Course No: BIOG523   Course Title: Advanced Applied Microbiology
Duration: 3 hours   Marks: 100   Date: 15.12.2023

NOTE: Submit answer sheet of CB and take fresh sheet for OB. Attempt all questions and their subparts in sequence. Be precise and answer in points. Give examples wherever possible.

Closed Book (Dur: 1.5 hr)

Section-A

Q1. Study the adjacent table and tell which organism most easily causes an infection. Justify. **(1.5)**

Bacterium	ID$_{50}$
E. coli O157:H7	20
Legionella pneumophila	1
Shigella	10
Treponema pallidum	57

Q2. Explain in brief five antibiotic resistance mechanisms adopted by bacteria. **(2.5)**

Q3. Explain in brief the difference and connection between local and focal infection. **(3)**

Q4. What do you understand by epidemiological bathtub? Represent it with the help of a diagram. Explain in brief each of its components. **(6)**

Q5. Choose the best possible option. Answer any 10 questions **(0.5x10 = 5)**

1) At high cell density, _____ blocks the yeast to filamentous transition in *Candida albicans*.
     A. Mevalonate   **B.** Farnesol   **C.** Sequalene   **D.** Autoinducer II

2) *S. aureus* utilizes a standard two-component Gram-positive QS system where a _____ binds to _____ triggering a phosphorylation cascade that ultimately controls the expression of virulence genes.
     A. ArgC; AIP   **B.** ArgD; AIP   **C.** AIP; ArgC   **D.** Autoinducer II; ArgD

3) *S. mutans* hinders *C. albicans* germ tube production_____
     A. even when physically separated, through the secretion of CSP
     B. only when in physical contact
     C. through the production of AI-2 that inhibits the germ tube protein
     D. when physically separated, through the secretion of AI-2

4) Bacteria uses _____ for trafficking hydrophobic autoinducers within a population.
     A. Membrane transporters
     B. Outer membrane vesicles
     C. Contact-dependent secretion into neighbouring bacteria
     D. Conjugation tubes

5) QS pathway allows *Vibrio* phage (VP882) to produce an antidepressant called ____ that promotes the lysis of host cells by interfering with the prophage inhibitory factor
     A. Qtip   **B.** VqmA   **C.** AHL   **D.** AI-2

6) Bacterial _____ play a significant role in halting primary root growth and enhancing lateral branches
     A. CSP   **B.** AHL   **C.** AI-2   **D**. AIP

7) Bacterial species use tit-for-tat method that is _____

A. contact-dependent communication system exhibited by T6SS
B. contact-independent communication system exhibited by effectors of T6SS
C. contact-dependent communication system exhibited by T3SS
D. contact-independent communication system exhibited by effectors of T3SS

8) QS can be inhibited by all except_____
    A. Targeting signalling molecules   B. Targeting signal biogenesis
    C. Targeting signal detection   D. Targeting signal transduction

9) Under low nitrogen condition, *S. cerevisiae* use the Ehrlich pathway, which consists of all except _____ steps to produce specific amino acids.
    A. Transamination   B. Decarboxylation   C. Reduction   D. Hydroxylation

10) Release of QS molecules occurs through_____
    A. Passive diffusion only   B. Active secretion only
    C. Both passive diffusion and active secretion   D. Active ATP-dependent transport only

11) _____ undergoes a conformational change upon binding of bacterial QS molecules, thereby enhancing its translocation into the nucleus.
    A. ArgC   B. AHR   C. GPCR   D. ARNT

Section –B (It is Quiz-2, not a part of Compre)

1. The ID_{50} is
A) a measure of pathogenicity.
B) the dose that will cause an infection in 50 percent of the test population.
C) the dose that will kill some of the test population.
D) the dose that will cause an infection in some of the test population.

2. Endotoxins are
A) associated with gram-positive bacteria.   **B)** molecules that bind nerve cells.
C) part of the gram-negative cell wall   **D)** excreted from the cell.

3. The fimbriae of *Neisseria gonorrhea* and enteropathogenic *E. coli* are examples of
A) adhesins.   **B)** ligands.   **C)** receptors   **D)** adhesins and ligands.

4. Endotoxins in sterile injectable drugs could cause
A) infection.   **B)** septic shock symptoms.   **C)** giant cell formation.   **D)** nerve damage.

5. In which of the following patterns of disease does the patient experience no signs or symptoms?
A) prodromal **B)** decline   **C)** convalescence   **D)** incubation **E)** incubation and convalescence

6. Koch observed *Bacillus anthracis* multiplying in the blood of cattle. What is this condition called?
A) bacteremia   **B)** focal infection   **C)** local infection   **D)** septicemia

7. If a prodromal period exists for a certain disease, it should occur prior to
A) incubation   **B)** illness.   **C)** decline.   **D)** convalescence.

8. Koch's postulates can be utilized in _____
A) determining the development of antibiotic resistance in a hospital diagnosis lab
B) discovering a new antibiotic-producing bacteria in a pharmaceutical lab
C) determining the causative agent of a patient's illness in a hospital microbiology lab
D) formulating a vaccine against a new pathogen in a genetic engineering lab

9. Which of the following statements best defines an altruistic act?
A. benefits the performer and another individual
B. benefits another individual at some cost to the performer
C. benefits another, related individual at some cost to the performer
D. imposes a cost on the performer and another individual
E. imposes a cost on the performer without benefiting another individual

10. Which of the following statements are true for combinatorial approaches?
 i. Approaches targeting several pathways in combination
 ii. Approaches operating on similar target in combination

   A. Only i   **B.** Only ii   **C.** Both i and ii   **D.** None is correct.

Section-C

**Q6. Answer the following questions with appropriate justification.**

(i) Out of rhizospheric, rhizoplanic, and endophytic bacteria, which will be the most efficient biofertilizer candidate and why? **(4)**

(ii) What do you mean by Induced systemic tolerance? Elaborate one example. And, how does it differ from induced systemic resistance? **(3)**

(iii) What is global transcription machine engineering? Explain and highlight the advantages. **(5)**

Section- D

Q7. (i) As per the following dilution and plating scheme, calculate the total number of colony-forming units per ml of original stock. **(1.5)**
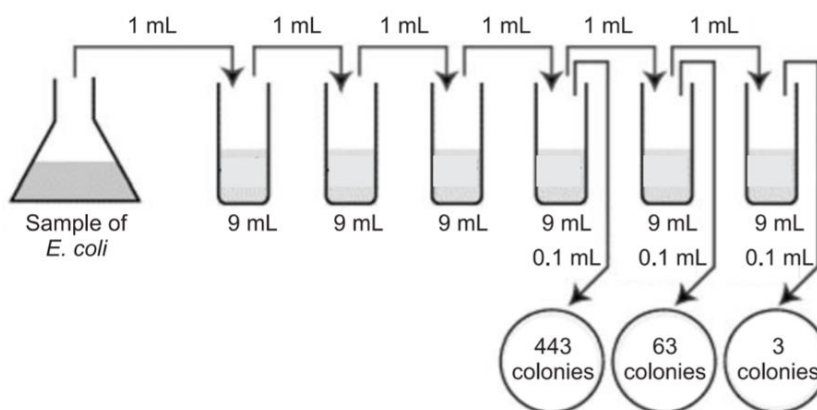
(ii). 20 Phosphate Solubilizing Bacterial strains were incubated in LB nutrient broth overnight at 180 rpm, 28 °C, and 1 mL of the culture medium of each PSB strain was centrifuged at 5000 rpm for 2 min, washed three times with sterile saline solution for bacterial cell collection and used for genotyping by amplification of ERIC-PCR. However, no bands were detected on the PCR gel. Can you enlist reasons as to what could have gone wrong? **(1.5)**

(iii) Discuss the principle of EMB agar media. **(2)**
(iv) What is the molarity of a 38% solution of HCl with a density of 1.20 g/ml? **(1)**

Q8. An experiment is designed to evaluate the degradation of biphenyl by GC-MS after inoculation of a strain P1 of *Pseudomonas* in a minimal medium (MM).
   A. For the PCB-77 assay, the inoculating minimal medium is supplemented with 50 mg/l biphenyl dissolved in _____. **(1)**
   B. Biphenyl degradation potential of the strain P1 can (increase/decrease/remain unchanged) after the addition of 0.2% glucose as a co-substrate to the Minimal medium. **(1)**
   C. A calibration curve of Biphenyl/PCB-77 is plotted using different concentrations of standard solutions (0 to 20 ug/ml; y = 0.0160x+0.0047, R^2 = 0.9897). Find the quantity of biphenyl/PCB-77 in a sample that gives a measured absorbance of 0.264 when blank for the same experiment has an absorbance of 0.095. **(1)**
   D. Name two solvents commonly used for extraction of biphenyl form MM media post-inoculation. **(1)**

PART A

Q1. Answer the following in brief.
  (i) During the recent COVID pandemic, we witnessed an increase in SARS-CoV2 infections and deaths in the second wave. Based on the content covered in the lectures, comment on the probable logical reason(s) for the same. Justify. *Please exclude the cases of insufficient medical supplies like oxygen from your answer.* **(3)**
  (ii) Thanks to the vaccination, the spread of the infection was controlled. Imagine if we were not privileged to have vaccines. Do you think over a period, the disease could have spread and wiped out the human population altogether? Justify your answer. **(3)**

Q2. Study the graphs obtained from infection studies carried out in mice and comment on what can we elucidate from this data. Would it have any clinical relevance? Justify. **(4)**
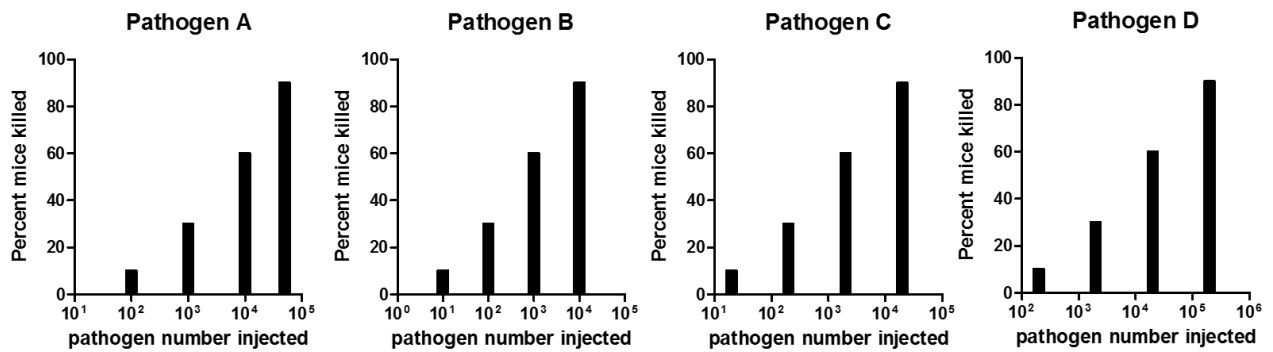
Q3. A research scholar in her first year is studying microbial pathogenesis. Looking at the results of her experiments, the supervisor interprets that the pathogen behaves as if *it is born to run*. Based on your understanding from the course, elaborate on why he made such a comment. Explain the molecular basis. **(6)**

Q4. After a few days of pacemaker insertion surgery, the patient was kept on antibiotic therapy for a week to treat bacteremia. However, every subsequent month, he suffered from recurrent bacteremia for which he received the same antibiotic therapy. The physician advised him to (i) have food rich in pro and prebiotics and (ii) replace the pacemaker to solve the problem. Do you think, his advise is wise and has long-term implications on the patient's overall health? Justify. **(4+4)**

PART B

Q5. You have isolated a bacteria from an extreme environment. On 16S rRNA sequence analysis, it shows closest similarity with *Halomonas elongata* having 90% match on performing BLAST. Can you infer it belongs to given species? What additional molecular characterization you should perform for in-depth molecular characterization and accurate identification (other than genome sequencing). In case, you find the given isolate to be novel, what steps are required for its taxonomic classification? **(5)**

Q6. With the development in synthetic biology, some scientists have been making attempts to synthesize non-natural nucleotide base pairs and have been successful in doing so. Cite few examples. How will it be useful in Biotechnology? What are the limitations and challenges to execute at the functional level? **(6)**

Q7 (i) Following data is obtained from the rhizosphere of corn plant. What ecological information do you get from following data w.r.t diversity of bacteria? **(3)**

Species	Sample A	Sample B	Sample C
Azotobacter	10000	320	0
Azospirillum	11000	15	0
Rhizobium	9500	115	0
Bacillus	11200	25000	0
Pseudomonas	0	237	54298
Azoarcus	9800	600	12
Acenetobacter	12100	31213	5

(ii) In 1977, many animal forms were discovered proliferating well in a hydrothermal vent, which was much below the sea surface where light was not available. What type of nutrition majorly drives the existence of all these organisms? **(4)**

Q8. Many bacteria are known to promote plant growth under abiotic stress, such as salt stress. Despite this fact, there are few PGPR that are used as biofertilizers to promote plant growth and ameliorate salt stress under actual farming conditions. Mention major stress amelioration mechanisms and challenges in using such bacteria as effective PGPR. Also, suggest a strategy for enhanced and sustainable agriculture employing microbial inoculation under such stress. **(8)**

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