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mutation. Finally, bacteria do not perform many of the posttranslational modifications that occur in eukaryotes, so the protein would not be processed properly.

Section: 5
Level of Difficulty: difficult

44) How could site-directed mutagenesis be used to investigate the kinetic properties of an enzyme?

Answer: If the gene for an enzyme is changed by site-directed mutagenesis, the relationship between structure and function can be determined. For example, altering a key amino acid to one with a very different ionic character would likely cause a change in enzymatic activity.

Section: 5
Level of Difficulty: difficult

45) Describe the human disease that has been successfully treated by gene therapy.

Answer: Children with SCID (severe combined immunodeficiency disease) received bone marrow transplants containing cells with a vector harboring the proper cytokine receptor.

Section: 5
Level of Difficulty: moderate
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responsible for the defect and possibly to identify a protein product. A region of DNA
was identified on chromosome 7, and further work made it possible to deduce the
putative structure and function of the protein (a membrane channel for ions) from its
gene sequence. Thus, identification of the mutation causing cystic fibrosis was found
by identifying and characterizing the gene.

Section: 4, Box 3-2
Level of Difficulty: difficult

40) Describe the cloning vectors that would be used to clone each of the following DNA
fragments: 1000 bp cDNA, 8000 bp eukaryotic gene fragment, 100,000 bp segment of a
chromosome.

Answer: A plasmid would be used to clone the small cDNA fragment. Plasmids are small,
replicate easily, contain multiple restriction sites for ligation, and carry genes such as
antibiotic resistance. Bacteriophages (such as \( \lambda \)) are used for larger fragments up to
16kb. The recombinant molecules are packaged into phage particles for growth and
maintenance. Very large pieces of DNA containing hundreds of kb are cloned into
bacterial or yeast artificial chromosomes (BACs or YACs). These structures replicate
and segregate during cell division.

Section: 5
Level of Difficulty: moderate

41) Assume you have successfully cloned a small (50 bp) fragment of DNA into the lacZ site
of a pUC18 cloning vector. Describe the appearance of transformed colonies you would expect
to see on each of the following plates: plain media, media containing ampicillin, media
containing tetracycline, media containing ampicillin and X-Gal.

Answer: Colorless colonies would grow on the plain media, media containing ampicillin, and
media containing ampicillin and X-Gal. No colonies would grow on the media
containing tetracycline. None of the plates would have blue colonies. However, a
control plate of pUC18-transformed cells (without the 50-bp insert) grown on media
with ampicillin and X-Gal would show blue colonies.

Section: 5
Level of Difficulty: difficult

42) Does a wild-type gene differ from a normal gene?

Answer: No. A wild-type gene is the form commonly found in nature.

Section: 5
Level of Difficulty: moderate

43) Would a genomic clone of a eukaryotic protein be properly expressed in a bacterial cell?

Answer: No, a genomic clone of a eukaryotic protein would not be expressed properly, if at all,
for several reasons. The clone might contain control sequences not utilized by the
bacterial system. Furthermore, the bacterial cell could not splice out introns, so an
expressed protein might contain additional sequences, and/or possibly a frame shift
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can specify the sequence of a poly[peptide (translation) but that genetic information does not flow from polypeptide sequences to RNA or DNA (see Figure 3-13).

Section: 3
Level of Difficulty: moderate

36) Why are Type II restriction enzymes preferred for laboratory manipulation of DNA?

Answer: Type II restriction enzymes recognize and cleave within a specific sequence of bases, forming smaller fragments with defined sequences at their ends. Types III and I do not cut within the recognition sequences and so are less useful for making recombinant DNA.

Section: 4, Box 3.1
Level of Difficulty: difficult

37) What are RFLPs? Provide an example of how they are used in research and/or diagnosis.

Answer: RFLP is the acronym for Restriction Fragment Length Polymorphism. In RFLP analysis, fragments of homologous DNA that vary in their number of restriction sites are digested with restriction enzymes, and the fragment sizes are compared. This technique can be used to determine paternity or identity (using hypervariable regions). Certain genetic diseases can be identified and characterized, such as mutations that cause alterations in the cleavage site (sickle cell anemia) or that cause an increase (Huntington’s disease) or loss (muscular dystrophy) of DNA within the target region.

Section: 4
Level of Difficulty: difficult

38) Draw a restriction map for a DNA given the following information about the sizes of DNA fragments produced from restriction digests.

<table>
<thead>
<tr>
<th>Enzyme</th>
<th>Size of fragments produced (kb)</th>
</tr>
</thead>
<tbody>
<tr>
<td>BamHI</td>
<td>10 kb</td>
</tr>
<tr>
<td>EcoRI</td>
<td>2.8, 2.9, 4.3</td>
</tr>
<tr>
<td>HindIII</td>
<td>5</td>
</tr>
<tr>
<td>PstI</td>
<td>1, 9</td>
</tr>
<tr>
<td>EcoRI + HindIII</td>
<td>2.1, 2.2, 2.8, 2.9</td>
</tr>
<tr>
<td>EcoRI + PstI</td>
<td>1, 1.9, 2.8, 4.3</td>
</tr>
<tr>
<td>HindIII + PstI</td>
<td>1, 4, 5</td>
</tr>
</tbody>
</table>

Answer: 0 1 2.9 5 7.2 10kb
/PstI/EcoRI/HindIII/EcoRI/
(BamH1 does not cleave this fragment. This fragment can be drawn in either orientation.)

Section: 4
Level of Difficulty: difficult

39) Summarize the search for the molecular defect in cystic fibrosis?

Answer: Unlike diseases such as sickle cell anemia, for which a protein defect was known, cystic fibrosis had no known molecular defect. The experimental approach was to compare DNA of patients and family members in order to identify the region of DNA
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Short Answer Questions
Write your answer in the space provided or on a separate sheet of paper.

31) The molecule ATP plays many roles in organisms. Describe some of the processes in which ATP participates.

Answer: ATP is a component needed to make RNA and DNA and other molecules. It is also an energy carrier critical to metabolic processes, and it can influence enzyme catalytic activity.
Section: 1
Level of Difficulty: difficult

32) What information about DNA was discovered from the compositional analysis performed by Erwin Chargaff?

Answer: Chargaff identified the ratios of specific bases. He discovered that there is a 1:1 ratio of A to T and C to G; these ratios are known as Chargaff’s Rules.
Section: 2
Level of Difficulty: easy

33) What evidence from the lab of Rosalind Franklin did Watson and Crick use to formulate their model of DNA?

Answer: Rosalind Franklin and colleagues had performed X-ray crystallography on DNA samples, and the photos indicated that the Watson-Crick model of DNA was correct. The X-ray diffraction photographs of DNA fibers that Rosalind Franklin and her colleagues obtained indicated that DNA is double helical and that the bases are stacked on one another.
Section: 2
Level of Difficulty: difficult

34) Describe the major features of the DNA double helix.

Answer: The DNA double helix contains two antiparallel strands of DNA, wound in a right-handed helix. The bases are found on the inside, stacked along the axis of the helix, and the sugar-phosphate chains are located on the surface of the helix. The strands form a major and minor groove that run the length of the molecule. The bases are hydrogen bonded in complementary pairs of A:T and G:C.
Section: 2
Level of Difficulty: difficult

35) What is the central dogma of molecular biology?

Answer: The central dogma of molecular biology was proposed by Crick to explain the relationship between DNA, RNA and protein. It indicates that DNA can specify its replication and its transcription to form complementary RNA molecules, and that RNA
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27) Alterations in genetic information are caused by:
   A) point mutations
   B) faulty recombination
   C) transposition of genes
   D) A and B
   E) A, B and C

Answer: E
Section: 4
Level of Difficulty: moderate

28) In order to perform PCR, the following reagents must be included:
   A) DNA fragment, primers flanking the region of interest, dNTPs, DNA Polymerase
   B) DNA fragment, primers flanking the region of interest, dNTPs, ddNTPS, DNA Polymerase
   C) DNA fragment, one primer, dNTPs, DNA Polymerase, DNA ligase
   D) DNA fragment, primers flanking the region of interest, dNTPs, DNA Ligase
   E) none of the above

Answer: A
Section: 5
Level of Difficulty: moderate

29) Which of the following statements about PCR is(are) true?
   A) Small amounts of DNA can be easily amplified to millions of copies.
   B) PCR is used in forensic and clinical diagnosis.
   C) PCR reaction products can be used in cloning
   D) A and B
   E) A, B and C

Answer: E
Section: 5
Level of Difficulty: difficult

30) A gene knockout is:
    A) a gene that has been inactivated or removed from an organism.
    B) a dominant gene that “knocks-out” expression of other genes.
    C) a gene inserted in place of another gene.
    D) a particularly attractive gene.
    E) none of the above

Answer: A
Section: 5
Level of Difficulty: moderate
The most common tautomeric form of the purine and pyrimidine bases in nucleic acids is the:

A) enol form
B) imino form
C) keto form
D) ester form
E) none of the above

Answer: C

Which of the following DNA sequences is (are) palindromic?

A) AGCT
B) AAGNCTT
C) AGGA
D) A and B
E) A and C

Answer: D

DNA sequencing using the Sanger method requires:

A) template, primer, DNA polymerase, mRNA, dNTPs, ddNTP
B) template, primer, DNA polymerase, dNTPs, ddNTP
C) template, primer, DNA polymerase, rRNA, dNTPs, ddNTP
D) template, primer, DNA polymerase, mRNA, dNTPs
E) none of the above

Answer: B

Which of the following conclusions about the human genome project are true?

A) Only about 28% of the genome is transcribed to RNA, and only about 1-2% codes for protein.
B) Most of the proteins found in humans are unique to vertebrates.
C) Nearly half of the human genome consists of repetitive DNA sequences.
D) A and C
E) A, B and C
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17) A set of DNA fragments made from the entire genome of an organism is called a ______ library.

Answer: genomic
Section: 5
Level of Difficulty: easy

18) A set of DNA fragments made from the expressed sequences found in a particular cell of an organism is called a ______ library.

Answer: cDNA
Section: 5
Level of Difficulty: easy

19) The process of transferring colonies from a master culture plate to plates containing differing types of growth media is called _____.

Answer: replica plating
Section: 5
Level of Difficulty: moderate

20) To express a particular protein, the gene is cloned into a(n) ______.

Answer: expression vector
Section: 5
Level of Difficulty: moderate

Multiple Choice Questions

21) What group is attached to the ring in thymine and is not found on uracil?
   A) ribose
   B) CH₃
   C) NH₃
   D) OCH₃
   E) none of the above

Answer: B
Section: 1
Level of Difficulty: easy

22) Nucleotides contain phosphate groups bonded to the:
   A) C3’ or C5’ atoms
   B) C3 or C3’ atoms
   C) C5 or N9 atoms
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10) E Section: 5 Level of Difficulty: easy

Terms not used: L) purine, H) exons

Fill In Questions

11) The concentration of cellular ATP is approximately _______.
Answer: 5 mM
Section: 1
Level of Difficulty: moderate

12) The structure of the DNA helix suggests that each strand can serve as a _____ for synthesis of the other strand.
Answer: template
Section: 2
Level of Difficulty: easy

13) RNA can base-pair intramolecularly by forming ______ structures.
Answer: stem-loop
Section: 2
Level of Difficulty: easy

14) If the DNA sequence read directly from the dideoxy sequencing gel is 5′CCATCG3′ then the template sequence is _______.
Answer: 5′CGATGG3′
Section: 4
Level of Difficulty: moderate

15) The greater complexity of vertebrates is partially due to their gene structure, which allows genes to undergo differential ______, so that various options in RNA processing can create diverse protein products.
Answer: gene-splicing
Section: 3,4
Level of Difficulty: moderate

16) Infectious phage particles growing on a bacterial lawn form a small circlet of lysed bacteria, called a _______.
Answer: plaque
Section: 5
Level of Difficulty: moderate
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Matching Or Fill In
Choose the correct answer from the list. Not all the answers will be used.

1) Cytosine, uracil, and thymine are structural derivatives of ______.
2) Nucleotides are linked by a ______ bond.
3) Human genomes have two sets of chromosomes, and are ______ organisms.
4) Experiments by Avery and colleagues proved that DNA was the substance that ______ a non-pathogenic pneumococcus strain to a virulent strain.
5) The method that Sanger developed for sequencing DNA using dideoxy nucleotides is called the ______ method.
6) Plasmids are small, circular DNA molecules that are used in ______.
7) Certain plasmids contain genes for resistance to ______.
8) A small fragment of labeled DNA or RNA used in hybridization is called a ______.
9) After hybridization, the fragment of interest can be detected by ______.
10) Eukaryotic genes often contain regions that are transcribed and later excised from the RNA, called ______.

Answers 1-10:
1) D Section: 1 Level of Difficulty: easy
2) B Section: 2 Level of Difficulty: easy
3) A Section: 2 Level of Difficulty: easy
4) G Section: 3 Level of Difficulty: easy
5) K Section: 4 Level of Difficulty: easy
6) F Section: 5 Level of Difficulty: easy
7) I Section: 5 Level of Difficulty: easy
8) C Section: 5 Level of Difficulty: easy
9) J Section: 5 Level of Difficulty: easy