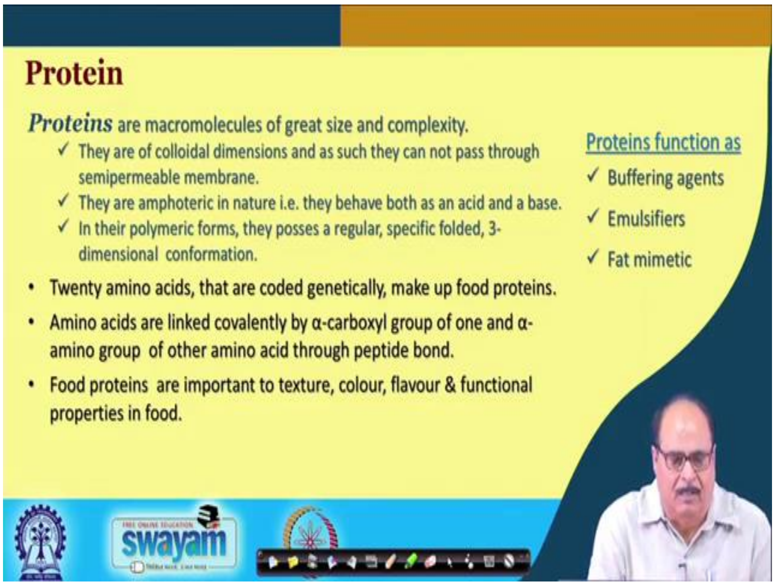
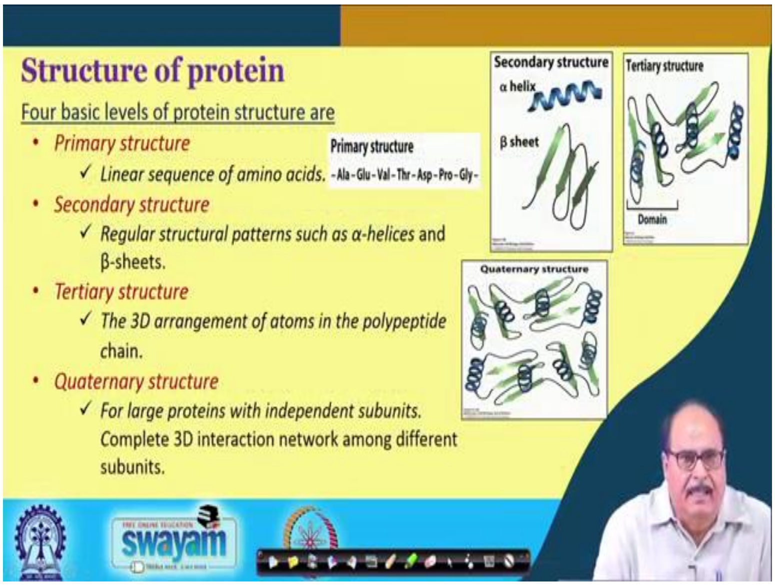
Novel Technologies for Food Processing and Shelf Life Extension
Prof. Hari Niwas Mishra
Department of Agricultural and Food Engineering
Indian Institute of Technology, Kharagpur

Lecture – 07
Food Proteins
(Denaturation, Foaming & Gelation)

An important process relating to protein foods including the chemistry of processes like denaturation, foaming and gelation, and the influence of processes on the characteristics and properties of the food materials in different operations will be discussed in this lecture.



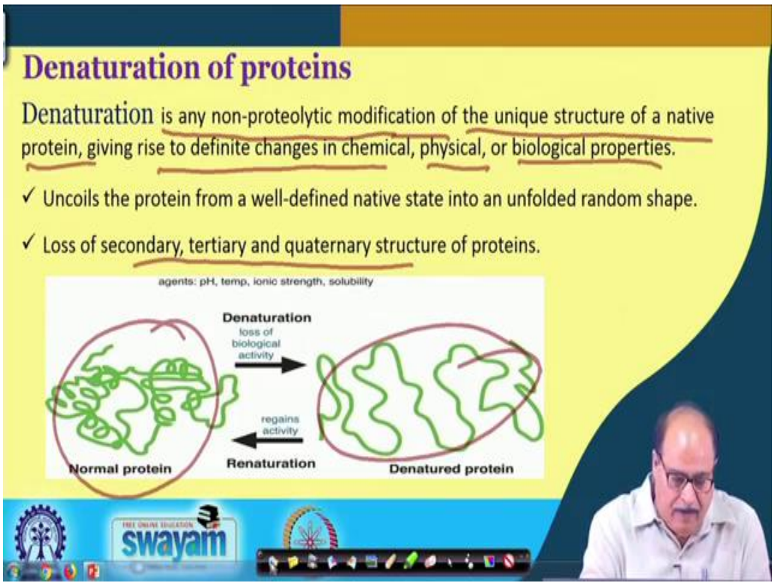
Proteins are macromolecules of great size and complexity. They are of colloidal dimensions, and as such they cannot pass through semipermeable membrane. They are amphoteric in nature; i.e. they behave both as an acid and as a base. In their polymeric forms, they possess a regular specific folded 3-dimensional confirmation. About 20 amino acids are coded genetically, which make up food proteins. These amino acids are linked covalently by α-carboxyl group of one amino acid and α-amino group of the other amino acid through peptide linkage. Food proteins are important to texture, colour, flavor, and functional properties of food. Proteins function as buffering agents, emulsifiers, and fat mimetic agents.



Structure of protein can be categorized into four levels: Primary structure; which implies the linear sequence of amino acids, i.e. how these amino acids are sequenced. Even the specific position of the amino acid is very important. So, number of amino acids and linear sequence is taken in the primary structure.

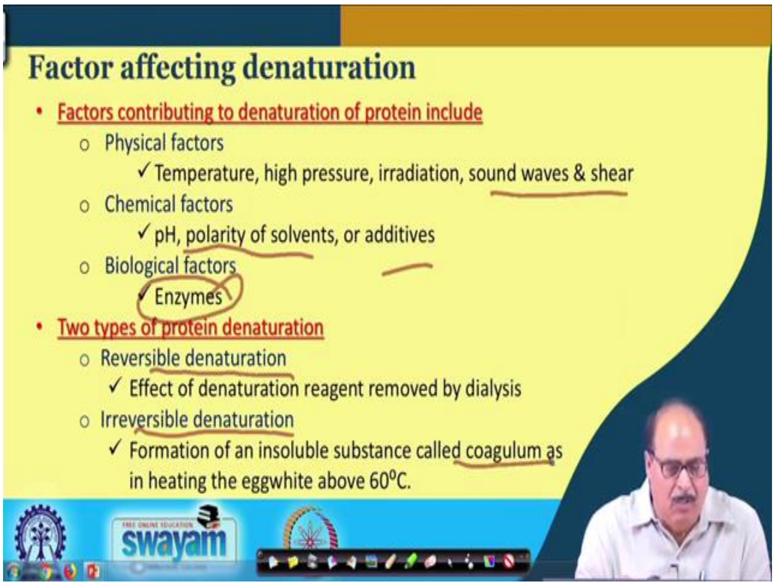
Secondary structure of a protein involves regular structural patterns, such as α-helical or β-sheet configurations which a protein might take up. In the tertiary structure, 3D arrangements of the atoms in the polypeptide chains are understood. That is how a specific random coiling takes place and then folding in the secondary and tertiary structures.

So, these three primary, secondary and tertiary structures normally deal with about single polypeptide chain which are called subunits. And it may so happen that, in a food protein there might be more than one subunits. So, how these different subunits interact among each other to give the final shape of the protein molecules are understood in quaternary structure.



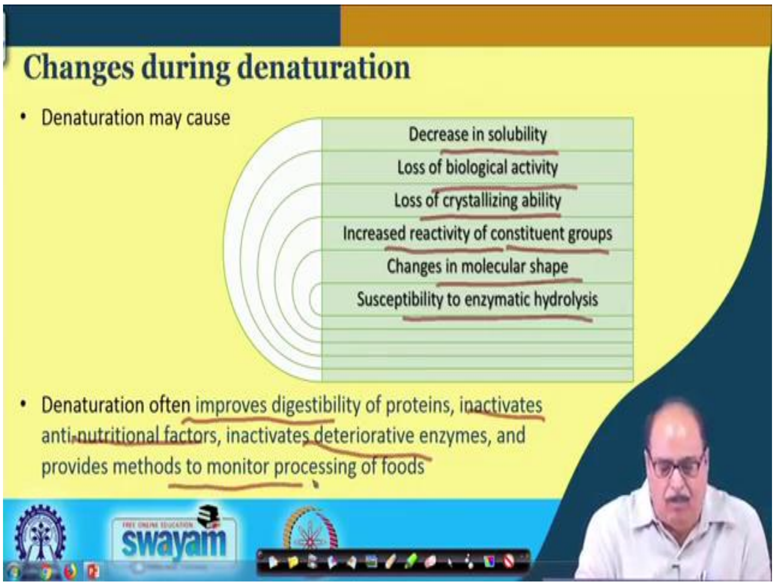
When these food proteins come across different agents, different conditions during processing and handling, certain changes take place in the characteristics, structure and properties of the protein. This is called as “denaturation”. So, the denaturation of a protein may be any non-proteolytic modification of the unique structure of a native protein, giving rise to definite changes in its chemical, physical or biological properties. There are no proteolytic decomposition, no proteolysis, there is no breakage of that peptide linkage during denaturation process. It may be unfolding, uncoiling of the organized protein structure.

It can be seen from the picture that, the normal protein is structured; during denaturation process, the organized structure is opened or unfolded. In some cases, it may again renature to get the structure. So, actually in denaturation there is loss of secondary, tertiary, and quaternary structure of protein, but definitely not the primary structure.



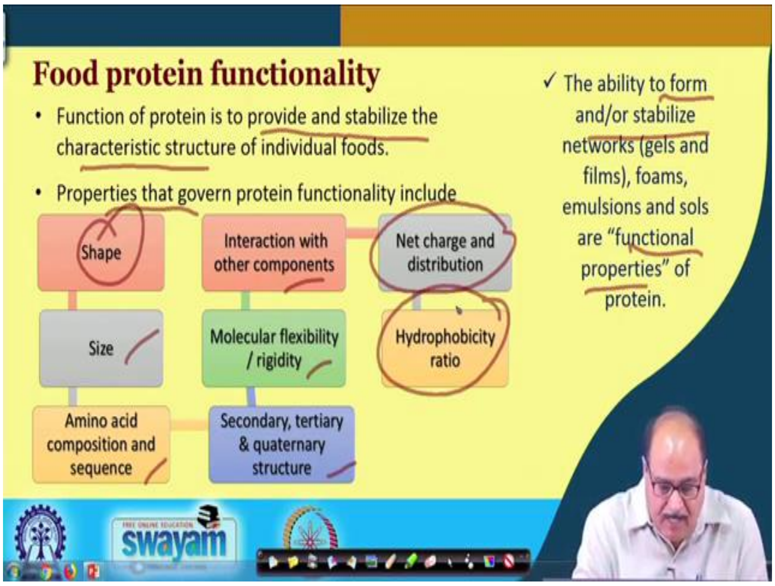
Thus, there are various factors which influence the denaturation process. These factors may be the physical factors, like temperature, high pressure, irradiation, sound waves, and shear forces etc. to which the proteins come across during processing or handling. The chemical factors like pH, polarity of solvents or additives, even the biological factors such as enzymes influence the rate of denaturation process in protein foods.

There may be two types of denaturation reactions or processes depending upon the factors, food characteristics, and the severity of the process to which these food proteins are exposed to. It may be a reversible denaturation or irreversible denaturation. In the reversible denaturation, the effect of denaturation reagent, if they are removed by processes like dialysis etc. then the protein may again renature. But in the case of irreversible denaturation, there is formation of an insoluble substance called generally as coagulum. As happens, when the egg is boiled, while heating the egg white above a certain temperature may be 50-60 °C, the coagulation of protein takes place; this is an example of irreversible denaturation process.



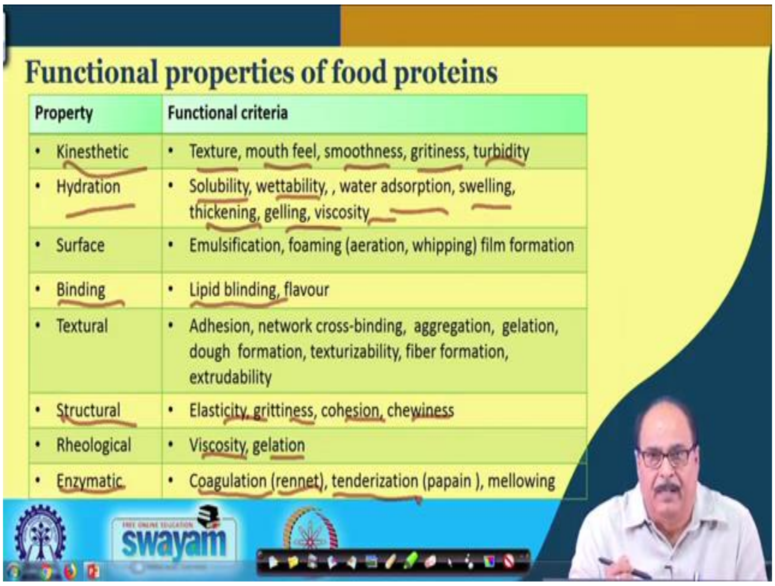
The different changes that take place in protein characteristics during denaturation include: decrease in its solubility, there will be loss of biological activity of the protein, it might not be possible to crystallize the protein. Once it is denatured there might be increased reactivity of constituent groups. There may be a change in the molecular shape of the protein or it may become more susceptible to enzymatic hydrolysis.

Denaturation often improves the functionality of the protein, like digestibility, it also inactivates sometimes the anti-nutritional factors, it might inactivate deteriorative enzymes, and may also provide method to monitor processing of food.

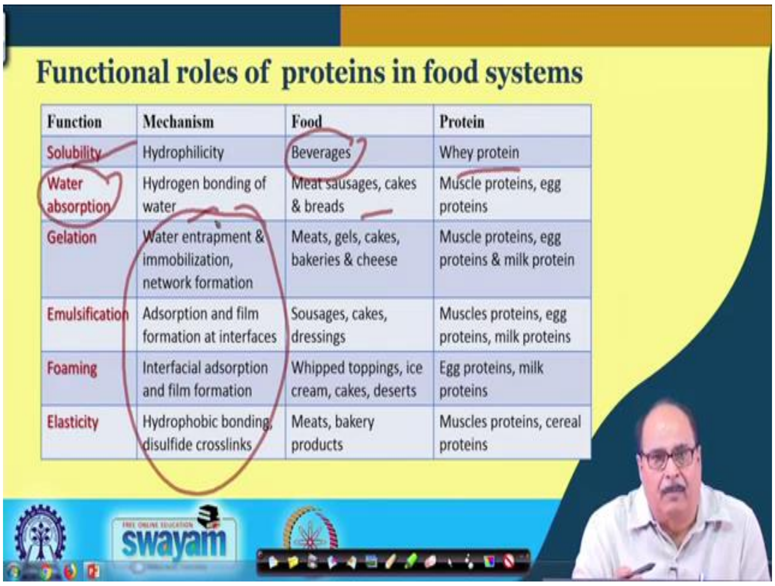


Functional characteristics of the protein food:

The function of protein is to provide and stabilize the characteristic structure of an individual food. The ability to form or stabilize networks i.e. gels, films, foams, and emulsions etc. or the sols are described as functional properties of the protein. Various characteristics that govern the protein functionality include shape of the protein, its size, amino acid composition and sequence, how the protein interacts with other components, and how the molecular flexibility or rigidity is: its secondary, tertiary or quaternary structure. Even the net charge on the protein molecule or the distribution of the protein molecule or even hydrophobicity ratio of the protein will influence its functionality.

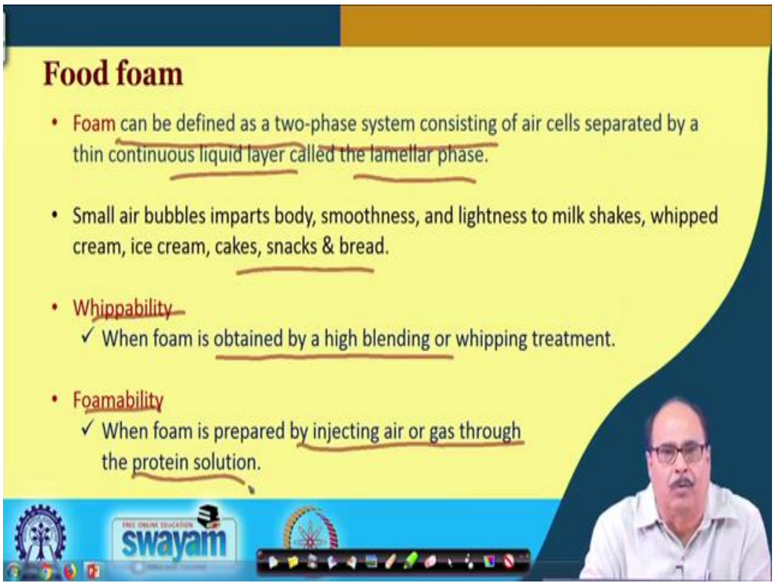


Descriptions about functional properties like hydration, textural attributes (mouth feel, smoothness, grittiness, cohesion, chewiness or turbidity), binding property, structural characteristics, rheological properties like elasticity, viscosity, gelation etc. of the food proteins are provided. Solubility, wettability, water absorption, swelling, thickening, gelling, all these characteristics might be influenced by the hydration characteristics of a protein. Even the enzymatic properties i.e. there might be coagulation, like in the cheese making, the rennet enzyme coagulates the protein. There may be certain enzymes which may cause tenderization of the protein. Example: the papain enzyme causes the softening of the hard tissues of meat i.e. meat becomes tender, there might be mellowing etc.

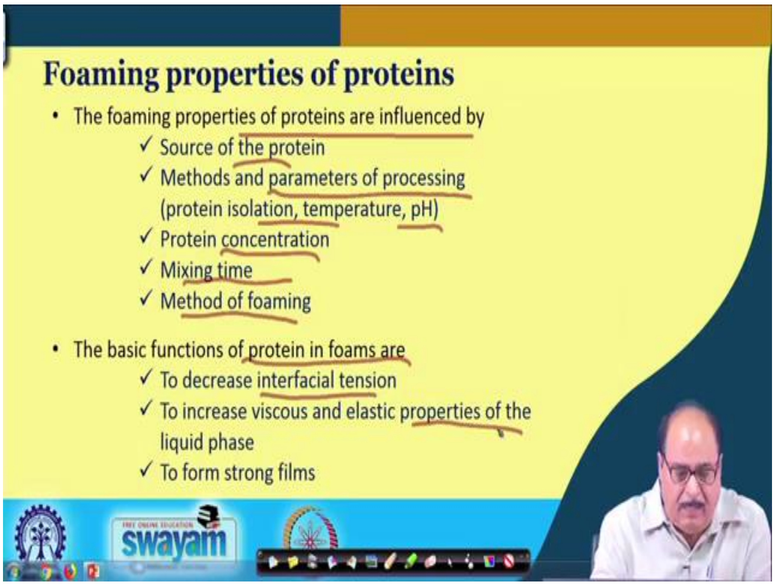


Function	Mechanism	Food	Protein
Solubility	Hydrophilicity	Beverages	Whey protein
Water absorption	Hydrogen bonding of water	Meat sausages, cakes & breads	Muscle proteins, egg proteins
Gelation	Water entrapment & immobilization, network formation	Meats, gels, cakes, bakeries & cheese	Muscle proteins, egg proteins & milk protein
Emulsification	Adsorption and film formation at interfaces	Sousages, cakes, dressings	Muscles proteins, egg proteins, milk proteins
Foaming	Interfacial adsorption and film formation	Whipped toppings, ice cream, cakes, deserts	Egg proteins, milk proteins
Elasticity	Hydrophobic bonding, disulfide crosslinks	Meats, bakery products	Muscles proteins, cereal proteins

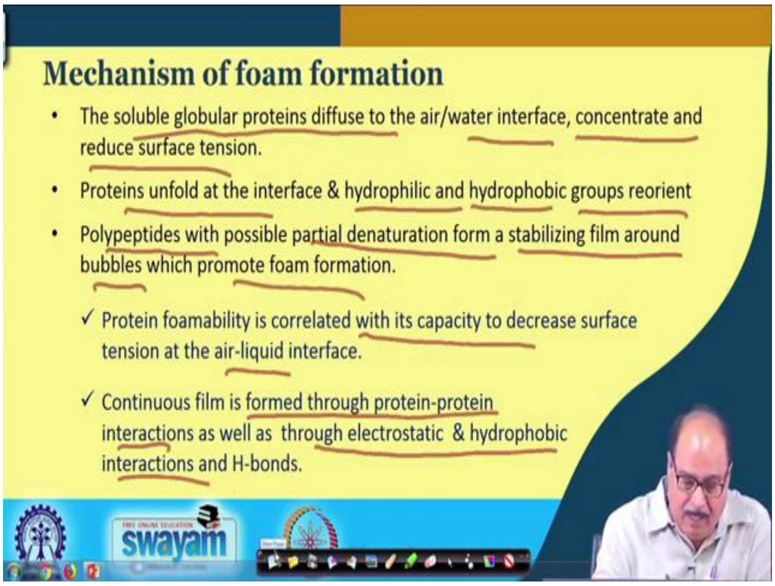
The different mechanisms that decide a food's functionality can be seen here in the table. Few examples like, in beverage and foods like milk, whey protein etc., hydrophilicity of the protein will ultimately influence its solubility. Similarly, the water absorption characteristics of the protein may be influenced by its ability to bind with water i.e. to form hydrogen bonds with water. This happens generally in the case of meats, sausage cakes, breads etc., where these different proteins bind with the hydrogen bonds and give the desired characteristics. Similarly, the other processes like gelation, emulsification, foaming, elasticity etc. influence the functionality. In case of gelation, the water entrapment and immobilization which might ultimately result to the network formation is the mechanism. The mechanism of emulsification might be adsorption and film formation at the interfaces. Elasticity may be because of the hydrophobic bonding or even formation of disulphide cross linkages. So, this explains briefly how these different functional characteristics of the proteins are influenced.



A foam can be defined as a two phase system consisting of air cells separated by a continuous liquid layer which is generally called the lamellar phase. Small air bubbles impart body, smoothness, and lightness to milkshakes, whipped cream, ice cream, cake, snacks and bread, etc. There are two important characteristics of the foam: whippability and foamability. When the protein containing materials are whipped or blended by some mechanical means, some foam is generated. A good example is the foam formation during beating of egg yolk. Another property is the foamability in which, foam is prepared by injecting air or gas through the protein solution. So, foamability and whippability are the two important characteristics which govern the formation of foam into the food materials.

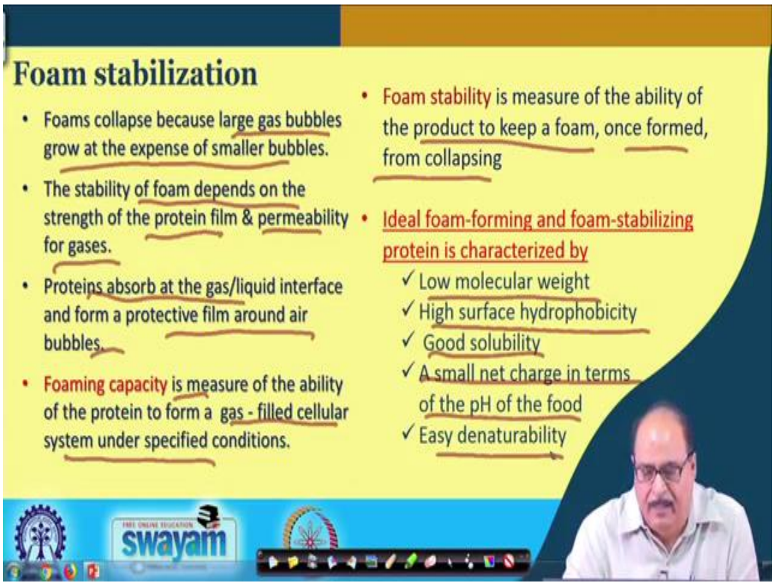


Foaming properties of the food proteins are influenced by source of protein, method and parameters of processing, like temperature, pH, concentration of protein, mixing time, and the method of foaming; whether it is whipping or other method. The main basic functions of protein in foams are to decrease interfacial tension, to increase viscous and elastic properties of the liquid phase, as well as to form strong films.



How the foam formation takes place? Generally the soluble globular proteins diffuse to the air water interface, they concentrate and reduce the surface tension. Proteins unfold at the interface, and hydrophilic as well as hydrophobic groups reorient. Even the polypeptides with possible partial denaturation form a stabilizing film around the bubbles which promote the foam formation.

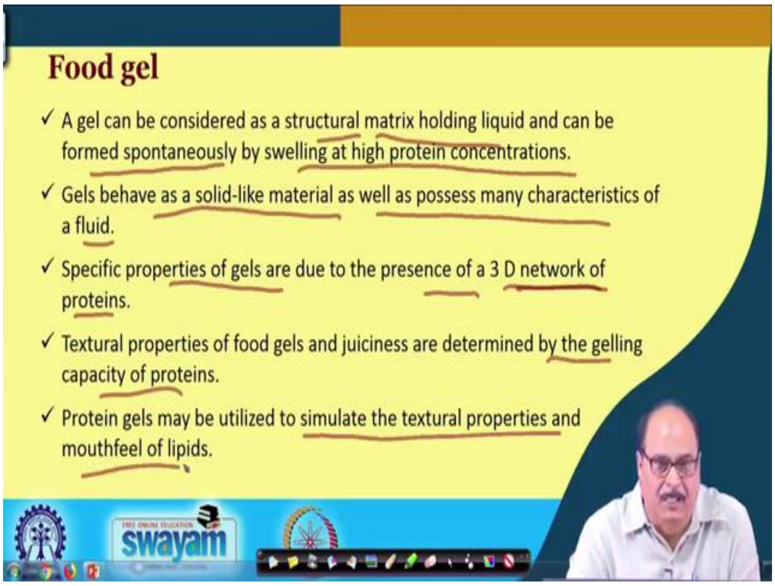
Protein foamability is correlated with its capacity to decrease surface tension at the air-liquid interface. The continuous film is formed through the protein-protein interactions as well as through electrostatic and hydrostatic interaction, or even hydrogen bond formations also influence to some extent the film formation characteristics of a protein.



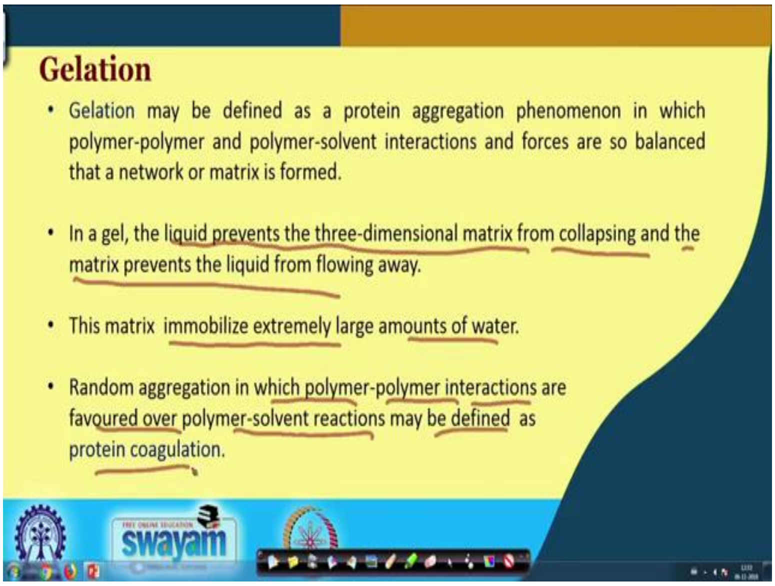
The foams once formed it is desired that it should be stable, and should not collapse. An important process in the foam mat drying, where by foaming process, the surface area is increased and the liquid foods are put into the dryer in a thin sheet of the foam which reduces the drying time. But the important consideration or characteristic of the foam should be that it should not collapse during the drying process. So, the stability of foam is a very important characteristic.

Stability is a measure of the ability of the product to keep a foam once formed and prevent it from collapsing. And, there might be various reasons for the collapse of the foam. It may be because the large gas bubbles grow at the expense of smaller bubbles. The stability of the foam may also depend on the strength of the protein film and permeability of gases; i.e. how this film is permeable or impermeable to different gases. Even proteins absorbed at the gas liquid interface form a protective film around the bubbles. Foaming capacity is a measure of the ability of the protein to form a gas filled cellular system under specified conditions. Foaming capacity and stability are important characteristics of foam forming system.

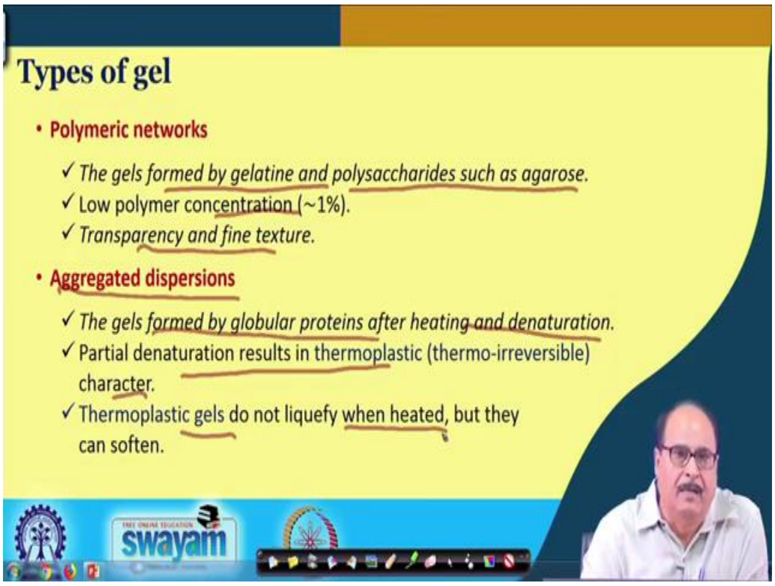
Ideal foam forming and foam stabilizing proteins are characterized by low molecular weight, high surface hydrophobicity, good solubility, small net charge in terms of pH of the food, and easy denaturability. If a protein is denatured easily, it may be a good source for providing a good foaming ability, as well as a good stable foam formation.



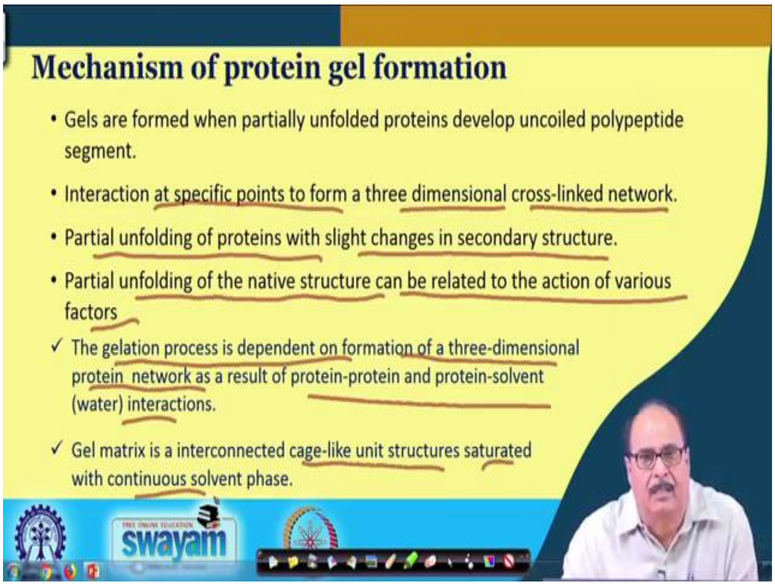
A food gel can be considered as the structural matrix holding liquid and can be formed spontaneously by swelling at a high protein concentration. Gels behave as a solid like material and they possess many characteristics of a fluid. Specific properties of the gel are due to the presence of 3D networks of proteins. Textural properties of food gels and juiciness are determined by the gelling capacity of the proteins. Protein gels may be utilized to simulate the textural properties and mouthfeel of the lipids in the food system.



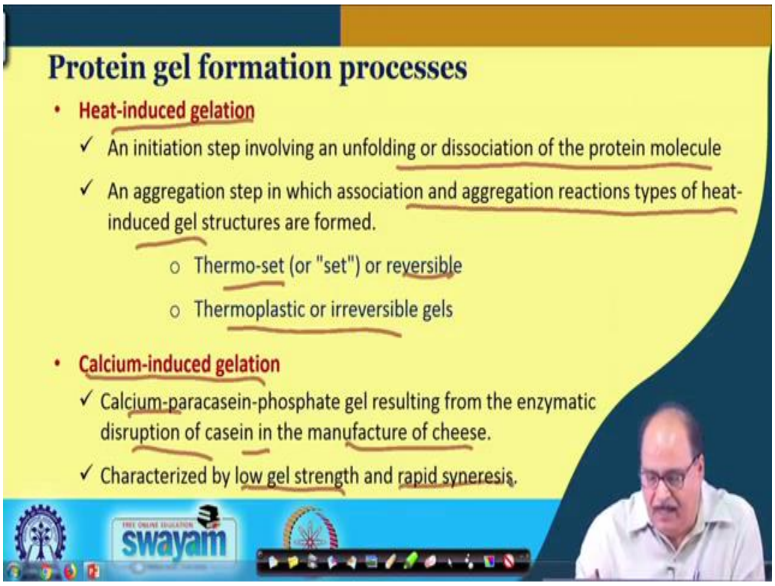
How gelation takes place? Gelation is defined as protein aggregation phenomenon in which polymer-polymer and polymer-solvent interactions and forces are so balanced that a network of matrix is formed. In a gel, the liquid prevents the 3-dimensional matrix from collapsing and the gel matrix prevents the liquid from flowing away. This matrix immobilizes extremely large amounts of water. The random aggregation in which polymer-polymer interactions are favoured over polymer solvent interaction may be defined as a protein coagulation.



There are two types of gel: polymeric networks or aggregated dispersion. Polymeric networks are the gel formed by gelatine and other polysaccharides as agarose. They are normally lower polymer concentrations; transparency and texture are generally better in this type of gels. On the other hand, aggregated dispersions are the gel formed by globular proteins after heating and denaturation. Partial denaturation results in thermoplastic character of the gel; these thermoplastic gels do not liquefy when heated but they can soften.

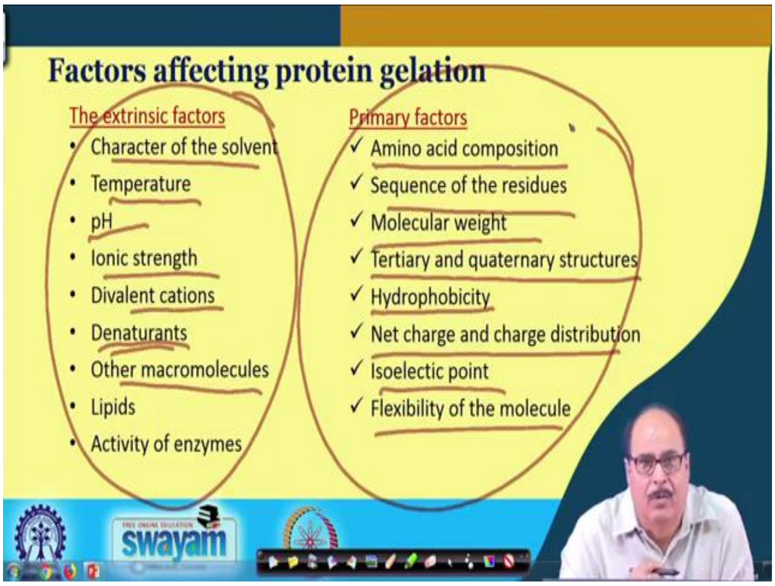


These gels are formed when partially unfolded proteins develop uncoiled polypeptide segment. Interactions at specific point to form a 3-dimensional cross linked network also result in the formation of gel. Partial unfolding of protein with slight change in secondary structure, partial unfolding of the native structure can also be related to the action of various factors. The gelation process is dependent on the formation of a three-dimensional protein network as a result of protein-protein, protein-solvent interactions. Gel matrix is an interconnected cage like unit structure saturated with continuous solvent like phase. So, these are the processes or mechanism of gel formation in various food systems.



The protein gel formation may be of two types. It may be heat induced gel or calcium induced gelation. In the heat induced gelation, an initial step involving the unfolding or the dissociation of the protein molecules takes place and an aggregation step in which association and aggregation reactions type of heat induced gel structures are formed. It may be thermo set or reversible, or thermoplastic or irreversible gels.

The calcium induced gelation is easily seen in the food. When the milk proteins are coagulated, i.e. calcium paracasein-phosphate gels are resulted from the enzymatic disruption of casein in the manufacture of cheese. They are characterized by low gel strength and rapid syneresis.



Different factors that influence the protein gelation processes include the extrinsic factors of the protein. Like, character of the solvent, temperature to which the proteins are exposed, pH, ionic strength, divalent cations, and even presence or absence of the denaturants, other macromolecules, lipids, enzymes, etc. So, these are some of the factors which are considered extrinsic factors and which influence the gel forming ability of the proteins.

Other factors, say primary factors, include the composition of the amino acids; it may also influence the acidic and basic nature of the protein. So that also might influence the gelation characteristics. Sequence of the amino acid residues, molecular weight, tertiary and quaternary structure of a protein, its hydrophobicity or the net charge and charge distribution on the protein, its isoelectric point, flexibility of the protein molecules etc. are the other important factors which influence gelation of a food protein.

A brief overview that how different processing conditions to which food proteins either in their native form or isolated/precipitated forms are exposed has been provided. The changes in their functional characteristics, their behaviors, denaturation, gel formation, foam formation, their usability in various food formulations, food preparations etc. are explained.