

Your name:

---

---

1. (3 points) You isolate DNA from yeast that has been arrested in G1 (prior to the initiation of DNA synthesis) or in G2 (prior to the initiation of mitosis). In separate reactions of the same volume you allow DNA from the same number of cells (e.g.  $10^8$ ) from each preparation to denature and hybridize. You measure hybridization rate as the time required for half of the DNA to become double-stranded (not the initial reaction rate).

**d) DNA from the G2-arrested cells would hybridize faster by a factor of 2**

2. (3 points) You repeat this experiment using the same amount of DNA (e.g. exactly 100 micrograms) from each sample. Once again, you use the same volume. This time

**c) DNA from the two samples would hybridize at the same rate** (you should be able to see that there ought to be no difference between these samples).

(3 points each for questions 3-28). In each of the following there are two or more statements. One is true (generally, it is taken directly from your textbook or another reliable source) and the others have been modified so as to be untrue, misleading, false, wrong, unlikely or bogus. Circle, check or otherwise designate the correct statement. Ambiguous marks (checking both, placing a mark between the two statements, etc.) will be considered wrong.

3. **a)** Human alpha globin and mouse beta globin are **homologs**.
4. **b)** In *E. coli*, *trpA* and *trpB* are two **genes**.
5. **b)** The bacterial origin of DNA replication can "fire" **before** the prior round of DNA replication is complete.
6. **a)** In conjugation, donor DNA is **transferred directly** to the recipient through a connecting tube.
7. **b)** The recipient of (successful) transfer from an Hfr strain **remains F<sup>-</sup>**.
8. **a)** Common plasmid vectors are capable of carrying up to **15 kb** of foreign DNA.
9. **b)** A single base mismatch in an allele-specific oligonucleotide used in **hybridization** will provide maximal discrimination when located **in the center** of the oligonucleotide.
10. **b)** A single base mismatch in an allele-specific primer used in **PCR amplification** will provide maximal discrimination when located **at the 3' end** of the primer.
11. **b)** **Microsatellites** are highly polymorphic DNA markers that are useful in linkage studies.
12. **a)** CODIS, the combined DNA index system, makes use of **STR** (short tandem repeats) polymorphisms.

Your name:

---

---

13. a) In PCR-based mutagenesis methods desired sequence changes are designed into the 5' end of the PCR oligonucleotides.
14. a) The initiating event for meiotic recombination is a double-strand break generated by a cellular protein.
15. b) For automated sequencing, the Sanger protocol is performed with all four individually labeled terminating nucleotides present in a **single** reaction.
16. a) New methods for the rapid and inexpensive sequencing of entire genomes are under active development and **dramatic improvements are expected**.
17. a) **DNA footprinting** uses end-labeled DNA to map the position of protein binding sites.
18. c) The human genome is about 1,000 times the size of bacterial genomes.
19. a) Mammals have **less than 15** times as many genes as typical bacteria.
20. b) In shotgun sequencing, fragments are sequenced using a common oligonucleotide primer that is complementary to a vector sequence that is immediately adjacent to a **random and unknown insert**.
21. a) **Intron** sizes vary more between species with genomes of different sizes (e.g. humans vs. Arabidopsis)
22. a) **Myc/Max heterodimers** bind to DNA and activate the transcription of genes that control growth but **Max/Max homodimers bind without activating**.
23. b) The **shotgun sequencing** protocol is a reasonable and commonly used approach to sequence whole genomes.
24. b) Long terminal repeats are found in mobile genetic elements that use **reverse transcription** during their replicative cycle.
25. b) Cytosine deamination in DNA is repaired by a mechanism involving **base** excision repair.
26. a) A lysogenic bacterium **carries a prophage**.
27. a) Different **sigma** factors allow *E. coli* RNA polymerase to recognize different promoters.
28. b) A core promoter **has little or no** transcriptional activity *in vivo* in the absence of additional regulatory sequences.

**Your name:**

---

---

29. (8 pts.) Rank the following types of DNA with respect to the fraction of the human genome it makes up (place a 1, 2, 3 or 4 next to each type of DNA where 1 indicates the largest amount of DNA, 2 the next largest, 3 the third and 4 the least abundant class of DNA).

- a) 4 Sequences that code for amino acids.
- b) 1 Non-LTR retroelements such as SINEs and LINEs.
- c) 2 LTR-retrotransposon and retroviral proviruses.
- d) 3 Conserved (and therefore presumably functional) noncoding sequences.

30. (8 pts.) Explain why CG dinucleotides are so rare in mammalian DNA and why they are more frequent in the vicinity of active genes.

4 points: Why CG dinucleotides are rare. Deamination of C to form U is a frequent spontaneous mutation. Normally, the resulting U residue is recognized and removed by base excision repair (the sequential action of uracil N-glycosylase, AP endonuclease, DNA polymerase and finally, DNA ligase). However, CG dinucleotides are a frequent site of methylation. 5-methyl cytosine when deaminated yields T, which is not recognized by the repair pathway. Thus, CG dinucleotides have the highest mutation rate of all nucleotides due to deamination of <sup>5m</sup>CG to TG.

4 points: CB dinucleotides are more frequently found near active genes because CG dinucleotides near active genes are less likely to be methylated (methylated DNA is a characteristic of inactive DNA) and therefore less likely to be mutated by the mechanism just described. Another reason (either answer was accepted) is that mutations near active genes are more likely to be eliminated by selection while mutations in other CG dinucleotides are more likely to be neutral.