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Exam 4 May 11, 2010

1 A	2 B	3 B	4 A	5 B	6 B	7 D	8 C	9 C	10 D
11 A	12 A	13 A	14 A	15 C	16 D	17 A	18 C	19 D	20 C

21 (2) How is priming okazaki fragments different in eukaryotes (yeast) vs bacteria (E coli)
E coli uses primase (DnaG) to synthesize RNA primer. In Euk Primer is synthesized by DNA pol alpha/ primase complex (RNA- DNA primer) clue in MCQ9

22 (2) Adenovirus has a double strand linear genome. How is DNA synthesis initiated as the ends of the Adenovirus genome? priming nucleotide provided (presented) by a protein

23 (2) How is replication terminated in E coli?
The *E. coli* genome contains terminator sequences (Ter) that are bound by the protein Tus. Tus blocks the movement of the replication fork in one direction / replication forks trapped .(optional: The replisomes are disassembled and type II topoisomerase (topoisomerase IV) separates the interlinked daughter DNA molecules) extra info must be correct or points deducted

24 (2) A) Why can't the DNA polymerase move continuously on both strands? **Because the DNA polymerase is capable of adding new nucleotides only at the 3' end of a DNA strand** (continuous synthesis could happen on top strand but corresponding region on bottom strand would end in 5' not 3')

25 (2)Removal of the primer is especially important on the lagging strand. Why must the primer be removed? Primer contains RNA (and initial DNA component is error-prone

26 (2) The lagging strand contains one primer for each Okazaki fragment. How is each primer removed in E coli? (DNA polymerase III Replaced by) **DNA polymerase I, which has a 5' to 3' exonuclease and removes the primer**,

27 (2pts) Primer removal in eukaryotes? **NO T similar**—eukaryotes do not have a DNA polymerase with 5' to 3' exonuclease activity primer displaced then removed via FEN endonuclease / RNAseH

28 (2) In E coli the A of the sequence GATC is methylated. What is the role of methylation in mismatch repair? **Detect daughter vs parent strand** .(For a short time after replication (1.5 min on ave) DNA is hemimethylated – parent strand is methylated and daughter strand is not) clue in MCQ2

29 (2) How does methylation of GATC regulate initiation of replication in E coli? **Replication can only be initiated at origins that are fully methylated**

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30 (2) What is the role of the septum in bacterial cell division? Divides bacterial cell

31. (2 points)Mitochondria genomes and bacterial genomes are usually circular. What Is one major differences between mitochondrial replicons and bacterial replicons (2) clue in MCQ4

Mito: separate origins for each strand, replication via displacement synthesis, multicopy

Ecoli: one origin functions on both strands, bidirectional replication with replication fork,

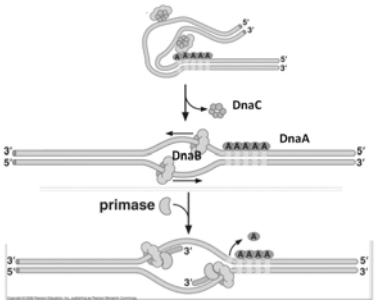
single copy genome any PAIR is fine (i.e mito multicopy/ Ecoli single copy)

32 (4)Compare rolling circle replication of lambda phage to E coli replication clue in MCQ5

	Bacteriophage lambda	E coli
Replication initiated :	nick	Replication fork (or ori C, dnaA at oriC)
Chromosome(genome) copy number:	multiple	one

33 (2) Why do origins of replication contain a core of AT rich DNA?

More easily melted



34 (2 points) a) How are replication forks initiated at the E. coli origin of replication?

DnaA proteins bind and promote DNA melting

DnaC (helicase loader) brings DnaB to melted region

35 (2) b) What is the function of Dna B?

Helicase –unwinds the double helix into single strands (uses ATP) clue in MCQ6

36 (2) Why are single stranding binding proteins SSB and RPA important in DNA replication?

Single stranded binding protein – prevents melted sequences from reannealing

37 (2pts) Why is sliding clamp important in DNA replicaton? Assures that DNA polymerase remains associated with strand being replicated

38 (5) What is the primary role of each of the following DNA polymerases?

E coli DNA polymerase I repair synthesis (primer removal also acceptable)

E coli DNA polymerase II restart stalled replication forks

E coli DNA polymerase III The major replicating polymerase of E. coli

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E coli DNA polymerase IV error-prone / translesion repair / SOS response

Eukaryotic DNA polymerase delta The major replicating polymerase of eukaryotes

39 (2) Why do all cell have error-prone DNA polymerases since their use is guaranteed to make mistakes?

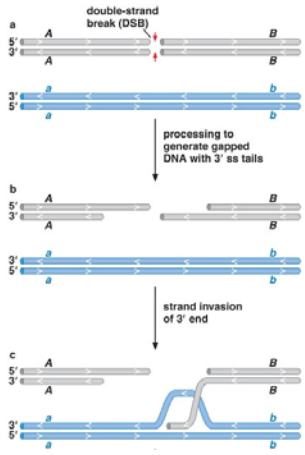
Enable replication fork to bypass damage that would otherwise halt replication

40 (4) How does repair DNA synthesis differ from replication DNA synthesis? (list two ways)

Possible answers include: Uses different polymerases(pol III for replic—others in repair),

repair enzymes not as much proofreading,

replication only once a cell cycle/ repair anytime

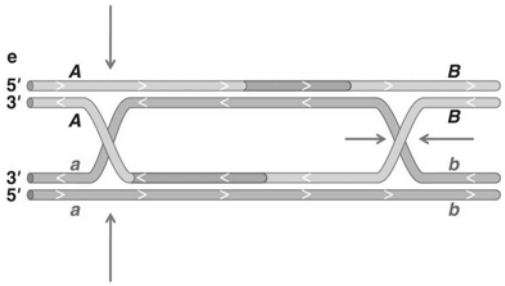


41 (3) Describe the major steps in recombination) clues in MCQ10-12 and fig

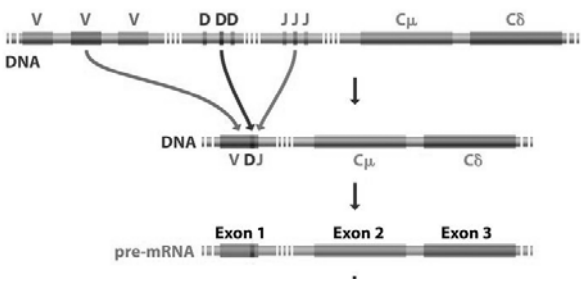
Generate double strand break – 5' resection to yield 3' single stranded overhang – strand invasion at homologous region of other duplex.

Can go on to describe branch migration- second strand invastion.

42. (2pts) What is the result if nicks are made in the non exchanged strands in the A/a region and in the exchanged strands in the B/b region? Genetic crossover / splice recombinant (Ab and aB)



43 (4points) Figure illustrates generation of immune diversity



a) These genome rearrangements occur in: lymphocytes (Bcells/ Tcells/ immune cells)

b, How is VDJ exon generated? RAG recombinase binds sequences flanking V D J sites (RSS- recombination signal sequences) to **enable site specific recombination**

combine 12bp spacer site with 23 bp spacer site (clue in MCq13)

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44 (2pts) If a replication error that creates an AG mismatch is not repaired before the next round of replication, what will be the sequences of the two daughter DANs after replication?

Upon replication AG mismatch becomes AT on one daughter strand and GC on the other

45 (2pts) Deamination of cytosine yields uracil and a U-G base pair. How is this repaired in somatic cells? Uracil is recognized as a "damaged" / incorrect base. Removed by uracil glycosylase. repair

46 (2pts) What happens if C deamination yields U-G base pair in the variable region of an immunoglobulin gene?

deamination of C- base excision but don't repair, during replic add any base

47 (2pts) How are thymidine dimers repaired in E coli in daylight? E.coli has photolyase that can directly reverse damage

48 (2pts) How are thymidine dimers repaired in humans? **Excision repair** (nucleotide excision NOT base excision): **damaged region recognized, endonuclease cleaves** on both sides, **damaged strand removed**, **synthesize new strand** using undamaged as template (clue in MCQ14)

(involves XP genes some of which are components of TFII H – not UvrA, B, C –deduct 1 point for Uvr genes)

49 (2pts) Why are transcriptionally active genes preferentially repaired?RNA polymerase stalls at damage/ TFIIH recruits repair

50 (2pts) Double stranded breaks in a chromosome can have disastrous effects if not repaired, therefore, most cells have two pathways for repair.

a non homologous end joining

b homologous recombination

51 (2pts)What are the essential features of an insertion sequence (IS)? Transposase flanked by inverted terminal repeats (clue in MCQ 15)

52. (2points) Insertion of the transposon results in a direct repeat of the target site. How does this happen?

Transposase **makes a staggered cut at target site.** Transposon is inserted (ligate end free 3'OH) then fill **in staggered region**. This results in direct repeat of staggered cut region

53.(2 points) Retroviruses have an RNA genome (**R-U5–protein coding region–U3–R)** that is replicated into DNA (**U3 –R –U5 – protein coding region U3 – R – U5)** prior integration into host genome.

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First strand is synthesized by reverse transcriptase

Where does synthesis initiate? Near U5 (just 3' to U5) region of **RNA genome**

68 (4 points) Distinguish between LINEs and SINEs Circle correct answer

0.5/correct ans	LINEs	SINEs
Move via RNA intermediate	Yes	Yes
Encode reverse transcriptase	Yes	no
Transcribed by pol III	no	Yes
Autonomous	Yes	No

Bonus

30 (2 points) Lambda is a bacteriophage (bacterial virus) that can integrate into the E. coli genome. HIV (a human retrovirus) can integrate into the human genome.
How does the integration of lambda into the E. coli genome differ from integration of HIV in the human genome?
CLUE in MCQ18

Lambda integration: **site specific integration** (staggered cuts of attP/ attB common core (O) regions) HIV integration: (convert RNA genome into dsDNA (cDNA)) **insert similar to transposons- relatively randomly**. (Insertion results in direct repeats)