MICRB265

General Microbiology

3 Credits

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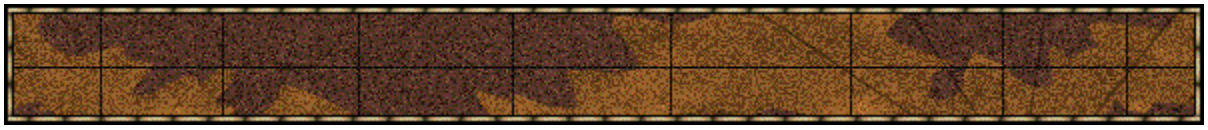

General Microbiology

Calendar Description

This course focuses on the structure and physiology of free-living and pathogenic bacteria. The diversity of their metabolic activities, the interaction of microbes with their environment, symbiotic relationships and cell-to-cell communication are major topics. Lectures and laboratory exercises are coordinated to explore topics in basic microbiology, environmental microbiology, molecular microbiology, and the production of economically or medically important products through microbial biotechnology.

Rationale

MICRB 265 introduces the student to the world of microscopic life, in particular that of bacteria. The ultrastructure, chemistry and function of bacterial cell components, bacterial motility and chemotaxis, mibrobial diversity, nutritional types and metabolism, biogeochemical cycles, pathogenic and symbiotic relationships, genome recombination and replication, gene expression, regulation and transcription, antibiotics and resistance, molecular cloning and microbial biotechnology are some major topics. In the laboratories, emphasis is placed on practical work skills and safe handling of bacteria.

MICRB 265 is the key prerequisite for advanced studies in microbiology, and is needed for several advanced level courses in other fields. The course is of interest to students in agriculture and forestry, arts, elementary and secondary education, human or veterinary medicine, pharmacy, and science.

Prerequisites

BIOL107 and CHEM161

Co-Requisites

None

Course Learning Outcomes

Upon successful completion of this course, students will be able to (cognitive skills)

1. distinguish cells based on their ultrastructural and biochemical components in the three domains and six kingdoms of life.
2. classify bacteria, archaeans, viruses, viroids, prions, some fungi and protistans based on structural, biochemical, nutritional, toxin-resistance tests, or molecular signature sequences.
3. explain the target of stains of subcellular components such as inclusions, including taxonomic stains, such as the Christian Gram stain, and various types of microscopic imaging techniques.
4. epitomize the nomenclature (Latin, genus, species), based on classical cell shape and biochemical methods, or on molecular DNA or RNA signature sequences.
5. outline microbial defenses against osmotic shock (peptidoglycan), chemical lysis, or predator and host attacks (capsule, M-protein); adhesion to substrates (fimbriae, capsule) and host cells.
6. describe the prokaryotic cytoskeleton and cellular motor (basal body) control, chemotaxis, the diversity of cell motility via flagella, axial filaments, gliding, twitching (type IV pili), ratcheting, slime propulsion, or gas vesicles.
7. compare the peptidoglycan wall chemistry, the periplasmic space in Gram negative versus Gram positive bacteria, and the action of penicillin antibiotics or lysozyme enzyme.
8. contrast cytoplasmic membrane with the Gram negative outer membrane lipids, porin protein permeability, pyrogenic lipopolysaccharide endotoxins; biofilms, extra- and intracellular pathogens.
9. itemize Robert Koch's postulates that link a pathogenic organism to an infectious disease (tuberculosis), nosocomial infections, antimicrobial agents and widespread resistance.
10. discuss the primary nutritional types based on carbon and energy source, the function of enzymes in anabolism and catabolism, bioassay growth optima, thermophiles, psychrophiles, acidophiles, halophiles.
11. assign reactants or products to metabolic pathways, such as glycolysis, Entner-Doudoroff pathway, fermentation (e.g. mixed acid, 2,3-butanediol, homolactic), Krebs cycle, Calvin cycle, electron transport chain, chemiosmosis, hydrothermal vents.
12. list ATP-generating reactions as substrate-level, oxidative or photophosphorylation, in aerobic or anaerobic respiration and oxygenic or anoxygenic photosynthesis, relative to the presence or production of oxygen.
13. specify microbial sewage treatment processes, obligatory anaerobic respiration, autotrophy, and methanogenesis, rumen cellulytic syntrophy and H_2 transfer, bioremediation of oil spills, oxygenases in aliphatic and aromatic hydrocarbon degradation.
14. quote the bacterial growth curve (lag, log, stationary phase, death and lysis), quorum sensing, binary fission, recombination, antibiotic secondary metabolites, cell differentiation, endospores.

15. define autotrophy, endosymbiosis, methanogens and methylotrophs, gene expression, operons, *Rhizobium* nitrogen fixation and legume root nodules, *Agrobacterium* and plant crown gall tumors.
16. formulate procedures in bioengineering, molecular cloning, biotechnology, biodegradable plastics, probiotics, botox, *Bacillus thuringensis* insecticide.

Upon successful completion of this course, students will be able to (applied skills)

17. recognize the colony morphology of pure and mixed bacterial cultures by their form, elevation, margin, surface, color and transparency on TCS, MCA, and EMB agar plates.
18. name the parts of the microscope, and correctly adjust a light microscope, bright or dark field, phase-contrast, live or stained bacterial smears, which includes the use of immersion oil.
19. draw or photograph bacteria viewed under the microscope, calculate their real size using micrometer scales, the magnification of the image, and the resolution of the lens.
20. isolate, streak out and culture bacterial cells on solid agar medium or in liquid broth culture, using an inoculation loop or Pasteur pipet, flaming the flask lip, replacing lid or cotton plug under sterile conditions.
21. stain bacteria to reveal subcellular structures, for example Gram stain, acid fast stain (demo), malachite green endospore stain, Einar Leifson flagellum stain, or capsule stain.
22. identify to genus some of the more common microorganisms according to cell shape, arrangement, Gram-positive or Gram-negative staining, size, specific enzymatic reactions in tests, and growth on differential solid media.
23. search bioinformatics databases such as GeneBank (BLAST) or Ribosomal Database Project, to match a DNA sequence for 16S rRNA of known and named species to a signature sequence new unknown isolate.
24. calculate original total microbial cell numbers from direct Petroff-Hausser cytometer counts or indirect turbidity measurements, and cell viability by counts of colony-forming units of microbial cells cultured at serial dilution.
25. tabulate cell shape, cell size, and cell arrangement of Gram-stained microbes from biofilms, such as dental plaque, rock or leaf surfaces, gut lining, and from plain or probiotic yogurt.
26. write laboratory reports that include title, abstract, introduction (hypothesis), methods, results (graphs, standard curves, tables), discussion (conclusions), citations in APA format from database searches or secondary sources.
27. incubate aseptic alfalfa *Melilotus sativus* seeds in plastic pouch cultures inoculated with rhizobia (*Sinorhizobium meliloti*), observe root nodulation and bacteroids; set up anaerobic bacterial cultures.
28. quantify nitrogen fixation by gas chromatography based on nitrogenase activity, using acetylene gas as an artificial substrate instead of the ubiquitous natural nitrogen gas.

29. monitor fecal indicators in ground beef by standard plate counts of *Escherichia coli* CFU, and count of *Enterobacter faecalis, Staphylococcus aureus*, and *Salmonella* after enrichment culture; exo- and enterotoxin predictions.
30. determine the relative effectiveness of antimicrobial agents against common bacteria in the human environment by the zone of inhibition in the Kirby-Bauer disc diffusion assay, using phenol solution as a standard.
31. decrease of ammonification by increased acidity in Peptone broth cultures of soil samples (unknown bacteria), known bacteria (*Bacillus cereus, Pseudomonas putida*), and sterile control plates, tested with the Nessler reagent.

Resource Materials

Required Texts:

Madigan M. T., Bender K. S., Buckley D. H., Sattley W. M., and Stahl D. A. (2021). *Brock biology of Microorganisms* (16th ed.). Hoboken, New Jersey, U.S.A.: Pearson Education Inc.

Boucher, Y., and Mah, R. (2020-2021). *Microbiology 265, Introductory Microbiology. Laboratory manual.* Department of Biological Sciences, University of Alberta, Edmonton, AB.

Cuny, R. (2021). MICRB 265 General Microbiology. Course notes and online if given. Lakeland College, Lloydminster, AB.

Reference Texts:

Garrity, G.M. (Editor-in-Chief). 1984-2012. Bergey's manual of systematic Bacteriology 2nd ed. Volume 1, Archaea and basal phototrophs, R.D. Boone. (Editor); Volume 2, Proteobacteria, D.J. Brenner, N.R. Krieg, J.T. Stanley. (Editors); Volume 3, Firmicutes, A.C. Parte. (Editor); Volume 4, Bacteroides, Spirochetes, Chlamydiae, Planctomycetes, A.C. Parte. (Editor); Volume 5, Actinobacteria, A.C. Parte (Editor). Springer, New York, NY.

Holt, J.G. (Editor). 1994. Bergey's manual of determinative Bacteriology. 9th ed. Lippincott, Williams, and Wilkins, Baltimore.

Conduct of Course

This is a 3 credit course with 3 hours of lecture and 4 hours of lab per week. (3-0-4).

Lectures - Three hours per week

The lectures are supported by PowerPoint data projection, white board, Smartboard, and occasionally by a movie. The printed course notes and electronic files placed on Desire-2-Learn must be supplemented by notes taken by the students. The library can be used to access the biological literature and on-line databases. Students are expected to do the assigned reading in the textbook and lab manual on a weekly basis. Students will recognize the importance of microbes in our world, their vast functional diversity, and their incredible potential in biotechnology.

Labs - Four hours per week

In the laboratory the students perform experiments on their own or they work in groups. Chemical and biological safety rules must be adhered to, and l**ab coats must be worn when in the lab**. The objective is to train the students in the safe handling, culture, and containment of potentially pathogenic bacteria, and to let them perform biochemical tests. For some procedures students must wear dust masks, face shields, goggles, surgical gloves, and they must be familiar with oil-immersion microscopy, containment hoods, inoculation, incubation, processing and storage of bacterial cultures. The scheduled lab time is often insufficient to complete the experiments, and students are expected to complete the work during posted open lab times.

In addition to the weekly worksheets, three lab activities are graded: database primary literature search (Lab 3), and two full lab reports (Lab 3 and Lab 5). All students must have passed a lab safety quiz (by Lab 2) before continuing with labs. Although the laboratory work may be performed in groups of up to 4 students, each student is responsible for an independent data analysis, and individual interpretation of the results. Citation of primary research follows a scientific format (APA).

The WHMIS Workplace Hazardous Materials Information System requires the safe handling and storage of chemicals, as specified in the MSDS Materials Safety Data Sheets. Microorganisms on Schedule 2 of the Human Pathogens and Toxins Act fall under the regulations of the Pathogens Regulation Directorate of the Public Health Agency of Canada, and are to be handled under supervision of qualified staff, must be fully contained, and shall not be disposed of in the regular waste, or poured down the drain. All laboratory equipment is operated as specified in the Operation Manuals.

Evaluation Procedures

The student's performance is evaluated in terms of percentage points that reflect the number of correct answers out of the total number of questions asked on exams. The final mark is the aggregate of the evaluations; however, students must achieve a mark of 50% or higher in the

laboratory component, which includes the laboratory safety quiz (for new students only), practical work and biosecurity observations, worksheets, a draft, laboratory reports, laboratory quizzes, and laboratory practical final exam combined.

The weighting of the course components is as follows:

Laboratory Component:		
Laboratory Safety Quiz (For new students only; you must pass.)	0%	
Worksheets and Assignments (6)	8%	
Laboratory Quizzes (2)	4%	
Laboratory Reports (2)	8%	
Laboratory Practical Work and Biosecurity	5%	
Laboratory Practical Final Exam	15%	
	40%	
Lecture Component:		
Lecture Quiz	5%	
Mid-term Lecture Exam	20%	
Final Lecture Exam	35%	
	60%	
Total		**100%**

The lecture exams are composed of a 2:1 mixture of multiple choice questions and short answer questions. **No supplemental assignments or exam re-writes are allowed in the University Studies Department**. The Final Laboratory Exam is a practical exam about the laboratory experiments; stations are set up and students take turns answering question at each station. In the Laboratory Practical Exam component students demonstrate that they can safely culture, isolate, serially dilute, fix and stain bacteria, as well as prepare media. The laboratory reports do not exceed 2 pages single spaced, excluding drawings, lists, tables, or graphs, and they are due in two weeks after the assignment. They follow a scientific format: title, author's name and address, abstract, brief introduction with hypothesis, methods, results with tables or graphs, discussion, reference list (APA), and they answer questions posed in the lab manual or on the worksheets. Worksheets provide a record of the data collected and their analysis, and they are due in one week. Late submissions of assignments suffer a 5% deduction per day on the mark, except under documented extraordinary circumstances. Database literature searches are guided by the librarian.

Cheating, falsification or fabrication of laboratory data, plagiarism, and non-compliance with all safety regulations or the code of conduct are academic and professional offences. Depending on the severity of the offence, a student may be reminded, sent out of the class room or laboratory, reported to the department head, may have marks deducted, assigned a failing grade in the course, or may be expelled from the college.

Grade Equivalents and Course Pass Requirements

A minimum grade of D (50%) (1.00) is required to pass this course.

Letter	F	D	D+	C-	C	C+	B-	B	B+	A-	A	A+
Percent Range	0-49	50-52	53-56	57-59	60-64	65-69	70-74	75-79	80-84	85-89	90-94	95-100
Points	0.00	1.00	1.30	1.70	2.00	2.30	2.70	3.00	3.30	3.70	4.00	4.00

Students must maintain a cumulative grade of C (GPA - Grade Point Average of 2.00) in order to qualify to graduate.

Attendance

Regular attendance is essential for success in any course. Absence for any reason does not relieve a student of the responsibility of completing course work and assignments to the satisfaction of the instructor. Poor attendance may result in the termination of a student from a course.

If you do not meet the established attendance requirements, your instructor will recommend that the Registrar withdraw you from the course. A failing grade of RW (Required to Withdraw) will appear on your transcripts.

Instructors have the authority to require attendance at classes.

1. All labs are mandatory. If more than 1 lab is missed, excused or unexcused, the student will either be required to withdraw (RW) or will be assigned a failing grade (F) for the entire course.
2. Students are only allowed to hand in laboratory worksheets, or laboratory reports for labs that they have attended.

   If the student's absence is excusable, the missing laboratory work will not be counted. If the absence is inexcusable, the laboratory work will be assigned a mark of 0.
3. Make-up laboratories are difficult or impossible to set up. It depends on the lab missed, but only students with an excused absence can sometimes attend a make-up lab, at the discretion of the lab technician and lab instructor.
4. Not wearing a lab coat and required attire, not having the current lab manual, or violating lab safety regulations will ban you from the laboratory, and this will result in an inexcusable absence.

Course Units/Topics

Week	Type	Title: MICRB 265 GENERAL MICROBIOLOGY
I. MICROBIAL CELL STRUCTURE AND FUNCTION, ROLE IN THE ENVIRONMENT		
1	Lec 1	Introductions, course format, goals, labs; some new discoveries
	Lab 0	Microbiology laboratory safety orientation (LAB SAFETY QUIZ for new students only)
	Lec 2	history of microbiology; origin of the 3 domains of life, prokaryotes, eukaryotes
	Lec 3	Microscopes, cell shape, arrangement, size; Gram stain and other stains
2	Lec 4	Cell membranes and walls, outer and inner membranes, periplasm, transport of molecules in and out of cells
	Lab 1	Laboratory procedures; bacterial colonies, cell morphology, and arrangements; microscopy, and stained bacterial smears (prepared examples)
	Lec 5	Cell walls, $Gram^+$ and $Gram^-$; peptidoglycan and pseudopeptidogycan, S-protein layer, pili, fimbriae, capsule, porins
	Lec 6	Cell differentiation, endospores, heterocysts; motility, flagella, axial filaments, gliding, soaring, slime propulsion, gas vesicles, chemotaxis, tropism
3	Lec 7	Osmosis, storage granules, polymer inclusions, stains, membrane-bound organelles: mesosomes, magnetosomes, chlorosomes, thylakoids, and chromatophores
	Lab 2	Aseptic technique and containment, isolation of bacteria from a mixture, subcultures; planning of isolation of bacteria from the environment
	Lec 8	Microbial growth, wall septum, bactoprenol, bacterial cytoskeleton (FtsZ, MreB, crescentin); binary and multiple fission, nucleoid, plasmids, ribosomes, recombionation, gene regulation review
	Lec 9	Clonal colony growth (CFU, lag, log), chemostat, growth optima; sterilization (autoclave, flame, UV), containment, chemical disinfectants
II. MICROBIAL COLONY GROWTH AND METABOLISM, BIOGEOCHEMICAL CYCLES		
4	Lec 10	Genomic sequencing, Sanger, next generation, metagenome, transcriptome, proteome, metabolome, biofilm, microflora
	Lab 3	Identification of $Gram^+$ and $Gram^-$ bacteria by biochemical taxonomy; selective and differential culture; colony growth characteristics
	Lec 11	LECTURE QUIZ; nutritional types, energy (photo-, litho-, chemotroph), carbon sources (auto-, organotroph), micronutrients

	Lec 12	Obligate and facultative aerobes and anaerobes, fastidious microbes, hyperthermophiles, acidophiles, halophiles
5	Lec 13	Viruses: virion, capsid, receptor, viral classification and diversity, lytic and lysigenic cycles, virulence and latent infections. viroids, prions
	Lab 4	LAB QUIZ 1; finish bacterial identification; home environmental survey, media preparation, and collecting tools picked up; dental plaque biofilm tools lined up
	Lec 14	Metabolic diversity, enzymes, redox potential, glycolysis, fermentation, aerobic and anaerobic respiration, citric acid cycle, electron transport chain, proton-motive force, ATP synthase
	Lec 15	Photosynthesis, light-harvesting complexes, bacteriorhodopsisns, oxygenic or anoxygenic; carbon fixation, carboxysomes, Calvin cycle, reverse Krebs cycle, hydroxypropionate pathway, acetyl-coenzyme A pathway
6	Lec 16	Biotechnology, metabolism of lipids, proteins, carbohydrates including cellulose, hydrocarbons, phenolics including lignin, vitamins, syntrophic consortia, bioreactors
	Lab 5	Home environmental survey and dental plaque biofilm, colony and cell morphology: heat-fix, Gram stain, and microscopy; effect of acidity on ammonification tube culture set up
	Lec 17	Methanogenesis, methylotrophs, interspecies hydrogen transfer, biogeochemical cycle of carbon, petroleum and aromatic hydrocarbons, bioremediation
	Lec 18	Nitrogen fixation, symbiotic rhizobia in plant root nodules, assimilative and dissimilative nitrate reduction, biogeochemical cycle of nitrogen, anammoxosomes, nitrification and denitrification, ammonification
7	Lec -	-
	Lab -	MIDTERM READING WEEK
	Lec -	-
	Lec -	-
8	Lec 19	Biogeochemical cycle of sulfur, hydrothermal vents, hot springs, geysers; biogeochemical cycle of iron, other metals, and ore leaching
	Lab 6	Effect of acidity on ammonification: ammonia test with Nessler's reagent; yogurt production: homolactic and heterolactic fermenters, probiotics
	Lec 20	MIDTERM LECTURE EXAM

III. Normal Human Microbiome, Invasion Barriers, Pathogenicity Factors, Innate and Adaptive Immunity		
	Lec 21	Normal human microbiome: skin, oral, digestive, respiratory, and urogenital systems, natural barriers, dental plaque biofilm
9	Lec 22	Pathogenesis, virulence factors, acute and chronic disease, attenuation, opportunistic pathogens in compromised host, adherence, AB-type, cytolytic, superantigen exotoxins, enterotoxins, endotoxins and fever
	Lab 7	Yogurt data analysis, Gram stain; activity spectrum of antibiotics on various bacteria, using the Kirby-Bauer test; anaerobic culture demonstration
	Lec 23	Innate immunity, myeloid phagocytes, pathogen-associated molecular pattern PAMP, toll-like receptors, inflammation, fever, all cells antigen-presenting via major histocompatibility complex MHC I, complements, opsonization, natural killer cells
	Lec 24	Adaptive immunity, lymphoid T (thymus) and B (bone marrow) cells, MHC II, differentiate to produce antigen-specific immunoglobulins, memory cells, plasma cells, passive/active immunity
IV. CLINICAL MICROBIOLOGY, PATHOGENS, TOXINS, and HUMAN DISEASES		
10	Lec 25	Disease diagnostics, containment, biosecurity, personal protective equipment, nosocomial infection, safe sample collection, biochemical tests, specimen culture on selective or differential media, DNA or RNA tests
	Lab 8	LAB QUIZ 2; analysis of Kirby-Bauer test: zone of inhibition; bioinformatics: 16S rRNA and rDNA signature sequences of unknown bacteria (GeneBank database BLAST)
	Lec 26	Vaccination: ajuvants, synthetic, genetically engineered, or conjugate vaccines, monoclonal antibodies
	Lec 27	Antibiotics: antibacterial, antifungal, antiprotistan, antiviral drugs: action spectrum, intracellular target, antibiotic resistance, bacteriophage injection
11	Lec 28	Epidemiology, pandemics, hygiene, immunization, water filtration, chlorination, globalization
	Lab 9	Meat-borne pathogens, enterotoxins, standard plate count: CFU/gram (colony-forming units); total count with the Petroff-Hausser cytometer
	Lec 29	Person-to-person bacterial and viral diseases, including airborne, contact, and sexually transmitted diseases.
	Lec 30	Vectorborne or zoonotic and soilborne diseases
12	Lec 31	Waterborne and foodborne parasitic, bacterial and viral diseases: coliform test, water treatment, botulism, food poisoning, norovirus

	Lab 10	End of meat-borne pathogen analysis; symbiotic nitrogen fixation in legume root nodules: squashed, heat-fixed, and nigrosine-stained
	Lec 32	Eukaryotic parasites: host alternation, infection cycles; protistans, fungi and worms; travel, water filtration, treatment
V. MICROBIAL DIVERSITY AND CLASSIFICATION		
	Lec 33	Microbial taxonomy and evolution, homologs, paralogs, horizontal gene transfer; Domains Eukarya (eukaryotes), Archaea, and Bacteria (prokaryotes)
13	Lec 34	Domain Bacteria: stem-thermophiles, phylogeny, ultrastructure; KingdomsAquificae to Deinococci and Chloroflexi, Chlamydiae to Planctomycetes
	Lab 11	FINAL PRACTICAL LABORATORY EXAM
	Lec 35	Domain Bacteria: $Gram^+$ Kingdoms Firmicutes to Actinobacteria, Cyanobacteria, Spirochaetes, Bacteroidetes, Chlorobi to Proteobacteria
	Lec 36	Domain Archaea: phylogeny, ultrastructure; Kingdoms Thaumarchaeota (nitrogen), Crenarchaeota (sulfur thermophiles), Korarchaeota (filamentous), Nanoarchaeota (ectoparasitic), Euryarchaeota (methanogens)
14	Lec 37	Domain Eukarya: serial endosymbiosis, phylogeny, ultrastructure; Kingdoms Protista, Fungi, Plantae, Animalia, microbial examples only
	Lab -	-
	Lec -	FINAL LECTURE EXAM WEEKS
	Lec -	-
15	Lec -	-
	Lab -	-
	Lec -	-
	Lec -	-

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