REVIEW
OF
LITERATURE
The literature is made pertaining to the work reported on the vegetable proteins, particularly oil seed proteins, their preparation, utilization and their relation with cardiovascular disease. The review also includes the preparation of protein hydrolysates, short peptides and their nutritional significance with reference to coronary heart disease.

Proteins are an essential component of the diet which is needed for survival of animals and humans. Its basic function in nutrition is to supply adequate amount of different amino acids. The protein quality is evaluated on the basis of its amino acid content and on the physiological utilization of specific amino acids after digestion, absorption and rates of oxidation. Metabolism of amino acids is determined by the proportion of amino acids used for protein synthesis. Rates of oxidation of amino acids are low until the amount consumed exceeds the amount needed for protein synthesis; oxidation then increases rapidly. The availability of amino acids is dependent on several factors. It varies with protein source, processing treatment, and interaction with other components of the diet.

Dietary protein falls into two categories defined by origin: animal protein, primarily meat and dairy; and vegetable protein, primarily grains
and legumes. The protein from vegetable origin is an alternative to animal protein for food and cosmetic applications, due to renewability of raw material and variety of sources (especially legumes, cereals, and oil seeds). World oil seed production reached 390 million metric tones in 2007/2008 and world protein meal production brought about 212 million metric tones. Soybean, rapeseed, cottonseed, sunflower, peanut etc are the most abundant protein meals. Beside these major oil seeds, sesame, mustard, rice bran are also contributing a significant level in the production of oil and oil meals particularly in India.

As population growth continues to increase and as the main sources of food may be approaching maximum per capita output, demand seems to outpace food production. A large segment of population will be affected by malnutrition and starvation due to inability to earn the means to buy food (Sen, 1993). The problem of obtaining sufficient protein is further complicated because the mean daily protein requirement for young adult males of 0.6 g/kg of highly digestible good quality protein should be increased to 0.8 g/kg. (Zello et al. 1992).

The incidence of several chronic diseases in Asian countries is lower in comparison with United States and other western nations. Asian diets are tending to be lower in animal protein and higher in protein from plant
sources. In fact Asians typically consume 20-50 times more plant foods than do Americans (Kim et al. 1998). Epidemiological study conducted by Campbell (1990) also support these observations that a diet rich in protein, particularly animal protein may have the greatest potential for enhancing risk for chronic degenerative diseases such as heart disease and cancer. The study of cardiovascular disease (CVD) is complicated by the fact that it has no one single cause. The reduction of elevated blood LDL cholesterol levels is given the highest priority in the prevention of cardiovascular disease. The association between the type of dietary protein consumed and the development of CVD is not new information. The role of plant proteins to reduce CVD risk was studied by several workers (Van Raaji et al. 1981; Sirtori et al. 1985; Nilausen et al. 1998).

The functionality of oilseed proteins in relation to food applications has been reviewed by different authors, the emphasis of their works being focused on different industrial crops (cereals, oilseeds, legume seeds) (Gueguen et al. 1999; Lampart-Szczapa 2001; Sosulski 1977) soybean (Barraquio et al. 1988; Johnson 1970; Kinsella 1976; Kinsella et al. 1985), sunflower (Sosulski et al. 1977), peanut (El-Zalaki et al. 1995), and legumes (Braudo et al. 2001; Singh 2001). The composition, classification and hydration related properties of beans were reviewed by Sathe (2002).
The gelation and interfacial properties of vegetable proteins were reviewed by Van Vliet et al. (2002). The importance of the protein structure in relation to the functional properties (Schwenke 2001) and interaction with polysaccharides in legumes (Braudo et al. 2001) has been reported. Koumanov et al. (2001) reviewed the electrostatic interactions of proteins in relation to their functional properties. The effect of protein modification on the functionality of proteinic products and the potential of knowing protein structure modifications resulting from controlled conditions was addressed by Kinsella (1979) and by Blenford (1994).

Until recently, there has been little work on the effect of dietary protein on plasma lipoprotein patterns. Target dietary recommendations as a percentage of energy has changed little over the years (Report on the national cholesterol education programme expert panel 1988; Second report on the national cholesterol education programme expert panel 1993; Executive summary of the third report on the national cholesterol education programme expert panel 2001; Anonymus 1972; Krauss 2000).

There has been considerable interest in recent years about the absolute level of protein on plasma lipoprotein patterns; it has not been possible to untangle the independent effect of the presence of protein from that of the
absence of carbohydrate and or a change in body weight. During the past
decade, attention has been focused on the potential unique properties of one
type of vegetable protein, soy protein, on plasma lipoprotein patterns
(Anderson et al. 1995). Some work suggests that the initial beneficial
effects on plasma lipoprotein concentrations relative to other types of
protein, most commonly casein, have been more modest than originally
thought (Sacks et al. 2006; Kreijkamp-Kaspers et al. 2004; Lichtenstein
et al. 2002).

Work done in India and elsewhere on the upgradation of oilseed meals for edible uses has proved beyond doubt the importance of oilseed protein concentrates in supplementation of diets to improve food quality (Parpia 1988). Legumes are an excellent source of protein containing almost double the quantity found in common cereals such as wheat and rice. Legume proteins are much cheaper than animal protein (Swindale 1998). At present, only soybean protein (containing 55-60% protein) is mainly utilized in food industries for direct human consumption. The quality of soy protein is next to that of animal protein and better than that of cereal and pulses. Soy-based food products are suitable for diabetes, as they contain less carbohydrates and high protein and oil. It is also good to the patients allergic to animal protein or milk (Kale 1930). Purkrtova et al. (2008) studied the
structure and function of seed lipid-body-associated proteins. Many organisms among the different kingdoms store reserve lipids in discrete subcellular organelles called lipid bodies. In plants, lipid bodies can be found in seeds but also in fruits (olives), and in leaves (plastoglobules). These organelles protect plant lipid reserves against oxidation and hydrolysis until seed germination and seedling establishment. They can be stabilized by specific structural proteins. They presented an overview of the data on the structure of these proteins, which are scarce, and sometimes contradictory and on their functional roles.

Sesame (Sesamum indicum L.), which is an important oilseed crop (Carter et al. 1961), grown extensively in tropical and subtropical areas of the world. The world production of sesame seed was approximately 2 million metric tonnes and that of sesame oil was 850,000 MT. About 35% of the world sesame area and 27% of its production was recorded in India (FAO 1992). Sesame seed was cultivated as a Kharif crop (June-October/November) in western and southern states and that as a Rabi crop (November-February/March) in the eastern region. Different varieties of sesame seed (white, brown, red and black) are cultivated in India. Among them, white seed is much preferred in export market due to its higher quality than black seed and it is primarily grown in the western states of India.
(Weiss 1983). The importance of sesame seed is continuously increasing due to high yield and quality of both oil and meal (Lyon 1972).

The remarkable stability of unrefined sesame oil is due to the presence of endogenous phenolic antioxidants like sesamin, sesamolin and sesamol collectively known as sesame lignans. Sesame lignans are known to have multiple physiological functions like antioxidant activity (Rivas et al. 1981; Johnson et al. 1979), anticarcinogenicity (Johnson et al. 1979) and antihypertensive effects (Salunkhe et al. 1971). Latest research on lipid metabolism and nutrigenomics study reveals that the impact of sesame lignans on gene expression profiles and fatty acid oxidation in rat liver (Ide et al. 2009). The impact of sesamin, episesamin and sesamolin (sesame lignans) on hepatic gene expression profiles was compared with a DNA microarray. This study also confirmed that dietary sesame seed affected the expression of genes related to fatty acid oxidation in a manner similar to isolated lignan compounds. Utilisation of sesame seed meal in broiler chicken diets is also studied by Rama Rao et al. (2008).

Several physical and biochemical properties of the sesame seeds have been reported by Yamashita et al. 1992. It has also been reported that sesame plants have an unusual capacity for accumulation of trace metal like
lead (Pb), the level of which may be 0.13-0.22 mg/100 gm (Yannai et al. 1973).

The use of sesame flour and meal in fortifying some feeds has been reported by several workers (Parpia 1966; Rooney et al. 1972). The use of sesame meal in the diet of children suffering from kwashiorkor has been found to be beneficial, because of its high protein content. This is mainly due to high content of sulphur containing amino acids, especially methionine (Block et al. 1957; Evans et al. 1967). They have also prepared a snack food product of high nutritive value by using 70% chickpea and 30% sesame flours.

Sesame seed have been developed by the Indian Council of Medical Research for use in rural areas for production of a number of ready to use infant foods (ICMR 1977). Micro-atomized protein foods suitable for feeding un-weaned babies from a mixture of groundnut, soy and sesame flours in the ratio of 4:4:3. When reconstituted in boiling water and sweetened with sugar, a free flowing emulsion suitable for use in feeding bottles was produced. There is an increasing interest in fortification of bread and cookies by replacing a portion of wheat flour with non-wheat flours, especially protein concentrates, isolates and oil seed meals. High protein
biscuits are prepared by mixing wheat flour with roasted Bengal gram and roasted sesame flour (ICMR 1977).

Defatted sesame meal, rich in protein (40-50%), is composed of globulin (67.3%), albumin (8.6%), prolamin (1.3 %) and glutelin (6.9%) (Rivas et al. 1981). It may be an excellent protein source for supplementing soybean, peanut and other vegetable proteins that lack sufficient sulphur-containing amino acids, mainly methionine (Johnson et al. 1979; Daghir et al. 1967).

The major protein of sesame flour, being globulins are salt soluble and about 84% of the total meal protein can be solublised by extraction with successive water and salt solutions (Kinsella et al. 1985). Rivas et al. (1981) had tried to extract protein from oil-expelled cakes of sesame seed and also from sesame flour. The nitrogen extractability of sesame flour in water is greatest using a flour : solvent ratio of 1:40 and an extraction time of 15 min. Sesame flour proteins are mainly salt soluble, alkali isolate proteins mainly water soluble (41%) and alkali soluble (41%), and salt isolate proteins mainly alkali soluble (35%). They also observed poor nitrogen extractability from oil-expelled cakes. This is primarily due to denaturation of proteins by high temperature processing.
Guerra et al. (1975) have observed that maximum extraction of sesame protein is obtained above pH-9.0 at solvent (water) meal ratios of 15:1. They also found that the solubility of protein increases with the increase in concentration of NaCl or CaCl₂ up to 1M. However, sesame protein can be maximally extracted at pH-6 if the ionic strength of the solutions increased to 1 M with sodium or calcium chloride (Rivas et al. 1981; Guerra et al. 1975).

In plant species, the most abundant proteins are globulins with molecular masses ranging from 320 to 450 KDa. They are assembled in structures composed of six subunit pairs that interact noncovalently and are known by their sedimentation coefficients as 11S globulins. This is the case of sesame, whose major protein is the insoluble 11S globulin that amounts from 60-70% of the total seed proteins (Casey et al. 1986). The remaining proteins are composed mainly of soluble fractions such as 2S albumin (Tai et al. 1999). Sesame has been a useful crop since ancient times due to its palatable oil. Its seed contain approximately 50% oil and defatted flour has high protein content (45-50%). Protein additives are of great importance due to their functional properties, among which their foaming and emulsifying capacity, gel formation, viscosity, texture and the ability to bind fat or water are the most relevant in food product development. Lopez et al. (2003)
studied the functional properties and potential use of sesame protein isolate from sesame seed flour as protein source in a liquid nutritional supplement. It was compared to a commercial soybean isolate and results showed that its emulsifying properties are better than those of its soybean counterpart. The sensory tests indicated that sesame isolate formulation was superior to the experimental soybean formulation and to the commercially available casein based product.

Khalid et al. (2003) investigated the effect of pH and salt concentration on solubility and functional properties of sesame protein isolate. The minimum protein solubility was at pH 5.0 and maximum was at 3.0. The emulsifying capacity, activity and emulsion stability as well as foaming capacity and foam stability were greatly affected by pH levels and salt concentrations. Lower values were observed at acidic pH and high salt concentration. Its water holding and fat holding capacities, bulk density and other properties were remarkable for application in food formulation systems.

The protein efficiency ratio (PER) of sesame seed, meal and isolated protein are 1.86, 1.35 and 1.2 respectively (Narasinga et al. 1985). Supplementation of sesame seed protein with lysine can increase it’s PER to
2.9. Sesame seed is almost free of antinutritional factors and is suitable for human consumption as such or after processing.

Prakash et al. (1977) observed the association and denaturation of α-globulin sub-units in acid or alkaline conditions. Impurities in the flour might also decrease the solubility of the protein. Sesame seed contains around 5% phytate that is known to form insoluble complexes with seed proteins. Addition of NaCl or NaOH might dissociate sesame phytate complexes. The purified protein would then demonstrate different characteristics in the same solvents.

Dehulling of sesame seed is necessary as hull contains a very high amount of oxalic acid (2-3%), which could complex with calcium and reduces its mineral bioavailability (Kinsella et al. 1985). O’Dell and de Boland (1976) have investigated the relationship between phytate and protein in sesame seed. They have extracted phytate from the sesame meal by dilute hydrochloric acid (0.3 M) and precipitated it with sodium hydroxide. Dehulling improves the nutritional and flavouring characteristics of the meals and leads to the production of a glossy white product (Johnson et al. 1979; Inyang et al. 1996), it also improves enzymatic digestibility of the protein.
Hull also contains indigestible fiber, which impairs digestibility of the protein and imparts dark colour to the meal. Dehulled and defatted meal is rich in protein (about 60%) and does not contain undesirable components (Salunkhe et al. 1971) and hence it may be an excellent protein source for supplementing soybean, peanut and other vegetable proteins, which lack sufficient methionine.

Several dehulling methods have been reported by various investigators (Kinsella et al. 1985; Abou EL-Khier et al. 1987). Wet processing methods including lye treatment process and mechanical treatments have been tried for dehulling (Ramachandra et al. 1970). Non-aqueous methods, such as sieving or air classification have also been utilized to remove hulls after flaking and oil extraction. To increase their nutritive qualities, the undesirable components such as fiber, soluble sugar and oxalates can be eliminated to a large extent by producing protein isolates or concentrates from dehulled and defatted meal (O’ Dell et al. 1976).

In recent years the availability of industrial proteases, mainly from bacteria and fungi, has enabled production of protein hydrolysates in large scale. Most of the hydrolysates are obtained from milk, soybean proteins, although other sources such as meat, eggs, cereals and plant proteins have been used (Alder-Nissen 1986). Villanueva et al. (1999) have prepared
protein hydrolysate from sunflower protein isolate using an endopeptidase (Alcalase) and exopeptidase (Flavourzyme) or both enzymes sequentially. Combined use of these proteases generated the highest degree of hydrolysis, 54.2% and highest solubility around 90%, between pH 2.5 and 7. Vioque et al. (1999) have also produced protein hydrolysates from rapeseed protein isolate by using sequentially an endopeptidase (Alcalase) and exopeptidase (flavozyme). Some of these fractions enriched in certain amino acids could be used for supplementation or treatment of determined clinical syndromes.

The peptides that are produced by partial hydrolysis of proteins have smaller molecular size and less secondary structure than the original proteins. The protein solubility, emulsifying properties and foaming capacities can be improved with a limited degree of hydrolysis (Villanueva et al. 1999; Chobert et al. 1988; Kim et al. 1990), whereas excessive hydrolysis often causes loss of some of these functionalities (Kuehler et al. 1974). They also studied the enzymatic hydrolysis of recovered protein from frozen small croaker and functional properties of the hydrolysates (Yeung et al. 2009). The application of enzyme technology to convert fish processing wastes or under utilized species into protein concentrates for food ingredients has become a considerable interest. Hydrolysis can be used to improve or modify the physicochemical, functional, and/or sensory
properties of native proteins without losing nutritional value (Kristinsson et al. 2000a).

Das et al. (2009) prepared sesame protein isolates and hydrolysates with papain and further fractionated by using ultrafiltration membrane of rotating disk module. They observed the improved functional properties of the protein hydrolysates produced by membrane process.

Enzymatic hydrolysis of proteins, especially partial hydrolysis, is often employed to improve the functionalities of food proteins (Althouse et al. 1995; Kristinsson et al. 2000b; Kuipers et al. 2005). The functionalities of food proteins vary according to the protein source, the proteases used, the degree of hydrolysis (Kristinsson et al. 2000a), the pH and ionic strength of buffer (Doucet et al. 2003), and heat treatment (Feng et al. 2003). Protein hydrolysate characteristics that are often related to emulsion properties are the degree of hydrolysis and the apparent molecular weight distribution (Van der Ven et al. 2001). The solubility is an important measure for liquid foods, while emulsion and water adsorption is important for semisolid foods. To widen the application of protein hydrolysate in various foods, the hydrolysates must be fractionated into soluble and insoluble parts after the process of enzymatic hydrolysis before determining their functionalities.
Most published studies cover the biochemical properties of the hydrolysate itself without fractionation.

Lv et al. (2008) have studied the effect of soluble soybean protein hydrolysate-calcium complexes on calcium uptake by Caco-2 cells. Soybean protein hydrolysates (SPHs) bind with calcium, forming soluble SPH-calcium complexes via the carboxyl groups of glutamic and aspartic acid residues. However, their effect on calcium uptake is still unclear. In this study, Caco-2 cells were used to estimate the effect of SPH-calcium complexes with different molecular weights on calcium uptake in vitro. Their results indicate that soybean protein itself might be responsible for promoting calcium absorption.

Mustard (Brassica juncea, L.), one of the most important oil seed crops, is widely grown in the northern parts of the country and has shown promise both under normal as well as late sown conditions. Mustard is the second largest oil seed crop in India. Mustard seed meal is rich in protein (about 45-55%) with balanced amino acid composition, which is comparable with milk protein casein (Sen et al. 2000). Mustard meal should serve as a suitable raw material in the manufacture of protein ingredients for the food and non-food industrial sectors.
Das et al. (2009) developed ultrafiltration/diafiltration based technique for simultaneous preparation of protein isolate and recovery of phenolic rich fraction from mustard seed meal with a high shear rotating disc membrane module with emphasis on reduced membrane fouling effect. They prepared a high purity of protein isolate (96%) with improved functional properties like emulsion activity index, emulsion stability index, solubility etc. The phenolic content in the protein was found to be reduced from 7.23 mg/g of protein to as low as 0.6 mg/g of protein in the above process. Finally they concluded that industrialization of the process will open up a potential route of seed meal utilization.

Aluko et al. (2005) examined the functional and electrophoretic properties of various mustard seed protein products and the relationship between polypeptide composition and functional characteristics. They studied five varieties of mustard seeds for polypeptide composition and functional properties. The emulsifying activity indices (EAI) of meals and protein concentrates from Brassica seeds were significantly higher than those obtained for similar products from S. alba seeds. It was concluded that the disulfide bonded 50- and 135-kDa polypeptides may have contributed to increased rigidity of S. alba meal proteins, which resulted in poor EAI
when compared to the *Brassica* meals, which do not contain these polypeptides.

Hydrolysis of glucosinolates, present in mustard seed can be prevented by very fast deactivation of myrosinase in the first stage of seed processing, which will result in extracted meal of superior quality. Removing seed hulls also improves the quality of the meal and several methods have been developed in this respect *(Schneider 1983; Kozlowska et al. 1984)*. The fiber content in the defatted meal should be eliminated to enrich protein in the final product.

A number of methods were developed in the seventies to obtain protein preparation from seeds. Most frequently proteins were concentrated by extracting undesirable and other soluble substances with water, using whole or ground seed *(Kozłowska et al. 1972; Ohlson et al. 1979; Eapen et al. 1969; Bhattay et al. 1972)*. Flavours and other soluble substances are removable best with polar solvents especially by alcohol solution *(Berot et al. 1983)*. Defatted rapeseed is used to produce protein isolates of good quality by various workers *(Vioque et al. 2000; Vioque et al. 1999)*. Several workers have examined the biorenewable solvent process for recovery of oil and mustard seed protein concentrates and later the concentrates were
evaluated in terms of their nutritional quality vis-à-vis casein in rats as the experimental animal (Sen et al. 2000; Sen et al. 2003).

The undesired components of sesame such as fibre, soluble sugar, phytates, oxalates and antinutritional components of mustard such as glucosinolates, phenolics and fibre can be eliminated to a large extent by producing protein isolates and hydrolysates (Inyang et al. 1996, Bandyopadhyay et al. 2002). Influence of the rapeseed protein hydrolysis process on CHO cell growth has already been studied by Chabanon et al. (2008). Their results showed that the positive influence of the rapeseed hydrolysates on cell growth was not only due to a nutritional support tied to the addition of small peptides but may be related to the presence of peptides exhibiting growth or survival factor effects. Furthermore, total substitution of proteins (transferrin, albumin and insulin) in the cell culture medium by some rapeseed hydrolysates appeared to be a promising alternative to improve the cell growth in protein-free media.

Diet content of proteins (Beynen et al. 1983) is important for atherosclerosis development and diet manipulations can retard or accelerate the progression of this pathology. Soy protein has been extensively studied and reported to be an effective weight reducer (Aoyama et al. 2000) and has been found to have hypocholesterolemic effect compared with animal
proteins such as casein (Damasceno et al. 2001). It markedly decreases the activity of hepatic lipogenic enzymes (Iritani et al. 1986). Soy protein in relation to casein tends to reduce the metabolism of linoleic and arachidonic acid and hence the production of several eicosanoids (Koba et al. 1994). Soy protein isolate, in comparison with casein, induces a low postprandial insulin/glucagons ratio (Sanchez et al., 1991) and decreases the lipid peroxide, cholesterol and triglyceride content of atherogenic lipoproteins, which has beneficial effects over atherosclerosis progression in cholesterol-fed rabbits (Damasceno et al. 2001).

Most researchers have worked mainly on soy bean protein because of its high protein content and its unique ability to reduce blood cholesterol and to favorably affect other biological parameters. A great deal of work has been done on animals as well as direct human trials (Kurowska et al. 1994). Animal studies (Wong et al. 1998) indicated that certain amino acids especially lysine increases blood cholesterol level while arginine counteracts this effect. Therefore, the ratio of arginine to lysine in the diet is crucial in explaining elevated blood cholesterol level and the resulting atherosclerosis. The theory is supported by the fact that soy protein provides more favourable arginine to lysine ratio than casein. David Kritchevsky have also observed that animal protein (usually casein) is more cholesterolemic and
atherogenic than vegetable protein (Kritchevsky 1979; Kritchevsky 1993; Campbell 1990).

Rajamohan and Kurup (1997) have already studied the effect of sesame protein globulin fraction containing lysine to arginine ratio 0.67 on cholesterol metabolism in rats. Sen and Bhattacharyya (2001) have also observed the cholesterol lowering effect of sesame protein fraction extracted with isopropanol.

Attempts have been made to explain the mechanism by which the vegetable proteins exert hypocholesterolemic effect in experimental animals. Soybean protein compared with casein with or without dietary cholesterol (Choi et al. 1989) lowers plasma cholesterol and triacylglycerol concentrations in rats. Several studies have suggested that the hypocholesterolemic effect of vegetable protein, particularly soy protein is largely attributed to higher fecal steroid excretion as a consequence of reduction in intestinal absorption. Iwami et al. (1986) reported that soy protein isolate was inferior to casein in digestibility and suggested that the hydrophobic properties of soy protein that remain after digestion bind well to bile acids and serves as a cholesterol lowering factor.

Huff and Carrol (1980) reported that a cholesterol free purified diet (soy protein) increased fecal excretion of bile acids relative to casein when
fed in rabbits. These observations are also supported by a number of studies using rats fed a cholesterol free purified diet, (Beynen et al. 1986, Nagata et al. 1982) and is considered to be one of the main mechanisms of hypocholesterolemic activity of soybean protein in rats as well as rabbits.

Studies also showed that an amino acid mixture simulating soy protein demonstrated hypocholesterolemic effects without influencing fecal bile acid excretion in rats (Nagata et al. 1981) and rabbits (Huff et al. 1977) compared with an amino acid mixture simulating casein. On the basis of the studies of the metabolites of methionine, Sugiyama et al. (1986) reported that the methyl group of methionine is responsible for the cholesterol elevating effect of methionine in rats. In contrast, Moundres et al. (1995) reported that when rats were fed soybean protein at suboptimal level (13%), serum cholesterol concentration was significantly higher than that in rats fed a 13% casein diet, and this higher cholesterol level was counterattacted by supplementation of diet with 0.4% of methionine.

Morita et al. (1997) examined the role of methionine in hypocholesterolemic effects of some dietary proteins in rats fed cholesterol free purified diets with different levels of methionine. They concluded that methionine contents in the dietary proteins would be more responsible for cholesterol lowering effect of soy protein, rice protein and potato protein.
rather than the fecal steroid excretion. Sugiyama et al. (1993) clearly showed that supplementation of 2% glycine to a 250 g casein/kg diet significantly reduced serum cholesterol concentrations compared with un-supplemented diet, indicating that dietary glycine is hypocholesterolemic. Finally, they concluded that the mechanism by which the methionine:glycine ratio could regulate serum cholesterol is still unclear.

Non-protein components (such as fibre, phytic acids, minerals and isoflavones) associated with protein may also affect cholesterol metabolism (Potter 1995). Soy protein may stimulate hepatic activities of hydroxy methyl glutaryl CoA (HMG-CoA) reductase, the rate limiting enzyme in the biosynthesis of cholesterol (Nagata et al. 1982) and cholesterol 7α-hydroxylase, the key enzyme that converts cholesterol to bile acids (Beynen 1990). Dietary protein can also modify essential fatty acid metabolism (Huang et al. 1993). Soy protein compared with casein reduces Δ6 (n-6) desaturase activity in liver microsomes (Koba et al. 1993) in rats. This is the enzyme responsible for polyunsaturated fatty acid biosynthesis. Similarly, the lower 20:4(n-6)/18:2(n-6) ratio, the linoleate desaturation index, that is observed in liver microsomes of rats fed soy protein compared with casein, has also been attributed to reduced Δ6(n-6) desaturase activity (Choi et al. 1989).
Madani et al. (1998) investigated the effects of highly purified soy protein (98%) vs casein with or without addition of cholesterol (0.1%) in plasma and liver cholesterol concentration, fecal steroid excretion and liver enzymes involved in cholesterol metabolism and fatty acid biosynthesis. Cholesterol supplementation (0.1%) did not affect plasma cholesterol, but increased liver cholesterol and triacylglycerol concentrations and reduced HMG-CoA reductase activity. Cholesterol 7α-hydroxylase activity was diminished only in rats fed casein. Desaturase activities and particularly Δ5(n-6) activity were lowered by cholesterol supplementation in rats fed both protein diets, including a significant lowering of 20:4(n-6)/18:2(n-6) ratio in liver microsomal lipids and liver phospholipids. Finally, they concluded that dietary proteins have no effect on serum cholesterol in rats, they affect enzyme activities involved in cholesterol metabolism and fatty acid desaturation.

Current nutritional research and product development is focused on products that help to reduce or control diet-related chronic diseases such as atherosclerosis, cancer or liver failure as well as special products for hospitalized patients (Weaver et al. 1993). Proteolytic enzyme modification of proteins is an effective way to improve the various functional properties and to increase the field of application of the proteins (Panyam et al. 1996;
Wu et al. 1998; Vioque et al. 2000). Protein hydrolysates are widely used as nutritional supplements, functional ingredients and flavour enhancers in foods, coffee whiteners, cosmetics, personal care products, and confectionary, and in the fortification of soft drinks and juices. Protein hydrolysates are also used in soups, sauces, gravies, snacks, meat products and other savoury applications (Weir et al. 1986; Giese 1994).

Nutritional support in patients in liver disease is one of the most important areas of treatment. Currently, treatment under investigation includes the administration of insulin growth factors, branched-chain amino acids (BCAA), structured lipids and polyunsaturated fatty acids (McCullough et al. 1989). Protein hydrolysates enriched with up to 40-50% BCAA have been recommended for patients (both children and adults) with liver disease, because BCAA may reduce plasma concentration of aromatic amino acids. BCAA are preferentially taken up by muscle and are therefore theoretically available for peripheral metabolism even in advanced liver disease (Fischer 1990) Therefore, preparation of products with high concentration of BCAA, are desirable in diet formulation for patients with liver disease. Bautista et al. (2000) have prepared sunflower protein hydrolysates from globulin fraction-II, which is rich in BCAA about 29.7%. Subsequent purification by ultrafiltration and enzyme treatment
(carboxypeptidase-A), the BCAA concentration has increased to 37.4%. These products are used enteral, parenteral and oral nutrition for the treatment of patients with liver failure.

Cho et al. (2007) studied the mechanism and component of the cholesterol lowering activity of soybean. They proved that peptides in soybean protein hydrolysates (SPH) made by certain proteases have a hypocholesterolemic effect. Among the mechanisms suggested, blockage of bile acids and/or cholesterol absorption, inhibition of cholesterol synthesis and stimulation of low-density lipoprotein receptor (LDL-R) transcription, SPH appeared to stimulate LDL-R transcription. They concluded that the dietary upregulation of LDL-R transcription by soybean may be consequent to an enhanced catabolism or a reduced synthesis of intracellular cholesterol. Finally, they suggested that the soy peptides can effectively stimulate LDL-R transcription in the human liver cell line and reduce blood cholesterol level.

Yang et al. (2008) investigated the effects of soy protein hydrolysate on blood pressure and angiotensin I-converting enzyme activity in rats with chronic renal failure. They suggested that soy protein hydrolysate was shown to be as effective as soy protein in preventing the elevation of blood pressure, the progression of renal failure, and decreases in kidney TNF-α
level, plasma ACE activity, and insulin concentration. So, the beneficial effects of consuming soy protein on blood pressure and renal function may be mediated mostly by its pepsin-digested hydrolysate through its ACE inhibitory activity.

Ikeda et al. (2007) studied the effect of dietary sesame seed and sesamin on the ascorbic acid concentration in rat tissues. The dietary sesame seeds and sesamin elevated ascorbic acid concentrations in the liver and kidney and increased urinary excretion in those Wistar rats. The dietary sesamin also elevated the hepatic mRNA levels of cytochrome P450 (CYP) 2B, uridine diphosphate (UDP)-glucuronosyltransferase (UGT) 1A and 2B. The results suggest that dietary sesame seed and its lignan stimulate ascorbic acid synthesis as a result of the induction of UGT1A and the 2B-mediated metabolism of sesame lignan in rats. The study also suggests that dietary sesame seed enhances antioxidative activity in the tissues by elevating the levels of two antioxidative vitamins, vitamin C and E.

Plant protein hydrolysates are a source of bioactive peptides. There are peptides that decrease the micellar cholesterol solubility from bile acids and therefore may reduce in vivo cholesterol absorption. The presence of these peptides in sunflower protein hydrolysates has been studied by Christina et al. (2009). They have studied the generation of inhibitory
peptides of cholesterol incorporation to micelles, after the hydrolysis of sunflower proteins with bacterial proteases (alcalase and flavourzyme) or digestive enzymes (pepsin and pancreatin). The protein hydrolysates, either with microbial or digestive proteases, represent a source of peptides that inhibits cholesterol incorporation to micelles. This high hydrophobicity probably favours their inclusion in the lipid micelles. In vivo, this inhibition may translate in a decrease of cholesterol absorption. Reported results show that a combination of different characteristics such as peptide size or hydrophobicity may be responsible of the inhibitory activity of generated peptides.

Seed protein isolates and hydrolysates (Vioque et al. 2000; Bautista et al. 2000) of improved functional properties are already very popular in various food items. Two membrane based processes were tested for protein purification from yellow mustard meals (Xu et al. 2003) and also improvement of functional properties, chemical composition and protein characterization by preparation of protein hydrolysate has already been published by Pedroche et al. (2004). Sadeghi et al. (2006) has reduced the anti nutritional factors by preparation of protein isolate by steam injection from mustard meal. The purity of the protein isolate was 95%. The hydrolysed products of glucosinolates like isothiocyanates and oxazolidine
thione levels, phenolics and phytic acid levels were low in protein isolate. The *in vitro* digestibility of the protein isolate was 92.4% compared to the 80.6% in the meal. The calculated nutritional indices, essential amino acid index, biological value, nutritional index and C-PER of protein isolate were higher compared to meal.

In the last two decades, fundamental studies have opened a new field of research related to bioactive food components that not only help in adequate nutrition, but may provide specific health benefits. The term bioactive peptides is used to identify molecules of peptidic nature or origin which display a biological behaviour or activity which can be developed at the industrial level for pharmaceutical, diagnostic, chemical and agro-food applications. In particular, food-derived bioactive proteins and peptides are claimed to be health-enhancing components used to reduce the risk of disease or to enhance a certain physiological function. Activated peptides may exert quite different bioactivities, such as opioid, angiotensin I-converting enzyme (ACE) inhibitory, immunomodulatory, antimicrobial, mineral binding, antimitogenic and cytomodulatory effects.

Ochi *et al.* (1995 and 2002) have isolated three peptides with ACE inhibitory activities, Met-Leu-Pro-Ala-Tyr, Val-Leu-Tyr-Arg-Asp-Gly and Ile-Val-Tyr, from a thermolysin hydrolysate of sesame proteins. Each
peptide also showed an anti hypertensive effect in spontaneously hypertensive rats when orally administrated at the dose of 100 mg/kg. Nakano et al. (2006) were isolated sesame peptide powder (SPP) which exhibited strong ACE inhibitory activity and significantly and temporarily decreased the systolic blood pressure (SBP) in spontaneously hypertensive rats by a single administration (1-10 mg/kg). Repeated oral administration of SPP also lowered both SBP and the aortic ACE activity in spontaneously hypertensive rats. These results demonstrate that SPP would be a beneficial ingredient for preventing and providing therapy against hypertension and its related diseases.

Murray and Fitz Gerald (2007) underline that the biofunctional peptide activity currently most studied in food proteins, appears to be those that inhibit Angiotensin-converting-enzyme (ACE), which plays a central role in the regulation of blood pressure. Parodi (2007) focuses his attention on the influence of certain dietary proteins and peptides on carcinogenesis. There is now ample evidence that whey proteins from bovine milk and their peptides have the potential to inhibit cancer development at some sites, particularly the colon and breast. It is suggested that the anticancer action of whey protein is due to its content of cystine/cysteine and γ-glutamylcystine dipeptides.
Valuable information on the superior functionality of sesame will promote strongly the use of sesame seed in the daily diet worldwide. There are various forms and methods of using sesame seed and oil in Asian countries and people in these countries enjoy many kinds of foods containing sesame products with superb taste and flavour. There is enough scope to utilize sesame protein in various food formulations supplementing soy bean and other proteins. By considering the enormous importance of sesame protein, it will be worth to investigate the effect of seed protein isolates and hydrolysates rich in small peptides on the lipid profiles and lipid peroxidation and their probable correlation with cardiovascular disease.