Original Research Article

ASSESSING THE IMPACT OF GAS FLARING ON TEA PLANTS IN THE SOUTH OF KOTHALONI OCS IN THE DISTRICT OF DIBRUGARH, ASSAM, INDIA

ABSTRACT

Effect of gas flaring on physiology and growth of tea plants was done by using Randomized Complete Block Design (RCBD) and analysis was done accommodating two seasons and five levels of distances during September 2019-March 2020. Plant samples were collected from (40-50) metres, (50-60) metres, (60-70) metres, (70-80) metres and control site (150-160) metres away from the tea garden situated south of kothaloni OCS in rainy and autumn seasons. The experimental plot was laid out at a distance of 40 metres away from the flare pit. Plant parameters namely relative turgidity, water saturation deficit, tea leaf area ,specific leaf weight, stomatal count, caffeine content, chlorophyll content, polyphenol content and plucking point density were studied. From the study, plant parameters viz. relative turgidity, stomatal count, specific leaf weight, tea leaf area, chlorophyll-a content and chlorophyll-b content, caffeine content, polyphenol content, plucking point density were found to be decreased at distances nearer to the gas flaring site which significantly increased with distances away from the gas flaring site. Water saturation deficit of tea leaves recorded highest at distances closest to the flaring site and decreased with distances away from the gas flaring site.

***Keywords**: Gas flaring, seasons, distance,OCS(Oil Collecting Station)*

1.INTRODUCTION

Tea [*Camellia sinensis* (L) O. Kuntze] is one of the ancient and most popular beverages in the world. India is one of the leading countries in the world with respect to global tea consumption, production and exports. The state of Assam in India lying on either side of the Brahmaputra river is one of the most prolific tea producing regions because of its long generous rainfall and growing season.

Assam is enriched with natural resources like natural gas, oil and coal. The petroleum industry of Assam plays an important role in the process of industrialization in the state and the crude oil is one of the valuable natural resources of Assam.. The excess or unwanted gases which are produced during different operations such as crude oil extraction get burnt through a vertical device and the process of burning of the unwanted gases through the vertical device can be called as gas flaring. Many oil drilling sites of upper Assam are located near the tea estates.

Gas flare consists of a mixture of different gases and its composition greatly depends on the gas source that is entering to the system of flare. During oil-gas production, associated gases that are released mainly contain natural gas and natural gas is consisted of more than 90% methane (CH4) with ethane, a small amount of other hydrocarbons and inert gases such as N_2 and CO_2 [1]. Gas flaring from refineries and other different operations in general contain a mixture of hydrocarbons and sometimes hydrogen. Carbon dioxide (CO_2) and methane (CH_4) are the Green House Gases which increase the temperature of the atmosphere when directly released into the air. Some substances released during the production of oil are of poisonous nature and are detrimental to the environment. Gas flaring does not only destruct vegetation and wild life but it also significantly impacted the microclimate in vicinity of the flare sites [2], which may reduce the growth of plants and it's resistance to abiotic and biotic stresses, making them more susceptible to pathogens.

Crude oil and associated gas flaring are the major reasons for the occurrence of acid rain [3] and the acid rain causes vegetation loss [4]. The impact of acid rain on vegetation is more severe near the gas flare stack and when leaves get washed with acid precipitation, the protective waxy coating of leaves get away which affects their photosynthetic activity. Transpiration and photosynthesis get reduced as deposited oil penetrates into leaves and leaves turn yellow when light is the pollution.

As research work related effect of to gas flaring on tea plant is very limited,so an assessment was taken to study its effect on growth of tea plants adjacent to Kothaloni OCS South in the district of Dibrugarh of Assam. Keeping in view all the above points. The present investigation has the following objective: To study the effect of gas flaring on physiology and growth of tea plants.

2.MATERIAL AND METHODS

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A tea garden south of Kothaloni OCS, Dibrugarh district was selected for this study. The study was done during September 2019-March 2020. Experimental Design selected was Randomized Complete Block Design accomodating five distances namely (40-50) metres, (50-60) metres, (60-70) metres, (70-80) metres and control site (150-160) metres away from the gas flaring site and two seasons namely rainy and autumn seasons.

A pond was there in between the flare site and the selected tea garden and so the experimental plot was laid out at 40 metres away from the flare pit. The flaring site was surrounded by a concrete wall. Plots were selected at 10 metres interval within the experimental design. DC denotes control plot.

2.1.Relative turgidity and Water saturation deficit

The relative turgidity technique was determined by Weatherley [5]. Relative turgidity (RT) was expressed in percentage (%) and water saturation deficit (WSD) was also expressed in percentage (%) and determined using the following formula:
R T (%) = {Average fresh weight (mg)−Average dry weight (mg)} /{Average saturation weight (mg)−Average dry weight (mg)} X 100
WSD (%) = {Average saturation weight (mg)− Average fresh weight (mg)}/ {Average saturation weight (mg)− Average dry weight (mg)} x100

2.2.Specific leaf weight

Fifty leaves without the petiole were selected from each planting material and their individual leaf area was measured. Specific leaf weight of tea leaves can be calculated by using the following formula:

$$SLW(g/cm^2) = \frac{\text{Dry weight of leaf (mg)}}{\text{Total leaf area } (cm^2)}$$

2.3.Stomatal count

The estimation of the stomatal count was determined by the method given by Beakbane and Mazumdar [6].

2.4.Leaf area of tea leaves(cm^2)

Individual leaf was taken and tracing was done over graph paper and the covered grids by the leaf are counted to give the area.It is expressed in cm^2.

2.5.Chlorophyll content

The chlorophyll extraction was done using methanol given by Taylor [7]. Buds and two leaves weighing 0.5g were taken and chopped into pieces. A liquid mixture was made by adding 25 ml of methanol slowly to the chopped pieces placed in mortar and pestle. In 25 ml conical flask,2ml of supernatant was transferred and conical flask was covered with carbon paper which was kept separately in a dark place for about 30minutes. The mixture was then centrifuged at 5000 rpm for 5 minutes. In 2 ml of the mixture, 8 ml methanol was added after centrifuge to make a volume of 10 ml. Then spectrophotometric observation of diluted solutions was recorded in different wavelengths namely 653 nm for chlorophyll-a and 666 nm for chlorophyll-b. It wasexpressed as mg/g.

The formulas used were as follows-

$$\text{Chl-a}=15.65A_{666}-7.34A_{653}$$

$$\text{Chl-b}=27.05A_{653}-11.21A_{666}$$

2.6.Caffeine content

Caffeine estimation was done by boiling 200 ml of water containing 20 g of two leaves and a bud for 30 minutes in a 400 ml beaker and should be filtered while hot. The filtrate was collected in an another clean beaker and of Lead acetate of 10% aqueous solution was added in the filtrate that was taken in a clean beaker followed by centrifuged at 5000 rpm for 5 minutes. The volume of the supernatant was reduced to 25 ml by boiling and cooled it at room temperature. The mixture was extracted with 25 ml chloroform in a separating funnel after cooling the reaction. The chloroform was distilled off from the chloroform extract on a water bath using a water condenser. By using a spatula, the dried residue was scrapped from the flask and weighed was taken. Weight of fresh tea leaves and weight of crude caffeine in gram gives the amount of caffeine. The total caffeine in tea leaves was expressed in percentage (%).

2.7. Total polyphenol content

Polyphenol content was estimated with Folin-Ciocalteu reagent, based on the reaction between phenols and phosphomolybdate that results in the blue complex formation [8].

2.8. Plucking point density

The method used for counting plucking points was same as the method to the method described by Barua and Dutta [9]. A light bamboo grid of 10 cm square sides was placed on the top of the bush and in the randomly selected plant, the number of plucking points at the the end of the season was collected.

3. RESULTS AND DISCUSSIONS

3.1. Relative Turgidity

Relative turgidity of tea leaves had shown significant differences among distances and seasons. Relative turgidity was found to be lower near the gas flaring site which increased with distances away from the gas flare. Among the distances, D_2, D_3, D_4 and D_C were significant to D_1. A lowest mean value was recorded at D_1 (79.68%) and the highest mean value was seen at D_C (84.51%) as mentioned in Table 1. This might be due to high temperature stress that decreased fresh and dry weight of leaves of tea plant which reduced its relative turgidity. This was agreed with the findings of Naz [10], who reported that relative turgidity of potato plants were successively reduced by increased heat stress. In this study, relative turgidity of tea leaves had shown decreased mean value in autumn season as compared to rainy season. This might be due to increased water stress in the season of autumn which led to reduction in relative turgidity of tea leaves. These results corroborates the findings of Waheed [11] who reported that relative turgidity of tea leaves were successively decreased during drought stress.

3.2. Water Saturation Deficit

Water saturation deficit was significantly varied among distances. Among the distances, D_2, D_3, D_4 and D_C were significant to D_1. There was no significant variations observed In rest of the distances. Highest mean value was found in D_1 (20.51%) and the lowest mean value was recorded in D_C (16.01%) as mentioned in Table 2. Higher water saturation deficit near the gas flaring site as compared to far away distances might be due to high temperature stress that reduced tea plants‘ relative water content and it successively increased water saturation deficit of tea. The relative humidity around the gas flare was relatively reduced and might dessicate leaves of plants that are vulnerable to dessication [12]. In this study, water saturation deficit of tea leaves was increased in autumn season than rainy season. This might be due to water stress intensified by dry season that led to increase in water saturation deficit of tea leaves.

3.3. Number of stomata

Number of stomata of tea leaves varied significantly among distances. Among the distances, variations were observed between all the distances except D_3 and D_4. The lowest mean stomata was recorded at D_1 (19.66 stomata/mm^2) which gradually increased with increase in distance from the flare site and the highest value was found in control site at 150 m-160 m (22.83 stomata/mm^2) as shown in Table 3. Significant variation was observed among seasons and autumn season recorded the lowest mean value (20.93 stomata/mm^2) This might be due to presence of pollutants in the surrounding environment of gas flare that changed biochemical composition along with morphology of plants leading to the nitrogen limitation for the plant. In a similar findings of Sarma [13], who reported that stomata of *Cassia tora* in Sivasagar district of Assam was reduced due to CO_2 stressed environment .

3.4. Leaf area of tea leaves

It was observed that tea leaf area varied significantly among distances. Among the distances, D_4, D_C are significant to D_1. Between rest of the distances no significant variations were observed . D_1 recorded the lowest mean value of leaf area (36.37 cm^2) and the highest mean value was observed in D_C (49.76 cm^2) as mentioned in Table 4. This might be due to decreased stomatal conductance and reduced nitrogen content in plant growing under enriched air pollutants level for which the photosynthetic activities might be suppressed that led to reduction in leaf area of tea. The findings of Sarma [13] also reported reduction in leaf area of *Cassia tora* near the gas flaring site. However, tea leaf area had shown no significant differences in seasons.

3.5. Specific weight of tea leaves

Specific leaf weight varied significantly among distances. Within the distances, significant variations were found between D_1 and D_3, D_1 and D_4, D_1 and D_C, D_2 and D_3, D_2 and D_4, D_2 and D_C. The D_1 and D_2 recorded the lowest mean specific leaf weight (0.007 g/cm^2) whereas, D_4 and D_C recorded the highest mean specific leaf weight (0.011 g/cm^2) as mentioned in Table 5. This might be due to the reason that plants growing under enriched CO_2 stress might reduced leaf area of tea plants resulted in lowering its specific weight near gas flaring site. The findings of Sarma [13] also reported reduction in leaf area of *Cassia tora* near the gas flaring site.

Seasons had shown significant variation where autumn season recorded the lowest mean value of specific weight of tea leaves (0.009 g/cm^2). This might be due to the seasonal variation of air pollutants and decreased photosynthetic activities during post monsoon period [13].

3.6. Chlorophyll content

It was found that chlorophyll acontent (mg/g) and chlorophyll b content (mg/g) were significantly varied along distances.. D_1 recorded the lowest mean chlorophyll a content (0.96 mg/g) and highest value was found in D_C (2.31mg/g) and mentioned in Table 6. Lowest mean chlorophyll b content was observed in D_1 (0.42 mg/g) and highest mean value was found in D_C (0.85 mg/g). Combination of both drought and heat stress might be the reason of low chlorophyll content near the gas flaring site. Chlorophyll content might be reduced due to heat alone[14] and shortage of water might disrupt chloroplast structure which might led to destruction of Chlorophyll. This is in confirmation with the findings of Isichei and Sanford [12] who reported Chlorophyll content of *Eupatorium odoratum* was decreased in the distances nearer to gas flaring site.

Chlorophyll a and chlorophyll b contents had shown significant variation along seasons. Mean chlorophyll a and mean chlorophyll b contents were recorded highest in rainy season having 1.66 mg/g and 0.67 mg/g respectively.

3.7. Caffeine content

Caffeine content in tea leaves had shown significant variations with distances. Distances nearer to gas flaring site had shown lower value as compared to distances away from the gas flaring site. Lowest mean caffeine content was observed in D_1 (2.35%) and the highest mean value was found in D_C (3.48%) as shown in Table 7. This might be due to drought stress that inhibited the expression of genes related to caffeine biosynthesis and thereby reducing the accumulation of Caffeine in *Camellia sinensis*. This corroborates the findings of Song [15], Lee [16] and Zhang [17] who reported caffeine levels decreased with an increase in light.

3.8. Polyphenol content

Significant variation was seen in polyphenol content (%) of tea leaves along distances and the significant variations were observed between all the distances except D_2 and D_3. Lowest mean value for total polyphenol content was recorded in D_1 (21.37 %) and the highest mean value was observed in D_C (23.27 %) as recorded in Table 8. It was also observed that mean total polyphenol content was varied significantly in seasons and was higher in rainy season (22.67 %). Polyphenol content was observed to be lowest in distances closest to the gas flaring site and also observed lowest in autumn season as compared to rainy season. This might be due to the alteration of biochemical constituents necessary for tea quality by drought stress. Erturk [18] also reported significant decrease in total polyphenol content that reduced leaf quality of *Camellia sinensis* as affected by drought stress.

3.9. Plucking point density of tea bush

It had shown significant differences with respect to distance. The highest mean value was observed in DC (41.50 no./2500 cm^2) and the lowest mean plucking point density was observed in D1 (31.33 no./2500 cm^2) as mentioned in Table 9. Plucking point density had also shown significant variation in seasons and was higher in rainy season (36.99 no./2500 cm^2).

Plucking point density of tea bush was lowest in distances nearer to the gas flaring site. Air pollutants was higher in the surrounding environment of gas flare [19] which might reduce chlorophyll content of tea leaves that affected growth of tea bush reducing its plucking point density. This study finds similarity with the findings of Sarma [13].

Table 1. Relative turgidity of tea leaves as affected by gas flaring

Seasons / Distances (m)	Relative turgidity (%)			
	Rainy season (S1)	**Autumn season (S2)**	**Mean**	
D1(40-50)	86.45	72.90	79.68	
D2(50-60)	90.32	76.04	83.18	
D3(60-70)	88.34	78.68	83.51	
D4(70-80)	89.36	78.09	83.72	
DC(150-160)	89.86	79.16	84.51	
Mean	88.86	76.97		
Factors	**C.D.**	**SE(d)**	**SE(m)**	**Significance**
Distance(D)	2.48	1.17	0.83	S
Season(S)	1.57	0.74	0.52	S
Distance X Season(DXS)	N/A	1.659	1.17	NS

**S= Significant at 5% probability level; NS = Non Significant; N/A= Not Applicable*

Table 2. Water saturation deficit of tea leaves as affected by gas flaring

Seasons / Distances (m)	Water saturation deficit (%)		
	Rainy season (S1)	**Autumn season (S2)**	**Mean**
D1(40-50)	13.58	27.45	20.51
D2(50-60)	9.73	23.86	16.79

D_3(60-70)	11.30	21.54		16.42
D_4(70-80)	10.39	21.67		16.03
D_C(150-160)	11.00	21.01		16.01
Mean	11.20	23.10		
Factors	**C.D.**	**SE(d)**	**SE(m)**	**Significance**
Distance(D)	2.46	1.16	0.82	S
Season(S)	1.56	0.77	0.52	S
Distance X Season(DXS)	N/A	1.65	1.16	NS

**S= Significant at 5% probability level; NS = Non Significant; N/A= Not Applicable*

Table 3. Stomatal count of tea leaves as affected by gas flaring

Seasons / Distances (m)	Stomatal count (number/mm^2)			
	Rainy season(S1)	**Autumnn season(S2)**		**Mean**
D_1(40-50)	20.00	19.33		19.66
D_2(50-60)	21.00	20.33		20.66
D_3(60-70)	21.66	21.33		21.50
D_4(70-80)	22.00	21.00		21.50
D_C(150-160)	23.00	22.66		22.83
Mean	21.53	20.93		
Factors	**C.D.**	**SE(d)**	**SE(m)**	**Significance**
Distance(D)	0.49	0.23	0.16	S
Season(S)	0.31	0.15	0.10	S
Distance X Season (DXS)	N/A	0.37	0.23	NS

**S= Significant at 5% probability level; NS = Non Significant; N/A= Not Applicable*

Table 4. Leaf area of tea as affected by gas flaring

Seasons / Distances(m)	Leaf area (cm^2)		**Mean**
	Rainy season (S_1)	**Autumn season (S_2)**	
D_1(40-50)	38.66	34.08	36.37
D_2(50-60)	45.89	36.75	41.32

D_3(60-70)	44.49	43.76	44.12	
D_4(70-80)	49.76	49.18	49.47	
D_C(150-160)	50.15	49.37	49.76	
Mean	45.79	42.62		
Factors	**C.D.**	**SE(d)**	**SE(m)**	**Significance**
Distance(D)	8.78	4.15	2.93	S
Season(S)	N/A	2.62	1.85	NS
Distance X Season (DXS)	N/A	5.87	4.15	NS

**S= Significant at 5% probability level; NS = Non Significant ; N/A= Not Applicable*

Table 5. Specific leaf weight of tea leaves as affected by gas flaring

Seasons / **Distances (m)**	**Specific leaf weight (g/cm^2)**		**Mean**	
	Rainy season (S_1)	**Autumn season (S_2)**		
D_1(40-50)	0.008	0.007	0.007	
D_2(50-60)	0.008	0.007	0.007	
D_3(60-70)	0.011	0.010	0.010	
D_4(70-80)	0.012	0.010	0.011	
D_C(150-160)	0.012	0.010	0.011	
Mean	0.010	0.009		
Factors	**C.D.**	**SE(d)**	**SE(m)**	**Significance**
Distance(D)	0.002	0.001	0.001	S
Season(S)	0.001	0.001	0.000	S
Distance X Season (DXS)	N/A	0.001	0.001	NS

**S=Significant at 5% probability level; NS=Non Significant; N/A=Not Applicable*

Table 6. Chlorophyll content of tea as affected by gas flaring

Seasons / Distances(m)	Chlorophyll a content (mg/g)				Chlorophyll b content (mg/g)			
	Rainy season (S_1)	Autumn season (S_2)		Mean	Rainy season (S_1)	Autumn season (S_2)		Mean
D_1(40-50)	1.00	0.93		0.96	0.43	0.41		0.42
D_2(50-60)	1.19	1.15		1.17	0.53	0.51		0.52
D_3(60-70)	1.58	1.39		1.48	0.67	0.57		0.62
D_4(70-80)	2.18	2.12		2.15	0.86	0.73		0.79
D_C(150-160)	2.35	2.28		2.31	0.88	0.82		0.85
Mean	1.66	1.57			0.67	0.60		
Factors	**C.D.**	**SE(d)**	**SE (m)**	**Significance**	**C.D.**	**SE(d)**	**SE (m)**	**Significance**
Distance	0.13	0.06	0.04	S	0.05	0.02	0.02	S
Season	0.08	0.04	0.03	S	0.03	0.02	0.01	S
Distance X Season (DXS)	N/A	0.09	0.06	NS	N/A	0.03	0.02	NS

**S= Significant at 5% probability level; NS = Non Significant ; N/A= Not Applicable*

Table 7. Caffeine content of tea as affected by gas flaring

Seasons / Distances (m)	Caffeine content (%)		Mean
	Rainy season (S_1)	Autumn season (S_2)	
D_1(40-50)	2.38	2.33	2.35
D_2(50-60)	2.46	2.44	2.45
D_3(60-70)	3.01	2.71	2.86
D_4(70-80)	3.29	3.19	3.24

D_C(150-160)	3.54	3.43		3.48
Mean	2.93	2.82		
Factors	**C.D.**	**SE(d)**	**SE(m)**	**Significance**
Distance(D)	0.10	0.05	0.03	S
Season(S)	0.06	0.03	0.02	S
Distance X Season (DXS)	N/A	0.07	0.05	NS

**S= Significant at 5% probability level; NS = Non Significant ; N/A= Not Applicable*

Table 8. Total polyphenol content of tea leaves as affected by gas flaring

Seasons / Distances (m)	Total polyphenol content (%) Rainy season (S_1)	Autumn season (S_2)		Mean
D_1(40-50)	21.72	21.03		21.37
D_2(50-60)	22.24	21.78		22.01
D_3(60-70)	22.79	22.06		22.42
D_4(70-80)	23.13	22.80		22.96
D_C(150-160)	23.48	23.06		23.27
Mean	22.67	22.14		
Factors	**C.D.**	**SE(d)**	**SE(m)**	**Significance**
Distance(D)	0.49	0.23	0.16	S
Season(S)	0.31	0.15	0.10	S
Distance X Season (DXS)	N/A	0.33	0.23	NS

**S= Significant at 5% probability level; NS = Non Significant; N/A= Not Applicable*

Table 9. Plucking point density of tea as affected by gas flaring along

Distances (m) \ Seasons	Plucking point density (No./2500 cm^2) Rainy season (S_1)	Plucking point density (No./2500 cm^2) Autumn season (S_2)	Mean	
D_1(40-50)	32.00	30.66	31.33	
D_2(50-60)	33.66	32.66	33.16	
D_3(60-70)	37.00	36.33	36.66	
D_4(70-80)	40.33	38.66	39.49	
D_C(150-160)	42.00	41.00	41.50	
Mean	36.99	35.86		
Factors	**C.D.**	**SE(d)**	**SE(m)**	**Significance**
Distance(D)	1.55	0.73	0.52	S
Season(S)	0.98	0.46	0.33	S
Distance X Season (DXS)	N/A	1.04	0.73	NS

**S= Significant at 5% probability level; NS = Non Significant; N/A= Not Applicable*

4. CONCLUSION

Gas flaring has effects in physiology of tea plants with respect to growth in the area under study where the studied plant parameters had shown an increasing trend with distances away from the gas flaring site except water saturation deficit of tea leaves which increased with the distances nearer to the gas flaring site.

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