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YUMPU automatically turns print PDFs into web optimized ePapers that Google loves. Copy Chapter 10 - Molecular Biology of the Gene Extended embed settings Chapter 10 Molecular Biology of the Gene Molecular Biology: Study of DNA and how it serves as the molecular basis of heredity. In 1920 s it became clear that chromosomes contained genes for genetic traits. However, chromosomes are made up of both protein and DNA. Which one was the genetic material? Before the 1940 s most scientists believed that proteins were the genetic material of cells, and that nucleic acids (DNA and RNA) were too simple to code for genes. Molecular Biology: How was the genetic material identified? A number of experiments were important in establishing that DNA was indeed the genetic material of living organisms. u Frederick Griffith's experiments (1928) u Oswald Avery's experiments (1944) u Alfred Hershey and Martha Chase experiments (1954) I. Frederick Griffith's Experiments (1928) Griffith was studying two strains of the bacteria Streptococcus pneumoniae, which causes pneumonia and other infections. u Smooth strain (S): Produced a polysaccharide capsule that gave its colonies a smooth appearance. Because of the capsule, the bacteria can evade the immune system and cause disease. u Rough strain (R): Does not produce a capsule, rough appearance. Not pathogenic, doesn't cause disease. I. Frederick Griffith's Experiments (1928) Griffith treated his mice with the following bacterial preparations: Treatment Outcome 1. Live rough Mouse lives 2. Live smooth Mouse dies 3. Heat killed smooth Mouse lives 4. Heat killed smooth + live rough Mouse dies To his surprise, he did not get the expected results in the last treatment (#4). The mice got sick and died. Additionally, he recovered smooth bacteria from these mice. Griffith's Experiments: Transformation of Harmless Bacteria into Deadly Bacteria I. Frederick Griffith's Experiments (1928) Why were smooth bacteria present in the last group of mice (live rough + dead smooth)? n Griffith concluded that the genetic instructions to make capsules had been transferred to the rough bacteria, from the dead smooth bacteria. n He called this phenomenon transformation. n However, he was unable to identify what type of molecule was responsible for transformation. II. Oswald Avery's Experiments (1944) Avery repeated Griffith's experiments using purified DNA, protein, and other substances. He showed that the chemical substance responsible for transformation was DNA and not protein. While many biologists were convinced, others remained skeptical. III. Hershey and Chase Experiments (1952): Definitive proof that DNA rather than Protein carries the hereditary information of life E. Coli bacteriophage: A virus that infects bacteria. Bacteriophages only contain a protein coat (capsid) and DNA. They wanted to find out whether the protein or DNA carried the genetic instructions to make more viruses. They labeled either the viral proteins or DNA: u Protein capsid: Labeled with radioactive sulfur (35 S) u DNA: Labeled with radioactive phosphorus (32 P) Radioactive labeled viruses were used to infect cells. Bacteriophages Are Viruses that Infect Bacteria Either Bacteriophage DNA or Proteins Can be Labeled with Radioactive Elements Hershey Chase Experiment: DNA is Genetic Material III. Hershey and Chase Experiments (1952): Bacterial cells that were infected with the two types of bacteriophage, were then spun down into a pellet (centrifuged), and examined. Results: 1. Labeled viral proteins did not enter infected bacteria (found in supernatant). 2. Labeled viral DNA did enter bacteria during viral infection (found in cell pellet). Conclusion: Protein is not necessary to make new viruses. DNA is the molecule that carries the genetic information to make new viruses!!!! IV. DNA and RNA are Polymers of Nucleotides Each nucleotide has: 1. A 5 carbon sugar (pentose); u Deoxyribose in DNA u Ribose in RNA 2. A phosphate group. 3. A N-containing organic base: u DNA: Adenine (A), Guanine (G), Cytosine (C), and Thymine (T). u RNA: Adenine (A), Guanine (G), Cytosine (C), and Uracil (U). u Purines: Bases with two rings (A and G) u Pyrimidines: Bases with one ring (C, T, and U) DNA are Polymers of Nucleotides IV. Structure of DNA Molecule Erwin Chargaff (1947) Chargaff's Rule: In the DNA of all living organisms, the amount of A = T and the G = C n No matter which species on earth he studied, the DNA showed the same relative ratios u Adenine = Thymine u Guanine = Cytosine n These results suggested that A & T and C & G were somehow paired up with each other in a DNA molecule. IV. Rosalind Franklin, James Watson, and Francis Crick (April 1953, Nature) u Rosalind Franklin: X-ray crystallography of DNA, trying to determine the structure of the molecule. Franklin's work laid the foundation for Watson and Crick. Died of cancer in 1958. u James Watson and Francis Crick: Determined the exact three dimensional structure of DNA as a double helix held together by H bonds. Won 1962 Nobel Prize. u DNA is an antiparallel double helix: 5' end of one strand is paired to 3' end of other strand. F A & T and G & C are paired up by hydrogen bonds F Two strands are complementary to each other. F If you know sequence of one strand, can determine sequence of the other one. DNA Structure: Double Helix Held Together by H Bonds Hydrogen Bonding Between Complementary Nucleotides Explains Chargaff's Rule V. How exactly does DNA replicate? Several models for DNA replication were proposed: 1. Conservative model: Two completely new strands are formed, which coil together. Original strands stay together. 2. Semiconservative model: One original strand pairs up with one new strand. 3. Dispersive model: Each strand is a mixture of old and new DNA. Three Models of DNA Replication V. How exactly does DNA replicate? Findings: u Replication is carried out by DNA polymerase. F 50 nucleotides per second in mammals F 500 nucleotides per second in bacteria u DNA strands unzip and each one acts as a template for the formation of a new strand. u Nucleotides line up along template strand in accordance with base pairing rules. u Enzymes link the nucleotides together to form new DNA strands. u Semiconservative replication: Each new helix will contain one new strand one old strand. DNA Replication is Semiconservative V. How exactly does DNA replicate? u Strands are antiparallel: Run in opposite directions. u DNA polymerases can only add nucleotides to one end of the strand (3' end). u New strands grow in a 5' to 3' direction u Replication fork with: F F Leading strand: Made continuously. Lagging strand: Made discontinuously in Okazaki fragments which are then joined together. DNA Strands are Antiparallel DNA Replication: Double Helix Must Unwind Two Strands are Made Differently DNA Replication: Leading Strand is Made Continuously; Lagging Strand is Made in Fragments VI. DNA Genotype is Expressed Phenotypically as Protein or RNA product. Fundamental unit of heredity. DNA sequences specify order of amino acids in protein; but do not produce protein directly. u Proteins are crucial to cell activity F Cell movement F Oxygen F Active F Cell and carbon dioxide transport across membranes division F Enzymatic reactions (respiration, digestion, etc.) VII. Flow of Genetic Information in the Cell u DNA does not produce protein directly. Genetic blueprints do not leave nucleus. u Transcription: An RNA copy of gene is made (messenger RNA or m. RNA). F In eukaryotes m. RNA is made in nucleus and then goes to cytoplasm. u Translation: Process in which protein is made from instructions contained in messenger RNA. Translation is carried out by ribosomes in the cytoplasm or rough E. R VII. Flow of Genetic Information in the Cell Transcription DNA Translation m. RNA Protein Transcription and Translation Occur in Different Parts of Eucaryotic Cells VIII. RNA is made by Transcription u Occurs in the cell's nucleus. u Transcription is carried out by RNA polymerase. u A single strand of DNA ("coding strand") serves as the template for RNA synthesis. u RNA nucleotides are matched to complementary DNA nucleotides in 5' to 3' direction. u RNA contains uracil (U) instead of thymine (T). u m. RNA transcript leaves nucleus through nuclear pores and goes to cytoplasm. Transcription of a Gene IX. Proteins are made by Translation u Occurs on ribosomes in the cell's cytoplasm or rough ER. u Translation is a complex process, carried out by several types of RNA molecules: 1. Messenger RNA (m. RNA) 2. Transfer RNA (t. RNA) 3. Ribosomal RNA (r. RNA) Necessary Components for Translation: 1. Messenger RNA (m. RNA): Encodes for a specific protein sequence. F Variable length (depending on protein size) F Information F 64 is read in triplets (codons) possible codons (4 x 4 = 64 = 43) • 61 codons specify amino acids • 3 codons are termination signals. m. RNA is complementary to DNA and read in triplets (codons) Necessary Components for Translation: 2. Transfer RNA (t. RNA): Brings one amino acid at a time to the growing polypeptide chain. F Small molecule (70 to 90 nucleotides) F Forms a cloverleaf structure F Anticodon: Base pairs to m. RNA codon during translation. F Amino acid binding site: At 3' end of molecule. Transfer RNA (t. RNA) Carries Amino Acids to the Growing Polypeptide Chain Necessary Components for Translation: 3. Ribosomal RNA (r. RNA): Ribosome is site of protein synthesis. F Facilitates F Huge coupling of m. RNA to t. RNA molecule: Large and small subunits must assemble for translation. F Ribosome protein composition: 60% r. RNA and 40% Ribosome is the Site of Translation STEPS OF TRANSLATION: 1. INITIATION: F Messenger F Transfer RNA (m. RNA) and ribosome come together. RNA (l. RNA): Carrying first amino acid (methionine) has anticodon which binds to start codon (AUG). 2. ELONGATION F One amino acid at a time is added and linked to growing polypeptide chain by a peptide bond. 3. TERMINATION F Stop codons: UAA, UAG, or UGA F Ribosome/m. RNA complex dissociates. Translation: Initiation at Start Codon Translation: During Elongation one Amino Acid is Added at a Time Elongation: Ribosome Travels Down m. RNA, Adding One Amino Acid at a Time Termination: Once Stop Codon is Reached, Complex Disassembles X. Genetic Code u Twenty amino acids are found in proteins u Four nucleotides are found in RNA and DNA u How can nucleic acids with only 4 bases encode proteins with 20 amino acids? u Each amino acid is encoded by more than one nucleotide F If 1 base = 1 amino acid, Can only determine 4 amino acids F If 2 bases = 1 amino acid, Can only determine 16 amino acids F If 3 bases (Codon)= 1 amino acid, Can determine 64 amino acids u Genetic code is redundant, more than one codon per several amino acids. u Genetic code is universal Universal Genetic Code Mutations are permanent changes in DNA u DNA replication is never 100% accurate u Bases may be inserted, deleted, or mismatched during replication. u Mutations: Any mistakes that cause changes in the nucleotide sequence of DNA. u Mutations may be either harmful, beneficial, or have no effect on a cell or individual. XI. Mutations: Permanent changes in DNA n There are several possible types of mutations: I. Substitution mutation: One nucleotide is replaced by another. May result in: F Missense: Different amino acid. May or may not have serious consequences. Example: Sickle cell anemia. F Nonsense: Stop codon. Protein is truncated. Usually has serious consequences. F Silent: No change in amino acid. No consequence. Missense Mutation in Sickle Cell Anemia Base substitution results in a single amino acid change Glu --> Val XI. Mutations: Permanent changes in DNA n There are several possible types of mutations: II. Frameshift Mutation: Nucleotides which are inserted or deleted may change the gene's reading frame. Usually serious, because entire protein sequence after mutation may be disrupted. Effects of Different Types of Mutations Many Viruses Cause Disease in Animals Reproductive Cycle of an Animal Virus 1. Entry: Virus gets inside cell. F Attachment: Virus attaches to a specific receptor on cell surface. F Penetration: Virus fuses to cell membrane and enters cell. 2. Uncoating: Viral capsid releases genetic material. 3. Synthesis: Genetic material is copied, viral proteins are made. 4. Assembly: Genetic material is packaged into capsids. u Release: New viruses (50 -200) leave the cell through: F Lysis: Cells burst and die. F Budding: Cell does not necessarily die. Life Cycle of the Influenza (Flu) Virus The AIDS Virus Makes DNA from RNA u Human Immunodeficiency Virus (HIV): Causes AIDS. HIV is a retrovirus, which contains the enzyme reverse transcriptase. u Flow of genetic information is reversed: Transcription DNA Translation RNA Protein Reverse Transcription Viral DNA is inserted into host chromosome as a provirus. u HIV Contains Unique Enzyme Reverse Transcriptase Infection of a Cell by HIV

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