**Waste Water Treatment Inverse Fluidisation Method Using Algae**

**Vivek Kumar Soni1, Bhojram Sahu2**

1MTech Scholar, Environmental & Water Resource Engineering, Bharti Vishwavidhyalaya, Durg, Chattisgarh, India

2Assistant Professor, Civil Engineering, Bharti Vishwavidyalaya, Durg, Chattisgarh, India

**ABSTRACT**

**Chemical Oxygen Demand (COD) and Biological Oxygen Demand (BOD) play important role in determining the quality of wastewater. Hence it is necessary to calculate COD and BOD of water before setting up of wastewater treatment plant. Algae has been used for decades for various purposes. It is one of the important characteristics is to nitrogen, phosphorus etc. ,which are harmful for drinking and other purposes but they act as food for algae. Thus in this study COD and BOD analysis is done for sterilized and non-sterilized wastewater after and before treating it with algae in inverse fluidization under aerobic condition, for different time interval and found that percentage reduction in COD and BOD for sterilized waste water gives greater value than non-sterilized water the reason for this difference being the decrease in the competition between algae and other micro-organism which are present in raw waste water. And COD % reduction is 65-70 % and BOD % reduction is 68.75- 70.5%.**

**Keywords:** Chlorella Scenedesmus ,COD, BOD, wastewater, inverse fluidization unit

1. **INTRODUCTION**

Among many conventional processes available for wastewater treatment, inverse fluidisation process, which is a three-phase fluidisation process, has been widely used for many applications such as hydro-treating and conversion of heavy petroleum and synthetic, crystallization, food processing, biomedical engineering, methanol production, treatment of municipal sewage wastewater, and similarly many processes. Some of the benefits which one’s process can gain if this unit is used are: easy to handle, less consumption of power, low space requirement, less chemical waste, and eco-friendly as it does not produce any chemical as its waste after the process. Indeed, the most significant feature of it is high efficiency as compared to the other conventional fluidization processes. The name inverse fluidisation comes from the direction of flow of liquid and gas which depends upon the density of the particle. Here, the liquid is fed continuously from the top using a pump if it is a continuous process, and gas is released from using a sparger from the bottom after it has been compressed in a compressor. Thus, it makes the process a counter-current flow process. In this counter-current flow process, the density of the particle is lesser than that of the liquid, which is in a continuous phase. With the rapid growth in population and industrialization, it is leading to the depletion of natural resources and causing major environmental problems such as water pollution, soil pollution, etc. The environmental problem which is of our concern is water pollution, which is mainly caused due to the discharge of heavy metals from steel, dairy, and fertilizer industries and nitrogen, phosphorus, sulphides, and chlorides. Due to the rapid use of nitrogen in fertilizer industries, an excessive amount of it may cause several health-related problems and causes eutrophication and acidification of water bodies. To overcome this process, there are various methods which have been used for decades, but the question that arises is: which process is more economical and offers numerous benefits over others.

**Why inverse fluidization technique and not the conventional one?**

* The bio film thickness which grows very fast on the surface of the solid particle ,if provided proper conditions. Sometimes it also happens that bio film thicknes increases so much that it causes bloom and proper mixing and growth of film is degraded. Thus some new particle have to be added to provide new surface to the biomass from time to time. The advantage of IFBR lies here that it controls biofilm thickness in a very narrow range.
* Due to power failure sometimes it needs to start the fluidization process from the beginning itself but with the IFBR this problem is almost sorted out as we can re- fluidize the process.
* The growth of microorganism is very faster as seen from the literature survey due to high mass transfer rate.
* Carry over of particleis minimized due to low particle or solid attrition.

 **Type of Algae and why it is used in waste water treatment process.**

* Algae involves a process which is very similar to the green plants, and the most common process in plants is photosynthesis. Algae absorbs sunlight, which is a source of carbon dioxide for it, and converts it into oxygen, and photosynthesis takes place through chlorophyll present in it. Algae size varies from single-cell to branched size of visible length. Some of the algae which grow in wastewater are Chlorella sp., Spirulina sp., Microactinium sp., and some more.

* The treatment of wastewater can be achieved by biodegradation of it using bacteria or algae. Biodegradation converts organic matter into smaller molecules which requires oxygen for the process. And the supply of oxygen is tedious and costly. Thus, it is better to use natural abundance sources of oxygen, which can give a lot of benefits apart from biodegradation. Algae absorbs various compounds and nutrients such as nitrogen, phosphorus, and metals required for its growth.

In all other conventional method for wastewater treatment which does not uses algae the treatment process produces lots of sludge which eventually goes to off-site for its disposal and maintaining sludge which is diurnal and seasonal is a costly process. Some of the benefits which is prominent in today’s century are reduction in green-house -gases and production of useful products from end product which is a highly rich nutrient containing algae itself and can further be used for production of bio fuel and diet supplementary. Aeration is an energy intensive process and accounts for 45-70 % of total energy cost of treatment plant. Algae consumes CO2 in a larger amount than it is released during the process. ChlorellaScenedesmus is one among the fastest growing genus of single celled green algae, includes 14%-22% of lipid, 51%-58% of protein, 12%-17% of carbohydrates, and 4%-5% of nucleic acid.Algae can act as a bio-filter for nutrient laden, CO2 laden, and can convert low oxygen water into highly rich oxygen water. Thus any wild algae can be grown in the area where the wastewater is reserved. End use of algae can be in production of biodiesel or biofuel as compared to soyseed (60-100gallons), coconut (230 gallons), and palmoil(500 gallons) can produce 5000 or more gallons per acre of area.

 **Biological Oxygen Demand (BOD) and Chemical Oxygen Demand (COD)**

COD test is used to measure the amount of organic compounds in water. In other words, we can say that it is the amount of oxygen required to chemically oxidize the pollutants. The applicable range of COD is 3–900 mg/ml.BOD test is used to determine the amount of oxygen required by the microorganisms to break the organic material present in the sample at a particular temperature over a specific period of time. Generally, the time taken for the test is 5 days at a temperature of 20 degrees Centigrade. It is also a principal test which predicts the biodegradability of any water or wastewater sample. The efficiency of wastewater is measured by measuring the effluent BOD and influent BOD of the sample taken. Any effluent to be discharged into the water should have BOD less than 30 mg/ml. COD value is always greater than BOD value. It is found from the research that the COD values for domestic and industrial wastewater is about 2.5 times the BOD value. The ratio of BOD to COD, if greater than 0.8, then it is considered that the water is highly polluted and amenable to biological treatment.

1. **METHODOLOGY**

2.1 Materials Required:

1. Algae

-Chlorella Scenedesmus and local algae from pond

 -Quantity used: 250ml

Table-1: Nutrients required for growing Chlorella Scenedesmus

|  |  |  |
| --- | --- | --- |
| S.No. | Compounds Name | Quantity per litre |
| 1. | Fog’s Medium* Magnesium sulphate hepta-hydrate (MgSO4.7H2O)
* Dipotassium hydrogen phosphate (K2HPO4)
* Micro nutrients solution
* Calcium chloride hydrated (CaCl2. H2O)
* Fe-EDTA solution
* Distilled water
* Agar (Difco)
 | 0.2g0.2g1ml 0.1g5.0ml1.0L12.0g |
| 2. | Micronutrient solution* Hydrated Manganese Chloride (MnCl2.4H2O)
* Boric Acid (H3BO3)
* Zinc sulphate hepta hydrate(ZnSO4.7H2O)
* Sodium Molybdate (Na2MoO4.2H2O)
* Copper Sulphate penta-hydrate (CuSO4.5H2O)
* Distilled water
 | 181.0mg286.0mg22.0mg39.0mg8.0 mg 100.0ml |
| 3. | Fe-EDTAIn hot water 745.0 mg of Na2 EDTA was dissolved and then557.0 mg of FeSO4.7H2O was added. The solution was boiled for few minutes and the volume was made to 100.0 ml. |  |

2.Wastewater from Rourkela Steel plant

- Quantity used: 1 litre

Table-2: Composition of waste water obtained from Rourkela Steel plant, Rourkela, Orissa

|  |  |
| --- | --- |
| Component | Amount in ppm |
| Phenol | 70-72 |
| Sulphate | 76.8 |
| Chloride | 192-223 |
| Nitrite | 0.2-0.34 |
| Ammonia | 116.8 |
| Total Kjeldah l Nitrogen (organic nitrogen) | 246.6 |

3. Polypropylene balls

-Density: 910kg/m3

 4. Glass-wares

#  2.2 Procedure for growing algae:

1. Petridishes containing growth medium with 1–1.5% agar medium was prepared. And the agar medium should be ½ to 2/3 the depth of dish.
2. 1-2 drops of algae sample from the slant was placed near the periphery of the agar. The wire loop was sterilized using burner.
3. The petriplate was covered and sealed with parafilm. Then it was incubated in a low light at constant temperature.
4. The colonies were selected which are free of other organism for further isolation process.
5. The sample was removed using sterile wire loop and placed in a drop of sterile culture media on a glass slide.
6. Then the species was checked microscopically for whether the species is uni-algal or not.
7. The streaking procedure was repeated with a single colony and again allowed to colonies to develop.
8. The second streaking is done to reduce the possibility of bacterial contamination and species containing more than one algal species.
9. Then the selected colonies were transferred to the liquid nutrient medium and allowed to grow in an incubator shaker for temperature maintenance of around 20–25C̊. The alternative for maintenance of temperature is by keeping it in an AC room and for stirring keep it in a magnetic stirrer at low rpm.
10. After 5 -10 days growth is observed in a beaker of liquid medium and the growth substantially increases but pH and nutrient level in a medium must be checked and maintained.

#  2.3 Experimentation

 Inverse Fluidisation Unit

2.3.1Designof IFBR:

* + - 1. The unit consist of long perplexed glass tube–
* Height=1.240m

* Diameter= 10cm
* Wall thickness= 3mm

1.Centrifugal pump

* Power0.5HP
* Head= 14ft
	+ - 1. Calibrated Rota meter
* For water= 0-100LPM
* For Gas=0-200m3/hr
	+ - 1. Manometer
* Number = 4
* Length = 1m
	+ - 1. Circular pith distributed plate
			2. Conical heads (at the top and bottom)
* Apex angle=60̊
* Inner diameter=10cm
* Height=30 cm

 Figure– 5: Outline sketch of the IFBR unit

# Experimental Procedure for operation in IFBR:

* + - 1. The column was loaded with some amount of polypropylene balls.
			2. Fill the liquid storage tank with10 litre of water and mix waste water around 200ml to it. Then add 250 ml of algae sample to the tank and mix it very well.
			3. Pump the water from liquid storage tank to the vertical unit with the liquid flow rate of 10LPM and till certain height is reached in the bed measured from the scales tick to it on the outer surface.
			4. The pressure drop across the test section is measured with the help of manometer connected across the bed.
			5. The flow rate of the gas is slowly increased to bring the bed into the state of mixing , as mixing provides better growth of microbes due to continuous interaction with each other .
			6. The bed continuously kept under light of intensity which is required for the growth of algae.
			7. The mixture of wastewater, algae and nutrients was kept in fluidization for hours and sample was taken for COD and BOD analysis after 6hr, 24hr, 32hr, 48hr, 96hr, 120hr.
			8. Two wastewater samples were taken untreated and sterilized wastewater for the treatment.
	1. COD Analysis

# Materials required**:**

1. Potassium dichromate
2. Concentrated sulphuric acid
3. Ferro in indicator
4. Ferrous Ammonium Sulphate(FAS)
5. Mercuric sulphate
6. Distilled water
7. Glassware’s ( conical flask, beaker, heater, stirrer, measuring cylinder )

# 2.4.1Procedure:

* + 1. Potassium Dichromate (K2Cr2O7) solution

 -12.259 g of K2Cr2O7 was dissolved in 1000ml distilled water.

* + 1. FAS solution
			- * 98 g of FAS is dissolved in distilled water and then 20ml of Conc.Sulphuric acid was added and the solution is diluted to 1000ml

Molarity of FAS can be calculated as

 **Molarity FAS= Volume of K2Cr2O7 inml ∗0.25**

 **Volume of FAS used in ml**

* + 1. Now 20ml of the sample was taken in a 500ml flask

* + 1. Then10ml of K2Cr2O7 was added to it.
		2. 30 ml of conc.H2SO4 was added slowly and cautiously.
		3. 0.4gm of Mercuric sulphate was then added then the sample was heated at 120̊C for Around 10min.
		4. Then the sample was cooled to room temperature
		5. The solution was diluted to two times its volume with distilled water.
		6. Fill the burette with FAS solution and add 2–3 drops of Ferroin indicator to the diluted solution and titrate it against FAS solution.
		7. The end point of the titration is determined by sharp colour change from blue green to reddish brown which persisted for 1 min.
		8. Similarly the waste water sterilized and untreated were also titrated to check the COD before process

# 2..4.2 Sample Calculation

Molarity of FAS=0.1M

* For Waste water before sterilization

COD=(A-B)\*M\*8\*1000/Volume of the sample used (2)

(Source-APHA standard method for examination of water and waste water,20thedition, Method 5220C)

Where;

A=Volume of FAS for blank = 13.4

B=Volume of FAS for sample=3.0 M = molarity of FAS solution = 0.1 M Volume of the sample used = 20ml COD = 416 mg/ml

* For waste water after sterilization COD measured = 380 mg/ml

### BOD Analysis

# **Materials Required:**

* + - 1. Potassium hydrogen phosphate (KH2PO4)
			2. Di-potassium hydrogen phosphate (K2HPO4)
			3. Di-sodium hydrogen phosphate (Na2HPO4.7H2O)
			4. Ammonium chloride (NH4Cl)
			5. Magnesium sulphate hepta-hydrate (MgSO4.7H2O)
			6. Calcium chloride (CaCl2)
			7. Ferric Chloride (FeCl3.6H2O)
			8. Sodium sulphite (Na2SO3)
			9. Distilled water
			10. Glassware’s(test tubes, beaker, conical flask)


# **Procedure for preparation of solution:**

* + - 1. Phosphate buffer solution

8.5g of KH2PO4 ,21.75g of K2HPO4, 33.4 g of Na2HPO4.7H2O and 1.7g of NH4Cl

Was dissolved in 500 ml distilled water and diluted it to 1000ml. Make sure that the pH is adjusted to 7.2.

* + - 1. Magnesium Sulphate solution

22.5g MgSO4.7H2O was dissolved in distilled water and dilute it to1litre.

* + - 1. Calcium Chloride solution

27.5g of CaCl2 was dissolved in 1000 ml distilled water.

* + - 1. Ferric Chloride solution

0.25g of Ferric chloride solution was dissolved in 1000ml of distilled water.

* + - 1. Sodium sulphite solution

1.575g of sodium sulphite is dissolved in 1000ml of distilled water.

**NOTE: All the solutions must be prepared daily because they are not stable.**

# **Procedure:**

* + - 1. The 20 ml sample was kept in a 1 litre flask
			2. Then 1 ml magnesium sulphite solution, 1 ml calcium chloride solution, and 1 ml ferric chloride solution were added to 1 litre of distilled water.
			3. If the solutions are acidic or alkaline, then they must be neutralised before use, and this can be done by adding sodium thio-sulphate solution to destroy residual chlorine.
			4. The sample must be diluted as follows:
* Strong water =0.1,0.5,or 1%
* Settled domestic sewage= 1,2.5,or5%
* Treated effluents =5, 12.5or25%
* River water= 25to100%
	+ - 1. The sample was diluted with distilled water and mixed nicely.
			2. The diluted sample was the taken in two BOD bottles.
			3. The DO of diluted water and diluted waste water was taken immediately.
			4. The other two bottles were kept at 20degreeC for 3–5 days and the sample was incubated.
			5. After 3 days the DO of sample was taken.
			6. The procedure for DO analysis follows this.

# **Procedure for Dissolved oxygen analysis:**

* + - 1. The two BOD bottles were taken and 2ml ofalkali–iodize-azidewasaddedtoitbelow the liquid level.
			2. The bottle must completely air tight so that no air should enter into it.The sample was mixed properly. The presence of oxygen is indicated by the appearance brownish – orange cloud of precipitate or floc. This floc can be disappeared byturning the bottle upside down and allowing it to settle.
			3. Then2mlofsulphuricacidwasaddedtoitviaapipetteholdingitjustabovethesurface of the sample. Againthebottleisinvertedaftercarefullypluggingthestopperintoitto dissolve the floc. Then the sample is kept for 8 hr.
			4. Filled the burette with sodium thiosulfate solution.
			5. 2 ml starch solution was added so a blue colour forms.
			6. The sample was titrated slowly till the end point .And end point is determined when the blue colour disappears.
			7. The concentration of dissolved oxygen can be determined by the number of milli litres titrant used. A search ml of sodiumthio-sulphate added equals1mg/l dissolved oxygen.

# **Sample Calculation forBOD**

Initial DO of diluted sample, Do= 8.2

 DO at the end of 3 day D3=6.08

Blank correction, BC = 0.2

Volume of sample diluted, Vd=500ml

 Volume of sample taken ,Vs=20ml

BOD =(Do–D3-BC)\*Vd/Vs (3)

(Source: APHA standard methodforexaminationofwaterandwastewater,20th edition, Method 5220C)

=(8.2–608-0.2) \*500/20

= 48mg/ml

## RESULTAND DISCUSSION

* 1. For non-sterilized waste water after treatment with Chlorella Scenedesmus at Vb/Vr=0.5 Initial Value of COD before treatment is 416 mg/ml, pH = 6.7

After treatment the pH is 8.5 at the end of 192 hr

Table–3:Variation of COD with time for non-sterilized wastewater

|  |  |  |
| --- | --- | --- |
| S.No: | Number of Hours ofoperation | COD in mg/ml |
| 1 | 0 | 416 |
| 2 | 6 | 400 |
| 3 | 24 | 346 |
| 4 | 48 | 277 |
| 5 | 72 | 236 |
| 6 | 168 | 149.76 |
| 7 | 192 | 146.6 |

450

400

350

300

250

CODINMG/ML

200

150

100

50

0

0 50 100 150 200

TIMEINHOUR

Figure–7:Variation of COD with time for non-sterilized wastewater

Table-4:BOD analysis of non-sterilized wastewater

|  |  |  |
| --- | --- | --- |
| S.No.: | Number of hours of operation | BOD in mg/ml |
| 1 | 0 | 48 |
| 2 | 72 | 28 |
| 3 | 144 | 15 |

60

50

40

30

BODIN MG/ML

20

10

0

0 20 40 60 80 100 120 140 160

TIMEINHOURS

Figure-8:BOD vs time for non-sterilized wastewater

* 1. For wastewater(sterilized) after treatment with Chlorella sp.at Vb/Vr=0.5 Initial value of pH before treatment is 8.3 and after treatment

After treatment pH was 8.9 for the total time duration of 192 hours.

Table-5: Variation of COD with time for sterilized wastewater

|  |  |  |
| --- | --- | --- |
| S.NO | Number of hours ofoperation | COD in mg/ml |
| 1 | 0 | 380 |
| 2 | 6 | 368 |
| 3 | 24 | 310 |
| 4 | 48 | 246.4 |
| 5 | 72 | 195.4 |
| 6 | 168 | 126 |
| 7 | 192 | 114 |

400

350

300

250

CODINMG/ML

200

150

100

50

0

0 50 100 150 200 250

TIMEINHOURS

Figure–9: COD vs time for sterilized wastewater

Table-6: BOD vs time for sterilized wastewater

|  |  |  |
| --- | --- | --- |
| S.NO. | Number of hours of operation | BOD in mg/ml |
| 1 | 0 | 44 |
| 2 | 72 | 23 |
| 3 | 144 | 13 |

50

45

40

35

30

BODIN MG/ML

25

20

15

10

5

0

0 20 40 60 80 100 120 140 160

TIMEINHOUR

Figure-10 BOD vs time for sterilized wastewater

### Calculation of percentage reduction in COD and BOD after treating it with algae

% Reduction in COD for non-sterilized wastewater

= [(initial value of COD-final value of COD)/ initial value of COD] \* 100

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| % | Reduction | in | COD | for | sterilized | === | (416–146.67/416) \*10065%[(initial value of COD-final value | of |
| wastewater |  |  |  |  | COD)/initial value of COD]\* 100=((380–114)/380)\*100=70% |  |
| %Reduction wastewater | in | BOD | for | non-sterilized | =[(initial value of BOD-final value BOD)/ initial value of BOD] \* 100=((48–15)/48)\*100=68.75% | of |
| %Reduction wastewater | in | BOD | for | non-sterilized | =[(initial value of BOD-final value BOD)/ initial value of BOD] \* 100=((44-13)/44)\*100=70.5% | of |

Table–7:%Reduction in COD and BOD of Chlorella Scenedesmus with other species of Algae

|  |  |  |  |
| --- | --- | --- | --- |
| S.No. | Name of the Algae species | % Reduction of BOD | % Reduction of COD |
| 1 | No stoc Muscorum(Ref. :3) | --- | 20–57.1 |
| 2 | Chlorella.Pyrenoidosa(Ref. :5) | 92 | 86 |
| 3 | Euglena(Ref-12 ) | 96 | 80 |
| 4 | Chlorellasp(Ref–4) | ------ | 50.8 |
| 5 | Chlorella Scenedesmus (Species used for this project) | 68.75 | 70 |

### Algae Identification

Algae Name: Chlorella Scenedesmus

Figure-11 :Chlorella (Microscopic view of chlorella viewed in the range of 10 microm)

Figure–12:Scenedesmus (Microscopic view of chlorella in the range of 10 microm)

## CONCLUSION

COD and BOD analysis of wastewater is one of the basic step which is needed to set up any waste water treatment plant and to control losses to the sewer system. Many ways of chemical treating wastewater has been proved to be very expensive and produces harmful end product which is very necessary to be avoided in today’s century. This study which includes treatment of steel plant waste water with the most abundantly available resource i.e., algae shows a new path way to achieve two major goals of any waste water treatment plant first being the economy and second being the efficiency in reduction of harmful components present in industrial , domestic or municipal wastewater. Treatment in inverse fluidization unit is very economical as it very cheap to procure, easy to handle and require low power to operate and in addition to this using Algae in it for degradation of hazardous components sorts out problems such as cost of oxygen supply needed for conversion of organic compounds and moreover algae can further be used as a source of biofuel and diet supplementary as some of the species are very effective for it. Continuous mixing with the help of solid particles in fluidization unit helps Algae to grow on its surface. Thus, this type of study is necessary before setting up any wastewater treatment plant.

**FUTURE WORK:**

* + 1. Measurement of the COD and BOD content of the outlet stream from the inverse fluidization unit by varying parameter such as :
			- Gas flow rate
			- Concentration of effluent in water
			- Different strains (Spirulinaandmix)
			- Ratio of volume of bed and volume of the reactor
		2. Comparing this method of using algae with other biological methods.
		3. Comparing it with other conventional method for wastewater treatment.
		4. Analysing the bio-hydrogen evolution from biomass under anaerobic condition

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15. **RESULTS AND DISCUSSION**

In this Section results and discussion of the study is written. They may also be broken into subsets with short, revealing captions. This section should be typed in character size 10pt Times New Roman.

**Table 1.**Sample Comparison

|  |  |  |
| --- | --- | --- |
| SN. | Sample | Quantity (Liter) |
| 1 | Fluid A | 22 |
| 2 | Fluid-B | 15 |
| 3 | Fluid-C | 12 |
| 4 | Fluid-D | 10 |
| 5 | Fluid-E | 27 |
| 6 | Fluid-F | 32 |



**Figure2:**10 liter capacity vessel (Font size-10)

Unless or otherwise specified specific gravity values reported shall be based on waterat 270C. So the specific gravity at 270C= K Sp. gravity at Tx0C. The specific gravity of the soil particles lie with in the range of 2.65 to 2.85. Soilscontaining organic matter and porousparticles may have specific gravity values below2.0. Soils having heavy substances may have values above 3.0.

1. **CONCLUSION**

All the main points of the research work are written in this section. Ensure that abstract and conclusion should not same. Graph and tables should not use in conclusion.

**ACKNOWLEDGEMENTS (optional)**

The authors can acknowledge professor, friend or family member who help in research work in this section.

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