Biochemical Oxygen Demand
5-Day (BOD)
(The same as DO)

Reagents:

1- Phosphate buffer solution:
- Dissolve 8.5 gram KH$_2$PO$_4$
  21.75 gram K$_2$HPO$_4$
  33.4 gram Na$_2$HPO$_4$.7H$_2$O
- 1.7 gram NH$_4$Cl. In about 500 ml dH$_2$O,
- Dissolve well dilute up to 1L, pH should be 7.2

2- Magnesium sulfate:
- Dissolve 22.5 gram MgSO$_4$.7H$_2$O, up to 1L d H$_2$O

1- Calcium chloride
- Dissolve 27.5 gram CaCl$_2$, up to 1L d H$_2$O or 36.42 gram CaCl$_2$.2H$_2$O →1L

2- Ferric chloride
- Dissolve 0.25 gram FeCl$_3$.6H$_2$O, up to 1L d H$_2$O

5-Nitrification Inhibitor:
Dissolve 10 mg/L

Procedure
1- Prepare 1 liter dilution (or aerated) water at 20 °C overnight
2- Add 10 mg nitrification inhibitor/L dil. H$_2$O
1 ml MgSO$_4$, 1 ml CaCl$_2$, 1 ml FeCl$_3$, 1 ml phosphate buffer
3- Take 25%-100% of your water sample of the BOD bottles (0.5-2.5) ml of waste water sample
4- place in 3 BOD bottles
5- Complete the 3 bottles by the dilution water prepared on (1)
6- Incubate (2) bottles under 20.5 °C for 5 days
7- Use the third bottle for DO$_0$ determination as usual

DO measurement
a- Add 1 ml MnSO$_4$
b- 1 ml Alkali-Azide.Iodide solution
c- Add 1 ml conc. H$_2$SO$_4$,
d- take appropriate sample for DO determination
d- Add few drops of starch solution, continue
*Titrate against 0.025M Na$_2$S$_2$O$_3$
  mls titrant = DO mg/L
6- After the 5 days, test the DO$_5$ value of the other (2) bottles by the same step
7-Calculation

**DO > DO** \(_{5 \text{ days}}\)

\[ \text{BOD} = \frac{\text{DO}_0 - \text{DO}_5}{\text{P}} \]

As \[ \text{P} = \frac{\text{mls} \_ \text{sample}}{\text{Vol.BOD bottle}} \]