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Original Research

Effects of Seed Proportion and Planting Pattern on Dry Matter Yield, Compatibility and Nutritive Value of *Panicum coloratum* and *Stylosanthes guianensis* Mixtures under Bako Condition, Western Oromia, Ethiopia

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Abstract

Panicum coloratum (PC) and *Stylosanthes guianensis* (SG) mixed pasture was established to assess the effect of seed proportion and planting pattern on Dry Matter Yield (DMY), compatibility and nutrient content of the mixed stand. Seeds of PC and SG were mixed as 50%PC+50% SG and 25% PC+75% SG and sown in same row, alternate row and broad casting pattern. A randomized complete block design with four replications was used to layout the experiment. The 50%PC+50%SG proportion resulted in significantly (p<0.01) higher DMY for PC (p<0.0001) while the 75% SG+25%PC mixture resulted in significantly higher DMY of SG (p<0.0001). For PC and SG, and total herbage yield, an alternate row planting pattern gave significantly highest (p<0.0001) yield, followed by broadcasting. Planting along the same row resulted in inferior values (p<0.0001) for all parameters, which perhaps is induced by the severity of competition between species. The RY (Relative Yield), LER (Land Equivalent Ratio), and CR (Competitive Ratio) values also imply similar trends for DMY of the component species. The legume DMY steadily increased from first to third year for the 75% SG+25% PC proportion. It can thus be concluded that the 50%PC+50SG proportion sown in alternate pattern was favorable for DMY in PC whereas the 25%PC+ 75%SG proportion combined with alternate row pattern was appropriate for the legume. Intercropping SG into PC sward at 25%PC+75%SG mixed proportions also improved the CP content, IVDMD and reduced NDF content.

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INTRODUCTION

Tropical pastures, in addition to their scarce availability (Birhanu *et al.,* 2013; Nina *et al.,* 2012), are low in quality, which among other factors, can be due to deficiency in soil nitrogen content. Other authors (Webster *et al.,* 1980; Albayrak and Türk 2011; Springer *et al.,* 2001; Byron *et al.,* 2000) also claimed similar phenomenon and suggested the need for integrating grass and legume species to produce herbage stands of better quality. But achieving balanced grass and legume mixed stands requires consideration of the interactions existing between the species, which can in turn be manipulated by the relative seed proportions and the pattern in which they are planted, among others (Hadji, 2000; Sanderson *et al.,* 1999).

Herbage DMY of *Panicum coloratum* and *Stylosanthes guianensis* mixed pasture under varying relative seed proportion was previously evaluated during establishment year (Diriba, 2002). The study showed 25%PC+75%SG planting proportion to be optimum when a respective base seed rate of 10 kg and 14 kg was used for grass and legume components, respectively. It was further stated that grass competition was severe, leading to inferior legume performance. To enhance the contribution of the legume component, optional agronomic strategies that help manipulate interspecies interaction and ensure balanced contribution of the component species to the total herbage mass and quality need to be designed (Diriba and Adugna, 2013). Information on these types of alternative options is generally inadequate. In the present study, it was thus hypothesized that planting pattern and seed rate proportion may manipulate interspecies interaction in a mixed stand leading to variable yield and quality contribution of the component species. The aim of this study thus was to assess the effect of planting pattern and seed proportion on herbage yield and nutrient content of mixed stands of PC and SG.

MATERIALS AND METHODS

Study Area

The experiment was conducted at Bako Agricultural Research Center (BARC) located in east Wollega zone of

Oromiya regional state, western Ethiopia from 2004 to2006. BARC is located at 9°6'N latitude and 37° 09' E longitude at a distance of 260kms west of Addis Ababa. Its altitude is 1650 masl. The months of rainy seasons at this area are from March (Bako Agricultural Research Center metreological summary report, 2006) to October with the peak rainfall in July. Mean annual temperature is 20°C with mean maximum temperature of 27°C and mean minimum 13°C.

Land Preparation and Planting

A fine seed bed was prepared using tractor drawn implements before the experimental plots were laid out. Then the plots were uniformly fertilized with DAP at a rate of 100 kg/ha during the year of establishment and the same rate of fertilizer was used during the later years. For same row and alternate row planting patterns 30cm between row spaces were maintained whereas the seeds for broadcasting were uniformly distributed over the plots to avoid, as much as possible, irregular spacing of the seeds. Weeds were removed manually as required to protect their negative effect on forage growth and yields and herbage was harvested each year (2004, 2005 and 2006) in October.

Treatments and Experimental Design

Seeds of *Panicum coloratum* (PC) cv. coloratum and *S.guianensis* (SG) cv. cook were mixed at two varying relative seed proportions (50%PC+ 50% SG and 25% PC+ 75% SG) and sown in three patterns (in the same row, alternate row and broad casting) in early June of 2004 with base seed rate of 10 kg/ha for PC and 14 kg/ha for SG on 3m x 4m plots. The plots were laid out in randomized complete block design with four replications.

Data Collection

Dry matter yield determination

Two middle rows (0.3m x 3m=0.9m^2) were randomly selected and harvested for forage yield determination from plots planted using a row planting pattern when the grass reached 50% flowering stage. In plots where broadcasting was used, a net area of land similar to that of the two rows was harvested from the middle of the plots. The herbage was cut manually using sickle and fresh weight for each sample was recorded in the field using a field balance immediately after mowing. Chopped subsamples (300gm) of each treatment were dried in the oven at 105^{0}C for 24 hours to determine the DMY on hectare basis. Samples from each treatment used for quality assessment were oven dried at 60°C for 72 hours and maintained for laboratory analysis packed in a paper bag. The percentage contribution of the component species to the total DM yield was determined by dividing the DM yield of each of the components by the total DM yield of each treatment.

Land Equivalent Ratio (LER)

The LER is defined as the amount of land required under monoculture to obtain the same dry matter yield as produced in the intercrop. It is calculated according to the equation proposed by (Willey and Rao, 1980) as follows:

LERab = (Yab/ Yaa) + (Yba/ Ybb)

Where, Yaa = sole crop yield of species 'a'; Ybb = sole crop yield of species 'b'; Yab = inter crop yield of species 'a' in combination with species 'b' and Yba = inter crop yield of species 'b' in combination with species 'a'. If LERab > 1, there is yield advantage.

The relative DM yields (RY) of the components in the mixtures were calculated using the equations of De Wit (1960) as:

RYG = DMYGL/DMYGG (1)
RYL = DMYLG/DMYLL (2)

where DMYGG is the dry matter yield of any perennial grass 'G' as a monoculture; DMYLL is the dry matter yield of any perennial legume 'L' as a monoculture; DMYGL is the dry matter yield of any perennial grass component 'G' grown in mixture with any perennial legume 'L'; and DMYLG is the dry matter yield of any perennial legume component 'L' grown in mixture with any perennial grass 'G'.

Relative total yield (RTY) was calculated according to the formula of De Wit (1960):

RTYGL = (DMYGL/DMYGG) + (DMYLG/DMYLL) (3)

Competitive ratio (CR) was calculated according to Willey and Rao (1980) as:

$$CR_{ab} = (DMY_{ab}/DMY_{aa}) \div (DMY_{ba}/DMY_{bb})$$
$$CR_{ba} = (DMY_{ba}/DMY_{bb}) \div (DMY_{ab}/DMY_{aa})$$

Where, $_{ab}$ is the performance of *P.coloratum* 'a' mixed with *S. guianensis* 'b'; $_{ba}$ is the performance of sweet clover 'b' mixed with Rhodes 'a'; $_{aa}$ is the performance of *P. coloratum* 'a' as a monoculture; and $_{bb}$ is the performance of *S.guianensis* 'b' as a monoculture. Z is the sown proportion/ratio.

Chemical Composition

DM and ash were determined following AOAC (1980). Neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) were determined according to Van Soest and Robertson (1985). Hemicelluloses and cellulose were calculated as NDF-ADF and ADF-ADL, respectively. The N content of the samples was determined by the micro-Kjedal method and CP was calculated as N x 6.25. The *in vitro* DM digestibility was determined using Tilley and Terry (1963) *in vitro* method as modified by Van Soest and Robertson (1985).

Statistical Analysis

The DM yield and chemical composition data were analyzed using the General Linear Model Procedure of the SAS computer software (SAS, 1996). The following model was fitted to the data:

$$Y_{ijk}=\mu + \tau_i + \beta_j + \varepsilon_{ijk}$$

where,
μ=overall mean of the population
τ_i= The ith treatment effect
β_j= The jth block effect and
ε_{ijk}=random error associated with yij

Significant differences between means were separated at $p\leq0.05$ using Tukey honestly significant difference test.

RESULTS AND DISCUSSION

Table 1 shows the statistical effects of seed proportion treatments on herbage DMY of the component species. The grass component gave significantly higher yield (p<0.01) in 50%SG+50 PC% proportion compared to 75% SG+25%PC. However, the 75%SG+25%PC proportion resulted in better (p<0.0001) yield for SG as compared to 50%PC+50%SG. The total DMY was also higher for the 50%PC+50%SG, and this observation agrees with results reported from a similar previous study. In a study in which mixtures of PC and *Desmodium unicenatum* (DU) were evaluated. Diriba *et al.* (2004) for example reported higher total DMY (11.63t/ha) for the 50%PC+50%DU proportion. The highest (p<0.0001) total DMY (3.74t/ha) for 50%PC+50%SG seed proportion compared to 75%SG+ 25%PC suggests the aggressivity of the grass component even at low proportion in total seed mass. Baba *et al* (2011) also reported comparable results in Guinea grass and Centro mixtures. In spite of higher proportion of the legume in 75%SG+25%PC mixture, the grass gave close to 5 times more yield than the legume, which is apparently an indicative of its higher aggressivity.

Table 1: Effect of seed proportion of component species herbage DMY of component species.

Seed proportions	DMY		
	PC	SG	Total
50%PC : 50%SG	3.46^a	0.26^b	3.74^a
25%PC : 75%SG	2.48^b	0.50^a	2.98^b
SE	**0.22**	**0.11**	**0.23**
Significance level	****	****	****

Means with different superscripts within each column are significantly different. LER= land equivalent ratio; SE= standard; PC= *P.coloratum*; SG=*S.guianensis*

The herbage yield of component species as influenced by planting pattern is presented in Table 2. Component species and total stand yields were significantly (p<0.0001) higher for alternate row pattern followed by broadcasting. The with-in same row pattern resulted in lower (p<0.0001) herbage yield values for both species, which is possibly due to a negative competitive interaction between the species. It was also apparent that with-in same row pattern negatively affected the legume component as compared to broadcasting. In plots where broadcasting was used, seeds of the two species are normally distributed at relatively distant positions spatially, leading apparently to low competitive interaction as opposed to with-in same row pattern. In the latter case, seeds of the two species fall closer to each other, leading to severe competition, resulting in poor performance of the legume counterpart. The alternate row planting pattern was found to be the most favorable strategy for the grass, the legume and their mixed stand as compared to with-in same row and broadcasting patterns.

Table 2: Effect of planting pattern on herbage DMY of PC and SG mixed pasture.

Planting pattern	DMY		
	PC	SG	Total
Same row	2.50^c	0.26^b	2.79^c
Alternate row	3.54^a	0.56^a	4.10^a
Broadcasting	2.87^b	0.32^b	3.1^b
SE	**0.22**	**0.11**	**0.23**
***p*-value**	**0.0001**	**0.0001**	**0.0001**

Means with different superscripts within each column are significantly different. LER= land equivalent ratio; SE= standard; PC= *P. coloratum*; SG=*S. guianensis*

The relative yields (RY) of PC and SG as affected by seed proportion and planting pattern were given in Table 3. Relative yield of the grass component at 50%PC+50%SG proportion (0.96) and at 25%PC+75%SG (0.69) were significantly (p=0.0001) higher than the legume at both seed proportions, respectively. The legume had superior RY values at 25%PC+75%SG than at 50PC+50SG proportions (Table 3). Similarly, Lithourgidis *et al* (2006) reported that for common vetch grown in mixture at 65% vetch+35% oat, the legume component performed better than the grass. Though the 25%PC+75%SG proportion was seemingly appropriate for the legume, yet the grass was found to have higher RY values in the order of 3% more than that of the legume, which evidently suggests the higher competitive ability of PC over SG. The total RY for 50PC+50SG proportion (1.07) indicates that there was 7% more biomass yield in mixed sward of the grass and the legume than either of their sole stand whereas the 0.91 total RY for 25%PC+75%SG proportion shows that there was a loss of 9% yield which could be achieved when the grass was established alone.

A similar trend was also observed for planting pattern treatments where the grass was dominant across the patterns evaluated too. The RY values of both components followed similar pattern with that of DMY (Table 1) in that the alternate row pattern resulted in significantly (p=0.0001) higher RY followed by that of broadcasting, and with the least value for the with-in same row pattern. This implies that the RY is directly proportional to the DMY of each of the components in the mixture. The grass in alternate row pattern also resulted in higher RY values (0.98), being nearly fourfold higher than that of the legume component. As can be inferred from the main effects of both factors, the legume performed poorly in relation to the grass for the on agronomic traits assessed.

Table 3: Relative yields (RY) of PC and SG as affected by seed proportion and planting patterns.

Seed proportions	RY		
	PC	SG	Total
50%PC : 50%SG	0.96^a	0.11^b	1.07^a
25%PC : 75%SG	0.69^b	0.22^a	0.91^b
SE	**0.0011**	**0.0041**	**0.0010**
Significance level	****	****	****
Planting patterns			
Same row	0.70^c	0.11^c	0.81^c
Alternate row	0.98^a	0.25^a	1.23^a
Broadcasting	0.80^b	0.14^b	0.93^b
SE	**0.024**	**0.0067**	**0.0058**
Significance level	****	****	****

Means with different superscripts within each column are significantly different. RY= relative yields; SE= standard error; PC= *P. coloratum*; SG=*S. guianensis*

Results from the analysis of variance for competitive (CR) and land equivalent (LER) ratios of the mixtures as affected by seed proportion and planting pattern is presented in Table 4. The 50%PC+50%SG combination gave significantly (p=0.0012) higher CR than the 25%PC+75%SG. A reverse pattern to that of the grass was observed for the legume component for the seed proportion treatments. For 50%PC+50%SG combination, the CR of the legume was less by 64% than the 25%PC+75%SG, suggesting SG to be less competent in 50%PC+50%SG. Regarding the effect of planting pattern, the grass was more favored in the with-in same row pattern (p=0.0001). The least CR for the grass was observed in alternate row indicating that its negative competitive effect over the legume was less in this pattern compared to with-in same row. On the contrary, the legume appears to be more competent (p=0.001) in alternate row planting pattern followed by the broadcasting pattern.

The 50%PC+50SG proportion had 11.5% yield advantage over the sole planted grass. On the other hand, 75%SG+25%PC mixture gave 9% less yield advantage compared to its sole stand. This indicates that SG was not favored by the 50%PC+50SG proportion unlike the latter case. On the other hand alternate row planting resulted in 19.35 more yield over the sole grown stands. Broadcasting and same row planting patterns resulted in 2% and 17% less yield advantages, respectively. This indicates that competition between the grass and legume in alternate row planting pattern was inferior compared to that of within same row and alternate row ones. This could be due to relatively wider space between the rows of the grass and legume stands and less competition for soil nutrients among them. This suggests that the alternate row method of sowing created better opportunity for the legume component to compete the grass in the mixture.

Table 4: Competitive (CR) and land equivalent (LER) ratios of PC and SG mixture as affected by seed proportion and planting patterns of the component species.

Seed proportions	Parameters		
	CR (PC)	CR (SG)	LER
50%PC : 50%SG	8.80^a	0.114^b	1.13^a
25%PC : 75%SG	8.15^b	0.318^a	0.91^b
SE	**0.139**	**0.003**	**0.14**
***p*-value**	**0.0012**	**0.0004**	**0.003**
Planting patterns			
Same row	5.956^a	0.168^b	0.83^b
Alternate row	3.273^b	0.314^a	1.24^a
Broadcasting	5.701^a	0.176^b	0.98^b
SE	**0.255**	**0.022**	**0.14**
Significance level	****	***	****

Means with different letters within each column are significantly different. CR= competitive ratio; SE= standard error; PC= *P.coloratum*; SG=*S.guianensis*

The effect of year on herbage DM yield of the mixture is presented in Table 5. Significantly (p<0.0001) higher PC and total stand DMY was recorded during second year. Similarly, legume DMY was observed to exhibit an increasing trend with age. During the first year, the legume DM yield was inferior; but ended to rise with age, while the grass DMY declined in the third year. The total DMY was least in the third year, during which yield values for the legume was also superior while that of PC was in declining status. This indicates that SG contributed less to the total DMY especially in the establishment and second years as compared to the grass component. Although the proportion of SG reached 43% during the third year, the total DMY remained least. The declining trend in total mixture DMY with advancing stand age concurs with previous observations (Diriba, 2002) and suggests the need for partial or total renovation. The change in DMY with advancing stand age is depicted in Figure 1.

Table 5: Year effect on DMY of PC and SG mixed pasture

Year	Parameters			
	LER	DMY (PC)	DMY (SG)	DMY (total)
1	1.21^a	3.17^b	0.12^c	3.30^b
2	1.10^a	3.88^a	0.28^b	4.20^a
3	0.74^b	1.84^c	0.75^a	2.59^c
SE	**0.18**	**0.27**	**0.13**	**0.28**
***p*-value**	**0.0001**	**0.0001**	**0.0001**	**0.0001**

Means with different letters within each column are significantly different. SE= standard error; PC= *P. coloratum*; SG= *S. guianensis*

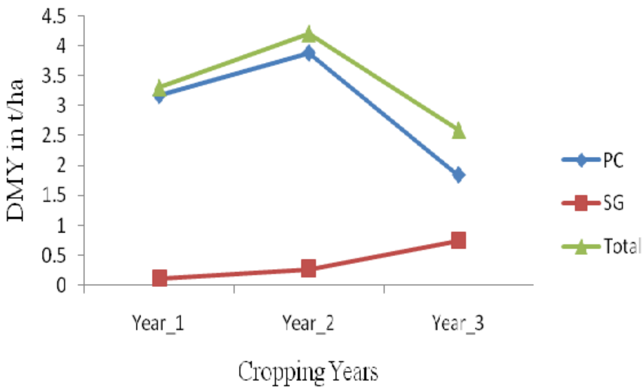

Figure 1: Effect of years on DMY of PC & SG mixed pasture.

The combined effect of the various factors on herbage DMY is given in Table 6. Significantly (p<0.0001) higher herbage yield was achieved from the mixed stand as compared to the sole grass stand. The mixed stands produced 27.75% more herbage yield than the sole grass stand, which agrees with reports in related literatures (Sturludóttir, 2011; Hadji, 2000). The 50%PC+50%SG mixture in combination with alternate row planting pattern gave significantly (p<0.0001) higher herbage DM yield values for consecutive three years.

Table 6: The effect of seed proportions and planting pattern on DMY of PC and SG mixed pasture as affected by year.

Treatments	Years		
	Year one	Year two	Year three
Sole PC stand	3.28^{bc}	3.96^{bc}	3.54^a
50%PC+50%SG, same row	2.83^{cd}	4.13^{bc}	2.04^{bc}
50%PC+50%SG, alternate row	4.54^a	5.49^a	3.71^a
25%PC+75%SG, same row	3.69^b	4.32^b	2.95^{ab}
25%PC+75%SG, alternate	2.43^d	3.34^c	2.00^{bc}
50%PC+50%SG, broadcasting	3.50^{cd}	4.50^b	2.86^{abc}
25%PC+75%SG, broadcasting	2.81^{cd}	3.40^c	1.97^c
SE	**0.27**	**0.29**	**0.32**
LSD	**0.81**	**0.87**	**0.95**

Means with different letters within each column are significantly different; SE= standard error of the mean; LSD= List significant digits; PC= *P. coloratum*; SG=*S. guianensis*

Chemical composition and *in vitro* DM digestibility of the grass and legume samples in pure stands and mixtures is presented in Table 7. The mean DM content of the grass and the legume components are comparable. The ash content of the legume was slightly higher than that of the grass. As would be expected the mean CP content of the legume was much higher than that of the grass component, but differences within the mixture treatments was narrow for both. The NDF was higher for the grass and lower for the legume, but the reverse was true for ADF and ADL contents. The mean IVDMD was slightly higher for the legume component.

Table 7: Chemical composition and *in vitro* DM digestibility (% DM) of the grass and the legume component grown in pure stands and mixtures.

Grass	Chemical composition and *in vitro* digestibility						
	DM	**Ash**	**CP**	**NDF**	**ADF**	**ADL**	**IVDMD**
100PC	92.22	9.34	8.69	73.83	47.36	6.65	55.74
50 PC+50 SG	92.39	9.63	8.69	72.69	43.84	6.17	60.00
25 PC+75 SG	92.1	9.64	9.56	71.98	43.79	5.98	59.76
Mean	**92.24**	**9.54**	**8.98**	**72.83**	**45.00**	**6.27**	**58.50**
Stylosanthes							
50SG+50 PC	91.46	11.36	18.87	56.2	49.77	15.24	61.49
75 SG+25 PC	91.57	10.72	19.81	57.61	50.78	15.52	60.51
100 SG	92.15	10.97	19.56	55.85	50.21	16.63	63.16
Mean	**91.73**	**11.02**	**19.41**	**56.55**	**50.25**	**15.80**	**61.72**

PC= *P. coloratum*; SG=*S. guianensis;* DM= dry matter; CP=crude protein;
NDF= neutral detergent fiber; ADL= acid detergent lignin; IVDMD= *In vitro* dry matter digestibility

Legumes, grasses and grass-legume mixtures containing greater than 19% CP are rated as having prime standard and those with CP values lower than 8% are considered to be of inferior quality (Kazemi *et al*., 2012). The mean CP content of Stylosanthes in the present study (19.41%) is comparable to the indicated critical value but that of the grass is remarkably lower. Also, it is apparent that the legume component had CP levels greater than 15%, a level which is usually required to support lactation and growth (Nsahlai *et al*., 1996), suggesting the apparent role of legume integrations in improving overall nutritional quality of mixed stand herbage.

According to Singh and Oosting (1992), feeds containing NDF values of less than 45% are classified as high, those with values ranging from 45% to 65% as medium, and those with values higher than 65% as having low quality. Generally NDF values in both samples were higher, with that of the legume component falling in the medium quality range. The quality of the grass component was evidently low, and this high cell wall content can be a limiting factor to feed intake as DM intake and NDF content are negatively correlated (Ensminger *et al*., 1990). The mean NDF content of the legume (56.55%) observed in the present work is higher than that reported previously (47%) for eleven herbaceous (Seyoum *et al*., 1996) and eight browse legume (46%) species (Seyoum *et al*., 1999). The mean IVDMD for both components was higher than the critical threshold level of 50% required for feeds to be considered as having acceptable digestibility (Owen and Jayasuriya, 1989).

CONCLUSION

Both the legume and the grass DMY performances were highest for the alternate row planting pattern. The DMY of the legume steadily increased from first to third year for the 75% SG to 25% PC seed proportions in the mixture. The total DMY of the mixed components was found to be determined more by DMY of the grass during the first and second years indicating that the legume couldn't compete well during these years. Even though the legume component shown better compatibility at the third cropping year, the total DMY was less than both the first and second years. The competition indices, RY LER and CR, all indicated that the competition was severe against the legume in 50% PC: 50%SG seed proportions and same row planting patterns. Hence it can be concluded that 50% seed proportion for each of the components sown in alternate rows were the best practice in favor of forage biomass yield whereas the 25%PC with 75%SG seed proportions in alternate row planting methods was for the better persistence of the legume component in the mixture according to this experiment. Intercropping SG into PC sward at 25%PC with 75%SG mixed proportion also improved the CP content, IVDMD and reduced NDF content.

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