

LECTURE NOTES



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PREFACE

Text book on Medical Bacteriology for Medical Laboratory Technology students are not available as need, so this lecture note will alleviate the acute shortage of text books and reference materials on medical bacteriology.

Since it comprises most of the contents of course outline on medical bacteriology to nursing, pharmacy and environmental science students, it can be used as a main learning material to these category of students.

This lecture note gives emphasis on the knowledge and procedures of medical bacteriology to common pathogens in our country.

At last but not least, the quality of this lecture note is kept updated by continous comments made by users of this lecture note.

Abilo Tadesse

Meseret Alem

ACKNOWLEDGEMENT

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Our deepest gratitude goes to Prof. Dennis Carlson for his invaluable technical and moral support for the completion of this work.

We also extend our appreciation to those individuals who reviewed this lecture note in different teaching institutions for the materialization of this lecture note.

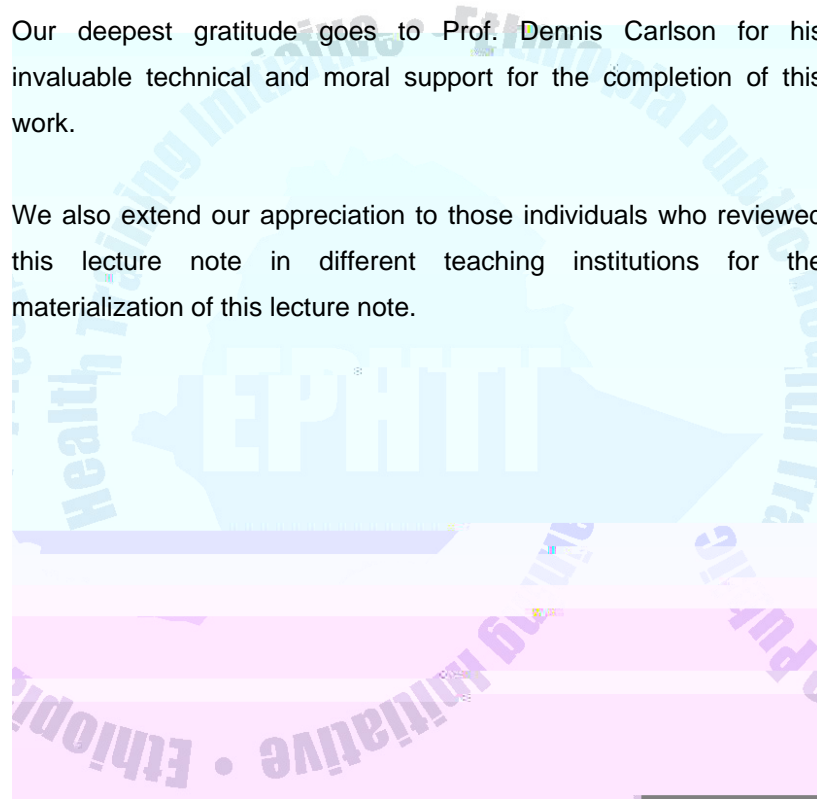


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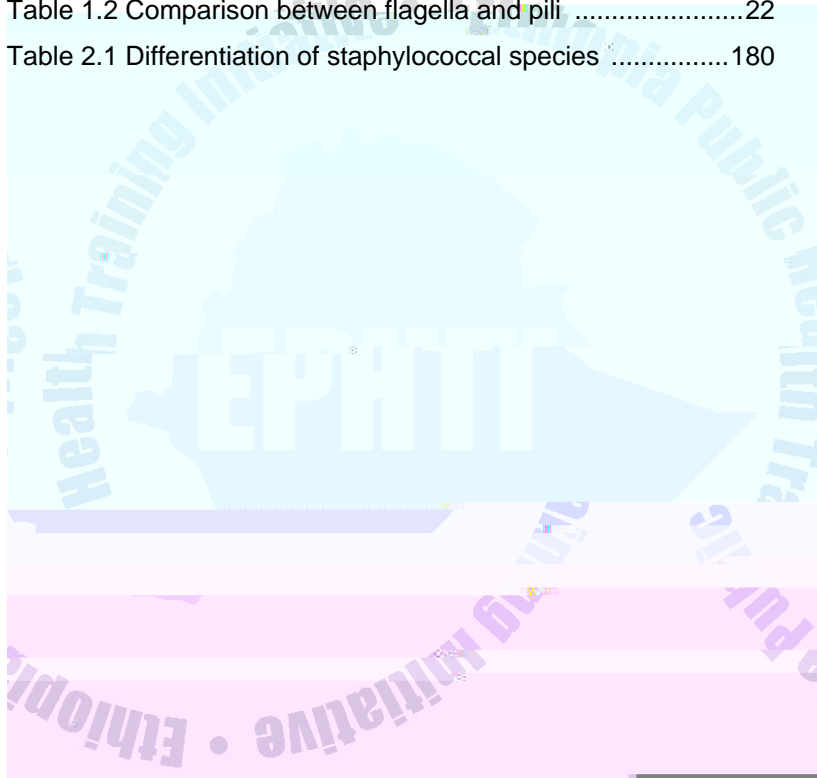
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ABBREVIATIONS

. AIDS.....	Acquired immunodeficiency syndrome
. AFB	Acid fast bacilli
. ATP	Adenosine triphosphate
. CO ₂	Carbon dioxide
. CSF.....	Cerebrospinal fluid
. CNS	Central nervous system
. DNA	Deoxy ribonucleotide
. DNase	Deoxy ribonucleotidase
. GIT	Gastrointestinal tract
. HIV	Human immunodeficiency virus
. HPF	High power field
. IP	Incubation period
. LGV	Lymphogranuloma venereum
. NADase	Nicotinamide adenine dinucleotidase
. NB	Notta Bonne
. O _c	Degree of Celsius
. P ^H	Hydrogen ion concentration
. RBC	Red blood cell
. RNA	Ribonucleotide
. RPR	Rapid plasma reagin
. SS agar	Salmonella-Shigella agar
. STD	Sexually transmitted disease
. UTI	Urinary tract infection
. VDRL	Venereal disease research laboratory test
. WBC	White blood cell

CHAPTER ONE

Learning Objective

-

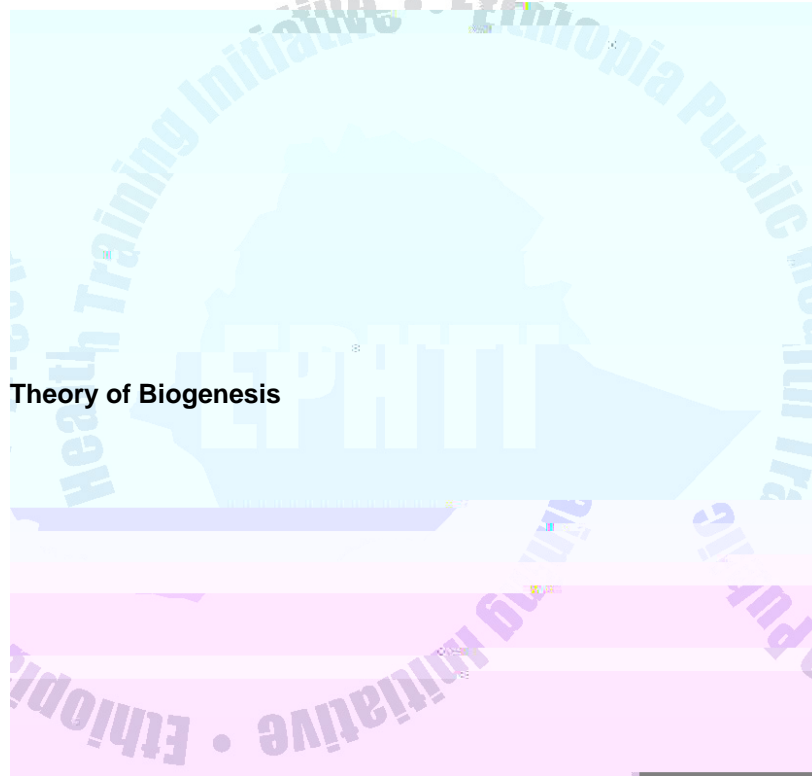


Subdivision of microbiology

History of Microbiology



Theory of Abiogenesis



Major contribution of Louis Pasteur

The germ theory of disease

Major achievements of Robert Koch

Koch's postulates: proof of germ theory of disease

Exceptions to Koch's postulates

1.2. THE MICROBIAL WORLD

TAXONOMIC CLASSIFICATION OF ORGANISMS



Escherichia coli
, *Escherichia*
coli)

Escherichia coli)

Escherichia “









PROKARYOTIC CELL



Bacterial Cell

-
-
-
-
-
-

1.3. STRUCTURE OF BACTERIA



Functions of cell wall



B. Cell membrane

Function of cell membrane

2. Cellular element enclosed with in the cell envelope

A. Mesosomes

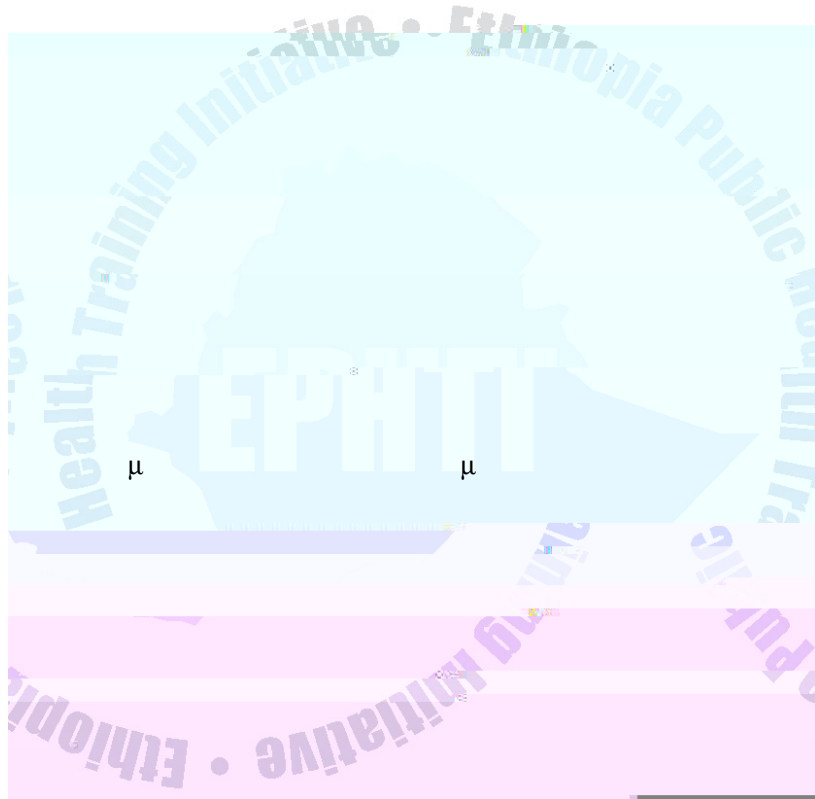
B. Ribosomes

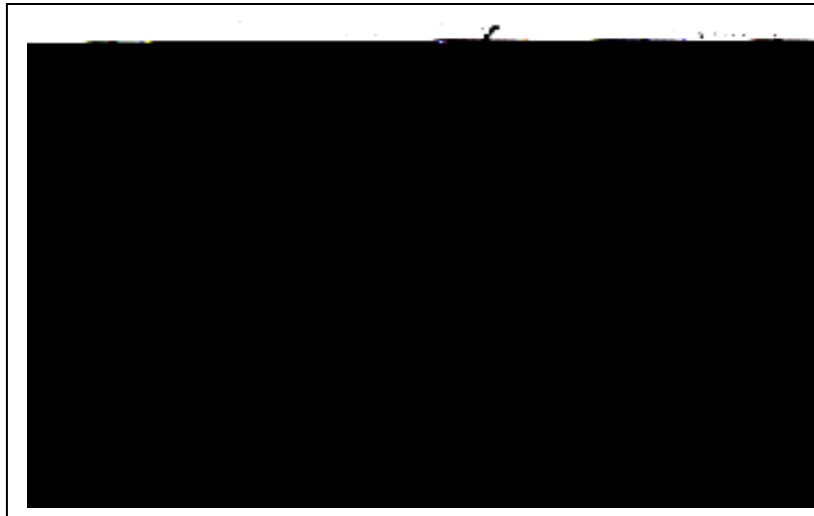
C. Polyamines





B. Flagellum





Flagellar arrangements





Endoflagella (axial filament)

C. Pili (fimbriae)

Two types (Based on function)

1.4. Classification of bacteria

1. Morphology of bacteria

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μ

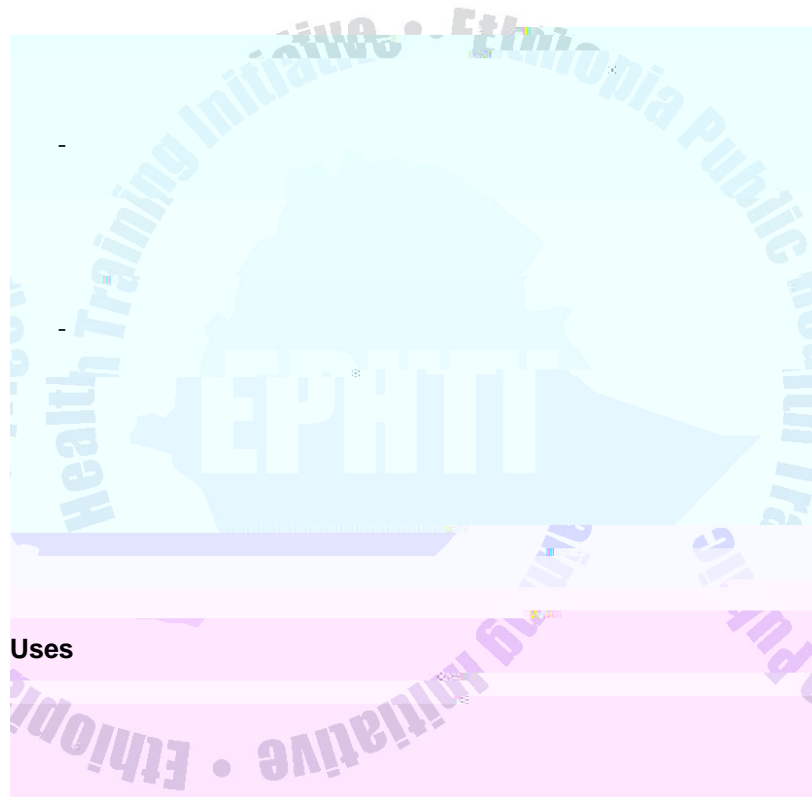
μ



2. Staining of bacteria







Uses

Types of microbiological stains

NB:

Basic stains

Acidic stains

Neutral stains

Types of staining methods

1. Simple staining method

Procedure:



Required reagents:

Procedure:



B. Ziehl-Neelson staining method

Reagents required:

Procedure for Ziehl-Neelson staining method



3. Special stains

a. Spore staining method

Procedure:



Peptone:

Meat extract:

Yeast extract:

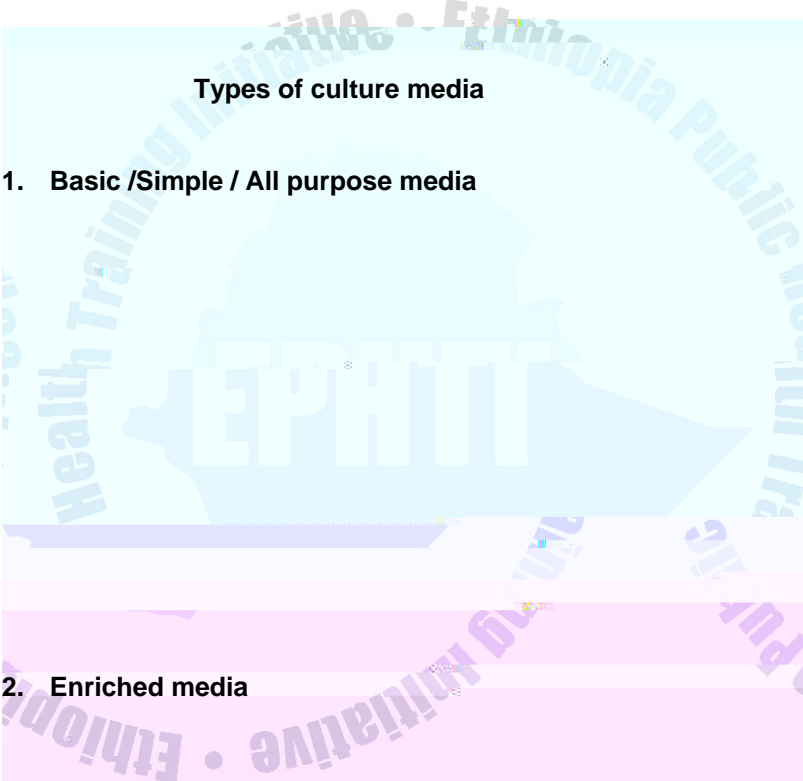
Mineral salts:

Carbohydrates:

Agar:

Property

Water

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Types of culture media

- 1. Basic /Simple / All purpose media**
- 2. Enriched media**

3. Enrichment media

4. Selective media

1. Differential media

2. Transport media

-
-
-
-
-

2. Semisolid culture media

Uses:

3. Fluid culture media

Uses :

Preparation of culture media

-
-
-

2. Sterilization and sterility testing

A) Autoclaving

B) Steaming at 100 °C

C) Filtration

Sterility testing

3. Addition of heat-sensitive ingredients

4. pH testing

5. Dispensing of culture media

Dispensing agar media in petridish

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-
-

-

6. Quality control

-

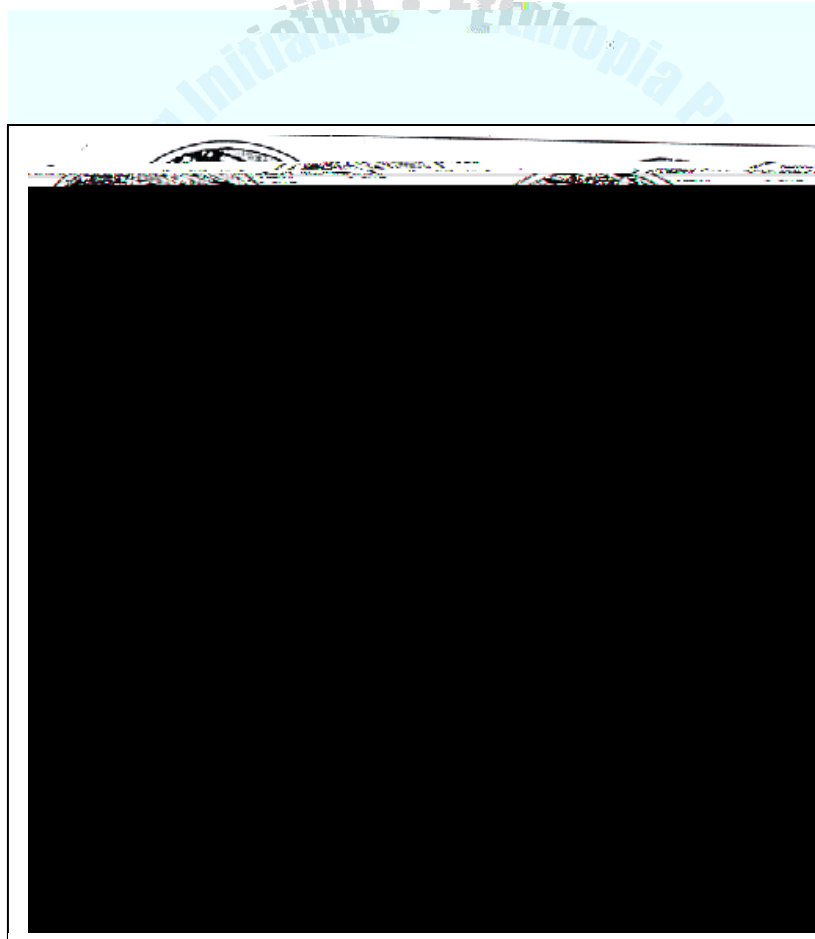


Aseptic technique during inoculation of culture media



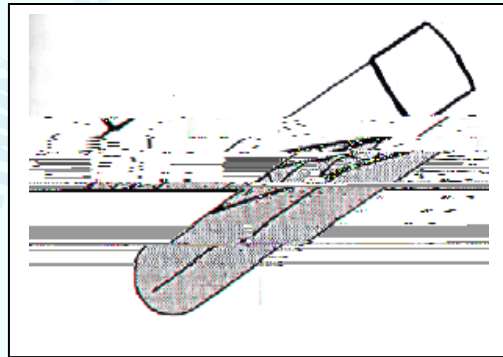
Fig. 1.6

Inoculation of media in petridishes

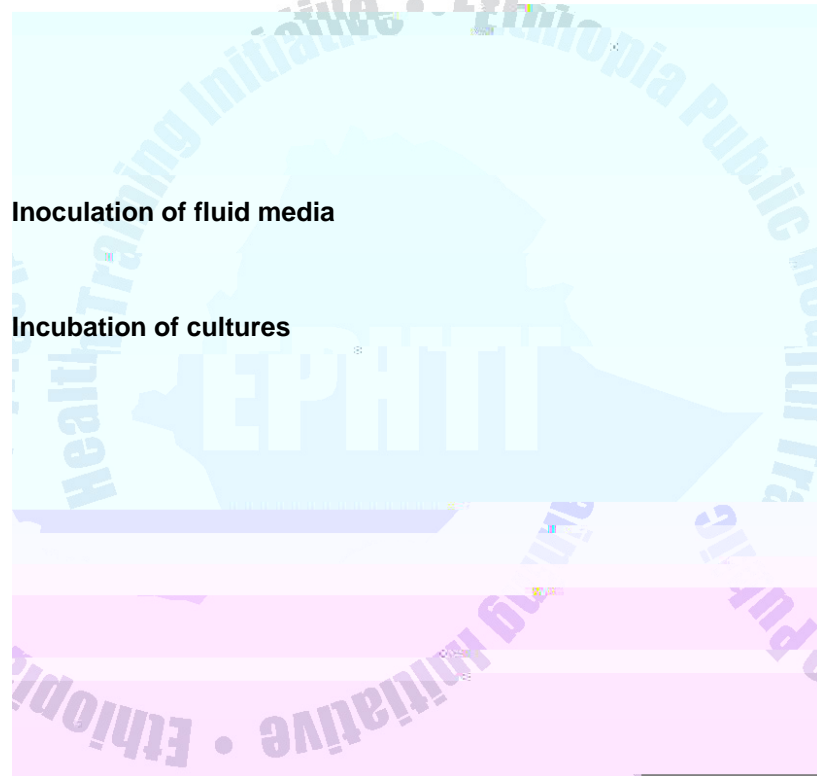


Inoculation of butt and slant media

Inoculation of slant media



Inoculation of stab media



Inoculation of fluid media

Incubation of cultures

Hydrogen and oxygen

-
-

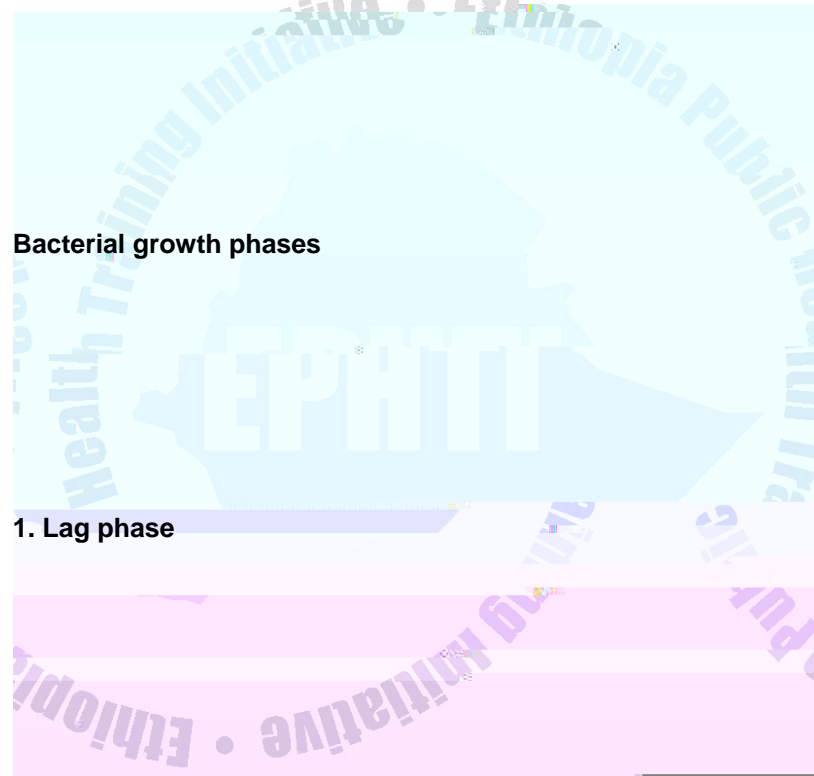
Nitrogen

-
-

Growth factors

1.7. BACTERIAL GROWTH

Generation time



3. Maximal stationary phase



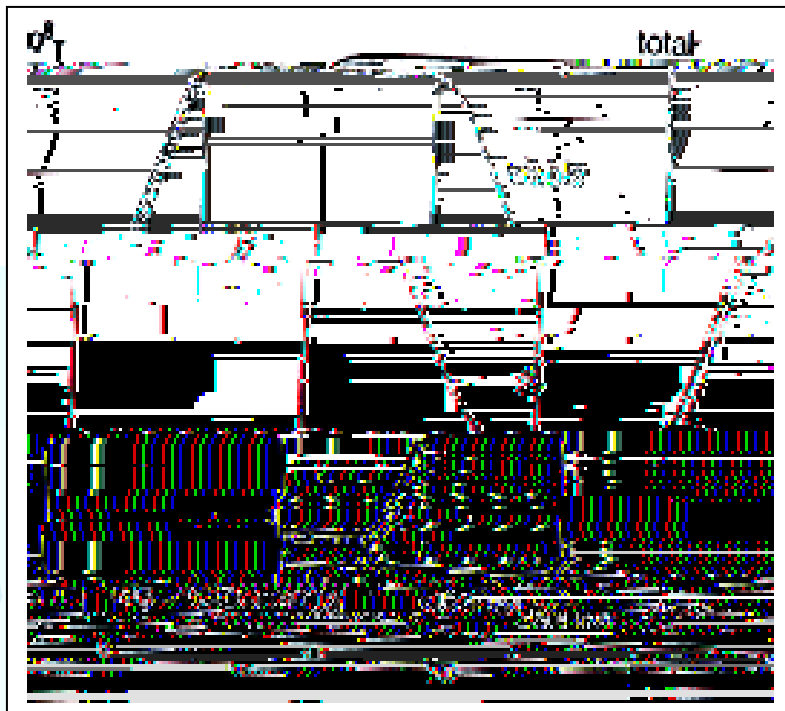
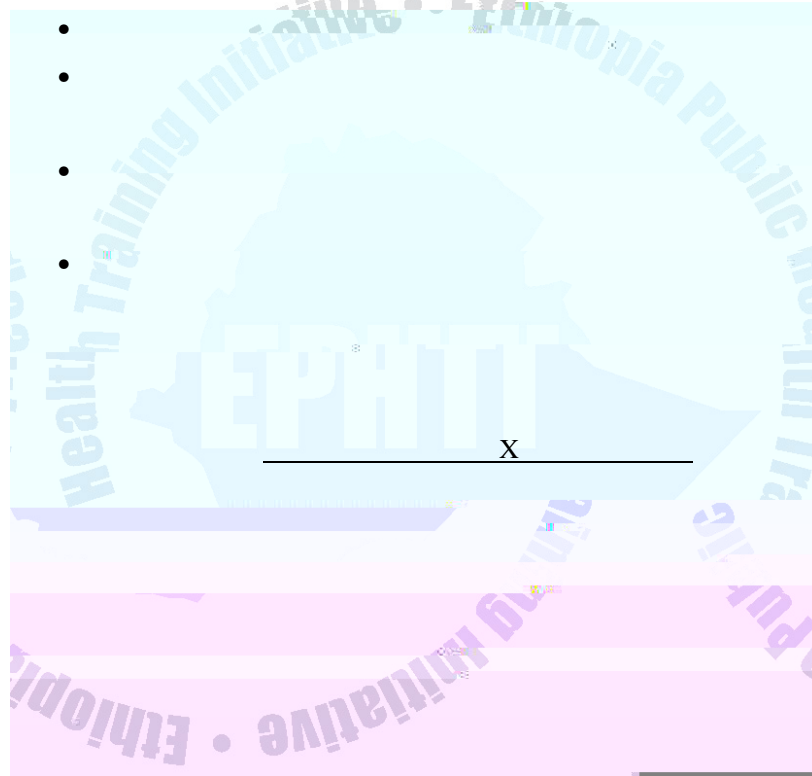


Fig. 1.11

Quantitative measurement of bacterial growth

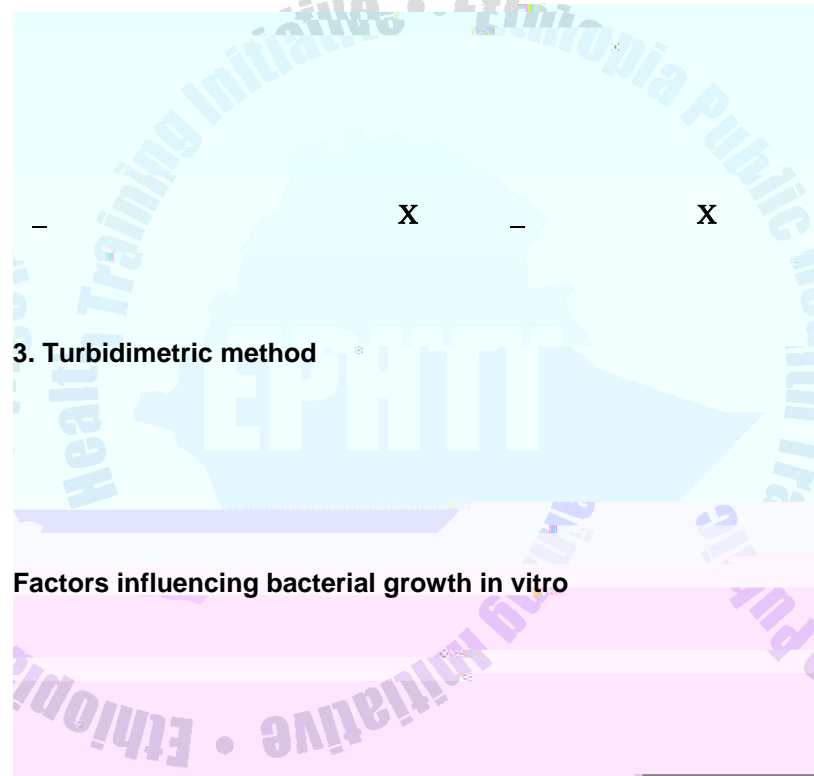
1. Viable plate count

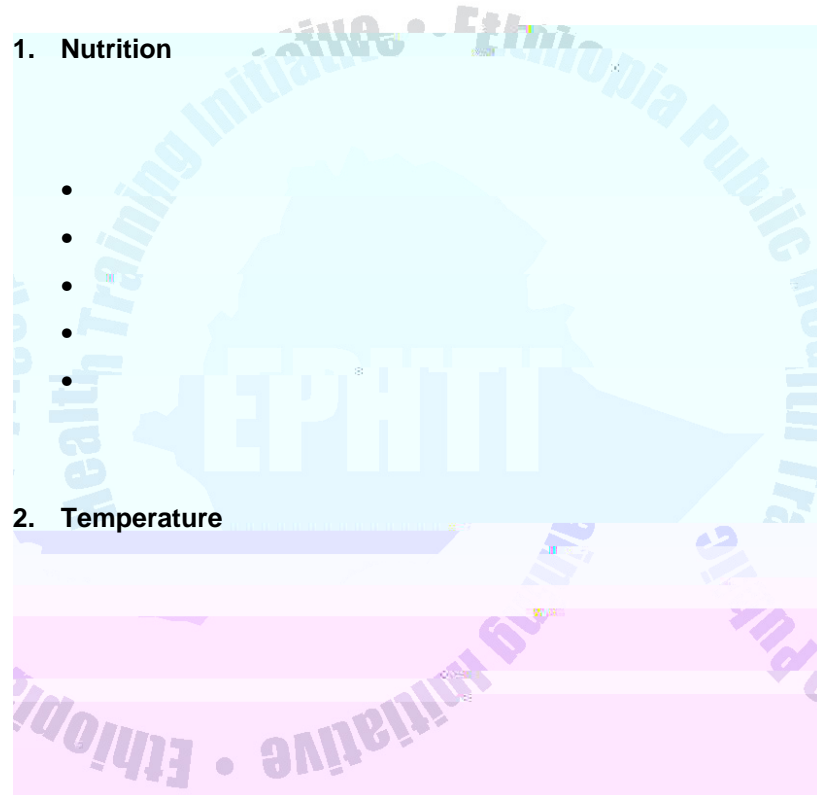
Procedure



2. Direct count

Procedure





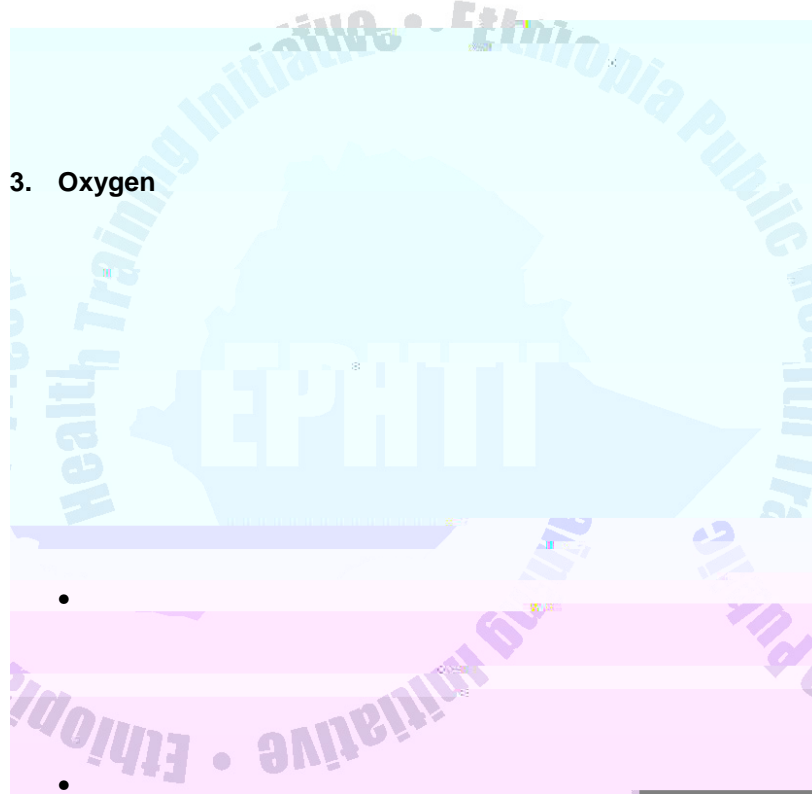
1. Nutrition

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-
-

2. Temperature

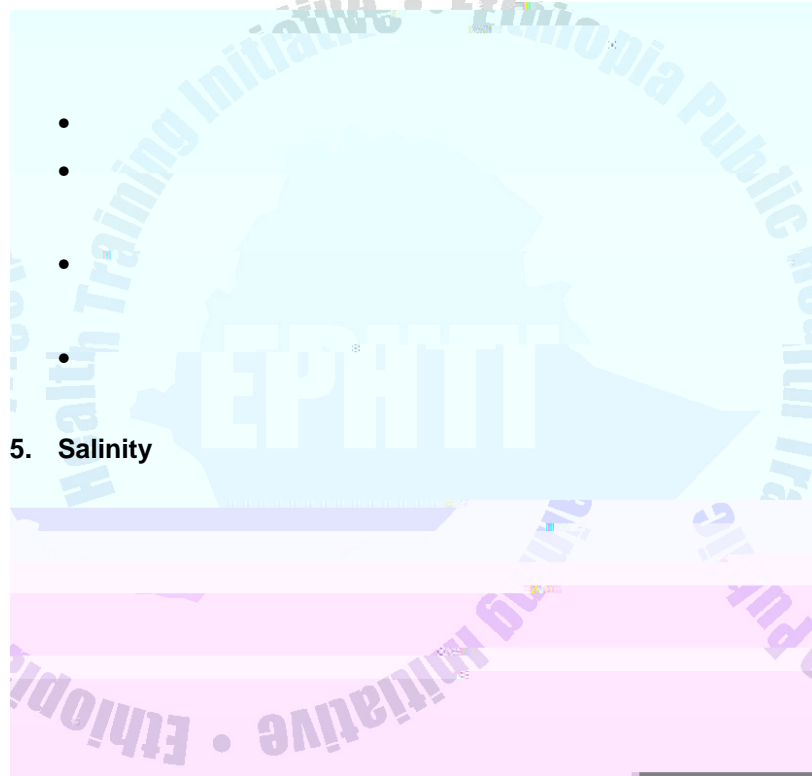
Optimal growth temperature

3. Oxygen

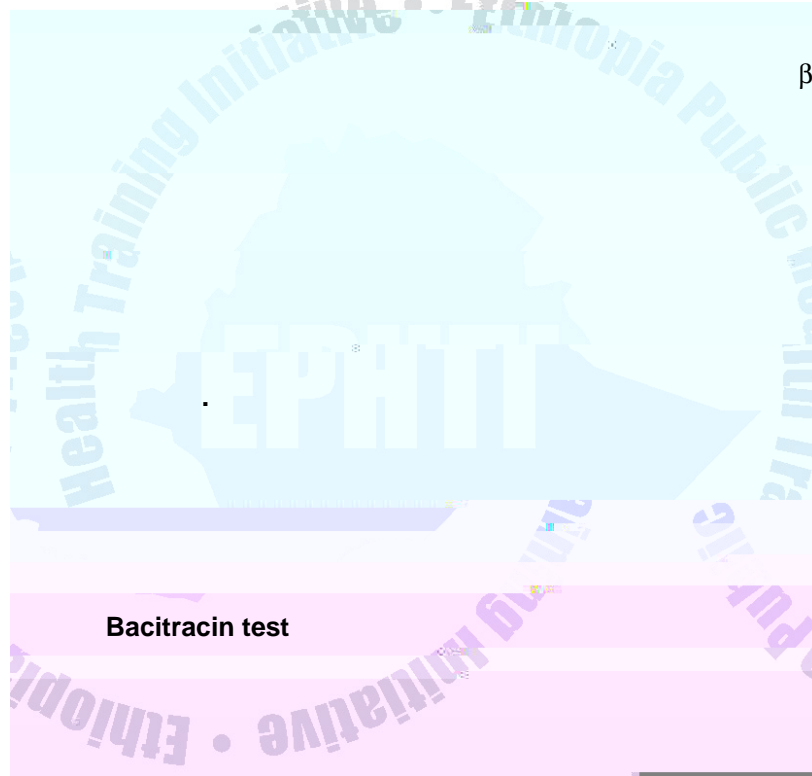


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4. Hydrogen ion concentration



CAMP test



Optochin test



BILE SOLUBILITY TEST

Principle

Required

Method

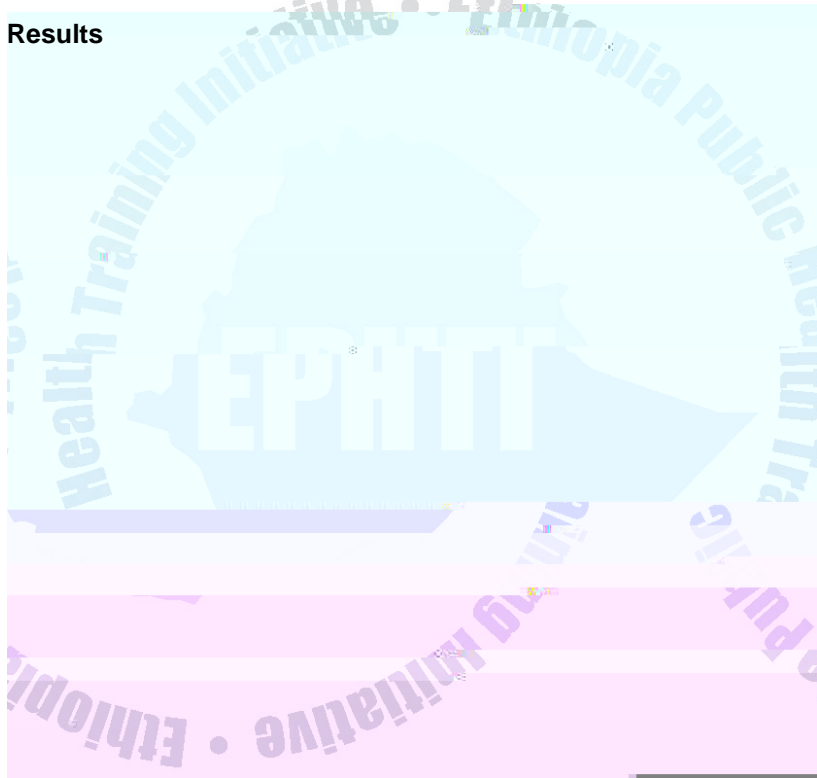
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Results



Principle



Required

Method

-

-

Note

-

Results

Note:

Caution:

Controls

CITRATE UTILIZATION TES

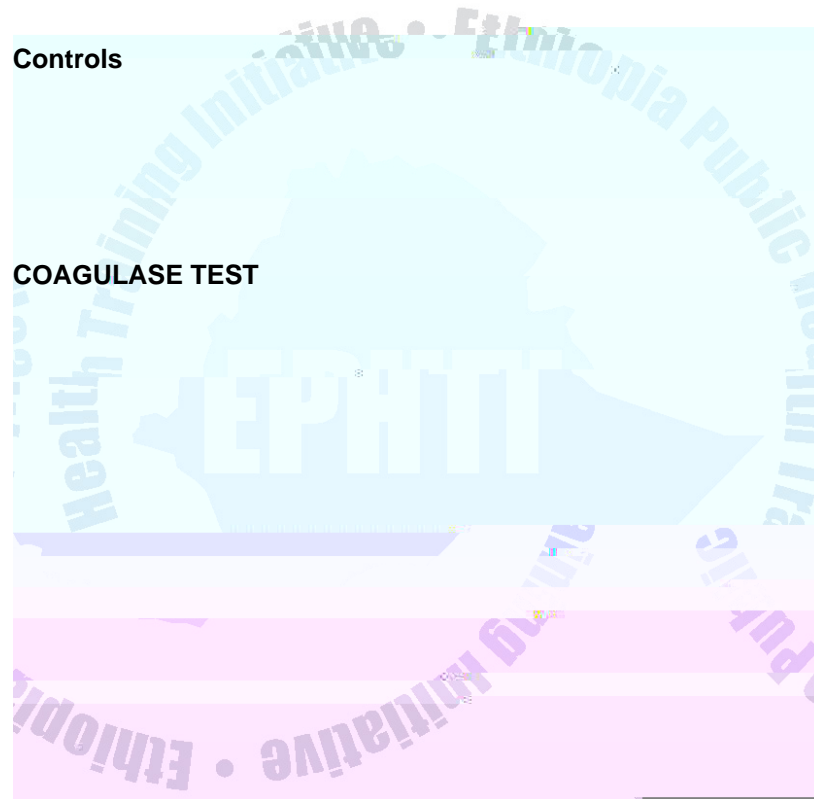
Principle

Required

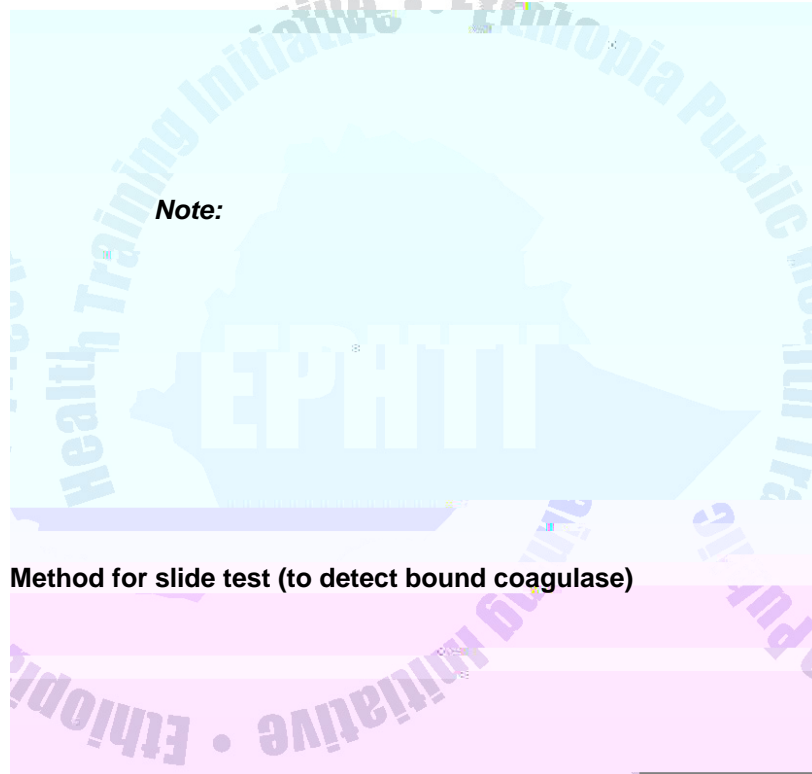
Method

Note:

Results



Required



Note:

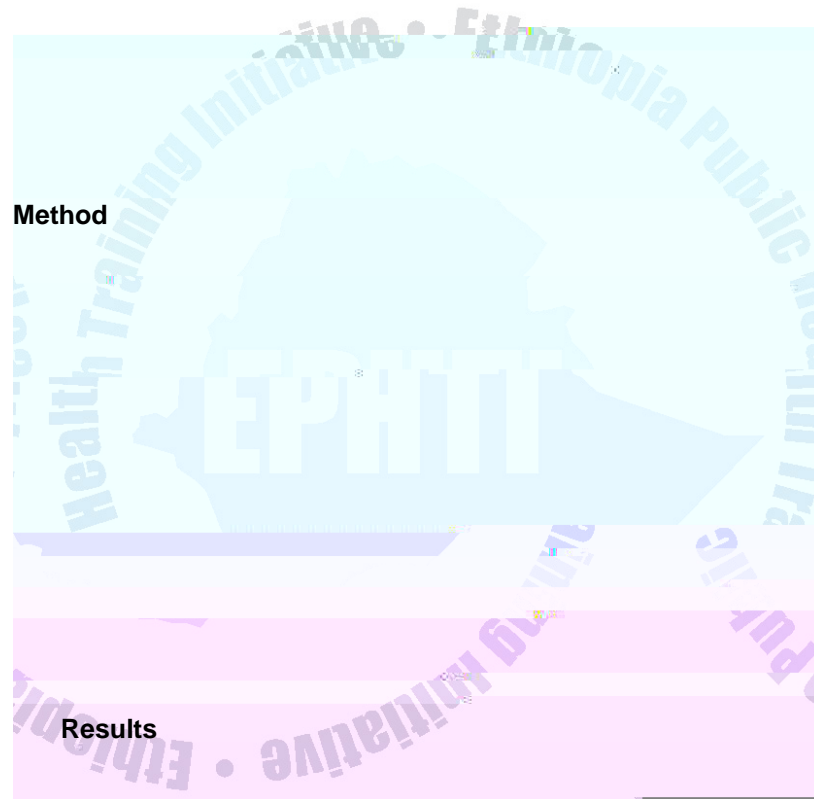


Results

DEOXYRIBONUCLEASE (DNase) TEST

Principle

Required



HYDROGEN SULPHID (H₂S) PRODUCTION

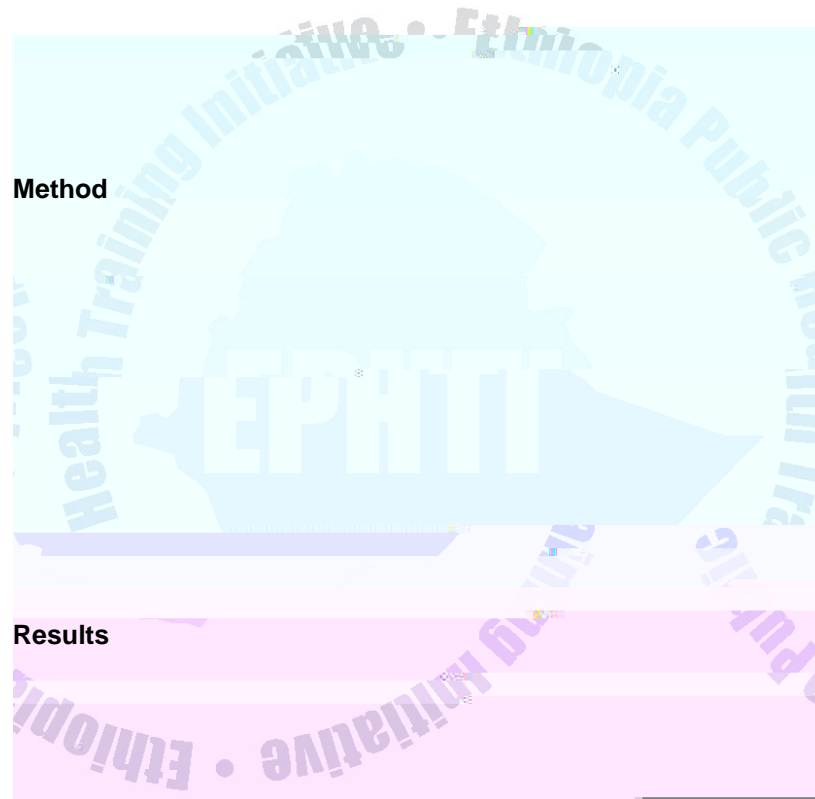
Use of Kligler iron agar (KIA) to detect H₂S

Lead acetate paper test to detect H₂S

Results



Required

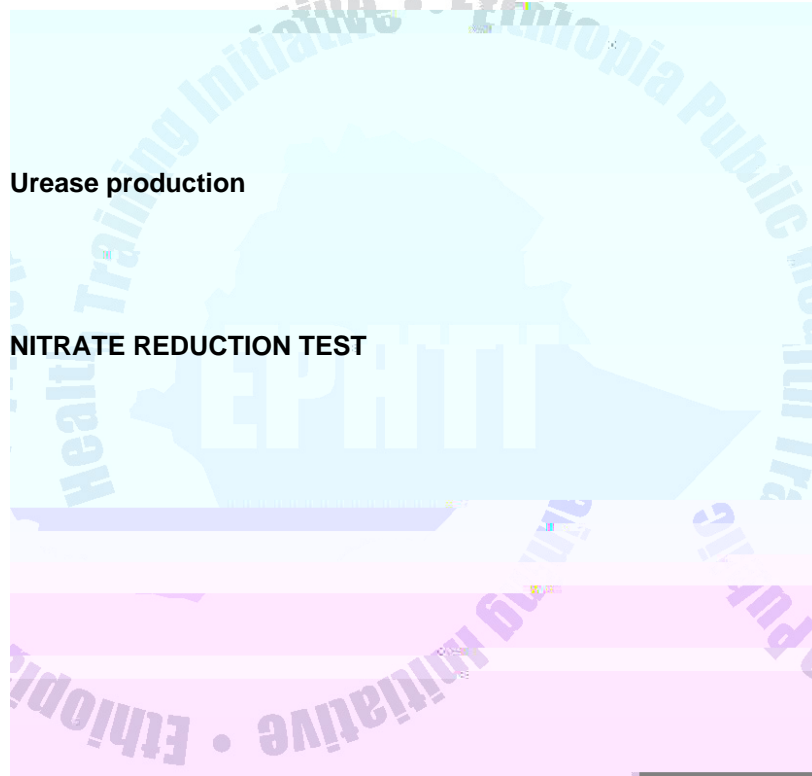


Controls

Motility Test

Urease production

NITRATE REDUCTION TEST





Controls

OXIDASE TEST (Cytochrome Oxidase)

Principle

Important

Required

- Oxidase reagent

Freshly prepared

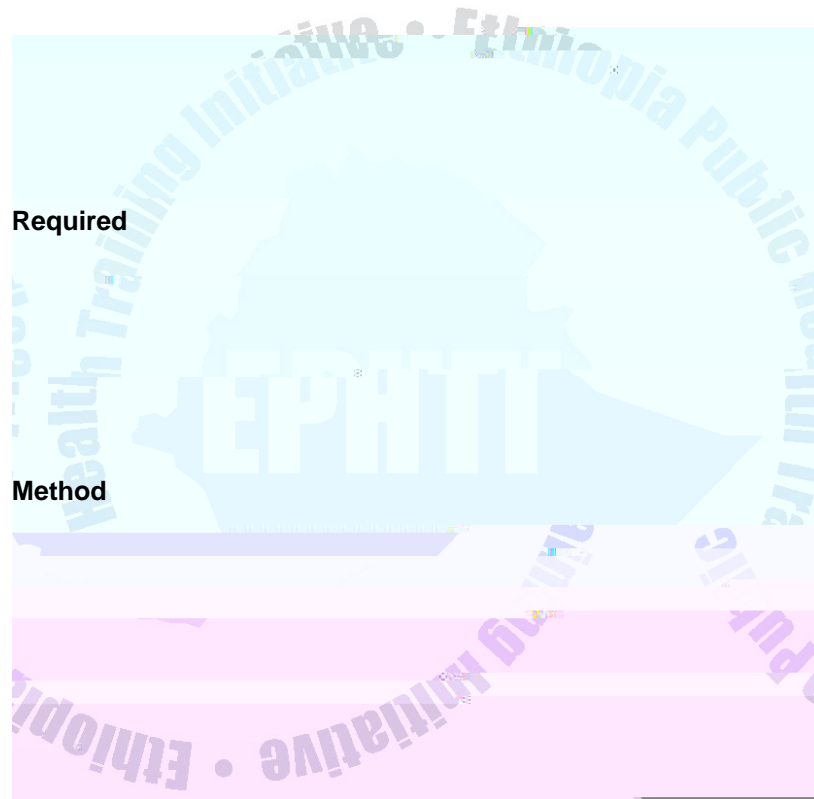
Method

Results

Controls

OXIDATION – FERMENTATION (O-F) TEST

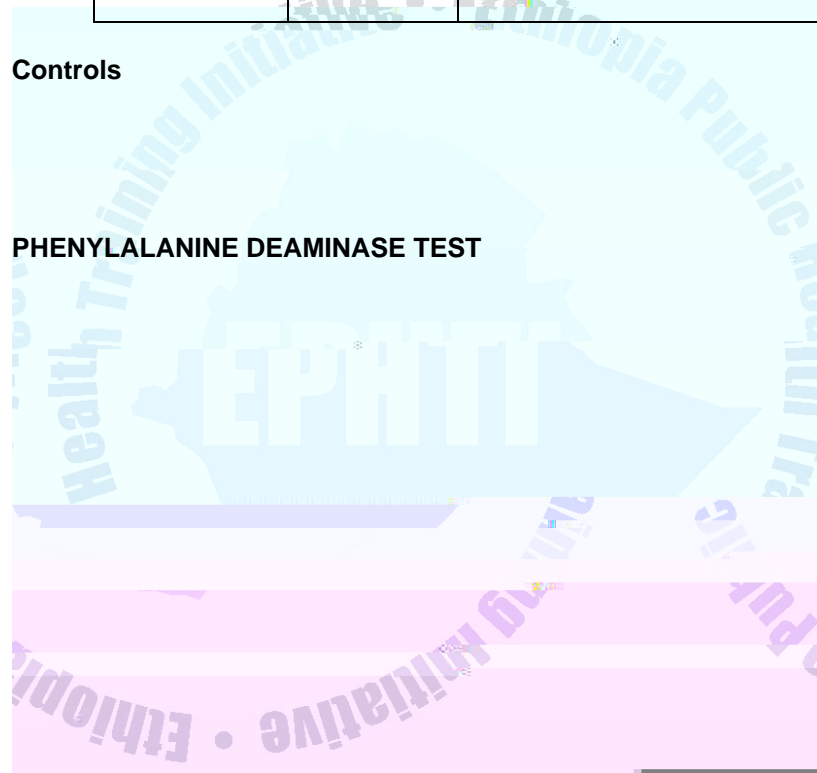
Principle



Results

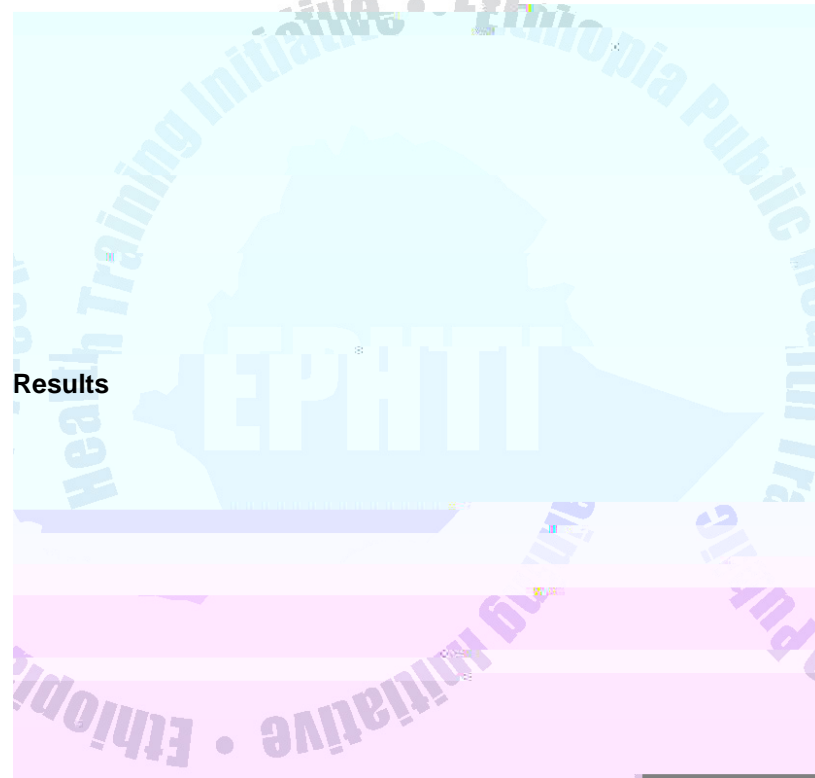
Controls

PHENYLALANINE DEAMINASE TEST



Required

Method



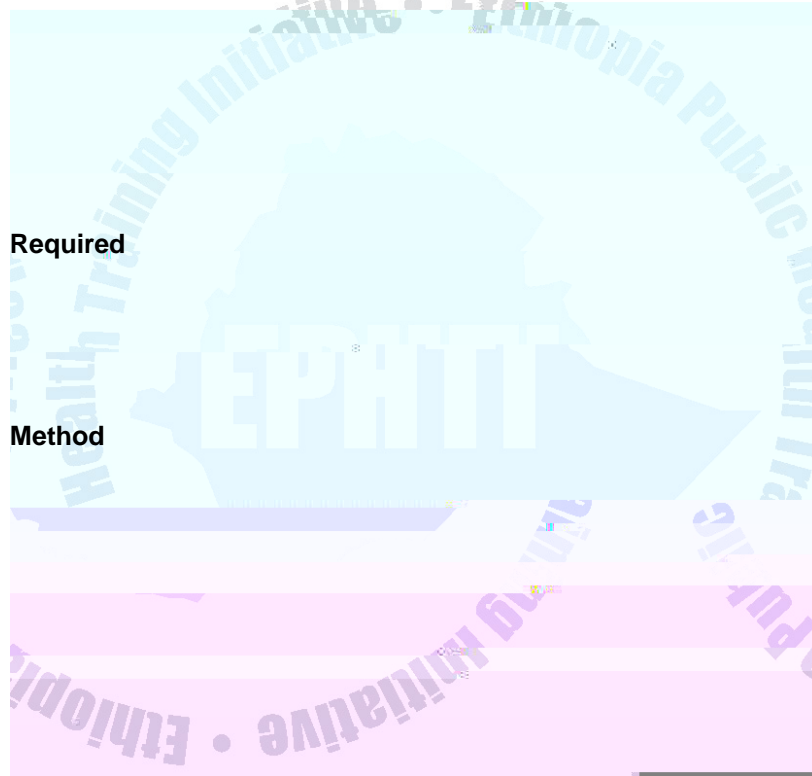
Results

TWEEN 80 HYDROLYSIS TEST

Principle

Required

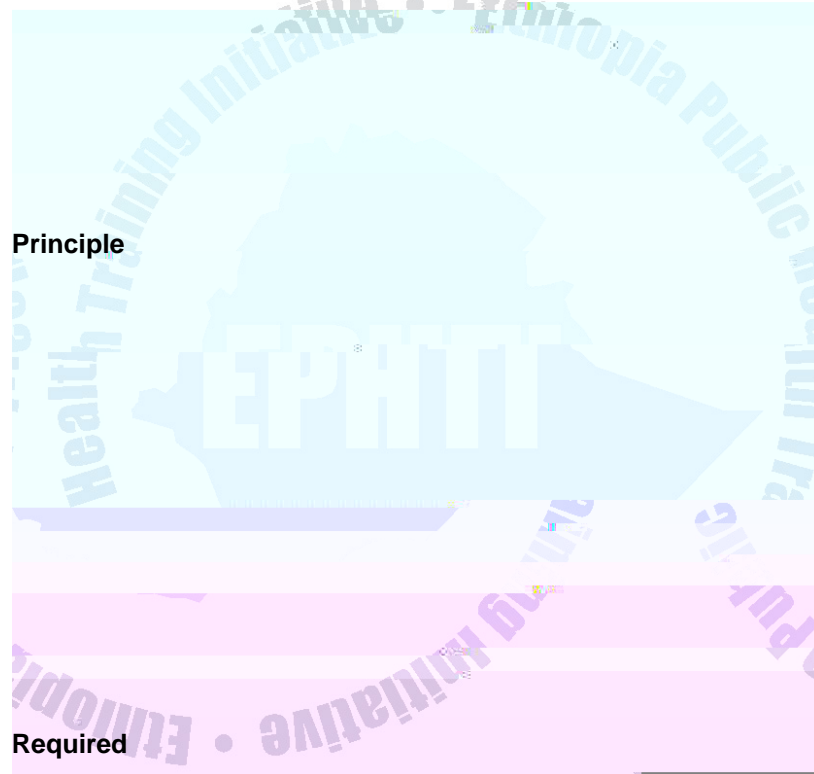
Method



Controls

UREASETEST

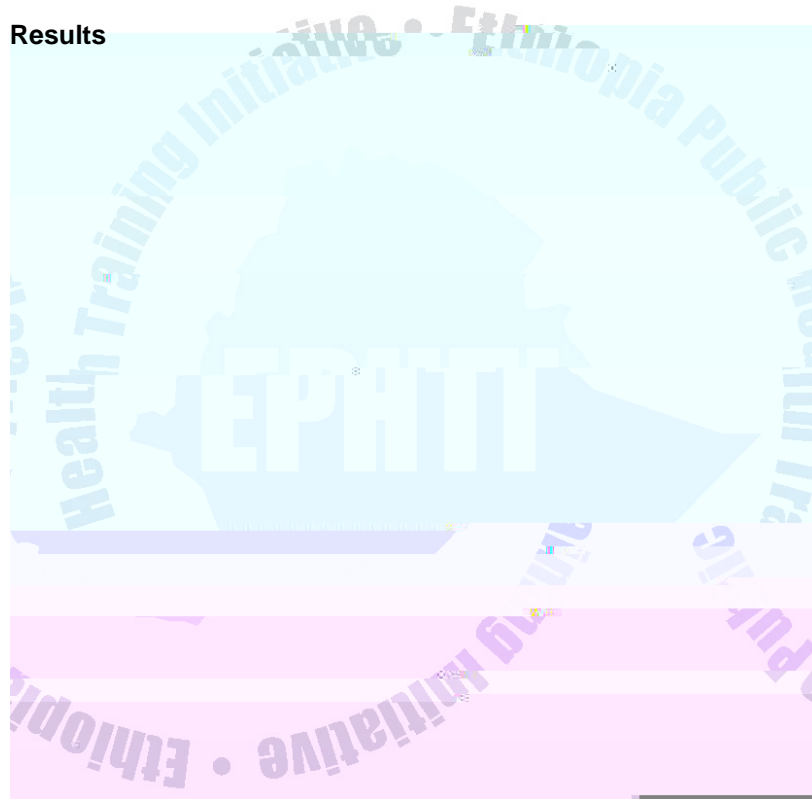
Principle



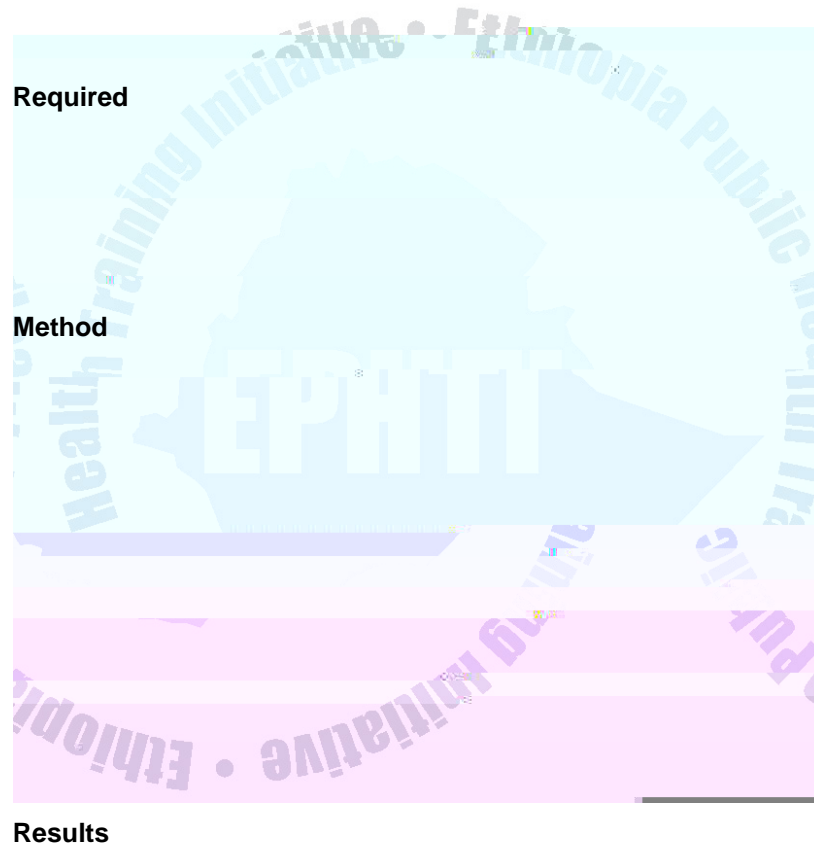
Required

Method

Results



Principle



Controls

1.8. BACTERIAL GENETICS

The bacterial chromosome

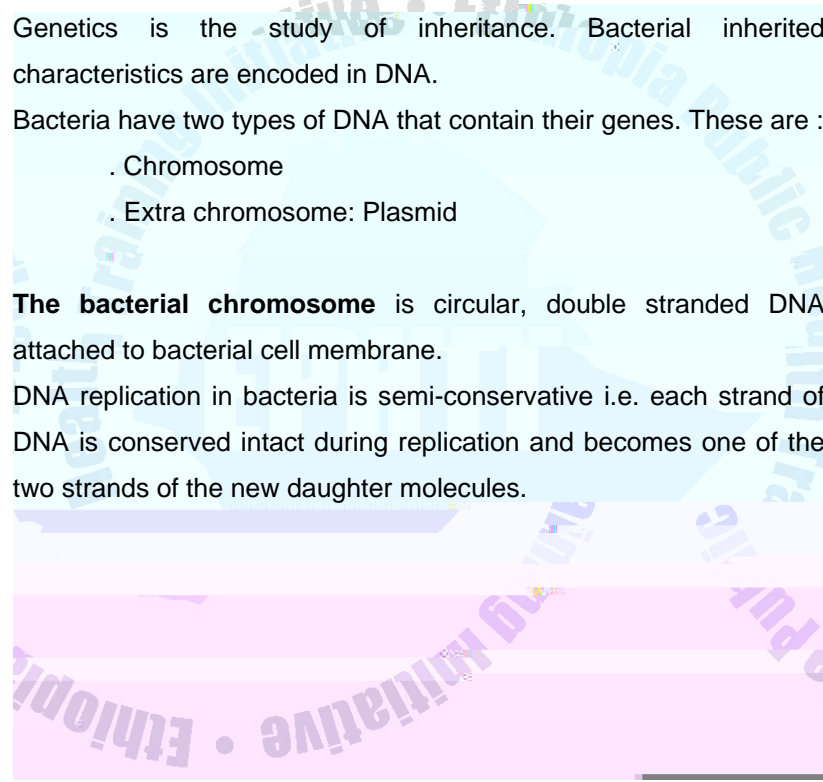
The image contains a diagram of a bacterial chromosome. At the top, there is a circular map of the chromosome with various genes and plasmids indicated by small colored dots. Below this, there is a linear representation of the chromosome, showing a series of colored bands (purple, pink, and light blue) representing different regions or genes. The text 'The bacterial chromosome' is positioned to the left of the diagram. The background of the diagram features a watermark for 'EPHITI' (Ethiopia Public Health Training Initiative) and a map of Ethiopia.



Fig. 1.12

Plasmids

Plasmid types

Genetic variation in Bacteria

Mechanisms:

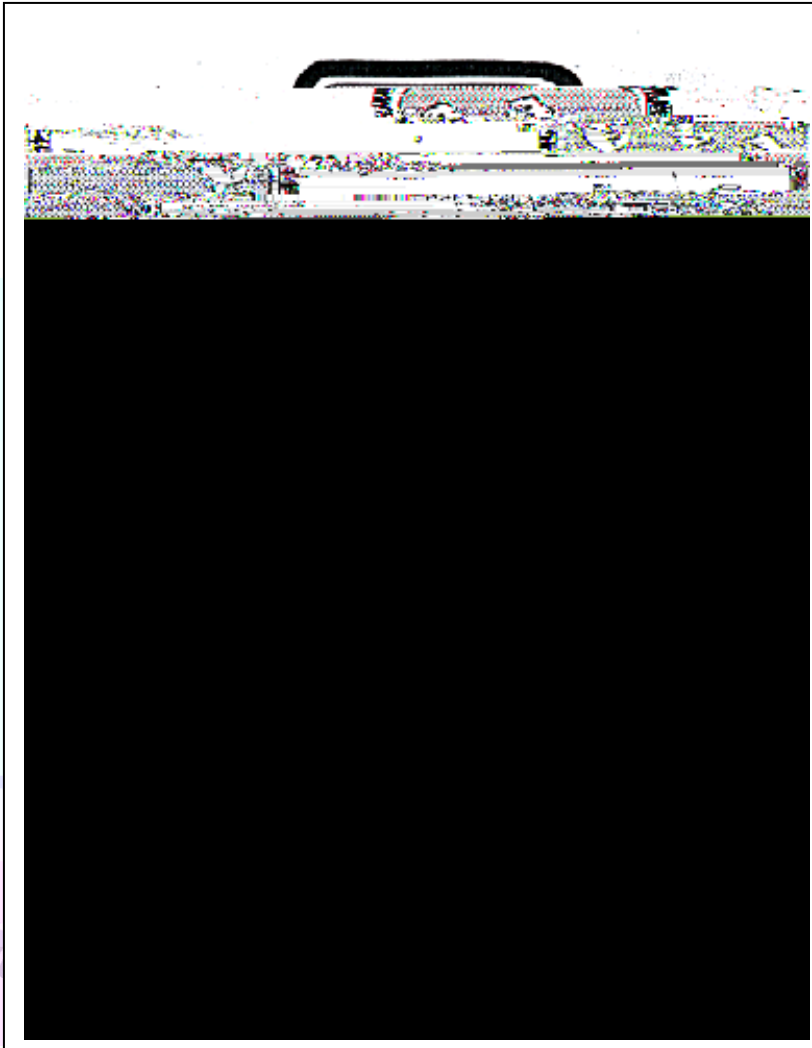
1. Mutation:

Types of mutation

2. Gene transfer

Transformation





Conjugation



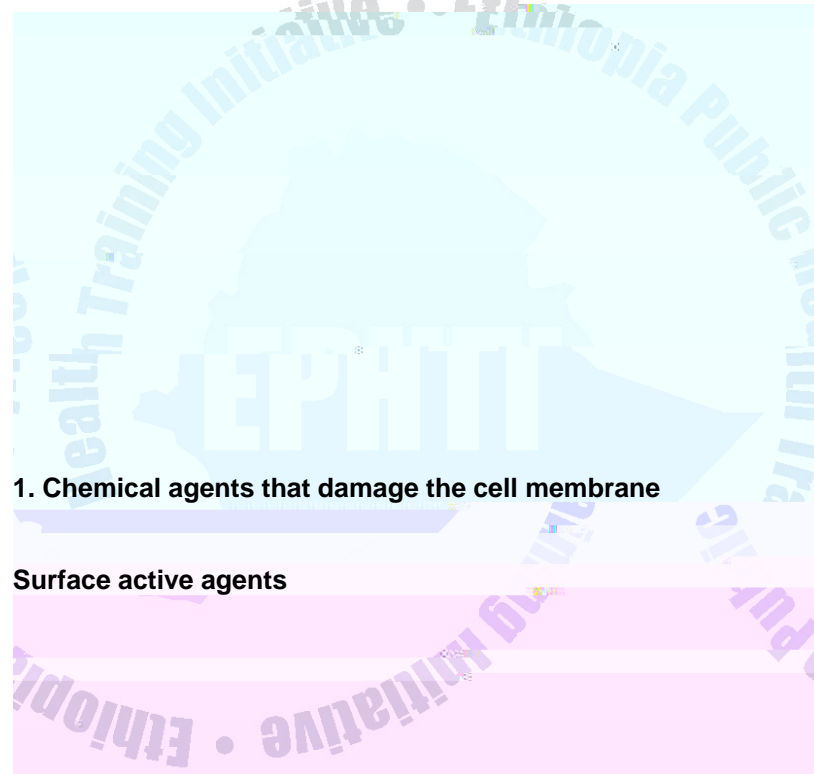
Fig. 1.15



1.9.1. Chemical methods of sterilization and disinfection



Classification of chemical methods of sterilization and disinfection

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1. Chemical agents that damage the cell membrane

Surface active agents

-

-



Organic solvents

Chemical agents that denature proteins

Chemical agents that modify functional groups of proteins and nucleic acids

Heavy metals

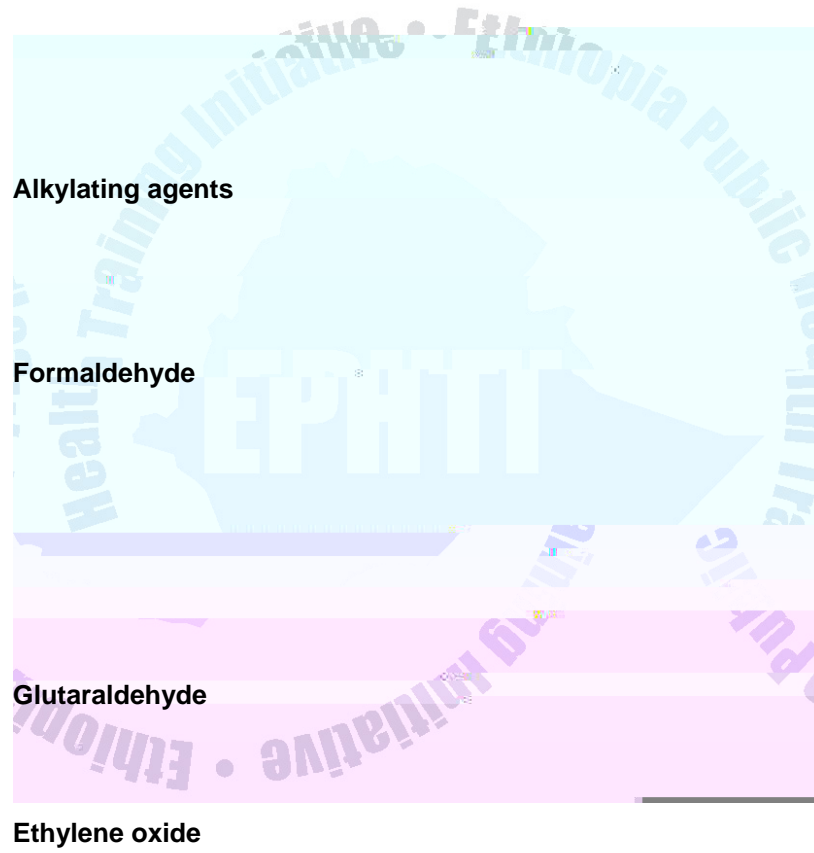
Oxidizing agents

Preparations and uses:

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-
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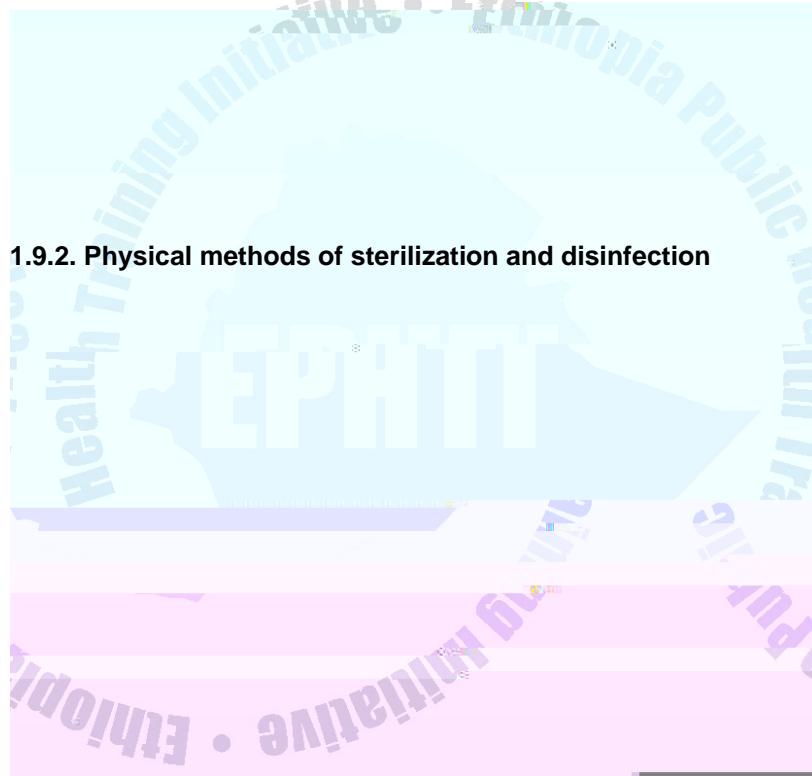
Preparations:

Dyes

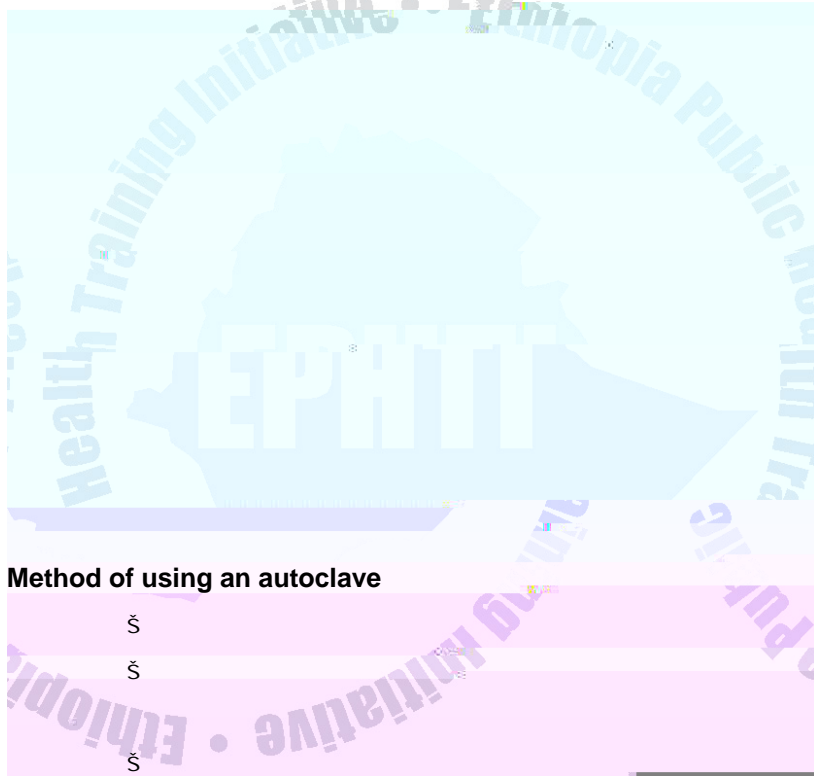


Antiseptic agents:

1.9.2. Physical methods of sterilization and disinfection







Method of using an autoclave

§

§

§

§

§

§

Time –Temperature-Pressure level relationship in moist heat sterilization (autoclaving)

Methods of controlling sterilization



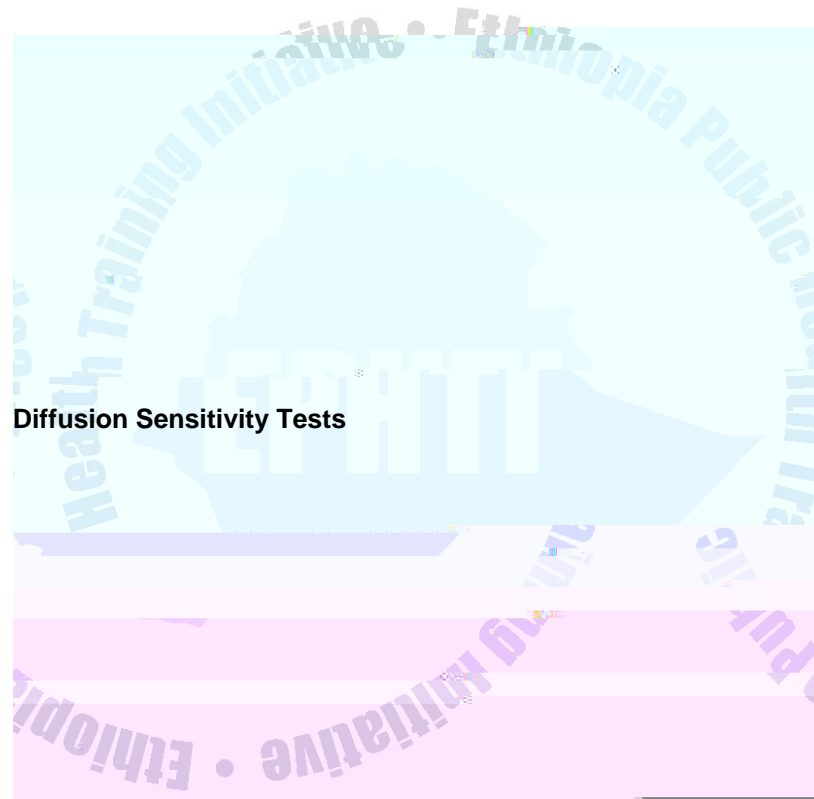
1.9.3. Anti-Microbial agents and Sensitivity Testing

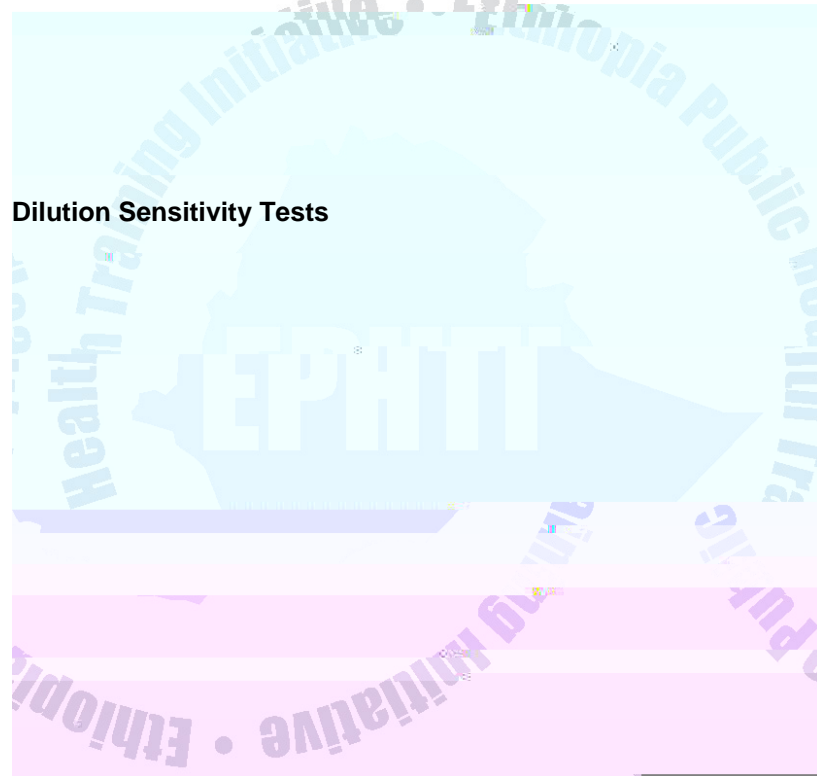
Anti- Microbial drugs

Mechanism of action of anti-microbial drugs



Anti-microbial sensitivity testing





Dilution Sensitivity Tests

Factors affecting anti-microbial activity in vitro





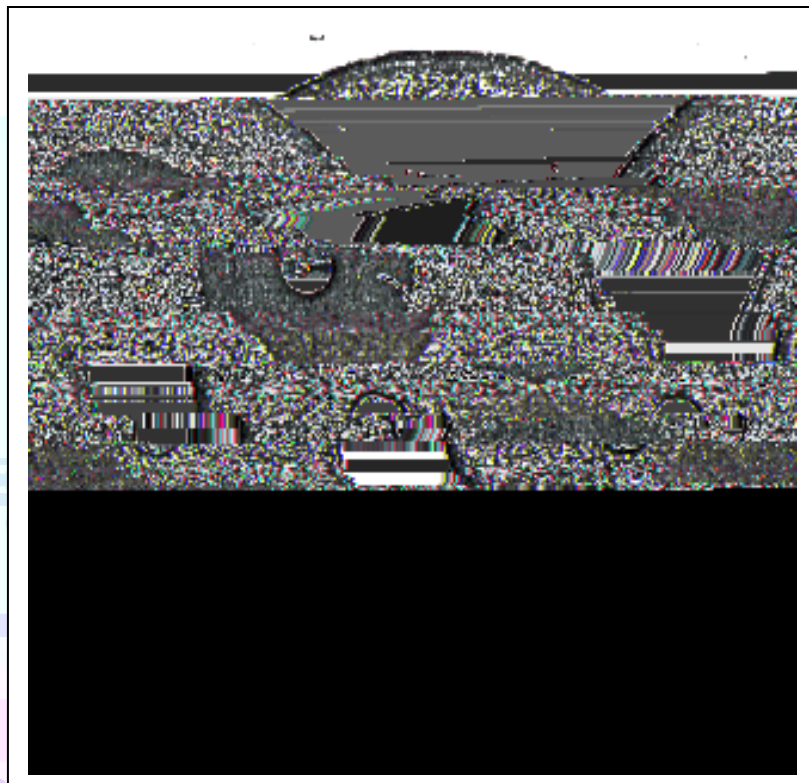


Fig. 1.16

Review Questions



CHAPTER TWO

COLLECTION, TRANSPORT AND EXAMINATION OF SPECIMEN

◆

◆

Type of specimen

:

N.gonorrhoeae

Time of collection

3

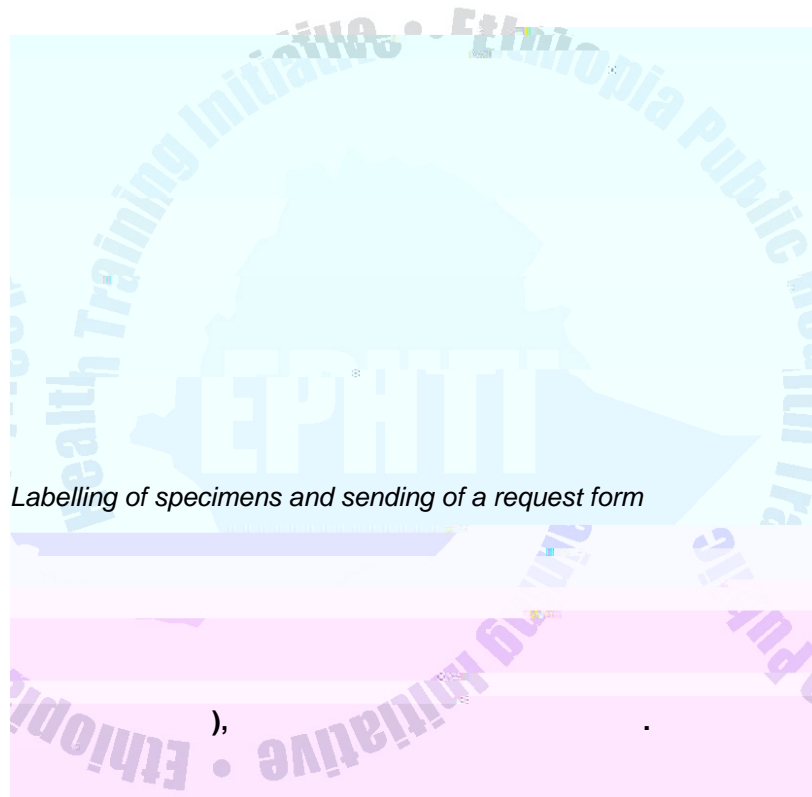
3

3

3

Collection techniques





Labelling of specimens and sending of a request form

Specimen containing dangerous pathogen

3

HIGH RISK

HIGH RISK

- *M. tuberculosis*
- *V. cholerae*
- *S.typhi*
- *anthrax bacilli*

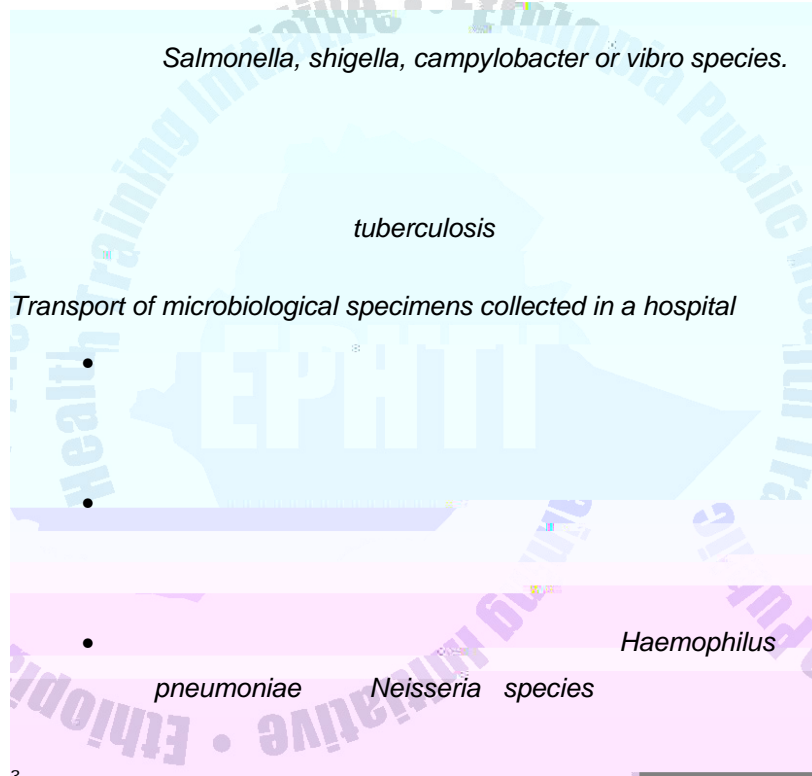
HIGH RISK

Preservatives and transport media for microbiological specimen

3

3

Neisseria gonorrhoeae



Collection, Transport and examination of sputum

Possible pathogens

Gram positive

Streptococcus pneumoniae
Staphylococcus aureus
Streptococcus pyogenes

GRAM NEGATIVE

Haemophilus influenzae
Klebsiella pneumoniae
Pseudomonas aeruginosa
Proteus species
Yersina pestis

Sputum commensals

Gram positive

Staphylococcus aureus
Staphylococcus epidermidis
Streptococcus Viridans
Streptococcus pneumoniae
Enterocci
Diphtheroids
Yeast-like fungi

Gram negative

Neisseria
Branhamella catarrhac's
Haemophilus influenzae
Fusobacteria
Coliforms

In a hospital with a microbiology Laboratory.

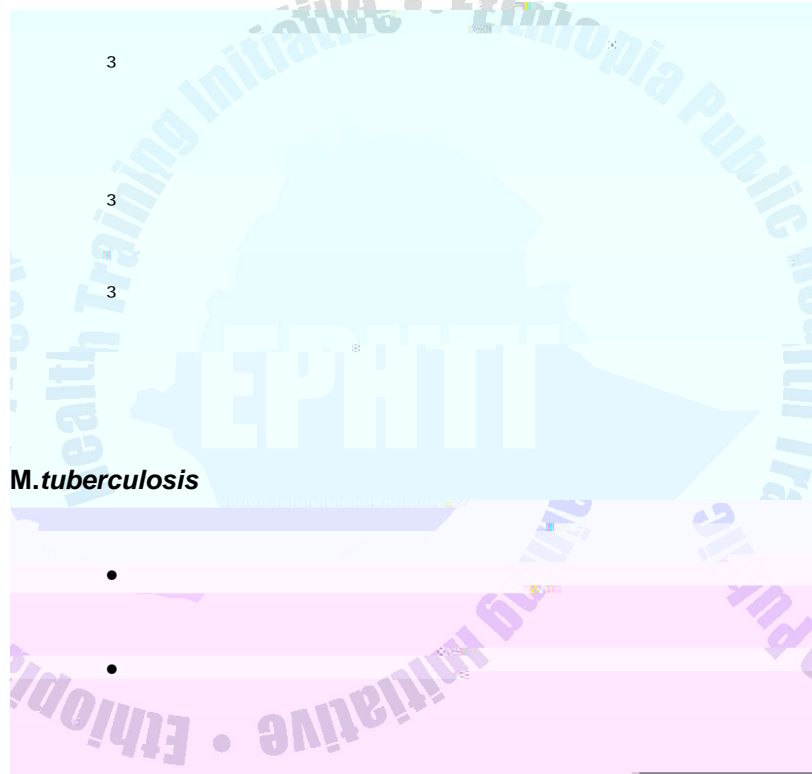


For dispatching to a microbiology laboratory

pneumonia

tuberculosis

Pneumonia and Bronchopneumonia pathogen



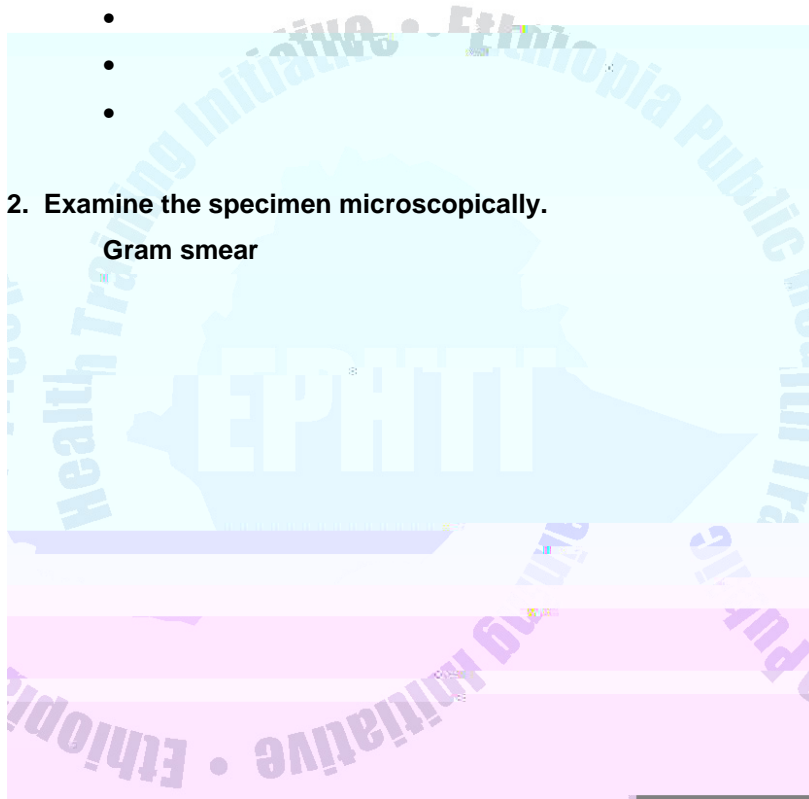
Laboratory examination of sputum

1. Describe the appearance of the specimens

-
-
-
-
-

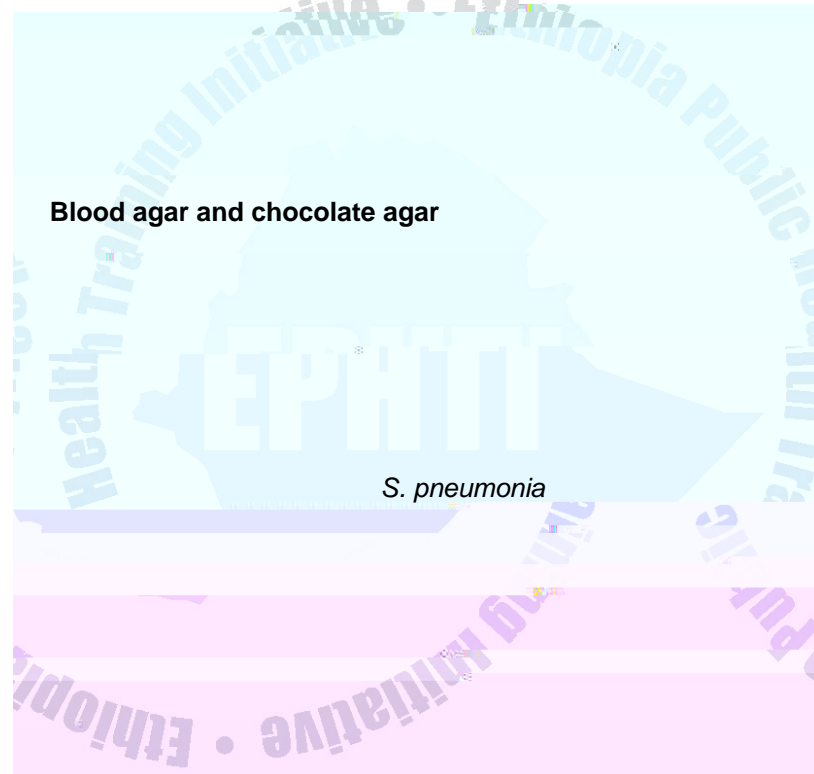
2. Examine the specimen microscopically.

Gram smear



Giemsa smear

3) Culture the specimen



Lowenstein Jensen medium, if pulmonary tubercles is suspected.

4. Examine and report the culture

- S. pneumonia*
- S. aureus*
- H. influenzae*
- K. pneumoniae*
- Pse. Aeruginosa*

Examin

TUBERCULOSIS

Collection and Transport of Throat and Mouth swabs

-

In a hospital with a microbiology laboratory



•

Dispatching to the microbiology laboratory

Laboratory Examination of throat and month swabs

1. culture the specimen

Blood agar

S. pyogenen.



-

-

-

Additional

Albert stained smear

-

-

3. Examine and report the cultures.

Blood agar culture

-

-

Reporting of the throat swab cultures:

-

-

Additional:

Modified Tinsdale medium (MTM) and tellurite blood agar (TBA) Cultures.

-

-

Sabouraud agar culture **Look for candida albicans**

Collection transport and examination of Nasopharyngeal aspirates and Nasal swabs

Nasopharyngeal Aspirates and perinasal swabs

<u>Gram positive</u>	<u>Gram negative</u>
<i>Streptococcus pneumoniae</i>	<i>Haemophilus influenzae</i>
<i>Corynebacterium diphtheriae</i>	<i>Neisseria meningitidis</i> (carriers) <i>Bordetella pertussis</i> <i>Bordetella parapertussis</i> <i>Klebsiella species</i>
Also <i>M. leprae</i>	
Viruses	
Anterior Nasal Swabs Possible pathogens	

•

Gram positive

Gram negative

Nasopharyngeal Aspirates, perinasal and Anterior Nasal swabs

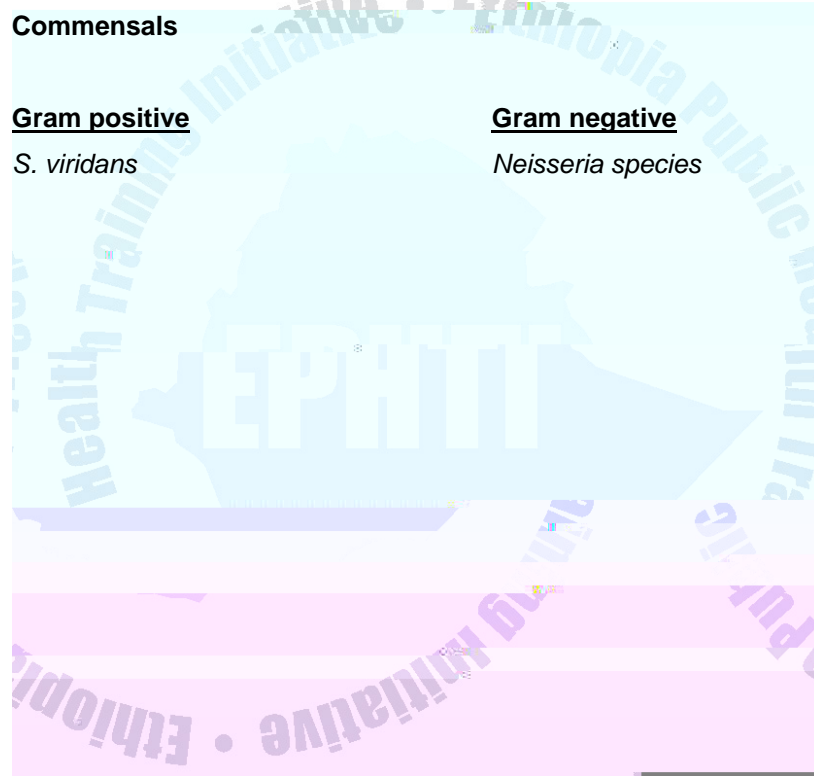
Commensals

Gram positive

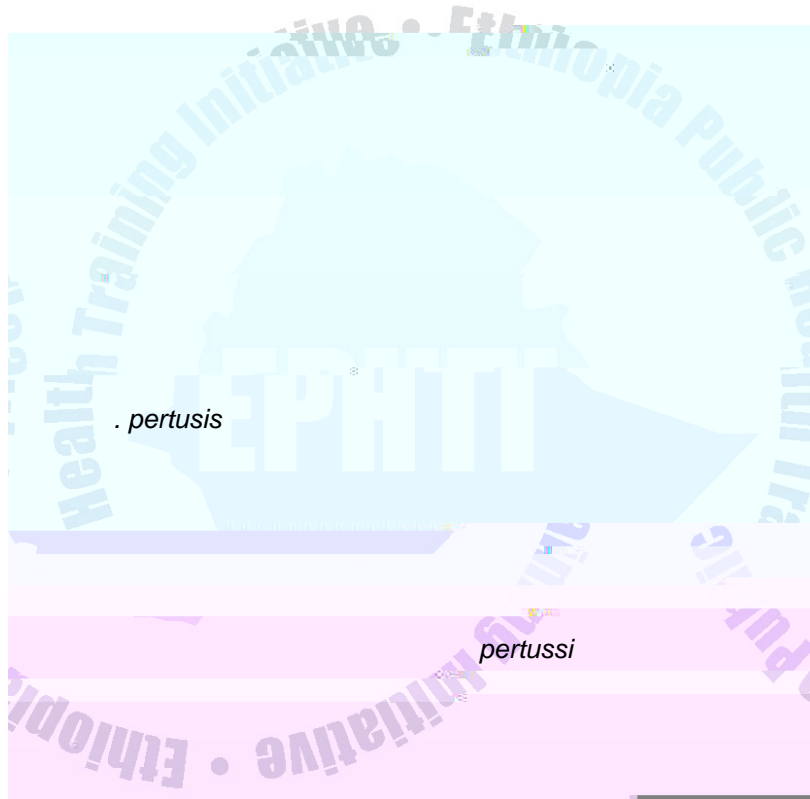
S. viridans

Gram negative

Neisseria species



Pernasal swabs for the culture of *B. pertussis*:



Anterior nasal swabs to detect carriers:

Laboratory examination of upper respiratory tract specimen

1. culture the specimen

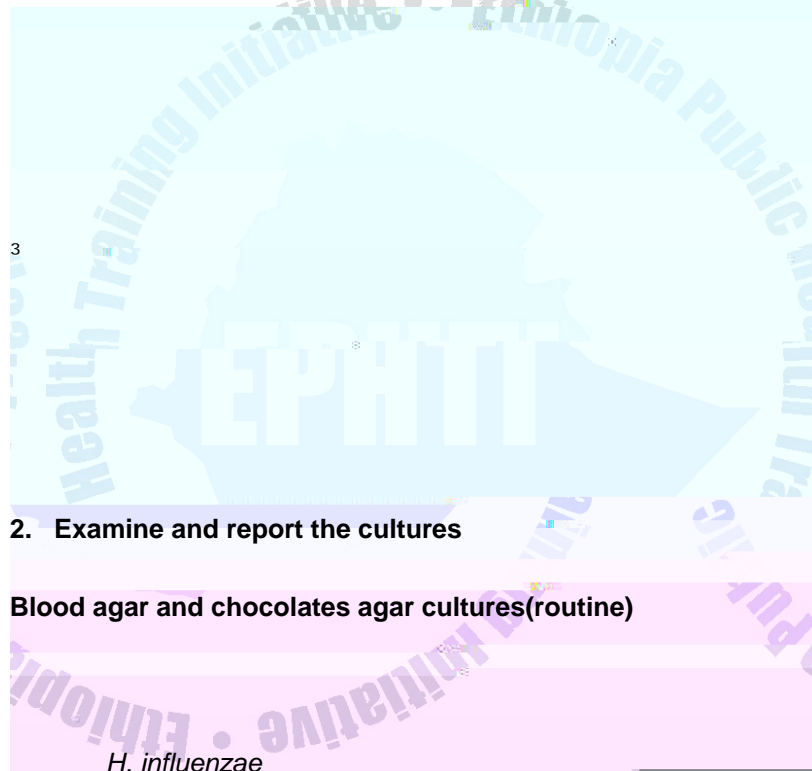
Blood agar and chocolate agar

- *S. pneumoniae*, *H. influenzae*, *N. meningitidis*, *S. aureus*

Additional

Charcoal Cephalexin blood agar if whooping cough is suspected

Culture of swab received in bordetella transport medium:



Neissena mengitdis

S. aureus

S. pyogenes (Group A)

Charcoal ceplea lecin blood agar (CCBA) culture

berdetela

B. pertussis *B. parapertussis*

Collection, Transport and examination of Ear Discharges

Possible pathogens

Gram positive

S. aureus

S. pyogenes

Other beta-haemolytic streptococci

S. pneumoniae

Gram negative

P. aeruginosa

H. influenzae

Klebsiella specia

proteus species

E.coli and other coliforms

Bacteriodes species

Fungi

3

S. aureus

S. pyogenes

P. aeruginos.



Laboratory examination of Ear Discharges

Blood agar and Macconkey agar

Blood agar (Kanamycin) for anaerobic incubation if the infection is chronic

Sabouraud agar if a fungal infection is suspected

2. Examine the specimen Microscopically

-
-
-

Additional:

Potassium hydroxide preparation if a fungal infection is suspected

Look for:

-
-

Collection, transport and examination of eye specimens

Possible pathogens

Gram positive

Staphylococcus aureus

Streptococcus pneumoniae

Streptococcus agalactiae (Group – B)

Streptococcus Pyogenes (Group – A)

Other B- hemolytic streptococci

Gram negative

Nesseria gonorrhoeae

Haemophilus influenzae

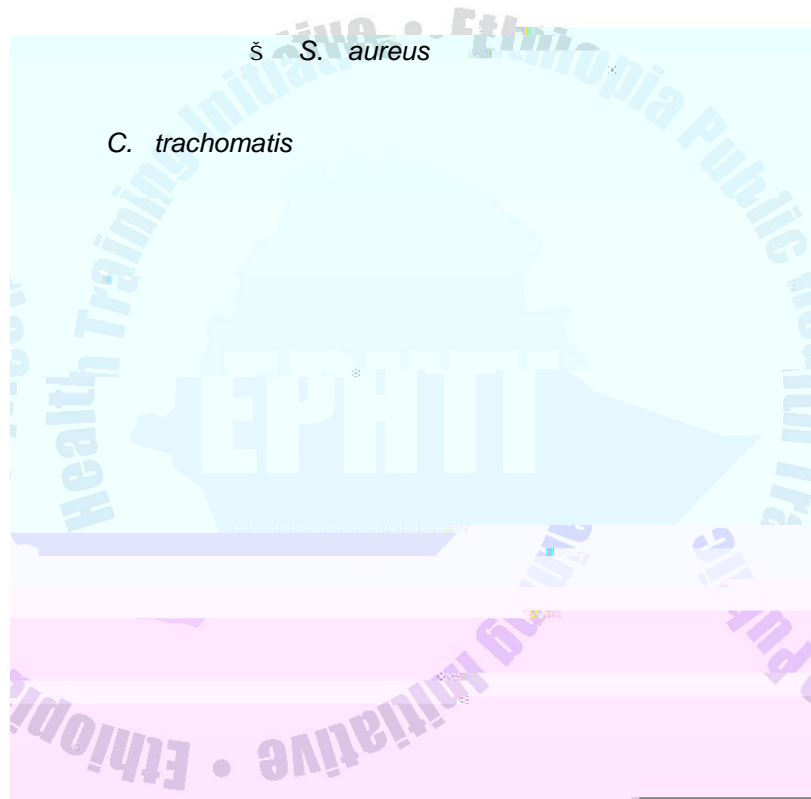
Haemophilus aegyptius

Pseudomonas aeruginosa

Moraxella lacunata

§ *Streptococcus Group B (S.agalactiae*
B-hemolytic streptococci

§ *C. trachomatis*





-
-

Additional

MNYC selective medium if gonococcal conjunctivitis is suspected (infant less than 3 weeks old).

-
-

Loeffler serum slope if Moraxella infection is suspected:

-
-

2. Microscopically examination

Gram smear

-

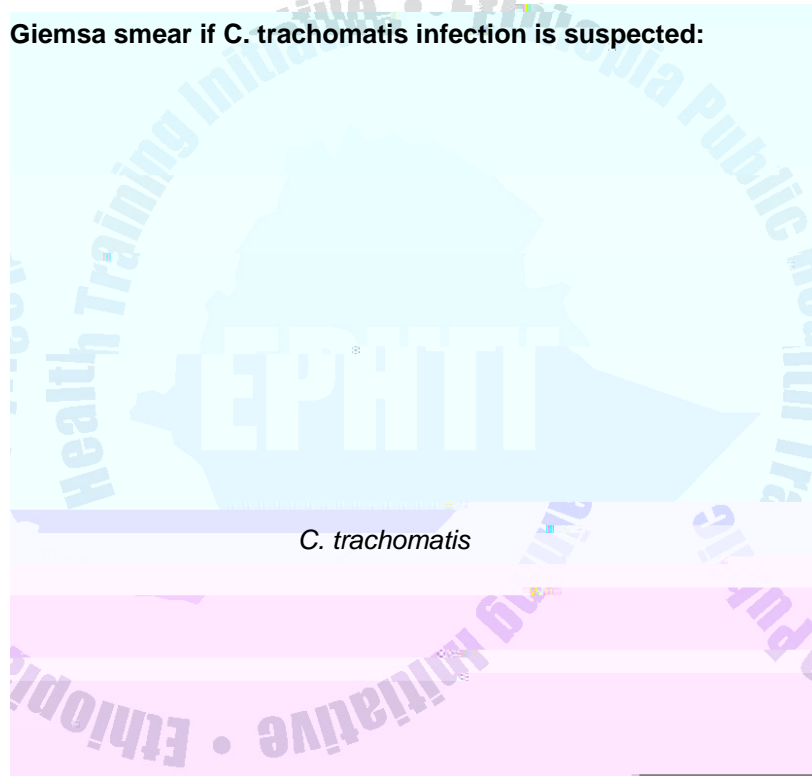
Gonorrhoeae

N.

N.gonorrhoeae

-
- *S.aureus*
-

Giemsa smear if *C. trachomatis* infection is suspected:



J

J

J

Collection, transport and examination of skin specimen

Possible pathogens

Gram positive

S. aureus

S. pyogenes

Enterococci

Anaerobic streptococci

Bacillus anthracis

Corynebacterium ulcerans

Gram negative

Escherichia coli

Proteus

Pseudomonas aeruginosa

Yersinia pestis

Vincent's organisms

M. leprae

M. ulcerans

T. Pertenue

T. Carateum

Commensales

Gram positive

Staphylococci

Micrococci

Anaerobic cocci

Viridans streptococci

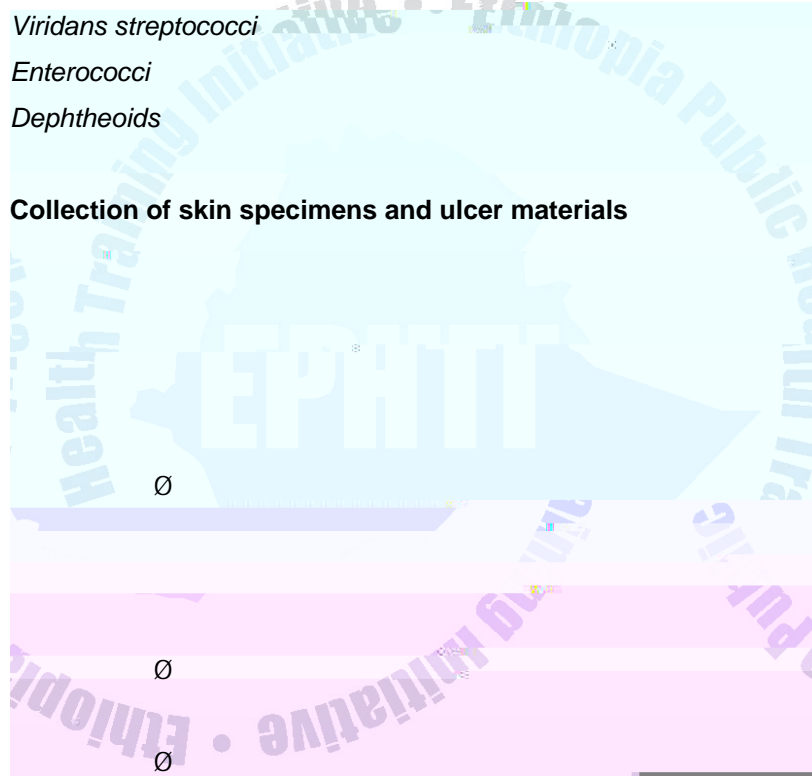
Enterococci

Diphtheroids

Gram negative

Escherichia Coli and

Collection of skin specimens and ulcer materials



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Laboratory examination of skin specimens

1)

Blood agar and MacConkey

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-

Sabourand agar if a fungal infection is suspected

-
-

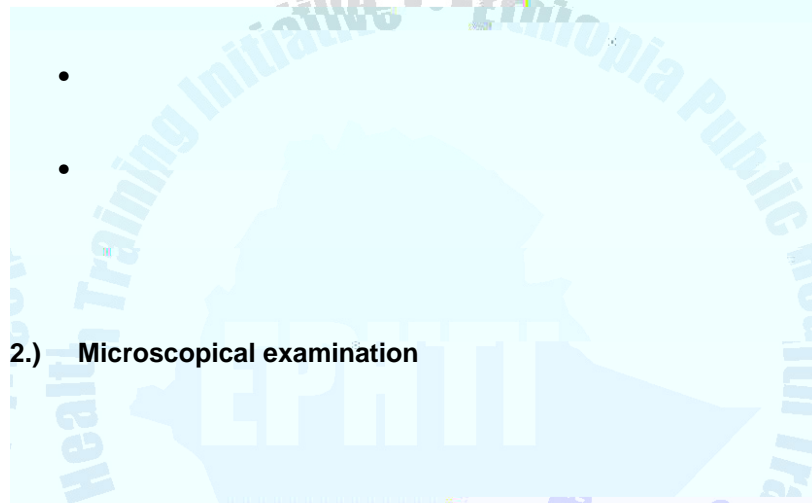
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- J

Lownstein Jensen (LJ) Medium if Buruli ulcer is suspected:

-



2.) Microscopical examination

- *S. aureus*
- *S. pyogenes*
- *p. aeruginosa proteus*
- *speices, E.coli*

J **If tropical ulcer is suspected**

J **If cutaneous anthrax is suspected**

B. anthracis

<i>Enterococci</i>	Lowenstein Jensen
<i>Proteus species</i>	-
<i>Escherichia coli</i>	MTM

-

Collection, Transport and Examination of urogenital specimens
possible pathogens

Urethral swabs

- *N. gonorrhoeae*
- *S. Pyogenes*
- *Ureaplasma urealyticum*
- *Chlamydia trachomatis*
- *Trichomonas vaginalis*

Cervical swabs from non-puerperal women:

- *N. gonorrhoeae*
- *S. pyogenes*
- *Other B.hemolytic streptococci*
- *Chlamydia trachomatis and*
- *herpes simplex virus*

Cervical swabs from women with puerperal sepsis or septic abortion:

- *S. pyogenes*
- *other B – haemolytic*
- *streptococci*

- *anaerobic streptococci*
- *enterococci*
- *S. aureus*
- *clostridium perfringes*
- *Listeria monocytogenes*
- *Bacterioles species*
- *protens species*
- *E. Coli & other coliform*

Vaginal Swabs:

Fluid and pus from genital ulcers

Collection and transport of urogenital specimen

-

Specimen required for diagnosis of gonorrhoea



-
-

Laboratory examination of urogenital specimen

- **MNYC medium**

⇒ *Neisseria gonorrhoeae*

Microscopy

⇒

Suspected gonorrhoeae:

vaginitis:

puerperal sepsis or septic abortion

chanchroid

Additional culture

Blood agar (aerobic and anaerobic), macCokey agar, and cooked meat medium, if puerperal sepsis or septic abortion is suspected

⇒ *H. ducreyi*

Microscopy

Collection, transport and examination of cerebrospinal fluid

S. pneumoniae

S. aureus

S. agalactiae (Group B)

Listeria monocytogenes

Bacillus anthracis

Gram negative

Neisseria meningitidis

H. influenzae type b

Escherichia coli

Pseudomonas aeruginosa

Proteus species

Salmonella species

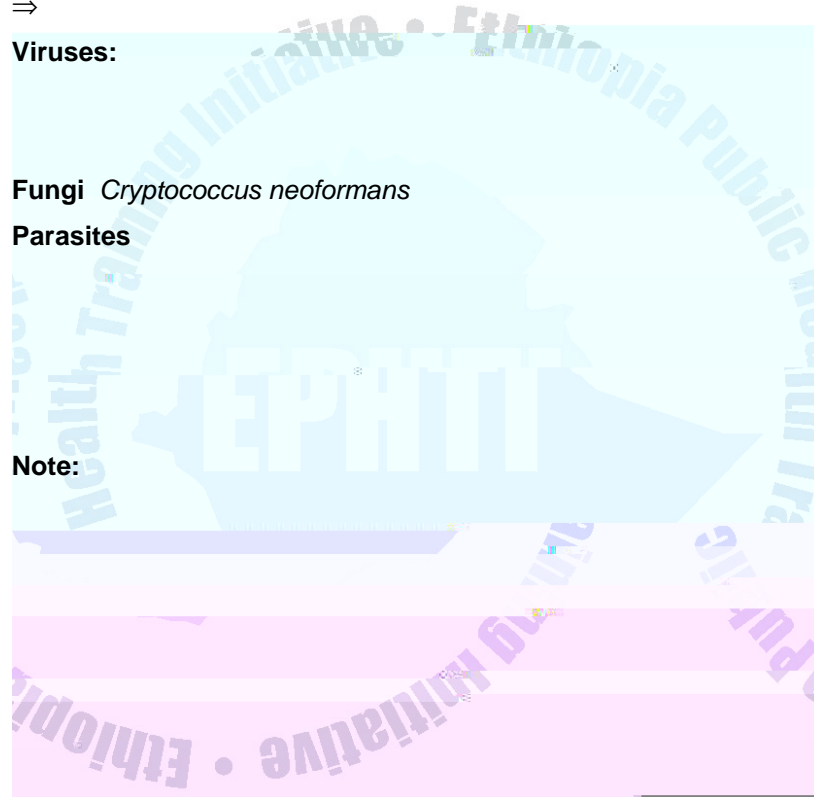
⇒

Viruses:

Fungi *Cryptococcus neoformans*

Parasites

Note:

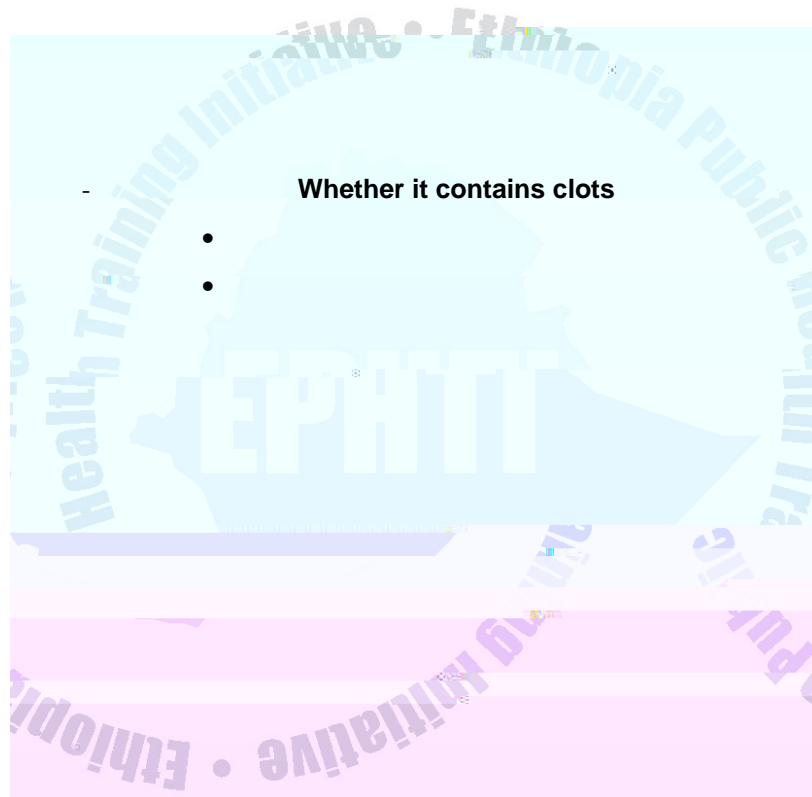




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4. Culture the specimen (sample No 1)

Note

Chocolate (heated blood) agar

- N. meningitides*
- S. Pneumonia*
- H. influenza*

MacConkey and blood agar

- *E.coli* or other coliform
- *S. agalactee* (Group B)
- *Lesteriae monocytognes*
- *S. aureus*


Lowenstein Jensen medium

Sabourand agar

5. Microscopy

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Ziel-Neelsen – smear
M. tuberculosis

Indian ink preparation *cryptococcal meningitis*

Wet preparation

Giemsa stain

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Collection, transport and Examination of Blood AND Bone marrow

Possible pathogens

Gram positive

S. aureus
Viridans streptococci
S. Pneumoniae
S. pyogenes
Enterococci
Anaerobic streptococci
Clostridium prefringes

Gram negative

Salmonella typhi
Other salmonella
Brucella species
H. influenzae
P. aeruginosa
Klebsiella strains
E. coli
Proteus species
Bacterioides species
Neisseria meningitidis
Yersinia pestis

Fungi:

Parasites

Note:



J

Tryptone soya (tryptic soy) diphasic medium

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Liquid:

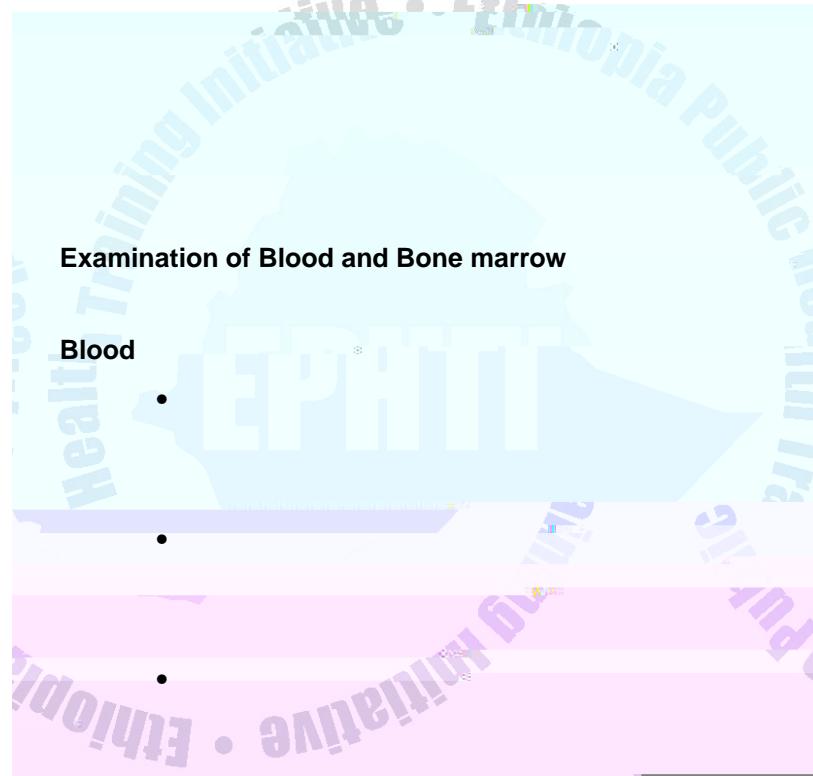
P. Aminobenzoic acid

Ø

Thioglycollate broth

Ø

Ø



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Thioglycollate broth



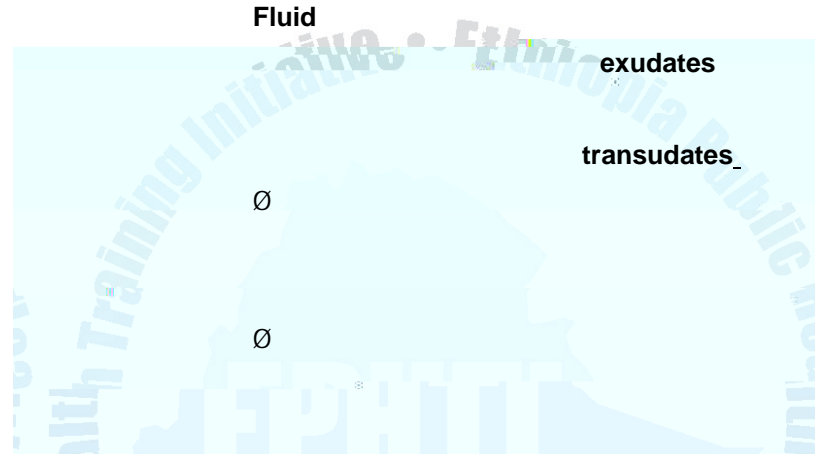
Tryptone soya diphasic medium



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Collection, transport and examination of effusions (synovial, pleural, pericardial, ascitic and hydroceles fluids)

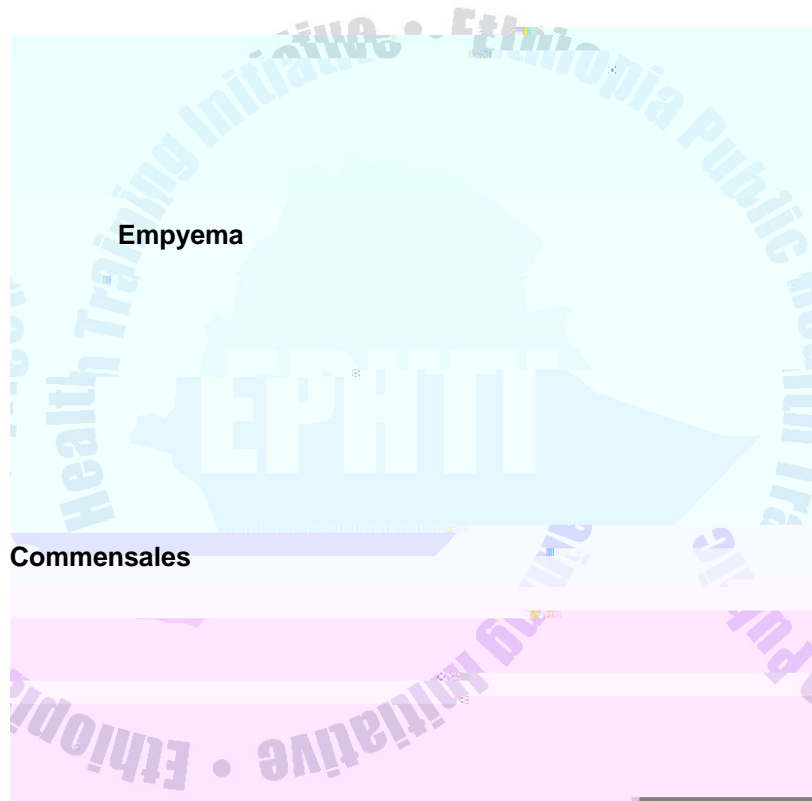
Ø



Fluid	Origin

Synovitis

arthritis.



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CHAPTER THREE

Learning Objective

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GRAM POSITIVE COCCI

2.1.1. GENUS: STAPHYLOCOCCI

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Table 2.1 DIFFERENTIATION OF SPECIES

_____	_____	_____	_____	_____
_____	_____	_____	_____	_____
_____	_____	_____	_____	_____
_____	_____	_____	_____	_____
_____	_____	_____	_____	_____

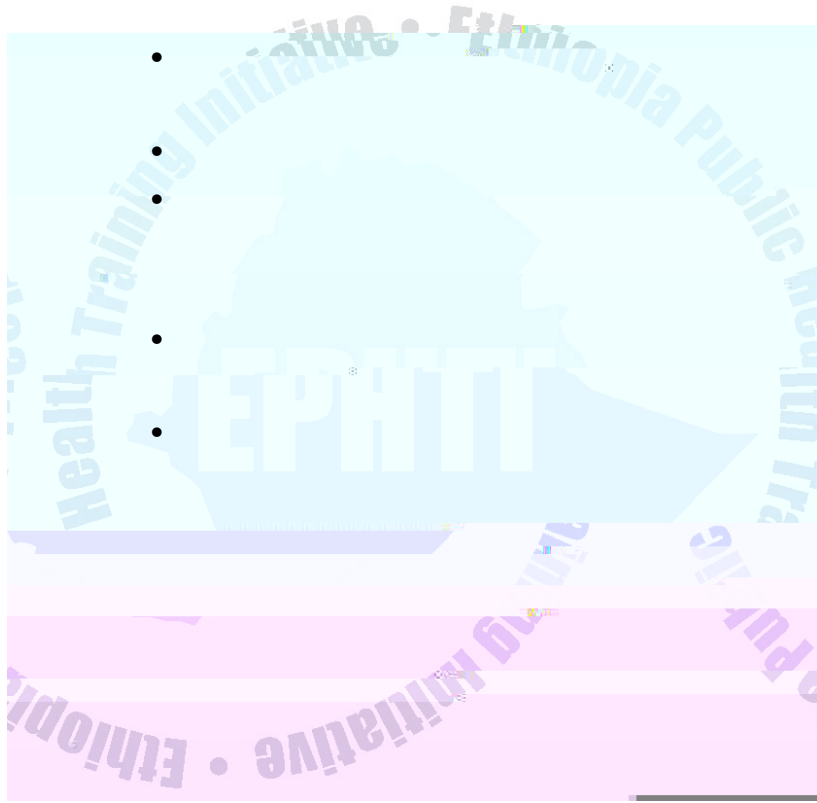
2.1.2. GENUS: STREPTOCOCCI

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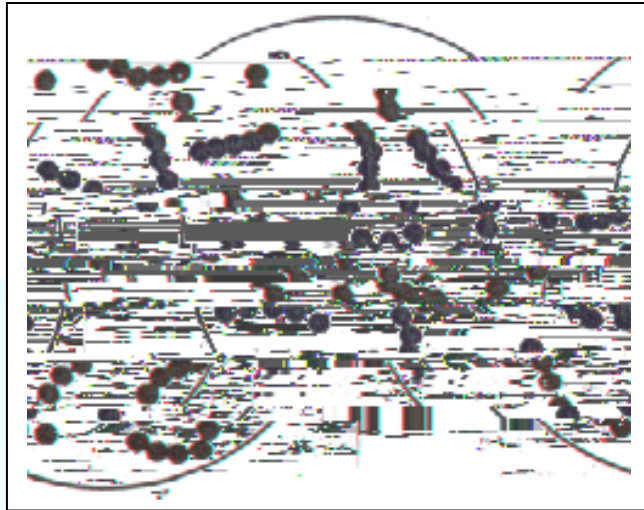
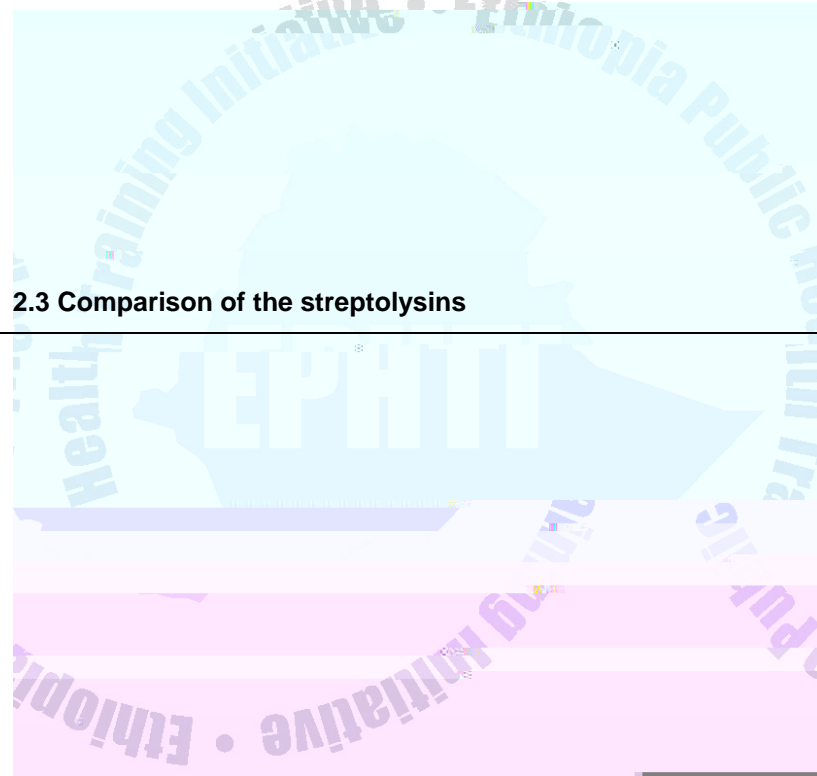


Fig. 3.2

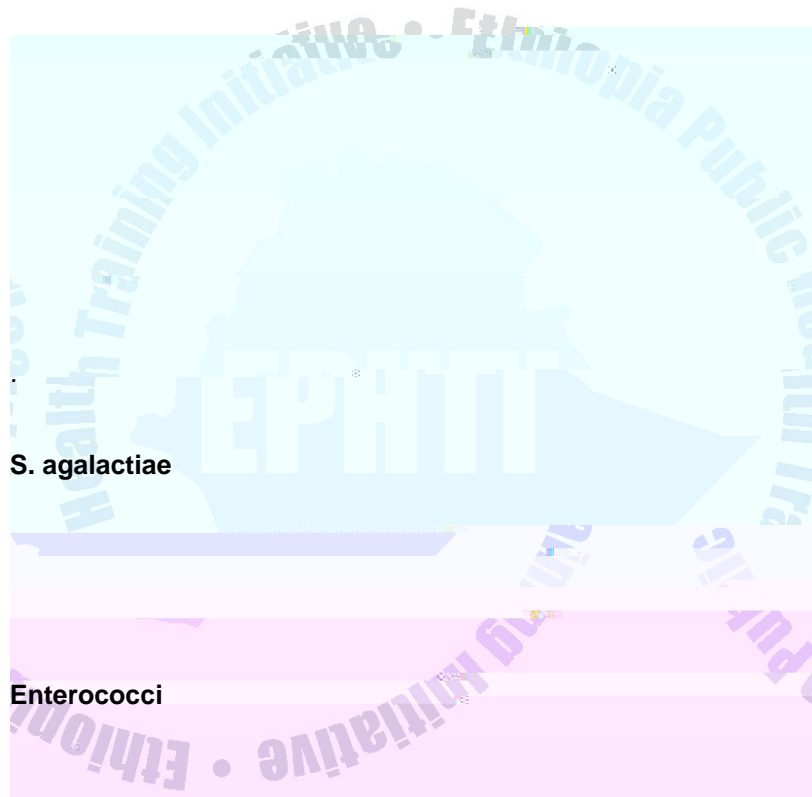
Table 2.2 Hemolytic reaction of streptococci:







2.3 Comparison of the streptolysins



Viridans streptococci

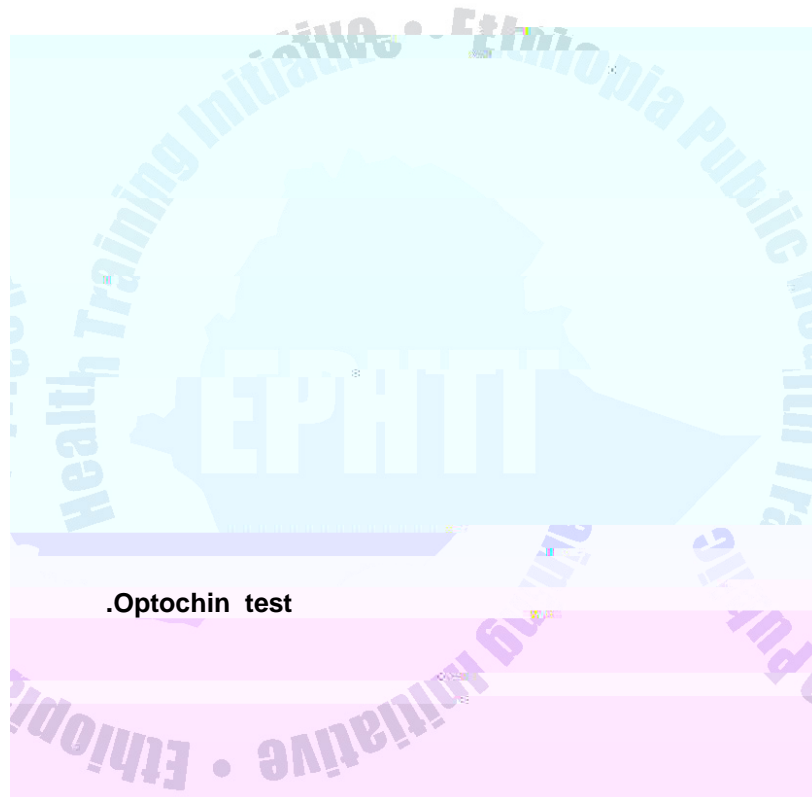


Streptococcus pneumoniae

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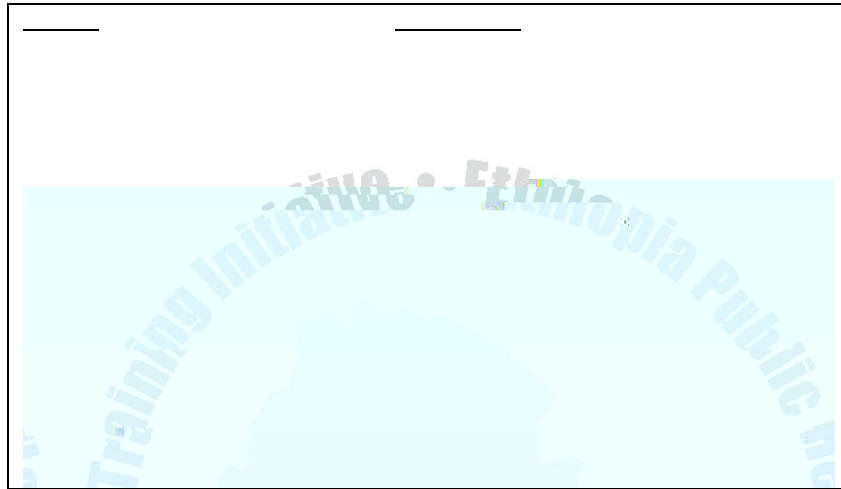






.Optochin test

Table 2.4 Differentiation of streptococcus species



GRAM POSITIVE SPORE FORMING RODS

2.2.1. GENUS: BACILLUS

Characteristics:

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Bacillus anthracis

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- 3. Bacteremic anthrax: presents with clinical features of sepsis**

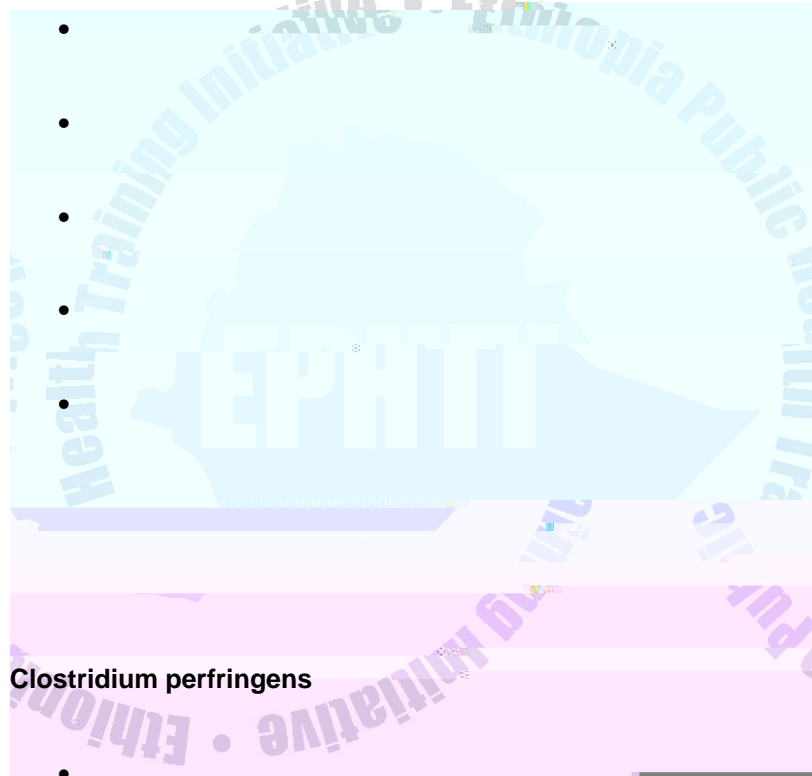




Bacillus cereus



2.2.2. Genus: Clostridium









Clostridium tetani

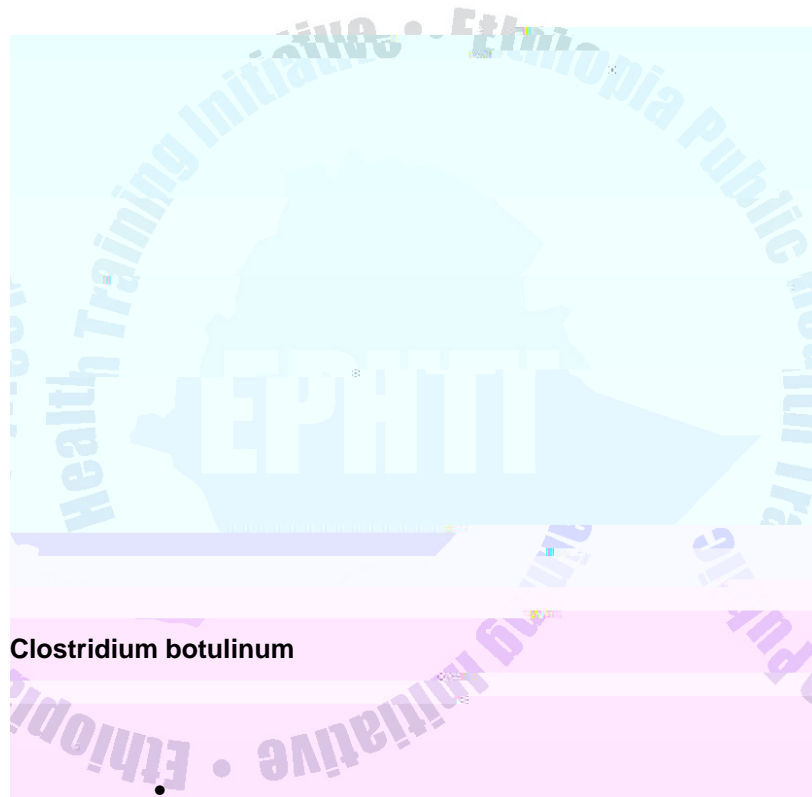
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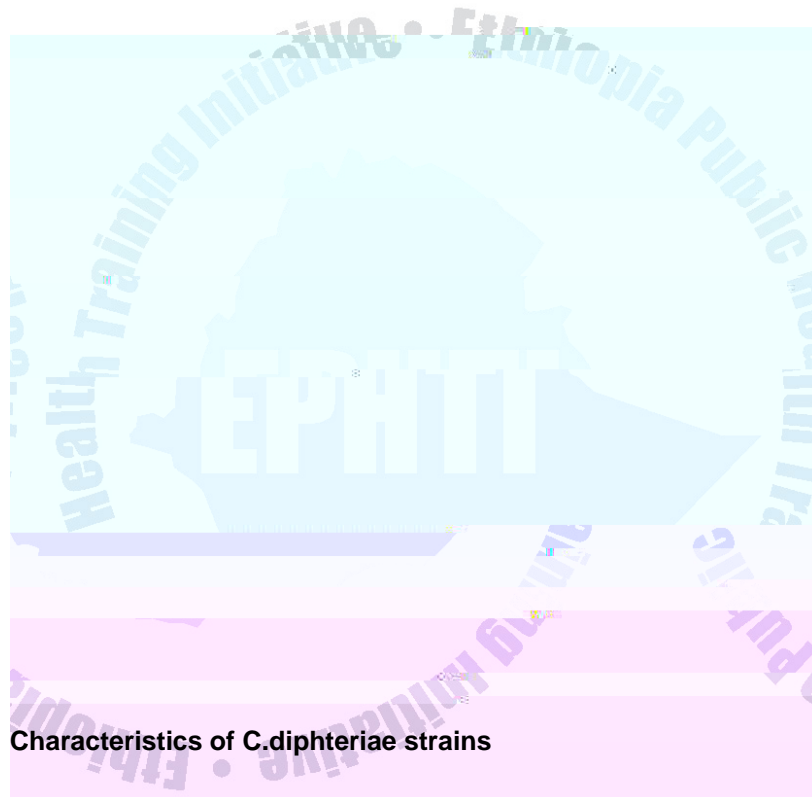






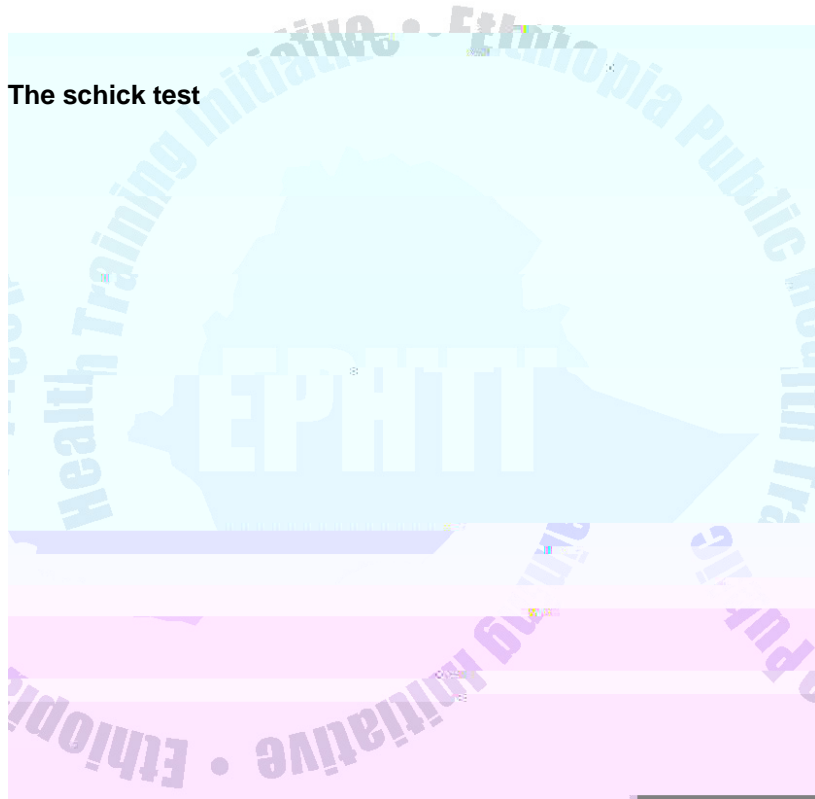








The schick test



GENUS: LISTERIA

L. monocytogens





GENUS : ERYSIPELOTHRIX







Fig. 3.4.







Neisseria meningitidis

-
-



Neisseria meningitidis





2.6. Comparison features of N.gonorrhoea and N.meningitidis

Features	N.gonorrhoea	N.meningitidis

2.3.2. GRAM NEGATIVE COCCOBACILLI

GENUS: HAEMOPHILUS

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-
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H. ducreyii

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-
-
-
-

H. aegyptius

2.3.3. GENUS: BORDETELLA

C





GENUS BRUCELLA

-

-











2.4.1. ENTEROBACTERIACEAE



GENUS:ESCHERICHIA

Escherichia coli





Genus: Klebsiella

K.pneumoniae



GENUS: CITROBACTER

GENUS: SALMONELLA





Variables Enteric fever Septicemia Enterocolitis



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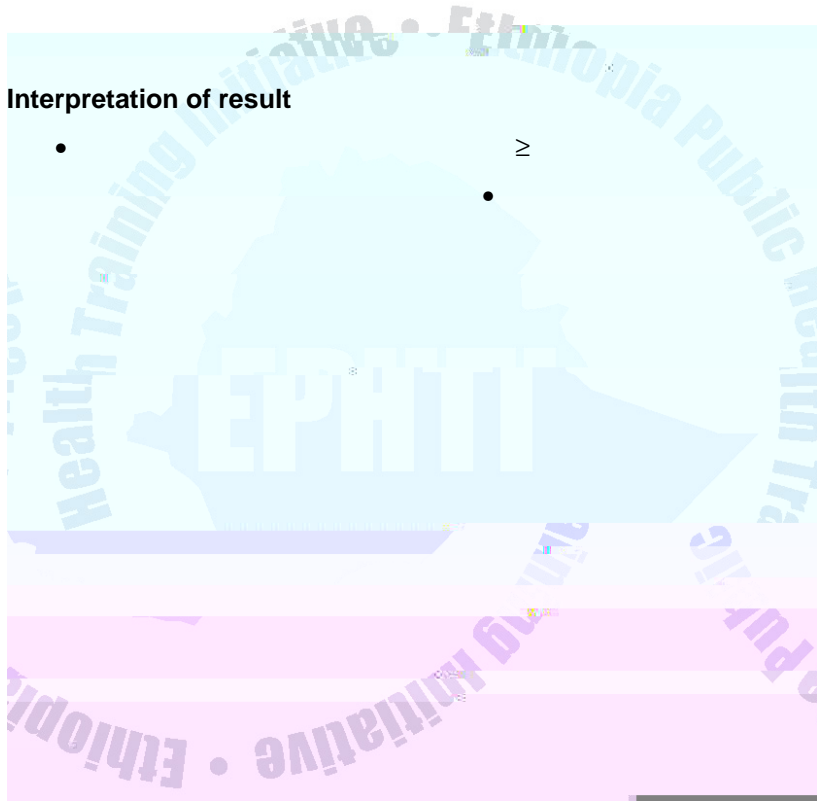
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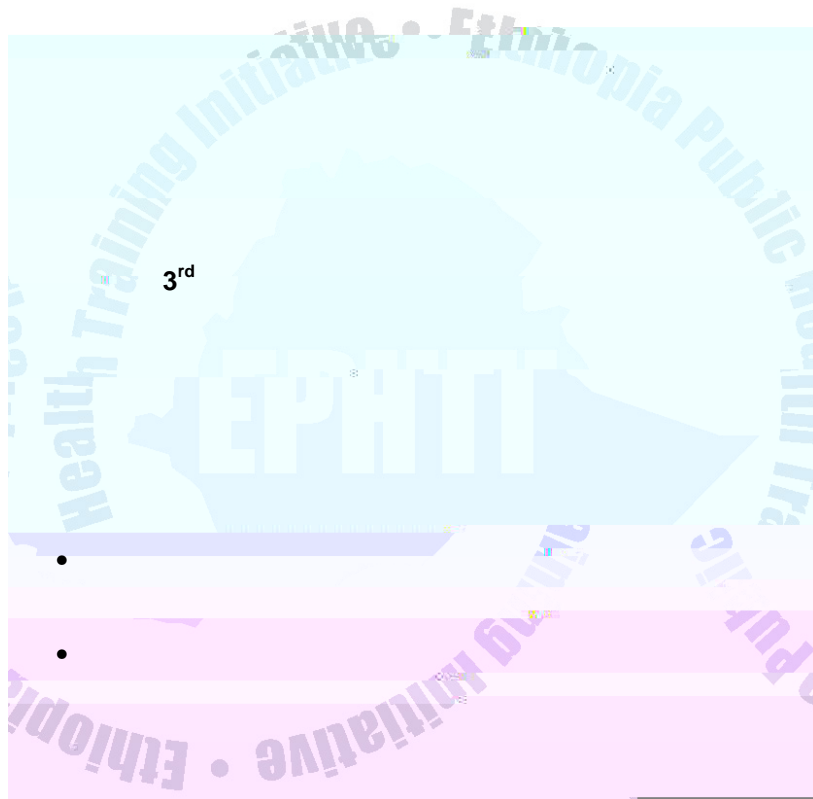
Interpretation of result

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GENUS: SHIGELLA







GENUS YERSINIA

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Yersinia pestis









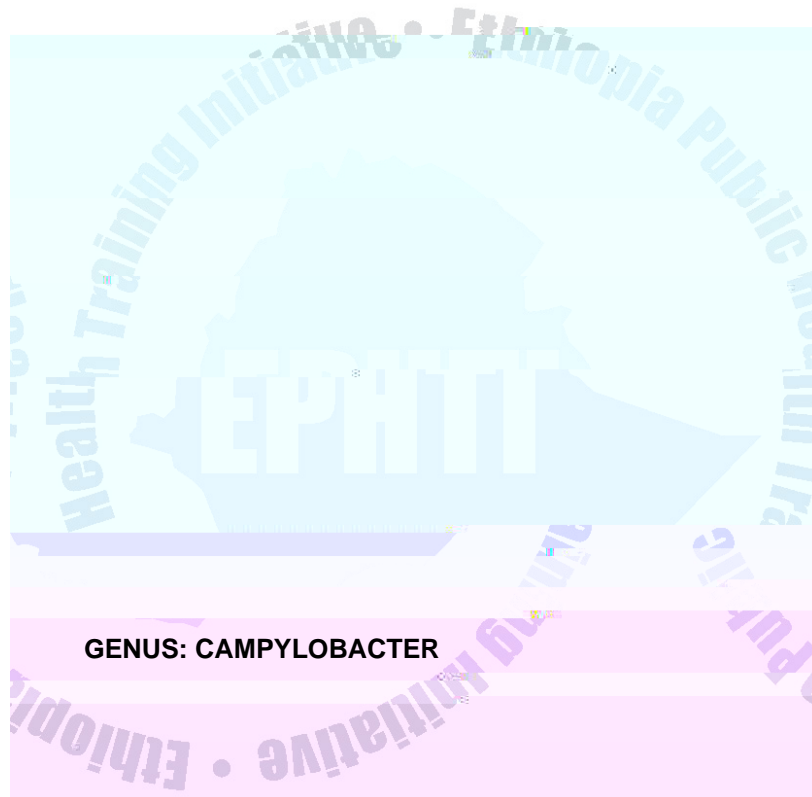
Pseudomonas aeruginosa











Campylobacter jejuni and *Campylobacter coli*







GENUS: LEGIONELLA

L. pneumophila







Representative anaerobic infections

**Commonly
isolated anaerobic
bacteria**





2.5. GENUS: MYCOBACTERIA







Table 2.7. Comparison of the two types of leprosy

Characteristics	Lepromatous leprosy	Tuberculoid leprosy
Bacterial index (BI)		

GRADING:

≥

≥

≥

NB: Count BI for each smear and calculate the average to give an over all BI

Morphologic index

Methods of collecting skin smears and staining of *M. leprae*

Procedure:



ACTINOMYCETES







6. SPIROCHETES



Treponema pallidum





B. Congenital syphilis







2.6.2. GENUS: BORELLIA



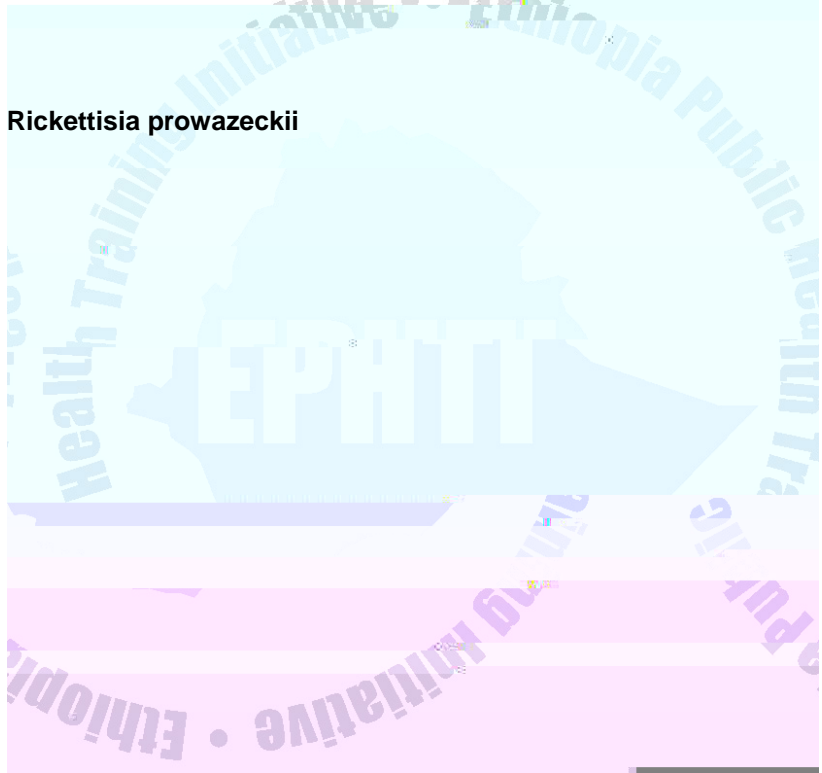


GENUS LEPTOSPIRA

L. interrogans











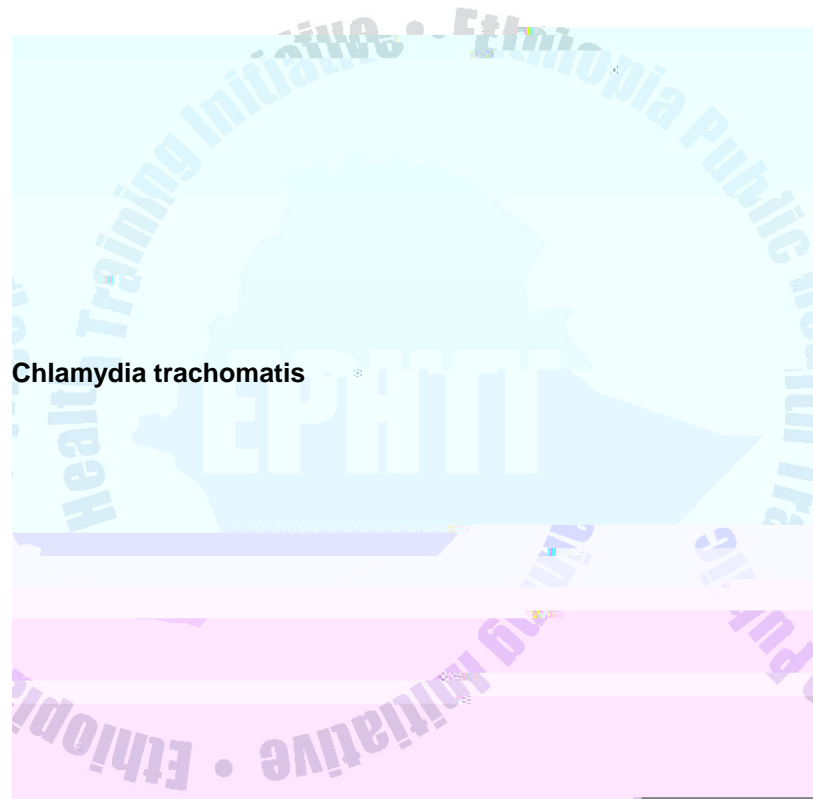


Mycoplasma hominis and Ureaplasma urealyticum

2.9. GENUS: CHLAMYDIA



Developmental cycle of chlamydia





Chlamydia pneumoniae



Review Questions

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Major events in phagocytosis



Major functions of complement system



2. Specific defense mechanisms

The humoral response

Functions of antibodies

The cell mediated response

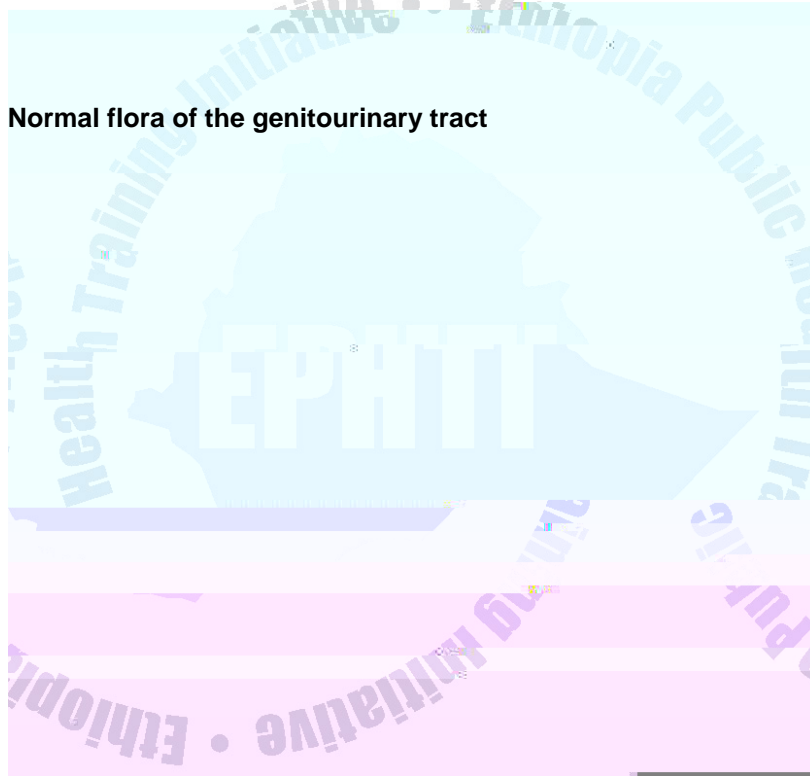
Functions of lymphokines

4.2. Normal microbial flora





Normal flora of the genitourinary tract



Normal flora of the eye

Normal flora of the external auditory meatus

4.3. INFECTION OF SKIN AND WOUND

A. Infection of skin

1. Superficial skin infection



B. Infection of wound



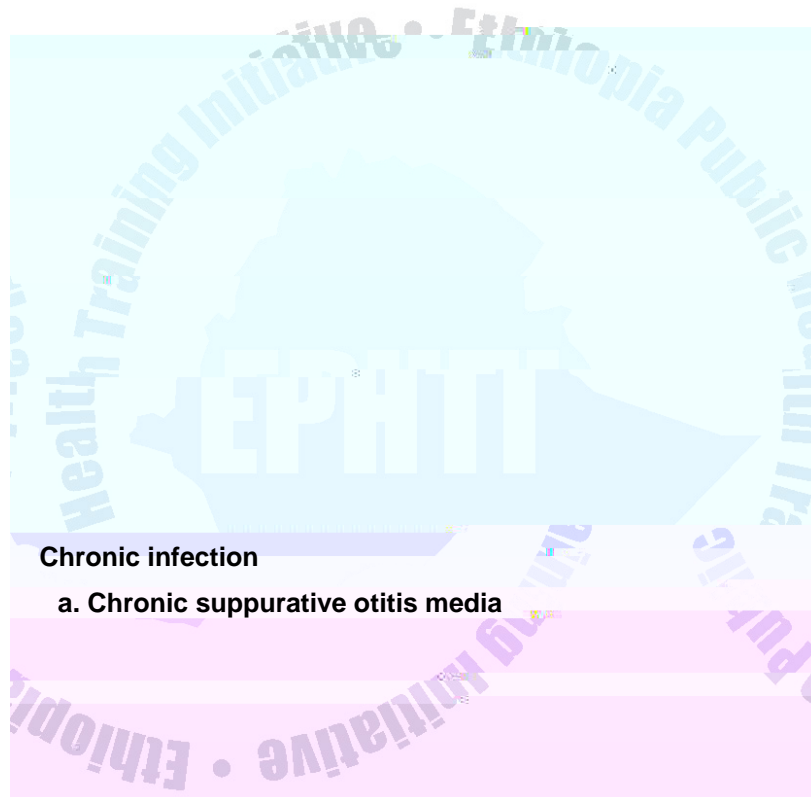
4.4. Infection of Respiratory Tract

Infection of middle ear and sinuses

Acute infection

a. Acute otitis media

. Acute sinusitis

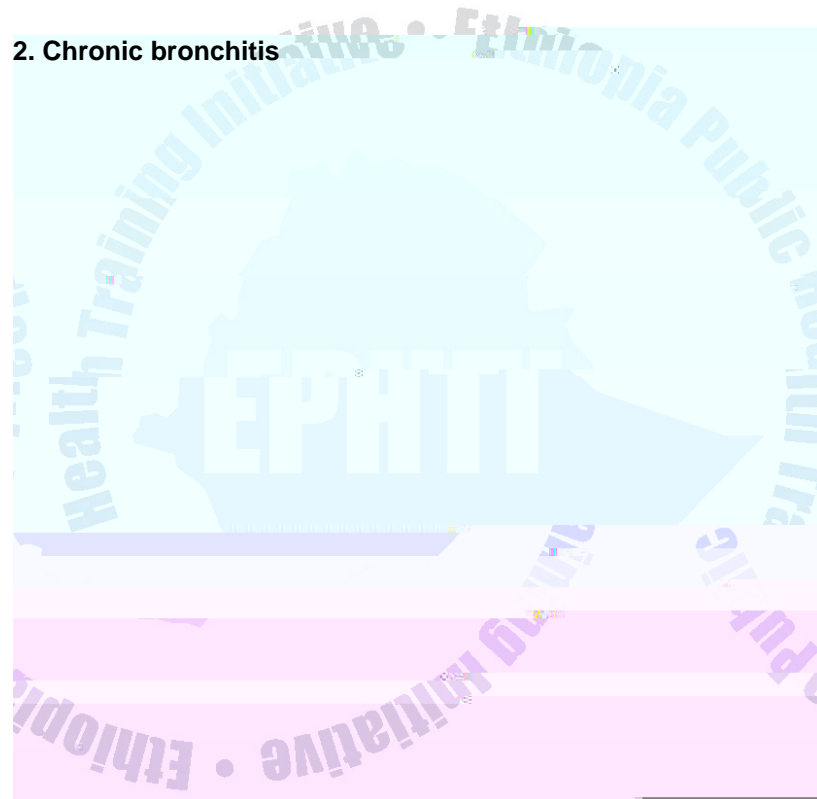




Bronchitis

1. Acute bronchitis





2. Chronic bronchitis

Pneumonia







Types of diarrhea



Complication of diarrhea



FOOD POISONING

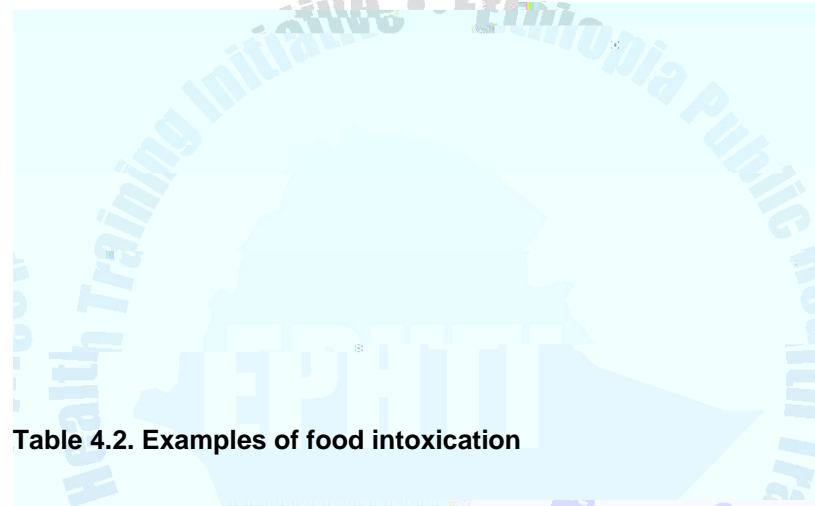
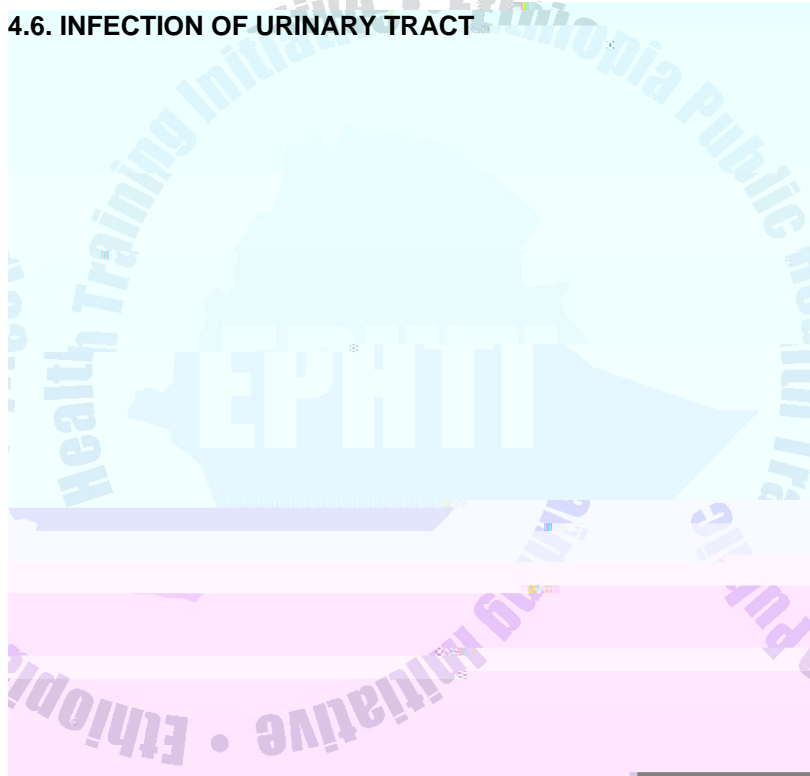


Table 4.2. Examples of food intoxication

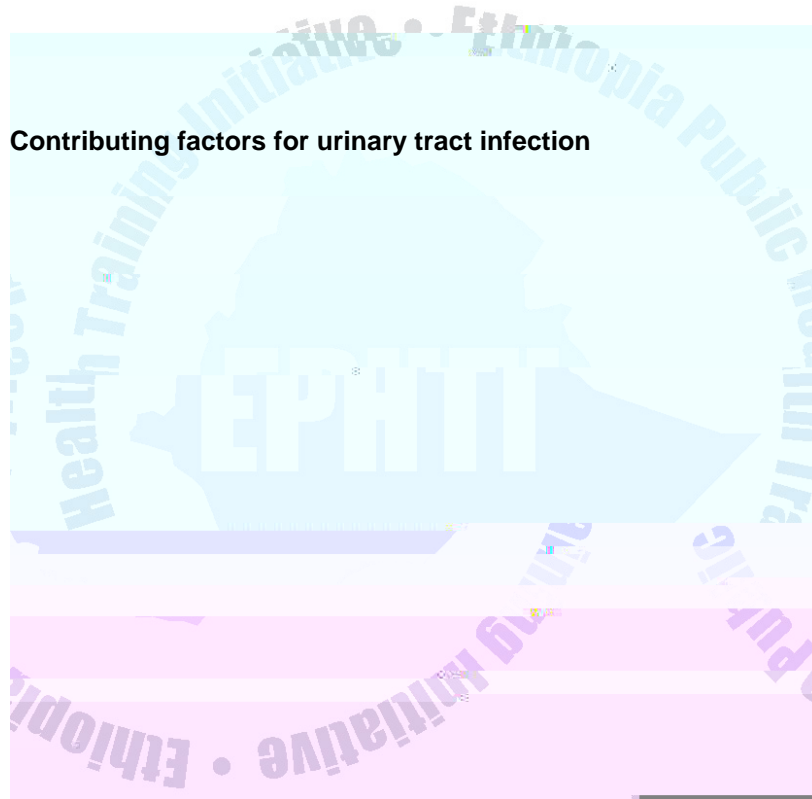
Organism	Incubation period	Clinical findings	Related food item

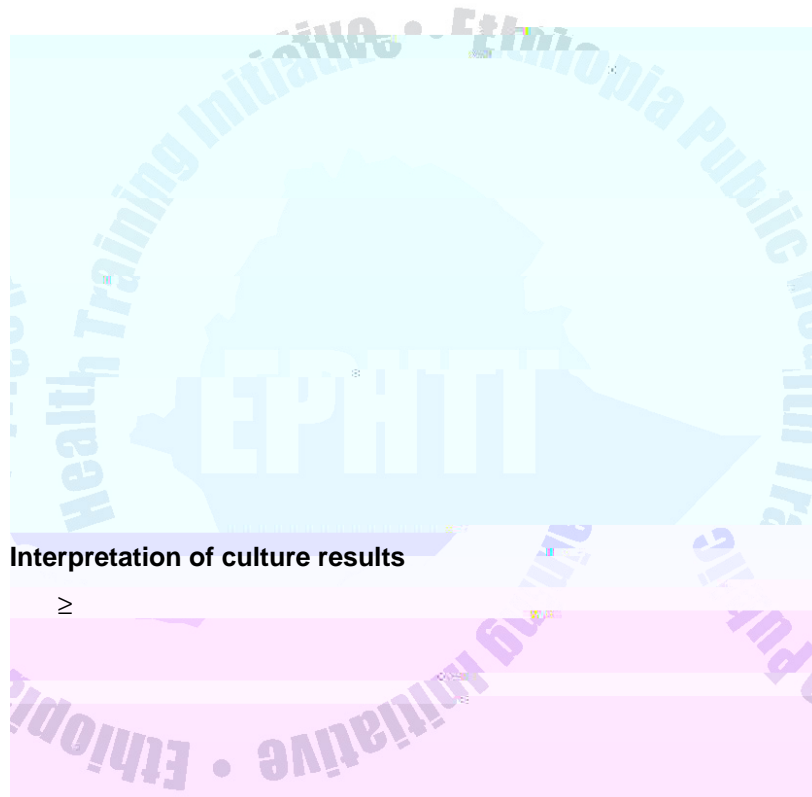
4.6. INFECTION OF URINARY TRACT



Routes of infection

Contributing factors for urinary tract infection





4.7. INFECTION OF GENITAL TRACT

Table 4.4. Genital tract infection manifests as either genital discharge or genital ulceration with or without inguinal lymphadenitis.


Causative agents	Diseases
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[Content obscured by watermark]	[Content obscured by watermark]

Urethral and vaginal discharge

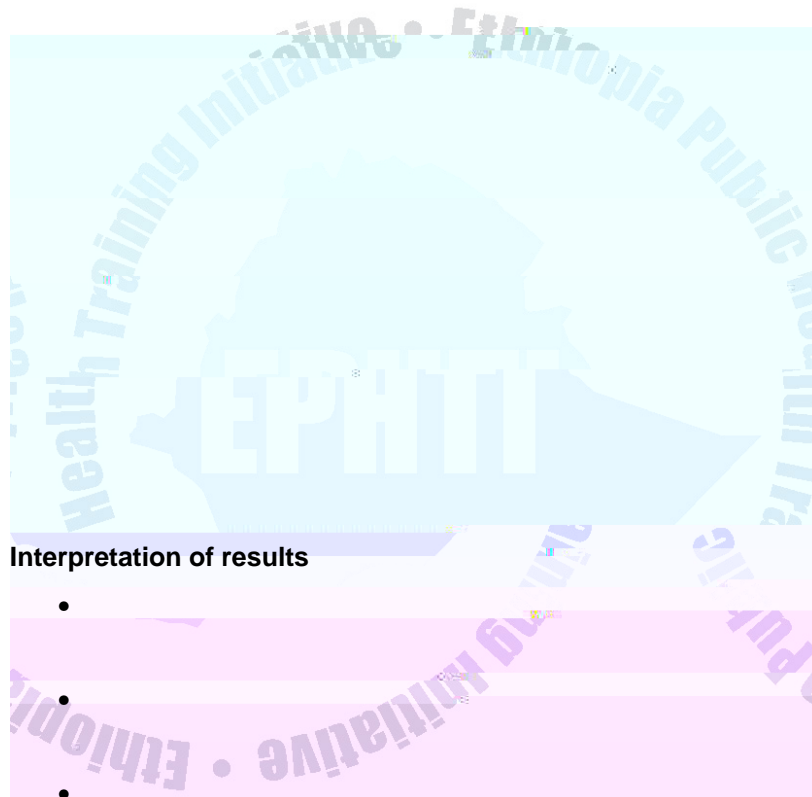




Table 4.5. Genital ulceration with or with out regional lymphadenopathy

Disease	Lesion	Inguinal lymphadenopathy
		





Common contaminants of blood culture

4.9. INFECTION OF CENTRAL NERVOUS SYSTEM







Complication of meningitis

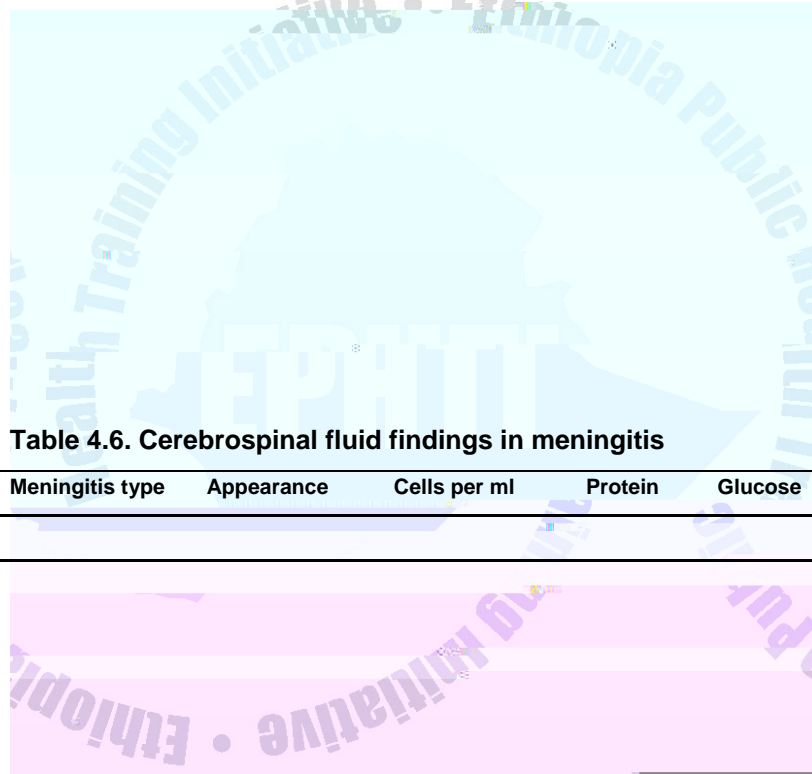
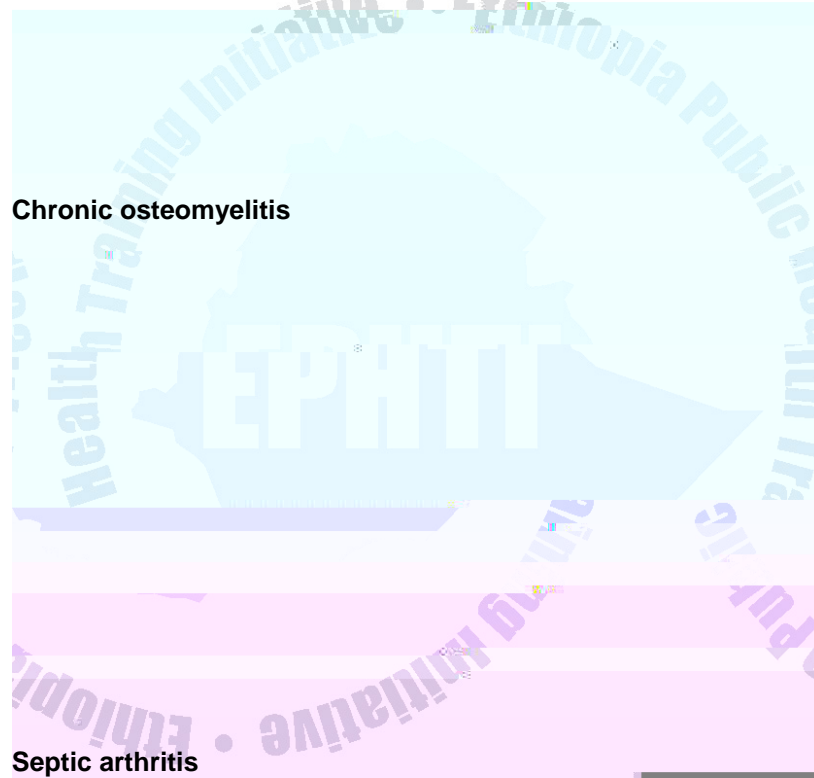


Table 4.6. Cerebrospinal fluid findings in meningitis

Meningitis type	Appearance	Cells per ml	Protein	Glucose







Review Questions

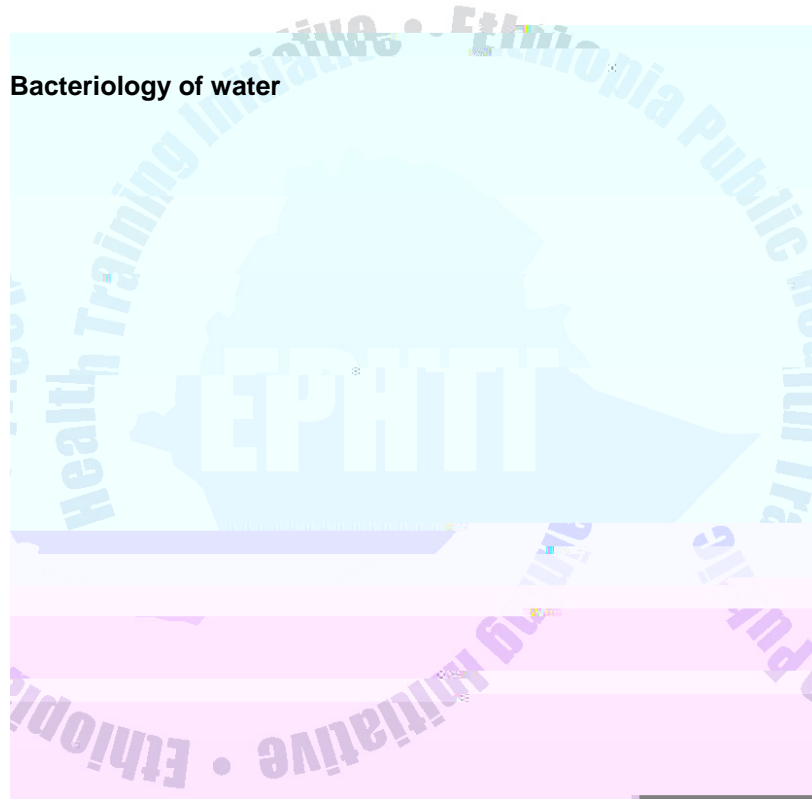
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CHAPTER FIVE

Learning objective:

Bacteriology of water



Collecting a sample from an open well:

Transport of water sample

Frequency of sampling

Population served **Sampling interval**

Multiple tube technique for counting fecal coliforms



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Interpretation of results

E. coli count	Comment
—	

E. coli count Category Comment

Review question



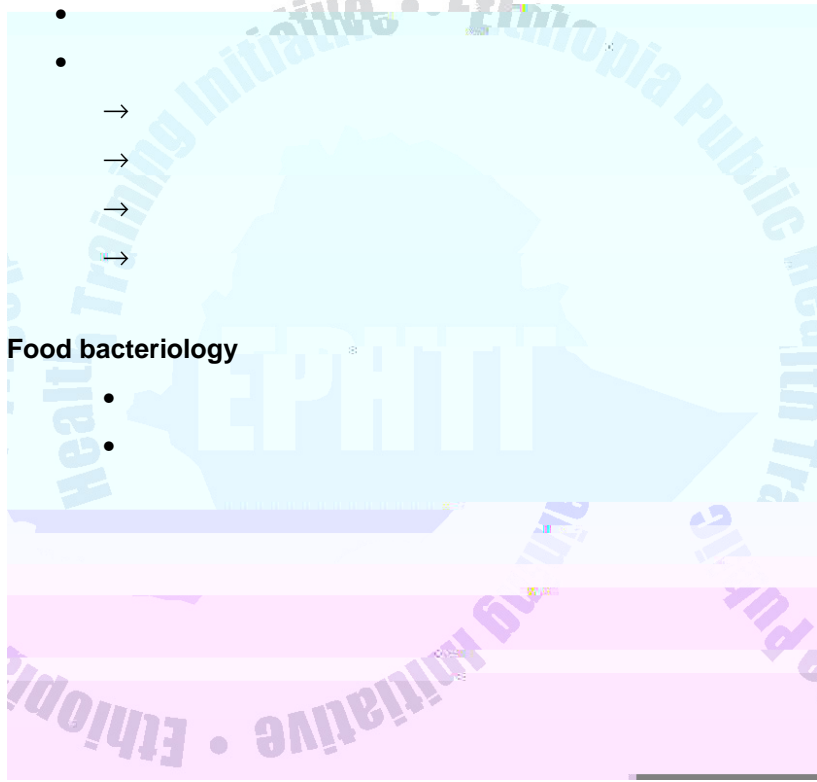
CHAPTER SIX

Learning objective

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Food bacteriology

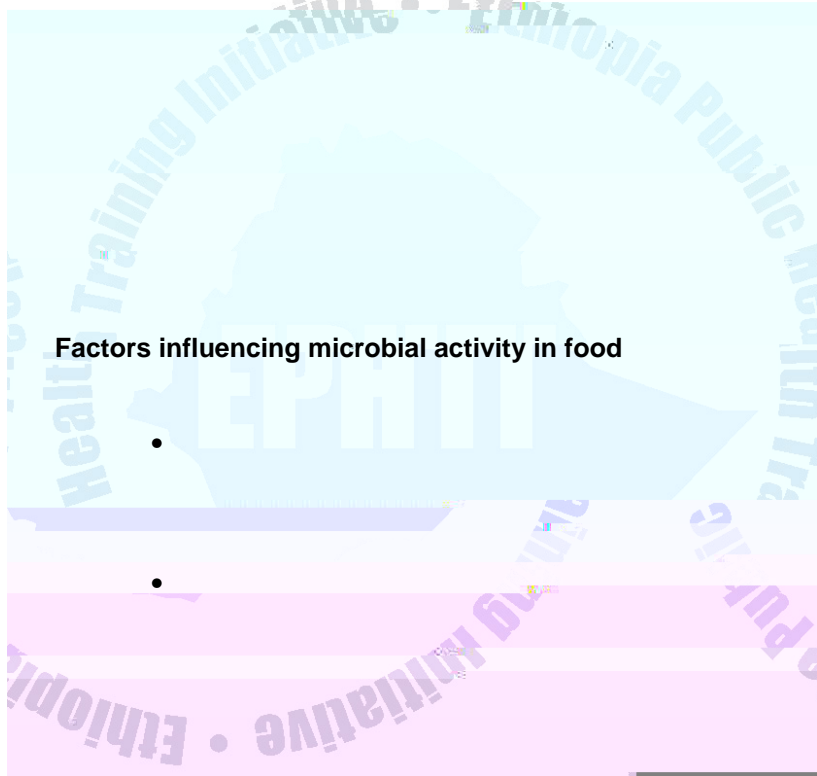
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Sources of food contamination





Factors influencing microbial activity in food

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§

Differentiation of faecal from non faecal coliform



Organisms	Indole	Methyl red	V – P	Sodium citrate

Elevated temperature test

§

Index organisms

iii) Food poisoning organisms

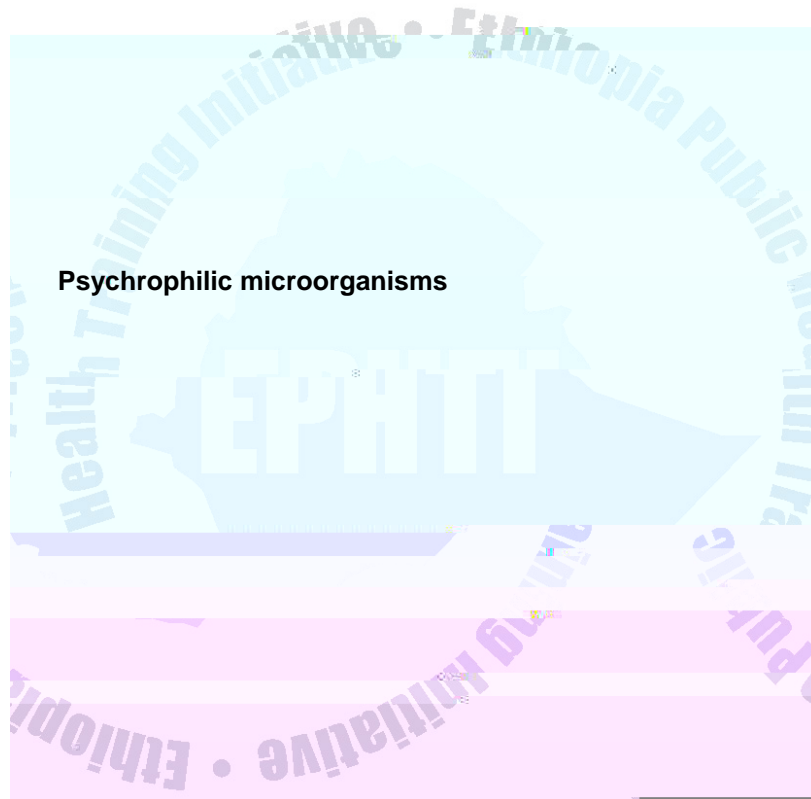
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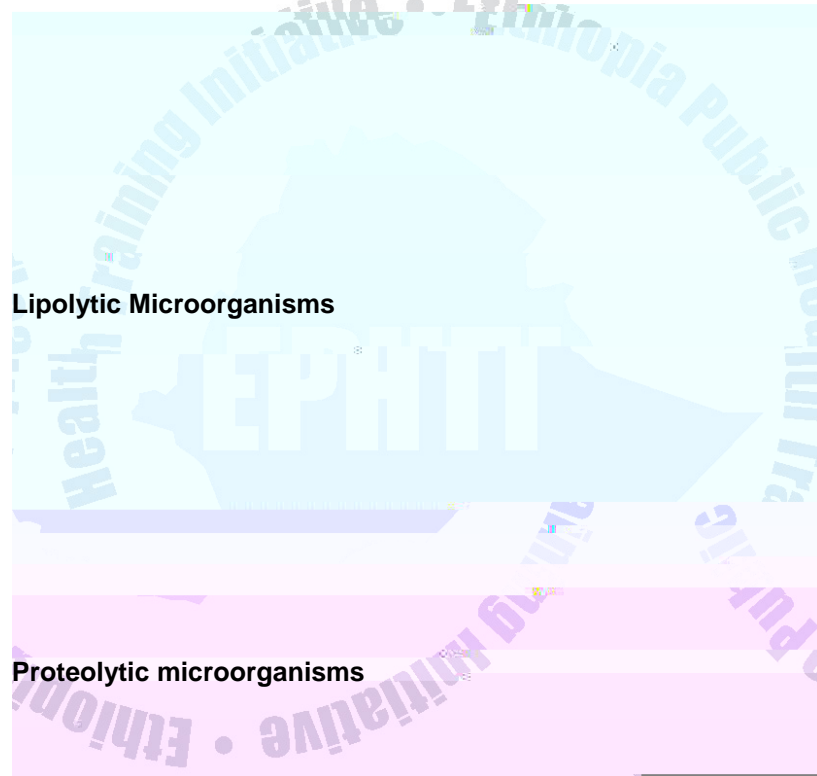
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iv) Infectious microorganisms

The spoilage micro-organisms



Thermoduric microorganisms



Halophilic microorganisms



Osmophilic microorganisms

Pectinolytic microorganism

Acid producing microorganisms

Yeasts and moulds

Mesophilic spore forming aerobes

Thermophilic anaerobes

Microbiological Examination of Food
Sampling

-
-
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TM

TM

TM

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MICROBIOLOGICAL EXAMINATION OF DIFFERENT FOOD

For example Egg and egg products ‘

Types of contaminating microorganisms

TM

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Microbiological examination

Methods of analysis

Sampling plan and microbiological limit



METHODS OF MICROBIOLOGICAL ANALYSIS OF FOOD AND WATER

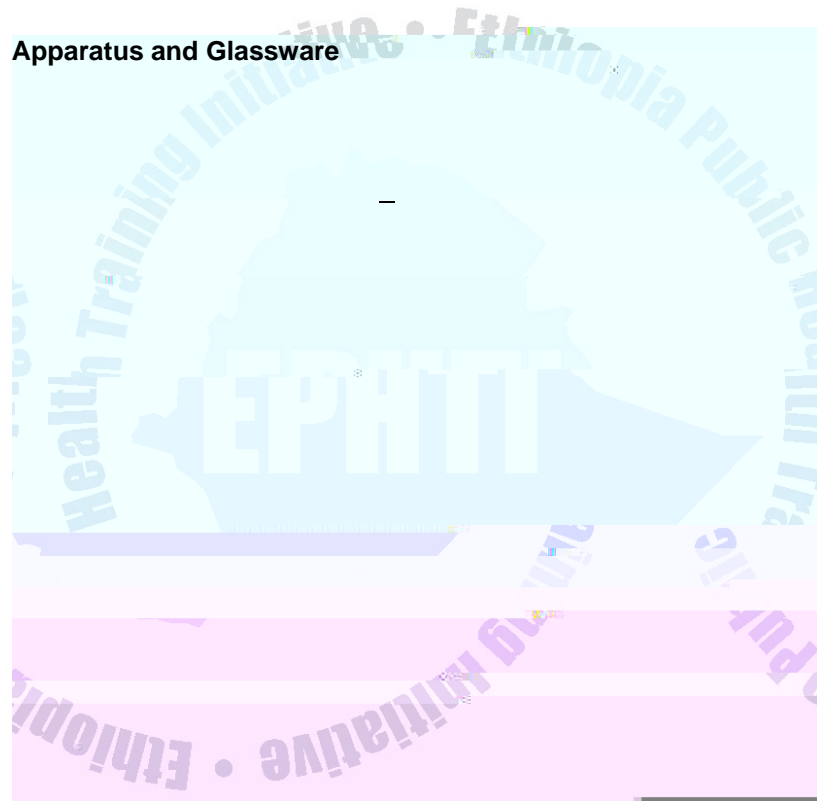
Principle



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TM

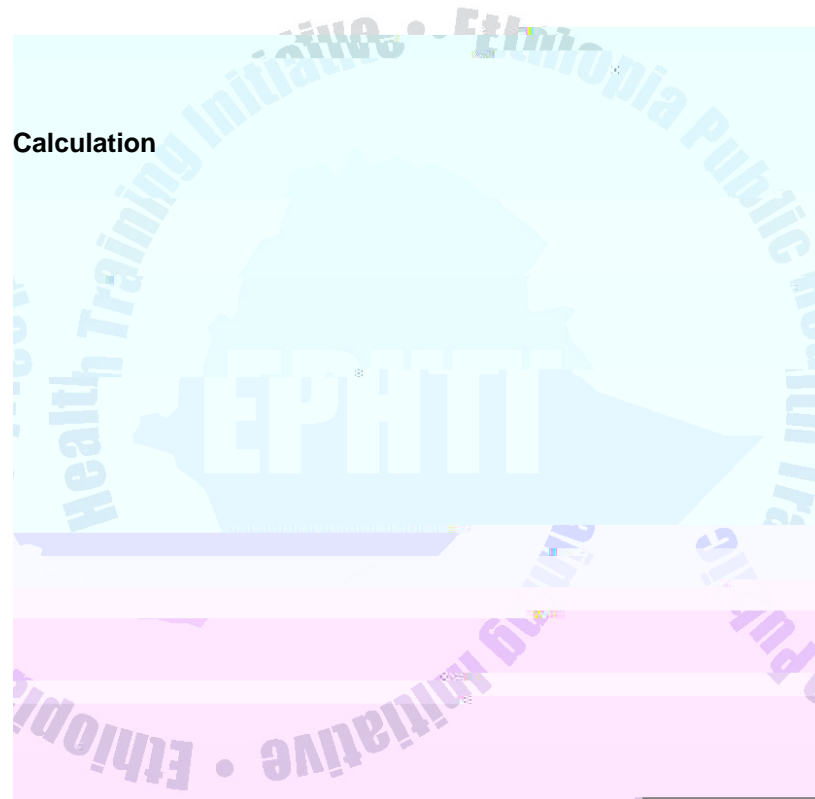
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Incubation

Counting the colonies



Calculation

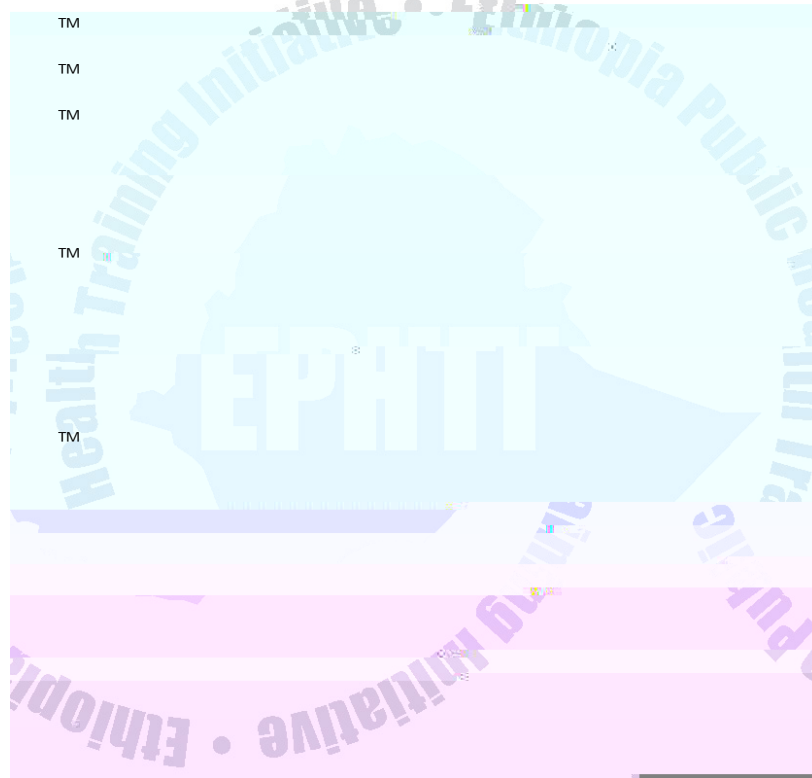
Calculation

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ENUMERATION OF COLIFORM BACTERIA

MPN

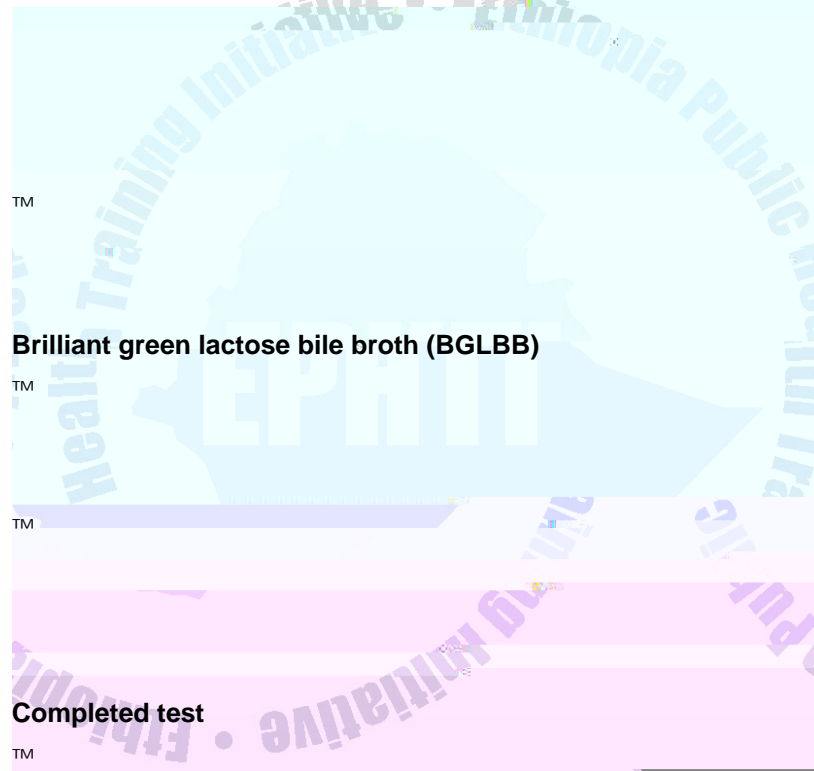
Principle



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Brilliant green lactose bile broth (BGLBB)

TM

TM

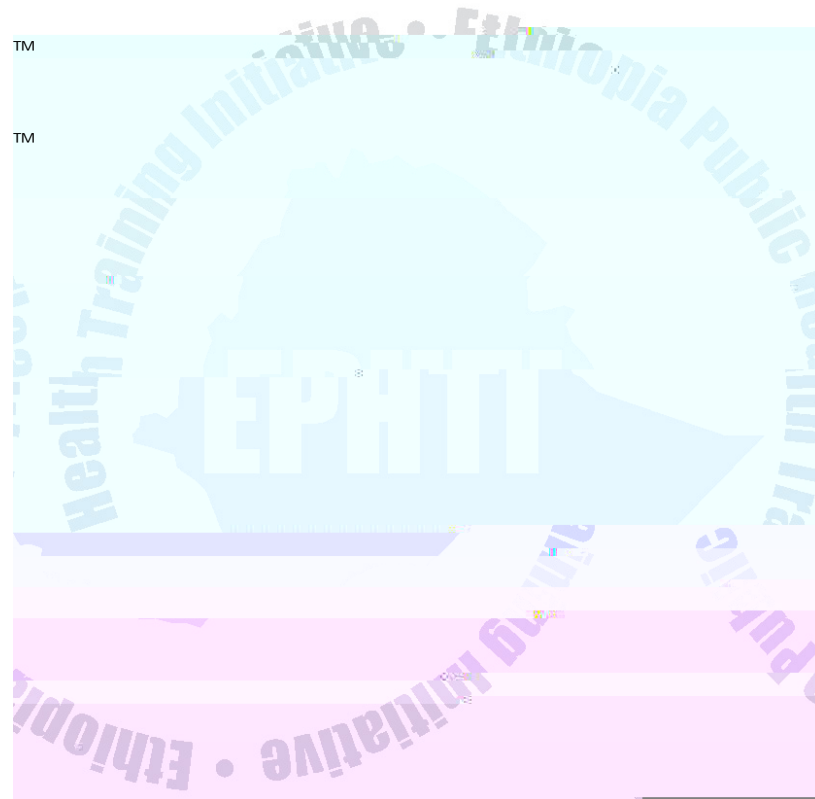
Completed test

TM

TM

The purpose of the completed test is to determine

TM



Procedure

Preparation of food homogenate

-

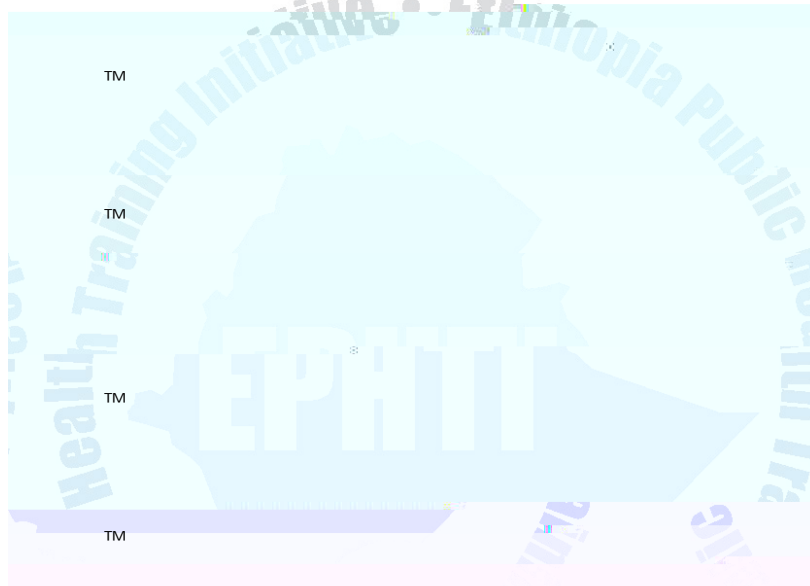


Dilution

-



Differentiation of non-faecal coliforms faecal coliforms



Classification of Coliforms by IMViC test

Indole	MR	VP	Citrate	Type
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Procedure

Preparation of food homogenate

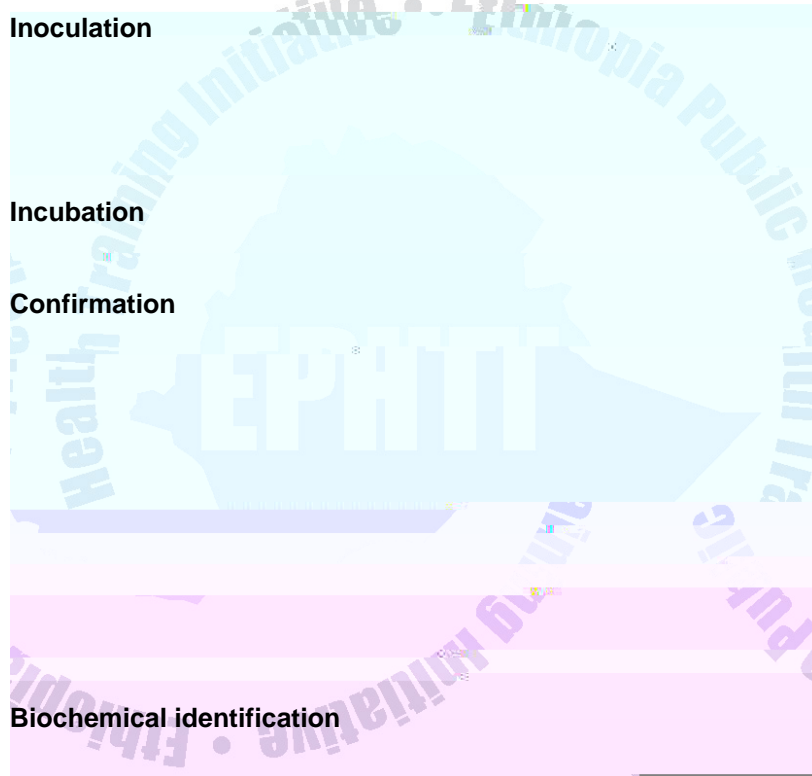
Dilution

Inoculation

Incubation

Confirmation

Biochemical identification







Calculation

ENUMERATION OF BACILLUS CEREUS

Principle

Apparatus and Glassware

Culture media and reagents

Incubation

Counting of the colonies (presumptive *B. cereus*)

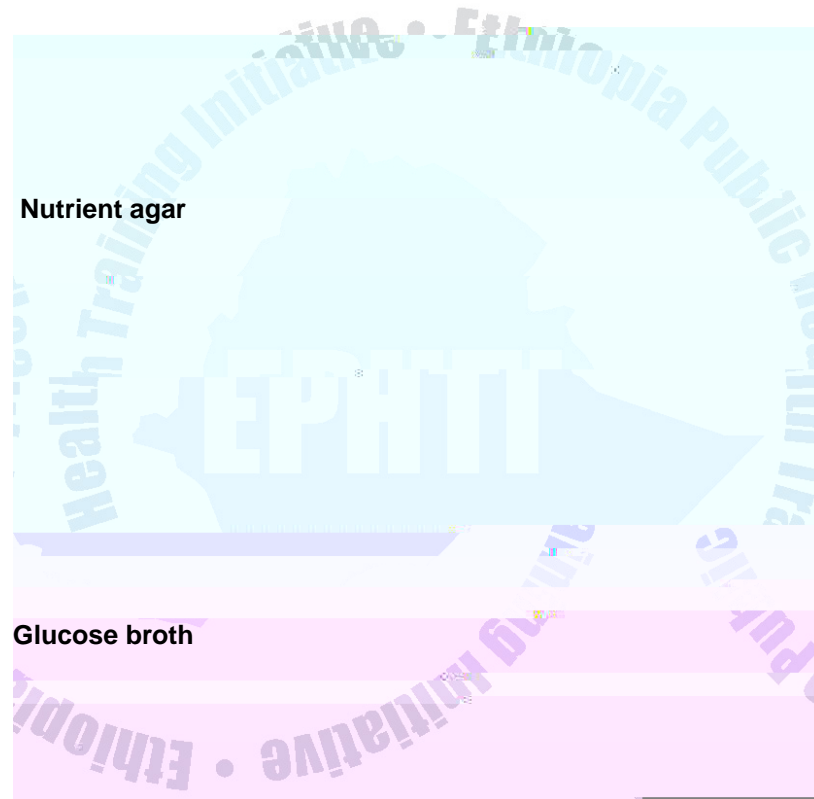


Calculation





Nutrient broth



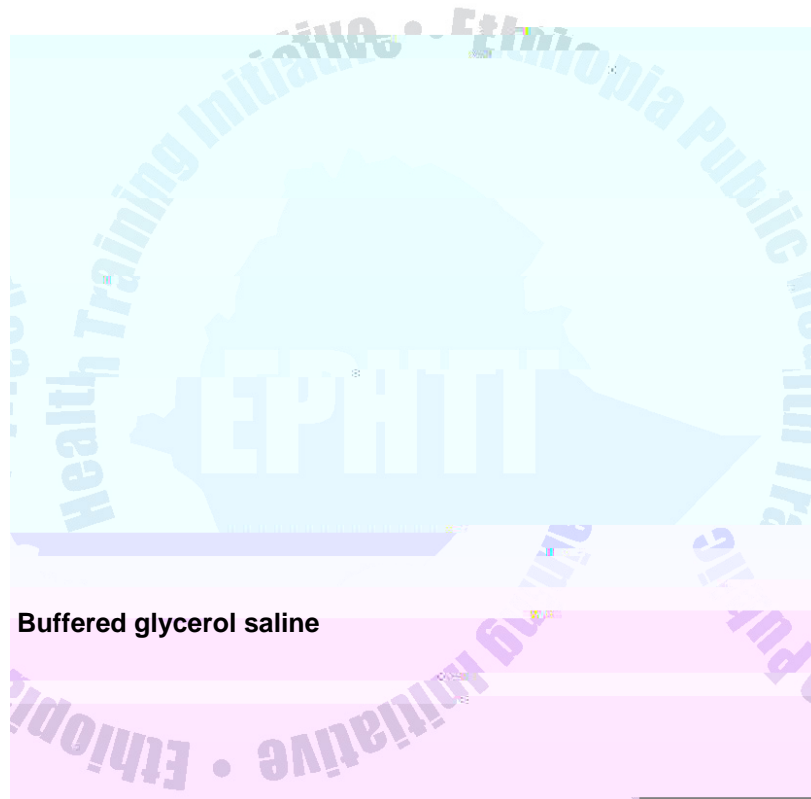
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Blood agar

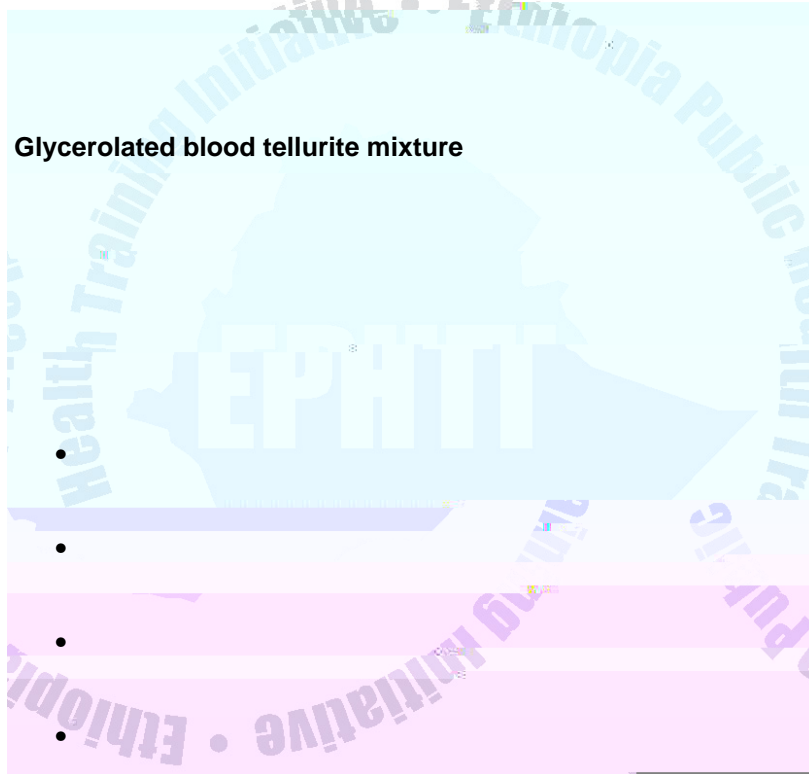


XLD agar



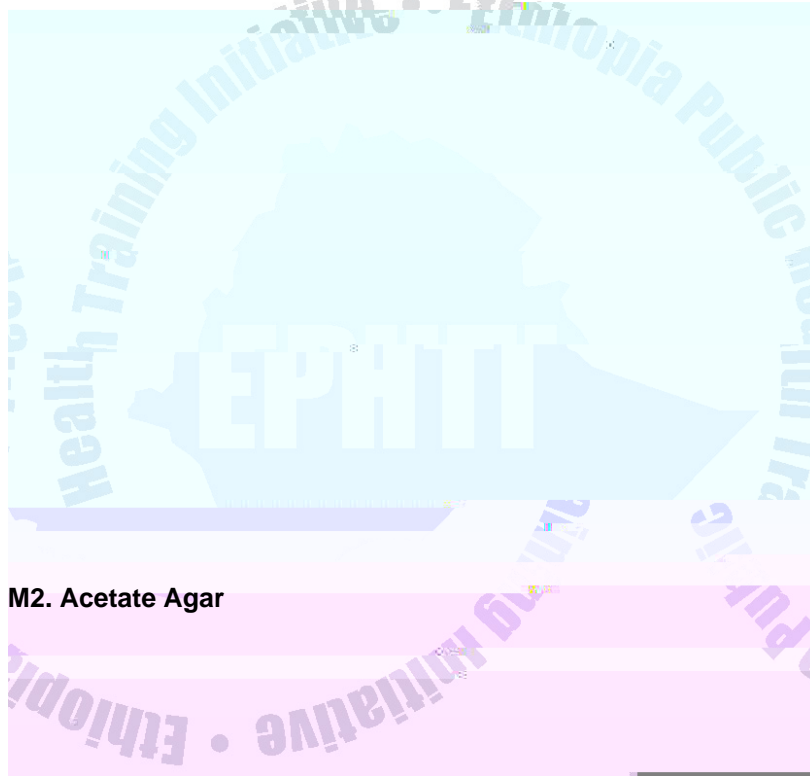
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Preparation of complete medium

M1. A-1 Medium





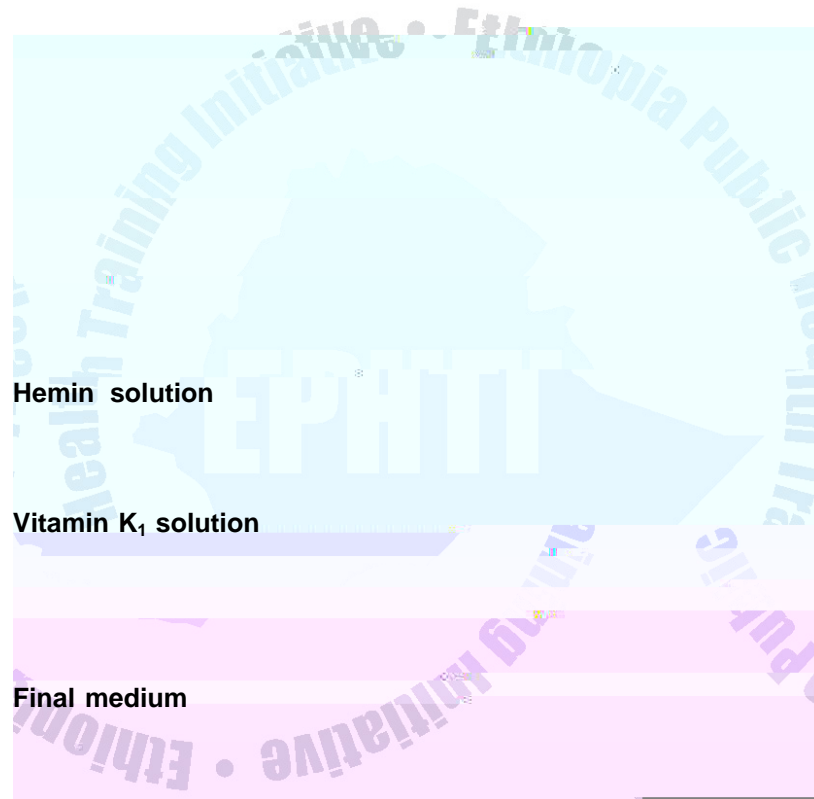
M5. Alkaline Peptone Agar

M6. Alkaline Peptone Salt Broth (APS)

M7. Alkaline Peptone Water

M8. Anaerobe Agar

Base



M9. Anaerobic Egg Yolk Agar

Agar base



M10. Antibiotic Medium No. 1 (Agar Medium A)

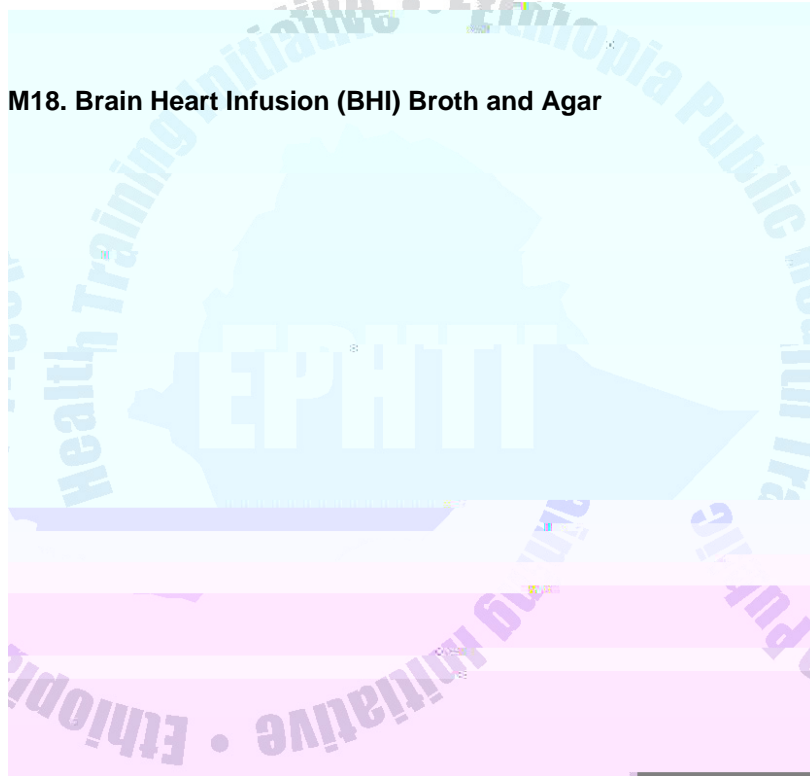






M17. Brain Heart Infusion (BHI) Agar (0.7%)
(for staphylococcal enterotoxin)

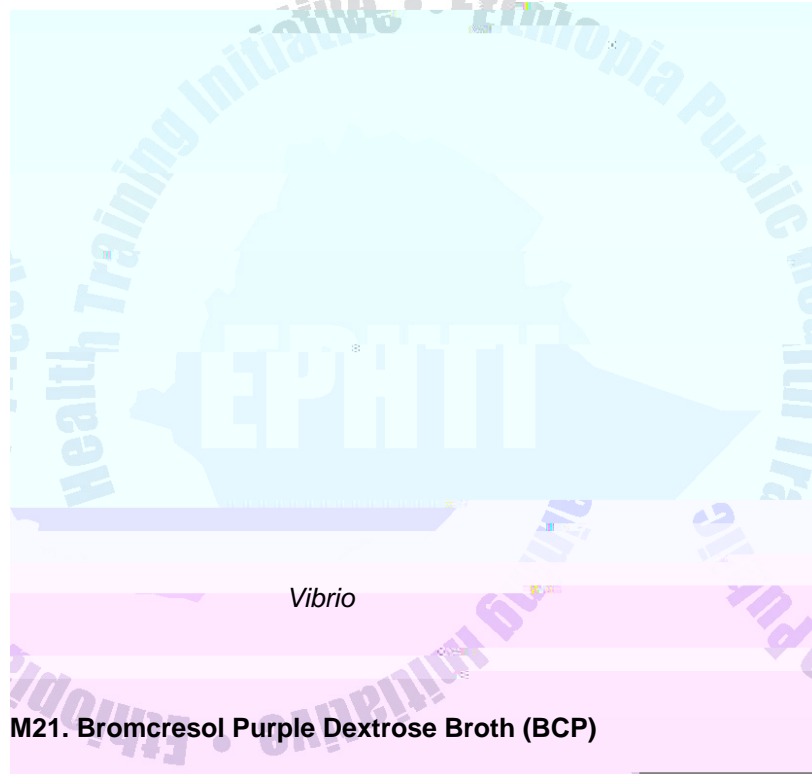
M18. Brain Heart Infusion (BHI) Broth and Agar

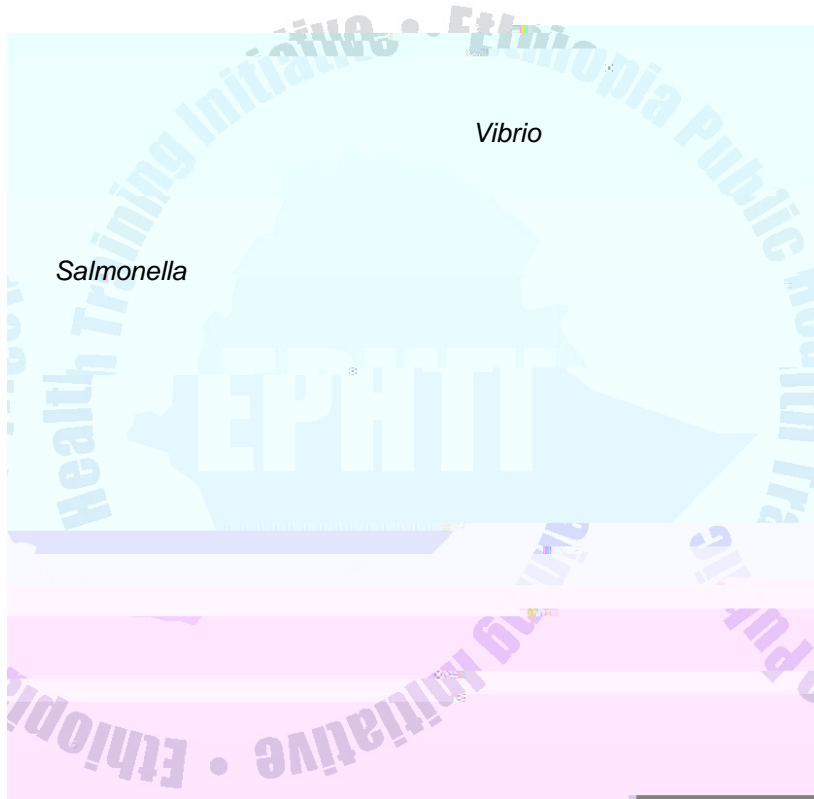




M20. Bromcresol Purple Broth

Base



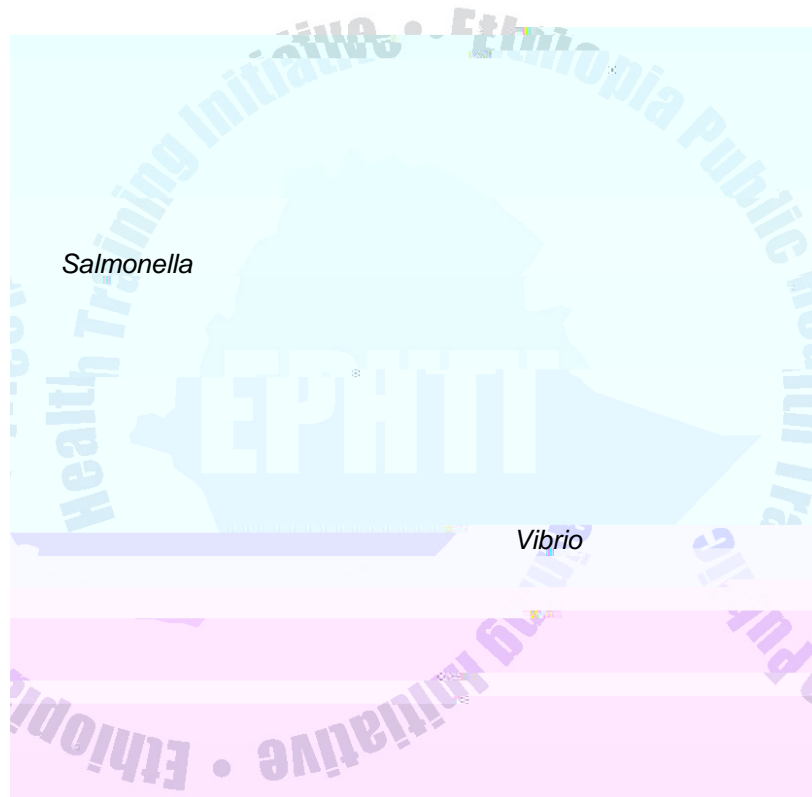


Vibrio





















Vibrio





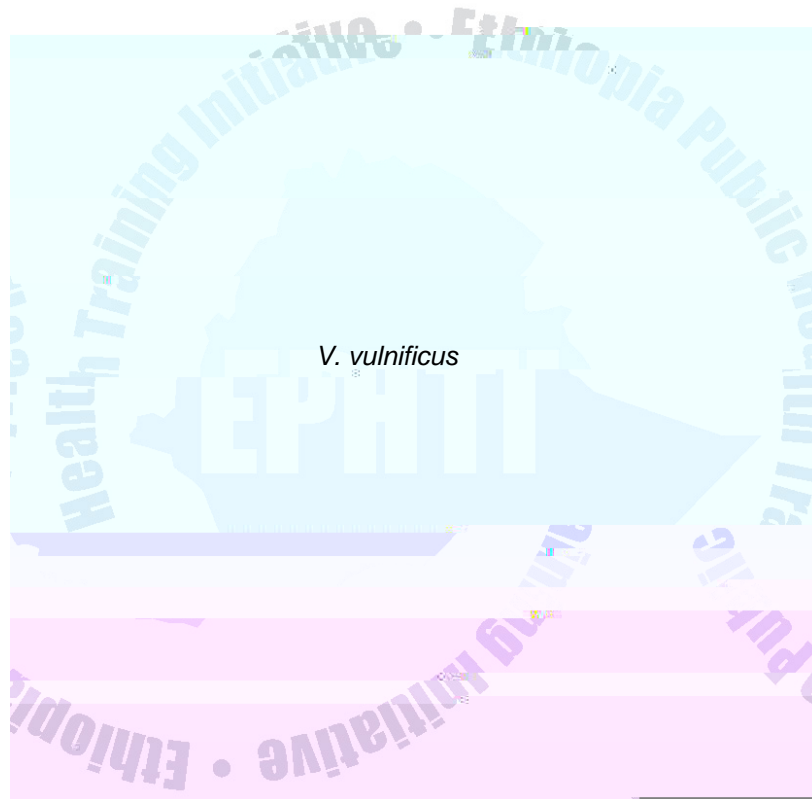










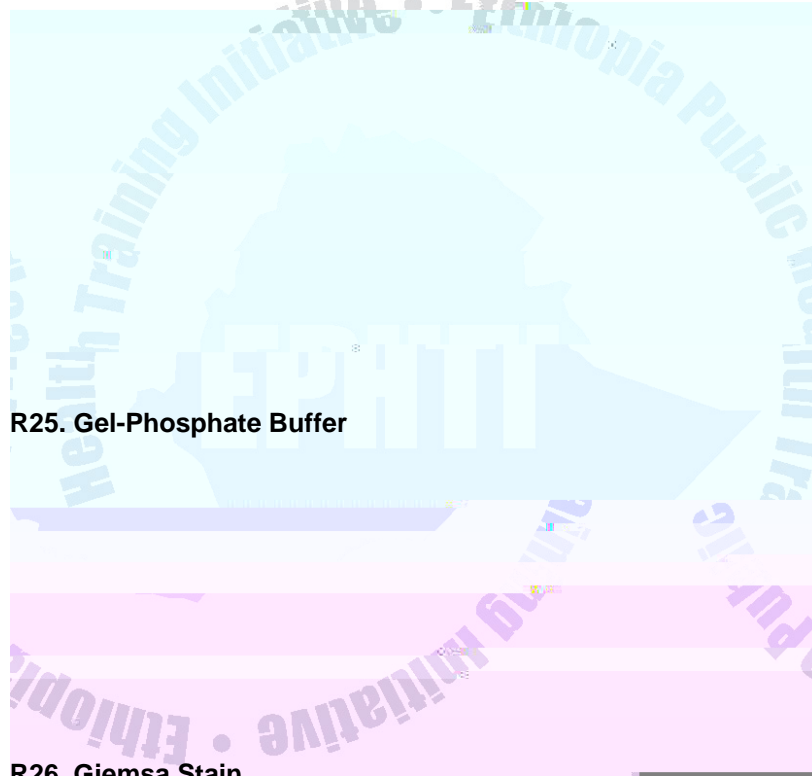




V. vulnificus



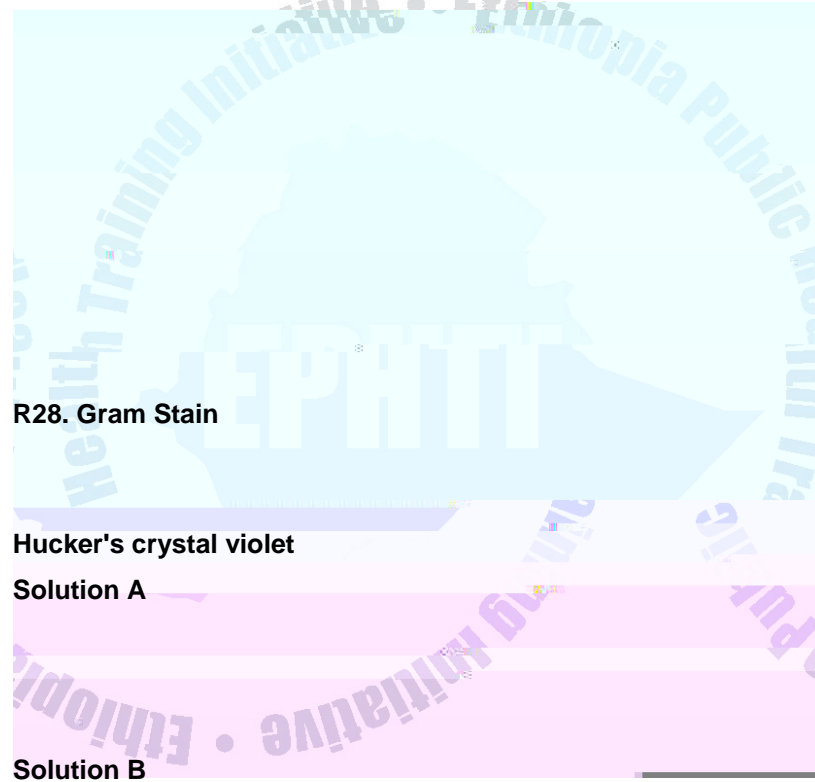
R24. Gel Diffusion Agar. 1.2%



R25. Gel-Phosphate Buffer

R26. Giemsa Stain

R27. Glycerin-Salt Solution (Buffered)



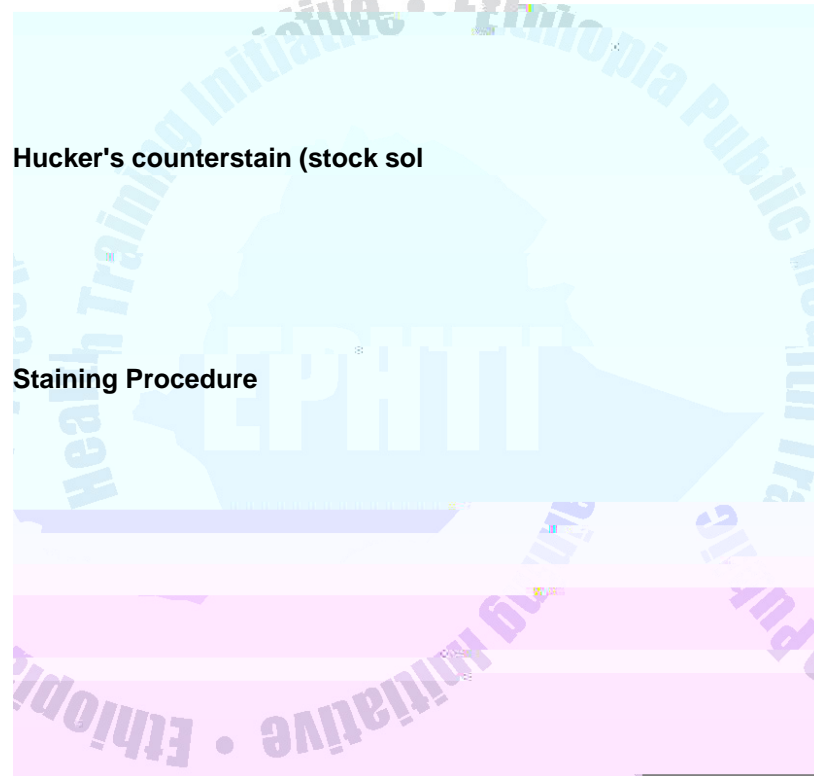
R28. Gram Stain

Hucker's crystal violet

Solution A

Solution B

Gram's iodine



R29. Endospore Stain (Schaeffer-Fulton)Sol

So(l).5n B

R30. Hippurate Solution, 1%

R31. Horseradish Peroxidase

Solution A (horseradish)

Solution B

R32. 1 N Hydrochloric Acid

R33. Kovacs' Reagent

p

p

R34. 0.1 N Lithium Hydroxide

R35. Lugol's Iodine Solution

R37. Methylene Blue Stain (Loeffler's)

Solution A

Solution B

R38. Nitrite Detection Reagents

A. Sulfanilic acid reagent

B. N-(1-naphthyl)ethylenediamine reagent

C. alpha-Naphthol reagent

Nitrate reduction test for enteropathogenic *E. coli*

D. Alternative test reagents

R39. Oxidase Reagent

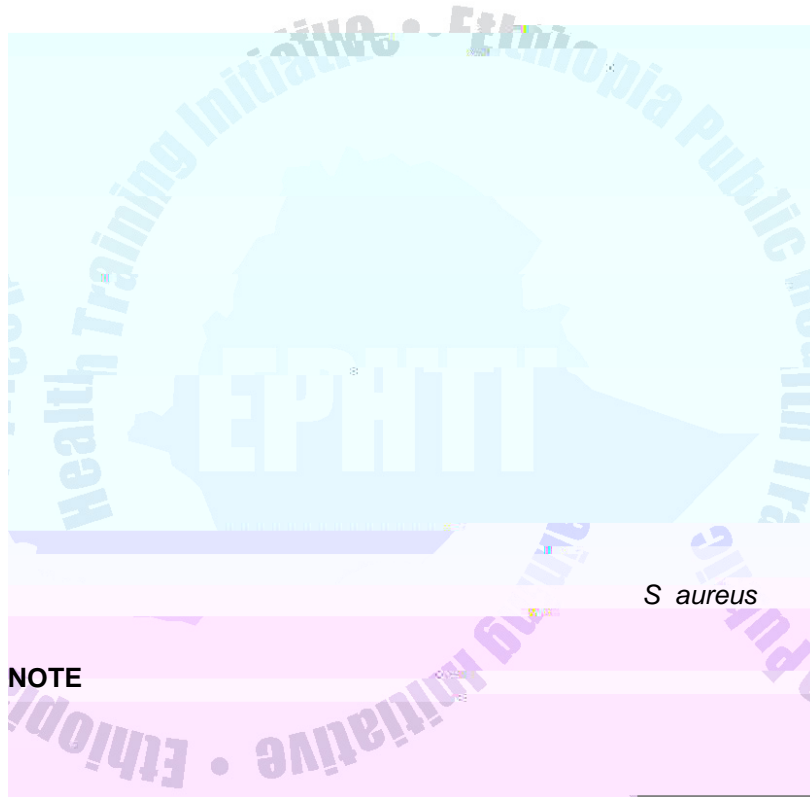
R40. Peptone Diluent, 0.1%

**R41. 0.01 M Phosphate-Buffered Saline (pH 7.5) Stock solution
(0.1 M)**

R42. 0.02 M Phosphate Saline Buffer (PH 7.3-7.4)

Stock solution 1

Stock solution 2



R44 Potassium Hydroxide Solution. 40%

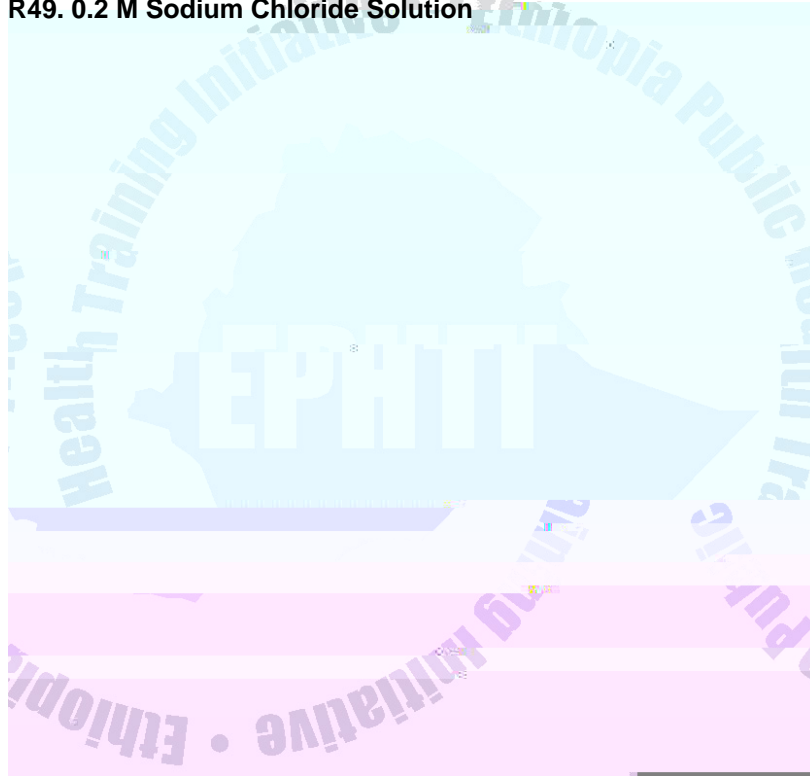
R45. Saline Solution. 0.5% (Sterile)

R46. Salts-Phosphate Buffered Saline Solution (Salts-PBS)

R47. Slide Preserving Solution

R48. Sodium Bicarbonate Solution. 10%

R49. 0.2 M Sodium Chloride Solution



6X SSC

3X SSC

2X SSC

R53. Tris-Buffered Saline (TBS) (PH 7.5)

R54. Tris-Buffered Saline (TBS), with Gelatin

1% solution

3% solution

R55. Tris-Buffered Saline (TBS)-Tween

R56. Tris-Buffered Saline (TABS), 1% or 3% Gelatin, or Tween
20

For 1% and 3% Gelatin-TABS,

For Tween-TABS,

R57. Voges-Proskauer (VP) Test Reagents

Solution 1

Solution 2

Voges-Proskauer (VP) test

GLOSSARY

Asexual reproduction:

Acute:

Antigen:

Antibody:

Blood Brain Barrier:

Chronic:

Convulsion:

Congenital:

Dehydration:

Disease:

DNA:

Endogenous:

Endoplasmic reticulum:

Electrolyte:

Fastidious:

Genome:

Hematogenous:

Histone:

Hydrocephalus:

Iatrogenic:

Incubation Period:

Infertility:

Lysosome:

Microscopic

Macroscopic

Microscope:

Microorganism

Mitochondria

Microtubule Microfilament

Nuclear membrane

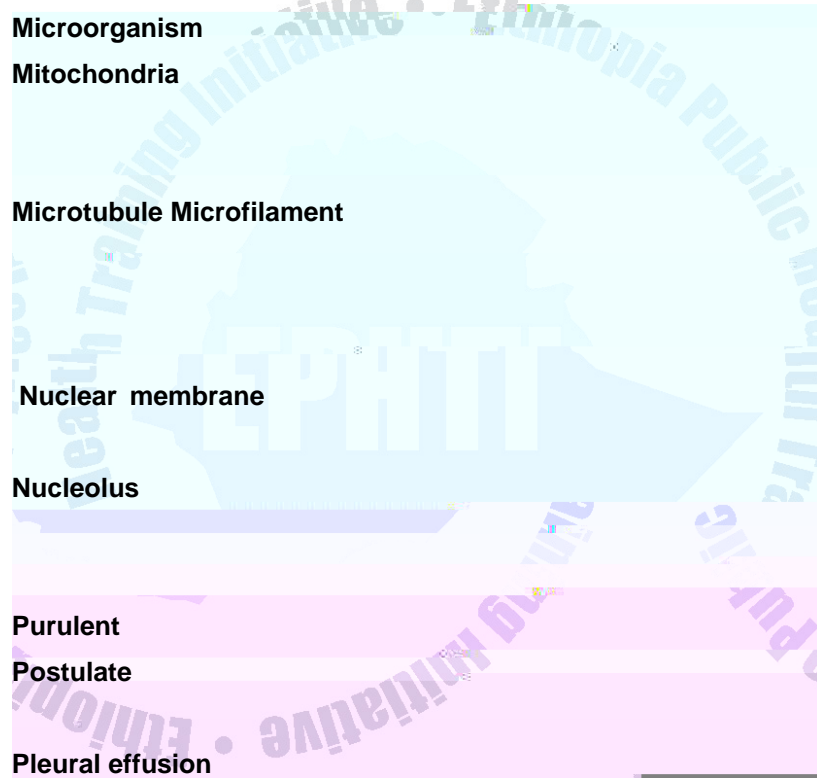
Nucleolus

Purulent

Postulate

Pleural effusion

Primary stain



Mordant

Decolorizer

Pathogen

Virulence

Invasiveness :

Toxicity:

Opportunistic

Infection:

