Dissolved Oxygen and Biochemical Oxygen Demand Analyses

Prepared By
Michigan Department of Environmental Quality
Operator Training and Certification Unit

Note: These Procedures are available in the OTCU Laboratory manual which is available on the OTCU website.

Dissolved Oxygen

D.O.

Amount of “FREE” Oxygen

O_2

In the Water

Required By :

FISH

MICROORGANISMS

(Bacteria)

RESPIRATION

Organic Matter Converted to:

Carbon Dioxide

and

Water

$$CH_2O + O_2 \rightarrow CO_2 + H_2O$$

RESPIRATION

$$CH_2O + O_2 \rightarrow CO_2 + H_2O$$

Reason for Treatment

Basis of
Secondary Treatment

Basis of BOD Analysis

Nitrification

BIOLOGICAL Oxidation of Nitrogen

From	to	to
AMMONIA	NITRITE	NITRATE
(NH_3)	(NO_2)	(NO_3)

$$NH_3 + O_2 \longrightarrow NO_2 + O_2 \longrightarrow NO_3$$

NITRIFICATION

$$NH_3 + O_2 \rightarrow NO_2 + O_2 \rightarrow NO_3$$

May be Required:

Oxygen Demand Reduction

Ammonia Removal (toxic)

Reason for D.O. Analysis

Used in BOD Analysis

Treatment Processes

Treatment Plant Effluents

Receiving Stream

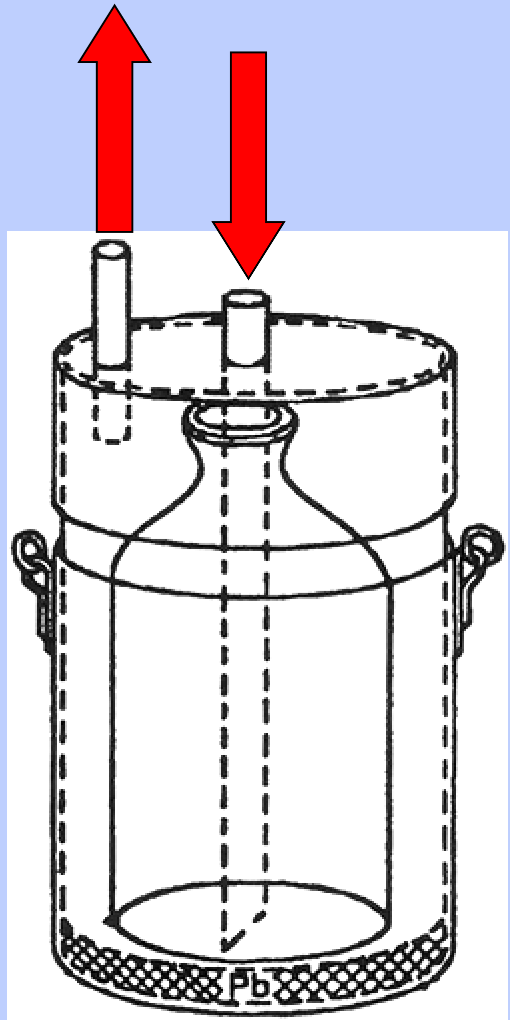
Maximum D.O. Concentrations in Water (Saturation)

Temperature ^0C	Max. Concentration mg/L
0	14.6
4	13.1
8	11.9
12	10.8
16	10.0
20	9.2
24	8.5
28	7.9
30	7.6

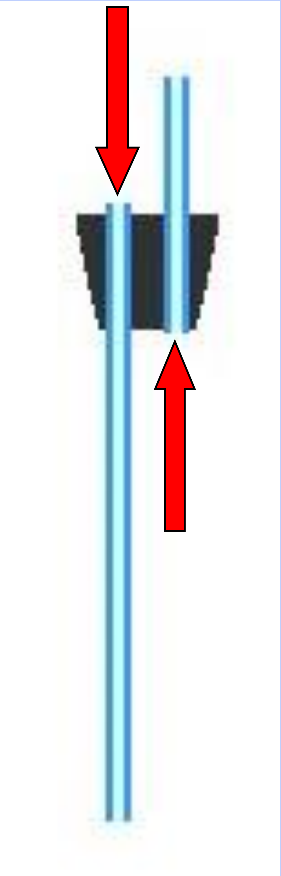
SAMPLE COLLECTION

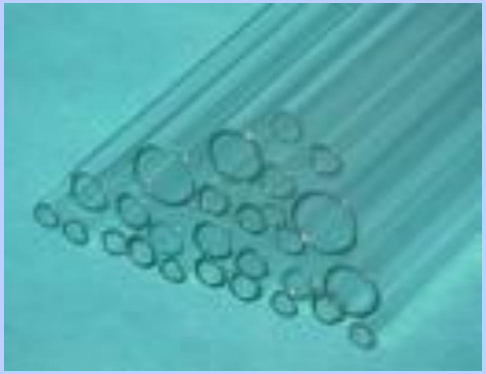
Sample	Atmosphere
0 to 10 mg/L	21 %
	210,000 mg/L

SAMPLE COLLECTION

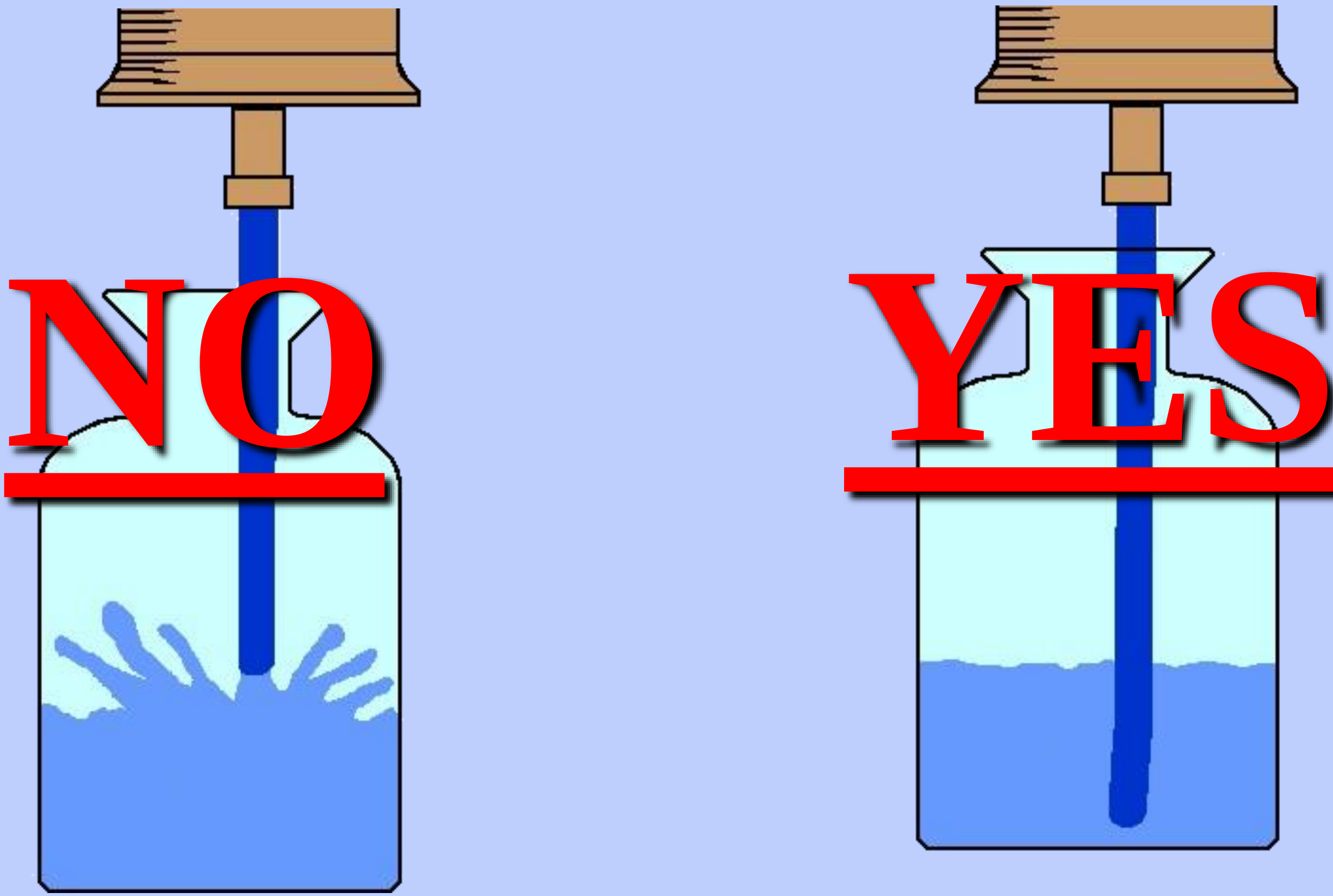


SAMPLE COLLECTION





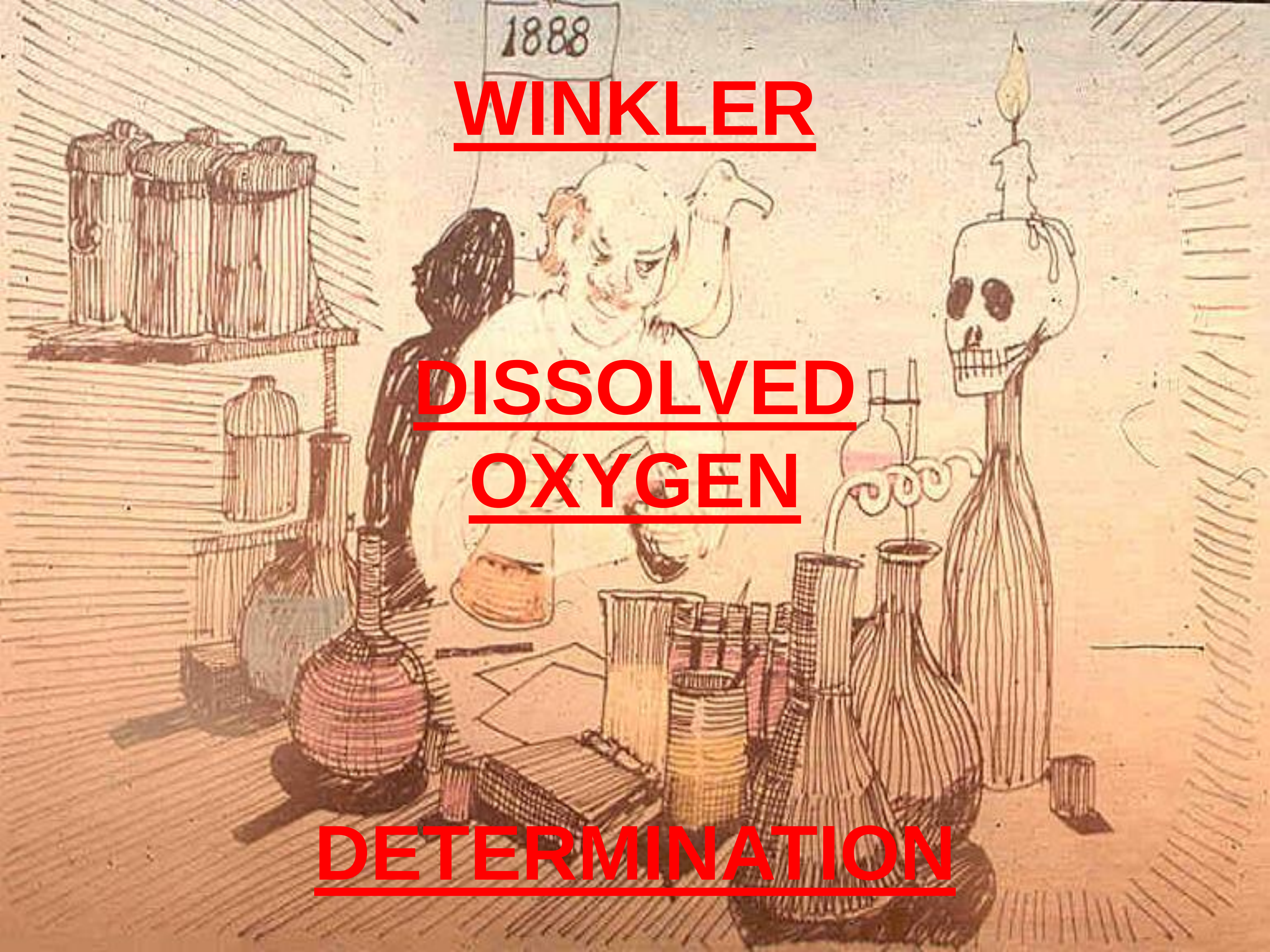
Do Not AERATE Sample



Do Not Splash Sample in Air
or
Let Air Circulate in Sample

D.O. Procedure

WINKLER

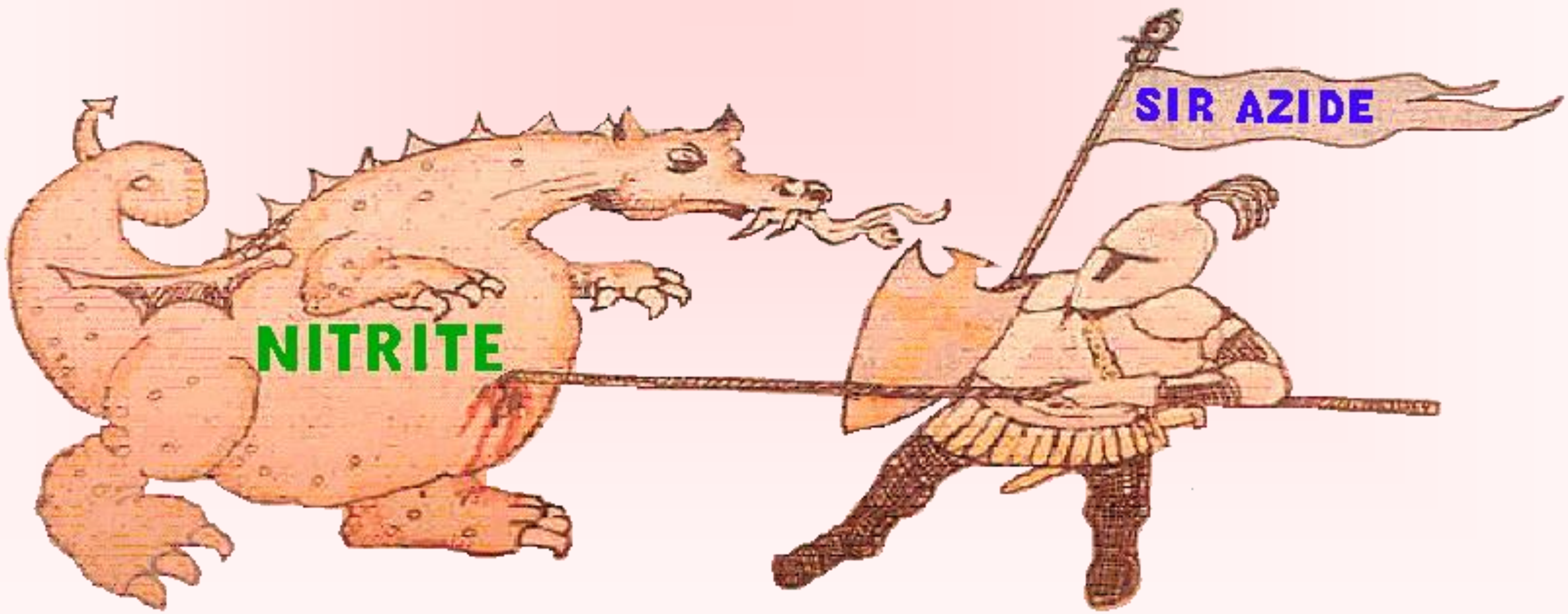


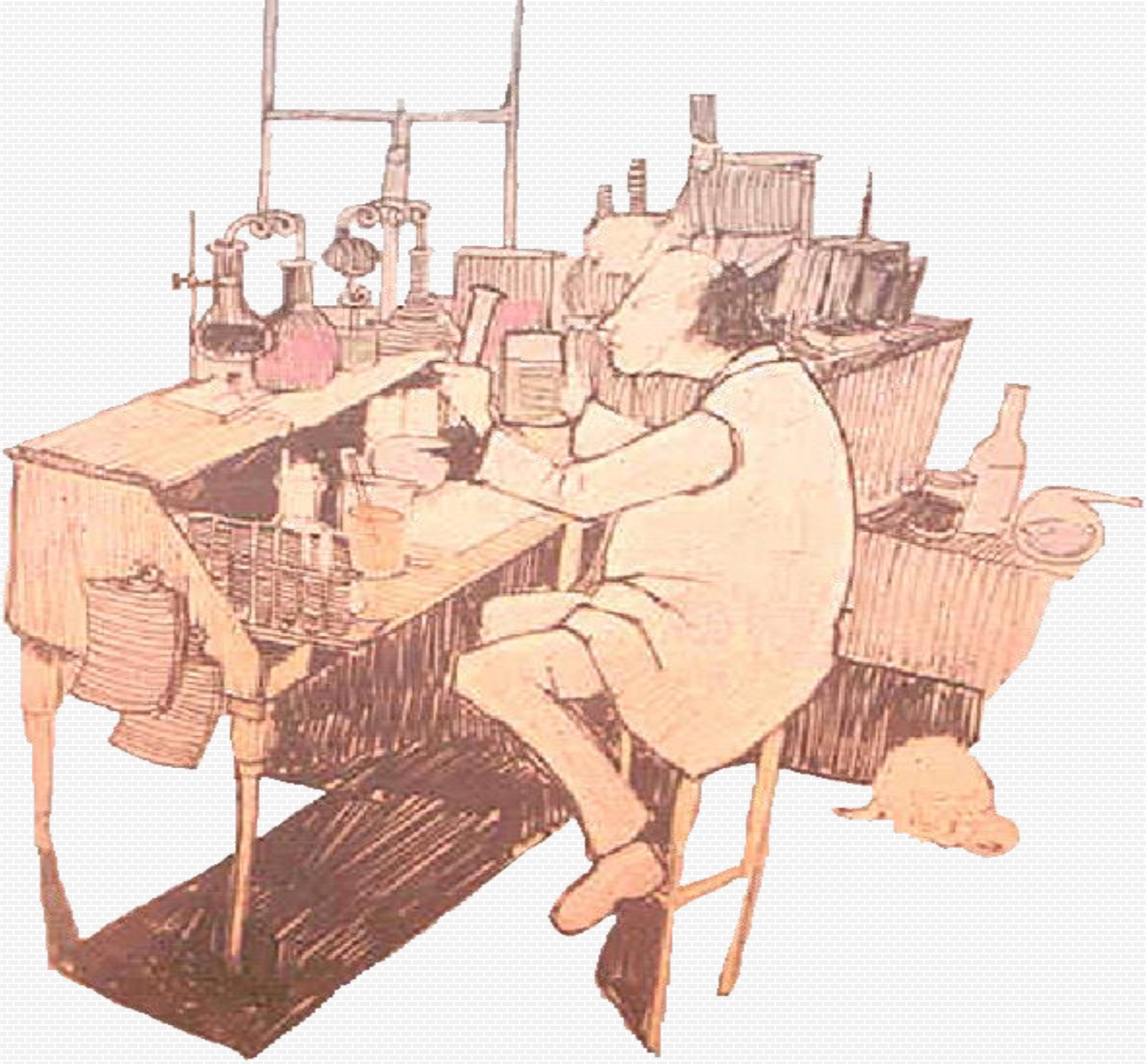
DISSOLVED OXYGEN

DETERMINATION

WINKLER Dissolved Oxygen PROCEDURE
with the
ALSTERBERG
AZIDE
MODIFICATION

AZIDE Destroys NITRITE





Winkler Method

Iodometric Method

$$MnSO_4 + 2\,KOH \longrightarrow Mn(OH)_2 + K_2SO_4$$

$$2\,Mn(OH)_2 + O_2 \longrightarrow 2\,MnO(OH)_2$$

$$MnO(OH)_2 + 2\,H_2SO_4 \longrightarrow Mn(SO_4)_2 + 3\,H_2O$$

$$Mn(SO_4)_2 + 2\,KI \longrightarrow MnSO_4 + K_2SO_4 + I_2$$

1. Takes Mixing

2. Takes Time

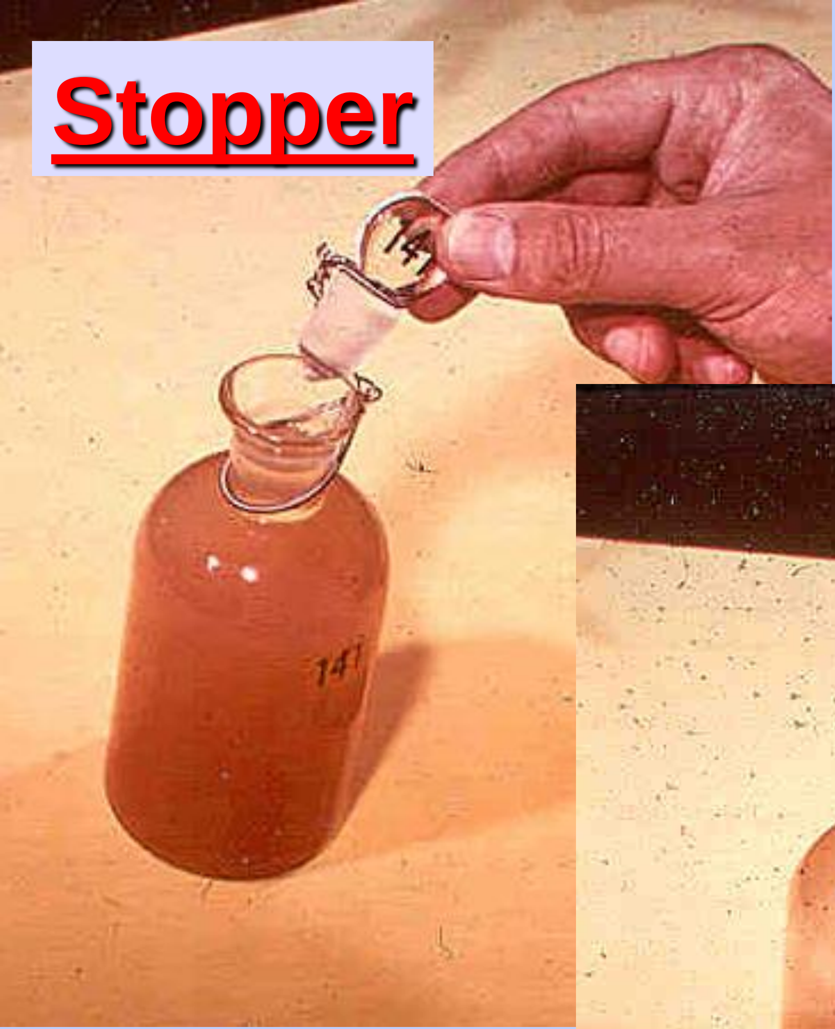
3. Free IODINE released in relation to D.O. in Sample

Winkler Procedure

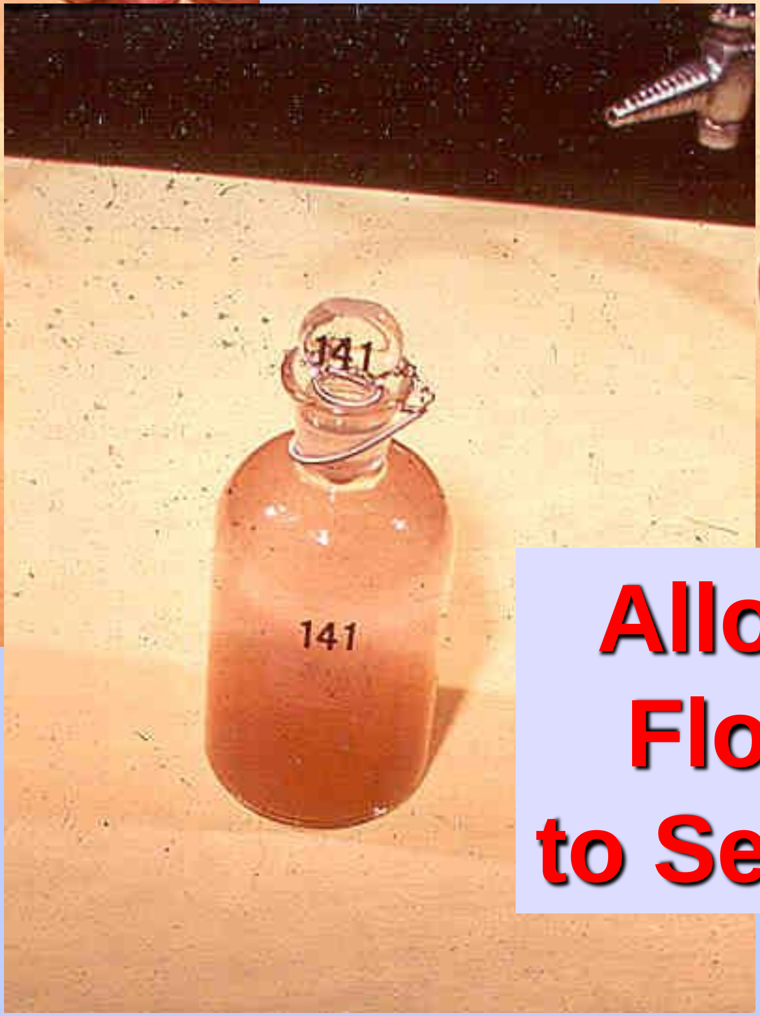
1 mL Manganous Sulfate

1 mL Alkaline Azide Solution

Stopper



Mix Well



Allow Floc to Settle

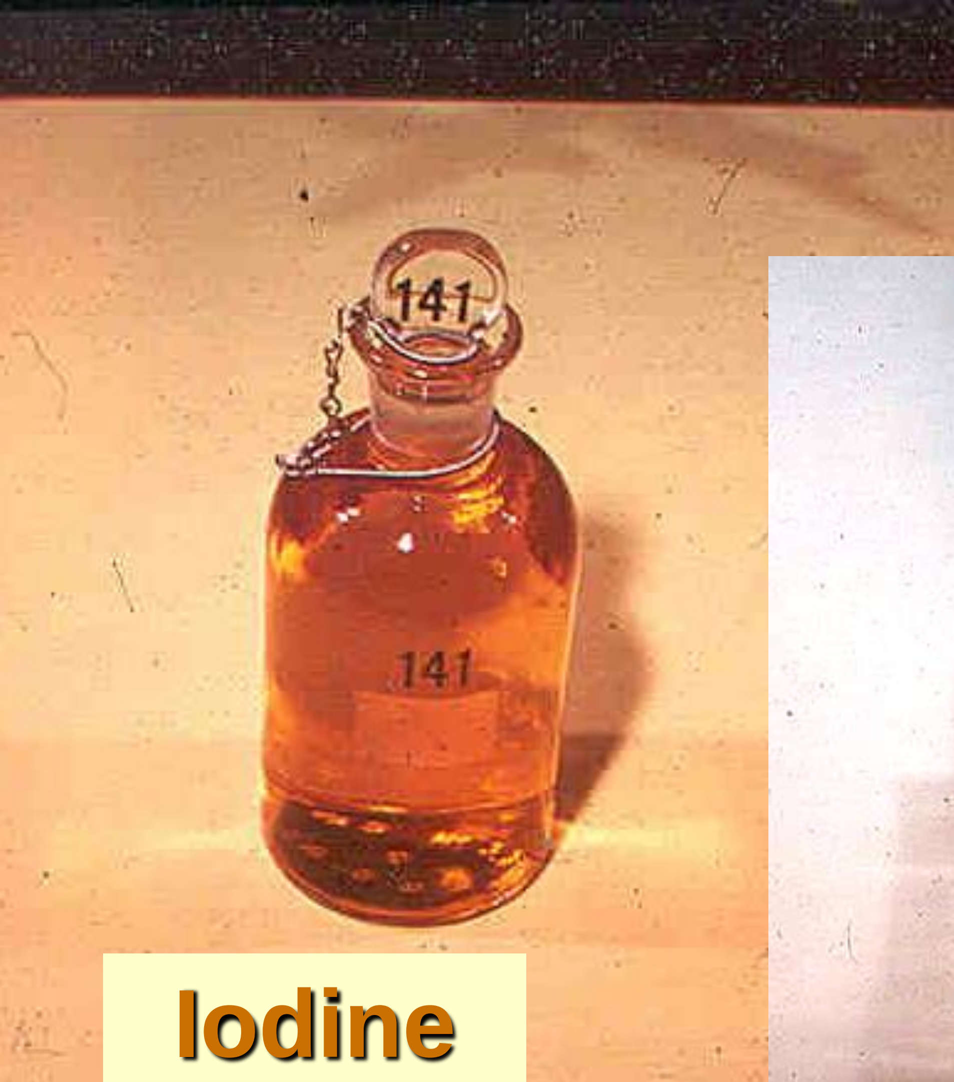

Repeat Mixing
Settle Again
141
141
141
Contact and Time

White - No Dissolved Oxygen

1 mL Sulfuric Acid

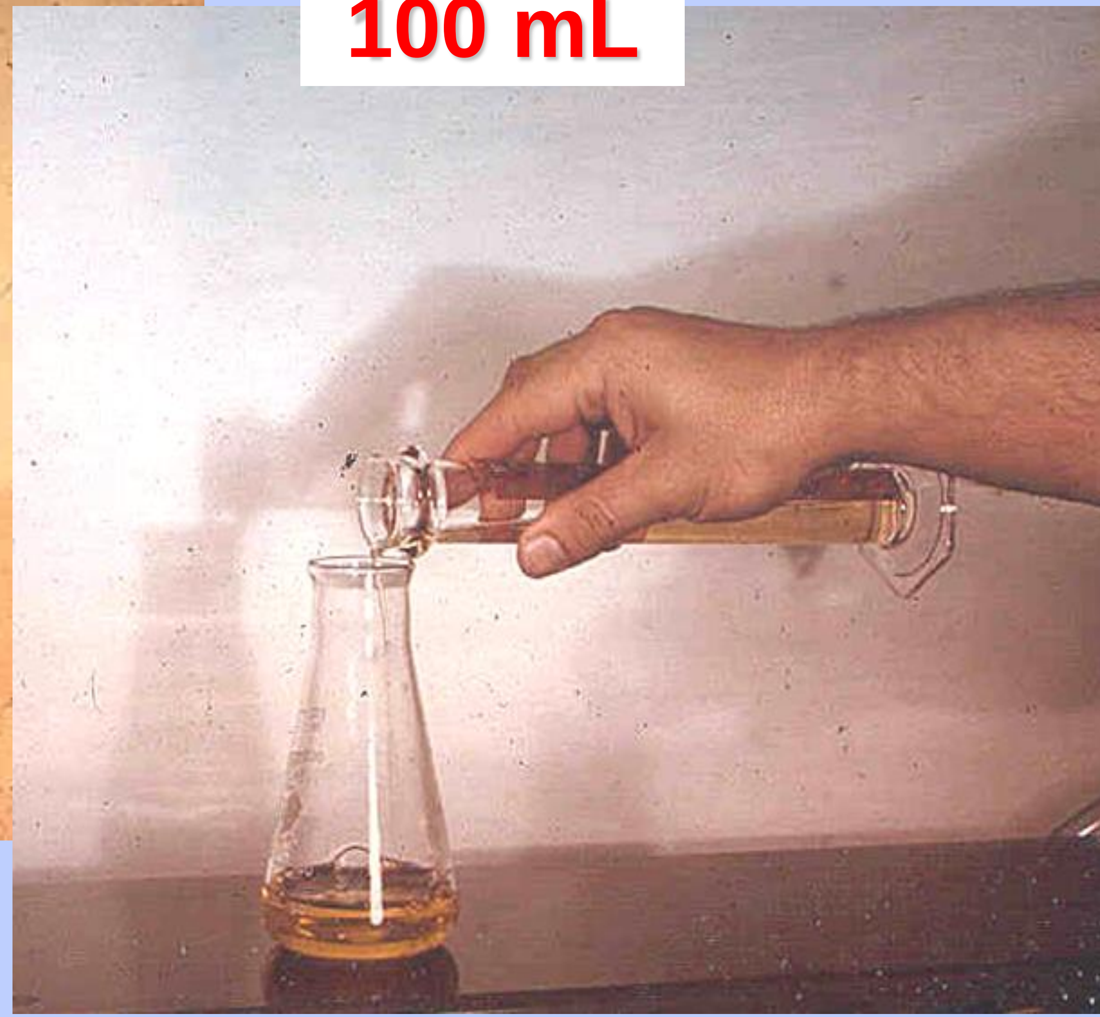
Sulfamic Acid

Mix



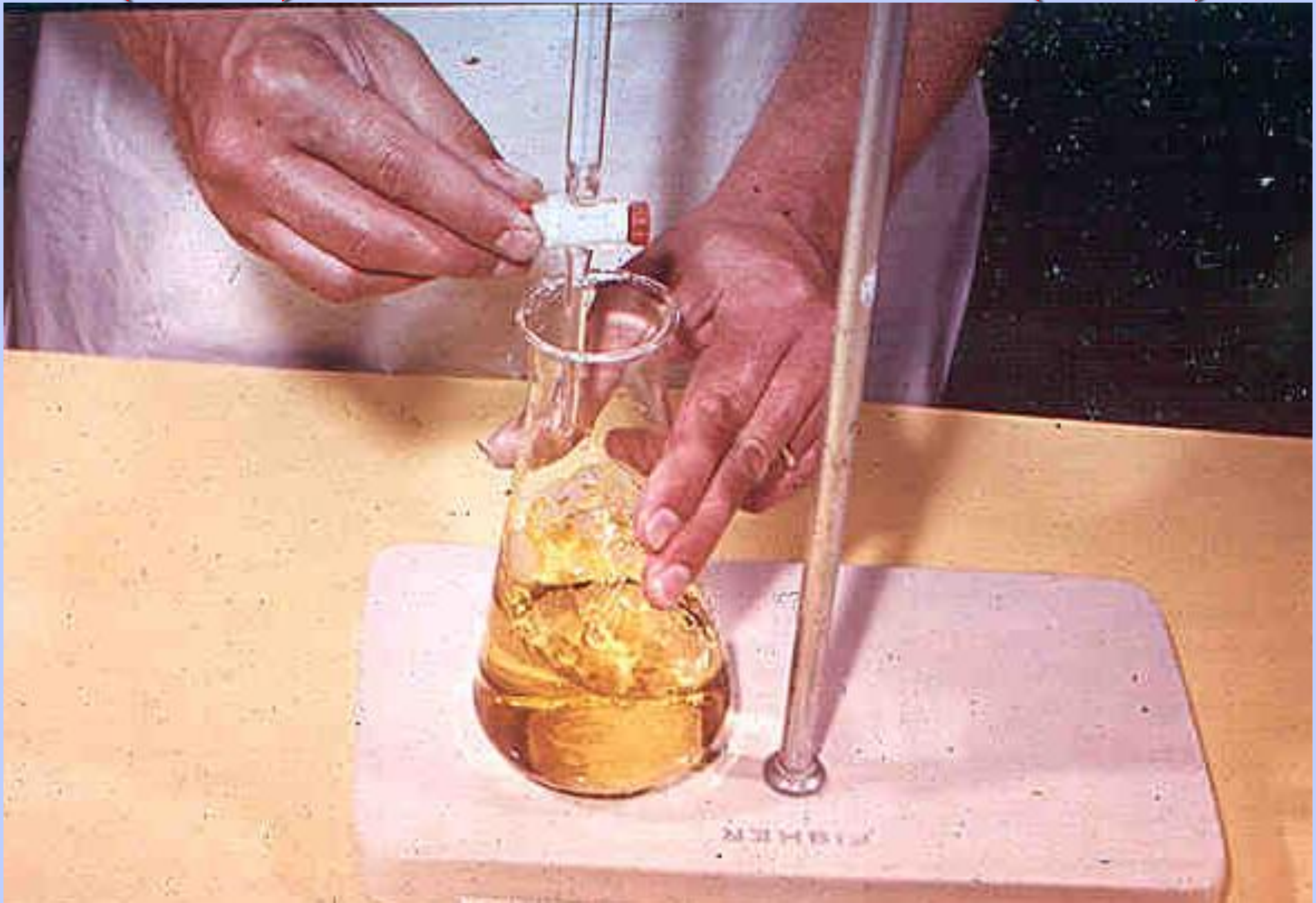
Iodine Solution

Measure 100 mL



Titration

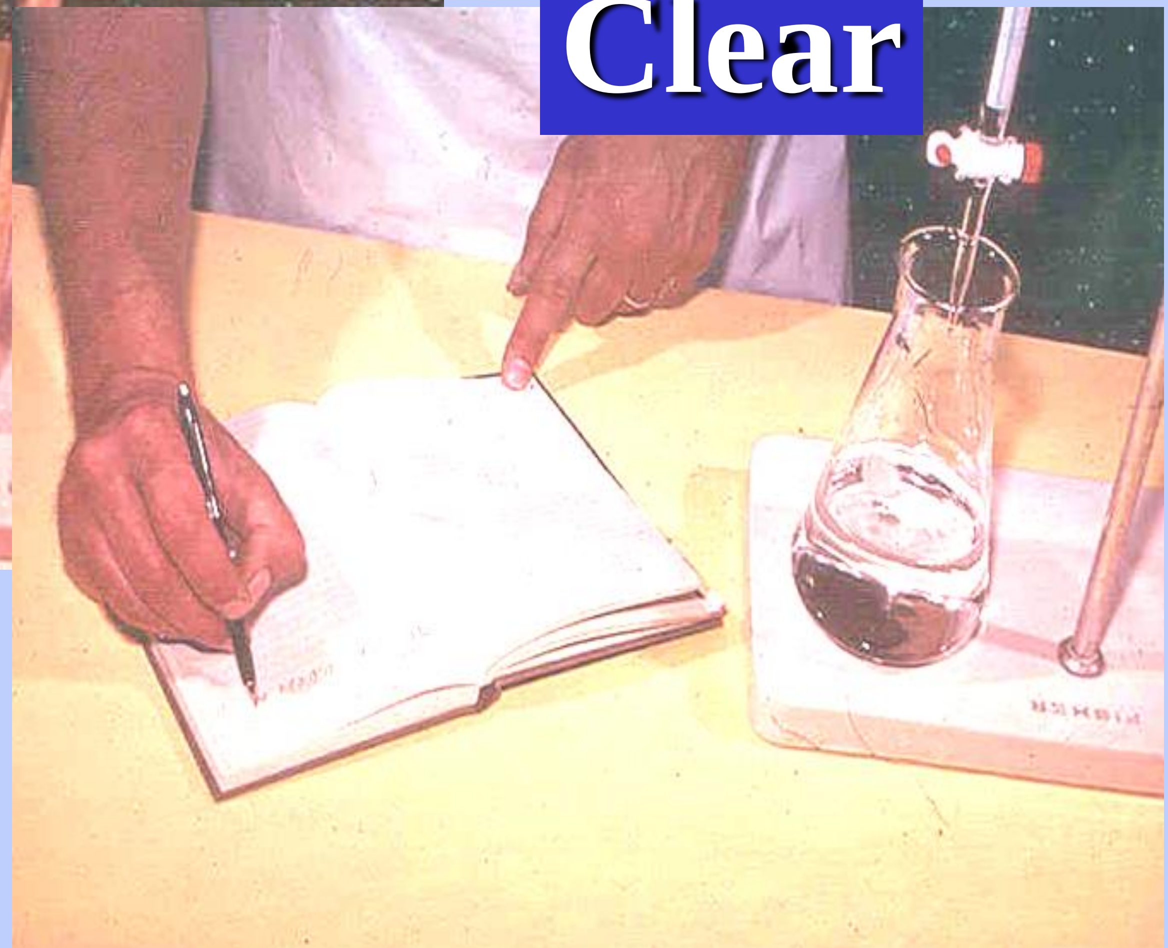
Sodium Thiosulfate (Thio) OR **Phenylarsene Oxide (PAO)**



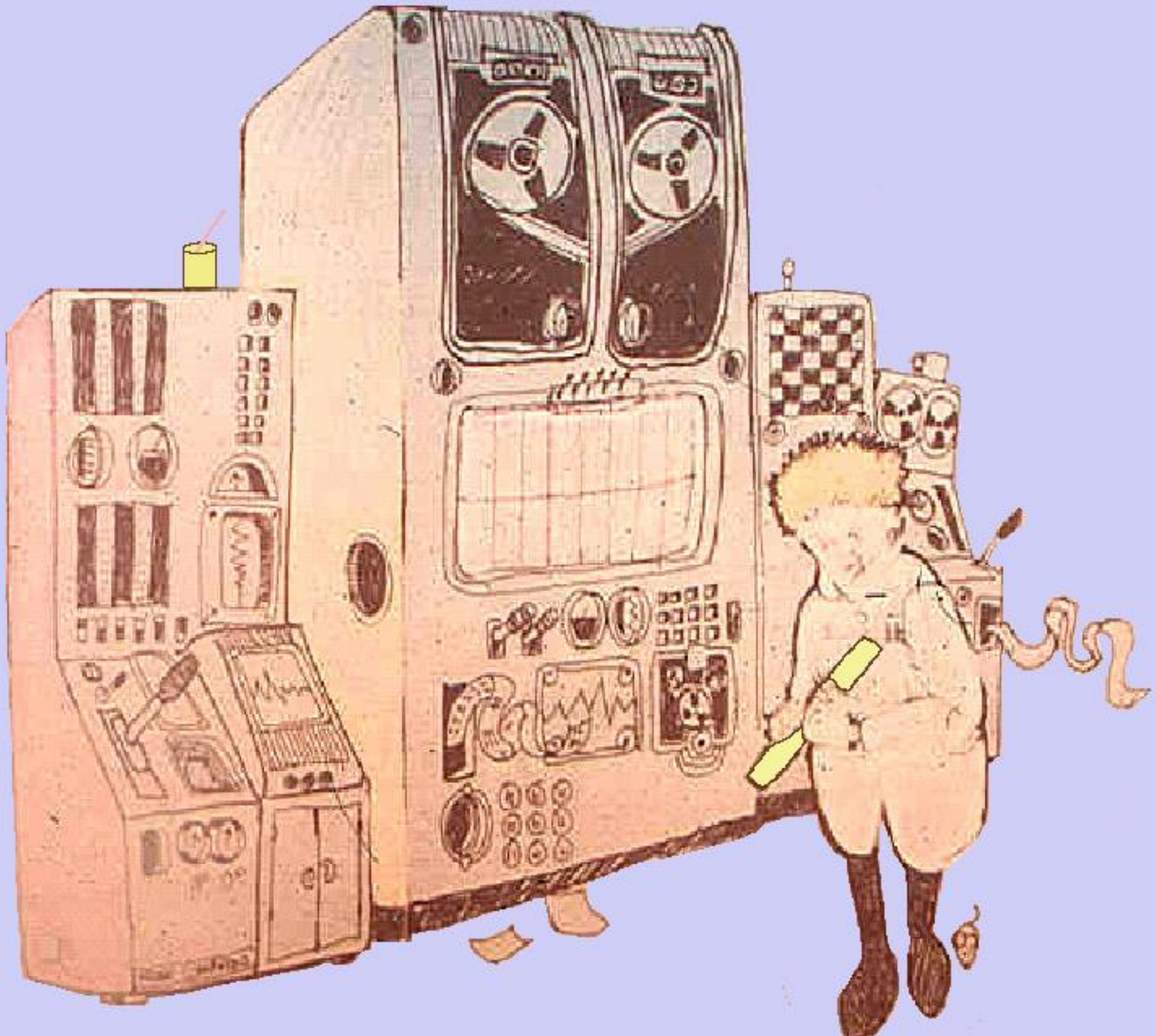
End Point Indicator

Starch

Blue



Clear



Mg/L D.O. =

$$\frac{\text{mL of Titrant} \times \text{Normality of Titrant} \times 8 \times 1000}{\text{mL of Sample}}$$

$$\frac{0.0125\ \text{N} \times 8 \times 1000}{100} = 1$$

Mg/L D.O. =

mL of Titrant X 1

When 100 ml of Sample is Titrated with 0.0125 Normal Titrant

Each mL used Equals 1 mg/L of D.O.

If

4.6 mL

is Used

Then the D.O. in the Sample is

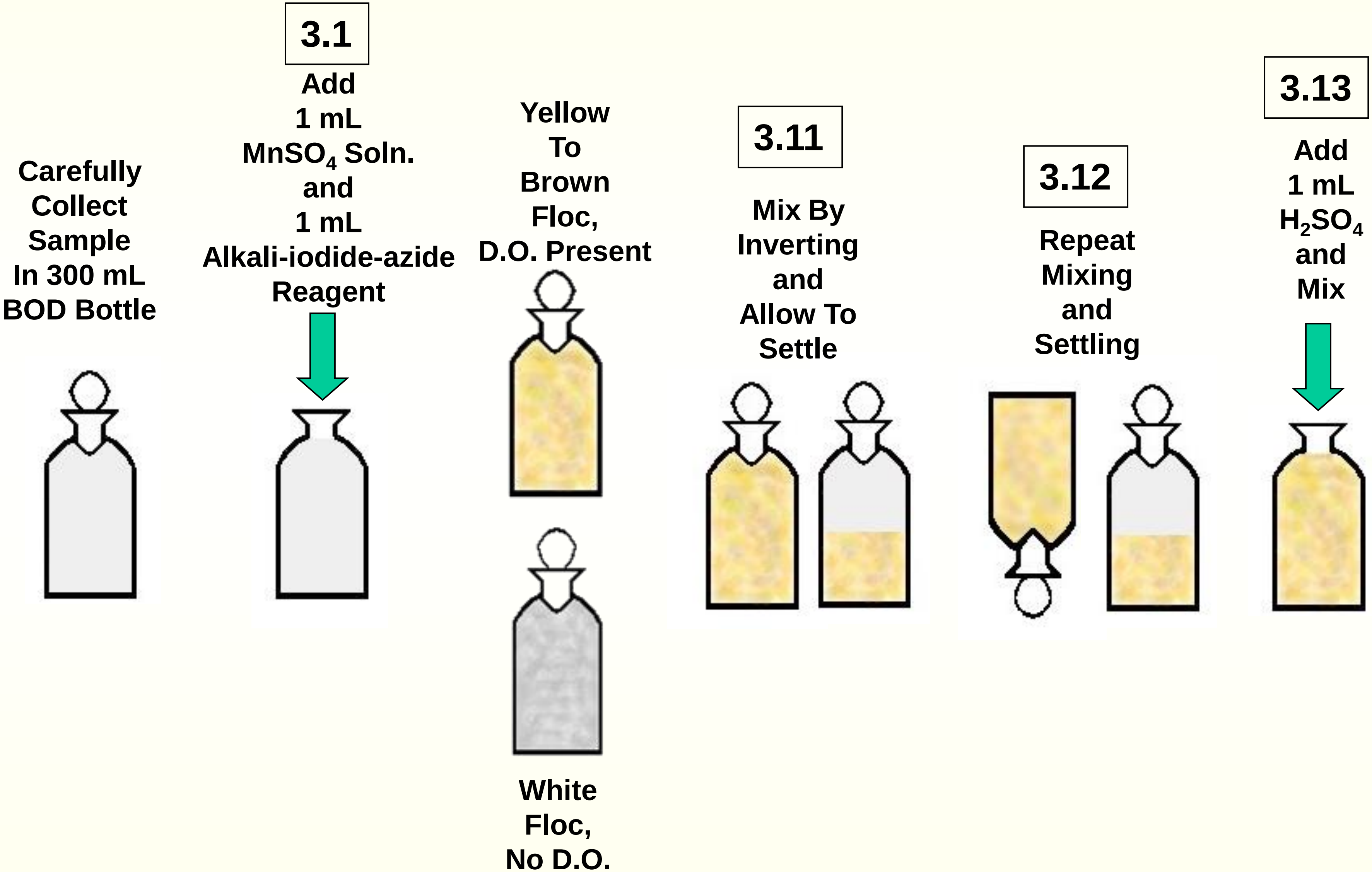
4.6 mg/L

1 mL of Titrant = 1 mg/L D.O.

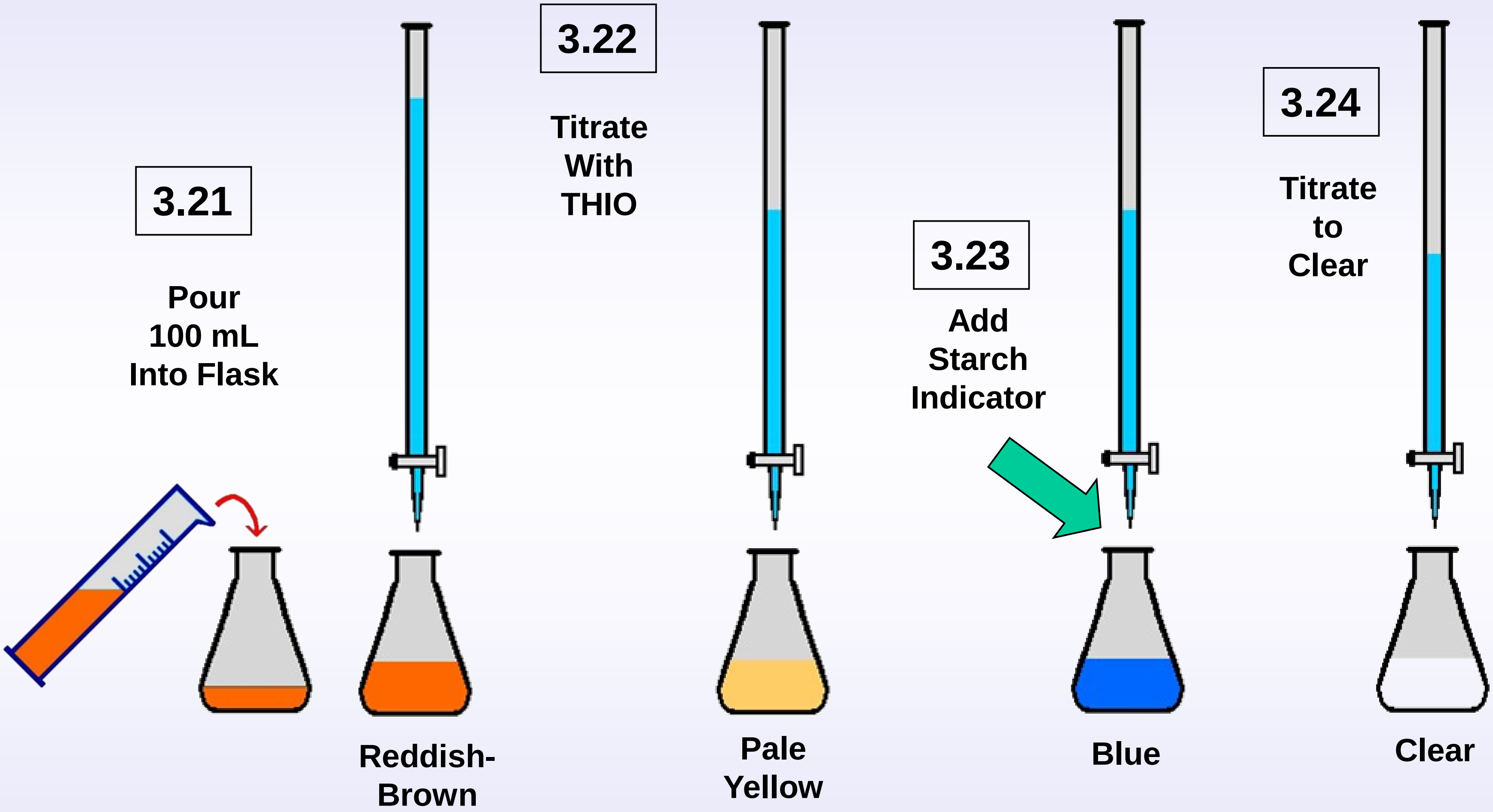
With Any of the Following Combinations of Sample Volumes and Normalities

Sample Volume	Normality
100 mL	0.0125 N
200 mL	0.0250 N
300 mL	0.0375 N

Outline Of Winkler Dissolved Oxygen Procedure



Titration of Iodine Solution



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1. Takes Mixing

2. Takes Time

3. Free IODINE released in relation to D.O. in Sample

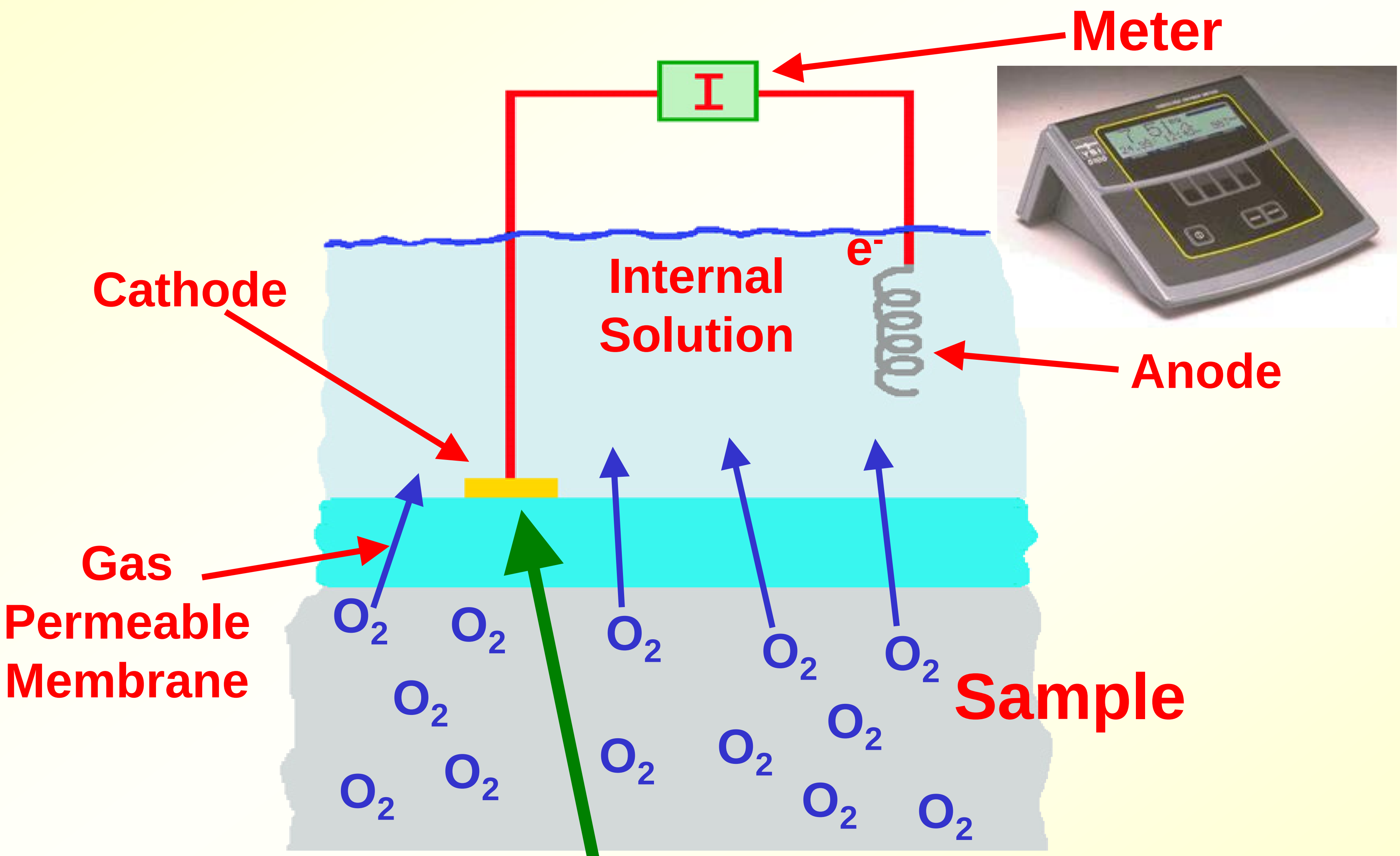
D.O. Procedure

Electrode Methods

MEMBRANE ELECTRODE METHOD

The membrane electrode is composed of two solid metal electrodes in contact with supporting electrolyte separated from the test solution by a gas permeable membrane. Oxygen dissolved in the sample diffuses through the membrane on the DO probe and is chemically reduced (accepts electrons), producing an electrical current between the anode and cathode in the probe. The amount of current is proportional to the concentration of DO. Following proper calibration, the meter relates this current to the concentration of DO.

CLARK ELECTRODE



$$O_2 + 4H^+ + 4e^- \longrightarrow 2H_2O$$

MEMBRANE ELECTRODE METHOD

Calibration

Comparison with Winkler Titration

1. Fill two BOD bottles completely full of BOD dilution water, being very careful not to introduce air into either bottle.

2. Analyze one bottle for D.O. using the Winkler titration.

3. Insert the electrode into the second bottle, turn on the stirring mechanism, and wait for the reading to stabilize.

4. Calibrate the meter to the D.O. value obtained in the titration.

5. The meter is now ready for sample analysis

MEMBRANE ELECTRODE METHOD

"AIR" Calibration

This procedure varies considerably among the various instrument models available. Therefore, the procedure must be obtained from the instrument manual, but the following points should be noted.

1. Where possible with the specific equipment being used, compensation should be made during calibration for <u>both</u> ambient temperature and local atmospheric pressure. This pressure should be determined using a reliable onsite barometer. The oxygen solubility table following this procedure may be used.

2. Carefully blot any water droplets from the membrane using a soft tissue.

3. During calibration, be sure the membrane is exposed to fresh air. Laying the electrode on the bench for calibration is usually adequate.

4. Complete the calibration as soon as possible before the electrode membrane begins to dry.

5. The temperature registered on the meter should be checked against a trusted thermometer often.

6. Daily calibration of the D.O. meter is required. Calibration should also be verified after every five or six sample measurements.

7. Assure sufficient sample flow across membrane surface during analysis to overcome erratic response.

	Onsite Barometric Pressure														
	Atm:	0.970	0.975	0.980	0.985	0.990	0.995	1.000	1.005	1.010	1.015	1.020	1.030	1.040	1.050
	mm Hg:	737	741	745	749	752	756	760	764	768	771	775	783	790	798
	Inch Hg:	29.02	29.17	29.32	29.47	29.62	29.77	29.92	30.07	30.22	30.37	30.52	30.82	31.12	31.42
Temperature° Celsius	15.00	9.78	9.83	9.88	9.93	9.98	10.03	10.08	10.13	10.19	10.24	10.29	10.39	10.49	10.60
	15.50	9.67	9.72	9.77	9.82	9.88	9.93	9.98	10.03	10.08	10.13	10.18	10.28	10.38	10.48
	16.00	9.57	9.62	9.67	9.72	9.77	9.82	9.87	9.92	9.97	10.02	10.07	10.17	10.27	10.37
	16.50	9.47	9.52	9.57	9.62	9.67	9.72	9.77	9.82	9.87	9.92	9.97	10.07	10.16	10.26
	17.00	9.37	9.42	9.47	9.52	9.57	9.62	9.66	9.71	9.76	9.81	9.86	9.96	10.06	10.16
	17.50	9.27	9.32	9.37	9.42	9.47	9.52	9.57	9.61	9.66	9.71	9.76	9.86	9.96	10.05
	18.00	9.18	9.23	9.27	9.32	9.37	9.42	9.47	9.51	9.56	9.61	9.66	9.76	9.85	9.95
	18.50	9.08	9.13	9.18	9.23	9.28	9.32	9.37	9.42	9.47	9.51	9.56	9.66	9.75	9.85
	19.00	8.99	9.04	9.09	9.13	9.18	9.23	9.28	9.32	9.37	9.42	9.47	9.56	9.65	9.75
	19.50	8.90	8.95	9.00	9.04	9.09	9.14	9.18	9.23	9.28	9.32	9.37	9.47	9.56	9.65
	20.00	8.81	8.86	8.91	8.95	9.00	9.05	9.09	9.14	9.18	9.23	9.28	9.37	9.45	9.56
	20.50	8.72	8.77	8.82	8.86	8.91	8.96	9.00	9.05	9.10	9.14	9.19	9.28	9.37	9.46
	21.00	8.64	8.68	8.73	8.78	8.82	8.87	8.91	8.96	9.01	9.05	9.10	9.19	9.28	9.37
	21.50	8.56	8.60	8.65	8.69	8.74	8.78	8.83	8.87	8.92	8.96	9.01	9.10	9.19	9.28
	22.00	8.47	8.52	8.56	8.61	8.65	8.70	8.74	8.79	8.83	8.88	8.92	9.01	9.10	9.19
	22.50	8.39	8.44	8.48	8.53	8.57	8.62	8.66	8.70	8.75	8.79	8.84	8.93	9.02	9.10
	23.00	8.31	8.36	8.40	8.44	8.49	8.53	8.58	8.62	8.67	8.71	8.75	8.84	8.93	9.02
	23.50	8.23	8.28	8.32	8.37	8.41	8.45	8.50	8.54	8.58	8.63	8.67	8.76	8.85	8.93
	24.00	8.16	8.20	8.24	8.29	8.33	8.37	8.42	8.46	8.50	8.55	8.59	8.68	8.76	8.85
	24.50	8.08	8.12	8.17	8.21	8.25	8.30	8.34	8.38	8.43	8.47	8.51	8.60	8.68	8.77
	25.00	8.01	8.05	8.09	8.13	8.18	8.22	8.26	8.31	8.35	8.39	8.43	8.52	8.60	8.69
	25.50	7.93	7.97	8.02	8.06	8.10	8.15	8.19	8.23	8.27	8.31	8.36	8.44	8.53	8.61
	26.00	7.86	7.90	7.94	7.99	8.03	8.07	8.11	8.15	8.20	8.24	8.28	8.36	8.45	8.53
	26.50	7.79	7.83	7.87	7.91	7.96	8.00	8.04	8.08	8.12	8.16	8.21	8.29	8.37	8.46
	27.00	7.72	7.76	7.80	7.84	7.88	7.93	7.97	8.01	8.05	8.09	8.13	8.22	8.30	8.38
	27.50	7.65	7.69	7.73	7.77	7.81	7.86	7.90	7.94	7.98	8.02	8.06	8.14	8.22	8.31
	28.00	7.58	7.62	7.66	7.70	7.75	7.79	7.83	7.87	7.91	7.95	7.99	8.07	8.15	8.23
	28.50	7.52	7.56	7.60	7.64	7.68	7.72	7.76	7.80	7.84	7.88	7.92	8.00	8.08	8.16
	29.00	7.45	7.49	7.53	7.57	7.61	7.65	7.69	7.73	7.77	7.81	7.85	7.93	8.01	8.09
	29.50	7.38	7.42	7.46	7.50	7.54	7.58	7.62	7.66	7.70	7.74	7.78	7.86	7.94	8.02
	30.00	7.32	7.36	7.40	7.44	7.48	7.52	7.56	7.60	7.64	7.68	7.72	7.79	7.87	7.95

NOTE: The first three lines are different units for the same pressure measurement.

CLARK ELECTRODE

Calibration

NOT REQUIRED

NO

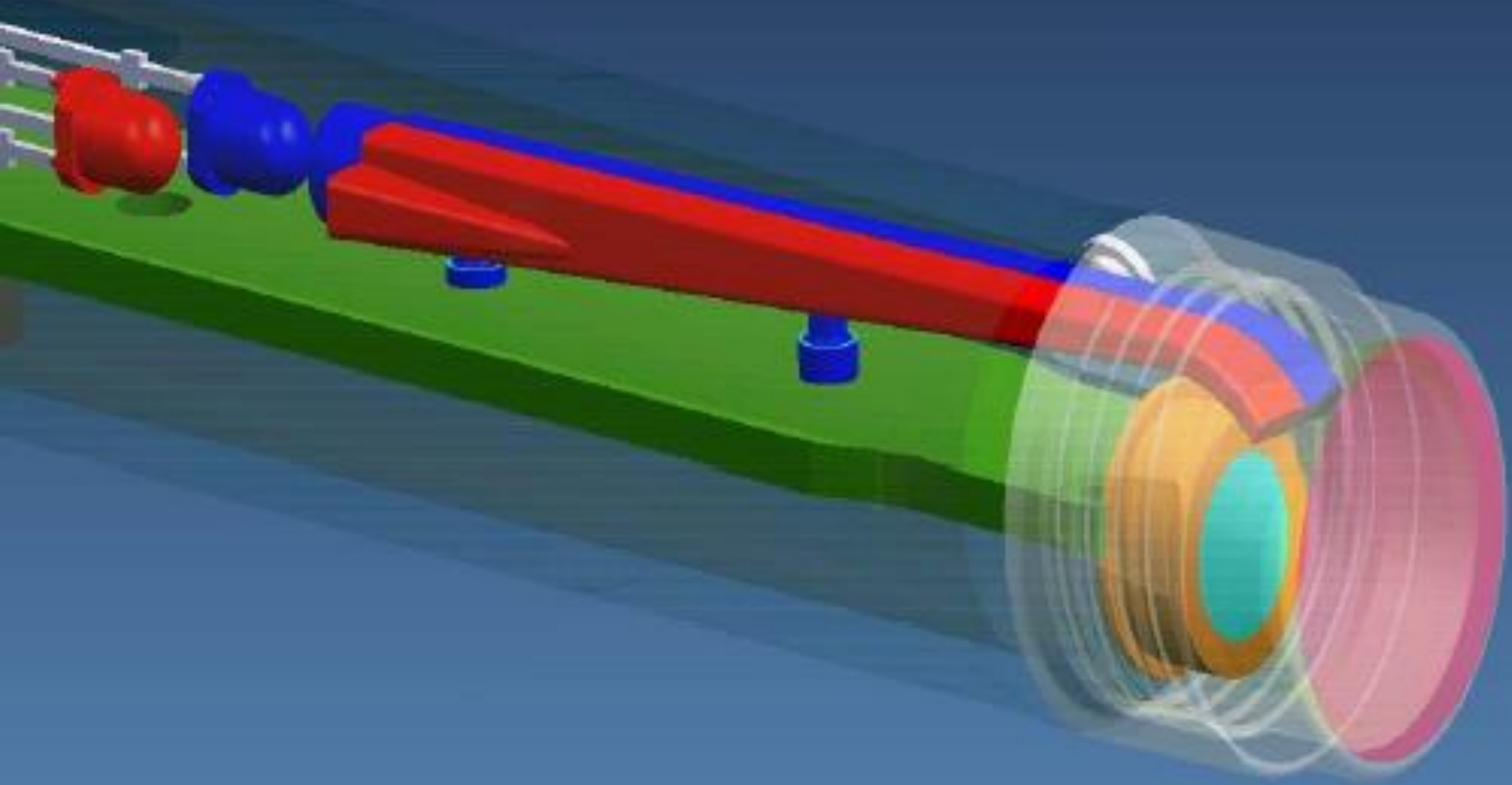
Temperature

Atmospheric Pressure

(Use a Reliable Barometer)

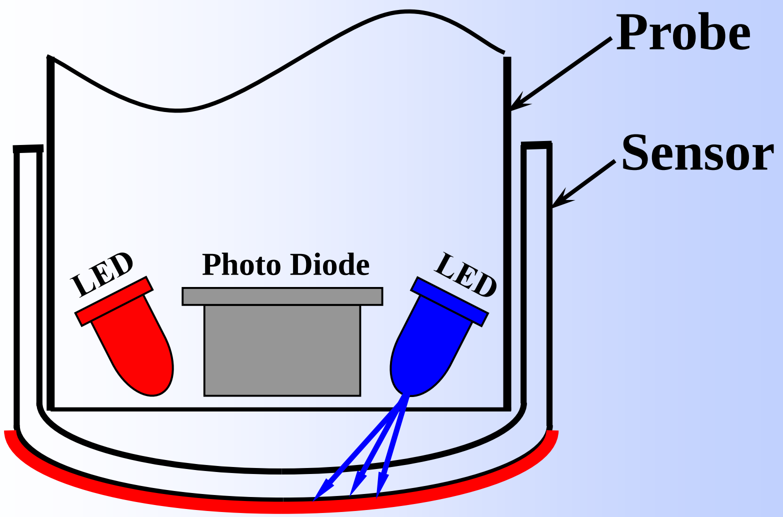
Luminescence D.O. Probe

Luminescence D.O. Probe



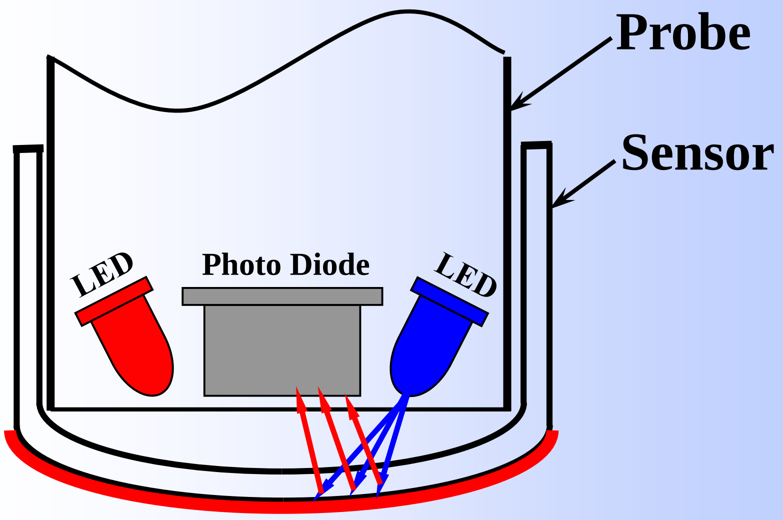
How Does LDO Work?

- A sensor is coated with a luminescent material.
- Blue light from an LED strikes the luminescent chemical on the sensor.
- The luminescent chemical instantly becomes excited.



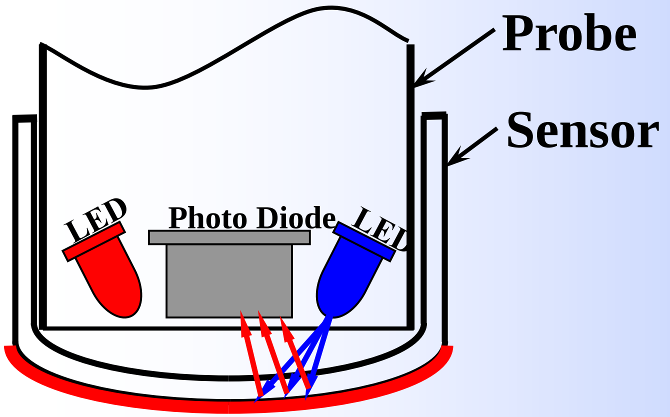
How Does LDO Work?

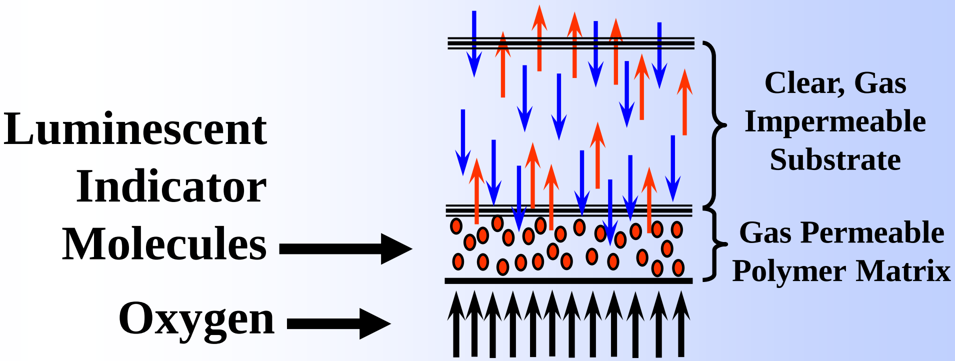
- As the excited chemical relaxes, it releases red light.
- The red light is detected by a photo diode.
- The time it takes for the chemical to return to a relaxed state is measured



How Does LDO Work?

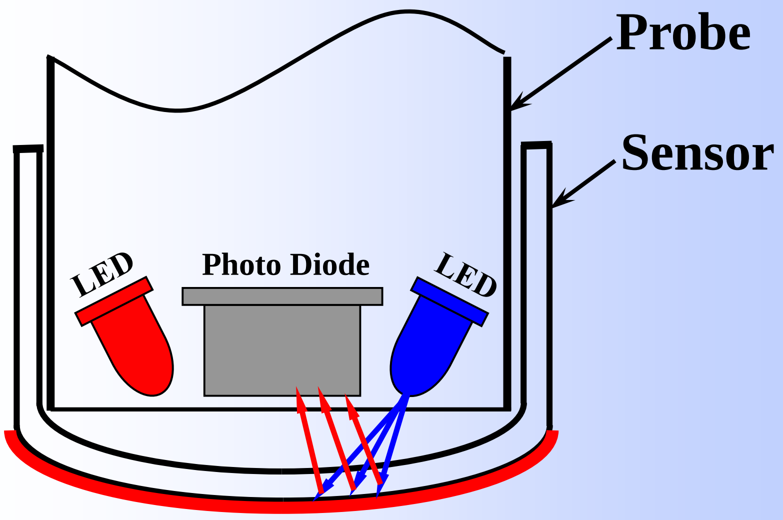
- When oxygen contacts the luminescent chemical, the intensity of the red light decreases
- The amount of time it takes for the material to relax is reduced





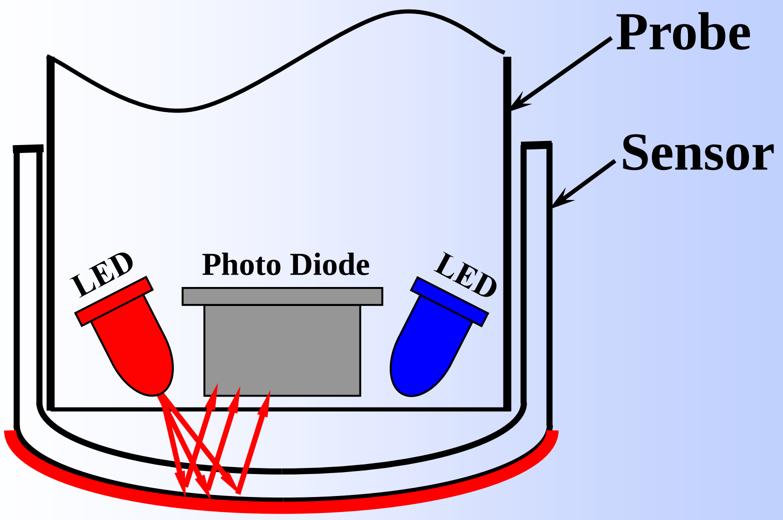
How Does LDO Work?

- The intensity of the red light is <u>not</u> what's being measured.
- What's being measured is the time it takes after excitation for red light to be given off.
  - Lifetime of luminescence



How Does LDO Work?

- A red LED is also present in the probe.
- Between flashes of the blue LED, a red LED of known intensity, is flashed on the sensor.
    - The red LED acts as an internal standard (or reference) for a comparison to the red light given off by the luminescent chemical.



Why is this a Big Deal?

Reduced Maintenance

- No membrane to replace
  - No more stretching of Teflon and worrying about air bubbles
  - No more punctured membranes
- No electrolyte to foul or poison
  - No H_2S poisoning of the electrolyte
- No anode or cathode
  - No cleaning of anodes
  - No more coating of electrodes

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No anode or cathode

- No cleaning of anodes
- No more coating of electrodes

Why is this a Big Deal?

- Frequent Calibration Not Required
  - No anode to consume and no electrolyte to deplete means extremely stable measurements
  - Internal standard with Red LED
  - No interference from pH swings, wastewater chemicals, H_2S, or heavy metals

Why is this a Big Deal?

- **Accurate and Stable Readings**
  - With nothing to interfere with the readings, LDO produces more stable measurements for a longer time
- **Speed!**
  - Turn it on and it's running!
  - Response time of less than 30 seconds to 90%!

Why is this a Big Deal?

- Simple Operation and Maintenance
  - Only one replacement part
  - Inexpensive sensor cap is simple to replace quickly

NOW EPA APPROVED

Listed In
Federal Register
Vol. 77,No. 97
Friday, May 18, 2012

Listed as ASTM Method D888-09 (C)

Footnote 63 – Hach Method 10360
(BOD and cBOD)
Footnote 64 In-Situ Method 1002-8-2009
(Dissolved Oxygen Measurement by Optical Probe)

URL - http://www.gpo.gov/fdsys/browse/collection.action?collectionCode=FR&browsePath=2012%2F05%2F05-18%5C%2F6%2FEnvironmental+Protection+Agency&isCollapsed=false&leafLevelBrowse=false&isDocumentResults=true&ycord=546

D.O. Meter

D.O. METER

Advantages

Saves Time

Continuous Monitoring

Less Chemical Interference

Portable

D.O. METER

Limitations

(Membrane Electrode)

Daily Calibration

Flow Past Membrane

Membrane May Foul

Requires Training

BOD_5

B.O.D.

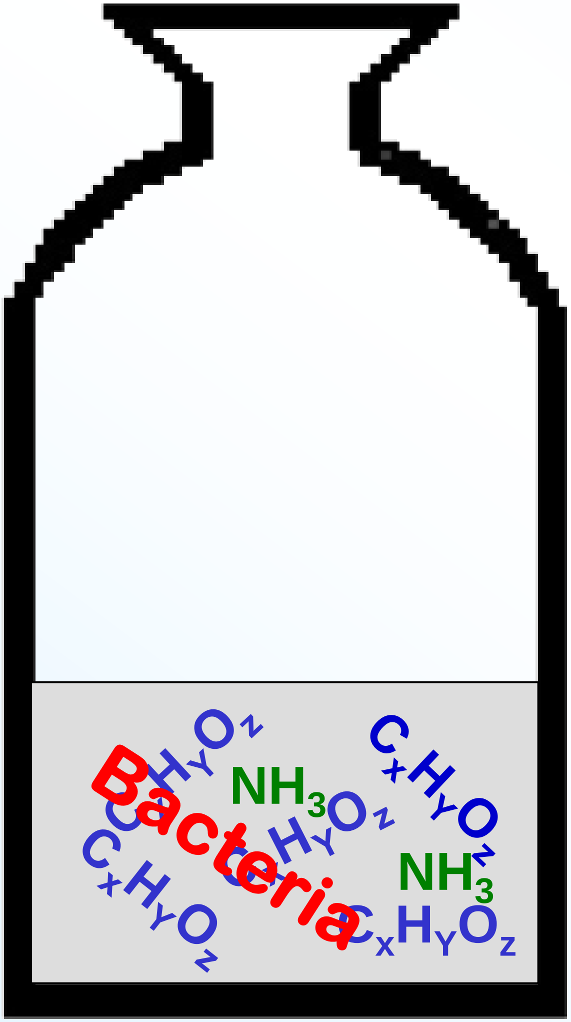
Biochemical Oxygen Demand

The Quantity of Oxygen Used in the Biochemical Oxidation of Organic Material.

Under:

Specified Time	**5 Days**
Specified Temperature	**20^0 C**
Specified Conditions	**In the Dark** **In the Presence of Bacteria**

Measured Volume of Wastewater is Added to BOD Bottle.



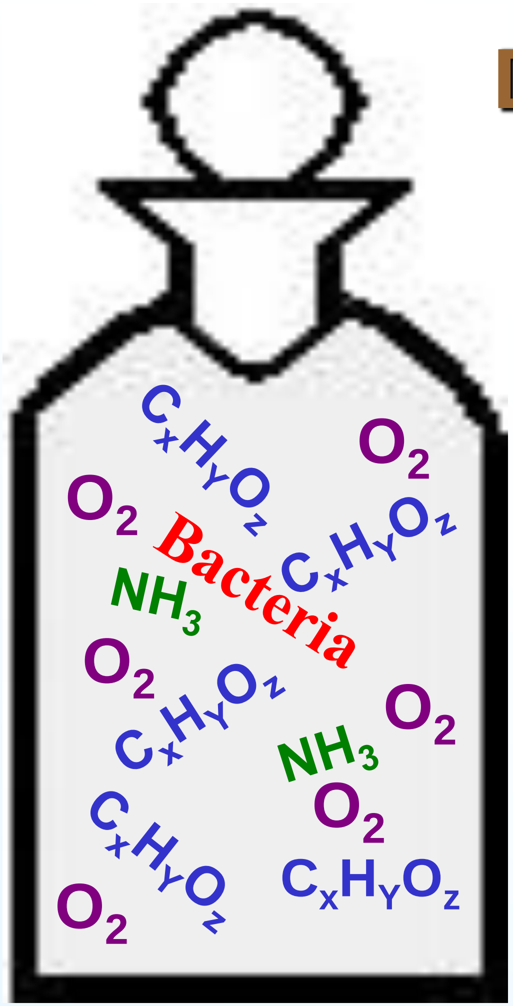
Contains:

Organics

Ammonia

Bacteria

Dilution Water Added



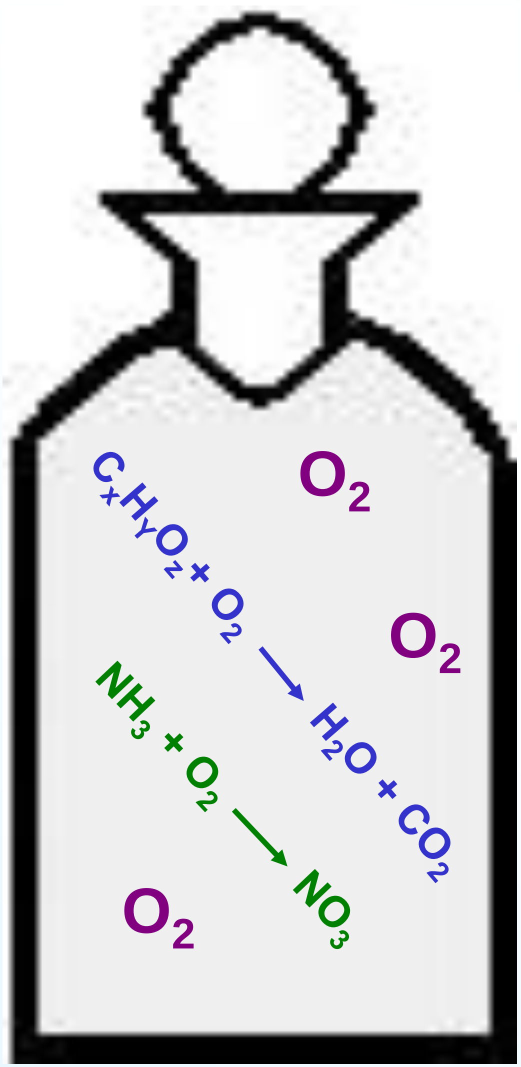
Dilution Water Contains:

Nutrients

Oxygen

Measure D.O. Concentration

Incubate 5 Days



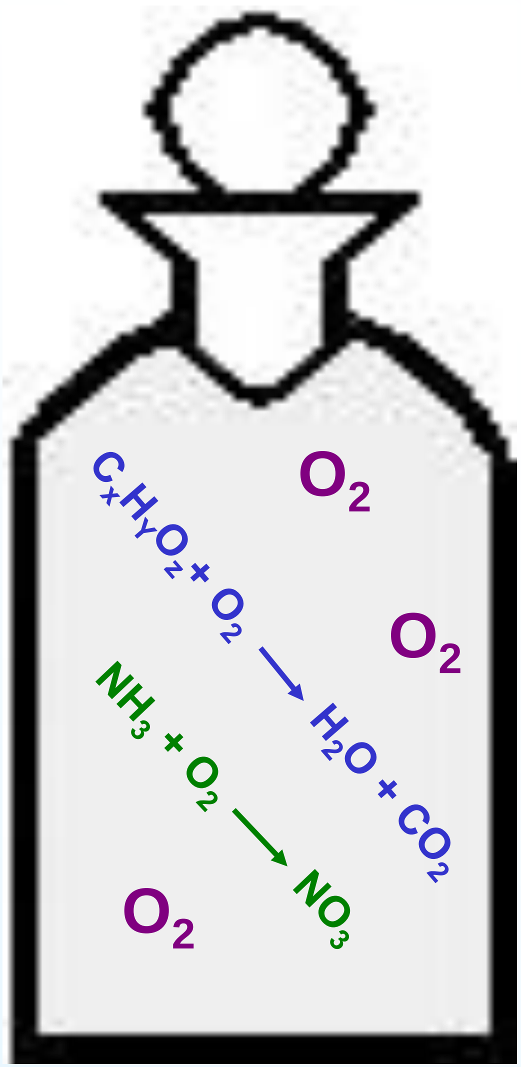
Some Oxygen Used:

Respiration

Nitrification

Measure
D.O. Concentration

Measure Oxygen Loss (Demand)



D.O. Day 1 - D.O. Day 5

= Oxygen Demand

(Of What Is In The Bottle)

BOD

REAGENTS

Distilled Water

High Quality

Free of Toxic Material

Free of Oxygen Demanding Substances

BOD

REAGENTS

Distilled Water

Phosphate Buffer
Magnesium Sulfate
Calcium Chloride
Ferric Chloride

Provide Essential Nutrients
Buffer pH

OTHER REAGENTS

DECHLORINATING AGENT
Sodium Sulfite - Na_2SO_3

NITRIFICATION INHIBITOR
CBOD

QUALITY CONTROL CHECK
Accuracy
Glucose - Glutamic Acid Solution

SAMPLE PRETREATMENT

Temperature

Near 20^{0}C

pH

Between 6.5 and 7.5

(Adjust if > 8.5 or < 6.0 and seed)

Supersaturated D.O.

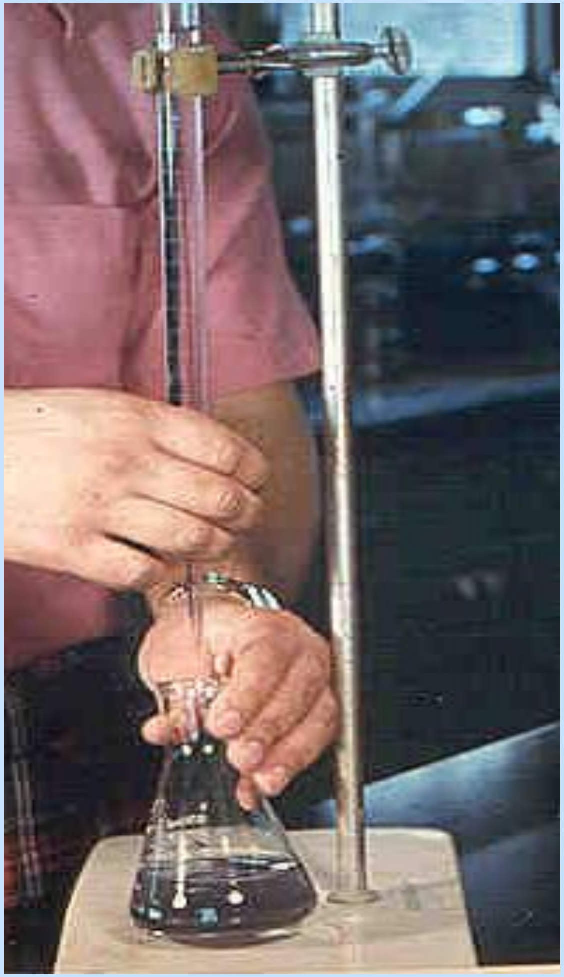
Agitate

Dechlorinate

Proper Amount of Sodium Sulfite

DECHLORINATION

100 mL of Sample
+ Potassium Iodide + Sulfuric Acid + Starch



Titrate with Sodium Sulfite

to Starch Iodide Endpoint

DILUTION WATER

Distilled Water

plus

BUFFER

plus

NUTRIENTS

High Quality
No Toxics
No Organics

pH 7.2

(Phosphorus
and Ammonia)
Magnesium
Calcium
Iron

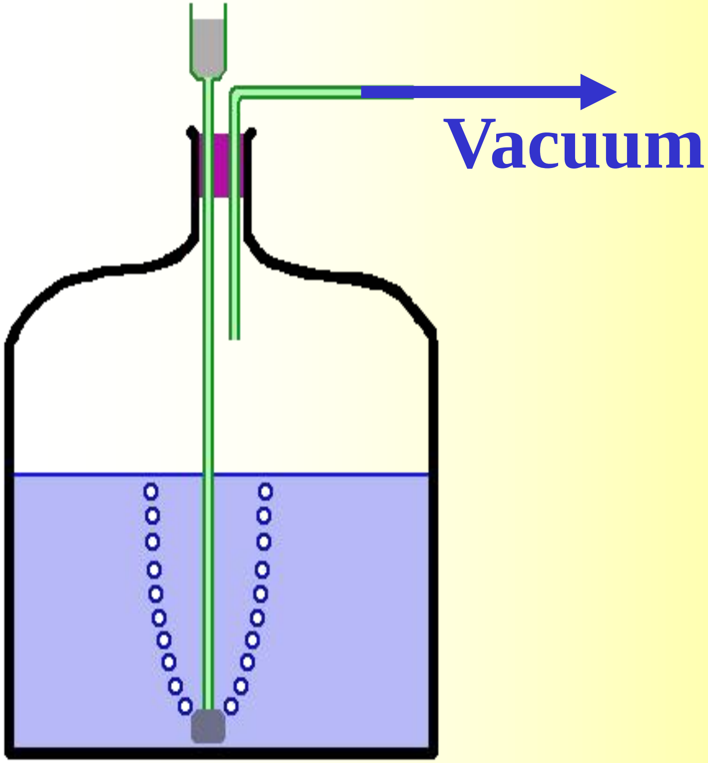
DILUTION WATER PREPARATION

NEEDED VOLUME for Each Day's Use

ADD NUTRIENTS (1 mL per Liter)

SATURATE WITH OXYGEN

Shake
(small volume)
or
Draw Vacuum



DILUTION WATER PREPARATION

NEEDED VOLUME

ADD NUTRIENTS

SATURATE WITH OXYGEN

STORE
In Incubator

ADD BUFFER
Day of Use

BOD PROCEDURE

DILUTE SAMPLE

Minimum Residual, 1.0 mg/L
Minimum Depletion, 2.0 mg/L
At Least Two Dilutions
Thoroughly Mix Sample

ADD NITRIFICATION INHIBITOR

If Required for CBOD

TCMP
0.10 gram/bottle
Two “shots”

BOD PROCEDURE

DILUTE SAMPLE

Minimum Residual, 1.0 mg/L
Minimum Depletion, 2.0 mg/L
At Least Two Dilutions
Thoroughly Mix Sample

ADD NITRIFICATION INHIBITOR

If Required for CBOD

Add SEED (Bacteria)

If Required

Disinfected Samples
Industrial Samples
Reference Samples

BOD PROCEDURE

Source of Seed (bacteria)

Settled Sewage

Primary Effluent

Commercially Available

Adapted Seed

BOD PROCEDURE, (cont.)

FILL BOTTLE (with dilution water)

MEASURE INITIAL D.O.

STOPPER

(No Air Bubbles)

SEAL

(Water and Cover)

BOD PROCEDURE, (cont.)

INCUBATE

20 ± 1°C

5 Days ± 6 hour

MEASURE FINAL D.O.

(wash bottles)

DILUTION WATER BLANK

CHECK ON QUALITY

MAXIMUM DEPLETION 0.2 mg/L

NOT USED IN CALCULATIONS

B.O.D. Calculation

D.O. In - D.O. Out

= DEPLETION

= Oxygen Demand of Diluted Sample

$$\boxed{C_1 \times V_1 = C_2 \times V_2}$$

$$\text{Sample BOD} \times \text{Sample Volume} = \text{BOD Diluted} \times \text{Diluted Volume}$$

$$\text{BOD Sample} = \frac{\text{BOD Diluted}}{\text{Sample Volume}} \times \text{Volume Diluted}$$

$$\text{BOD Sample} = \frac{\text{Depletion}}{\text{Sample Volume}} \times 300\text{ mL}$$

$$\text{B.O.D. mg/L} = \frac{\text{D.0. DEPLETION (mg/L)}}{\text{SAMPLE VOLUME (mL)}} \times 300 \text{ mL}$$

D.O. DEPLETION = D.O. Initial - D.O. 5-Day

Minimum Depletion - 2.0 mg/L

Minimum Residual - 1.0 mg/L

B.O.D. Example Problem

Calculate the B.O.D.:

Initial Sample D.O.	= 8.1 mg/L
5-Day Sample D.O.	= 2.1 mg/L
Vol. Of Sample in 300 mL Bottle	= 60 mL

$$\text{B.O.D., mg/L} = \frac{\text{D.O. Depletion, mg/L}}{\text{Volume Sample, mL}} \text{ X 300 mL}$$

$$= \frac{\text{Initial D.O. - Residual D.O.}}{\text{Volume Sample, mLs}} \text{ X 300 mL}$$

$$= \frac{\text{8.1 mg/L - 2.1 mg/L}}{\text{60 mL}} \text{ X 300 mL}$$

$$= \frac{\text{6.0 mg/L}}{\text{60 mL}} \text{ X 300 mL}$$

$$= \text{0.1 mg/L X 300 } \boxed{= \text{30 mg/L}}$$

B.O.D. PRACTICE PROBLEM

Calculate the B.O.D. value to be reported for each of the samples below. Be sure to consider the minimum depletion and residual requirements.

Sample 1	Dilution A	Dilution B
mL Sample	15	30
Initial D.O., mg/L	~~8.1~~	8.2
5-Day D.O., mg/L	~~6.6~~	4.2
Depletion	~~1.5~~	4.0

$$\text{B.O.D., mg/L} = \frac{\text{D.O. Depletion, mg/L}}{\text{Volume Sample, mL}} \text{ X 300 mL}$$

$$= \frac{4.0 \text{ mg/L}}{30 \text{ mL}} \text{ X 300 mL}$$

$$\boxed{= 40 \text{ mg/L}}$$

B.O.D. PRACTICE PROBLEM

Calculate the B.O.D. value to be reported for each of the samples below. Be sure to consider the minimum depletion and residual requirements.

Sample 2	Dilution A	Dilution B
mL Sample	15	30
Initial D.O., mg/L	8.0	~~8.2~~
5-Day D.O., mg/L	4.3	~~0.7~~
Depletion	3.7	

$$\text{B.O.D., mg/L} = \frac{\text{D.O. Depletion, mg/L}}{\text{Volume Sample, mL}} \times 300 \text{ mL}$$

$$= \frac{3.7 \text{ mg/L}}{15 \text{ mL}} \times 300 \text{ mL}$$

$$\boxed{= 74 \text{ mg/L}}$$

B.O.D. PRACTICE PROBLEM

Sample 3	Dilution A	Dilution B
mL Sample	15	30
Initial D.O., mg/L	8.1	8.1
5-Day D.O., mg/L	5.6	3.3
Depletion	2.5	4.8

$$BOD_A = \frac{2.5\ mg/L}{15\ mL} \times 300\ mL = 50\ mg/L$$

$$BOD_B = \frac{4.8\ mg/L}{30\ mL} \times 300\ mL = 48\ mg/L$$

$$\frac{50 + 48}{2} \boxed{= 49\ mg/L}$$

BOD PROCEDURE

DILUTE SAMPLE

Minimum Residual, 1.0 mg/L
Minimum Depletion, 2.0 mg/L
At Least Two Dilutions
Thoroughly Mix Sample

ADD NITRIFICATION INHIBITOR

If Required for CBOD

Add SEED (Bacteria)

If Required

De-chlorinated Samples
Industrial Samples
Reference Samples

BOD PROCEDURE

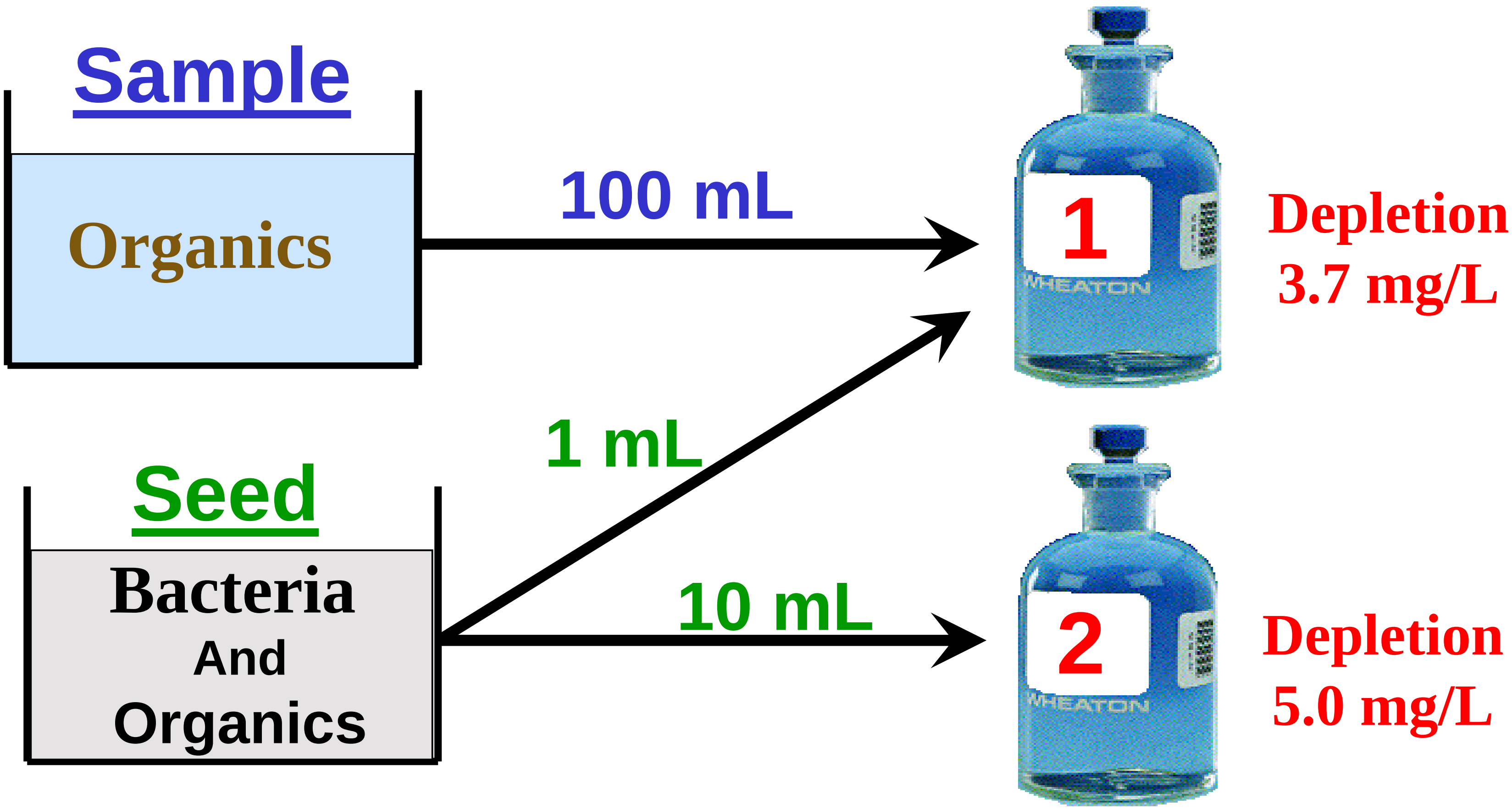
Source of Seed (bacteria)

Settled Sewage
Primary Effluent

Commercially
Available

Adapted Seed

SEEDED BOD PROCEDURE



10 mL of seed caused 5.0 mg/L Depletion

1 mL would have caused $\frac{5.0 \text{ mg/L}}{10 \text{ mL}}$ = 0.5 mg/L

So: 0.5 mg/L would be used by 1 mL of seed in seeded sample

SEEDED BOD CALCULATION

$$\text{BOD} = \frac{\text{Depletion Caused by Sample}}{\text{Volume of Sample Used}} \times 300 \text{ mL}$$

$$\text{BOD}_{(\text{seeded})} = \frac{D_1 - D_2}{\text{Sample Volume}} \times 300 \text{ mL}$$

SEEDED BOD CALCULATION

$$BOD_{(seeded)} = \frac{D_1 - D_2}{\text{Sample Volume}} \text{ X } 300 \text{ mL}$$

D_1 = Depletion Due To Sample and Seed

(Total Depletion in Bottle with Seed and Sample)

D_2 = D.O. Depletion Due to Just Seed

(Seed Depletion in Bottle with Seed and Sample)

?

$$BOD_{(seeded)} = \frac{D_1 - D_2}{\text{Sample Volume}} \times 300\ mL$$

D_2 = D.O. Depletion Due to Just Seed

(Seed Depletion in Bottle with Seed and Sample)

D_2 Must Be Calculated

Based on Bottle with Just Seed

$$D_2 = \frac{\text{D.O. Depletion of a Sample of Seed}}{\text{Volume of Seed Used}}$$

$$\boxed{\text{In The Bottle with Just Seed}}$$

$$BOD_{(seeded)} = \frac{D_1 - D_2}{\text{Sample Volume}} \times 300\ mL$$

D_2 = D.O. Depletion Due to Just Seed

(Seed Depletion in Bottle with Seed and Sample)

$$D_2 = \frac{\text{D.O. Depletion of a Sample of Seed}}{\text{Volume of Seed Used}}$$

In the Example

$$D_2 = \frac{5.0\ mg/L}{10mL} = 0.5\ mg/L$$

$$BOD_{(seeded)} = \frac{D_1 - D_2}{\text{Sample Volume}} \times 300 \text{ mL}$$

$$BOD_{(seeded)} =$$

$$\frac{3.7 \text{ mg/L} - 0.5 \text{ mg/L}}{100 \text{ mL}} \times 300 \text{ mL}$$

$$= \frac{3.2 \text{ mg/L}}{100 \text{ mL}} \times 300 \text{ mL}$$

$$= 9.6 \text{ mg/L}$$

D_2 = D.O. Depletion Due to Just Seed

(Seed Depletion in Bottle with Seed and Sample)

What If More Than 1mL Was Used for Seeding?

$$D_2 = \frac{\text{D.O. Depletion of a Sample of Seed}}{\text{Volume of Seed Used}}$$

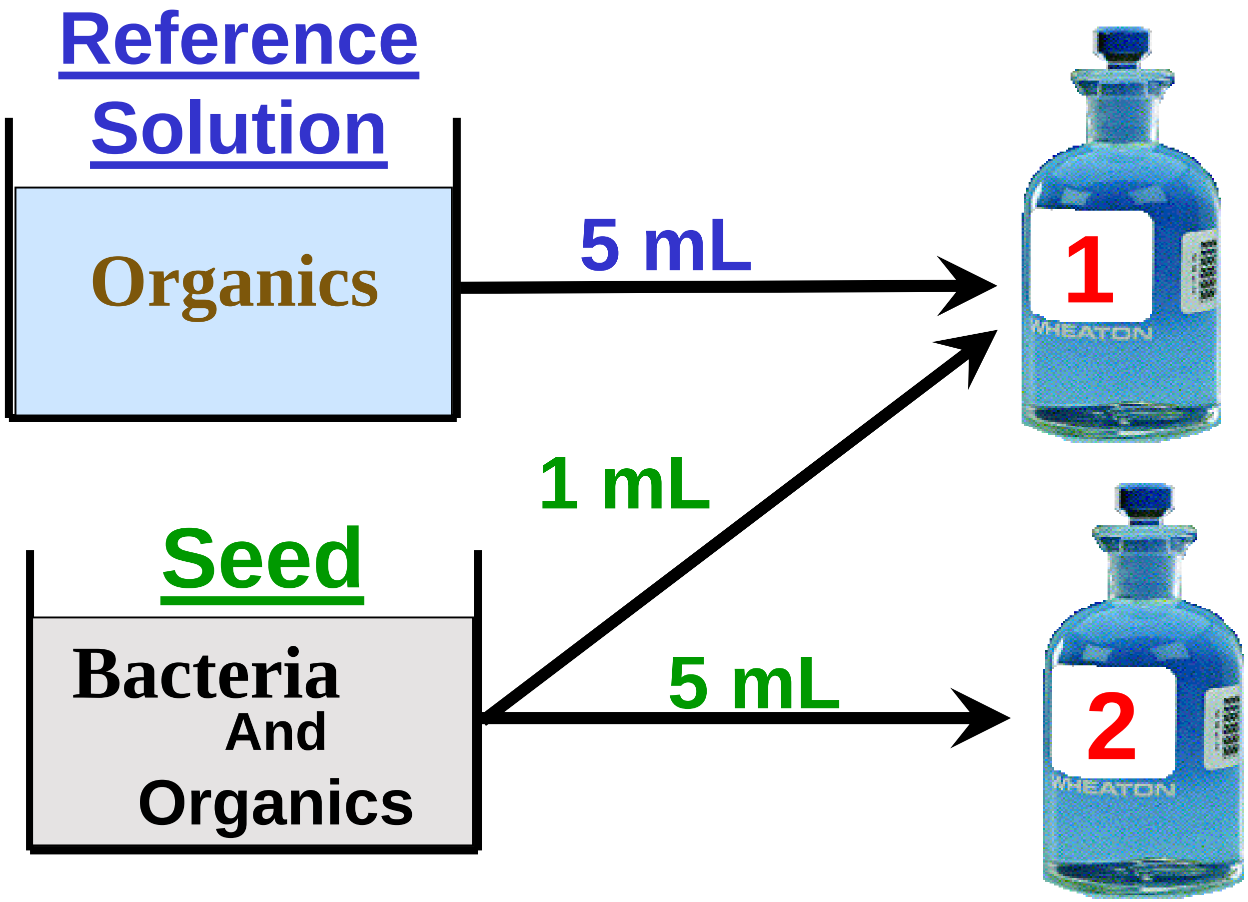
$$D_2 = \frac{5.0\ \text{mg/L}}{10\ \text{mL}} = 0.5\ \text{mg/L}\ /\text{mL}$$

<u>Multiply by mL Used for Seeding</u>

Example Seeded BOD Calculation.

Calculate the B.O.D. of a reference sample given that 1mL of seed material was used in the reference sample. (300 mL BOD bottles were used)

	Seed Material	Reference Sample
Volume Used	5 mL	5 mL
Initial D.O.	8.6 mg/L	8.7 mg/L
Final D.O.	5.8 mg/L	4.8 mg/L



Example Seeded BOD Calculation.

Calculate the B.O.D. of a reference sample given that 1mL of seed material was used in the reference sample. (300 mL BOD bottles were used)

	Seed Material	Reference Sample
Volume Used	5 mL	5 mL
Initial D.O.	8.6 mg/L	8.7 mg/L
Final D.O.	5.8 mg/L	4.8 mg/L

$$BOD_{seeded} = \frac{D_1 - D_2}{\text{Sample Volume, mL}} \times 300\ \text{mL}$$

D_1 = Depletion of Sample & Seed

$$D_1 = 8.7\ \text{mg/L} - 4.8\ \text{mg/L}$$

$$D_1 = 3.9\ \text{mg/L}$$

Example Seeded BOD Calculation.

Calculate the B.O.D. of a reference sample given that 1mL of seed material was used in the reference sample. (300 mL BOD bottles were used)

	Seed Material	Reference Sample
Volume Used	5 mL	5 mL
Initial D.O.	8.6 mg/L	8.7 mg/L
Final D.O.	5.8 mg/L	4.8 mg/L

$$BOD_{seeded} = \frac{D_1 - D_2}{\text{Sample Volume, mL}} \text{ X 300 mL}$$

D_2 = Depletion of Seed (In Seeded Sample)

$$D_2 = \frac{8.6 \text{ mg/L} - 5.8 \text{ mg/L}}{5 \text{ mL}}$$

$$D_2 = \frac{2.8 \text{ mg/L}}{5 \text{ mL}} = 0.56$$

Example Seeded BOD Calculation.

Calculate the B.O.D. of a reference sample given that 1mL of seed material was used in the reference sample. (300 mL BOD bottles were used)

	Seed Material	Reference Sample
Volume Used	5 mL	5 mL
Initial D.O.	8.6 mg/L	8.7 mg/L
Final D.O.	5.8 mg/L	4.8 mg/L

$$BOD_{seeded} = \frac{D_1 - D_2}{\text{Sample Volume, mL}} \times 300 \text{ mL}$$

$$BOD = \frac{3.9 \text{ mg/L} - 0.56 \text{ mg/L}}{5 \text{ mL}} \times 300 \text{ mL}$$

$$BOD = \frac{3.34 \text{ mg/L}}{5 \text{ mL}} \times 300 \text{ mL}$$

$$\boxed{BOD = 200.4 \text{ mg/L}}$$

Example #2 Seeded BOD Calculation.

Calculate the B.O.D. of an de-chlorinated effluent sample given that 1mL of seed material was used in the sample. (300 mL BOD bottles were used)

	Seed Material	Sample
Volume Used	10 mL	200 mL
Initial D.O.	8.3 mg/L	8.5 mg/L
Final D.O.	5.6 mg/L	3.7 mg/L

$$BOD_{seeded} = \frac{D_1 - D_2}{\text{Sample Volume, mL}} \times 300\text{ mL}$$

D_1 = Depletion of Sample & Seed

$$D_1 = 8.5\text{ mg/L} - 3.7\text{ mg/L}$$

$$D_1 = 4.8\text{ mg/L}$$

Example #2 Seeded BOD Calculation.

Calculate the B.O.D. of an de-chlorinated effluent sample given that 1mL of seed material was used in the sample. (300 mL BOD bottles were used)

	Seed Material	Sample
Volume Used	10 mL	200 mL
Initial D.O.	8.3 mg/L	8.5 mg/L
Final D.O.	5.6 mg/L	3.7 mg/L

$$BOD_{seeded} = \frac{D_1 - D_2}{\text{Sample Volume, mL}} \text{ X 300 mL}$$

D_2 = Depletion of Seed (In Seeded Sample)

$$D_2 = \frac{8.3\ \text{mg/L} - 5.6\ \text{mg/L}}{10\ \text{mL}}$$

$$D_2 = \frac{2.7\ \text{mg/L}}{10\ \text{mL}} = 0.27$$

Example #2 Seeded BOD Calculation.

Calculate the B.O.D. of an de-chlorinated effluent sample given that 1mL of seed material was used in the sample. (300 mL BOD bottles were used)

	Seed Material	Sample
Volume Used	10 mL	200 mL
Initial D.O.	8.3 mg/L	8.5 mg/L
Final D.O.	5.6 mg/L	3.7 mg/L

$$BOD_{seeded} = \frac{D_1 - D_2}{\text{Sample Volume, mL}} \times 300 \text{ mL}$$

$$BOD = \frac{4.8 \text{ mg/L} - 0.27 \text{ mg/L}}{200 \text{ mL}} \times 300 \text{ mL}$$

$$BOD = \frac{4.53 \text{ mg/L}}{200 \text{ mL}} \times 300 \text{ mL}$$

$$\boxed{BOD = 6.8 \text{ mg/L}}$$

BOD Practice Problems

1. Calculate the B.O.D. given:

D.O. IN	=	7.0 mg/L
D.O. OUT (5-day)	=	3.5 mg/L
Vol. Sample in B.O.D. bottle	=	15 mL

2. Calculate the B.O.D. of a reference sample given that 1mL of seed material was used in the reference sample.
(300 mL BOD bottles were used)

	Seed Material	Reference Sample
Volume Used	9 mL	5 mL
Initial D.O.	8.7 mg/L	8.6 mg/L
Final D.O.	5.1 mg/L	5.0 mg/L

Work Calculations on Separate Paper
Answers Given on Next Slides

1. Calculate the B.O.D. given:

D.O. IN	=	7.0 mg/L
D.O. OUT (5-day)	=	3.5 mg/L
Vol. Sample in B.O.D. bottle	=	15 mL

$$\text{BOD} = \frac{\text{Depletion, mg/L}}{\text{Sample Volume, mL}} \text{ X 300 mL}$$

$$\text{BOD} = \frac{\text{7.0 mg/L} - \text{3.5 mg/L}}{\text{15 mL}} \text{ X 300 mL}$$

$$\text{BOD} = \frac{\text{3.5 mg/L}}{\text{15mL}} \text{ X 300 mL}$$

$$\boxed{\text{BOD} = \text{70 mg/L}}$$

2. Calculate the B.O.D. of a reference sample given that 1mL of seed material was used in the reference sample. (300 mL BOD bottles were used)

	Seed Material	Reference Sample
Volume Used	9 mL	5 mL
Initial D.O.	8.7 mg/L	8.6 mg/L
Final D.O.	5.1 mg/L	5.0 mg/L

$$BOD_{seeded} = \frac{D_1 - D_2}{\text{Sample Volume, mL}} \text{ X 300 mL}$$

D_1 = Depletion of Sample & Seed

$$D_1 = 8.6 \text{ mg/L} - 5.0 \text{ mg/L}$$

$$D_1 = 3.6 \text{ mg/L}$$

D_2 = Depletion of Seed (In Seeded Sample)

$$D_2 = \frac{8.7 \text{ mg/L} - 5.1 \text{ mg/L}}{9 \text{ mL}}$$

$$D_2 = \frac{3.6 \text{ mg/L}}{9 \text{ mL}} = 0.40$$

2. Calculate the B.O.D. of a reference sample given that 1mL of seed material was used in the reference sample.
(300 mL BOD bottles were used)

	Seed Material	Reference Sample
Volume Used	9 mL	5 mL
Initial D.O.	8.7 mg/L	8.6 mg/L
Final D.O.	5.1 mg/L	5.0 mg/L

$$BOD_{seeded} = \frac{D_1 - D_2}{\text{Sample Volume, mL}} \text{ X 300 mL}$$

D_1 = Depletion of Sample & Seed
D_2 = Depletion of Seed

$$BOD = \frac{3.6 \text{ mg/L} - 0.40 \text{ mg/L}}{5 \text{ mL}} \text{ X 300 mL}$$

$$BOD = \frac{3.2 \text{ mg/L}}{5 \text{ mL}} \text{ X 300 mL}$$

$$\boxed{BOD = 192 \text{ mg/L}}$$

Dissolved Oxygen and Biochemical Oxygen Demand Analyses

Prepared By
Michigan Department of Environmental Quality
Operator Training and Certification Unit