PLASMA PROTEINS

Plasma is non-cellular portion of blood. The total plasma protein level ranges from 6-7 gm/dl. Plasma contains many structurally and functionally different proteins. Plasma proteins are divided into two categories.

1. **Albumin:** Not precipitated by half-saturated ammonium sulfate.
2. **Globulin:** Precipitated by half-saturated ammonium sulfate.

The albumin constitutes over half of the total protein. Albumin level ranges from 3.5-5.5 gm/dl. Globulin ranges from 2-3 gm/dl. After the age of 40, albumin gradually declines with an increase in globulins. Albumin is found to be simple protein and a single entity. But globulin has been found to contain many components. Subglobulins are detected as bands on electrophoresis. They are α1, α2, β and γ-globulins.The different plasma protein bands are semi-quantitated using densitometer

Characteristics of Plasma Proteins

1. They are all glycoproteins except albumin. Sialic acid is the most important of all the sugars present in plasma proteins. Removal of sialic acid decreases the life span of plasma proteins.
2. Each plasma protein has defined life span. The half life of albumin is 20 days and haptoglobin life span is 15 days.
3. Liver is the sole source of albumin, prothrombin and fibrinogen. Most of the α and β globulins are also of hepatic origin. γ-globulins are derived from lymphocytes.

Albumin

Liver produces about 12 gms of albumin per day.

Structure

It consists of single polypeptide chain of 584 amino acid residues with a molecular weight of 66,300. Charged amino acids (glutamate, aspartate and lysine) make up a quarter of the total amino acid residues. The acidic residues out number the basic amino acids hence molecule is highly negative charged which accounts for the high mobility of albumin towards anode. Secondary structure of the protein is over half is in the α-helical conformation. 15% as β-pleated structure and remaining in random coil conformation. The tertiary structure is that of globular protein. The overall shape resembles ovoid. The hydrophobic amino acid residues are present in the hydrophobic interior and polar amino acids are arranged to face the exterior of the albumin. This accounts for the high solubility of the albumin in water (aqueous solutions).

Functions

1. Albumin accounts for 75% of the osmotic pressure (25 mm Hg) in blood and responsible for maintenance of blood volume.
2. Albumin has major role in the regulation of fluid distribution.
3. One gram of albumin hold 18 ml of fluid in the blood stream. Decrease in albumin level leads to accumulation of fluid which results in edema.
4. It transports fatty acids from adipose tissue to liver. Albumin also binds many hydrophobic

substances like bilirubin and several drugs. The binding of bilirubin is critical in neo-natal period.
5. Albumin act as a reservoir for Ca^{2+} in plasma. About 40% of plasma calcium is bound to albumin.
6. Albumin is also involved in the transport of thyroid hormones, glucocorticoids and sex steroids.
7. Albumin function as protein source for peripheral tissues. Each day liver replaces about 12 gm of albumin taken up by peripheral tissues. In certain conditions like stress and starvation the turn over rate of albumin is increased. Albumin is in dynamic equilibrium.
8. Albumin acts as a buffer.

α1-Globulin: Mainly α1-antitrypsin. It is a protease inhibitor. It is the major component of α1-fraction and accounts more than 90%. It inhibits trypsin, chymotrypsin, elastase and neutral protease. The major function of α1-antitrypsin is the protection of pulmonary tissue and other tissues from the destructive action of proteases.

α1-Acid glycoprotein (AAG): It is another major component of α1-globulins. It increases in plasma in inflammatory conditions.

Other components of α1-globulins are

α-Lipoprotein: Functions in the transport of lipids (HDL). It transports cholesterol from extra hepatic tissue to liver.

Prothrombin: Blood clotting factor.

Retinolbinding protein: Transport of Vit A.

Thyroxine binding globulin: Transport of thyroxine.

α1-Fetoprotein: It is present only in fetal serum. Its presence in non-foetal serum indicates primary carcinoma of liver. It is referred as tumour marker.

α2-Globulins: The α2-fraction of globulins includes.

Haptoglobulin: It combines with haemoglobin in order to remove it from the circulation. Kidney cannot filter haemoglobin-haptoglobin complex because of its larger size.

α2-Macroglobulin: It functions as protease inhibitor. It combines with proteases and facilitates their removal from circulation. It also binds with cytokines and involved in zinc transport.

Ceruloplasmin: A copper binding plasma protein and function as ferrooxidase and converts $Fe^{2+} \rightarrow Fe^{3+}$

Erythropoietin: It is involved in erythropoiesis.

Pseudocholinesterase: It is only functional enzyme present in plasma. It hydrolyzes acetylcholine.

β-Globulins: They are

Transferrin: It accounts for about 60% of β-globulins. It is an iron transport protein.

β-*Lipoproteins:* Involved in the transport of cholesterol from liver to extrahepatic tissue (LDL).

Complement-3: It is one of the member of complement system present in plasma. It

is involved in phagocytosis.

Other globulins present in plasma are:

Fibrinogen: It is similar to globulins because it is precipitated by half saturation with ammonium sulfate. It is a fibrous or filamentous protein. It is the precursor of fibrin, the blood clotting substances.

Prealbumin: It is a component of globulin fraction. Though it is a globulin by nature it is named as prealbumin because it migrates ahead of a albumin in electrophoresis. It is a carrier of thyroxine, Vitamin A and binds calcium.

Other blood clotting factors, plasminogen and several non-functional enzymes are also present in plasma.

Acute Phase Proteins or Reactants (APR)

1. The concentration of these proteins increases markedly during acute inflammation.
2. They are α1-antitrypsin, haptoglobin, ceruloplasmin, complement-3, fibrinogen and c-reactive protein. Their concentration increases in conditions like surgery, myocardial infraction, infections and tumours.
3. Acute phase reaction is general to any infection. They all play part in complex defensive process of inflammation.
4. The synthesis of these proteins by liver is triggered by interleukin at the site of injury.
5. The plasma levels of these APR raises at different rates. The levels of c-reactive protein raises first followed by α1-antitrypsin. The level of complement-3 raises at the end

γ-Globulins

The immunoglobulins and c-reactive protein (CRP) constitutes this fraction. C-reactive protein is so called because it forms precipitate with somatic C-polysaccharide of pneumococcus bacteria.

IMMUNOGLOBULINS

They are globulins produced as body's immune or defence against infection. Invasion of body by virus or microorganisms or foreign molecules is called *infection*. They are produced by B-lymphocytes, bone marrow and spleen in response to infection. Entry of foreign molecule into body triggers the synthesis of specific globulin, which selectively combines with foreign molecule and lead to its inactivation. The foreign molecule is called as *antigen* where as globulin produced against it is called as antibody. Even without infection the normal plasma contains hundreds of different antibody molecules.

Classification

The immunoglobulin (Ig) proteins of plasma are divided into three major classes Ig G, Ig A, Ig M and two minor classes Ig D, Ig E based on their composition.

Structure

The composition and shape of various classes of immunoglobulins have similar pattern and are represented by the structure of major G class of molecule *i.e.*, Ig G. Each Ig G molecule consist of 4 polypeptide chains and molecular weight is 150,000. The four polypeptide chains are of two types. They are two heavy chains or H chains or about 450 amino acids (molecular weight

50,000) and two light or L chains or about 220 amino acids (molecular weight 25,000). Over all shape of the molecule represents 'Y'. Two heavy chains intertwine to form the base of the Y, a disulfide bond links the L chain to H chain to form arm of the Y. The two heavy chains are held together by disulfide bonds formed between them at the hinge region of the Y
The H chain contains variable region of domain (VH) at the N-terminus and three constant domains (CH1, CH2, CH3) at the C-terminus. Likewise L chain consists of variable domain (VL) at the N-terminus and a constant domain (CL) at the C-terminus. The carbohydrate is attached to CH2 of the heavy chain. The amino acid sequence in the variable regions of H and L chains varies and are specific to the type of antibody. In contrast amino acid sequence in constant region of H and L chains are same in each class of immunoglobulins. The antigen binding site is called as *Fab site*. It consists of light chain and N-terminal half of the heavy chain. The remaining part of the immunoglobulin is called as Fc (fragment with constant domain).
The different classes of immunoglobulins vary in their size, distribution, function and composition. The main chemical differences are found in their H chains. They are named according to the types of H chain present. There are five classes of H chains. They are γ, α, μ, δ, ε. However, there are only two classes of L chains κ or λ.

Different Classes of Immunoglobulins

1. Ig G class

It constitutes 70 to 80% serum immunoglobulins. Its composition is γ2L2 (γ2k2 or γ2λ2). It is the only class of antibody that is capable of crossing the placental barrier from the maternal to fetal circulation. It is the antibody of newborn until synthesis of immunoglobulins in the body *i.e.*, up to 2 years of age. Ig G antibodies bind to phagocytic cells thus making a link between antibody and phagocytes. Further, binding of Ig G to foreign cells increases their susceptibility to killer cell attack.

2. Ig A class

It accounts for 10-20% of immunoglobulins. Its basic composition is (α2L2), SCJ and it also exists as multimer of the basic unit (α2L2)*n* where *n* = 1, 2, 3 etc. It is the chief antibody present in mucous secretions of lungs and gastrointestinal tract. Mucosal cells add one more polypeptide chain known as *secretory component* (SC), joining H chains of Ig A dimers before passage into secretions. They form aggregates with antigen in the gut and lungs thus prevent the entry of such harmful substances into the body .

3. Ig M class

It accounts for about 5-10% of total immunoglobulins. Like Ig A class, it is also a multimer of basic tetramer. Its composition is (μ2L2)5 J *i.e.*, it is a pentamer of basic unit. The H chains are joined by JC chain. When these are present in secretions of mucous membranes they may contain SC component also. It is the largest of all the immunoglobulins.

IgM act as antigen receptor on B-lymphocytes. It is also involved in complement fixation.

IgM molecules are first to appear in infancy.

4. Ig D class

It accounts less than 0.5% of total immunoglobulins. Its composition is δ2L2. The biological activity of Ig D appears to be limited. It is not a secretory antibody. It is involved in the initiation of alternate pathway of complement fixation.

5. Ig E class

It is least concentrated and has shortest life span of all the immunoglobulins. Its composition is ε2L2. Ig E concentration increases in allergic reactions. It is a surface antibody of cells involved in anaphylactic response. The constant region of the antibody is bound to membrane receptor of leukocytes or mast cells and variable region is exposed to the outer surface. When the specific antigen reacts with antibody, it triggers the cells to release histamine and other vasoactive amines. The Ig E class also found in secretions of lungs and gut but the Ig Es lack the J chain and SC part found in Ig As and Ig M

Immunoglobulins Disorders

There are numerous disorders associated with different classes of immunoglobulins.

1. Multiplemyeloma

It is a malignant disease of single clone (cell type) of plasma cells of the bone marrow. These plasma cells proliferate throughout bone marrow. Other bone marrow cells are reduced. Tumours of the plasma cells produce myeloma proteins.

The incidence is low in individuals younger than 60 years but raises with age. Symptoms include recurrent infections, weight loss, bone lesions, anaemia and haemorrhages.

Bence-Jones proteins

They are immunoglobulins light chains present in plasma and urine of multiple myeloma patients. The molecular weight is 2500. They are found with γ-globulin fraction on electrophoresis. The characteristic property of these proteins is their behaviour on heating. The normal plasma proteins precipitates between 60-70°C. The Bence-Jones proteins precipitate at 40-60 °C completely. Redissolving of the precipitate occurs as the temperature reaches boiling point. Subsequent cooling reprecipitates the protein and boiling redissolves it. They are identified in the urine of the suspected individuals based on this property.

2. Agammaglobulinemia

It is x-chromosome linked and affects only males. γ-globulins are absent in plasma of these patients. So they are prone to infections.

3. Hypogammaglobulinemia

Production of γ-globulins is decreased in these cases.

4. Autoimmune disorders

Sometimes body rejects its own proteins which becomes antigenic. This results in auto immune disorders due to production of antibodies against its own proteins. Rheumatoid arthritis is known auto immune disorder.

Catalytic Antibodies or Abzymes

1. Immunoglobulins bearing catalytic activity of an enzyme are produced using an enzyme active site as the antigen.

2. The first step consists of producing an antibody A1 against the active site of an enzyme.
3. Enzyme inhibition studies are used to confirm that A1 contains active site close to enzyme active site.
4. Then A1 is used to produce second generation A2 antibodies having specific catalytic activity.
5. They are used to remove toxins or viral coat proteins present in the body.

NUCLEIC ACIDS

OCCURRENCE

Two types of nucleic acids are present in all mammalian cells including humans. They are DNA-deoxy ribonucleic acid and RNA-ribonucleic acid. DNA is present in nucleus and mitochondria. RNA is present in nucleus and cytoplasm. Nucleic acids are also present in bacteria, viruses and plants.

MEDICAL AND BIOLOGICAL IMPORTANCE

1. Nucleic acids serve as genetic material of living organisms including humans.
2. Nucleic acids are involved in the storage, transfer and expression of genetic information.
3. Nucleic acids contain all the necessary information required for the formation of individual or organism.
4. Nucleic acids determine physical fitness of an individual to life.
5. Some nucleic acids act as enzymes and coenzymes. For example, RNA, act as catalyst and RNA is coenzyme for telomerase which seals ends of chromosomes.
6. DNA exhibits structural polymorphism. It assumes several forms depending on certain conditions. Several DNA variants are known.
7. Some RNAs without protein products are found recently in mammals, yeast and bacteria. They are involved in cellular functions.
8. Human Genome Project (HGP) is completed in 2000. It is considered as a major achievement of man after landing on moon. It is useful for finding causes of several diseases whose causes are unknown till. It may also lead to development of new therapeutics as well as diagnostics.

Chemical nature of nucleic acids

Nucleic acids are acidic substances containing nitrogenous bases, sugar and phosphorus. Both DNA and RNA are polynucleotides. They are polymers of nucleotides.

Phosphodiester linkage

In polynucleotides, nucleotides are joined together by phosphodiester linkage. Diester linkage of phosphate joins 3′ OH and 5′ OH belonging two separate sugars (Figure 16.1).

Nucleic acid structure

Primary structure of nucleic acids

Nucleotide sequence of a polynucleotide is known as primary structure of nucleic acid. The primary structure confers individuality to polynucleotide chain. Polynucleotide chain has direction. They are represented in 5′ → 3′ direction only. However, the phosphodiester

linkage runs in 3′ → 5′ direction. Each poly nucleotide chain has two ends. The 5′ end carrying phosphate is shown on the left hand side and 3′ end carrying unreacted hydroxyl is shown on the right hand side . Primary structures of DNA and RNA exist in single stranded DNA and RNA organisms.

Since polynucleotide consists of various bases, sugars and phosphates writing a segment of polynucleotide showing structures of bases, sugars with attached phosphates is awkward or highly inconvenient. So, short hand or compact representation of polynucleotide has been proposed. In compact nomenclature or polynucleotide letters A, G, C and T represents nitrogenous bases adenine, guanine, cytosine and thymine, respectively. A vertical line represents sugar back bone. The branches of verticle lines with numerals 3′ and 5′ represents hydroxyl bearing carbon atoms of sugar. A branch at the middle of the verticle line represents hydroxyl bearing 3rd carbon atom of sugar. Another branch at the bottom of verticle line represents hydroxyl or phosphate bearing 5th carbon atom of sugar. The more compact representation of the same molecule is PAPCPGPTPA. Since primary structure is the sequence of nucleotides still more compact representation of the same molecule is ACGTA. In this primary structure, letters A, G, C, T stands for nucleotides and sequence is written from left to right. Therefore, in DNA and RNA, letters A, G, C, T stands for nucleotides and sugar is deoxy ribose if the polynucleotide is a segment of DNA and sugar is ribose if it is a RNA segment. Remember that letters A, C, U, G, T stands for nucleosides in the case of nucleotides.

Structure of DNA

E. Chargoff and his colleagues extensively studied base composition of DNA. Their studies provided valuable information on the structure of DNA.

Characteristics of DNA base composition

1. In DNA, number of adenine residues is equal to the number of thymine residues *i.e.*, A = T. Further number of guanine residues is equal to number of cytosine residues *i.e.*, G = C. As corollary sum of purine residues is equal to sum of pyrimidine residues A + G = C + T.
2. DNAs from different tissues of same species have same base composition.
3. Base composition of DNA varies from one species to another species.
4. DNAs from closely related species have similar base composition.
5. DNAs of widely different species have different base composition.
6. DNA base composition of a species is not affected by age, nutritional state and environment.

In 1953, J.D. Watson and F.H.C. Crick proposed precise three dimentional model of DNA structure based on model building studies, base composition and X-ray diffraction studies. This model is popularly known as DNA double helix. Using this model, they also suggested a precise mechanism for the transfer of genetic information to daughter cells from parent cells.

Salient features of double helix

1. Two polynucleotide chains are coiled around a central axis in the form of right handed double helix. It represents secondary structure of DNA. It is present in double stranded DNA containing organisms.
2. Each polynucleotide chain is made up of 4 types of nucleotides. They are adenylate, guanylate, thymidylate and cytidylate.
3. Each polynucleotide chain has direction or polarity. Further each polynucleotide chain has 5′ phosphorylated and 3′ hydroxyl end.
4. The back bone of each strand consist of alternating sugar and phosphates. The bases projects inwards and they are perpendicular to the central axis.
5. The two strands run in opposite direction, i.e., they are anti-parallel.
6. The strands are complementary to each other. Base composition of one strand is complementary to the opposite strand. If adenine appears in one strand thymine is found in the opposite strand and vice versa. Where ever guanine is found in one strand cytosine is present in the opposite strand and vice versa.
7. **Base pairing** Bases of opposite strands are involved in pairing. Pairing occurs through hydrogen bonding and it is specific. Adenine of one strand pairs with thymine of opposite strand through two hydrogen bonds. Guanine of one strand pairs with cytosine of opposite strand. Three hydrogen bonds between GC pair makes it more stronger than two hydrogen AT pair .
(*a*) DNA double helix
(*b*) Base pairing among complementary bases of opposite strands
(*c*) Alternating sugar and phosphate form back bone of strand. Bases project inwards and perpendicular to central axis
8. Complementarity of strands and base pairing are the outstanding features of Watson-Crick model. Specific base pairing immediately suggests a copying mechanism for DNA.
9. The large number of hydrogen bonds along entire length of DNA makes DNA molecule highly stable.
10. Major and minor grooves are present on double helix. They arise because glycosidic bonds of base pairs are not opposite to each other.
11. The base pairs are stacked and 3.4 Å apart. The pitch of the helix (One turn) is 34 Ao and accommodates ten base pairs.
12. Apart from hydrogen bonding, the double helix is stabilized by hydrophobic attraction between bases.
13. The width of double helix is 20 Å.
14. Watson-Crick model is known as B-DNA. Majority of the nuclear DNA is in B-form.

Functions of DNA

1. DNA is the genetic material of living systems. It is super chip ever made by man present in living systems.
2. DNA contains all the information required for the formation of an individual or organism.
3. The genetic information in DNA is converted to characteristic features of living organisms

like colour of the skin and eye, height, intelligence, ability to metabolize particular substance, ability to with stand stress, susceptibility to disease and unable to produce or synthesize certain substances etc.

4. All the above phenotype characters of living organisms are intimately related to functions of proteins. Thus, DNA is the source of information for the synthesis of all cellular proteins. The segment of DNA that contains information for a protein is known as *gene*.
5. DNA is transmitted from parent to off spring and hence DNA flows from one generation to other in a given species. Further, DNA provides information inherited by daughter cells from parent cells.
6. The amount of DNA per cell is proportional to the complexity of the organism and hence to the amount of genetic information. The amount of DNA in mammalian cell is 1000 times more than bacteria. Likewise, bacteria contain more DNA than virus and plasmids.
7. The amount of DNA in any given species or cell is constant and is not affected by nutritional or metabolic states.

DNA as the gene

Studies on bacterial transformation carried out by Avery and his colleagues provided first experimental evidence to prove DNA is genetic material in living organisms. They used two types of pneumococci. They are virulent (pathogenic) and avirulent (non-pathogenic) types. DNA isolated from heat killed virulent organism when introduced into avirulent organism it transformed avirulent organism into virulent organism. Deoxy ribonuclease treatment of DNA isolated prior to introduction destroyed transforming capacity of DNA. These observations indicated that DNA is a genetic material.

Mitochondrial DNA

Eukaryotic mitochondria contains DNA. It is different from DNA present in nucleus. It account for 1% of cellular DNA. Base composition of mitochondrial DNA is different from nuclear DNA. Mitochondrial DNA is double stranded and circular.

Bacterial DNA

Bacteria like *E. Coli* contains single molecule of double stranded DNA. *E. Coli* DNA is 1.4 mm long which is 700 times bigger than the size of bacteria. Hence in bacteria also DNA is tightly packed or folded. In *E. Coli* the two ends of DNA are joined to form circular DNA. Histones are not used for packing of bacterial DNA because they are absent in bacteria. Super coiling of circular DNA allows its containment with in nuclear zone. Super-coiled DNA may be in association with some proteins, which stabilizes super coil.

Viral DNA

Viruses are extremely small particles. They are composed of a piece of DNA, which is surrounded by protein coat called *capsid*. Viral DNA may be single stranded or double stranded. Adeno virus (cold virus), Herpes virus and Pox virus are examples for double stranded viruses. Parvo virus is a example for single strand DNA virus.

Plasmids

They exist in bacteria as circular DNA molecules. Plasmid DNA is different from bacterial DNA. They are present in anti-biotic resistant bacteria. They contain genes for inactivation of anti-biotics. pBR 322 of *E. Coli* is an example for plasmid. Plasmids are used as vectors in genetic engineering.

Denaturation of DNA

When DNA molecule is heated it denatures and strands separate. Thermal denaturation of DNA is known as melting of DNA. Melting point of DNA is known as *Tm*. It is a characteristic of given DNA. If the heat denatured DNA is cooled base pairing occurs between strands and reformation of double, stranded molecule takes place. This process is known as *annealing*. It is very useful in genetic engineering particularly in DNA hybridization techniques

Ribonucleic acids (RNAs)

Ribonucleic acids are present in nucleus and cytoplasm of eukaryotic cells. They are also present in prokaryotes. They are involved in the transfer and expression of genetic information. They act as primers for DNA formation. Some RNA act as enzymes as well as coenzymes. RNA also function as genetic material for viruses.

Chemical nature of ribonucleic acids

Like DNAs, RNAs are also poly nucleotides. In RNA polymer, purine and pyrimidine nucleotides are linked together through phosphodiester linkage. The sugar present in a RNA is ribose.

There are mainly three types of RNAs in all prokaryotic and eukaryotic cells. The three types of RNA are 1. Messenger RNA or m-RNA, 2. Transfer RNA or t-RNA, 3. Ribosmal RNA or r-RNA. They differ from each other by size, function and stability.

Messenger RNA

It accounts for 1-5% of cellular RNA.

Structure

1. Majority of mRNA has primary structure. They are single-stranded linear molecules. They consist of 1000-10,000 nucleotides.
2. mRNA molecules have free or phosphorylated 3’ and 5’ end.
3. mRNA molecules have different life spans. Their life span ranges from few minutes to days.
4. Eukaryotic mRNA are more stable than prokaryotic mRNA.
5. The mRNA nucleotide sequence is complementary from which it is synthesized or copied.
6. Some eukaryotic mRNA molecules are capped at 5’ end. The cap is methylated GTP (m7 GTP). Some mRNA contain internal methylated nucleotides. Capping protects mRNA from nuclease attack.
7. At 3′ end of most of eukaryotic mRNA, a polymer of adenylate (poly A) is found as tail. Poly A tail protects mRNA from nucleaes attack.
8. In prokaryotes 5′ end of mRNA contains a sequence rich in A and G. Such sequence is known as *Shine-Dalgarno sequence*. It helps attachment of mRNA with ribosome during

protein synthesis.
9. Some prokaryotic mRNA has secondary structure. Intrastrand base paring among complementary
bases allows folding of liner molecule. As a result hairpin, or loop like
secondary structure is formed. (Figure 16.7b).

Functions

1. mRNA is direct carrier of genetic information from the nucleus to the cytoplasm.
2. Usually a molecule of mRNA contains information required for the formation of one protein molecule.
3. Genetic information is present in mRNA in the form of genetic code.
4. Sometimes single mRNA may contain information for the formation of more than one protein.

Transfer RNA

t-RNA accounts for 10-15% of total cell RNA.

Structure

They are the smallest of all the RNAs. Usually they consist of 50-100 nucleotides. They are single strand molecules. t-RNA molecules contain many unusual bases 7-15 per molecule. They are methylated adenine, guanine, cytosine and thymine, dihydrouracil, pseudo uridine, isopentenyl adenine etc. These unusual bases are important for binding of t-RNA to ribosomes and interaction of t-RNA with aminoacyl-t-RNA synthetases. About half of the nucleotides in t-RNA are involved in intrachain base pairing. As a result, double helical segments are formed in t-RNA. Further some bases are not involved in the base pairing resulting in loops and arms formation in t-RNA. Thus, folding in primary structure generate secondary structure. Though t-RNAs differ in chain lengths they have some common features with regard to secondary structure.

Secondary structure of t-RNA

Secondary structure of all the t-RNAs is in the form of clover leaf

1. An amino acid arm where amino acid is attached to 3′-OH of adenosine moiety of t-RNA. ACC is the common base sequence at this 3′-end.
2. Tϕc arm, which contains sequence of ribothymidine-pseudouridine-cytidine. Greek alphabet ϕ (Psi) stands for pseudo uridine. Thymine and pseudouracil are the two unusual bases found in this arm.
3. An anti-codon arm, which recognizes codon on mRNA.
4. DHU arm, which contains many dihydrouridine (UH2) residues.
5. The 5′ end of t-RNA is phosphorylated and residue is guanosine.
6. About 75% t-RNA molecules have extra arm. It consist of 3-5 base pairs. It is found between TϕC and anti-codon arm.

Tertiary structure of t-RNA

X-ray diffraction analysis indicated complex three-dimentional structure for t-RNA molecule. Three-dimentional structure of t-RNA looks like inverted or tilted L. The anti-codon arm is

at the tip of the vertical arm of tilted L. The acceptor arm is at the tip of horizontal arm of tilted L. The D loop and TϕC loop are pushed into corner of tilted L.

Functions

1. It is the carrier of amino acids to the site of protein synthesis.
2. There is at least one t-RNA molecule to each of 20 amino acids required for protein synthesis.
3. Eukaryotic t-RNAs are less stable where as prokaryotic RNAs are more stable.

Ribosomal RNA

Ribosomal RNA or r-RNA accounts for 80% of total cellular RNA. It is present in ribosomes. In ribosomes, r-RNA is found in combination with protein. It is known as *ribonucleoprotein*. The length of r-RNA ranges form 100-600 nucleotides. Both prokaryotic and eukaryotic ribosomes contain r-RNA molecules. r-RNAs differ in sedimentation coefficients (S). There are four types of r-RNAs in eukaryotes. They are 5, 5.8, 18 and 28S r-RNA molecules. Prokaryotes contains 3 types of r-RNA molecules. They are 5, 16 and 23S r-RNA molecules.

Structure

r-RNA molecules have secondary structure. Intra strand base pairing between complementary base generates double helical segments or loops. They are known as domains. 16S r-RNA with 1500 nucleotides has four major domains (Figure 16.8c). The three-dimentional tertiary structure of r-RNA is highly complex.

Functions

1. r-RNAs are required for the formation of ribosomes.
2. 16S RNA is involved in initiation of protein synthesis.

Differences between DNA and RNA

DNA	**RNA**
1. Sugar moiety is deoxy ribose	Sugar moiety is ribose
2. Uracil, a pyrimidine base is usually absent	Thymine, a pyrimidine base is absent
3. Double-stranded molecules	Single stranded molecules
4. Sum of purine bases is equal to sum of pyrimidine bases A + G = C + T A + G # C + T	Sum of purine bases is not equal to sum pyrimidine bases
5. Resistant to hydrolysis by alkali because of absence of hydroxyl group on 2 carbon atom of deoxyribose	Because of presence of hydroxyl group on 2 carbon atom of ribose RNA is easily hydrolyzed by alkali
6. Bases are not modified	Bases are modified
7. No catalytic activity	Some RNA are catalytically active
8. Only one form or type	More than three types
9. Usually not subjected to degradation in cell	Degraded in the cell by nucleases

REFERENCES

1. Freifelder, D. Molecular Biology. 2nd ed. Jones and Bartlett Publishers, Boston, 1987.

2. Watson, J.D. and Crick, F.H.C. Molecular structure of nucleic acids. A structure for DNA. Nature **171**, 737-738, 1953.
3. Saenger, W. Principles of nucleic acid structure. Springer-Verlag, New York, 1984.
4. Schimmel, P. Soll, D. and Abelson, J. Eds. Transfer RNA. Cold spring Harbor Laboratory. New York, 1979.
5. Van Holde, K.E. Chromatin. Springer–Verlag. New York, 1988.
6. Davidson, J.N. The biochemistry of nucleic acids. Academic Press, New York, 1972.
7. Rich, A. and Raj Bhandary, V.L. Transfer RNA: Molecular structure, sequence and properties. Ann. Rev. Biochem. **45**, 805, 1976.
8. Brown, T and Brown, T.W. Genomes. Wiley-Liss, 2002.
9. Gesteland, R.F., Cech, T.R. and Atkins, J.F. (Eds.). The RNA World. Cold Spring Harbor Laboratory, NY, 1999.
10. Watson, J.D.A. passion for DNA: genes, genomes and Society. Cold Spring Harbor Laboratory Press, 2000.
11. Driel, R.V. and Arie, P.O. Nuclear organization, chromatin structure and gene expression. Oxford University Press, NY 1997.
12. Donald M. Crothers, Nucleic acids: structure, properties and functions, University Science Books, 2000.
13. Olby, R. Quiet debut for the double helix. Nature **421**, 402-405, 2003.
14. Parkinson, G.N. Lee M.P.H. and Neidle, S. Crystal structure of parallel quadruplexes from human telomeric DNA. Nature **417**, 876, 2002.
15. Sumen, N.C. DNA in material world. Nature **421**, 427-431, 2003.
16. Ariyoshi, M. *et al*. Crystal structure of the holliday junction DNA in complex with a single RuvAtetramer. Proc. Natl. Acad. Sci. USA **97**, 8257-8262, 2002.
17. Contor, C.R. and Smith C.L. Genomics: the science and technology behind human genome project (E-book). J. Wiley. New York, 2004.
18. Myers, E.W. Sutton, G.G. Smith, H.O. Ademson, D. and Venter, J. Craig. On the sequencing and assembly of human genome. Proc. Natl. Acad. Sci. USA **99**, 4145-4146, 2002.
19. Venter, J.C. *et al.* The sequence of human genome. Science. **291**, 1304-1351, 2001.
20. International human genome sequencing consortium (HGSC). Nature. **409**, 860-921, 2001.
21. Vanderpool, C.K. and Gottesman. S. Non-coding RNAs at the membrane. Nature Structural and Molecular Biology. Vol. 12, April 2005.