CAMBRIAN COLLEGE OF APPLIED ARTS & TECHNOLOGY

HEALTH SCIENCE PROGRAMS

MEDICAL LABORATORY TECHNOLOGY

MICROBIOLOGY (GENERAL)

BIO 1022-4

SEMESTER 2 - 4 HOURS/WEEK

APRIL 1976

INSTRUCTOR: M. FRENCH

CHAIRMAN: A.J. QUAIATTINI

CAMBRIAN COLLEGE OF APPLIED ARTS & TECHNOLOGY

Course: Microbiology (General) Course No: BIO 1022

Duration: One Semester (16 wks) Credits: 4

Hours/wk: Lecture: 4

Total: 64

General Description & Objectives:

This course is a general introduction to the science of microbiology with emphasis on those areas that apply to the hospital laboratory.

History of microbiology, bacterial morphology, groups of microorganisms, bacterial taxonomy and nomenclature, growth and death of bacteria, effects of environment on bacteria, sterilization and disinfection, microscopic preparations - staining methods, pure culture study methods, culture media.

Prepared by: M. French

Date: January, 1976

Text: Microbiology, McGraw Hill, 1972, 3rd. ed.

Pelczar, Michael J. Reid, Roge D.

Reference Material

- Manual of Clinical Microbiology, Blair, J.E., Lennette, E.H., Truant, J.F. 2nd ed., 1974, American Society for Microbiology, Bethesda, Md.
- Medical Microbiology, Cruikshank, R., 12th ed., 1973 E. & S. Livingstone, Ltd., Edinburgh and London
- Review of Medical Microbiology, Jawetz, E. et al, 1974 Lange Medical Publications
- Medical Laboratory Technology and Clinical Pathology, Lynch, M.J., et al 2nd ed., 1969, Saunders Co., Philadelphia, Pa.
- Clinical Diagnosis by Laboratory Methods, David Sohn, I., Henry, J.B. (Todd-Sanford), 14th ed., 1969, W.B. Saunders Co., Philadelphia, Pa.
- BBL Manual of Products and Laboratory Procedures
 Becton, Dickinson and Company

Method of Evaluation:

Mid-Semester Exam 25%
Periodic Tests 25%
Semester Final Exam 50%

- A) Mid-Semester Examination Duration 50 minutes Examination on material covered from the beginning of the course up to the time of the examination.
- B) Semester final exam Duration 2 hours
 Examination material all topics covered during the semester.
- C) Periodic Tests on a regular basis, of approx. 15 20 minutes duration, usually with approx. 1 weeks notice, done at the end of a topic or as seen fit by the instructor.

A pass mark is 60% (Grade - C)

80% - 100% = A

70% - 79% = B

60% - 69% = C

Under 60% = F

Course Description - Details

A. Basic Microbiology

1. Definition, Scope, and History of Microbiology

The student shall:

- define the terms "microbiology" and "microorganism" and indicate the scope of the science.
- know and list characteristics of plants and animals
- list with examples, characters which relate bacteria to the plant kingdom
- list with examples, characters which relate bacteria to the animal kingdom
- describe distinguishing characteristics for procaryotic and eucaryotic cells nuclear division, nuclear structure, cytoplasmic structure, flagella, chemical composition
- define Protista (Procaryota) and differentiate between higher and lower protists
- indicate the major contributions of Leeuwenhock, Pasteur, Koch, Jenner, Lister, Fleming
- state Koch's postulates

2. Anatomy of the Eubacterial Cell

The student shall:

- identify on a schematic drawing:

capsule (slime layer, microcapsule, sheath, envelope), cell wall, cell membrane, cytoplasm, nucleus, septum, flagellum, fimbrae, volutin granules, endospores, mesosomes, ribosomes

- state the function of these structures
- state the chemical composition of capsules, cell walls, cell membrane, flagella, ribosomes, nucleus, fimbrae, cytoplasm
- state the differences in the cell wall composition of gram positive and gram negative cells

- describe the shapes and location of endospores and name the bacterial genera which form endospores
- compare the characteristics of endospores and the vegetative cell
- describe the different forms of flagellation
- define "protoplast" and "spheroplast" and describe how these forms may be caused by penicillin and lysozyme
- describe with examples the following shapes and groupings assumed by bacteria: - bacilli, fusiform, spirochaete, spirillum, vibrio, streptococcus, tetracoccus, cubiodal arrangement, staphylococcus, streptobacillus, palisade arrangement
- state the relative sizes of cocci, bacilli and spiral cells

3. Bacterial Growth & Reproduction

The student shall:

- be able to discuss the meaning of bacterial growth
- describe the means of reproduction among microorganisms, especially that of transverse binary fission
- define generation rate
- describe the bacterial "growth curve"
- describe methods for the enumeration of bacteria, especially for viable counts

4. Bacterial Metabolism

- be able to describe, in an elementary fashion, the characteristics of enzymes
- differentiate between exoenzymes and endoenzymes
- define constitutive enzymes and induced enzymes
- relate catabolism and anabolism to exerg**o**nic and endergonic reactions
- define biological oxidation reduction
- indicate energy transfer compounds
- define aerobic respiration, anaerobic respiration, fermentation
- describe the nutritional types of bacteria

- list the nutritional requirements common to all biological systems and describe in an elementary fashion the roles played by these substances in bacterial nutrition and growth
- define optimum, minimum, and maximum growth temperatures
- describe the temperature requirements of psychrophilic, mesophilic, and thermophilic organisms
- discuss briefly the thermal resistance and susceptibility of bacteria
- describe the gaseous requirements of aerobic, anaerobic, facultative, and microaerophilic bacteria
 - describe the growth pH range and optimum pH for growth of pathogenic bacteria
 - briefly discuss how pH changes may occur in a bacterial culture
 - define buffer and indicate what substance(s) are most commonly used as buffers for bacterial culture
 - define hypertonic, hypotonic and isotonic solutions and describe the effect on bacteria of the first two
 - describe the effects on bacteria of dessication and vacuum describe the process of lyophilization
 - describe the effects of heavy metal ions on the growth of bacteria

5. Culture Media

- define the terms culture and culture medium
- describe the forms of culture media liquid, solid, solidreversible-to-liquid
- discuss agar in terms of source, chemical composition, melting and gelling points, characteristics making it suitable for bacteriological purposes, the different concentrations used with effects and purposes
- define synthetic or chemically defined medium, non-synthetic medium, living medium
- define with examples and describe differences between a basal, selective, differential, enriched, enrichment, transport or maintenance medium

- list the ingredients commonly found in nonsynthetic media and state their purpose:
 - peptones (state sources and how obtained, composition of, usual concentration)

- meat extracts and infusions

- yeast extract
 bile products
- carbohydrates
- dyes and indicators (pH, redox, selective agents, indicators of growth eg. T.T.C.)

- reducing agents

- antibiotics eg. V C N

- penicillinase and para-aminobenzoic acid (PABA)

- serum - collection and sterilization of

- blood - types commonly used, collection of, anticoagulants for, including Alsever solution, defibrination, Fildes' peptic digest of blood, usual concentration used in blood media

- activated charcoal

- list the principle ingredients and their function of the following media:

- blood agar

- MacConkey agar (with and without crystal violet)
- Salmonella Shigella agar (S.S.)

- Brewer's thioglycollate medium

- Chocolate agar

- Mannitol Salt agar
- Phenylethanol agar
- Triple sugar iron agar (interpret reactions)

- Selenite broth

- Stuart's Transport Medium
- describe the preparation and storage of culture media
- describe the process of tyndallization and indicate its uses and limitations
- describe inspissation and state two examples of media that are inspissated
- indicate how culture media are usually sterilized and describe the application of filtration for use with certain heat-sensitive ingredients
- list the pH indicators commonly used in culture media and describe the colour changes
- describe the colour comparator eg. the Lovibond comparator and discuss its use in the determination and adjustment of the pH of culture media

6. Pure Culture Study and Characterization of Bacteria

The student shall:

- define mixed culture, contaminated culture and pure culture
- indicate the importance of studying pure cultures
- be able to explain and discuss the application of the following for obtaining a pure culture-

- streak plate

- pour-plate qualitative and quantitativeselective and differential media
- pretreatment of the specimen
- antibiotics
- discuss and compare methods for the maintenance and preservation of pure cultures
- be able to list the major characteristics used to identify an unknown microorgainism: -

a) Growth Characteristics

- shall describe bacterial colonies on agar media by size, shape, elevation, colour, and margin
- snall describe bacterial growth in broth media by pellicle formation, turbidity, sediment

b) Microscopic Examination

- (i) shall describe the preparation, examination and application of a wet preparation and a hanging drop slide
 - shall describe the preparation and interpretation of a negative stain for capsules and name two solutions used for this procedure
 - shall know the principles and application of darkfield illumination in microbiology
- (ii) Gram stain and Ziehl-Neelsen acid-fast stain -
 - shall state the principles, reagents used, describe the techniques and controls, and interpret results
 - shall list two genera of bacteria that have acidfast properties
 - discuss general differences between gram-positive and gram-negative bacteria

(iii) Other staining procedures

- shall describe Albert's staining method for corynebacteria
- shall describe the Malachite Green method for staining spores

c) Other Characteristics

i) Biochemical reactions

- be able to discuss briefly the use of carbohydrate utilization tests (fermentative and oxidative) in the identification of an unknown microorgainism
- discuss briefly protein hydrolysis as shown by gelatin liquefaction

ii) Antigenic Structure

- be able to define the terms antibody, antigen, and serotype
- list bacterial components that are antigenic
- discuss briefly the use of antigenic structure for identification of an unknown microorgainism

iii) Bacteriophage Typing

- be able to define bacteriophage
- discuss in an elementary fashion the specificity and application of phage typing as used in identification

iv) Inoculation of Experimental Animals

- be able to briefly discuss the use of animals in the identification of an unknown microorganism

7. Anaerobic and CO₂ Cultures

- differentiate between an anaerobic environment and an environment containing an increased CO₂ content
- discuss the benefit or necessity of an environment containing increased CO 2
- list body sites where anaerobic microorgainisms constitute part of the normal flora

- describe the principle and operation of hot and cold catalyst anaerobic jar techniques
- know controls used for these jars
- describe the principle and limitations of using reducing agents in culture media and be familiar with thioglycollate and cooked meat medium
- explain the use of indicators of eH
- describe one method of achieving 5 10% CO2 atomosphere

8. Sterilization and Disinfection

I - Introduction

The student shall:

- be able to define sterilization, sterile, disinfectant, disinfect, antiseptic, sanitizer, germicide, bactericide, bacteriostasis, fungicide, virucide
- have an understanding of the necessity for more than one means for the control or destruction of microorganisms
- discuss the factors influencing the destruction of microorganisms - type of organism, temperature, moisture, concentration, time of exposure, contact, pH, physiological state of the cells, nature of the material containing the organisms

II - Physical Methods

a) Heat (dry and moist)

- i) be able to discuss the resistance and susceptibility of different microorganisms and spores to moist and dry heat
- ii) explain the principles involved in incineration, dry heat, steam under pressure, boiling, steaming at 100° C single and intermittent exposures, and pasteurization
- iii) list times and temperatures for methods in ii)
- iv) list potential sources of error for methods in ii)
 - v) describe sterility check procedures for dry heat and steam under pressure
- vi) identify on a diagram controls, major parts, and steam pathways in the autoclave

- vii) describe the loading and operation of ovens and autoclaves (manual and automatic types)
- viii) be able to choose the most suitable method for items to be sterilized

III - Filtration

The student shall:

- i) be able to explain the principle of sterilization by filtration
- ii) identify asbestos pad and membrane-type filters
- iii) describe the method of sterilizing fluids using filter
 types listed in ii)
 - iv) describe controls for filtration methods
 - v) list potential sources of error and limitations for filtration methods
 - vi) choose materials suitable for sterilization by filtration

IV - Irradiation with Ultra-Violet Light

The student shall be able to:

- i) explain the principle involved
- ii) list exposure times and conditions for effective sterilization
- iii) list and explain the hazards and limitations of U-V sterilization
 - iv) choose items suitable for U-V sterilization
 - v) describe sterility checking methods

V - Chemical Methods

- a) Chemical Disinfectants: For the principle types of disinfectants (phenolics, halogens, aldehydes, alcohols, quaternary ammonium compounds, surfactants) the student shall list:
 - i) mode of action
 - ii) effective concentrations
 - iii) times of exposure
 - iv) uses and limitations
- b) Chemical sterilizers: The student shall be familiar with the conditions and limitations of the use of ethylene oxide.

VI - Terminal Decontamination

The student shall list the types of materials requiring, prior to disposal, the following methods of decontamination:

- a) autoclaving
- b) incinerating
- c) chemical disinfecting

9. Classification (Taxonomy) and Nomenclature

- i) define order, family, genus, species, strain, mutant, type
- ii) know the prefixes used to denote an order, family, tribe
- iii) list the general characteristics on which bacterial classification is presently based. (eg. as used in Bergey's Manual of Determinative Bacteriology, 8th edition)
 - iv) list characteristics (morphological and other) of viruses, mycoplasmas, rickettsia and chlamydia
 - v) indicate the culture medium (cellular or non-cellular) required for the groups of microorganisms listed in iv)
- vi) name some human diseases caused by organisms listed in iv)