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EFFECT OF DISTILLERY SPENT WASH (DSW) AND FERTILIZER ON GROWTH AND CHLOROPHYLL CONTENT OF SUGARCANE (*SACCHARUM OFFICINARUM* L.) PLANT

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Abstract
A field experiment was conducted with distillery spent wash (DSW) collected from Aska Co-operative Sugar Industries Ltd., Aska, Odisha, India and other two types of treatments viz; inorganic fertilizer and cow dung to observe a comparative effect on growth and chlorophyll content using sugarcane variety Co1274 as the test crop. The experiment was formulated with four treatments (50% DSW, 100% DSW, fertilizer & fertilizer + cow dung) each with three replicates and one set of control. The growth parameters such as height of the plant, length and girth of stem, breadth of leaves, number of leaves and number of tillers per plant, leaf area index and total chlorophyll content of sugarcane plant were estimated during different growth periods in all treatments. All the parameters showed an increasing trend from the control except 100% distillery spent wash, where a declining trend over the control in all parameters was observed. Maximum growth and chlorophyll content was observed in 50% distillery spent wash as compared with two different types of fertilizers.

Keywords: Distillery spent wash (DSW), Sugarcane growth, Height, Stem girth, Chlorophyll content

Introduction
One of the most important environmental problems faced by the world is management of wastes. Different industries create a variety of waste water pollutants; which are difficult and costly to treat. Waste water characteristics and levels of pollutants vary significantly from industry to industry. The use of industrial waste as soil amendment has generated interest in recent time. The waste water produced continuously could cater the needs of irrigated crops [1].

Production of ethyl alcohol in distilleries based on sugarcane molasses constitutes a major industry in Asia and South America. The world's total annual production of alcohol from sugarcane molasses is more than 13 million m^3. The aqueous distillery effluent stream known as spent wash is a dark brown highly organic effluent and is approximately 12-15 times by volume of product alcohol [2]. The disposal of distillery spent wash is of serious concern due to its large volume and high biological oxygen demand (BOD) and chemical oxygen demand (COD). Due to high concentration of organic load, distillery spent wash is a potential source of renewable energy. The effluent does not contain any toxic heavy metals as it is a waste from plant materials. It contains high amount of nutrients such as nitrogen, phosphorous, potassium, sulphur and a large amount of micronutrients. The land application of distillery spent wash often benefits water pollution control and utilization for agricultural production [3]. So it can be applied directly to the land as irrigation water as it helps in restoring and maintaining soil fertility, increasing soil microflora, improving physical and chemical properties of soil leading to better water retaining capacity of the soil. The effluent is ideal for sugarcane, maize, wheat and rape seed production [4]. It has been reported that waste water from different industries produced continuously could cater the needs of irrigated crops [5]. Thus the distillery spent wash will not only prevent waste from being an environmental hazard but also served as an additional potential source of fertilizer for agricultural use. Diluted spent wash increased the growth of shoot length, leaf number per plant, leaf area and chlorophyll content of peas [6]. It was also reported that the water holding capacity, cation exchange capacity, increases the availability of nitrogen, phosphorus, potassium, copper, zinc, iron, manganese; but with reduced biochemical oxygen demand (BOD) with addition of sewage sludge to a course textured sandy and calcareous soil [7]. An increase in the soil organic matter by 1% with sugar factory effluent applied to soils was observed in Cuba [8]. Many workers reported an adverse effect of higher concentration of different types of industrial effluents in the growth rate of different crops [9, 10, 11, 12].

There have been studies related to the application of distillery spent wash to agriculture in India as well as other parts of the world. Spent wash at the rate of 35-50 m^3 ha^{-1} was recommended as optimum dose for higher sugarcane yield in Brazil and in Australia. In Sau Paulo, Brazil, the crop productivity was 2-10 times higher as compared to the untreated lands. Distillery spent wash was found to increase the cane yield in sugarcane and decrease the potassium fertilizer need in a study conducted by Caroni Research Station, Trinidad and Tobago. In Philippines, spent wash application at the rate of 80-240 m^3 ha^{-1} in addition to chemical fertilizers increased the cane yield by 10-12 percent and sugar yield by 13-16 percent compared to normal irrigation. In Cuba, spent wash application at the rate of 90-150 m^3 ha^{-1} increased the potassium content of the soil, with increased cane yield and sugar recovery [13]. In a study conducted in Kiev, Ukraine has shown increased yield of grasses, maize and fodder beet by 45-100% using distillery effluent. In India, extensive studies on distillery spent wash have been carried out successfully with respect to various crops in different agro-climatic regions [13]. Sugarcane (*Saccharum officinarum* L.) has a worldwide significance as a major source of food (sugar) and by-products which are economically more important.

The present investigation aims to study the effect of different concentration of distillery spent wash, inorganic fertilizer and farmyard manure which are usually used by the local farmers, on the growth and chlorophyll content of sugarcane plant at Aska, Ganjam district, Odisha, India.

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Materials & Method

The field experiment was conducted with two different doses of distillery spent wash (50% & 100%), fertilizer used by the local farmers and fertilizer combined with organic manure using Co 1274 sugarcane variety (*Saccharum officinarum* L.) as test crop. The experimental field was divided into five sets of three equal parts of size 5 m×3 m each. Before plantation the land was ploughed, labelled and divided into ridges and furrows with uniform distance. The sugarcane test crop was collected from the local farmer for planting. The setts were treated with Bovistin and planted in the experimental field. After plantation with regular watering once in a week the plants were allowed to grow for three months. After a period of three months different growth parameters and chlorophyll contents were recorded and the fields were treated with different conditions (50 %, 100 % effluent, fertilizer and fertilizer + cow dung). Out of the five sets of fields with three replicates, one set was kept as control without any treatment. Other four sets of fields were treated with different conditions on every month and all the fields were irrigated with water on every week. Biometric observations were made randomly by selecting five plants in the net plot area of individual treatments on every month.

Distillery spent wash application

The distillery spent wash samples were collected from the Aska Co-operative Sugar Industries, Ltd., Ganjam district, Odisha. The physicochemical properties of spent wash were analyzed by standard methods of APHA [14] (Table 1).

Table 1- Chemical composition of DSW collected from Aska Co-operative Sugar Industries Ltd., Aska, Odisha.

Chemical parameters	Amount (mg l^{-1})
pH	7.23
Electrical conductivity (µs)	28700
Total solids	35340
Total Dissolved Solids	27240
Total Suspended Solids	9980
Settleable Solids	9860
Chemical Oxygen Demand(COD)	30520
Biological Oxygen Demand(BOD)	15300
Carbonate	Negligible
Bicarbonate	12200
Total Phosphorus	28.36
Total Potassium	6500
Calcium	920
Magnesium	753.25
Sulphate	5100
Sodium	420
Chlorides	5626
Iron	6.3
Manganese	1429
Zinc	1.09
Copper	0.265
Cadmium	0.036
Lead	0.19
Chromium	0.067
Nickel	0.145
Ammonical Nitrogen	636.25
Total Phosphorus	29.28
Total Potassium	7300
Sulphur	75.6

Raw effluent was diluted to 50% v/v with water. The 50% and concentrated (100%) effluents were taken for experimental studies.

Fertilizer application

In this experiment the fertilizer doses used by the local farmers were adopted for sugar cane crop. The inorganic fertilizer was applied to the demarcated plots at four different times i.e. NPK at the rate of 250 kg ha^{-1} before planting the setts; urea at the rate of 125 kg ha^{-1} in two doses at 30 days and 90 days after plantation; the last dose of inorganic fertilizer, urea at the rate of 60 kg ha^{-1}+ Potash @ 62 kg ha^{-1} after 180 days of plantation.

Organic manure (cow dung)

In addition to the fertilizer dose the organic manure- cow dung was applied to the experimental plots. The chemical composition of cow dung is presented in Table 2. Total organic carbon was measured by using the method by Nelson & Sommers [15]. Total nitrogen was determined by Bremmer & Mulvaney[16]. Total available phosphorus was determined by using the method given by Bansal & Kapoor [17]. Total potassium and calcium was determined by flame photometer [17]. The pH and electrical conductivity (EC) were determined by the method of Garg et al. [18]. Twenty kg of cow dung dissolved and mixed in 20 litre of water and applied to the fields (plots) over the fertilizers used.

Table 2. Chemical composition of cow-dung used in the experiment at Aska,Ganjam, Odisha.

Chemical Parameters	Amount
Total Calcium (g kg^{-1})	1.29 ± 0.3
Mg (g kg^{-1})	0.194 ± 0.26
Total Potassium (g kg^{-1})	4.69 ± 0.068
Total Phosphorus (g kg^{-1})	3.9 ± 0.04
Total Nitrogen(g kg^{-1})	5.9 ± 0.19
C:N ratio	81.2 ± 2.56
Total organic carbon (g kg^{-1})	480.65 ± 4.2
pH	7.6 ± 0.03
Electrical conductivity (ds m^{-1})	1.96 ± 0.02

Each value is the mean of five replicates ±SE

Soil analysis

The soil samples from the experimental site were collected, air dried, powdered and analyzed for physico-chemical properties. The physical properties like soil pH was estimated by pH meter and chemical characteristics was determined by following the methods of Chapman [19] and Wilde et al. [20] and presented in Table 3.

Table 3. Properties of the soil of the experimental site, Aska, Ganjam (Odisha).

Nutrients	Amount (ppm)
pH (1:2) solution	6.9
Organic carbon (%)	8.97
Available nitrogen	345
Available phosphorus	131
Available potassium	79
Calcium	139
Magnesium	226
Sodium	84
Available sulphur	231
Iron	192
Manganese	199
Copper	04

The growth parameters such as height of the plant, length and breadth of leaf, girth of the stem, number of leaves and number of tillers per plant and leaf area were recorded as per the standard procedure. From leaf area leaf area index was calculated [21]. The chlorophyll content of the sugarcane plant was determined by using method of Arnon [22] and expressed as mg g^{-1} fresh wt.

Data analysis

Data relating to different growth parameters and total chlorophyll content was analyzed by one way ANOVA. Results of different conditions were computed over control and their difference among treatments was tested through least significant difference (LSD). All the parameters are statistically significant.

Results and discussion

Distillery spent wash is the unwanted liquid waste produced during the production of alcohol and it is one of the most important environmental issues. The distillery spent wash with its characteristic unpleasant odor poses a serious threat to the water quality around the world [23]. The ever increasing amount of distillery spent wash and its disposal has stimulated the need for developing new technologies to process this effluent efficiently and economically including growth and yield of different crops in agriculture [24]. It was found that the growth and chlorophyll content of the sugarcane in different conditions of treatment showed an increasing trend over the control. Average height of the sugarcane plant after 210 days showed an increase of 13.45% in the 50% DSW treated plot over the control (Fig. 1). However, the growth showed negative trend in 100% DSW.) in all parameters. The average length of leaves of the test crop after 210 days of plantation showed an increase of 11.22% in 50% DSW treated plants over control and a negative trend in 100% DSW (Fig. 2). The average breadth of the leaves after 210 days of plantation showed an increase of 46.96% in the 50% DSW plants over control (Fig. 3) and a negative trend in 100% DSW.

Similarly the average girth of the stem, average number of leaves per plant, average number of tillers per plant, average leaf area index and chlorophyll content of the test crop after 210 days of plantation showed an increase of 16.79%, 21.18%, 57.5%, 62.84% and 28.99% respectively in 50% DSW treated plants over control (Fig. 4, 5, 6, 7, 8) and a negative trend in 100% DSW. The ANOVA showed that there was significant difference in all the stages of growth of the plant except in 90 days (Table 4, 5,6,7,8,9,10, & 11). All fertilizer and fertilizer+cow dung treated plants showed an increasing trend over control.

Fig. 1. Height of the sugarcane plant at different growth periods with different treatments.

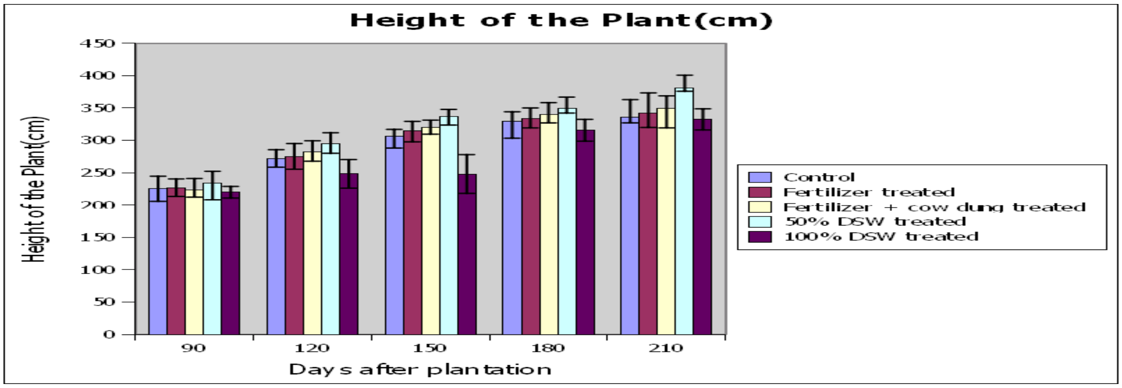


Table 4. Statistical analysis of the height of the sugarcane plant at different days of growth and different treatments

Days after plantation	F value for different treatments	Significance level	LSD
90	0.54	NS	23.96
120	4.62	0.05p	23.43
150	3.32	0.05p	22.92
180	3.45	0.05p	19.88
210	3.99	0.05p	28.89

NS – Not Significant

Fig. 2 . Length of leaves of sugarcane plant at different growth periods with different treatments.

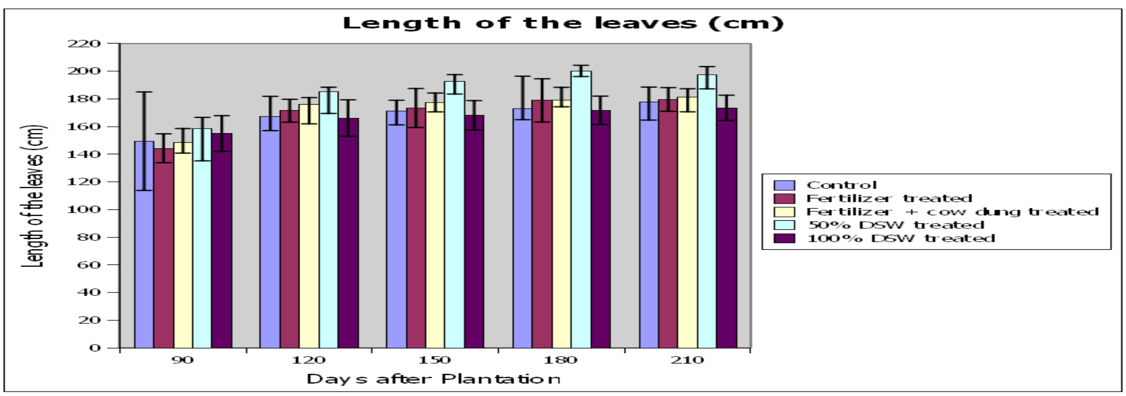


Table 5. Statistical analysis of the length of leaves of sugarcane plant at different days of growth and different treatments.

Days After Plantation	F value for different treatments	Significance level	*LSD*
90	0.46	NS	24.46
120	3.04	0.05p	12.98
150	5.27	0.05p	12.40
180	3.17	0.05p	18.64
210	6.1	0.05p	11.01

NS – Not Significant

Fig. 3. Breadth of leaves of sugarcane plant at different growth periods with different treatments.

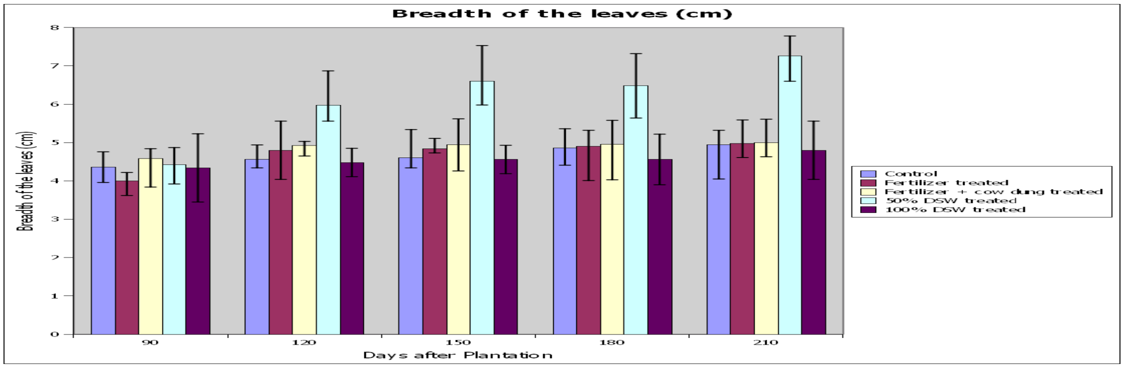


Table 6. Statistical analysis of breadth of leaves of the sugarcane plant at different days of growth and different treatments

Days after plantation	F value for different treatments	Significance level	LSD
90	0.88	NS	0.67
120	4.4	0.05p	0.85
150	8.02	0.05p	0.89
180	7.8	0.05p	0.83
210	16.5	0.05p	0.76

NS – Not Significant

Fig. 4. Girth of the stem of sugarcane plant at different growth period with different treatment

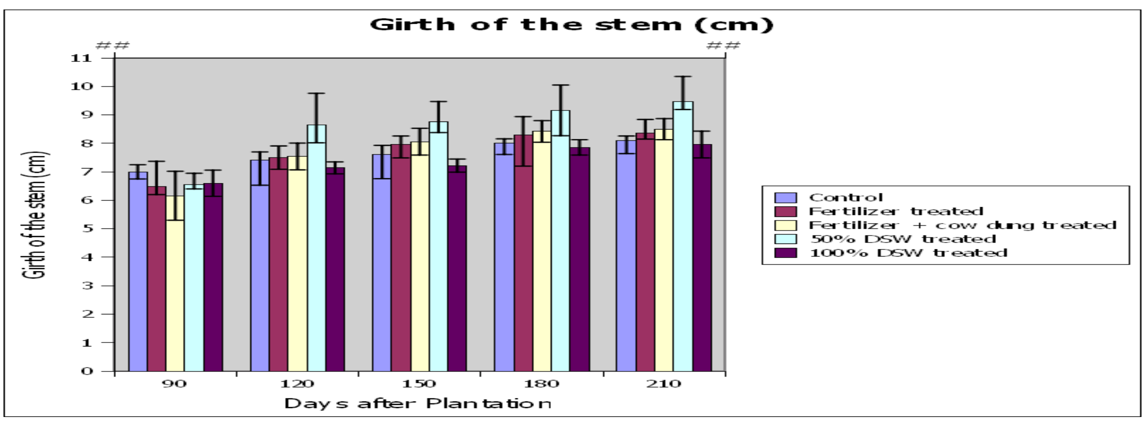


Table 7. Statistical analysis of the girth of the stem of the sugarcane plant at different days of growth and different treatment

Days after plantation	F value for different treatments	Significance level	*LSD*
90	1.16	NS	0.83
120	4.97	0.05p	0.78
150	7.68	0.05p	0.47
180	4.42	0.05p	0.50
210	6.14	0.05p	0.70

NS – Not Significant

Figure .5. Number of leaves per sugarcane plant at different growth periods with different treatments

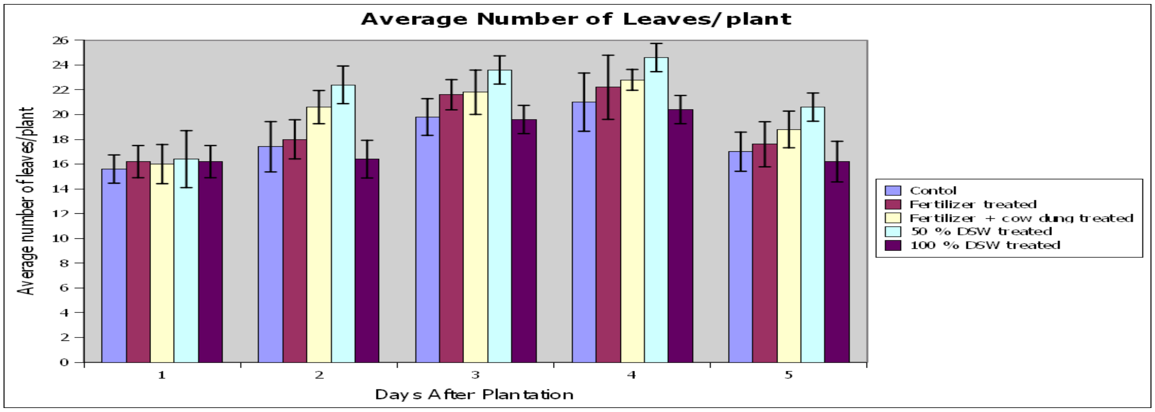


Table 8. Statistical analysis of number of leaves per sugarcane plant at different days of growth and different treatment

Days after plantation	F value for different treatments	Significance level	*LSD*
90	0.184	NS	2.09
120	11.57	0.05p	2.15
150	7.02	0.05p	1.82
180	4.36	0.05p	2.33
210	6.14	0.05p	2.04

NS - Not significant

Fig. 6. Numbers of tillers per sugarcane plant at different growth periods with different treatments

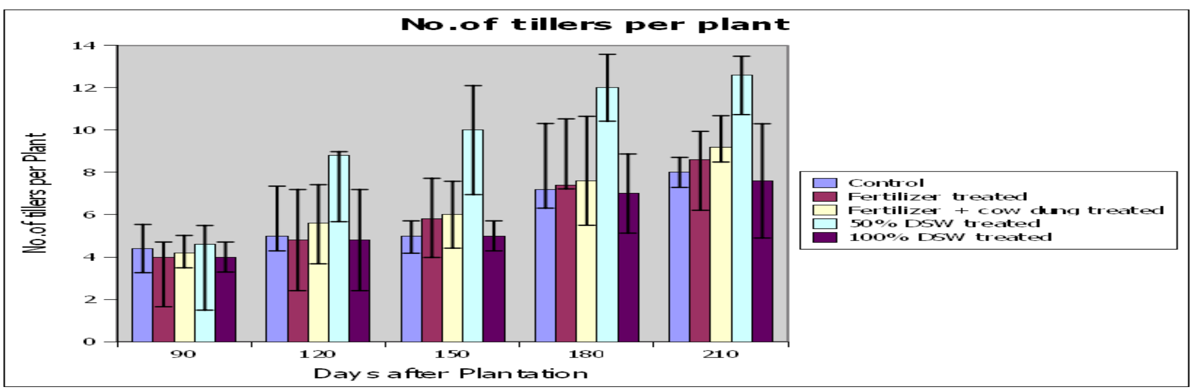
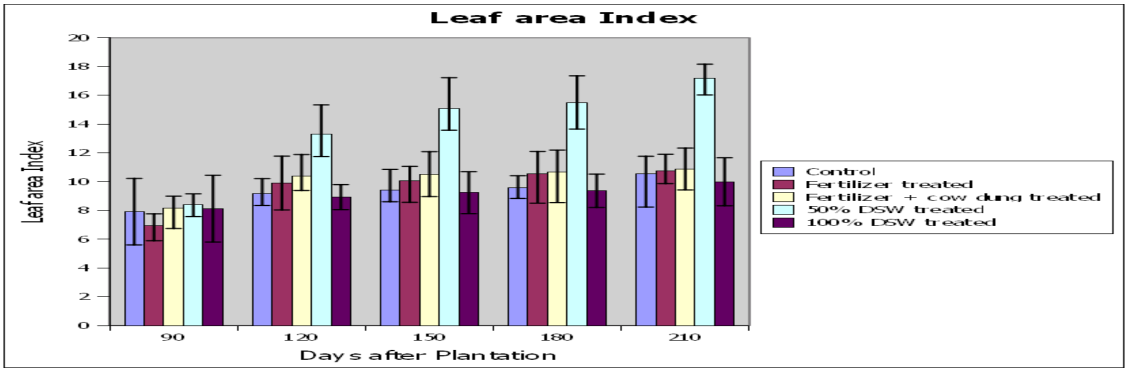


Table 9 .Statistical analysis of number of tillers per sugarcane plant at different days of growth and different treatment.

Days after plantation	F value for different treatments	Significance level	*LSD*
90	0.45	NS	1.15
120	3.49	0.05p	2.70
150	7.02	0.05p	2.13
180	3.29	0.05p	3.49
210	7.9	0.05p	2.10

NS – Not Significant

Figure. 7. Leaf area index of sugarcane crop at different growth periods with different treatments



analysis Table 10. Statistical of leaf area index of sugarcane plant at different days of growth and different treatment

Days after plantation	F value for different treatments	Significance level	LSD
90	0.63	NS	2.11
120	6.51	0.05p	2.03
150	12.07	0.05p	2.06
180	15.55	0.05p	1.88
210	25.83	0.05p	1.74

NS – Not Significant

Fig. 8. Chlorophyll content of sugarcane leaf at different growth periods with different treatments

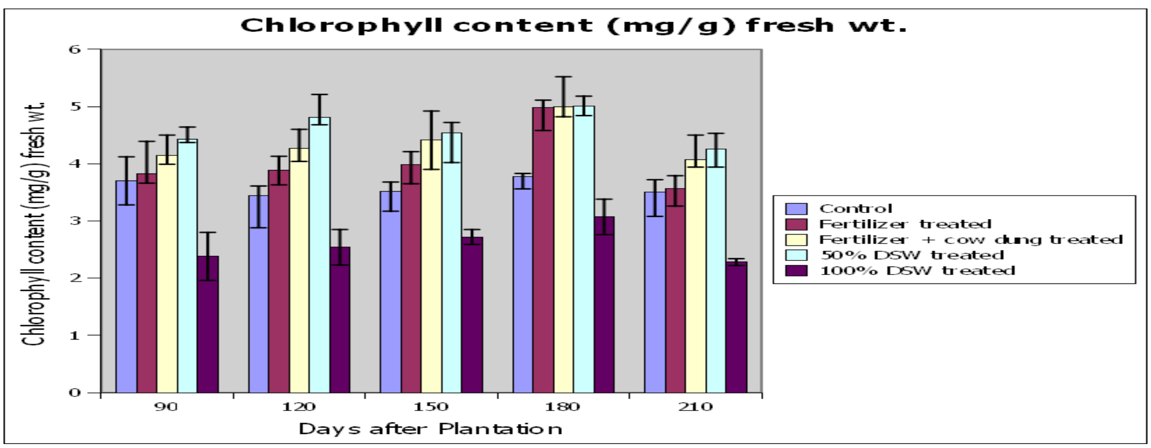


Table11.Statistical analysis of chlorophyll content of leaves of of sugarcane plant at different days of growth and different treatment

Days after plantation	F value for different treatments	Significance level	*LSD*
90	0.39	NS	0.57
120	4.02	0.05p	1.27
150	5.29	0.05p	1.17
180	3.28	0.05p	1.21
210	3.18	0.05p	1.26

NS – Not Significant

The spent wash treated soil is enriched with the plant nutrients such as nitrogen, phosphorus and potassium. Subsequent use of spent wash for irrigation enriches the soil fertility without any adverse effect and hence spent wash (50%) can be conveniently used for the irrigation of sugarcane crop without any external (either organic or inorganic) fertilizer application. Spent wash increases the soil enzymatic activity [25] which enriches the plant growth and development. The declining tendency of growth parameters in higher concentration may be due to the presence of higher amount of organic matter and biological oxygen demand (BOD) which might have lead to depletion of O_2 and accumulation of CO_2 in the soil.

The increase in chlorophyll content in 50% distillery spent wash treatment crop suggests that synthesis of chlorophyll is accelerated in low concentration. Similar findings have been reported by many workers [26, 27, 28]. The increase in chlorophyll content may be due to lack of heavy metals and in the effluent and probably the availability of Fe and Mg which are necessary for the synthesis of chlorophyll. Reduction of chlorophyll pigment in higher concentration (100%) may be associated with mineral ions. Some of the possible reasons for the decrease of pigment contents may be due to the formation of enzyme chlorophyllase which is responsible for chlorophyll degradation [29, 30].

Conclusion

From the experimental observations it may be concluded that distillery spent wash at 50% v/v concentration may serve as a good liquid fertilizer for sugarcane crop which is more effective than that of the fertilizer used by the local farmers as well as the inorganic fertilizer plus the organic manure (cow dung).

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