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# Chemical characteristics of different concentrations of sugarcane industry wastes for algal culture

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**Abstract**
Sugarcane industry wastes media made of press mud, sugar mill effluent and by-products before inoculation of *Chlorella vulgaris*, the chemical properties were analyzed such as dissolved oxygen (DO), hardness, chemical oxygen demand (COD), total solids (TS), total suspended solids (TSS), total dissolved solids (TDS). Different categories including some inorganic media 18 (eighteen) treatments were tested such as inorganic media enriched with sodium acetate, sugarmill effluent medium (SMEM), press mud medium (PMM), normal molasses medium (NMM) and red sugar medium (RSM) with a control Bold basal medium (BBM). Hardness and COD were determined maximum 85.5 mg/l & 2724.21 mg/l in $PMM_{1.5\ g/l}$ and $RSM_{1.2\ g/l}$ respectively and the minimum hardness 22.8 were found in $RSM_{0.4}$ g/l, $RSM_{0.8\ g/l}$, $RSM_{12}$ g/l. Similarly the COD was found minimum 826.96 in 25% SMEM. Proximate composition shows that maximum protein 10.35% were determined in press mud followed by 4.17% in molasses and 0.27% in red sugar. Similarly significantly ($P<0.05$) higher lipid was determined in press mud than those in molasses and red sugar.

**Keywords:** chemical properties, sugar mill effluent, press mud, molasses red sugar

## 1. Introduction
There are 17 sugar mills out of which 15 are running in the country and release huge amount of effluents, wastes and by-product during sugar production season. The effluents are found both in liquid and solid state which cause aquatic pollution when released in the surrounding environment. The washing discharges of the factory machineries, boilers, condensers etc. are liquid form of effluent and the discharge of untreated solids and the boiler's refusal (scum). The solid wastes being widely known as bagase and press mud. Molasses an important liquid discharge and bagase of sugar mill having limited economic use in different purposes. These effluent, press mud, bagase and molasses are inexpensive local ingredients usually available in sugar mill area during sugar production period.

The sugar mill effluent releases large quantities of organic and inorganic compound, some of which may produce toxic substances by decomposition and chemical changes [1]. During sugar production period (November to May) sugar mills discharge a large quantity of untreated liquid wastes and effluents [3]. The use of microalgae for nutrient recovery with biomass production is being studied as an alternative water treatment system to make the aquatic environment free from pollution [11]. Studies on rubber and palm oil mill effluents proved that these effluents are basically suitable media for algae growth [6, 8, 9, 14]. Microalgae could be grown in diluted agroindustrial rubber and palm oil mill effluents to grow rotifer and moina, and then to feed larvae of fish, shrimp and aquatic organisms both in freshwater and marine environment [7]. Keeping in mind the above factors, inexpensive algal culture media such as, sugar mill effluent media (SMEM), press mud media (PMM), normal molasses media (NMM), red sugar media (RSM) in different concentrations were made. Therefore, the present investigation was undertaken to determine some important physico-chemical characteristics of the media and chemical composition of these raw ingredients used for the media preparation.

## 2. Materials and Methods
Before inoculation of *Chlorella vulgaris* in different culture media made of sugarcane industry wastes in different concentrations sub samples were collected and chemical properties such as dissolved oxygen (DO), hardness, chemical oxygen demand (COD), total solids (TS), total suspended solids (TSS), total dissolved solids (TDS), were analyzed following standard methods [4]. Including some inorganic media 18 (eighteen) treatment were tested in different categories such as inorganic media enriched with sodium acetate, sugarmill effluent media (SMEM), press mud media (PMM), normal molasses media (NMM) and red sugar media (RSM) with a control medium Bold basal medium (BBM). Fresh press mud media (PMM) was first decomposed in three different concentrations (0.5 g/l, 1.0 g/l and 1.50 g/l) in room temperature. Then supernatant were taken and diluted with de-ionized water in different ratio to make different concentrations of the media. Proximate composition of raw ingredients such as press mud, molasses and red sugar were determined according to Horwitz method [10]. Means of the determinants were compared through statistical analysis one way ANOVA using Tukey Test.

## 3. Results and Discussions
Proximate composition (Table 1) shows that the maximum crude protein 10.35% was determined in press mud followed

by 4.17% in molasses and 0.27% in red sugar. Similarly significantly (P<0.05) higher lipid, crude fiber and ash content were determined in press mud than those in molasses and red sugar. NFE was found significantly (P<0.05) higher 84.60% in red sugar followed by 43.63% in molasses and 27.09% in press mud.

Average chemical contents determined in different media indicate that maximum TS obtained 1551.0 mg/l in $NMM_{1.5\ g/l}$ and minimum obtained 415.67 mg/l in $SMEM_{25\%}$ with a medium TS value 973.33 mg/l were recorded in BBM. Maximum TSS recorded in $NPKAC_2$ was 141.33 mg/l and minimum 5.33 mg/l found in $SMEM_{25\%}$. Maximum DO level 3.98 mg/l was determined in 25% SMEM and minimum 3.01 mg/l was detected in $NMM_{1.5\ g/l}$. Hardness and COD were determined maximum 85.5 mg/l & 2724.21 mg/l in $PMM_{1.5\ g/l}$ and $RSM_{1.2\ g/l}$ respectively and the minimum hardness 22.8 were found in $RSM_{0.4}$ g/l, $RSM_{0.8\ g/l}$, $RSM_{1.2}$ g/l. Similarly the COD was found minimum 826.96 in 25% SMEM.

Total solids (TS), total suspended solids (TSS), total dissolved solids (TDS), dissolved oxygen (DO), hardness and chemical oxygen demand (COD) of different media indicate that maximum TS 1551.0 mg/l was determined in $NMM_{1.5\ g/l}$ and the lowest TS 415.67 mg/l was recorded in 25% SMEM. Maximum TSS and TDS were determined 141.33 mg/l & 1417 mg/l in $NPKAC_2$ and $NMM_{1.5\ mg/l}$ respectively. Dissolved oxygen (DO)

**Table 1:** Proximate composition of raw ingredients of local inexpensive sugarcane industry waste and related materials

| Composition | Press mud | Molasses | Red sugar |
|---|---|---|---|
| Moisture | $10.81^{b} \pm 0.01$ | $27.01^{a} \pm 0.01$ | $9.63^{c} \pm 0.83$ |
| Protein | $10.35^{a} \pm 0.01$ | $4.17^{b} \pm 0.32$ | $0.27^{c} \pm 0.01$ |
| Lipid | $1.75^{a} \pm 0.01$ | $0.43^{c} \pm 0.01$ | $0.74^{b} \pm 0.11$ |
| Crude fiber | $13.21^{a} \pm 0.01$ | $0.42^{b} \pm 0.01$ | $0.32^{c} \pm 0.01$ |
| NFE | $27.09^{c} \pm 0.24$ | $43.63^{b} \pm 1.15$ | $84.60^{a} \pm 1.07$ |
| Ash | $36.80^{a} \pm 0.17$ | $24.33^{b} \pm 0.87$ | $4.44^{c} \pm 0.24$ |

Nitrogen free extract (NFE) were calculated as 100- % (moisture + protein + lipid + crude fiber + ash). Means (±SD) with different superscripts in the same row indicate significant differences (P<0.05).

**Table 2:** Average chemical contents (mg/l) determined in different concentrations of sugar mill effluent media (SMEM), press mud media (PMM), normal molasses media (NMM), red sugar media (RSM) and Bold basal medium (BBM) & other inorganic media before inoculation of *Chlorella vulgaris*.

| Medium | TS | TSS | TDS | DO | Hardness | COD |
|---|---|---|---|---|---|---|
| BBM | $973.33^{h} \pm 14.19$ | $41.67^{h} \pm 3.51$ | $929.67^{h} \pm 5.51$ | $3.74^{e} \pm 0.02$ | $74.1^{abc} \pm 9.87$ | |
| NPK | $1121.33^{c} \pm 9.50$ | $96.0^{b} \pm 5.0$ | $993.67^{f} \pm 6.66$ | $3.60^{g} \pm 0.05$ | $57.0^{c} \pm 9.87$ | |
| BBMAC | $1381.33^{b} \pm 15.01$ | $51.0^{g} \pm 2.64$ | $1317.0^{b} \pm 7.54$ | $3.78^{e} \pm 0.06$ | $74.1^{abc} \pm 9.87$ | |
| NPKAC 1 | $933.0^{i} \pm 11.79$ | $87.67^{c} \pm 3.51$ | $818.33^{k} \pm 6.66$ | $3.88^{b} \pm 0.04$ | $57.0^{c} \pm 9.87$ | |
| NPKAC 2 | $1065.0^{f} \pm 9.54$ | $141.33^{a} \pm 4.16$ | $847.33^{j} \pm 4.16$ | $3.68^{f} \pm 0.04$ | $62.7^{bc} \pm 9.87$ | |
| $SMEM_{100}$ | $752.33^{j} \pm 11.59$ | $53.67^{f} \pm 2.51$ | $665.0^{l} \pm 4.58$ | $3.78^{e}$ 0.06 | $74.1^{abc} \pm 9.87$ | $1639.94^{g} \pm 16.01$ |
| $SMEM_{75}$ | $646.33^{l} \pm 7.51$ | $33.33^{j} \pm 3.06$ | $615.33^{m} \pm 8.50$ | $3.84^{c} \pm 0.03$ | $74.1^{abc} \pm 9.87$ | $1274.06^{h} \pm 12.20$ |
| $SMEM_{50}$ | $625.33^{m} \pm 11.06$ | $28.00^{j} \pm 3.0$ | 579.0n ± 4.58 | $3.94^{a} \pm 0.03$ | $68.4^{abc} \pm 17.1$ | $1025.85^{j} \pm 13.48$ |
| $SMEM_{25}$ | $415.67^{o} \pm 8.62$ | $5.33^{k} \pm 0.58$ | $395.67^{q} \pm 4.51$ | $3.98^{a} \pm 0.03$ | $57.0^{c} \pm 9.87$ | $826.96^{l} \pm 12.54$ |
| $PMM_{0.5}$ | $562.0^{n} \pm 8.0$ | $38.33^{h} \pm 2.51$ | $540.0^{p} \pm 4.0$ | $3.86^{c} \pm 0.03$ | $79.8^{ab} \pm 9.87$ | $885.11^{k} \pm 9.26$ |
| $PMM_{1.0}$ | $618.0^{m} \pm 6.0$ | $57.67^{f} \pm 3.51$ | $559.33^{o} \pm 7.02$ | $3.88^{c} \pm 0.03$ | $79.8^{ab} \pm 9.87$ | $1190.15^{i} \pm 8.48$ |
| $PMM_{1.5}$ | $679.0^{k} \pm 7.55$ | $87.33^{c} \pm 4.04$ | $605.0^{m} \pm 6.25$ | $3.71^{f} \pm 0.03$ | $85.5^{a} \pm 17.1$ | $1280.13^{h} \pm 17.55$ |
| $NMM_{0.5}$ | $972.67^{h} \pm 8.02$ | $54.0^{f} \pm 3.0$ | $911.33^{i} \pm 11.06$ | $3.06^{j} \pm 0.03$ | $68.4^{abc} \pm 17.1$ | $1946.83^{f} \pm 16.93$ |
| $NMM_{1.0}$ | $1263.0^{d} \pm 11.0$ | $57.67^{f} \pm 3.51$ | $1191.33^{d} \pm 5.69$ | $3.08^{j} \pm 0.03$ | $74.1^{abc} \pm 9.87$ | $2234.33^{d} \pm 22.26$ |
| $NMM_{1.5}$ | $1551.0^{a} \pm 8.18$ | $69.0^{e} \pm 3.61$ | $1470.33^{a} \pm 9.50$ | $3.01^{j} \pm 0.06$ | $74.1^{abc} \pm 9.87$ | $2676.86^{b} \pm 11.06$ |
| $RSM_{0.4}$ | $1043.67^{g} \pm 7.50$ | $49.67^{g} \pm 2.51$ | $950.67^{g} \pm 4.04$ | $3.46^{h} \pm 0.06$ | $22.8^{d} \pm 9.87$ | $2121.18^{c} \pm 14.75$ |
| $RSM_{0.8}$ | $1127.33^{c} \pm 9.01$ | $80.33^{d} \pm 2.51$ | $1049.67^{e} \pm 8.50$ | $3.26^{i} \pm 0.06$ | $22.8^{d} \pm 9.87$ | $2263.71^{c} \pm 10.89$ |
| $RSM_{1.2}$ | $1345.0^{c} \pm 11.53$ | $88.0^{c} \pm 4.0$ | $1225.33^{c} \pm 8.33$ | $3.24^{i} \pm 0.05$ | $22.8^{d} \pm 9.87$ | $2724.21^{a} \pm 12.43$ |

Column means (±SD) with different superscripts indicate significant differences (P<0.05).

Ranged from 3.01 to 3.98 mg/l and found maximum in 25% SMEM and minimum in $NMM_{1.0\ g/l}$. Maximum hardness 85.5 mg/l was recorded in $PMM_{1.5\ g/l}$ and the minimum hardness 22.8 mg/l was found in all the treatments of RSM. The COD was determined maximum 2724.21 mg/l in $RSM_{1.2\ g/l}$ and minimum 826.96 mg/l in 25% SMEM.

Press mud media (PMM) show the level of DO, hardness and COD ranged from 3.70 to 3.85 mg/l, 79.8 to 85.5 mg/l and 885.11 to 1280.13 mg/l in different concentration of the media. The TS, TSS and TDS of the media ranged from 562.0 to 679.0 mg/l, 38.33 to 87.33 mg/l and 540 to 605 mg/l, respectively in different concentrations of PMM. Sugar mill effluent media (SMEM) obtained a TS ranged from 415.67 to 752.33 mg/l, TSS ranged from 28.0 to 53.37 mg/l and TDS from 395.67 to 665.0 mg/l in different concentrations of the media. Sugar mill effluents always increase water temperature, COD, hardness, TSS, TDS, conductivity, chloride, total sulphide and $NH_3$ which can easily change water quality, increase axonic condition of water and load of biodegradable and non-biodegradable organic and inorganic matter [2, 13, 17, 18] The TSS, TDS, BOD and COD value indicate a greater degree of pollution by biodegradable and non-biodegradable organic matter [5, 7, 12] studied on chemical properties of different raw latex concentrate rubber effluent (LCRE), standard Malaysian rubber effluent (SMRE) and palm oil mill effluent and found high organic load.

Rajshahi sugar mill wastes were studied on stream water contaminated with effluents from sugar mill and its plankton population [3] and found high water temperature, high values of TSS, TDS, COD, electric conductivity, chloride, hardness, total sulphide, $NH_4$, $NH_3$ and $NH_4$-N, anoxic conditions and low Eh and $rH_2$ values. Some of these values analyzed during the study are shown in Table 3.

**Table 3:** Range of physico-chemical variables of Rajshahi sugar mill wastes

| Parameters | Nov-October | Parameters | Nov-October |
|---|---|---|---|
| Water temp.(°C) | 25-37.5 | $CO_2$ (mg/l) | 9-109 |
| TSS (mg/l) | 120-430 | Total hardness (mg/l) | 230-604 |
| TDS (mg/l) | 640-6140 | Ca hardness (mg/l) | 72-478 |
| DO (mg/l) | 0-1.5 | Mg hardness (mg/l) | 2-65 |
| BOD (mg/l) | 5.2-16.4 | $NH_4$ (mg/l) | 0.212-0.74 |
| COD (mg/l) | 892-2471 | $NH_3$ (mg/l) | 0.20-0.70 |
| Chloride (mg/l) | 98-142 | $PO_4$ (mg/l) | 0.008-0.014 |

A study on different physico-chemical properties of seven Mauritian sugar factories critical load to streams [18] explored COD varied from 90 to 1510 mg/l, TSS ranged from 10 to 252 mg/l, TDS from 370 to 654 mg/l, total hardness from 285 to 436 mg/l, $NH_3$ from 0.13 to 0.38 mg/l, and DO level from 2.3 to 3.2 mg/l. The findings were more or less similar to the findings of the present investigation.

Recently some researchers [16] investigated and analyzed the sugar industry waste water and found COD varies from 0.07-0.072, DO level from 6.88- 7.38, temperature 27.4 to 29.9, pH 5.14-7.94, TDS from 25.0 – 30.5, TSS varies 0.45 – 1.0, TS varies 1.25- 2.6. The results indicated more positive than present study. They also found BOD from 3.04 - 7.04, TR from 40.0- 219, and EC 45.5- 110.5 which were not studied in present investigation.

Some other researchers [15] studied the quality and management of waste water in sugar industry and found the physicochemical parameters shown in the following Table 4.

**Table 4:** Physico chemical parameters of sugar industry waste water

| Sl. No | Parameters | | Sl. No | Parameters | |
|---|---|---|---|---|---|
| 1 | Color | Dark yellow | 12 | Dissolved solid | 1650 |
| 2 | Odor | Fishery | 13 | Chloride | 250 |
| 3 | pH | 5.5 | 14 | Calcium | 361 mg/l |
| 4 | DO | 1.5 | 15 | Magnesium | 268 mg/l |
| 5 | BOD | 970 mg/l | 16 | Sulphate | 419 mg/l |
| 6 | COD | 3682 mg/l | 17 | Iron | 12.8 mg/l |
| 7 | Oil & grease | 12 mg/l | 18 | Lead | 0.065 mg/l |
| 8 | Temperature | 40º C | 19 | Zinc | 0.26 mg/l |
| 9 | EC | 2.23 s cm-1 | 20 | Copper | 0.135 mg/l |
| 10 | TDS | 1480 mg/l | 21 | Potassium | 113 mg/l |
| 11 | Suspended solid | 790 | 22 | Phosphate | 5.9 mg/l |

The researchers studied over a wider range of ingredients. The study is useful for comparative information related to common parameters yet the present investigation being conducted with sugar waste water very diluted form in prepared algal culture media.

At present algal culture for commercial live food development not yet developed in Bangladesh and the standard inorganic media like Bold basal medium (BBM) is expensive. So that it is not economic to produce microalgae using BBM for live food culture in aquaculture practice or other means. Sugar mill effluents, press mud and sugar mill by-product such as molasses are inexpensive and easily available ingredients in Bangladesh. These agroindustrial wastes and byproduct may be used as organic media for algal culture to fore commercial live food development and simultaneously the country will be free from aquatic pollution partially.

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