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TREATMENT AND RECYCLING OF WASTE WATER FROM DAIRY INDUSTRY USING *SPIRULINA PLATENSIS*

PATIL NILAM SARJERAO[1] AND KURHEKAR JAYA VIKAS[2*]

[1,2]Department of Microbiology, Bharati Vidyapeeth's, Dr. Patangrao Kadam Mahavidyalaya, Sangli 416 416, Maharashtra, India

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ABSTRACT

Dairy industry is considered as highly polluting industries in India. Dairy effluent contains high organic load. If partially treated or untreated waste water is disposed off in the natural water bodies, there is a possibility of eutrophication due to high content of elements like Phosphorus, Nitrogen and Carbon etc. Dairy waste is the rich source of proteins like Casein and sugars like Lactose. Biological treatment was observed as effective and ecofriendly, as compared to physicochemical process. Biotreatment involves microbial degradation of waste water. Microalgae are known for complete removal of contaminants like Phosphorus, Nitrogen etc. In the current study, *Spirulina platensis* (blue green algae) has been used for the treatment and recycling of physicochemically treated dairy waste water. Open air system was designed for treatment. *Spirulina platensis* was grown for 28 days in physicochemically treated waste. The process was found to be environmental friendly and low investment. Chlorophyll, protein and carbohydrate content of the *Spirulina platensis* was studied and found increased at various concentrations like 20% to 100% during the treatment period. The aim of present investigation was to estimate the application of *Spirulina platensis* in treatment and recycling of waste water from dairy industry.

KEY WORDS : Dairy waste water, Biotreatment, *Spirulina platensis*, Open air system, Recycling

INTRODUCTION

Domestic, industrial and agricultural wastes are major water pollution sources of throughout India. Worners (1976) 1960's U.S. senate committee noted that industry processing dairy products is second most important source of pollution load in natural water streams. In India, approximately 70% of fresh water has been polluted due to discharge of industrial waste and domestic waste to the natural water streams like lakes, ponds and rivers (Sangu and Sharma, 1987). In India, because of huge population, demand of dairy products is high for human consumption. To fulfill this demand, many dairy industries are processing and dispatching the variety of dairy products like milk pouches, yoghurt, shrikhand, amrakhand, buttermilk, curd etc. Enormous quantity of water is required bythe industry for different processes like clean in place (CIP), raw material washing and packaging, leading to high amount of dairy wateras waste received in Effluent Treatment Plant (ETP). Dairy industry influent of waste water contains lipids, proteins, washing chemicals etc. The high organic load of the waste water can be reduced by either chemical treatment or biological treatment. Ecofriendly process for the reduction of organic load is found to be more applicable for the treatment. Chemical treatment is known to have bad impact on the environment so now a days biological treatment are regularly used in all food processing industries. Biotreatment involves the use of efficient microorganisms like bacteria, microalgae having ability to degrade the waste water contents. For many decades, Microalgae are used for waste water treatment. Dairy waste water contains high amount of Nitrogen mainly from milk proteins and alkaline cleaners, inorganicand organic Phosphorus, small

amounts of Na, Mn, K, Cl, Ca, Co, Mg, Ni (Demirel *et al.,* 2005 and Prakash *et al.,* 2011). Microalgae grow in sunshine and utilize large amount of Phosphorus, Nitrogen and other mineral nutrients. Microalgae play a major role in the recycling of waste water from various sources like dairy, herbal, pharmaceutical industries, by different metabolic processes like enzymatic degradation, accumulation and absorption (Vanerkar *et al.,* 2015). There are many reported Microalgae having a role in utilization of waste water like *Scenedesmus quadricauda, Spirulina platensis, Chlorella sp*. (Vonshak *et al.,* 1983). *Spirulina platensis* is a blue green algae growing in strong sunlight, under high temperature and alkaline conditions (Dineshkumar *et al.,* 2016).

In the present study, *Spirulina platensis* culture was used for the treatment of physicochemically treated dairy waste water. Open air system was designed for the treatment. Study was employed for 28 days using dairy waste water of various concentrations like 20, 40, 60, 80 and 100%. Growth of *Spirulina platensis* was measured at every four days interval period. It was observed that physico-chemical parameters of waste water from dairy like COD (Chemical Oxygen Demand), BOD (Bio-chemical Oxyġen Demand), TSS (Total Suspended Solids), TS (Total Solids) etc. were reduced after treatment.

MATERIALS AND METHODS

Collection and Identification of Algae Sample

Samples of algae were taken from a local pond of Sangli, Maharashtra, India. Sampling was done in three sterile glass bottles. Samples were transported immediately to the laboratory and preserved at 4 ºC. The *Spirulina platensis* culture was microscopically identified by Olympus microscope BX51 at 100X and 400X magnification (Olympus, Japan) and confirmed using the reported keys (Prescott,1978).

Enrichment of *Spirulina platens* is Culture

Enrichment of *Spirulina platensis* was done in the 250 mL capacity Erlenmeyer flask containing 100 mL of sterile Zarrouk's medium. As per investigations by Dinesh Kumar *et al.,* (2016) *Spirulina platensis* showed maximum growth in Zarrouk's medium of pH 8.8 to 9.0 at 30 °C. Shaking was done manually, twice a day.

Collection and Physicochemical Characterization of Waste Water from Dairy Industry

Waste water of dairy industry was taken from a reputed dairy industry Effluent Treatment Plant located in Sangli, Maharashtra, India. Sterile glass bottles were used for collection and sample was kept at 4R ºC in the refrigerator. Waste water sample was checked for physicochemical parameters as pH, Temperature, Color, Turbidity, Total Dissolved Solids (TDS), Total Suspended Solids (TSS), Total Solids (TS), Chemical Oxygen Demand (COD), Biochemical Oxygen Demand (BOD), Chloride, Sulfate, Lipids, Oil and grease, Nitrogen and Phosphorus (APHA, 2005).

Treatment of Waste Water from Dairy Industry Using *Spirulina platensis*

Open air algal cultivation (Open air system) allows cost effectiveness, free solar energy and low energy for the mixing (Hwang *et al.,* 2016). Drawback of the process is that it has no temperature control. Physicochemically treated waste water was diluted to different concentrations like 20%, 40%, 60%, 80%, 100% and control, using water as a diluent. Erlenmeyer flask of 500 mL capacity was taken for each dilution and 200 mL sample was added in the flask. 5 mL of *S. platensis* culture was inoculated in all flasks, which were placed in shadow at room temperature (Dineshkumar *et al.,* 2016). Manual shaking was employed three times a day, temperature and light was measured every day. Physicochemical parameters of waste water from dairy were checked before and after treatment of *Spirulina platensis.* Growth rate of *S. platensis* was measured after every four days interval for 28 days by taking absorbance (optical density) at 660nm using spectrophotometer. Protein content of *S. platensis* was estimated (Lowry *et al*., 1951). Estimation of chlorophyll was done by pellets of algae dissolving in 80% acetone, measured using spectrophotometer (Vanerkar *et al.,* 2015). Anthrone reagent method was carried out for carbohydrate estimation (Herbert *et al.,* 1977).

RESULTS AND DISCUSSION

Identified *Spirulina platensis* culture was enriched in Zarrouk's Medium. Flask was kept at controlled temperature as well as light. Shaking was done manually thrice a day. Physicochemically treated waste water from dairy industry was collected from the Effluent Treatment Plant [ETP] of dairy industry located in Sangli. Sample was analyzed for all physicochemical parameters before the inoculation of *Spirulina platensis* culture. Waste water

Table 1. Composition of Zarrouk's medium (Dineshkumar *et al.*, 2016)

Sr. No.	Ingredients	Amount (gms/L)
1.	$NaHCO_3$	16.8
2.	$NaNO_3$	2.5
3.	NaCl	1.0
4.	K_2SO_4	1.0
5.	K_2HPO_4	0.5
6.	$MgSO_4.7H_2O$	0.2
7.	$FeSO_4.7H_2O$	0.01
8.	$CaCl_2.2H_2O$	0.04
9.	EDTA	0.08
10.	Distilled Water	1000 ml

characteristics of untreated, physicochemically treated (before the application of *S. platensis*) and after treatment have been shown in Table 2. Temperature was recorded as 30º C +/- 2ºC and pH was as 5.5, 6.5 & 6.8 of untreated, before and after treatment of *S. platensis* respectively. BOD value of waste water was reduced up to 95 mg/L. However, COD was recorded 140 mg/L after treatment. Reduction was observed for other parameters like Sulphate, Chlorides, Nitrogen and Phosphorus. Similar results were recorded by Phang *et al.*, (2000) reported reduction in COD (Chemical Oxygen Demand), active Phosphorus and ammonical nitrogen from sago industry waste water by 98, 99.4 and 99.9% respectively after the application of *Spirulina platensis*.

An open air system was provided for the treatment. Different concentrations of waste water from dairy industry were collected in six flasks like 20, 40, 60, 80, and 100% and control respectively. 200 mL of waste water samples was collected in individual flasks. Fresh water sample was kept as a control. However 5 mL *S. platensis* culture was inoculated in every flask and flasks were kept under moderate sunshine for 28 days. Shaking was done manually thrice a day. Growth of *S. platensis* was observed every four days of interval, taking absorbance at 660 nm (Table 3). At 12th and 16th day of incubation, more absorbance means high growth was observed, after 16th day onward, growth reduced as per the absorbance. Results indicate that *S. platensis* has an ability to utilize all nutrients present in waste water and as the concentration of

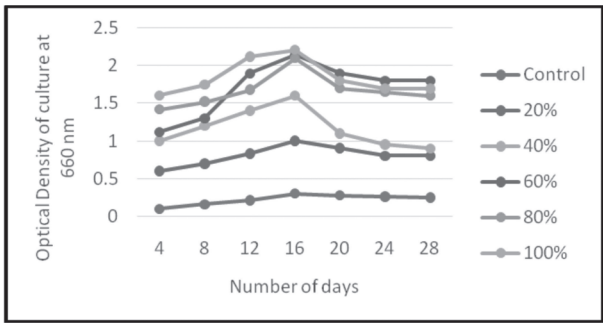

Fig. 1. Growth rate of *Spirulina platensis* at different concentrations of effluent

Table 2. Physicochemical characteristics of waste water from dairy industry untreated, before and after application of *Spirulina platensis*

Parameters	Untreated Dairy waste water	Before application of *S. platensis* (physico-chemically treated waste water)	After application of *S. platensis*
pH	5.5	6.5	6.8
Color	Milky white	Light milky white	Clear water
Temperature	30º+/- 2ºC	30º +/- 2ºC	30º +/- 2ºC
Turbidity	1230	450	50
Total Solids (TS)	1930	860	130
Total Dissolved Solids (TDS)	1250	360	40
Total Suspended Solids (TSS)	680	500	90
Biological Oxygen Demand (BOD)	1050	786	95
Chemical Oxygen Demand (COD)	3057	1965	140
Chloride	485	310	59
Sulfate	390	250	40
Oil and Grease	157	96	10
Lipids	630	495	25
Nitrogen	530	140	95
Phosphorus	380	90	20

* Except pH and Color, all values are expressed in mg/lit and Turbidity [NTU].

waste water increases from 20% to 100%, *Spirulin aplatensis* growth increases. This indicates that *S. platensis* has a capacity to tolerate high organic load. *Spirulina platensis* growth in control sample was low, throughout 28 days of incubation

After the 28 days treatment, algae culture was tested for protein, chlorophyll and carbohydrate content (Table 4). The increased values of proteins, chlorophyll and carbohydrate indicating increased survival rate of *Spirulina platensis*. Algal treatment of industrial wastewater is found to have better efficiency than conventional tertiary treatments because this process is mediated through combination of high dissolved oxygen concentration, nutrient uptake and elevated pH, which is economically cheaper and ecofriendly (Fulke, 2013).

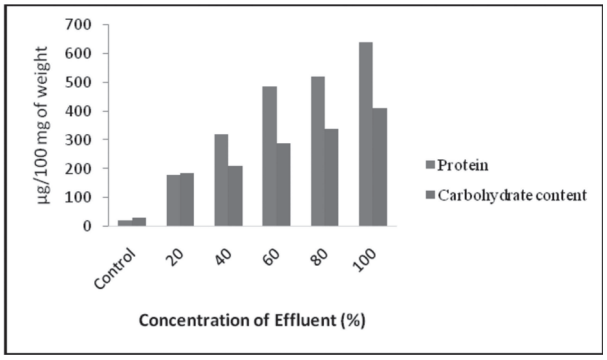

Fig. 2. Effect of dairy industry effluent on protein and carbohydrate content of *Spirulina platensis*

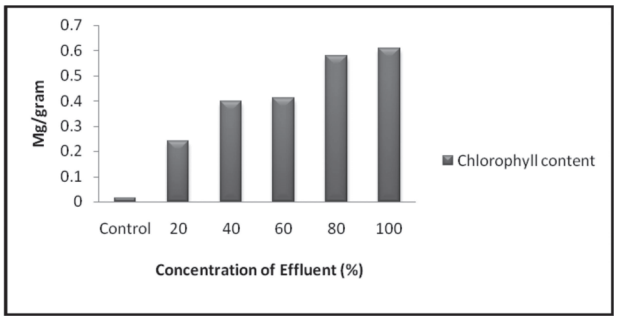

Fig. 3. Effect of dairy industry effluent onchlorophyll of *Spirulina platensis*

CONCLUSION

The present study results showed that Microalgae *(Spirulina platensis)* have an ability to utilize the nutrients of waste water from dairy industry and reduce organic load up to lower concentrations so that the treated water can be reuse. Recycling of treated water will help in conservation of water.

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Table 3. Growth rate of *Spirulina platensis* in dairy waste water

	Incubation Period (in Days)						
	4	8	12	16	20	24	28
Control	0.10±0.04	0.16±0.04	0.21±005	0.30±0.06	0.28±0.05	0.26±006	0.25±0.05
20	0.60±0.05	0.70±0.05	0.83±0.6	1.00±0.07	0.90±0.07	0.80±0.08	0.80±0.08
40	1.00±0.06	1.20±0.06	1.40±0.7	1.60±0.08	1.10±0.08	0.95±0.09	0.90±0.09
60	1.12±0.07	1.30±0.07	1.90±0.8	2.14±0.10	1.90±0.09	1.80±0.10	1.80±0.10
80	1.42±0.08	1.52±0.08	1.68±0.1	2.10±0.20	1.70±0.10	1.65±0.10	1.60±0.10
100	1.61±0.10	1.75±0.10	2.12±0.2	2.21±0.20	1.80±0.10	1.70±0.10	1.70±0.10

*Optical Density of culture measured at 660 nm.

Table 4. Effect of waste water from dairy industry on different metabolites and chlorophyll of *Spirulina platensis*

Concentration of Effluent (%)	Protein	Carbohydrate	Chlorophyll
Control (No effluent)	23	30	0.015
20	178	185	0.242
40	320	210	0.398
60	485	290	0.410
80	520	340	0.580
100	640	410	0.610

*Chlorophyll content was measured in mg/g, Protein and Carbohydrate content was measured in (µg/100 mg of weight).

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