

PLEASE WRITE LEGIBLY. IF THE GRADER CANNOT READ YOUR ANSWER IT WILL BE MARKED WRONG.

Multiple choice questions (20 pts/ 1 pt each) Please write the letter for the correct answer in the appropriate space. (circled answers will not receive credit)

82

- ✓ d 1. What event typically initiates homologous DNA recombination?
- A site-specific nick in one strand of duplex DNA
 - A site specific double strand break in duplex DNA
 - A random (not site specific) nick in one strand of duplex DNA
 - A random (not site specific) double strand break in duplex DNA
- ✓ a 2. In homology directed repair, single-strand invasion
- occurs when a free 3' single stranded region invades a region of a homologous duplex
 - occurs when a free 5' single stranded region invades a region of a homologous duplex
 - results in the generation of heteroduplex RNA
 - allows replication fork to bypass damaged bases
- C X 3. Integration of Lambda DNA into the *E. coli* genome involves
- homologous recombination into recombination hot spot
 - Xis, a serine recombinase
 - site-specific recombination between attB and attP
 - site specific recombination between attL and attR
- C X 4. Which of the following describes nucleotide excision?
- a single damaged nucleotide is removed and replaced with a new nucleotide
 - a single damaged base is removed and replaced with a new base
 - a region of single stranded DNA containing damaged nucleotides is removed and replaced with new DNA
 - a region of double-stranded DNA containing damaged nucleotides is removed and replaced with new DNA
- B X 5. Which of the following is feature common to an insertion sequence?
- ORF encoding reverse transcriptase
 - Short inverted terminal repeats
 - long terminal repeats
 - Chi sequence
- ✓ a 6. All of the following are ways cells minimize the potentially harmful effects of transposition EXCEPT
- immunoglobulins bind to the transposon-encoded proteins
 - ✓ differential splicing of transposase restricts activity to specific cell types
 - ✓ transposon sequences are methylated
 - ✓ piRNA base pairs with transposon mRNA
- ✓ a 7. NonLTR transposons that encode their own reverse transcriptases are
- long interspersed nuclear elements (LINES)
 - short interspersed nuclear elements (SINES)
 - composite transposons
 - P elements
- ✓ d 8. _____ have the intrinsic ability to excise and transpose.
- Transposase
 - Somatic controlling elements
 - nonAutonomous controlling elements
 - Autonomous controlling elements

- ☒ ~~B~~ ~~A~~ 9. What is the role of the 12bp and 23bp spacers in gene recombination of immunoglobulin variable (V-D-J) segments?
- a. prevents improper class switching
 - ☒ b. prevents a gene segment from recombining with another of the same type
 - c. promotes formation of IgG vs IgE
 - d. prevents premature termination codons in the generated ORF
- ☒ ~~d~~ 10. How does an *E. coli* cell attempt to replicate damaged DNA during SOS response?
- a. all DNA synthesis is stopped until the damage can be repaired via NHEJ
 - b. regions of damaged DNA are deleted from the genome
 - c. messenger RNA is converted to DNA which is inserted at the damaged sites by recombination
 - ☒ d. nucleotides are incorporated at random at the damaged sites via TLS polymerase
- ☒ ~~C~~ ~~X~~ 11. What event initiates the process of yeast mating type switching?
- ~~a.~~ A double strand break made at the MAT locus by Spo11 endonuclease
 - ☒ b. A single strand nick made at the MAT locus by Spo11 endonuclease
 - c. A double strand break made at the MAT locus by HO endonuclease
 - d. A single strand nick made at the MAT locus by HO endonuclease
- ☒ ~~b~~ 12. Deamination of cytosine in DNA results in
- a. A-T base pair
 - b. U-G base pair
 - c. G-C base pair
 - d. T-U base pair
- ☒ ~~C~~ 13. Which of the following is a common cause of errors in genome replication?
- a. formation of G-U base pairs at the replication fork
 - ☒ b. replication of regions of the genome that are being transcribed
 - c. a tautomeric shift within a nucleotide in the template DNA
 - d. the presence of nucleosomes attached to the DNA being replicated
- ☒ ~~A~~ 14. Glycosylases function by
- a. cleaving the bond between the base and the deoxyribose, leading to removal of the base
 - b. cleaving the ring structure of the nucleotide, leading to its removal
 - c. nicking the phosphodiester backbone on both sides of a base, leading to its removal
 - d. opening up the deoxyribose sugar ring, leading to its removal
- ☒ ~~C~~ ~~X~~ 15. Ultraviolet radiation causes which type of DNA damage?
- a. extensive single stranded DNA
 - b. base hydrolysis
 - c. pyrimidine dimers
 - d. base deamination
- ☒ ~~b~~ 16. Why does it make sense that the p53 gene is the most commonly mutated gene found in cancer cells?
- a. the p53 gene mutation actively promotes the uncontrolled growth of cancer cells
 - ☒ b. the p53 protein is responsible for initiating apoptosis of cells with DNA damage
 - c. the p53 protein is supposed to alert the immune system of any misgrowth in cells
 - d. the role of p53 in cancers is not clear

- A** ~~X~~ 17 Which of the following statements about CSSR is TRUE?
- CSSR resolvases are sequence specific topoisomerases
 - Cre recombinase, a serine recombinase, promotes recombination between lox sites
 - Cre recombinase requires accessory proteins to promote recombination between lox sites
 - Cre recombinase can only excise the region between a pair of lox target sites

- ✓ d 18. Which of the following describes the activity of the bacterial RecA protein?
- DNA helicase with 5'-3' polarity
 - DNA helicase with 3'-5' polarity
 - degrades DNA from the 3' end at a nick
 - coats single-stranded DNA and catalyzes homologous strand invasion

- ✓ a 19 AP endonucleases recognize what type of mutation in DNA?
- an abasic site created by the breaking of glycosidic bonds and loss of a base
 - purine dimers that result from UV damage
 - deamination
 - alkylated pyrimidines

- B** ~~X~~ 20. Ethidium bromide is a common intercalating agent used to visualize DNA in electrophoresis gels by associating with the DNA in the gels and fluorescing under UV light to reveal bands. How does ethidium bromide associate with the DNA?
- EtBr is charged end interacts with the polar backbone of DNA
 - EtBr inserts between the stacked bases in the DNA double helix
 - EtBr covalently bonds to nitrogen rich proteins associated with the DNA
 - EtBr forms hydrogen bonds with the guanines and cytosines in the DNA

21-25 (2 pts each) All statements are FALSE. Explain why the statement is false.

21. All DNA damage results from the action of chemical or physical mutagen

DNA damage can be caused by ~~the~~ many natural cellular pathways and metabolism.

22. Bacteriophage recombinases such as lambda integrase and Cre recombinase require host proteins for site specific recombination at target sites

the recombinases require only the site specific aspect to bind ~~the~~
~~additional proteins~~

23. RecA is the eukaryotic equivalent of MthH

RecA is the bacterial equivalent of ~~the~~ ^{RAD51} (for meiosis) in eukaryotes
OK

24. Base excision in humans involves XP genes, homologs of Ecoli Uvr genes

homologs of Ecoli UvrD genes

25. P elements are the major transposon in the human genome

Non-LTR elements, like *Alu*, make up the majority of transposons in the human genome

26. (4) Describe the process used by *E. coli* to repair mismatches detected during replication.

-3
if a mismatch is encountered during the replication process, the replication fork stalls. This leads to accumulation of proteins which ubiquitinate the PCNA clamp. The ubiquitinated clamp attracts the more flexible TLS polymerase which continues over the structural problem by inserting a random base to be repaired later through mismatch repair. If the fork stalls too long the cell may undergo apoptosis. Mut S, H, L

27. (2) What happens if a depurinated base is not repaired before replication?

-1
this can lead to a break during the replication fork where a double strand break occurs. point mutation

28. (4) Deamination of cytosine yields uracil and a U-G base pair. How is this repaired in somatic cells?

AP endonucleases excise the U base from the DNA (sometimes by flipping the base out) before the A base site is identified by the proteins, excized, and a new ~~base~~ nucleotide is inserted and ligated. ✓

29. (2) How are thymidine dimers repaired in *E. coli* in daylight?

the cell utilizes a special protein (photolyase sp?) which binds to the damaged region and uses light energy to repair the damage. ✓

30. (4) How are thymidine dimers repaired in humans?

Because the same protein is not available in placental mammals, humans utilize ~~AP endonuclease to remove the damaged~~ ~~section~~ nucleotide excision repair to repair the entire region around the damaged area. ✓

31. (2) How does the Ecoli alkyltransferase Ada repair methylated bases?

Ada accepts the methyl group from the bases, leaving a normal base and a methylated -Ada

32. (4) What are the major steps in repair of double strand breaks when sister chromatid is not available?

Ku proteins bind to the ends to be joined, some nuclease activity may occur (or blunt ends can be joined), the overlaps (or blunt ends) are ligated. This method of non-homologous endjoining can occur at any point in the cell cycle.

33. (4) Homologous recombination, homologous repair and gene conversion begin in a similar way. Describe the similar features of the initiation of these processes.

All begin w/ a double strand break which is identified by proteins which create a 3' end of ssDNA. the ssDNA invades the homologous chromosome, ~~and~~ and forms a D-loop which ~~then~~ diverges slightly between the 3 pathways.

34. (2) How are recombinases such as lambda integrase or Cre recombinase similar to type I topoisomerases?

they nick the DNA to allow for the recombination of ~~the~~ DNA.

35. (2) How is Cre recombinase activity distinct from topoisomerase activity?

Cre recombinase activity joins a different ssDNA molecule than the original one (~~the~~ topoisomerase) joins ends ok ✓

36. (2) Insertion of the transposon results in a direct repeat of the target site. How does this happen?

during insertion ~~the~~ the target site is converted to ssDNA which is then transcribed into a direct repeat. aligned ends

37. (2) How does a DNA transposon with intact terminal repeats but mutant/inactive transposase transpose?

the terminal repeats can still be recognized by transposase from a different transposon in the genome.

non autonomous

38. (3) Why is it that SINES in the human genome are more likely to cause mutations by unequal homologous recombination than by transposition?

sines transpose into regions that are rich in T's (due to the poly A tail) and such regions are not normally part of a coding gene, whereas unequal recombination may occur frequently since SINES are plentiful in the human genome.

39. (2) Trypanosomes evade the immune system of infected hosts by switching the expression of variant surface glycoprotein (VSG). What process is used for changing the identity of the expressed VSG?

gene conversion ✓

40. (2) How are heteroduplexes formed in homology directed repair?

the 3' end of the broken strand invades the homologous chromosome to form a D-loop. strand recombine

41. (3) How are the processes of homology directed repair and homologous recombination different?

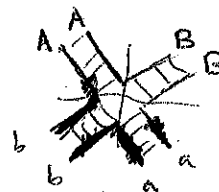
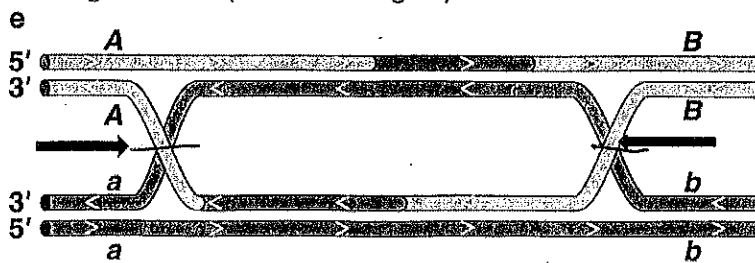
in HDR, the ~~broken~~ broken DNA will repair with the 3' overhang from the original broken DNA after gaining the necessary elongation.

in homologous recombination, a Holliday junction is formed where cleavage can occur in a number of ways to produce recombinant DNA sequences.

42. (2) Contrast Gene Conversion and homology directed repair. Identify 1 significant difference in these processes.

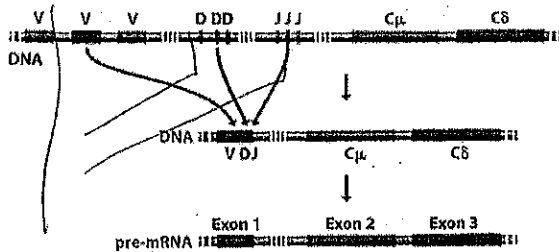
homology directed repair only elongates the 3' end of the broken DNA until it can re-pair with the other piece of the broken strand. gene conversion continues to the end of the chromosome which results in a loss of heterozygosity. spec. nuclease

43. (2pts) What is the result if resolution of the Holliday junction begins with nicks in both sets of exchanged strands (as shown in figure)?



non-recombinant ✓

44. (4) Figure illustrates generation of immune diversity. How is VDJ exon generated?



the VDJ exon is generated by first bringing ~~the D element~~ the D element over to the V and then bringing a J exon to join the VD.
detail

45. (4) How are mismatch repair and nucleotide excision repair similar?

both utilize a method of removal before the ssDNA is filled back in and ligated to yield a repaired double strand.

46. (2) What is the role of MRN in DNA damage repair?

MRN binds to double strand breaks and recruits ATM, a kinase which can effect change in the cell through mediators.

47. (2) Which two cellular events indicate to the cell that there is extensive DNA damage?

5sb + ddb
Haltting of the replication fork for an extended period of time can lead to a cellular apoptosis and excessive amounts of RecA indicate DNA damage.

48. (3) How does RPA initiate DNA damage response?

RPA ~~binds ATR~~ binds ATR and effects change by deactivating CDKs and preventing progression through the cell cycle

49. (2) Lox sites and att sites contain spacer regions (O). What is the role of the spacer region in the site specific recombination event?

the spacer region serves as the core of the SSR event, the region remains unchanged while the other parts around it are recombined.
orientation

50. (5) Compare the transposition of yeast Ty retroelements with the transposition of human L1 LINE elements. Clearly indicate 1 similarity and 2 differences in these processes.

Both processes result in the transposition of the specific sequence to an alternative locus in the genome ✓

L1 LINE excises from the DNA and reinserts into the genome at a different location

Ty retroelements utilize an mRNA intermediate which is reverse-transcribed into the genome.

2 diff's

Bonus (worth up to 10 points):

Complete student evaluation (online link at RPInfo)

Bonus score depends on the % of student submitting evaluation by 11:59pm Apr 30 (today!).

<50% response - no bonus

50-60% response: 1pts 60.1-70% response 3pts

70.1 80% 5pts

80.1-94.9% 8pts

>95% response 10pts

DATA