoid
6	30.15	831	1,2,3-Trimethyl-4-[(1E)-1-propenyl]naphthalene	Terpenoid
7	34.41	856	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	Phenol
8	35.09	759	(9Z)-9-Icosen-1-ol	Phenol
9	35.58	812	1,4-Eicosadiene	Terpenoid

3.1.7 Comparison of compounds identified in plant materials

Compounds identified in the stover of C, D, R, Mz& Mc donor plants are shown in Table 6. The majority were Phenols and Terpenoids. Three compounds in common were identified as 1, 4-Eicosadiene; 2, 5-di-tert-butylphenol and 3,7,11,15-Tetramethyl-2-hexadecen-1-ol R, D, Mz& Mc vegetative materials. Butylated Hydroxytoluene and (9Z)-9-Icosen-1-ol and 1,2,3-Trimethyl-4-[(1E)-1-propenyl] naphthalene terpenoid were extracted from NERICA 1, *D. uncinatum* and *Zea mays.* NERICA 1 and *D. uncinatum* stover produced three compounds namely; 2, 2'-Diethylbiphenyl; 1-Ethyl-2-(1-phenylethyl) benzene and 3, 4-Diethyl-1,1'-biphenyl' whereas NERICA 1 exclusively released Hexadecanoic acid ester as mucuna exclusively produced Hexa-hydro-farnesol phenol.

Table 6. Compounds in Cymbopogon, rice, desmodium, mucuna and maize stover

S/No	***Cymbopogon nardus***	***Oryza sativa*** **(NERICA 1)**	***Desmodium uncinatum***	***Mucuna pruriens***	***Zea mays*** **(LONGE 6H)**
1	Citronellal	1,4-Eicosadiene	1,4-Eicosadiene	1,4-Eicosadiene	1,4-Eicosadiene
2	Citronellyl butyrate	2,5-di-tert-butyl-Phenol	2,5-Di-tert-butylphenol	2,5-di-tert-butylphenol	2,5-Di-tert-butylphenol
3	β-Citral	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
4	cis-Geraniol	Butylated hydroxytoluene	Butylated hydroxytoluene	Butylated hydroxytoluene	Ionene
5	trans-Carane	(9Z)-9-Icosen-1-ol	(9Z)-9-Icosen-1-ol	(9Z)-9-Icosen-1-ol	(9Z)-9-Icosen-1-ol
6	Eugenol	1,2,3-Trimethyl-4-[(1E)-1-propenyl]naphthalene	1,2,3-Trimethyl-4-[(1E)-1-propenyl]naphthalene	Hexa-hydro-farnesol	1,2,3-Trimethyl-4-[(1E)-1-propenyl]naphthalene
7	Geraniol acetate	2, 2'-Diethylbiphenyl	2, 2'-Diethylbiphenyl		Falcarinol
8	β-Elemen	1-Ethyl-2-(1-phenylethyl)benzene	1-Ethyl-2-(1-phenylethyl)benzene		3,4-Diethyl-1,1'-biphenyl
9	Caryophyllene	3,4-Diethyl-1,1'-biphenyl	3,4-Diethyl-1,1'-biphenyl		Butylated hydroxytoluene
10	α-Gurjunene	Hexadecanoic acid ester			
11	γ-Cadinene				

3.2 EFFECTS OF DIFFERENT DONOR PLANTS LSR POWDERS

The results on effects of type and concentration of different LSR powders on germination of *B. pilosa* and volunteer weeds, weed biomass and number of broad and narrow leaved weeds are indicated in figures 1-6.

3.2.1 *Bidens pilosa* weed population

All LSR powders significantly (P <0.05) influenced the germination of sown *B. pilosa* weed population at 30 days after application (DAA) with higher population inhibitions and lower (4.5 weeds) *B. pilosa*

population at 45% concentration (Figure 1). MzMc based powders most effectively controlled weed germination (4.5-6.5 weeds) and recorded higher weed population inhibition compared to other treatments. The *Bidens pilosa* population significantly increased (5-6.5 weeds) with a lower population inhibition (36%) for R+C based extracts. The percent *Bidens pilosa* weed inhibition similarly reduced (6-42%) giving a higher weed population (5.5-8.5 weeds) with CD based extracts. The *B. pilosa* weeds population however significantly (P<0.05), enhanced (6.5-8.5 %) when R+C+D mixed powder concentration was raised from 15% to 30 %, but the population reduced by 3.5 %, at a higher (45 %) LSR powder concentration corresponding to increased (79%) inhibition effects. The highest *B. pilosa* (16.5 weeds) weed population were recorded in control.

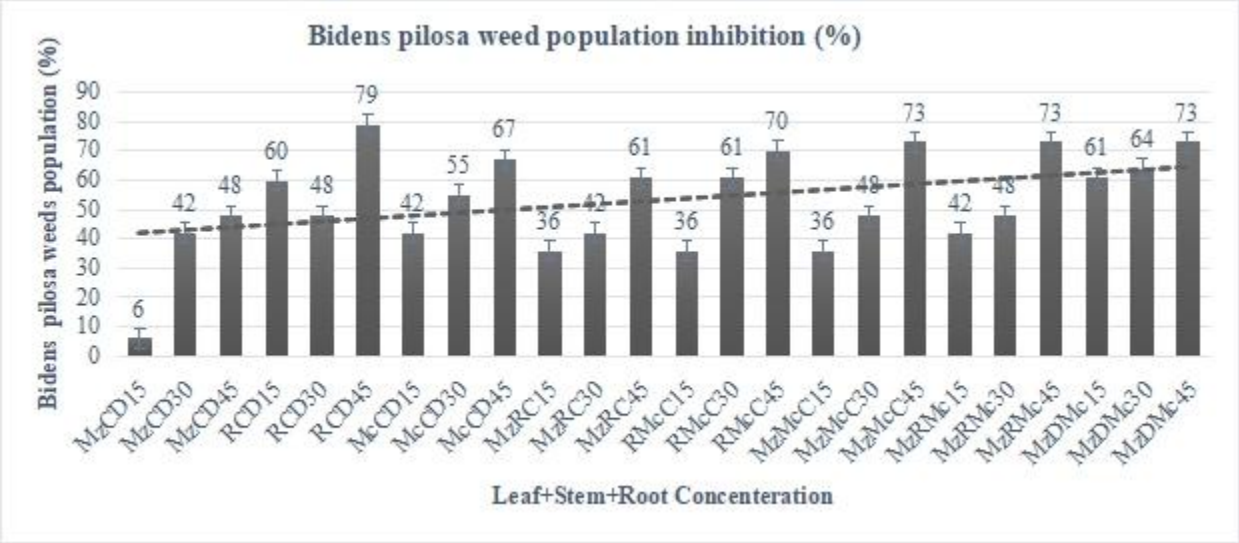

FIGURE 1: Effect of Leaf+stem+ root concentration on *Bidens pilosa* weed population

Note: mixed leaf/stem/root (LSR) powder concentrations (15-45 %) of rice (R), Cymbopogon (C), Desmodium (D), Mucuna (Mc) and Maize (Mz)

3.2.2 Total weeds population

Higher concentrations (15-45 %) of all LSR powders, significantly (P<0.5), reduced the total weed population at 30 DAA of the powder as indicated by the increasing inhibition on total weeds population with increase in the LSR (15-45%) concentration (Figure 2). The highest (26.5 weeds) total weed population was in the control treatment. The trend in total weed population inhibition was similar to that of Bidens pilosa in Figure 1. MzMc based extracts most effectively reduced the total population followed by RC extracts and lower inhibitions were with CD based LSR powder mixtures.

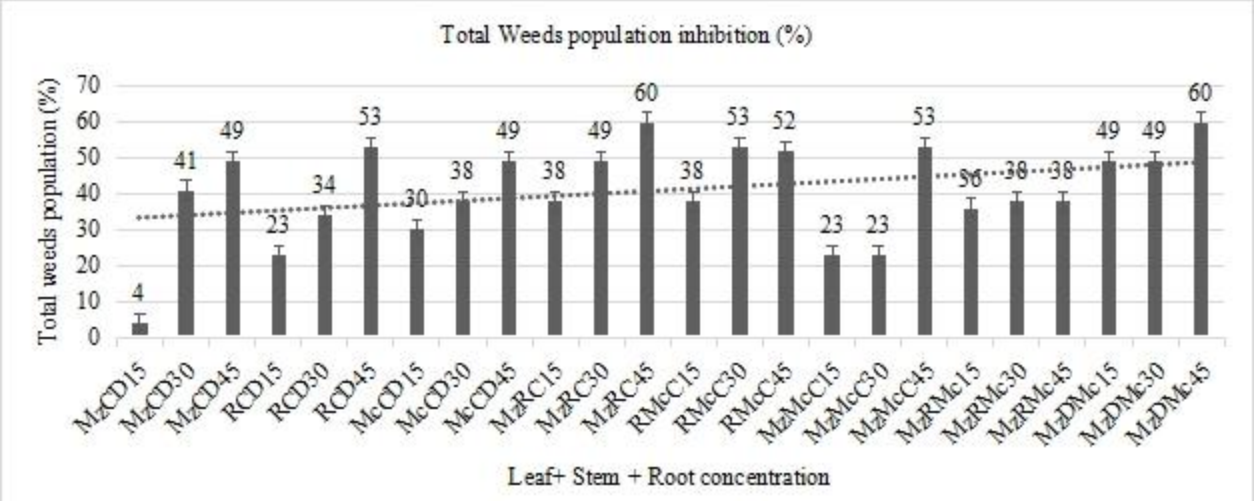

FIGURE 2: Effect of Leaf+stem+ root concentration on total weeds population
Note: mixed leaf/stem/root (LSR) powder concentrations (15-45 %) of rice (R), Cymbopogon (C), Desmodium (D), Mucuna (Mc) and Maize (Mz).

3.3.3 Volunteer weeds population

Volunteer weeds comprised of both monocot weeds (VMW) and dicot weeds (VDW) as indicated in Table 7. There was a total of 6 VMW and 6 VDW in all the treatments. The monocot weeds were majorly from the Poaceae, Cyperus and Poales families and the VDW were from the Amaranthaceae, Asteraceae and Euphorbiaceae families.

Table 7. Volunteer Monocot and Dicot weeds

Volunteer Monocot Weeds (VMW)				**Volunteer Dicot Weeds (VDW)**		
	English name	Botanical name	Family	English name	Botanical name	Family
1	Goose grass	*Eleusine indica*,	Poaceae	Cock's comb	*Celosia argentea*	Amaranthaceae
2	Finger grass	*Digtaria scalarum*	Poaceae	Goat weed	*Ageratum conyzoides*	Asteraceae
3	Nut grass	*Cyperus rotundus*	Cyperaaceae	Milk weed	*Euphobia heterophylla* L	Euphorbiaceae
4	Guinea grass	*Panicum maximum*	Poaceae		Gallinsogaciliata (Raf.) Blake	Asteraceae
5	Carpet grass	*Axonopuscompressus*	Poales	Slender amaranth	*Amaranthus gracilis*	Amaranthaceae
6	Swollen finger grass	*Chloris barbata*	Poaceae	Black jack	*Bidens pilosa*	Asteraceae

Higher concentrations (15-45 %) of LSR powders generally stimulated and enhanced the germination of volunteer weeds (Figure 3). Mz+R+C powders negatively and reduce the volunteer weeds population by 34-38 %, followed by Mz+C+D mixture (39-46 %) and the least effects were under R+C+D powders which stimulated the volunteer weeds population by 80-90%. Lower (51 %) volunteer weeds population

stimulation (4%) was recorded under RCD at 30 % LSR than at 15 % powder concentration (79 %). At 45 % RCD powder concentration higher (89 %) total volunteer weeds population stimulation was observed. The control treatment recorded reduced (38 %) total volunteer weeds

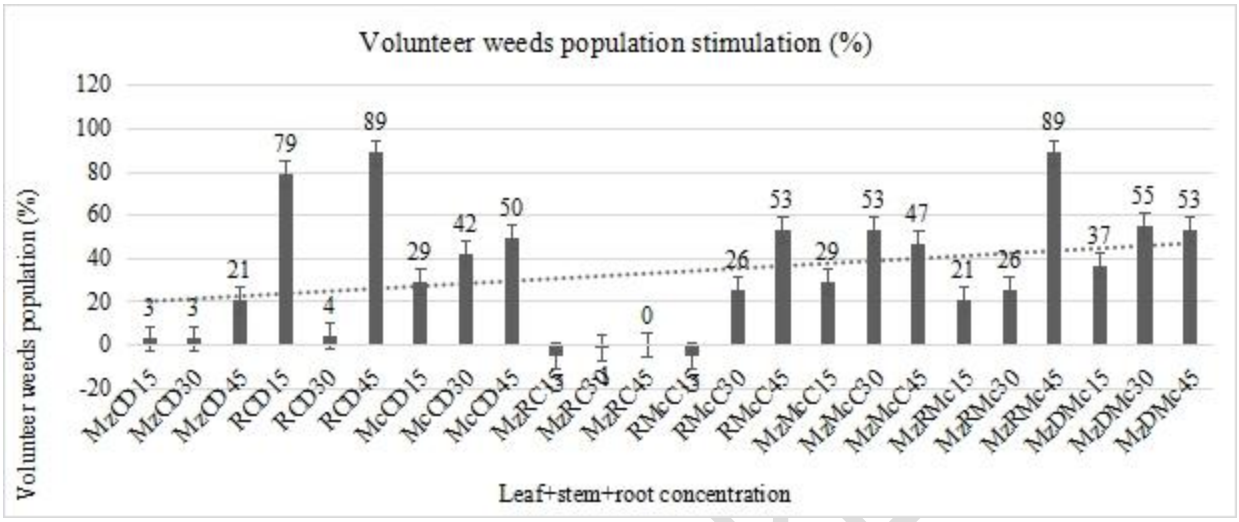

FIGURE 3: Effect of Leaf+stem+ root concentration on Volunteer weeds population

Note: mixed leaf/stem/root (LSR) powder concentrations (15-45 %) of rice (R), Cymbopogon (C), Desmodium (D), mucuna (Mc) and Maize (Mz).

3.3.4 Weeds biomass

The weed biomass significantly ($P < 0.05$), reduced with increases (15-45 %) in concentrations of LSR powders. This is shown by increasing percent biomass inhibitions/reductions in Figure 4. MzMc based powders had a higher biomass reduction effect at 45 % concentration (75%, 63% & 80%) followed by R+C (0.38g) and the least effect on weedbiomass was under C+D (0.45g, 0.46 g). Mz+C+D at 15 % LSR concentration recorded the highest total weed biomass (0.84 g) with lower weed biomass inhibition of 31%. The highest weed biomass (1.22 g) was under the control treatment.

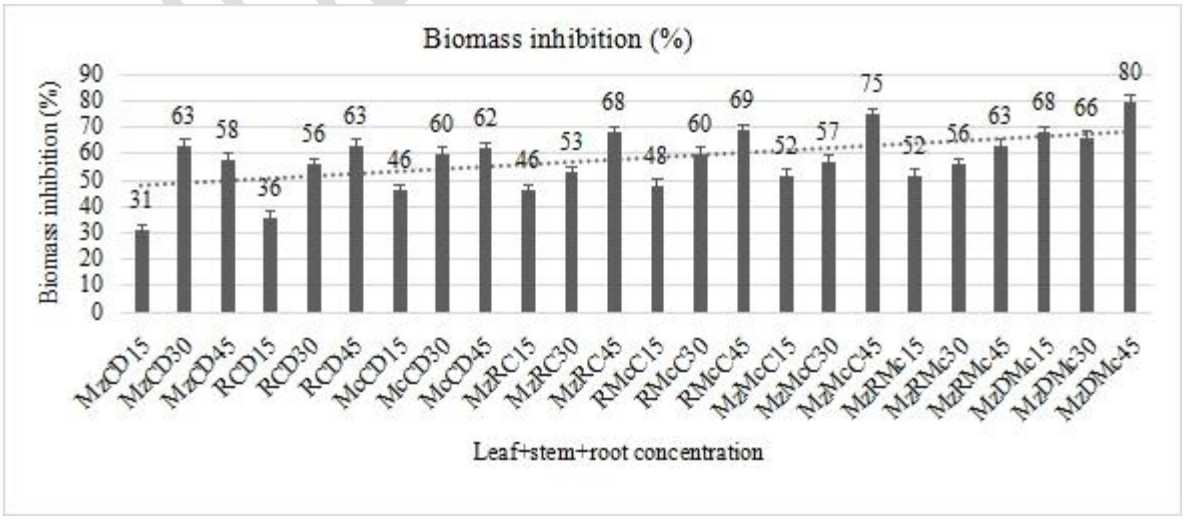

FIGURE 4: Effect of Leaf+stem+ root concentration on total weed Biomass
Note: mixed leaf/stem/root (LSR) powder concentrations (15-45 %) of rice (R), Cymbopogon (C), Desmodium (D), mucuna (Mc) and Maize (Mz)

3.3.5 Volunteer Dicot weeds (%)

Volunteer dicot weeds (VDW) were significantly stimulated at increasing (15-45 %) concentration of LSR powders (Figure 5). MzCD recorded lower (72 %) dicot weeds with 26% weed stimulation at 45 % concentration than other treatments. High populations of VDW (80 & 82 %) out of the total volunteer weeds were produced under C+D based mixtures.

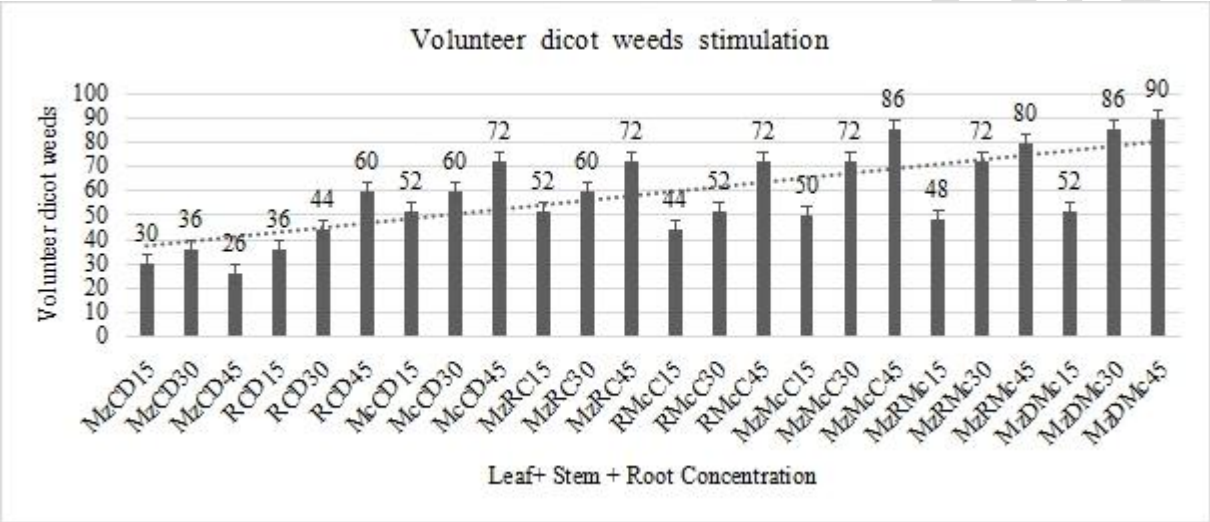

FIGURE 5: Effect of Leaf+stem+ root concentration on Volunteer dicot weeds percent stimulation
Note: mixed leaf/stem/root (LSR) powder concentrations (15-45 %) of rice (R), Cymbopogon (C), Desmodium (D), Mucuna (Mc) and Maize (Mz).

Higher VDW populations (86 %) were observed with increased VDW stimulation under R+C treatments. Significantly higher VDW (90, 93 & 96 %) were recorded under MzMc based mixtures that expressed higher weeds germination stimulation (80, 86 & 90%) at 45% LSR powder concentrations.

3.3.6 Volunteer Monocot weeds (%)

Increasing the concentration of LSR powders (15-45 %) increased the volunteer monocot weeds (VMW) inhibition and reduced the number of VMW (Figure 6). Addition of Mz+C+D extracts resulted into low inhibition on germination of VMW and subsequently recorded higher (17, 18 & 26 %) monocot weeds at 45 % concentration than other treatments. Low populations of VDW (14 %) were observed under R+C based LSR powders and the MzMc based mixtures recorded significantly lower (5, 7 & 9 %) VMW.

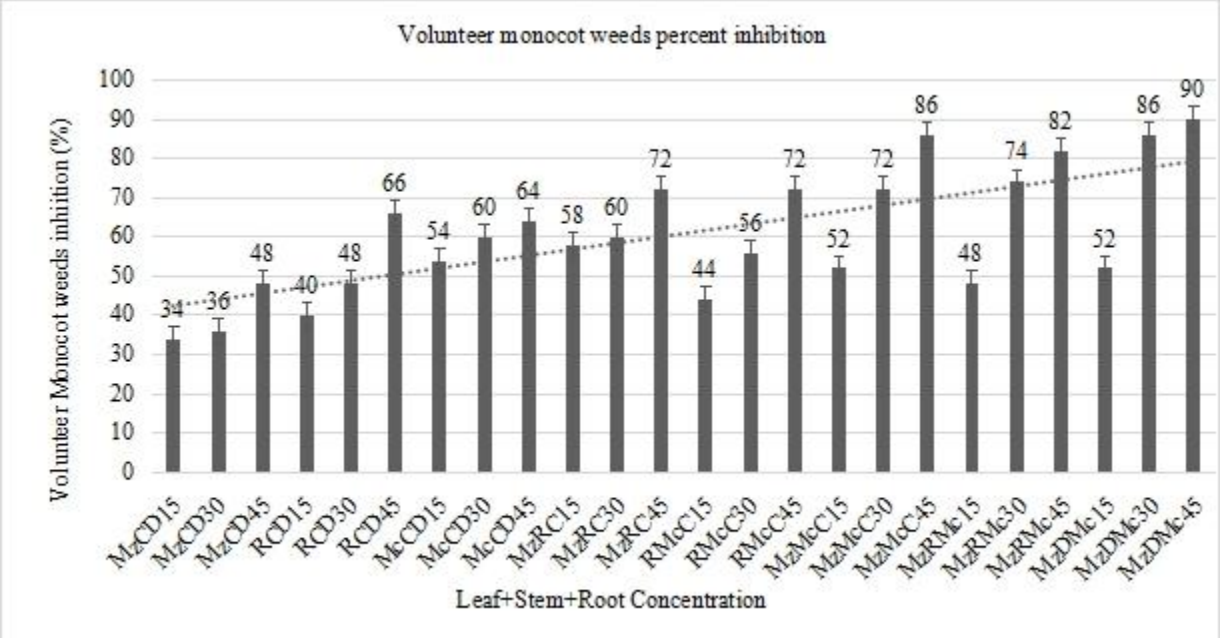

FIGURE 6: Effect of Leaf+stem+ root concentration on Volunteer monocot weeds percent inhibition
Note: mixed leaf/stem/root (LSR) powder concentrations (15-45 %) of rice (R), Cymbopogon (C), Desmodium (D), mucuna (Mc) and Maize (Mz).

4.DISCUSSION

4.1 *B. Pilosa*

Higher concentrations of all LSR powder increased percent inhibition and reduced the total germination of *B. pilosa*, total weedpopulation and biomass per treatment at 30 DAA. The *B. pilosa* weed population at 45 % was low under MzMc based powders but increased under R+C and a higher *B. pilosa* population was under C+D based powder. Relative reductions in *B. pilosa* weed germination, total weeds and biomass per treatment under MzMc based powders at 30 DAA may be attributed to increased negative allelopathic additive influences by the secondary metabolites in the powders on processes that promote plant growth and development such as radicle cell division and elongation, protein/ fatty acid synthesis and nutrient uptake. Two common terpenoids (Table 1) namely; 1, 4-eicosadiene and Butyrate hydroxytoluene besides the 3 phenols called; 2,5-di-tert-butylphenol; 3,7,11,15-tetramethyl-2-hexadecen-1-ol and (9Z)-9-icosen-1-ol, profiled from the stover of both Maize and Mucuna (20), could have additively inhibited germination, growth and development of *B. pilosa* and other weeds. The study crops were reported to exhibit allelopathic negative influences on weed growth and development (4, 5, 6).

MzMc based powders more effectively controlled the germination of *B. Pilosa,* population of volunteer weeds besides the total weed biomass than R+C and C+D based powders with increased (15-45 %) concentration. The lower weed control under R+C and C+D based mixed powder relates to possible weak synergy effects by common metabolites in NERICA 1 and *C. nardus* (Table 3.1) on processes that influence weed germination and development. Six terpenoids namely Butylated Hydroxytoluene; 3,4-Diethyl-1,1'-biphenyl, 1,2,3-Trimethyl-4-[(1E)-1-propenylnaphthalene; 2,2'-Diethylbiphenyl; 1-Ethyl-2-(1-

phenylethyl) benzene and 1,4-Eicosadiene besides 3 phenols called 2,5-di-tert-butyl- Phenol; 3,7,11,15-Tetramethyl-2-hexadecen-1-ol and (9Z)-9-Icosen-1-ol were commonly identified in NERCA 1 and *D. uncinatum* stover by (20). The terpenoids and phenols possibly weakly replaced each other on the basis of their biological exchange rate due to different molecular targets with the 10 unique terpenoids (citronellal, β-Citral, cis-geraniol, trans-carane, eugenol, geraniol acetate, β-elemen, caryophyllene, α-gurjunene and γ-cadinene) of *C. nardus*. The terpenoids and phenolic compounds were postulated to exhibit variable allelopathic inhibitive properties on weed growth and development as water extracts, (4) crop mulches (5, 6) and under intercropping ecosystems (18). *Cymbopogon citratus* essential oil was found to damage the membrane system of barnyard grass (*Echinochloa crus-galli* L.) by (19). Prapaipit *et al*., (2013) likewise noted inhibitory effects of *C. nardus* on the shoot and root growth of grass species at concentrations of 0.03 g dry weight equivalent extract/ ml. (20) similarly observed Citral of *C. citrates* to cause disruption of microtubules in wheat and *A. thaliana* L. roots. A putative allelopathic mechanism for *D. uncinatum* was found when an aqueous solution from *D. uncinatum* plants was applied (21). (22) reported that incorporation of fresh leaves of *M. pruriens* into the soil (1.0 % w/w) reduced succeeding emergence of *P. vulgaris* (60 %), and plant biomass (30 %). The results are further supported by researchers (12, 23) who reported that some phenolic compounds inhibited protein synthesis, amino acid transport and protein synthesis that reduced the integrity of DNA and RNA.

The population of *B. pilosa* increased following the rise of R+C+D (15-30 %) mixed powder concentrationbut the weed population reduced at 45 % concentration. The stimulated *B. pilosa* germination may probably be attributed to hormesis common with allelochemicals. Hormesis is the stimulation of growth due to enhanced physiological processes, at sub-optimal levels of allelochemicals common in auxin herbicides which mimic the growth hormone auxin, but which are lethal at higher doses. The observed hormesis may be accredited to antagonistic interactions between the 6 common terpenoids and 3 phenolic metabolites (24) observed in the stover of both NERICA 1 and desmodium with the 10 terpenoids and 1 ester (citronellal butyrate) of *C. nardus* profiled in the stover of the study plants (Table 1). Total number of weeds and biomass per R+C+D treatment however, declined with increasing (15-45 %) powder concentration due to probably increased synergistic effects of the common mixture of metabolites found in R & D. (25) indicated that herbicides applied in mixtures exhibited synergy compared with single herbicides under low infestation by weeds.

(26) reported that chemicals in a mixture have been reported to replace each other on the basis of their biological exchange rate or relative potency, due to similar molecular targets in the receiver plant and any departure of the effect is characterized by either reduced (antagonistic) or enhanced (synergistic) effects due to different molecular targets and exertion of biological processes such as photosynthesis, respiration, increased gaseous exchange and stabilization of the plants photosynthetic pigment. Hormesis was also earlier observed (26) when glyphosate promoted crop growth. Boosted weed growth due to application of allelopathic water extracts for weed control were reported (.27 28) The highest number of *B. pilosa* weeds under the control treatment, further supports the possible presence of Phyto-toxins in the

treatments. (29) remarked that the studies with herbicides are not done in sufficient detail to measure hormesis. Therefore, in many experiments hormesis has not been reported.

Mz+C+D exhibited the highest *B. pilosa* weed populationwith high weed biomass per treatment. This is linked to possible antagonism between molecules of the 4 terpenoids (1,4-Eicosadiene, Butylated hydroxytoluene, 1,2,3-trimethyl-4-[(1E)-1-propenyl]naphthalene & 3,4-diethyl-1,1'-biphenyl) and the 3 phenols [2,5-Di-tert-butylphenol, 3,7,11,15-tetramethyl-2-hexadecen-1-ol & (9Z)-9-icosen-1-ol)] commonly found in both maize and desmodium stover (Table 2). Antagonistic interactions cause no inhibition effect and result into action. The Terpenoids and Phenols possibly also exhibited weak synergistic molecular interactions with Ionene terpenoid and Falcarinol phenol of maize stover. The control treatment exhibited the highest number of *B. pilosa* weeds and total weed biomass per pot possibly due to lack of negative allelopathic effects in the receiver plant. (13) attributed hormesis to enhancement of positive effects on the physiological processes that promote germination and crop development.

4.2 Volunteer weeds

All the volunteer weeds (Monocot and Dicot) were promoted by higher LSR powder concentrations (15-45 %). This relates to the higher weed control of *B. pilosa* exhibited with increased LSR powder concentrations. As more of the sown *B. pilosa* weeds were controlled, more volunteer weeds germinated. This may be associated with increased exposure of the top soil in the pots to ambient conditions for volunteer weed seed germination under conditions of controlled *B. pilosa*. The similar VBW and VMW under the control treatment was a result of no treatment effect on the probably equally available seed bank for both classes of seeds. MzMc based powders presented higher efficacy in reducing weed germination and biomass relative to R+C and C+D based powders with increased (15-45 %) powder concentration. This relates to possible weak synergy effects by Terpenoids and phenolic acids in R+C and C+D relative to the additive effects of metabolites in MzMc (Table 1) and (24) on weed germination and development processes. R+C+D produced low total volunteer weeds at 30 % LSR powder concentration than at 15 % concentration but conversely a higher total volunteer weed population was at 45 % powder concentration. The declined volunteer weeds were possibly a negative response to the increased *B. pilosa* at 30 % concentration due to competition for resources such as water, light and nutrients. The control treatment similarly recorded a lower total number of volunteer weeds as a negative response to the high competition for resources with the high numbers of *B. pilosa* and VMW.

4.3 Dicot weeds

More volunteer dicot weeds (VDW) than volunteer monocot weeds (VMW) were produced at increased concentrations (15-45 %) of all the mixed LSR powders. The higher control of VMW may be attributed to additive and synergistic phytotoxic effects of molecules in the donor crops' stover, on enzyme processes involved in the germination and development of grass seedlings.

4.4 Monocot weeds

Germination of grass (Monocot) seeds is influenced by a chemical effect on amylase enzyme in seeds which catalyses the hydrolysis of starch, following imbibition of water, into simple sugars using Gibberellic

acid (GA). This is coupled with the hydrolysis of stored protein into amino acids. The germination of dicot seeds is not influenced by chemicals. The process involves imbibition of water and sugar is converted to simple sugars used in seed germination.

The SLR powders from NERICA 1, *C. nardus, Zea mays* and *M. pruriens* should be further investigated for potential production of triketone bio-herbicides. Bio-herbicides have high potential to control weeds since there are no reports of naturally occurring herbicide resistance to triketone herbicides and yet they are reported to have high efficacy (30). It is inferred from the studies that *Z. mays*, NERICA 1, *C. nardus* and *M. pruriens* have the potential to produce bio-herbicides based on terpenoids and phenolic acids that could effectively control weeds.

5.CONCLUSIONS

Major metabolites: Terpenoids and Phenols were identified from *C. nardus* classified as 10 terpenoids + 1 ester and each of NERICA 1 and *D. uncinatum* released 6 Terpenoids + 3 Phenols. Mucuna produced 2 Terpenoids + 4 Phenols and *Zea mays* vegetative material released 5 Terpenoids + 4 Phenols. The germination of *B.pilosa*, all weed and their biomass reduced with increasing concentrations of leaf, stem and root (LSR) powder. Volunteer dicot weeds however, increased at higher LSR powder concentrations. Raising the R+C+D; LSR powder concentration from 15 % to 30 %, on the contrary, increased the *B. pilosa* population due to hormesis. The MzMc and RC based mixed LSR powders most effectively reduced *B. pilosa* population, weed density, total weed biomass and volunteer monocot weeds. C+D based LSR powders, expressed lower inhibitory effects on all weeds seed germination and biomass. Inhibitory effects were attributed to possible influences of terpenoids and phenolic acids present in the crops' stover. Monocot weeds were more effectively controlled than broad leaved weeds by metabolites in the test plants. The study indicated that the terpenoids and phenolic acids in the vegetative parts of NERICA 1, *C. nardus*, *Z. mays* and *M. pruriens* inhibited the germination and growth of weeds. There is high potential to develop bio- herbicides for the control of weeds from the secondary metabolites of the donor crops.

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