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European Journal of Experimental Biology, 2014, 4(1):242-249



## Lipid extraction for biodiesel production from municipal sewage water sludge

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### ABSTRACT

*Municipal sewage is a source of various lipids where the lipids can be extracted and converted to biodiesel by transesterification reaction. The present study reveals the properties of extracted Lipids like, Viscosity mm<sup>2</sup>/S, Density, kg/m<sup>3</sup>, Acid value mg KOH/g, Iodine value g I<sub>2</sub>/100 g, Value of saponification(mg KOH/g), Free fatty acid (%),Capric acid (%),Lauric acid (%),Myristic acids (%),Stearic acid (%), Oleic acid (%),Palmitic acid (%),others are studied from waste water sludge collected from Hebbala, Nayandahalli and Vrishabhavathy treatment plants located in Bangalore in both summer and Rainy seasons and revealed in the paper. The lipid content was estimated to be maximum in summer season ranging from 785 mg/g in Hebbala and 486 mg/g in Nayandahalli sludge samples. Hence, the sewage sludge proved to be a good raw material which can be utilized for the synthesis of biodiesel economically and ecofriendly, with the extension of present investigation.*

**Key words:** Lipid, Biodiesel, Sewage water sludge

### INTRODUCTION

The production cost of biodiesel is high due to the fact that 75–85% of the total cost is related to the raw materials like vegetable lipids and animal fats (1, 2 and 3); therefore there is an urgent need for alternative and cheap sources of lipid to produce biodiesel. The amount of lipid-rich wastewater increases every year due to urbanization, lipids are one of the most important components of natural foods which constitute one of the major types of organic matter found in municipal wastewater (4, 5) which may find their way into surface waters. Municipal wastewater treatment plants in the USA produce over 6.2 metric tons of dried sewage sludge every year (6).The use of sewage as a fertilizer is restricted in many countries of world due to bad odor, heavy metals and toxic substances (7), the alternative to sewage management and disposal challenge is to utilize it as a source of lipid feedstock for biodiesel production. Lipid is a natural mixture of triglycerides, diglycerides, monoglycerides, cholesterol, free fatty acids, phospholipids, etc. (8), where the municipal wastewater contains a significant amount of lipid fraction that is a composite of lipids, greases, fats and long chain fatty acids originating from the direct adsorption of lipids from domestic or from the phospholipids in the cell membranes of microorganisms, their metabolites and byproducts of cell lyses. Research has indicated that the lipids contained in sewage are a potential feedstock for biodiesel (9). To avoid the interference in the biodiesel synthesis, lipids are usually extracted with organic solvents; several options have been attempted however the lipid extraction for biodiesel production from municipal sewage poses great challenges for commercial realization. In this contest the wastewater generated in Bangalore city (1500 MLD - million liters per day) is studied for extraction and characterization of lipid which has a dependable supply of sewage throughout the year. Resource demand analyses for lipid biofuel production highlighting the need for

sustainable wastewater treatment together with production of biofuel. The purpose of this research was to investigate the feasibility of using primary and secondary sewage sludge for lipid extraction to produce biodiesel.

### MATERIALS AND METHODS

The wastewater generated in the city flows across three main storm water channels in the three catchments where wastewater samples were periodically assessed in two seasons for lipid extraction.

**Sample collection and preparation:** The municipal primary and secondary sludge samples were collected from three principle waste water treatment plants like Hebbala, Nayandahalli and Vrishabhavati located in Bangalore. Raw sludge was allowed to settle for 24 h at 10°C. After discarding the supernatant liquid the resulting sludge was then centrifuged at 3000 rpm for 10 min for further dewatering. Dewatered sludge was spread on tray and put in a fume hood for 4 days to dry under vacuum at ambient temperature. The dried sludge contained approximately 94% solids measured by drying the sludge at constant mass in an oven at 120°C. Dried sludge was crushed in a mortar and pestle homogenized and then stored in a freezer prior to use. The powdered sample was then washed with distilled water in order to remove all the soluble impurities, then dried at 50°C overnight prior to use as a dehydrating agent for the extraction of total lipids (oil). Extraction from organic solvents was carried out by following the method of Bligh and Dyer, 1959 (10).

**Determination of the physico chemical properties of the lipid:** A.O.A.C standard methods were used to determine the properties like Lipid, Viscosity, Density, Acid value, Iodine value and Value of saponification.

**Determination of fatty acid composition:** Analysis of FAME was carried out on Gas Chromatograph (11). The GC was equipped with Flame Ionization Detector (FID) and stainless steel column, dimension 10 X 1/8, packed with 5 % DEGS-PS. The column was conditioned at 180°C about 2 hours for attaining thermal stability before use. The operating condition was programmed at oven temperature 150°C (hold time 5min) with increasing rate 8°C/min to 190°C (hold time 0 min), 2°C/min to 200°C (hold time 10min), injection temperature 250°C and detector temperature 250°C. About 10µL of sample dissolved in hexane was loaded onto the column. Nitrogen was used as a carrier gas with flow rate of 20 ml/min.

The concentration of individual fatty acids in the test samples were determined by comparing the peaks obtained from the GC analysis with the peaks of authentic standards and n-heptane was used as an internal standard. All tests were performed in triplicate the experimental design was completely randomized with three replicates. All data were expressed as mean values ± SE. The comparison between the mean values were tested using Duncan's new multiple range test and the ANOVA was also performed to find out the LSD ( $p = 0.05$ ).

### RESULTS AND DISCUSSION

The lipid content was more in Hebbala sample i.e. 78.5% and 65% compare to 48.6% and 55.6% in Nayandahalli and 58.7% and 60.2% in Vrishabhavathi (Summer and Rainy seasons respectively), the previous studies (20) reported that lipid from non edible lipid plant seeds yielded around 63% which is lesser compared to the present studies, hence sewage sludge samples could be used as a better and economical feedstock for biodiesel production compare to non edible plant (table 1,2,3 and figure 1)

**Table 1: Properties of lipid extracted from Hebbala**

Parameters	Summer season	Rainy season
Lipid (mg/g of sludge)	785.0	650.0
Viscosity mm <sup>2</sup> S	40.5	42.2
Density, kg/m <sup>3</sup>	1.10	1.05
Acid value mg KOH/g	2.34	2.22
Iodine value g I <sub>2</sub> /100 g	62.4	64.5
Value of saponification(mg KOH/g)	151.61	139.42
Free fatty acid (%)	0.89	0.95
Capric acid (%)	03.52	03.30
Lauric acid (%)	03.39	03.00
Myristic acids (%)	10.44	08.31
Stearic acid (%)	18.73	17.56
Oleic acid (%)	21.41	20.11
Palmitic acid (%)	40.03	38.01
others	02.05	09.71

Values are the means of three replicates each and two consecutive seasons. Data were subjected to analysis of variance and compared for significance according to DMRT ( $P=0.05$ ).

**Table 2: Properties of lipid extracted from Nayandahalli**

Parameters	Summer season	Rainy season
Lipid (mg/g)	486	556
Viscosity mm <sup>2</sup> S	38.69	39.85
Density, kg/m <sup>3</sup>	1.21	1.14
Acid value mg KOH/g	3.24	2.12
Iodine value g I <sub>2</sub> /100 g	83.2	74.5
Value of saponification(mg KOH/g)	149	140
Free fatty acid (%)	1.10	0.87
Capric acid (%)	03.02	03.00
Lauric acid (%)	02.98	02.81
Myristic acids (%)	09.41	09.13
Stearic acid (%)	14.91	14.57
Oleic acid (%)	21.31	20.88
Palmitic acid (%)	40.33	39.51
others	09.04	10.01

Values are the means of three replicates each and two consecutive seasons. Data were subjected to analysis of variance and compared for significance according to DMRT ( $P=0.05$ ).

**Table 3: Properties of lipid extracted from Vrishabavati**

Parameters	Summer season	Rainy season
Lipid (mg/g of sludge)	587	602
Viscosity mm <sup>2</sup> S	38.56	41.02
Density, kg/m <sup>3</sup>	1.04	1.18
Acid value mg KOH/g	3.39	2.24
Iodine value g I <sub>2</sub> /100 g	70.5	88
Value of saponification(mg KOH/g)	129	138
Free fatty acid (%)	00.68	00.76
Capric acid (%)	03.41	03.00
Lauric acid (%)	02.44	02.10
Myristic acids (%)	08.59	08.26
Stearic acid (%)	16.63	15.31
Oleic acid (%)	21.00	19.75
Palmitic acid (%)	38.36	37.56
others	09.57	14.02

Values are the means of three replicates each and two consecutive seasons. Data were subjected to analysis of variance and compared for significance according to DMRT ( $P=0.05$ ).

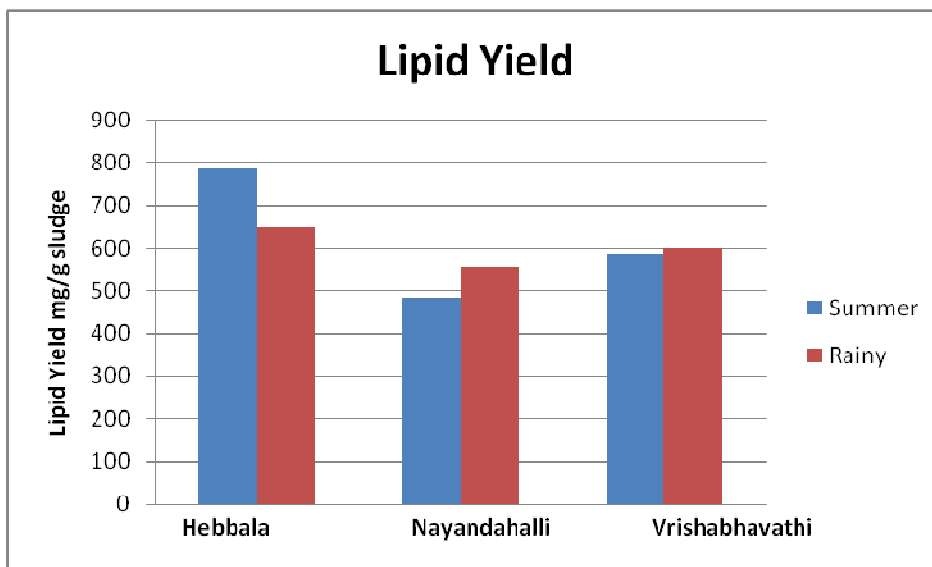


Figure 1: Comparison of the lipid yield

The density is a parameter used to determine the concentration of a solution (12) the density of the extracted lipid from all the three sites are denser in summer season and varied from 1.04 kg/m<sup>3</sup> (Vrishabhavathi) to 1.21 kg/m<sup>3</sup> (Nayandahalli) the data were not correlated with the reports from (13) (table 1,2,3 and figure 2).

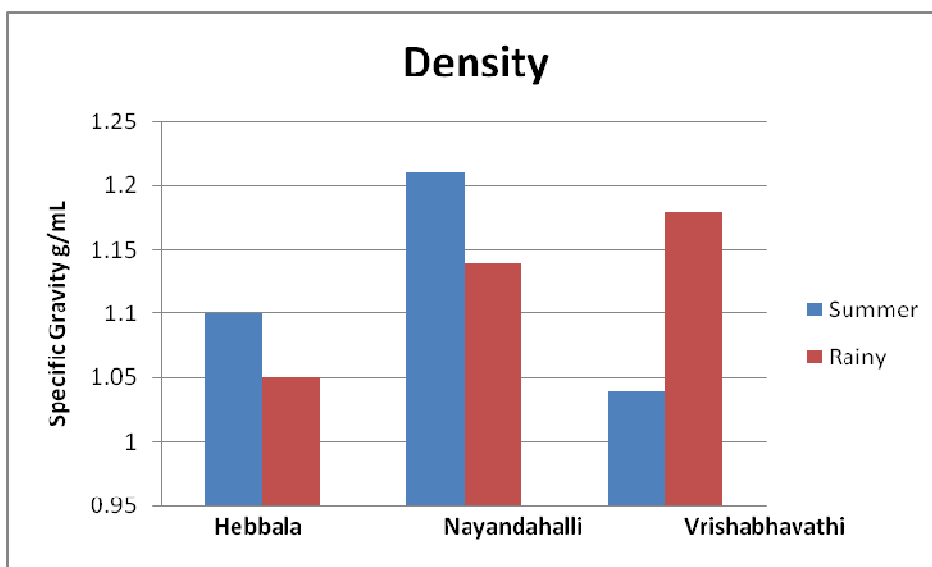


Figure 2: Comparison of the extracted lipid's Density (Specific gravity)

Viscosity is stated as the friction of a liquid to flow it increases with molecular weight and content of saturated fatty acids/decreases with increasing unsaturated level and temperature (14). The viscosity of the extracted lipid were varying from a highest value of 42.2 mm<sup>2</sup>/s in the sample of Hebbala plant during rainy season to 38.56 mm<sup>2</sup>/s in the sample of Vrishabhavathi during summer season. The viscosity of extracted lipid must be reduced for biodiesel application since the kinematic viscosity of biodiesel is lower as per international standards. Different approaches like blending, preheating and advance transesterification are being used to reduce the viscosity and make them suitable for engine applications (15, 16) (Figure 3).

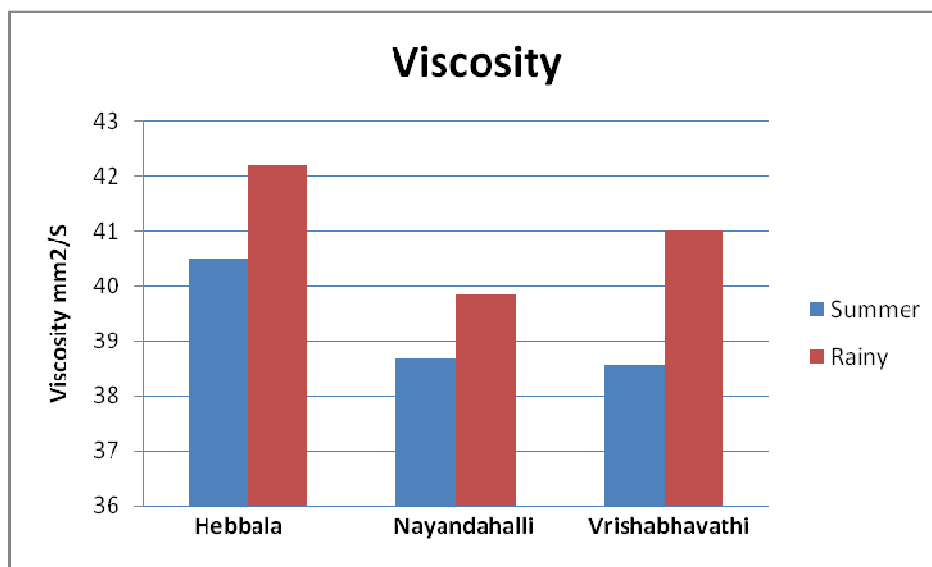


Figure 3: Comparison of the extracted lipid viscosity

The acid value is the concentration of total free fatty acids presents in a source this will not support the quality of biofuel source on higher values. The present investigation reveals that the acid value is highest in Vrishabhavathi sample (3.39 mg KOH/g) and lowest in Nayandahalli sample (2.12 mg KOH/g). Since, the extracted lipid is crude in nature; more than 1.0 mg/KOH reveals the lower quality of the lipid which needs to further be purified, processed and it is agreements with the findings of (17) (Figure 4).

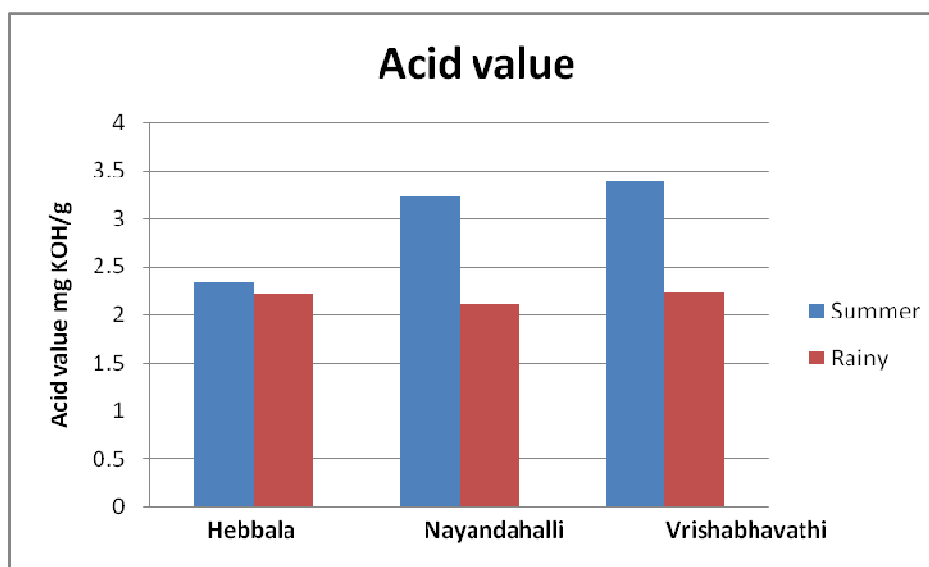


Figure 4: Comparison of the acid value in the extracted lipid

Iodine value is a measure of the unsaturation of fats and lipids high Iodine values reveal the high unsaturation (18). The Iodine values ranges from 62.4 to 88 g I<sub>2</sub>/100 g and the Standard iodine value is lesser than 120 as per EN 14214 specification for the raw lipid used to synthesize biodiesel. The limitation of unsaturated fatty acid is necessary due to, the fact that heating higher unsaturated fatty acids results in polymerization of glycerides which can results in deposition or deterioration of the lubrication property (19) (Figure 5).

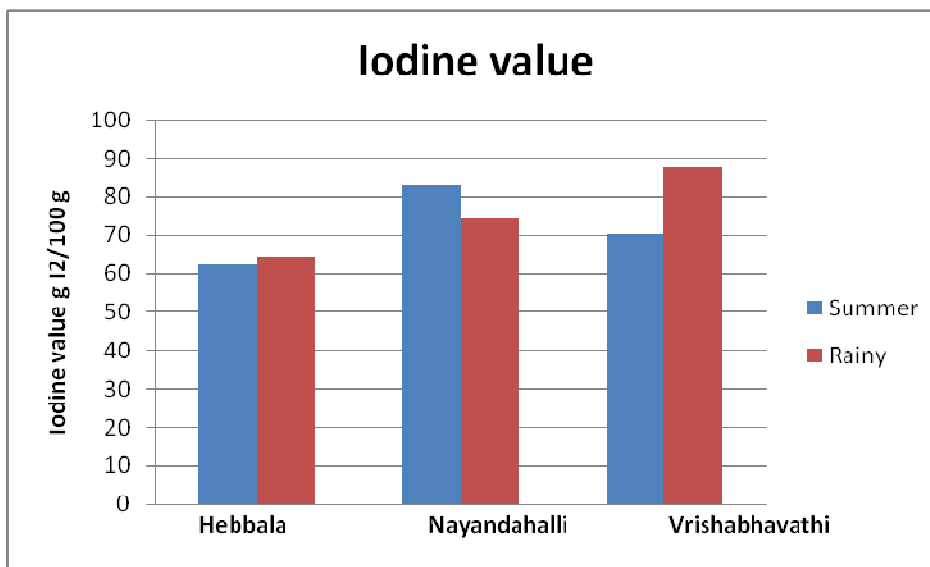


Figure 5: Comparison of the iodine value of the extracted lipids

The Saponification value were 151.61 and 139.42 (mg KOH/g) in Hebbala sample, 149 and 140 in Nayandahalli sample and 129 and 138 in Vrishabhavathi samples in Summer and Rainy seasons respectively. High saponification values indicate that, lipid samples are rich with triglycerides, diglycerides and monoglycerides can be useful in the production of soap and shampoo industries the present result is lesser than the work of (20) (Figure 6).

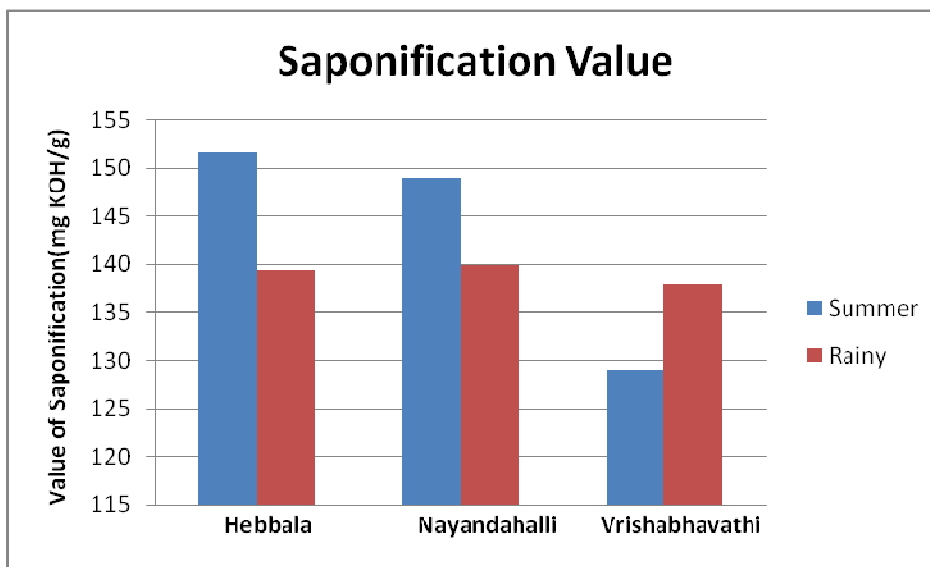


Figure 6: Comparison of the saponification value of the extracted lipids

The free fatty acid content with greater than 1%w/w, will lead to soap formation, the separation and purification will be very much difficult and will become a poor raw material for biodiesel production, in the present study, the free fatty acid content is less than one in all the six samples. The acid catalyzed esterification of the extracted lipid sample is an alternative (21) to solve the above problem, but is a slow reaction when compared to the base catalyzed transesterification reaction (Figure 7).

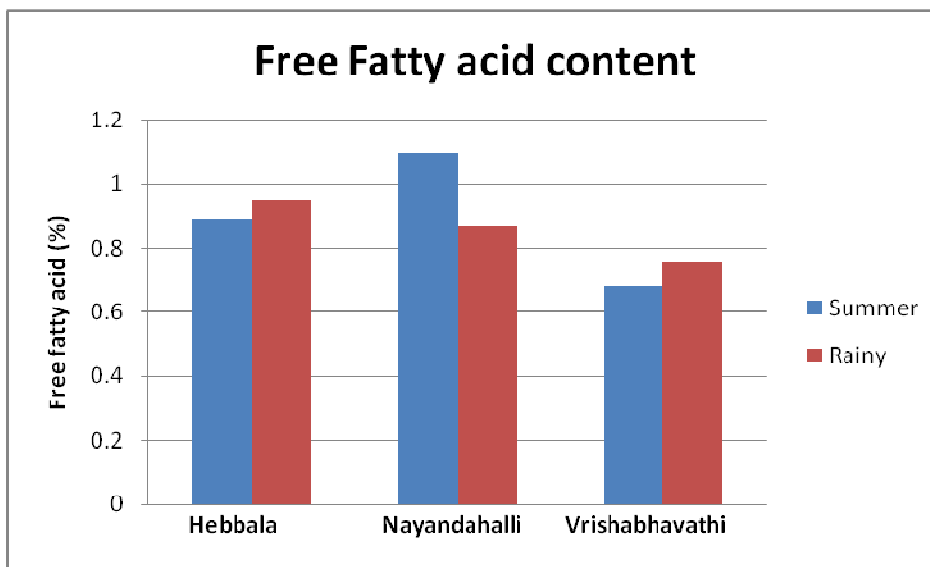


Figure 7: Comparison of the free fatty acid content

The fatty acid composition upon GC analysis was found to be with the varying concentrations of fatty acids with carbon chain length from ranging from C-4 to C-18 determined as follows. The content of was Capric acid (3.52% and 3.30%, 3.02 and 3.0, 3.41 and 3.0% , Lauric acid (3.39 and 3.00, 2.98 and 2.81, 2.44 and 2.10% , Myristic acids (10.44 and 8.31, 9.41 and 9.13 , 8.59 and 8.26 % , Stearic acid, 18.73 and 17.56, 14.91 and 14.57 , 16.63 and 15.31% ,Oleic acid, 21.41 and 20.11, 21.31 and 20.88 , 21 and 19.75 % , Palmitic acid 40.03 and 38.01 , 40.33 and 39.51 , 38.36 and 37.56 % ,others 2.05 and 9.71, 9.04 and 10.01, 9.57 and 14.02 % concentrations were present in Hebbala, Nayandahalli and Vrishabhavathi samples respectively during summer and rainy seasons.

### CONCLUSION

Municipal sewage sludge is readily available and is a potential source of lipid for biodiesel production. But there are few challenges for biodiesel production from sludge, lipid extraction from sludge is expensive and requires large volume of organic solvents and the amount of lipid depends on the sources and type of sludge. Solvent selection, sludge to solvent ratio, extraction time, temperature and solvent recovery are among the factors that affect lipid extraction efficiency and cost. Optimization of these factors is necessary for efficient lipid extraction.

### Acknowledgements

The authors are thankful to Bangalore University for providing infrastructural facility.

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