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Panicle removal increased stalk ethanol production of sweet sorghum [Sorghum bicolor (L.) Moench]

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Abstract: *The objective of this research was to increase the ethanol production of sweet sorgum stalks by panicle removal. This field experiment was conducted using a randomized complete block design with three replicates. The first factor was stages of panicle removal (50% booth, 100% flowering, and 50% grain filling) and non-panicle removal as control. The second factor was the variety of sweet sorghum (White Local Belu, Numbu, Kawali and Sugar Graze). Variables measured were seedling emergence, stages of 50% booth, 100% flowering and 50% grain filling, stalk fresh weight, juice production, sugar content (brix), ethanol content and ethanol production. Results showed that panicle removal increased the ethanol content and ethanol production of the three varieties except var. Numbu. Panicle removal at stage of 100% flowering or 50% grain filling resulted in the highest stalk ethanol content and ethanol production. Var. Sugar Graze gave the highest ethanol production (2084.51 L ha^{-1}) when panicle was removed at 100% flowering or 2080.43 L ha^{-1} when was done at 50% grain filling stages. The ethanol content of var. White Local Belu was lower than those of the other three improved varieties.*

Keywords: *Ethanol, Panicle removal, Sweet sorghum [Sorghum bicolor (L.) Moench, Varieties*

I. Introduction

The need for finding an alternative sustainable source of energy is important these days due to less and less available fossel sources. Sweet sorghum is one of cereal crops with several function such as grain or sweet forage (Almodares et al., 2008a). Although it produces grain, the essence of the crop is from the stalk wich contains high sugar contents (Almodares et al., 2008b). The sugar content in the stalk generally reaches 54 - 69 t/ha (Almodares et al., 2008b). The sugar contents in the sweet soghum juices vary with varieties. Brix among sweet sorghum varieties ranges between 14.32–22.35% (Almodares et al., 2006). The juice could be converted into 85% ethanol (54.4 L ethanol per 100 kg fresh stalk) (Rains et al., 1993). Sweet sorghum could produce bioethanol of 4000-7000 L/ha/year therefore for producing 60 000 000 KL/year as many as 15 000 0000 land was needed (Yudiarto, 2005). Results of previous experiment indicated that three sweet sorghum varieties originated from Japan (FS501, FS902 dan KCS105) and one local variety (Red Local Belu) were adapted to dryland of Jimbaran, Badung regency, Bali provinsi of Indonesia (Agung et al., 2013). Besides, sweet sorghums are tolerant to dry environment and highly efficient in using water and has high photosynthesis efficiency compared to other crops. Like other biofuel resource crops, sweet sorghum results in bioethanol which will not increase CO2 emission to the atmosphere.

Panicle removal before pollination and grain filling was reported to be able to influence sugar content of sweet sorghum due to photosynthates for grain set are able to store in the form of sugar in sorghum juices (Erickson et al., 2011). There are limited data available for supporting the hypothesis, but brix value was reported higher in sweet sorgum with panicle removed before grain setting (Broadhead, 1973). Rajendran et al. (2000) indicated that panicle removal increased sugar yields compared to non-panicle removal sweet sorghums. Information on increasing stalk ethanol production by removing panicles of sweet sorghum was limited particulaly of varieties grown in Indonesia. Therefore this research was needed to obtain high etanol production from the crops.

II. Materials and Methods

2.1 Study area

The field experiment was conducted in village of Biaung, Badung regency, Bali province at ± 10 m above sea level from May to November 2015. Analysis indicated that before the experiment the soil pH of 6.8 (normal), organic-C of 2.18% (moderate), total-N of 0.14% (low), available-P of 117.25 ppm (very high), available-K of 169.18 ppm (moderate), soil misture content at field capacity of 48.84% and claily-loam texture (Laboratory of Soil Science Faculty of Agriculture Udayana University, 2015). The field experiment was arranged as randomized complete block design, in which two treatment factors were imposed. The first factor were four sweet sorghum varieties (White Local Belu, Numbu, Kawali and Sugar Graze) and the second factor were three stages of panicle removal (50% booth, 100% flowering, 50% grain filling) and without panicle removal. Whole treatments were randomly arranged and replicated three times. The stage of fifty percent booth

(3 cm swollen heads), 100% flowering (full flowering at middle row plants, 50% grain filling (grains produced at middle row plants). The experiment was carried out for seven months (from May to November 2015).
The experiment consisted of 48 plots, which was 3.5 m x 1.5 m in size. Fertilizers of 50 kg SP36, 50 kg ha^{-1} KCl and 20 t ha^{-1} composts were applied a week before planting. Urea of 100 kg, was given twice i.e. seven days before planting and 30 days after planting (DAP). Five seeds were planted into a hole of 70 cm x 25 cm spacing and thinned at 14 DAP to keep one healthy seedling per hole or to maintain 30 plants per plot (population of 51.143 plants ha^{-1}). Spray-watering was given twice a day particularly at early plant growth until flowering and afterward watering was done when necessary. Weeding was carried out once at 14 DAP.

Panicle removal was done by removing panicle at 50% booth, 100% flowering, 50% grain filling stages after which the plants were harvested. Plants without panicle removed were maintained until heading stage. Observations and measurements were conducted to seedling emergence, stalk fresh weights, stalk sugar content (*brix)*, juice production, ethanol content and ethanol production after each harvest. Stalk fresh weights were determined from a quadran of 12 plants (1.26 m^2) in each plot, which then was converted into fresh weight per hectare. Juice production was measured by crushing the stalk through the sugar cane crusher after which the juices were collected. Sugar content was determined using hand refractrometer and the juice was then analyzed in the laboratory to determine ethanol content through fermentation and distillation process. Fermentation process was done for 60 hours and further continued with distillation process to primarily measured the ethanol content before the final content was determined using 3300 variant Gas Chromatography isothermally with stalk injector temperature of 150°C, column temperature of 150°C and detector temperature (FID) of 200°C (Laboratory of Analytic Udayana University, 2015). The ethanol production was calculated from the ethanol content (%) multiplied by juice production per hectare.

2.2 Statistical analysis

Data were subjected to ANOVA Statistical analysis using Costat and MstatC computer software and comparation of means were calculated using Duncan's Multiple range test and Least Significant Different analysis at 5% level (Gomez and Gomez, 2007). Data were transformed where necessary.

III. Results and Discussion

3.1 Percentage of seedling emergence

Among the four varieties, Numbu, Sugar Graze and Kawali apparently showed higher percentage of emergence at 3, 7 and 9 DAP compared to White Local Belu (Table 1). At 7 DAP those varieties had respectively 77.2%, 72.8% and 67.8% seedling emergence, while White Local Belu only had 62.8%. Varieties of Sugar Graze and Numbu had fully grown at 9 DAP, Kawali grew 89.2%, which was significantly higher than that of White Local Belu. These indicated that those three improved varieties had genetically better growth than Local variety.

Table 1. Percentage of seedling emergence at 3, 7 and 9 DAP.

Variety	Days after planting (DAP)		
	3	**7**	**9**
White Lokal Belu	50.8 ^{c}	62.8 ^{b}	76.9 ^{c}
Numbu	60.8 ^{ab}	77.2 ^{a}	94.7 ^{ab}
Kawali	54.7 ^{b}	67.8 ^{ab}	89.2 ^{b}
Sugar Graze	63.9 ^{a}	72.8 ^{a}	98.1 ^{a}
5% LSD	6.95	9.47	4.95

Notes: Means followed by the same letters at the same columns are not significantly different at 5% level of LSD.

3.2 Stages of 50% booth, 100% flowering and 50% grain filling of each variety

Kawali was the fastest booth (59.33 DAP) to reach the stage of 50%, followed by Numbu variety (Table 2), while Sugar Graze and White Local Belu were the slowest.Kawali and Numbu varieties also the fastest to reach the stages of 100% flowering and 50% grain filling, while Sugar Graze and White Local Belu varieties were the longest. The varieties with non-panicle removal (control) reached the age of harvest at 120 DAP.

Table 2. Stages of 50% booth, 100% flowering and 50% grain filling of each variety

Varietas	Stages of panicle removal			
	Harvest (Non-panicle removal)	50% booth	100% flowering	50% grain filling
White Local Belu	120	72.00^{a}	76.67^{a}	91.33^{a}
Numbu	120	64.00^{b}	72.00^{c}	84.00^{b}
Kawali	120	59.33^{c}	72.00^{c}	84.00^{b}
Sugar Graze	120	69.00^{a}	73.67^{b}	91.33^{a}
5% LSD	-	3.87	0.67	0.67

Notes: Means followed by the same letters at the same columns are not significantly different at 5% level of LSD.

3.3 Stalk fresh weights and juice production

The interaction between stages of panicle removal and variety did not significantly (p>0.05) affect stalk fresh weights and juice production. These parameters were also not affected by the stages of panicle removal but were significantly (p<0.05) affected by variety (Table 3). Sugar Graze variety had the highest (41.73 ton ha^{-1}), which was respectively 12.22%, 10.59% and 32.27% higher stalk fresh weights compared to White Local Belu, Numbu and Kawali. The highest stalk fresh weights of Sugar Graze variety was possibly due to the longest time to reach the stages of 50% booth, 100% flowering and 50% grain filling causing more assimilates stored in the stalks (Table 1).

Table 3. Stalk fresh weights and juice production of sweet sorghum varieties at different stages of panicle removal

Treatments	Stalk fresh weights (ton ha^{-1})	Juice production (KL ha^{-1})
Stages of panicle removal:		
Non-panicle removal	35.10^{a}	10.93^{c}
50% booth	36.86^{a}	13.17^{b}
100% flowering	39.16^{a}	15.29^{a}
50% grain filling	37.92^{a}	14.36^{a}
5% LSD	-	1.05
Variety:		
White Local Belu	37.40^{b}	7.94^{c}
Numbu	37.95^{b}	14.39^{b}
Kawali	31.73^{c}	14.72^{b}
Sugar Graze	41.97^{a}	16.70^{a}
5% LSD	3.96	1.05

Notes: Means followed by the same letters at the same columns and treatments are not significantly different at 5% level of LSD.

Juice production were affected significantly (p<0.05) by stages of panicle removal as well as variety (Table 2). Panicle removal at 100% flowering and 50% grain filling resulted in the highest (15.29 KL ha^{-1} and 14.36 KL ha^{-1} respectively) juice production, which were 39.89% and 31.38% higher than those of non-panicle removal treatment. The economic value of sweet sorghum is in the stem. The finding indicated that panicle removal resulted in assimilates supposedly used in grain formation and development had been diverted into the stems (Balole, 2001). Inhibition of flowering with some chemicals resulted in considerable increases in cane and sugar yields of sugarcane (Humbert et al.,1967). An increase of sucrose content in plants with panicle removal at boot stage compared to those at later stages was reported by Broadhead (1973). It was observed that juice quality was generally better in plants with panicle removal at early stages compared to those at the late developmental stages. That may be due to the fact that accumulation of sugar in sweet sorghum stems begins during the development of inflorescence (McBee and Miller, 1982). Variety of Sugar Graze produced the highest (16.70 KL ha^{-1}) stalk juice, which was 110.33%, 16.05% and 13.45% higher than those of White Local Belu, Numbu and Kawali respectively (Table 3). This finding was believed to be associated with the highest stalk fresh weights of Sugar Graze variety (Table 2).

3.4 Stalk sugar content (brix)

The stalk sugar content (*brix*) was significantly (p<0.05) affected by interaction between stages of panicle removal and variety of sweet sorghum (Table 4). Panicle removal increased stalk sugar content of two varieties except Numbu and Sugar graze. The sugar content (*brix*) of White Local Belu only increased when panicle were removed at 50% grain filling probably due to more assimilates accumulated in the stalk up to that stage although juice production were not significantly different among four varieties (Table 3). Panicle removal on Kawali variety at 50% booth stage resulted in the highest (11.10%) stalk sugar content, which was not significantly different from that of Numbu with panicle removal at stages of 100% flowering and 50% grain filling. Kawali variety reached 50% booth stage in the shortest time (Table 2) indicating the highest sugar content had been accumulated in the stalk and panicle removal afterwards therefore decreased the sugar content (Table 4).

Table 4. Effect of interaction between stages of panicle removal and variety on sugar content (brix)

Variety	Stages			
	Non –panicle removal	50% booth	100% flowering	50% grain filling
White Local Belu	8.00^{f}	6.93^{g}	7.87^{f}	8.80^{d}
Numbu	10.67^{b}	8.33^{ef}	10.73^{ab}	11.00^{ab}
Kawali	6.80^{g}	11.10^{a}	8.67^{de}	9.00^{d}
Sugar Graze	10.00^{c}	10.00^{c}	9.67^{c}	9.73^{c}

Notes: Means followed by the same letters at the same rows and columns are not significantly different at 5% Duncan’s Multiple Range Test.

Erickson et al. (2011) stated that sugar content of sweet sorghum was influenced by panicle removal before pollination and grain filling. That influence was due to assimilates allocated for grain set and development were translocated and accumulated in the form sugar in stalk sorghum juices. Broadhead (1973) also reported higher *brix* value when panicle of sorghum was removed before grain setting. Increased sugar yields were also observed in sorghums when their panicle were removed compared to non-panicle removal sweet sorghums (Rajendran et al., 2000). To maintain high sugar content in the stems, seed heads had to be removed when the seed is in the late milk stage (Bitzer, 2009). In the present study, panicle removal did not significantly affect the stalk sugar content of Numbu and Sugar Graze varieties. This was probably due to higher amount of assimilates had been accumulated in the stalk and smaller amount of them were translocated to the inflorescence.

3.5 Stalk ethanol content

The interaction between stages of panicle removal and variety significantly affected ($p<0.05$) stalk ethanol content. Ethanol content of Kawali started to increase when its panicle was removed at 100% flowering while that of Sugar Graze already increased when the removal of panicle occured at 50% booth (Table 5). The higher in stalk ethanol content of both varieties were associated with higher the sugar content due to panicle removal (Erickson et al. 2011). Ethanol content of Numbu variety did not significantly increase by panicle removal. This was associated with the insignificantly effect of the treatment on sugar content of that variety (Table 3). Stalk ethanol content of White Local Belu was significantly ($p<0.05$) lower than those of the improved varieties.

Table 5. Effect of interaction between stages of panicle removal and variety on stalk ethanol content (%)

Variety	Stages			
	Non –panicle removal	50% booth	100% flowering	50% grain filling
White Local Belu	8.80 ^{i}	9.31 h	9.63 ^{g}	9.63 ^{g}
Numbu	12.31 ^{a}	12.08 ^{b}	11.13 ^{c}	12.42 ^{a}
Kawali	10.64^{def}	10.75 de	11.09 c	11.01 ^{c}
Sugar Graze	10.47 ^{f}	10.61 ^{ef}	10.81 ^{d}	10.72 ^{de}

Notes: Means followed by the same letters at the same rows and columns are not significantly different at 5% Duncan's Multiple Range Test.

3.6 Stalk ethanol production

Ethanol production of sweet sorghum stalk was significantly ($p<0.05$) affected by interaction between stages of panicle removal and variety. The highest ethanol production was recorded on Sugar Graze variety with panicle removal at 100% flowering (2084.51 L ha^{-1}) or at 50% grain filling (2080,43 L ha^{-1}). The value was 10.30%, 20.75% and 127.43% respectively higher than those of Kawali, Numbu and White Local Belu with panicle removal at stage of 100% flowering (Table 6). Panicle removal increased stalk ethanol production of the four sweet sorghum varieties, although on Numbu ethanol production only increased (11.06%) when the panicle were removed at 50% booth. Meanwhile that of White Local Belu increased 34.58% when panicle removal occured at 100% flowering. Stalk ethanol production of Kawali and of Sugar Graze increased respectively 16.43% and 62.83 % when panicle removals occured at 50% booth and even increased by 48.73% and 84.28% when the panicles were removed at 100% flowering (Table 6). Stalk ethanol production of the latest two varieties increased more when panicle removals occured at 100% flowering. The increased in ethanol production was associated with increased juice production (Table 3) and ethanol content (Table 5) resulted from panicle removal at the stage of 100% flowering. This findings could help farmers to decide when panicle removal should be done in order to get higher stalk ethanol production.

Table 6. Effect of interaction between stages of panicle removal and variety on stalk ethanol production (L ha^{-1})

Variety	Stages			
	Non –panicle removal	50% booth	100% flowering	50% grain filling
White Local Belu	681.05 ^{ij}	610.06 ^{j}	916.55 ^{h}	773.64 ^{i}
Numbu	1666.58 ^{d}	1850.84 ^{b}	1726.31 ^{cd}	1638.94 ^{d}
Kawali	1270.40 ^{f}	1479.19 ^{e}	1889.46 ^{b}	1843.19 ^{bc}
Sugar Graze	1131.19 ^{g}	1841.89 ^{bc}	2084.51 ^{a}	2080.43 ^{a}

Notes: Means followed by the same letters at the same rows and columns are not significantly different at 5% Duncan's Multiple Range Test.

IV. Conclusion

Panicle removal increased stalk sugar content, ethanol content, juice production and ethanol production of sweet sorgum varieties. Sugar Graze variety had the highest stalk ethanol production (2084.51 L ha^{-1}) when the panicle was removed at 100% flowering or at 50% grain filling (2080.43 L ha^{-1}). Ethanol content of White

Local Belu was lower than that of improved varieties. Farmers can decide the right stage to remove the panicles of sweet sorghums to obtain high stalk ethanol production. Research on application of potassium fertilizer should be done on sweet sorgum with panicle removal to increase stalk ethanol production.

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