Miami Dade College

Course Description

BSC4422L | Biotechnology Methods and Applications - III Lab | 2.00 credits

This course provides students with hands-on laboratory experiences to supplement the BSC4422 lecture course. Students will learn how to perform advanced molecular biotechniques that build on previous knowledge. They will perform diagnostic assays, western blots, purifications, etc. and determine how to correlate findings with the basic research or clinical data. Prerequisites: BSC2427, 2427L, PCB3060, 3060L, BCH3023, 3023L. Corequisite: BSC4422.

Course Competencies:

Competency 1: The student will demonstrate knowledge of methods for manual and automated nucleic acid purification and characterization by:

1. Performing a manual DNA and RNA extraction from a sample specimen
2. Quantitating purified nucleic acid samples by UV spectrophotometry
3. Explaining the principles of gel and capillary electrophoresis of nucleic acids
4. Performing a gel-based electrophoretic analysis of DNA

Competency 2: The student will demonstrate knowledge of the elements of different methods based on the PCR reaction by:

1. Explaining the principles of the polymerase chain reaction (PCR)
2. Describing steps in the PCR reaction
3. Listing different applications of PCR (including reverse-transcriptase, multiplex, and quantitative PCR) in hematopathology, cancer, and infectious diseases
4. Carrying out an RT-PCR analysis for the molecular detection of chromosomal translocations associated
5. Performing a real-time PCR quantification of DNA with acute and chronic leukemia

Competency 3: The student will demonstrate knowledge of restriction analysis, physical mapping, and southern hybridization of DNA by:

1. Describing the mechanism of action of restriction enzymes
2. Listing the applications of restriction enzymes in the molecular diagnosis of hematopathology and other pertinent pathology specimens
3. Describing the following methods for nucleic acid detection: staining, blotting, hybridization, and fragment length polymorphism analysis

Competency 4: The student will demonstrate basic knowledge of tissue cell culture techniques by:

1. Comparing healthy, senescent, and transformed cells cultured in vitro
2. Explaining the processes of cellular division and growth about the cell cycle and how internal and external factors influence these processes
3. Listing current methods used for the development of primary cell cultures in vitro
4. Comparing the use of in vivo and in vitro cultures for the establishment, propagation, and/or growth of cell, tissue, and organ cultures
5. Discussing protocols for cellular enrichment and characterization in cell cultures
6. Explaining the development of cell lines from primary cell cultures
7. Discussing the use of cell lines as model systems for standard and disease states

Competency 5: The student will demonstrate knowledge of tissue protein purification and concentration by:

1. Discussing methods for the selective enrichment and/or over-expression of desired proteins
2. Evaluating preparative and analytical methods for the isolation and quantization of proteins
3. Listing procedures to reduce and/or control protein degradation or contamination

4. Explaining at least three distinct selective methods useful for protein purification and identifying the basis of separation for each type
5. Differentiating between the methods used for detecting and separating proteins
6. Surveying the methods for extraction, filtration, precipitation, and desalting of proteins
7. Using centrifugation to separate precipitated proteins from the supernatant
8. Discussing the principles and operation of dialysis, defiltration, ultrafiltration, and tangential flow filtration
9. Analyzing the solubilities of proteins for isolation purposes

Competency 6: The student will demonstrate knowledge analysis and characterization of proteins by:

1. Discussing the practical aspects of electrophoresis, such as staining and detecting bands, protein blotting, and analysis of the result
2. Describing the basis of polyacrylamide gel electrophoresis (PAGE), SDS (sodium dodecyl sulfate) PAGE, native gel electrophoresis, isoelectric focusing, and 2D electrophoresis
3. Comparing native gels and denaturing gels, non-reducing gels and reducing gels, gradient gels and straight gels, and between stacking gels and resolving gels
4. Interpreting the banding patterns from any of the listed gel types or combinations of gel types
5. Performing western blotting detects proteins that react with specific antibodies
6. Comparing the spectroscopic methods to determine protein concentration
7. Using spectroscopic techniques to determine protein concentration
8. Discussing hazards associated with gel electrophoresis and the purpose of different ingredients in the gel recipes and in sample preparations

Competency 7: The student will demonstrate knowledge of DNA molecular diagnosis by:

1. Explaining how the nucleotide sequence of DNA is determined
2. Listing applications of DNA sequencing to molecular diagnosis
3. Describing the method and interpreting results of the molecular diagnostic test for immunoglobulin heavy chain and t-cell receptor gene rearrangements
4. Describing the clinical settings in which various molecular tests are ordered, including the distinction between diagnostic testing and minimal residual disease testing, i.e., Detection of KRAS and EGFR mutations in colon and lung cancer. Patient outcome predictors in AML: FLT3, NPM1 and BRAF mutation analysis
5. Analyzing and interpreting molecular diagnostic data for hematopathology, genetics, and other pertinent pathology specimens; including appropriate reporting and medical significance of positive and negative test results
6. Correlating molecular diagnostic data with morphology and clinical information in diagnosing hematopathology, genetics, and other pertinent pathology specimens
7. Integrating molecular diagnostic results in hematopathology, genetics, and other pertinent reports

Learning Outcomes:

- Use quantitative analytical skills to evaluate and process numerical data
- Solve problems using critical and creative thinking and scientific reasoning
- Describe how natural systems function and recognize the impact of humans on the environment