Genetics and Genomics in Medicine Chapter 3 Questions

Multiple Choice Questions

Question 3.1

Which of the following statements, if any, is false?

a) Amplifying DNA means making many identical copies of one or more starting DNA sequences.
b) The object of DNA cloning is to amplify DNA.
c) The object of PCR is to amplify DNA
d) The object of DNA sequencing is to amplify DNA

Question 3.2

Which of the following statements, if any, is false?

a) Amplifying DNA always requires a DNA polymerase.
b) Amplifying DNA must be carried out in bacterial or yeast cells.
c) DNA cloning in cells involves attaching the DNA to be cloned to a vector DNA to form a recombinant DNA that may be circular or linear.
d) Vector molecules in DNA cloning must have a replication origin that confers the ability to replicate extrachromosomally.

Question 3.3

Which of the following statements, if any, is false?

a) Cloning DNA in bacteria typically requires a plasmid or a bacteriophage vector.
b) Plasmids and bacteriophages are both small circular double-stranded DNA molecules.
c) Plasmids and bacteriophages each contain a replication origin that allows them to replicate independently of the bacterial chromosome.
d) To be useful as vectors plasmids and bacteriophages need to be genetically modified.

Question 3.4

Which of the following statements, if any, is false?

a) The polymerase chain reaction (PCR) is a cell-free method of DNA amplification.
b) PCR is usually used to amplify a specific DNA sequence of interest using oligonucleotide primers that bind to closely flanking sequences.
c) PCR is superior to cell-based DNA cloning for two major reasons: it is much quicker and it allows much greater DNA amplification.

d) PCR requires the use of a heat-stable DNA polymerase to make copies of the template DNA.

Question 3.5

With respect to nucleic acid hybridization, which, if any, of the following statements is false?

a) The probe is a labeled nucleic acid or oligonucleotide that is expected to hybridize to a target DNA sequence within an unlabelled test nucleic acid population.
b) The probe is a known single-stranded nucleic acid or oligonucleotide that is intended to hybridize to complementary target sequences in a poorly understood nucleic acid test sample.
c) The object of a hybridization assay is to identify target sequences within a test sample that are related to the probe so that some new information is gained about the target sequences.
d) During a hybridization assay, a heteroduplex is formed by un-natural base pairing between complementary probe and target sequences that show a sufficiently high degree of base pairing across part or all of their lengths.

Question 3.6

With respect to nucleic acid hybridization, which, if any, of the following statements is false?

a) The strength of base pairing between a probe and a complementary target sequence depends on the number of stable base pairs that are formed.
b) Among other parameters, the hybridization stringency depends on the salt concentration and temperature of the hybridization reaction.
c) To identify a target sequence that is distantly related to the probe, high stringency hybridization needs to be used.
d) Under conditions that favor low hybridization stringency long heteroduplexes with significant base mismatching may be stable.

Question 3.7

Nucleic acid hybridization assays are normally carried out under relaxed hybridization stringency (to maximize the chances of heteroduplex formation) but afterwards, washes are carried out that can be designed to favor perfectly matched sequences only by changing some parameter. Which, if any, of the following changes would be consistent with that aim?

a) An increase in temperature.
b) An increase in salt concentration.
c) An increase in the concentration of a polar molecule, such as urea or formamide.

Fill in the Blanks Questions

Question 3.8

Fill in the blanks below.

In cell-based DNA cloning, a DNA population of interest (which consists of very long DNA fragments) needs to be cleaved by a _____1_____ ____2_____ into manageably short DNA pieces that can be transported more easily into cells. The resulting DNA fragments are joined by a ___3____ ____4_____ to a ____5____ DNA, resulting in the formation of a ____6____ ___3___. The ___5___ carries a replication origin that allows it, and the ____6____ ___3____ , to replicate within a suitable host dell, usually some type of ____7____ or ____8____ cell.

Question 3.9

Fill in the blanks below.

In cell-based DNA cloning a key step is ____ 1____ , the stage when the DNA of interest enters the provided host dells. The ____1_____ efficiency is usually very low, but when ____ 1____ occurs just a ___2____ DNA molecule usually enters the cell. As a result, a complex starting DNA population can be fractionated by the cells (which effectively act as sorting offices). A second key step allows identification of ____3_____ cells (in the case of bacteria, the host cells are genetically modified to be sensitive to some ____4____ , and the vector carries a gene conferring ____5____ to the ____4____) . Many of the transformed cells contain just the vector DNA instead of the desired ____5____ DNA. To identify a specific ____5____ DNA, a more specific assay is required that often involves ____6_____ using a closely related labelled DNA or RNA ____7____.

Essay and Lists Questions

Question 3.10
List four parameters that affect the stability of a heteroduplex and describe how they have an effect

Question 3.11
During PCR, each cycle has three defined steps. What are they, and what is involved?

Question 3.12
In cell-based DNA cloning two types of enzyme are critical for making recombinant DNA. What are they, and what roles do they carry out?

Question 3.13
Some restriction endonucleases cut DNA to produce *sticky ends.* What is meant by "sticky ends", and why are the restriction endonucleases that produce them so valuable for DNA cloning.

Question 3.14
In cell-based DNA cloning using plasmids, it is usual to use a plasmid that will allow maximum amplification (increase in copy number) of a recombinant DNA. Sometimes, however, that is not the aim. Explain why.

Question 3.15
Most PCR reactions are intended to amplify a specific DNA sequence of interest from within a complex starting DNA, often a genomic DNA sample. In addition to a sample of starting DNA of this type in an appropriate buffer with the correct ions to sustain the reaction, list four additional key requirements of the reaction to be successful.

Question 3.16
Describe the three recognized phases of a PCR reaction.

Question 3.17

What is the distinction, if any, between quantitative PCR and real-time PCR?

Question 3.18

Nucleic acid hybridization assays require one of the interacting nucleic acid populations to be labelled in some way. Why is that required and what does it involve?

Question 3.19

What is the essential difference between the Sanger dideoxynucleotide sequencing method and massively parallel (= next generation) DNA sequencing?

Question 3.20

Outline the different approaches to fractionating DNA using gel electrophoresis.