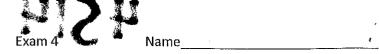PLEASE WRITE LEGIBLY. **IF THE GRADER CANNOT READ YOUR ANSWER IT WILL BE MARKED WRONG.**

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)

___B___ 1. If guanine is lost from DNA due to hydrolysis of the glycosidic bond, it is repaired via
a. DNA polymerase adding the guanine back by forming new glycosidic bond
b. Base excision repair of the AP site
c. Nucleotide excision repair of the AP site
d. Direct reversal of the DNA damage via a reverse glycosylase.

___B___ 2. In homology directed repair, single-strand invasion
a. occurs when a free 5'single stranded region invades a region of a homologous duplex
b. occurs when a free 3' single stranded region invades a region of a homologous duplex
c. allows replication fork to bypass damaged bases
d. results in the generation of heteroduplex RNA

___C___ 3. Which of the following repair systems is highly conserved in most organisms EXCEPT placental mammals?
a. Base excision   c. photoreactivation
b. Nucleotide excision   d. gene conversion

___D___ 4. Which of the following describes nucleotide excision?
a. a single damaged nucleotide is removed and replaced with a new nucleotide
b. a single damaged base is removed and replaced with a new base
c. a region of single stranded DNA containing damaged nucleotides is removed and replaced with new DNA
d. a region of double-stranded DNA containing damaged nucleotides is removed and replaced with new DNA

___D___ 5. Which of the following is feature common to DNA only transposons?
a. long terminal repeats   c. ORF encoding reverse transcriptase
b. Chi sequence   d. Short inverted terminal repeats

___D___ 6. All of the following are ways cells minimize the potentially harmful effects of transposition EXCEPT
a. differential splicing of transposase restricts activity to specific cell types
b. transposon sequences are methylated
c. piRNA base pairs with transposon mRNA
d. immunoglobulins bind to the transposon-encoded proteins

___D___ 7. Which bacteriophage lambda protein would you expect to be similar in function to RAG1 and RAG2?
a. Cro   c. integrase (Int)
b. cII   d. excisionase (Xis)

___A___ 8. Mobile DNA with the intrinsic ability to excise and transpose are called
a. Autonomous elements   c. non-autonomous elements
b. Somatic elements   d. replicative elements

___B___ 8. The Holliday junction:
   a. may occasionally form during transcription.
   (b) involves two DNA molecules connected by crossing DNA strands from homologous.
   c. involve a strand of DNA containing genetic information from two different chromosomes.
   d. may migrate and lead to formation of a three-stranded helix.

___B___ 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

___B___ 11. Homologous DNA recombination mainly benefits the cells by its role in
   a. Genetic recombination
   (b.) Post-replication repair of DNA
   c. Rearrangement of the genome at the immunoglobulin locus of B and T cells
   d. Preventing retroviral integration into the genome

___B___ 12. Deamination of methyl cytosine in DNA results in
   a. G-T base pair   c. G-C base pair
   (b.) U-G 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

___B___ 14. Glycosylases function by
   a. cleaving the ring structure of the nucleotide, leading to its removal
   (b) cleaving the bond between the base and the deoxyribose, leading to removal of the base
   c. nicking the phosphodiester backbone on both sides of a base, leading to removal of the base
   d. opening up the deoxyribose sugar ring, leading to its removal

___C___ 15. Ultraviolet radiation causes which type of DNA damage?
   a. extensive single stranded DNA   c. pyrimidine dimers
   b. base hydrolysis   d. base deamination

_____D__ 16. Which of the following describes the activity of the bacterial RecA protein?

a. DNA helicase with 5'-3' polarity

b. DNA helicase with 3'-5' polarity

c. degrades DNA from the 3' end at a nick

(d.) coats single-stranded DNA and catalyzes homologous strand invasion

_____A__ 17 Which of the following statements about CSSR is TRUE?

(a.) CSSR recombinases are sequence specific topoisomerases

b. Cre recombinase, a serine recombinase, promotes recombination between lox P sites

c. Cre recombinase requires accessory proteins to promote recombination between loxPsites

d. Cre recombinase can only excise the region between a pair of loxP target sites

_____A__ 18 Cells normally delay replication until DNA damage has been repaired because

(a.) Replication of damaged or mismatched DNA results in mutation

b. DNA polymerase cannot function when the DNA is damaged

c. Some repair may require recombination and that cannot occur during replication

d. Repair is more resource intensive after replication because it requires homologous recombination

_____B__ 19. TLS polymerases are able to bypass bulky lesions in DNA because

a. TLS polymerases have DNA repair domains that can repair DNA damage as the replication fork moves along

(b.) TLS polymerase active sites are much more open and flexible than more accurate polymerases

c. TLS polymerases have a clamp that can move the polymerase around the lesion and continue transcription

d. TLS polymerases are able to stall longer than replicating polymerases in order to wait for repair mechanisms to be carried out

_____C__ 20. In homologous recombination in *E. coli,* the protein that moves along a double-stranded DNA, unwinding the strands ahead of it and degrading them, is:

a. Rec A   (c.) RecBCD

b. RuvC   d. Rad51

21-25 (2 pts each)

21. ___B___ Which pair of statements is correct?

a. Deamination of cytosine changes the base to thymine. This is corrected by base excision repair

(b.) Deamination of cytosine changes the base to uracil. This is corrected by base excision repair.

c. Deamination of cytosine changes the base to thymine. This is corrected by nucleotide excision repair

d. Deamination of cytosine changes the base to uracil. This is corrected by base nucleotide excision repair.

22. __D__ Which statement is correct?
    a. Double strand break repair occurs via NHEJ in G2 and via HDR during G1
    b. Double strand break repair occurs via NHEJ in S and via HDR during G1
    c. Double strand break repair occurs via NHEJ in G2 and via HDR during G0
    (d.) Double strand break repair occurs via NHEJ in G1 and via HDR during G2

23. __C__ Compare with DNA polymerase, reverse transcriptase
    a. Does not require a primer to initiate synthesis
    b. Introduces no errors into the genome because it synthesizes RNA not DNA
    (c.) Makes more errors because it lacks the 3' to 5' proofreading exonuclease
    d. Synthesizes complementary strand in the 3' to 5' direction

24. __A__ Which statement is correct?
    (a.) Nucleotide excision in humans involves XP genes, functional equivalents of Ecoli Uvr genes
    b. Nucleotide excision in humans involves XP genes, homologs of Ecoli Uvr genes
    c. Base excision in humans involves XP genes, functional equivalents of Ecoli Uvr genes
    d. Base excision in humans involves XP genes, homologs of Ecoli Uvr genes

30, 3/5   25. ~~C~~. A The J segment of an immunoglobulin is
    (a.) Relatively short and codes for the first few amino acids of the constant region
    b. Relatively short and codes for the last few amino acids of the constant region
    (c.) Relatively short and codes for the first few amino acids of the variable region
    d. Relatively short and codes for the last few amino acids of the variable region

26. **(4) Describe the process used by _Ecoli_ to repair mismatches detected following replication. Be sure to identify key proteins involved**

E. coli solves mismatches using the mismatch repair mechanism where the primary proteins involved are Mut H L and S. Mut S ~~FOR~~ searches for mismatches and when it finds one it recruits Mut L which subsequently recruits Mut H. Mut H has endonuclease activity and can excise the mismatched base so that DNA polymerase can repair the site.

27. **(4) How is the collapse of replication forks repaired in E. coli**

In E. coli ~~replicator~~ collapsed replication forks are repaired by strand invasion of the broken 3' strand into the ~~forth~~ ~~intact~~ intact DNA. After strand invasion the 3' end acts as a primer for the ~~synthesis~~ lengthening of the 3' end of the broken strand. This opens the D-loop so that the 5' end can be lengthened. A Holliday junction is formed. Resolvases resolve the Holliday junction and the clamp is reloaded and replication continues.

4

13

42% **28. (4) How does DNA repair differ from DNA replication?**

In DNA repair specialized DNA polymerases add nucleotides to the ~~ends~~ repair site.

DNA repair is distributive whereas DNA replication is processive.

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

When a sister chromatid is not available, chromatids undergo Non-homologous End joining. RecBCD binds the ends of dsBreaks and cuts off nucleotides on both strands ~~as fast as~~ until a chi site is reached. Then RecD starts cutting off only bases on the 5' strand so that the 3' strand is longer. Then the chi sites from two ds breaks anneal together and DNA polymerase fills in the gaps, joining the two dSB ends together.

50% **30. (4) Homologous recombination, homologous repair and gene conversion begin in a similar way. Describe the similar features of the initiation of these processes.**

In homologous recombination, homologous repair and gene conversion all start with a strand of one DNA molecule base pairing with a homologous sequence on another molecule and then DNA synthesis occurs

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

A large amount of single stranded DNA from stalled replication forks and a large amount of double strand breaks from collapsed replication forks.

32. (6) Compare and contrast Cre recombinase with type I topoisomerases.

Cre recombinase and type I topoisomerase are similar because they both start out by ~~making a phosphodiester bond and attaching~~ nucleophilic attack on a phosphodiester bond by a tyrosine at the active site. Then both the recombinase and type I topoisomerase can rejoin nucleotides without the net expenditure of energy.

However Cre recombinase reattaches nucleotides to different DNA molecules (or different parts of the same DNA molecule whereas topoisomerase connects the nucleotide back to its ~~start~~ original positions.

600% **33. (2) Why is it that SINES in the human genome are more likely to cause mutations by unequal homologous recombination than by transposition?**

SINEs are very frequent in the human genome so unequal homologous recombination is very likely. ~~because~~ SINEs do not encode their own transposases so they can't move by transposition unless a transposase is being expressed by a LINE

34. (6) Compare and contrast the mechanism DNA only cut- and paste transposition and LTR retroelements transposition.

The DNA only cut-and-paste mechanism and LTR retroelement ~~cut~~ are similar in mechanism in that they both use their Terminal Repeats in the transposition mechanism. However DNA only cut-and paste transposition ~~only has either a single copy Tn~~ moves by the direct movement of DNA whereas LTR retroelements move via an RNA intermediate.

35. (2) Contrast Gene Conversion and homology directed repair. Indicate one significant difference in these two processes

In Gene Conversion, no new DNA is synthesized where as new DNA is synthesized for homology directed repair. In homology directed repair, the DNA invades as the homoduplex a single strand whereas in Gene conversion, the base pairing is between two fully intacted double stranded DNA.

6

36. (4) Contrast homology directed repair and homologous recombination. What are the major differences in these two processes.

78%

HDR sister chromatid same as HR

In homology directed repair the homologous chromosome acts as a template for the synthesis of new DNA ~~by~~ Where as in homologous recombination, DNA is already all present before the pairing of the strands.

1

37. (4) Describe the process used in B cells to generate the VDJ exon.

B cells use transposition to generate VDJ exons. Each component has a 7 base pair sequence followed by either 12 or 23 bp then a 9 bp sequence. The different elements are joined based on making sure that a 23 bp element only attaches to a 12 bp element. Exons are identified at each of the ends by some protein Rag 1/2 that cuts one end and forms a hairpin loop. The hairpin loop is then trimed to remove the loop which creates diversity of sequence. Then the two ends are joined together.

3

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

80%

Mismatch repair and nucleotide excision repair are similar in that the ~~repair~~ damaged DNA is identified and excised from the DNA molecule and then new nucleotides are added by DNA polymerase.

3

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

When a transposon is inserted, the DNA can only attach to the 3' end of each strand. This means that the 5' strands must be replicated in order to complete the insertion.

Stand w/ staggered cut

1

40. (2) What is the role of the Ku70/Ku80 dimer?

Ku 70 and Ku 80 bind to double strand breaks and keep the two ends together.

2

41. **(4) What is the role of the resolvase RuvC in homologous recombination in Ecoli**

RuvC binds to RuvAB at Holliday junctions and then cuts and rejoins the DNA so that it is no longer entangled. If it cuts in the proper way, homologous recombination will occur.

42. **(2) How does MRN initiate the DNA damage response?**

MRN initiates the DNA damage response by binding to single strand breaks and then phosphorylating proteins in the cell cycle ~~~~~~~~~~~~ in order to stop cell cycle progression.

(written in margin: Double ... single strand ... Recruit ATM Kinase ... cell cycle)

43. **(10) Compare and contrast the transposition an LTRretroelements (e.g. yeast Ty element)with the transposition of nonLTR retroelement (human L1). Be sure to address the following:**

LTR retroelements and non-LTR retroelements both transpose via a RNA intermediate. Both these elements are transcribed into mRNA and then reverse transcriptase makes a cDNA copy of the element in the cytosol. ~~~~~~~~~~~~~~~~ This reverse transcription requires the use of tRNA as a primer which for LTR retroelements, this primer attaches at ~~~~~~ adjacent to the US site. Then the cDNA copy of the element moves back into the nucleus to be integrated. LTR retroelements are integrated by their LTR mechanism, which is similar to the DNA only cut-and-paste mechanism. non-LTRs don't have Long terminal Repeats so they work by a different mechanism which involves the use of the non-LTR element's poly A tail.

(margin notes: LTR only ... LINE cDNA in nuc ... template primed)

nonLTR - mechanism?