SUPPLEMENTAL MATERIALS

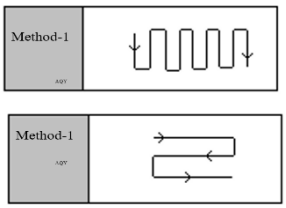
Appendix 1. Laboratory Curriculum Sample

Department of Microbiology, Immunology & Infectious Diseases

Problem	Chronic productive cough	MATERIALS MAY BE POTENTIALLY
Unit	III	INFECTIOUS; YOU ARE EXPECTED TO
Year	2	ADOPT THE STANDARD PRECAUTIONS (**SP**)*
Date	Monday-03.02.2020	
Time	10:00 am	

STATION-1

1. Discuss the proper sputum sample collection for mycobacteriological laboratory diagnostic methods.
2. Observe and discuss Ziehl Neelsen method while staining the provided **fixed** sputum smear.
   Ziehl- Neelsen (ZN) stain method
   a. **Flood** the slide with **carbol fuchsin**; **heat** from underneath till **steam rises** but ***don't*** *allow to* ***boil*** (re-heat at intervals). Stain for **7 -10** minutes.
   b. **Wash** in running tap water.
   c. **Decolorize** in 3 % acid alcohol till **excess** of stain **removes**. **(For spore staining decolorize using 0.25% to 0.5% sulfuric acid for 7-10 min)**
   d. **Wash** in running tap water.
   e. **Counterstain** with 0.5 % methylene blue or 0.5 % malachite green for **one minute**.
   f. **Wash** in running tap water.
   g. **Blot dry** and examine microscopically under X10, X40 and confirm with the X100 **oil immersion** objective
3. Examine the provided Ziehl Neelsen stained sputum smears.



Results

Acid fast bacilli	Red, straight or slightly curved rods, occurring singly or in small groups, may appear beaded.
Cell other than acid fast	Blue (if counterstain used is methylene blue) or Green (if the counterstain used is malachite green).
Background material	Blue (if counterstain used is methylene blue) or Green (if the counterstain used is malachite green).

STATION-2

1. Interpret the findings of stained Ziehl Neelsen smears and discuss how to report.

Number of AFB per oil immersion objective	**Result**	**Grading score**	**No. of fields to be examined**
No AFB/100 fields	Negative	0	100
1 or 2 AFB/100 fields*			Total in 100
1-9 AFB/100 fields	Scanty	Record exact number seen	Total in 100
10-99 AFB/100 fields	Positive	**1 +**	Total in 100
1-10 AFB/ field	Positive	**2 +**	In each of 50 fields
>10 AFB/ field	Positive	**3 +**	In each of 20 fields
***Request a further specimen to examine:** **(Those AFB might have come from tap water (saprophytic mycobacteria), or it may be scratch of glass slide or by the use of same piece of blotting paper while drying**			

2. Understand fluorochrome procedure using fluorochrome dye, Auramine-Rhodamine.

STATION-3

1. **Differentiate** on the pictures provided among different mycobacteria cultured on **Lowenstein Jensen (LJ) medium**.
2. Discuss advantages and disadvantages of **molecular techniques** in the diagnosis of TB. Polymerase chain reaction (**PCR** as an example).
3. Discuss antibiotic sensitivity testing and antibiotic resistant (MDR-XDR)

Appendix 2: Mini Cases

UNIT	**V**	**MATERIALS MAY BE POTENTIALLY INFECTIOUS; YOU ARE EXPECTED TO ADOPT THE STANDARD PRECAUTIONS (SP)***
PROBLEM	**A patient with Distended Abdomen & Hematemesis.**	
YEAR	**Year –3.**	
DATE	**Thursday - 02. January - 2020**	
TIME	**09:00 a.m.-12:00 p.m.**	
Presented by		
Edited by		

A 20-year-old female bank teller was evaluated for fatigue, malaise, nausea, and jaundice in **January** 2018. She had no history of alcohol or drug abuse. She was not taking any medications and had no previous history of liver disease.

Initial laboratory data showed **total bilirubin 13.6** mg/dl (H), **alanine aminotransferase** (**ALT**) **1,604** IU/L (H), and **normal alkaline phosphatase** and **serum albumin** levels.
IgM antibody to **hepatitis A** virus (**anti-HAV**) was **positive**, hepatitis B virus surface antigen (**HBsAg**) was **negative**, and hepatitis C virus antibody (**anti-HCV**) was **nonreactive**.

Hepatitis serological profile:

TEST	RESULTS	REFERENCE RANGE
HBs Ag	Negative	Negative
Anti-HBs	Negative	Negative
Anti-HBc	Negative	Negative
Anti-HAV	Positive	Negative
Anti-HAV IgM	Positive	Negative
Anti-HCV	Negative	Negative
Anti-HBs (SRU)	1	0-2
ALT (SGPT)	1604 H	3 -45

(SRU's) =sample ratio units

A diagnosis of **acute hepatitis A** was made; the patient appeared to recover. In **February**, her **bilirubin** level had fallen to 4.2 mg/dl and her **ALT** level was 180 IU/L.

1. **What is the usual clinical course of hepatitis A?**

In early **March**, however, she experienced an increase in **nausea** associated with several episodes of **vomiting**, progressive **fatigue**, deepening **jaundice**, and the new onset of **pruritus**.

Repeat laboratory data showed serum total **bilirubin 24 mg/dL**, and **ALT 2,420 IU/L**. Repeat **IgM anti-HAV** was **positive**; other serologies were still negative. Abdominal ultrasound revealed no abnormalities.

LIVER FUNCTION TESTS (LFT):

TEST	RESULTS	REFERENCE RANGE	UNITS
Alkaline Phosphatase	110	40-120	IU/L
ALT (SGPT)	2420 H	3 -45	IU/L
AST (SGOT)	2070 H	3 -45	IU/L
Total Protein	7.1	6.0 - 8.0	g/dL
Albumin	4.1	3.2 -5. 0	g/dL
Globulin	3.0	1.3 -3.6	g/dL
A/G Ratio	1.37	0.99 -3.55	
Total Bilirubin	24 H	0.1 -1.3	mg/dL
Direct Bilirubin	5.9 H	0.1- 1.3	mg/dL

AST (or **SGOT) SGOT** = **Serum glutamic-oxaloacetic transaminase**), **AST** = Aspartate aminotransferase

The patient was followed with serial visits and laboratory tests, which demonstrated **a slowly progressive fall in serum total bilirubin level to 14 mg/dl** in April, **8.2 mg/dL** in **May**, **2.0 mg/dl** in **July**, and **to normal** in October. During the same period, the **ALT level also gradually normalized**. The patient had **persistent fatigue, malaise, jaundice,** and **pruritus** that prevented return to employment until the late summer. Thereafter, the patient experienced complete recovery.

Interactive discussion and Learning objectives

1. Define the clinical characteristics of hepatitis A

2. Mention the laboratory abnormalities characterize HAV infection

3.Define the typical serologic events associated with hepatitis A

4. list the clinical manifestation of hepatitis A

5. Discuss the rationale for immunizing children and adults against hepatitis A

Appendix 3. Self-survey for PBL-based Microbiology Laboratory

Department of Microbiology, Immunology and Infectious Diseases
Problem: ……. Date:……
PART I
Grade the following on a scale of 5 where 5 - strongly agree 4- agree 3- cannot comment, 2-Disagree 1- Strongly Disagree

Sl. Number	Statement	Score
1.	The timing of the session was enough to achieve the objectives	
2.	The session was relevant to the concepts in the problem under study	
3.	Teaching materials and handouts are clear and easy to follow	
4.	Briefing before the session helped in understanding the concepts	
5.	The session was relevant to clinical diagnosis	
6.	The laboratory presentation was clear and specific	
7.	The presented tests were observed / visualized before	
8.	The case-based study helped to apply the concepts better	

PART II

1. What factors related to the session helped you better?
   ……
   ……
   ……

2. Did you face any difficulties/ challenges in learning? Yes / No

   If yes, Please specify
   ……
   ……
   ……

3. What changes do you recommend to make you learn better?
   ……
   ……

4. Do you have any other comments/ remarks/ suggestions?
   ……
   ……
   ……