

Short Communication

Influence of Senna Stems on Growth, Chlorophyll and Nitrate Reductase Activity of Pearl Millet

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Senna (*Cassia angustifolia*) is predominantly cultivated in semi-arid parts of Tamil Nadu, Rajasthan and Gujarat mainly due to the medicinal value of its leaves and pods which contain anthraquinone glycosides called the sennoside A and sennoside B, used in pharmaceutical industry for preparing preferred laxatives (Singh *et al.*, 1998). However, senna stems which are considered waste have also been found by some farmers of Rajasthan villages to be beneficial for crop growth and yield when used as soil application. Although there are numerous reports on the favorable effects of various crop residues on soil properties, plant growth and yield (Aggarwal *et al.*, 1997) but there is no such work on senna stems, their residue or extracts. Our preliminary work has shown the presence of triacontanol - a growth promoting naturally occurring regulator in senna stems (Garg *et al.*, 2003). The effects of triacontanol in enhancing plant growth and physiological processes are well known (Borowski *et al.*, 2000, Raichur *et al.*, 2001). These observations prompted the present study to explore the influence of senna stems and their crude extract in chloroform, on pearl millet growth and certain biochemical parameters.

The present investigation was conducted in the net house with pearl millet [*Pennisetum glaucum* (L.) R. Br. cv. HHB-67] under pot culture conditions. Experimental plants were raised in glazed pots (3 plants per pot) containing 40 kg loamy sand soil (Typic Camborthids having 7.1% clay, 5.6% silt, 63.1% fine sand and 24.1% coarse sand). Senna stems were obtained from the farmer's field during May 2002 and following seven treatments were imposed at the time of sowing in July 2002 with ten replicates under each treatment. T1- control, T2- soil application of senna stems @ 45 g per pot equivalent to 2.5 t ha⁻¹, T3- soil application of senna stems @ 90 g per pot equivalent to 5.0 t ha⁻¹, T4- chloroform extract of 45 g senna stems, T5- chloroform extract of 90 g senna stems, T6- residue left after extraction of 45 g senna stems and T7- residue left after extraction of 90 g senna stems.

For preparation of extracts of senna stems in chloroform, 450 g senna stems (material for 10 pots) were extracted in pure chloroform in a Soxhlet apparatus for 48 hours and the extract was concentrated and made to a volume of 100 ml. The identify of triacontanol (CH₃(CH₂)₂₈CHOH) was confirmed by its infrared spectroscopic

Table 1. Influence of senna stems, extract and residue on leaf area and fresh and dry weight of pearl millet at 20 and 40 days after sowing (DAS)

Treatments	Leaf area (cm ² plant ⁻¹)		Fresh weight (g plant ⁻¹)		Dry weight (g plant ⁻¹)	
	20 DAS	40 DAS	20 DAS	40 DAS	20 DAS	40 DAS
T1 Control	37.4	416.8	1.13	6.27	0.219	1.58
T2 Senna stems (45 g)*	49.1	498.1	2.47	8.41	0.475	2.50
T3 Senna stems (90 g)*	72.6	687.8	4.69	14.19	0.834	4.42
T4 Senna Extract (45 g)	41.7	388.6	1.92	6.85	0.344	1.85
T5 Senna Extract (90 g)	49.7	453.5	2.69	8.24	0.489	2.09
T6 Senna Residue (45 g)	42.4	541.6	2.07	7.50	0.379	2.14
T7 Senna Residue (90 g)	66.7	651.9	3.66	12.56	0.656	3.54
LSD (P =0.05)	10.4	48.8	0.35	1.04	0.080	0.29

*45 g and 90 g senna per pot equivalent to 2.5 and 5.0 tonnes ha⁻¹, respectively.

analysis as well as by co-TLC with an authentic sample This was then divided into ten equal parts of 10 ml (extract equal to that from 45 g senna stems) and each part of the extract was mixed with small amount of the experimental soil taken in the petri dishes collected from 10 pots separately. The soil mixed with the extract was allowed to dry completely so that all the chloroform evaporated. This dry soil containing the triacontanol was then mixed thoroughly with the respective ten pots (T2). The residue left after extraction was also dried, weighed and divided into ten equal parts which was added to 10 other pots separately (for T6). Similar procedure was followed for preparing the extract of 90 g senna stems where 900 g material was extracted in chloroform for ten pots (T3) as described above. Here also residue left after extraction was divided into 10 equal parts to add to respective pots for T7 treatment.

The addition of senna stems, chloroform extract and residue left after extraction was made one week before sowing and all the

pots including control were irrigated twice to thoroughly mix the senna stems, residue or extract. Sowing was done on 15th July 2002 and after germination 5 seedlings of uniform size were retained which were finally thinned to three seedlings after one week. Plants were watered frequently to keep the soil moisture close to field capacity.

Observations were recorded, in triplicate, on leaf area (using LICOR 3000 leaf area meter) and fresh and dry weight of plants at 20 and 40 days after sowing (DAS). At the same time fresh leaf tissue of two uppermost fully expanded leaves from 9 plants (3 pots) under each treatment was used for the estimation of total chlorophyll (Arnon, 1949) and nitrate reductase activity (Jaworski, 1971) at both the growth stages. Nitrate reductase activity was estimated *in vivo* from leaf discs using KNO₃ as substrate. Significance of the results was accessed through analysis of variance adopting completely randomized design.

Application of senna stems, their extract in chloroform and residue significantly

increased leaf area and fresh and dry weight of pearl millet seedlings within 20 days of growth (Table 1). Senna stems and residue were more effective in promoting growth than chloroform extract. Higher concentration of senna stems (90 g per pot) was more promotive than lower concentration. At 20 days after sowing (DAS) 90 g senna stems enhanced leaf area by more than 90% over untreated control plants while increase in plant fresh weight was of the order of 226% over control. Similar effects were observed at 40 DAS where senna stems followed by residue were found highly promotive for leaf area development and fresh and dry weight of plants.

These results indicate the possibility of the presence of a growth promotive substance in senna stems as such dramatic increase in growth within 20 days can not be ascribed to crop residue effects only. The higher promotion of growth by senna stems and their residue over chloroform extract suggest that besides triacontanol, there may be another growth promoting factor present in the senna stems. However,

significant promotion of plant growth by chloroform extract of senna stems does indicate the role of triacontanol as growth enhancing effects of this growth regulator are well known in higher plants (Sud and Thakur, 1998; Raichur *et al.*, 2001).

The increase in leaf area and plant growth by senna application was associated with a significant increase in total chlorophyll concentration and nitrate reductase (NR) activity at both the growth stages (Table 2). Interestingly lower concentration of senna stems, residue or extract was more promotive than higher concentration for increase in chlorophyll concentration. Among all the treatments senna stems were most effective followed by residue and extract.

The increase in NR activity by application of senna stems, residue and extract was also significant at both the growth stages. At 20 DAS, NR activity increased by 119.7% with 90 g senna stems and by 85.8% with 90 g senna residue as compared to control plants. Similarly at 40 DAS, the increase in NR activity

Table 2. Influence of senna stems, extract and residue on chlorophyll concentration and nitrate reductase activity of pearl millet at 20 and 40 days after sowing (DAS)

Treatments	Total Chlorophyll (mg g ⁻¹ dw)		Nitrate reductase activity ($\mu\text{mol NO}_2 \text{ g}^{-1} \text{ dw h}^{-1}$)	
	20 DAS	40 DAS	20 DAS	40 DAS
T1 Control	7.06	9.21	9.38	7.97
T2 Senna stems (45 g)*	10.35	11.76	12.17	11.71
T3 Senna stems (90 g)*	9.45	10.82	20.62	13.78
T4 Senna Extract (45 g)	9.73	10.91	13.00	11.57
T5 Senna Extract (90 g)	9.64	10.31	13.65	14.00
T6 Senna Residue (45 g)	10.27	10.42	13.61	11.47
T7 Senna Residue (90 g)	10.01	11.26	17.43	14.32
LSD (P =0.05)	0.87	0.53	1.99	1.26

*45 g and 90 g senna per pot equivalent to 2.5 and 5.0 tonnes ha⁻¹, respectively

was of the order of 72.9% and 79.7% by the above treatments, respectively. The chloroform extract of senna stems also increased the NR activity considerably and significantly at both the stages. The nature of active ingredient in senna stems is unknown at present but one of the component may be triacontanol as crude chloroform extract contained this compound as confirmed by IR spectroscopic analysis and co-TLC with an authentic sample. The role of triacontanol in enhancing photosynthetic efficiency and chlorophyll concentration is well documented (Souza *et al.*, 1999; Borowski *et al.*, 2000). Kumaravelu *et al.* (2000) also reported a significant increase in nitrate reductase activity in green gram seedlings by application of triacontanol. However, there may be other growth regulating substances in senna stems which increased chlorophyll and NRA in pearl millet plants. Clearly there is a need for further research to identify and isolate the growth promotive factors in the senna stems. However, the present results show the effectiveness of senna stems in enhancing plant growth of pearl millet and this seem to be mediated through enhanced photosynthetic efficiency and improved nitrogen metabolism.

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