

9. BIOCHEMICAL OXYGEN DEMAND (BOD)

TITLE

To determine the amount of Biochemical Oxygen Demand (BOD) in domestic waste water.

THEORY

Biochemical Oxygen Demand (BOD)

The amount of oxygen required by the bacteria while stabilizing decomposable organic matter under aerobic conditions. Decomposable means that organic matter can serve as food for the bacteria and energy is derived from its oxidation.

- Biochemical oxygen demand is a measure of the quantity of oxygen used by microorganisms (e.g., aerobic bacteria) in the oxidation of organic matter.
- Natural sources of organic matter include plant decay and leaf fall. However, plant growth and decay may be unnaturally accelerated when nutrients and sunlight are overly abundant due to human influence.
- Urban runoff carries pet wastes from streets and sidewalks; nutrients from lawn fertilizers; leaves, grass clippings, and paper from residential areas, which increase oxygen demand.
- Oxygen consumed in the decomposition process robs other aquatic organisms of the oxygen they need to live. Organisms that are more tolerant of lower dissolved oxygen levels may replace a diversity of more sensitive organisms.

BOD Level (in ppm)	Water Quality
1 - 2	Very Good-not much organic waste present
3 - 5	Moderately clean
6 - 9	Somewhat polluted
10+	Very polluted

Importance of BOD Test in Environmental Engineering

The BOD test is used to determine the relative oxygen requirements of wastewaters, effluents, and polluted waters. The test measures the oxygen utilized during a specified incubation period for the biochemical degradation of organic material. It is also used to determine treatment plant efficiency.

Determination of BOD

Principle:

The method consists of filling with sample, to overflowing, an airtight bottle of the specified size and incubating it at the specified temperature for 5 days. Dissolved oxygen is measured initially and after incubation, and the BOD is computed from the difference between initial and final DO. Because the initial DO is determined shortly after the dilution is made, all oxygen uptake occurring after this measurement is included in the BOD measurement.

Sampling and Storage:

Sample for BOD analysis may degrade significantly during storage between collection and analysis, resulting in low BOD values. Minimize reduction of BOD by analyzing sample promptly or by cooling it to near-freezing temperature during storage. However, even at low temperature, keep holding time to a minimum. Warm chilled samples to $20 \pm 3^\circ\text{C}$ before analysis.

Apparatus:

- a. Incubation bottles: Use glass bottles having 60 mL or greater capacity (300mL bottles having ground-glass stopper and a flared mouth are preferred).
- b. Air incubator or water bath, thermo-statistically controlled at $20 \pm 1^\circ\text{C}$. Exclude all light to prevent possibility of photosynthetic production of DO.

Reagents:

Prepare reagents in advance but discard if there is any sign of precipitation or biological growth in the stock bottles.

- a. Phosphate buffer solution: Dissolve 8.5 g KH_2PO_4 , 21.75 g K_2HPO_4 , 33.4 g $\text{Na}_2\text{HPO}_4 \cdot 7\text{H}_2\text{O}$, and 1.7 g NH_4Cl in about 500 mL distilled water and dilute

to 1 Lit. The pH should be 7.2 without further adjustment. Alternatively, dissolve 42.5 g KH_2PO_4 or 54.3 g K_2HPO_4 in about 700 mL distilled water. Adjust pH to 7.2 with 30% NaOH and dilute to 1 Lit.

- b. Magnesium sulfate solution: Dissolve 22.5 g $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ in distilled water and dilute to 1 L.
- c. Calcium chloride solution: Dissolve 27.5 CaCl_2 in distilled water and dilute to 1 L.
- d. Ferric Chloride solution: Dissolve 0.25 g $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ in distilled water and dilute to 1 L.
- e. Acid and alkali solution, 1N, for neutralization of caustic or acidic waste samples. 1) Acid-Slowly and while stirring, add 28 mL cone. Sulfuric acid to distilled Water. Dilute to 1 L. 2) Alkali-Dissolve 40 g sodium hydroxide in distilled water. Dilute to 1 L.
- f. Sodium sulfate solution: Dissolve 1.575 g Na_2SO_3 in 1000 mL distilled water. This solution is not stable; prepare daily.
- g. Nitrification inhibitor: 2-chloro-6-(trichloromethyl) pyridine (if nitrification inhibition desired).
- h. Glucose-glutamic acid solution: Dry reagent-grade glucose and reagent-grade glutamic acid at 103°C for 1 h. Add 150 mg glucose and 150 mg glutamic acid to distilled water and dilute to 1 L. Prepare fresh immediately before use.
- i. Ammonium chloride solution: Dissolve 1.15 g NH_4Cl in about 500 mL distilled water, adjust pH to 7.2 with NaOH solution and dilute to 1 L. Solution contains 0.3 mg N mL^{-1}
- j. Dilution water: Use demineralized, distilled, tap, or natural water for making sample dilutions.

Procedure: (with out seeding)

- 1) First of all it is important to know the amount of samples to be used for test. For this purpose the source of sample is to be recorded which will indicate the approximate value of BOD_5 for the sample.
 - (i) Domestic sewage $\text{BOD}_5 = 100\text{-}500\text{mg/L}$
 - (ii) Effluent from treatment plant = $20\text{-}80\text{mg/L}$
 - (iii) River water = $2\text{-}4\text{mg/L}$
- 2) Take 9 BOD bottles note their numbers and arrange them in 3 groups.

- 3) Fill each bottle half with dilution media ensuring that no air gets mixed with the media while fill in as in DO test.
- 4) Add 2ml sample in each of the three bottles marked as first group; 5 ml in each bottle pf 2nd group and 10ml in each bottle of the 3rd group.
- 5) Fill the bottle completely with dilution media and place the stopper such that no air bubbles are trapped.
- 6) Now take one bottle from each set and estimate its DO. This will be DO initial or DO 0days.
- 7) For comparison prepare two more bottles with blank dilutions media (with out sewage sample) and find the DO from one bottle.
- 8) Place the rest of the six bottles with sewage samples and one bottle for blank in the incubator at 20⁰c.
- 9) After 5 days find out DO in all bottles.
- 10) That value of oxygen depletion should be considered correct which gives an oxygen depletion of at least 2 mg/L. and which have at least 0.5 mg/L DO after 5 days of incubation.
- 11) Calculate BOD₅ at 20⁰c. for the sample using following relation ship.

$$\text{BOD}(\text{mg/L}) = \frac{\text{DO depletion (mg/L)} * 300}{\text{Volume of sample in bottle (ml)}}$$

Volume of sample in bottle (ml)

Observations:

At zero days.

Bottle#	Sample added (ml)	Volume of sample (ml)	Volume of Na₂S₂O₃	DO (mg/L)	Mean DO (mg/L)
	Blank				

After 5 days.

Bottle#	Sample added (ml)	Volume of sample (ml)	Volume of Na ₂ S ₂ O ₃	DO (mg/L)	Mean DO (mg/L)
	Blank				

DO Depletion:

Bottle#	Sample added (ml)	DO at Zero days (mg/L)	DO at 5 days (mg/L)	DO Depleted (mg/L)	BOD ₅ (mg/L)
	Blank				

Mean BOD₅ =

Comments:

Questions:

1). Define BOD.

2) Why natural water and tap water can not be used for preparing dilution media in the BOD test.

3) What is the role of nitrifying bacteria in BOD test?

4) What is the history of BOD test?

5) What is seeding?