Practical - Welsh Onion

03 October 2014 17:50

Allum Fistulosium

- Mitotic cell cycle
  - Sequence of steps between 1 division and the next
  - All cell multiplication
  - Phases
    - Interphase
      - G1
      - S
      - G2
    - M phase
      - Mitosis
        - Prophase
        - Metaphase
        - Anaphase
        - Telophase
      - Cytokinesis
    - Continuous
      - Divided for convenience

Aim

Look at mitotic cell cycle in root tip

Preparation

- Germinated, 1-2cm roots
- Seedlings fixed in 3:1 ethanol:acetic
- Hydrolysed in 1M HCl for 10 minutes at 60^0C
- DNA stained red/violet in Feulgen stain

1. Remove 1 seedling
2. Cut off stained tip
3. Drop of aceto-carmine stain
4. Apply cover slip
5. X10 then x40

C-Metaphase

- 0.2% colchicine
- Disrupts metaphase

        - Small, low lying
        - 30 survivors of 1780s typhoon
            - Bottleneck
        - One survivor carried rare allele for total colour blindness
            - Sampling error
        - 10% of population now carries that allele
            - Allele frequency of 0.32
            - Genetic drift
- **Assortative mating**
    - Preference towards a similar genotype when mating
    - E.g. Pigeons
        - Assortative mating with regard to size
        - Dissortative mating with regard to colour
            - Prefer differently coloured pigeons
- **Natural selection**
    - Some genotypes are fitter than others
    - Fitter individuals produce more offspring during their lifetimes
    - This may be because they
        - Live longer
        - Produce more offspring
    - Directional selection
        - One allele decreases, one increases
        - 1 will go extinct, the other fixated
        - E.g. Peppered moths
    - Balancing selection
        - Heterozygotes favoured over homozygotes
        - Preserves genetic diversity
        - E.g. Sickle-cell anaemia in malarial regions
    - Diversifying selection
        - Homozygotes favoured
        - Very few homozygotes
        - E.g. 3 spine stickleback
            - Forage near bottom or surface of lakes
            - Size difference
            - Differences in courtship
            - Hybrid disadvantage
- **Net mutation**
    - Mutations occur continuously
    - Rate of forward mutations may exceed reverse mutations

    - Particularly when mutations cause loss of function
        - Loss of function is more likely than gain
        - Mutation/selection balance often restores frequencies
    - E.g. Albinism
        - Mutation from wild type to albino
        - More frequent than the reverse mutation
        - More likely to be predated
            - Mutation/selection balance

- Importance
  - If heritability is high
    - Trait can be modified by breeding
  - If heritability is low
    - Trait can be modified most successfully by changing the environment

Additive genetic variation

- Several genes contribute to same trait
- E.g. two genes influence coat colour
  - Alleles A and B code for pigment
  - Alleles a and b code for no pigment
  - ADD PICTURE
- As the number of genes for the trait increases
  - Number of genotypes increases
  - With n genes there are 3n genotypes
  - Distribution of trait values approaches normal distribution

Quantitative trait loci (QTL)

- QTL mapping
  - Score members of population for trait
  - Determine genotype with respect to a marker gene
  - Look for association between genotype and trait
    - Does genotype explain the variance?
  - This shows that the marker is linked to a QTL
    - This doesn't necessarily mean that the marker *is* the QTL
  - Make linkage map to see how close the marker is to the QTL

Selection on Quantitative Traits

- Evolutionary biologists often assume genetic variation is continuous
  - Assume many genes influence a trait
  - Assume that genes contribute additively
- Selection
  - Acts upon the mean and variance of a trait
  - It is implied that this action reflects changes in the frequency of alleles
  - Directional, stabilising and diversifying selection
  - E.g. Peacocks
    - Long tail
      - More mates
      - Directional selection
    - Too long
      - Easy prey
      - Stabilising selection

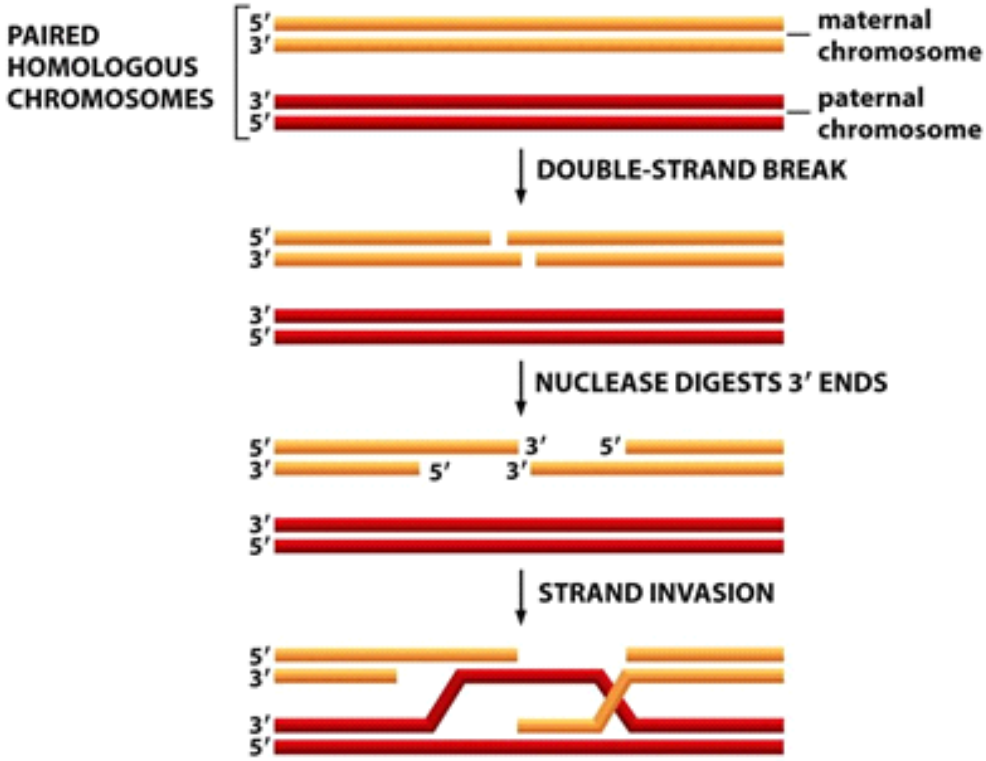

Figure 6-31 part 1 of 2 Essential Cell Biology 3/e (© Garland Science 2010)

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DNA synthesis and ligation

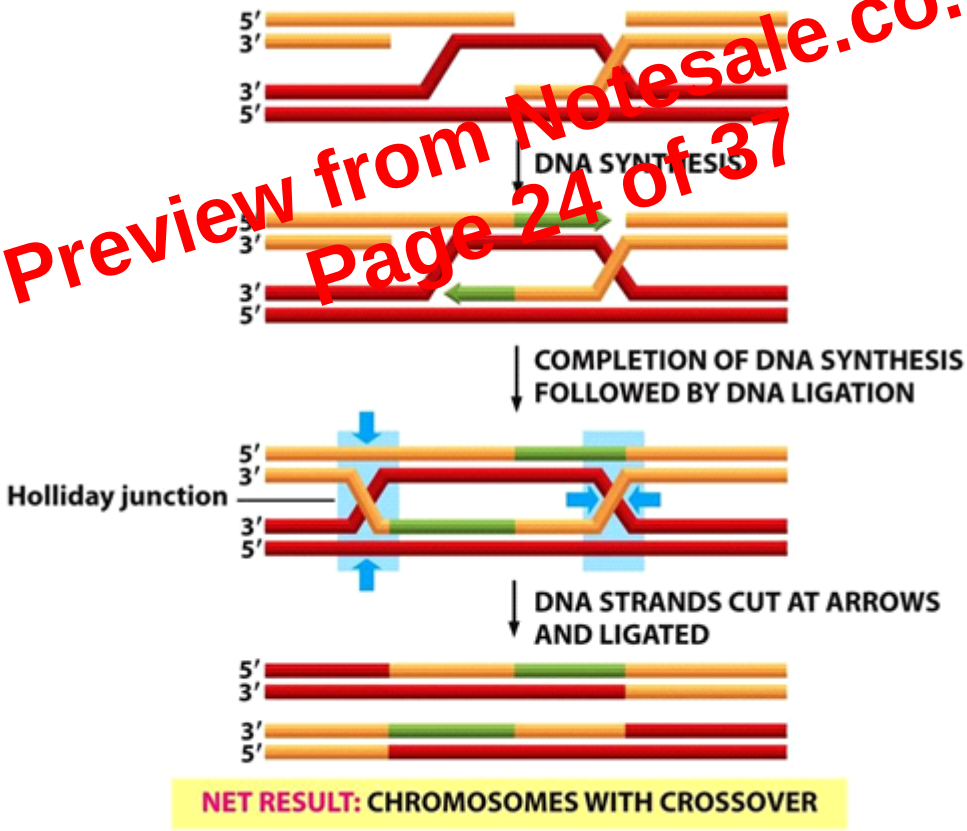

Figure 6-31 part 2 of 2 Essential Cell Biology 3/e (© Garland Science 2010)

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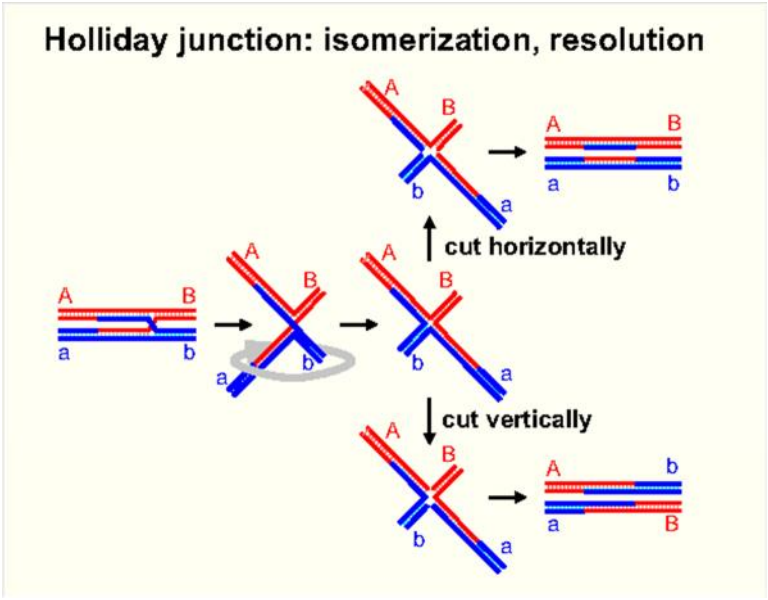
- Loop displaced from unbroken molecule
  - Pairs with non-invading strand of broken molecule
  - Template for DNA synthesis
- 2 Holliday junctions form between the DNA molecules
- Cross over.
- Holliday junction

- Migrates along chromosome
- Rotates
- Forms cross-shaped Holliday intermediate
- Cleaved in vertical or horizontal plane
- 

- Gene conversion
  - Occurs in heterozygous non-crossover chromosomes
- vertical : crossing over
- horizontal: no recombination
- **Homologous repair of double strand breaks**
  - Can be caused by
    - Radiation
    - Mutagen
  - Homologous chromosome used as template for repair
  - 
  - Homologous chromosome used as template for repair
  - Potentially the origin of crossing over

Recombination and evolution

- Shuffles alleles at different loci
- Genotypic diversity
  - Adaptive response to environmental change
- Decouple beneficial alleles from harmless ones

Mutation

25 November 2014 12:48

- An hereditary change in
  - DNA sequence
    - Gene mutation
  - Chromosome number, form, or structure
    - (DNA & Chromosomes)
  - Caused by
    - Errors in replication
    - Spontaneous damage
      - Radiation
      - Chemicals

Genetic diversity

- Polymorphic DNA
  - Several different alleles present at the same time
- Increased by
  - Mutation
  - Immigration
- Reduced by
  - Natural selection
  - Genetic drift

Types of gene mutation

- Substitution
  - Substitutes 1 nucleotide for another
  - Transition
    - A <--> G
    - C <--> T
  - Transversion
    - A <-> C, A <-> T, G <-> C, G <-> T
  - More possible transversions
    - But transitions more likely
      - Molecular mechanisms make them more likely to occur
      - More likely to persist
        - Less likely to change any amino acids

The genetic code

- DNA to mRNA to protein
- Codon
  - 3 base pairs
  - 1 amino acid
- Sequence of amino acids determines protein structure and function
- Reading frame
  - Describes how continuous DNA is divided into codons
  - 6 frames
  - 

    The six reading frames of DNA
  - Frameshift mutation
    - Substitution
      - Can change 1 codon
        - Amino acid changes
        - Slightly different protein
      - Stop codon
        - Truncated protein
        - TAC to TAA, TGC to TGA
    - Insertion or deletion
      - In multiple of 3 base pairs
        - Loss or gain of amino acid
      - Not in multiple of 3
        - Knock on effect down entire protein
        - Radical effect
      -

Classes of mutation

- Beneficial
  - Increase fitness
  - Occur rarely
    - Random changes unlikely to make improvements
  - Constantly fixed into genome by directional selection
    - Accumulate into evolution
    - Lost through genetic drift in small population
  - Rarely polymorphic
    - Pushed straight to fixation
  - E.g. deer mouse
    - Dark soil = dark fur. Most common
    - Sand Hills - Tan fur.
      - Mutation occurred 4000 years ago
- Deleterious
  - Reduce fitness
  - Frequent
    - Random changes much more likely to cause damage
  - Constantly removed
    - Purifying selection
    - Mutation/selection balance
  - Usually polymorphic and rare
    - But can be fixed into genome by drift in small populations
      - Pingalap atoll
  - Often recessive
    - Not exposed to purifying selection in heterozygotes
    - Lurks in heterozygotes
      - Isn't expressed
      - Isn't selected against
    - If dominant normally selected against
      - Unless presented in old age
  - E.g. Over 4000 human diseases caused by single gene mutation
- Neutral
  - Occur frequently
  - Non-coding mutation
    - In untranslated DNA
      - Introns
    - Intergentic regions