## JAYPEE UNIVERSITY OF INFORMATION TECHNOLOGY, WAKNAGHAT TEST -3 EXAMINATIONS-2022

M.Sc-II Semester (BT)

COURSE CODE (CREDITS): 20MS1BT211(3)

MAX. MARKS: 35

COURSE NAME: Genetic Engineering COURSE INSTRUCTORS: Dr. Anil Kant

RS: Dr. Anil Kant MAX. TIME: 2 Hours

Note: All questions are compulsory. Marks are indicated against each question in square brackets.

Q. 1

- A. You want to use an pET series vector for the expression of recombinant protein in E.coli. This protein also negatively affects the growth of *E.coli*. Illustrate how pET vectors regulate the expression of recombinant protein at un-induced and induced state.
- A. Demonstrate how "Yeast episomal plasmid Vector" and "Yeast integrating plasmid vector" works. Figure out a basic difference between these. Which one of these is expected to produce a more stable yeast cell line. Mention the reason supporting your choice of answer.
- B. Compare the contrast between shuttle and binary vectors? Discuss functional components of Ti plasmid based plant transformation vectors and their working procedure in plant transformation.
- Q. 2
  A. Value the innovation which made it possible to carry out PCR reaction in a single tube in modified Sangger's sequencing.
  - B. Let you try to express a transgene, sourced from eukaryotic organisms in E. coli. But the level of expression is not up to the mark. Based on your learning, priority wise enlist three factors along with reasons, which you would consider for enhancing the level of expression? 2
- Q.3
  A. Let you are trying to express a recombinant protein in E.coli. During the experimentation you came to know that recombinant protein is accumulating in the form of inclusion bodies. Interpret why such inclusion bodies form. Suggest and explain with examples at least two strategies which can be adopted to express this protein in E. coli in biological active form.
  - B. Let a recombinant protein be tagged with histidine or maltose binding tag. Demonstrate how it can be purified.
- Q.4
  - A. Calculate the minimum number of clones required in a gene library of *rice*? Given genome size  $5.7 \times 10^2$  MB, fragment size 10 KB, desired probability of finding the fragment 0.95.
  - B. Suggest method to accomplish the following during cDNA library construction. i) mRNA isolation ii) Second strand synthesis of cDNA 2
  - C. Let you interested in preparing a gene library of a eukaryotic species for gene isolation? Which type of library would you prefer to construct? Give any four reasons for your choice?

Q.5 Do any four of the following.

 $2 \times 4 = 8$ 

- A. CRISPR Cas is a better gene editing tool compared to earlier protein based tools. Mention two arguments?
- B. What role do following components play is CRISPR cas systems i) Cas proteins ii) tacr RNA iii) spacer or guide RNA
- C. Draw a well labeled general design of RNAi delivery or CRISPR delivery vector and mention key points about the design.
- D. Mention two key points in support of RNAi mediated gene silencing could be a better tool as compared to traditional gene knockout specially when targeting a gene of multigene family or polyploid species.
- E. Why blue colored roses could not be developed by just over-expression of missing gene function of 3'-5' Flavanone hydrolase, and required gene silencing of its DFR gene also. Mention only three quick points.