

POTENTIAL OF BIOACTIVE PROTEINS AND PEPTIDES FOR PREVENTION AND TREATMENT OF MASS NON-COMMUNICABLE DISEASES

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ABSTRACT: Chronic non-communicable conditions are, by far, the leading cause of mortality in the world. They are often linked to dietary factors. A functional food can be defined generally as any food which can provide a health benefit to one or more bodily functions beside that of basic nutrition. Numerous peptides with different functional properties can be found encrypted in the primary structure of many dietary proteins. These peptides can be released during digestion by proteolytic enzymes in the gastrointestinal tract or during fermentation and food processing. Bioactive peptides have physiological effect on the major body systems – the cardiovascular, digestive, endocrine, immune and nervous system. Sources of bioactive proteins and peptides can be divided to animal and plant sources. Cultures rich in proteins, such as cereals, pseudocereals and legumes are potential source of bioactive proteins and peptides with different activities, including antiproliferative, immunomodulatory, antioxidant, antithrombotic, antihypertensive, hypocholesterolemic and opioid activity.

Key words: *bioactive peptides, cancer, cardiovascular diseases, cereals, pseudocereals, legumes*

Abbreviations: Ala = alanine, Arg = arginine, Asn = asparagine, Asp = aspartic acid, Glu = glutamic acid, Gln = glutamine, Gly = glycine, His = histidine, Ile = isoleucine, Leu = leucine, Lys = lysine, Met = methionine, Phe = phenylalanine, Pro = proline, Ser = serine, Thr = threonine, Trp = tryptophan, Tyr = tyrosine, Val = valine, IC₅₀ = half maximal inhibitory concentration.

INTRODUCTION

In recent years, increasing epidemiological evidence is linking the prevalence of mass non-communicable diseases, such as obesity, hypertension, diabetes, hyperlipidemia, and even cancer, to dietary factors. A functional food can be defined generally as any food which can provide a health benefit to one or more bodily functions beside that of basic nutrition (Hernández-Ledesma et al., 2011). Bioactive proteins and peptides are derived from food and

they have physiological, hormone-like effect in human organism. They act in a direct manner, through their presence in the undisturbed food (Hartmann and Meisel, 2007). Bioactive peptides are encrypted in the primary structure of the protein, and they are inactive until released by enzymatic hydrolysis. It can occur during digestion by proteolytic enzymes in the gastrointestinal tract or during fermentation and food processing. Bioactive peptides can exhibit local effects in the gastrointestinal system, or cause systemic effects after intestinal absorption and entering the circulatory system (Erdmann et al., 2008). They range in size from 2 to 50 amino acid residues (Hernández-Ledesma et al., 2011). Bioactive proteins and peptides exhibit different activities, such as antimicrobial, antioxidant, antithrombotic, antihypertensive, hypocholesterolemic, hypoglycemic, immunomodulatory, opioid, and antiproliferative activities. They can affect the condition of major body systems,

and these are cardiovascular, digestive, endocrine, immune and nervous system (Hernández-Ledesma et al, 2011).

Bioactive peptides can be classified on the basis of their origin as animal origin peptides and plant origin peptides. Animal sources include dairy products, egg, meat, insects, fish and seafood (Wang et al., 2005). Plant sources usually include cereals, such as wheat, corn, rice, barley, rye and pseudocereals, such as buckwheat and amaranth. Other plant sources are legumes (soy, pea, and chickpea), *Brassicaceae* species (mustard, rapeseed) and other (sunflower). Among plant sources, soybean is the most studied source of bioactive proteins and peptides. This can be explained by the fact that soybean is an important protein source. On average, soybean contains about 40% protein (Wang et al., 2005). In Western countries, other mentioned cultures are grown more than soy and they should be further studied for their biological activity.

BIOLOGICAL FUNCTIONS OF BIOACTIVE PEPTIDES

Anticancer

Cancer is one of the leading causes of mortality worldwide, and only cardiovascular disease is a more important mortality

factor in the developed world (Maldonado et al., 2010). Carcinogenesis is a process that consists of several steps. Many different factors, both genetic and environmental, affect initiation and promotion of cancer (Ledesma et al., 2009). Chemical carcinogenesis and viral oncogenesis have been found to share common mechanisms involving changes in chromatin status. Histone acetylation and deacetylation are involved in chromatin remodeling, which is important in cell cycle control (Ledesma et al., 2009). Anticarcinogenic peptides have no common mechanism of action, but they eventually lead to the inhibition of mitosis and cell death.

Investigations of anticancer activity of peptides derived from plant proteins are presented in Table 1. It can be seen that most researchers have investigated anticancer activity of lunasin.

Lunasin is firstly discovered in soy (Hernández-Ledesma et al., 2009), and later in some cereals and pseudocereals, like wheat, barley, rye, rice and amaranth. It consists of 43 amino acid residues. The antimitotic activity of lunasin is explained by 8 Asp residues at carboxyl end of its chain which are able to bind to the hypoacetylated regions of chromatin (Hernández-Ledesma et al., 2009).

Table 1.
Examples of anticancer peptides derived from plant proteins

Disease/condition	Biological assay	Encoding protein/protein hydrolysate	Source	References
Cancer	<i>In vitro</i> and <i>in vivo</i>	Lunasin	Wheat	Jeong et al., 2007
Cancer	<i>In vitro</i>	Lunasin	Barley	Jeong et al., 2002
Cancer	<i>In vitro</i> and <i>in vivo</i>	Lunasin	Rye	Jeong et al., 2009
Cancer	<i>In vitro</i>	Lunasin from glutelin fraction	Amaranth	Silva-Sánchez et al., 2008; Maldonado-Cervantes et al., 2010
Cancer	<i>In vitro</i> and <i>in vivo</i>	Lunasin	Soy	Jeong et al., 2003, 2007; De Mejia et al., 2003, 2004; Park et al., 2005; Hernández-Ledesma et al., 2009
Cancer	<i>In vitro</i>	Pentapeptide	Rice	Kannan et al., 2010
Cancer	<i>In vitro</i> and <i>in vivo</i>	Bowman-Birk inhibitor	Soy	Park et al., 2005
Cancer	<i>In vitro</i> and <i>in vivo</i>	Lectins	Soy	De Mejia et al., 2003; Barać et al., 2005

Several *in vitro* and *in vivo* studies have shown its cancer preventive action. In addition, it has been showed that, in the absence of carcinogens, it does not seem that lunasin affects cell morphology and proliferation, but prevents transformation of cells in the presence of carcinogens. *In vitro* investigations have shown that while pure lunasin is easily hydrolyzed in gastrointestinal tract by pepsin and pancreatin, lunasin in soy protein is resistant to the action of these enzymes. Further bio-availability studies, carried out with animals, have confirmed these preliminary results obtained by *in vitro* analysis (Hernández-Ledesma et al., 2009). This is very important property of lunasin, because it implicates that this cancer-preventive agent can be taken orally.

Kannan et al. (2010) isolated and characterized novel anticancer pentapeptide derived from rice bran enzymatic hydrolysate, whose amino acid sequence is Glu-Gln-Arg-Pro-Arg. This peptide is resistant to gastrointestinal juices and possesses cancer growth inhibitory properties on colon, breast, lung and liver cancer cells.

One of the most extensively studied bioactive substances in soy is the Bowman-Birk protease inhibitor (BBI) (Jeong et al., 2003; Park et al., 2005; Hernández-Ledesma et al., 2009). BBI is a serine protease inhibitor that consists of a single chain of 71 amino acid residues. This chain is cross-linked by seven pairs of disulfide bonds. It has the ability to inhibit the action of trypsin and chymotrypsin (Park et al., 2005, Barać et al., 2005). BBIC (BBIC refers to BBI concentrate, crude form) has been shown to be cancer preventive *in vitro* models of carcinogenesis. In preclinical studies, BBIC has been found to interfere with the development of tumors induced by chemical carcinogens in a number of animal model systems (Park et al., 2005).

Lectins are glycoproteins that can selectively bind carbohydrates (Mejia et al., 2003). Lectins show several biochemical, physiological and nutritional effects after ingestion. Some of them are agglutination of red blood cells and stimulation of pancreatic enzyme secretion, resulting in reduced intestinal absorption of nutrients.

There are several reports that plant lectins may have antitumor and anticarcinogenic activities that could be useful in cancer treatment. The exact mechanism of the antitumor effect of plant lectins is not clear, although several mechanisms have been proposed. Some of these mechanisms are reduction of cell division, increasing the number of macrophages, increasing the susceptibility of tumor cells to macrophage attack and serving as a bridge between tumor cells and macrophages (Barać et al., 2005).

Regulation of the immune system

Immunomodulatory peptides can enhance immune cell functions, measured as lymphocyte proliferation, natural killer (NK) cell activity, antibody synthesis and cytokine regulation, reduce allergic reactions and enhance mucosal immunity in the gastrointestinal tract (Hartmann and Meisel, 2007). Activation of NK cells is effective in patients with autoimmune disease or cancer and in elderly people, who usually have low levels of NK cell activity (Horiguchi et al., 2005).

Effect of peptides on the immune system has been the topic of several researches, which are presented in Table 2. It is usually studied among other functionalities, but Dia et al. (2009) reported anti-inflammatory activity of lunasin. It prevents inflammation by inhibiting COX-2/PGE₂ and iNOS/NO pathways. Previous investigations of lunasin were focused on its cancer preventive activity. However, inflammation is a critical factor in tumor progression and cells that suffer inflammation produce various responses that can damage DNA and cause mutations that lead to tumor initiation and/or promotion. Horiguchi et al. (2005) reported that intake of wheat gluten hydrolysate can increase NK (natural killer) cell activity without severe side effects. NK cells play a major role in the rejection of tumors and cells infected by viruses. Oryzatesin, an ileum contracting bioactive peptide obtained from rice albumin, was shown to have immunostimulatory role that was mediated by histamine release. Its amino acid sequence is Gly-Tyr-Pro-Met-Tyr-Pro-Leu-Pro-Arg (Takahashi et al., 1996 in Kannan et al., 2010).

Table 2.

Examples of peptides derived from plant proteins with effect on the immune system

Disease/condition		Biological assay	Encoding protein/protein hydrolysate	Source	References
Immune system	Inflammation	<i>In vitro</i>	Lunasin	Soy	Dia et al., 2009
Immune system	Immunomodulation	<i>In vivo</i>	Wheat gluten hydrolysate	Wheat	Horiguchi et al., 2005
Immune system	Immunomodulation	Database	Glutelin tryptic digest	Amaranth	Silva-Sánchez et al., 2008
Immune system	Immunomodulation	N.A.	Oryzatensin from rice albumin	Rice	Takahashi et al., 1996 in Kannan et al., 2010
Immune system	Immunostimulating	Database	Glutelin tryptic digest	Amaranth	Silva-Sánchez et al., 2008

There were 36 sequences for amaranth seed proteins reported in the database (www.ncbi.nlm.nih.gov). These sequences were tested for all of the 1573 active peptides reported (www.uwm.edu.pl/biochemia) with 39 different activities, including immunomodulating and immune-stimulating activity (Silva-Sánchez et al., 2008).

Regulation of the cardiovascular system and metabolic disorders

Cardiovascular disease (CVD) is the leading cause of death for both males and females in the United States and other developed countries in the world. It usually ranks among the top five causes of death in less developed countries (Erdmann et al., 2008). The most frequent cardiovascular diseases are coronary heart disease, peripheral artery disease and stroke (Hernández-Ledesma et al., 2011). Prevention of cardiovascular diseases often includes improvements of risk factors such as high blood pressure, an unfavorable profile of blood lipids, insulin resistance, obesity, insufficient physical exercise and smoking.

High blood pressure is one of the major risk factors for CVD (Erdmann et al., 2008). Angiotensin I-converting enzyme (ACE, EC 3.4.15.1) is one of the main regulators of blood pressure because it acts on two body systems. Firstly, ACE

forms part of the renin-angiotensin system (RAS), converting angiotensin I to angiotensin II, a potent vasoconstrictor. Angiotensin II also induces the release of aldosterone and therefore increases the concentration of sodium in circulation and blood pressure. Secondly, ACE hydrolyzes bradykinin, which has a vasodilatory effect and forms part of kinin-kallikrein system. Different synthetic ACE inhibitors, such as Captopril and Enalapril, are being used worldwide to treat hypertension (Hernández-Ledesma et al., 2011). The renin-angiotensin system is presented in Figure 1.

Numerous peptides with beneficial effects on the cardiovascular system and related metabolic disorders have been discovered up to date. However, most of protein hydrolysates and identified peptides have hypotensive effect and they are presented in Table 3.

All these peptides have common mechanism of action - inhibition of angiotensin I-converting enzyme. It is supposed that peptides that consist of two or three amino acid residues could be absorbed directly from the gastrointestinal tract into the blood circulation. These intact peptides are able to exert physiological action (Wu et al., 2006). Peptides with Pro or hydroxy-Pro as C-terminus are usually resistant to degradation by digestive enzymes (Erdmann et al., 2008).

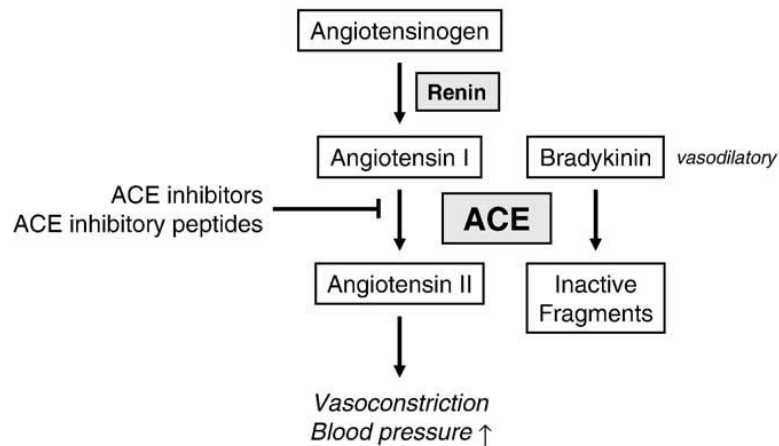


Figure 1. The renin–angiotensin system. (Erdmann et al., 2008, Fig 1, Pg. 644)

Table 3.

Examples of peptides derived from plant proteins with hypotensive effect

Disease/condition		Biological test	Encoding protein/protein hydrolysate	Source	References
Cardiovascular system	Hypertension (ACE inhibitor)	<i>In vitro</i>	Wheat germ protein hydrolysate	Wheat	Matsui et al.; 1999, Jia et al., 2010
Cardiovascular system	Hypertension (ACE inhibitor)	<i>In vitro</i> and <i>in vivo</i>	Gliadin hydrolysate	Wheat	Motoi et al., 2003
Cardiovascular system	Hypertension (ACE inhibitor)	<i>In vitro</i>	α -Zein hydrolysate	Corn	Yano et al., 1996
Cardiovascular system	Hypertension (ACE inhibitor)	Database	Glutelin tryptic digest	Amaranth	Silva-Sánchez et al., 2008
Cardiovascular system	Hypertension (ACE inhibitor)	Database	Peptides from globulin 11S	Amaranth	Silva-Sánchez et al., 2008
Cardiovascular system	Hypertension (ACE inhibitor)	<i>In vitro</i>	Protein hydrolysate	Pea	Humiski and Aluko, 2007; Li and Aluko, 2010
Cardiovascular system	Hypertension (ACE inhibitor)	<i>In vitro</i>	Legumin hydrolysate	Chickpea	Yust et al., 2002
Cardiovascular system	Hypertension (ACE inhibitor)	<i>In vitro</i>	Protein hydrolysate	Mustard	Pedroche et al., 2007
Cardiovascular system	Hypertension (ACE inhibitor)	<i>In vitro</i>	Helianthinin hydrolysate	Sunflower	Megias et al., 2004
Cardiovascular system	Hypertension (ACE inhibitor)	<i>In vitro</i> , prediction	Peptides from glycinin	Soy	Wu et al., 2006
Cardiovascular system	Hypertension (ACE inhibitor)	<i>In vitro</i> , prediction	Peptides from β -conglycinin	Soy	Wu et al., 2006; Chen et al., 1995
Cardiovascular system	Hypertension (ACE inhibitor)	<i>In vitro</i> , prediction	Peptides from legumin	Pea	Wu et al., 2006
Cardiovascular system	Hypertension (ACE inhibitor)	<i>In vitro</i> , prediction	Peptides from albumin	Pea	Wu et al., 2006
Cardiovascular system	Hypertension (ACE inhibitor)	<i>In vitro</i> , prediction	Peptides from vicilin	Pea	Wu et al., 2006
Cardiovascular system	Hypertension (ACE inhibitor)	<i>In vitro</i>	Protein isolate	Rapeseed	Yoshie-Stark et al. 2006, 2008

ACE inhibitory peptides were isolated from wheat germ and wheat gliadin hydrolysates and their sequences were Ile-Val-Tyr and Ile-Ala-Pro, respectively. It can be seen that they both are small peptides with hydrophobic amino acids (Matsui et al., 1999; Motoi et al., 2003). Peptides with hypotensive effect were also isolated from another cereal, corn. α -Zein hydrolysate contained many different small peptides with various potency of ACE inhibitory activity (Yano et al., 1996). Several peptides isolated from amaranth glutelin tryptic digest had the same amino acid sequences as some previously reported ACE inhibitory peptides. Tripeptides Leu-Pro-Pro, Leu-Arg-Pro, and Val-Pro-Pro as well the dipeptide Tyr-Pro have been shown to have ACE inhibitory activity in spontaneously hypertensive rats. Antihypertensive peptides were also found in the globulin 11S sequence. Di- and tripeptides as Gly-Lys-Pro, Leu-Phe, Tyr-Leu, Arg-Phe, and His-Tyr are reported in the literature as angiotensin-I-converting enzyme inhibitors (Silva-Sánchez et al., 2008).

Hydrolysates from legumes, pea and chickpea, also contain peptides with hypotensive effect. Li and Aluko (2010) reported three ACE inhibitory peptides from pea protein hydrolysate, whose sequences were Ile-Arg, Lys-Phe and Glu-Phe. Antihypertensive peptides isolated from chickpea legumin hydrolysate consist mostly of Met, Asp, Phe and Leu (Yust et al., 2002).

Other industrial cultures were also investigated for peptides with ACE inhibitory activity. Fractions obtained from *Brassica carinata* (Ethiopian mustard) protein hydrolysate showed ACE inhibitory activity (Pedroche et al., 2007). Megias et al. (2004) obtained an ACE inhibitory peptide with the sequence Phe-Val-Asn-Pro-Gln-Ala-Gly-Ser from helianthinin, the 11S globulin from sunflower seeds, which is the main storage protein in sunflower.

Some of bioactive peptides have not been discovered yet, but there are indications that they could be obtained by hydrolysis of natural proteins. Wu et al. (2006) constructed from published literature a database consisting of 168 dipeptides and 140

tripeptides to study the quantitative structure-activity relationships of angiotensin I-converting enzyme (ACE) inhibitory peptides. Three predicted dipeptides and four predicted tripeptides were located within the primary structure of food proteins. Then they were synthesized for validation of their IC₅₀ values through laboratory determination of inhibition of ACE activity. Three peptides determined in their study were more potent than the well-known milk tripeptides of Val-Pro-Pro and Ile-Pro-Pro. Their sequences were Leu-Arg-Trp, Ile-Lys-Pro and Phe-Trp. Leu-Arg-Trp and Phe-Trp were located in pea protein sequence, and Ile-Lys-Pro was located in soybean and pea proteins.

Yoshie-Stark et al. (2008) found that protein isolate from rapeseed might be a good source of ACE inhibitory peptides. The highest ACE inhibition was shown by pepsin-pancreatin digested precipitated protein isolate.

Oxidative stress, the increased production of reactive oxygen species (ROS), is another significant factor for the initiation or progression of several vascular diseases. ROS can cause extensive damage to biological macromolecules like DNA, proteins and lipids. Moreover, the oxidation of LDL leads to the increasing of its atherogenicity (Erdmann et al., 2008). Several amino acids, such as Tyr, Met, His, Lys, and Trp, are generally accepted to be antioxidants (Wang et al., 2005). High amounts of His and hydrophobic amino acids in peptides contribute to their antioxidant activity. Among all tested peptides, those with a Pro-His-His sequence showed the greatest antioxidant activity (Erdmann et al., 2008). Wheat germ and wheat gluten hydrolysates showed *in vitro* antioxidative and free-radical scavenging action. Defatted wheat germ protein isolates were hydrolyzed using alcalase. Biological activity of obtained wheat germ protein hydrolysates was determined by several tests: antioxidative activity in linoleic acid emulsion system, scavenging effect on DPPH free radical, superoxide radical-scavenging activity, hydroxyl radical-scavenging activity, reducing power and ferrous ion-chelating activity (Zhu et al., 2006). Enzymatic hydrolysis was also used for preparing hydrolysates from

wheat gluten and the enzyme used for the hydrolysis was papain. Antioxidative activity was measured using DPPH radical and TBA method (Wang et al., 2007). Iwami et al. (1987) showed that wheat gliadin, obtained by extraction of wheat gluten with 70% ethanol, has the most potent antioxidative action against peroxidation of linoleic acid. Other cultures whose protein hydrolysates showed antioxidative and free-radical scavenging activity are amaranth, pea and mustard.

Antioxidative activity of pea protein hydrolysate was confirmed by DPPH radical scavenging activity (Humiski and Aluko, 2007; Pownall et al., 2010), superoxide scavenging activity, hydrogen peroxide

(H₂O₂) scavenging activity, hydroxyl radical (OH•) scavenging assay, reducing power, metal chelating assay and inhibition of linoleic acid oxidation. The peptide fractions from pea had greater ability to inhibit linoleic acid oxidation and chelate metals than glutathione. However, they showed lesser ability to scavenge free radicals in comparison with glutathione (Pownall et al., 2010). Peptide fractions obtained by hydrolysis of *Brassica carinata* protein also showed antioxidative activity. It was confirmed by the antioxidant assay that used the discoloration of β-carotene, because β-carotene is extremely susceptible to oxidation caused by free radicals (Pedroche et al., 2007).

Table 3.

Examples of peptides derived from plant proteins with other effects on the cardiovascular system and metabolic disorders

Disease/condition		Biological test	Encoding protein/protein hydrolysate	Source	References
Cardiovascular system	Oxidation	<i>In vitro</i>	Wheat germ protein hydrolysate	Wheat	Zhu et al., 2006
Cardiovascular system	Oxidation	<i>In vitro</i>	Wheat gluten hydrolysate	Wheat	Wang et al., 2007
Cardiovascular system	Oxidation	Database	Glutelin tryptic digest	Amaranth	Silva-Sánchez et al., 2008
Cardiovascular system	Oxidation	<i>In vitro</i>	Protein hydrolysate	Pea	Humiski and Aluko, 2007; Pownall et al., 2010
Cardiovascular system	Oxidation	<i>In vitro</i>	Protein hydrolysate	Mustard	Pedroche et al., 2007
Cardiovascular system	Oxidation	<i>In vitro</i>	Wheat gliadin	Wheat	Iwami et al., 1987
Cardiovascular system	Oxidation	<i>In vitro</i>	Protein isolate	Rapeseed	Yoshie-Stark et al. 2006, 2008
Cardiovascular system	Thrombosis	Database	Glutelin tryptic digest	Amaranth	Silva-Sánchez et al., 2008
Cardiovascular system	Hyperlipidemia	<i>In vitro</i>	Protein hydrolysate	Mustard	Pedroche et al., 2007
Cardiovascular system	Hyperlipidemia	<i>In vitro</i> and <i>in vivo</i>	Peptides from globulin	Soy	Duranti et al., 2004, Pak et al., 2005
Cardiovascular system	Hyperlipidemia	<i>In vitro</i>	Proteins from wheat flour	Wheat	Tani et al., 1994
Cardiovascular system	Hyperlipidemia	<i>In vitro</i>	Wheat germ proteins	Wheat	Borel et al., 1989 in Möller et al., 2008
Gastrointestinal system	Hyperlipidemia	<i>In vitro</i>	Protein isolate	Rapeseed	Yoshie-Stark, 2006, 2008
Cardiovascular system	Diabetes	<i>In vitro</i> and <i>in vivo</i>	Wheat albumin	Wheat	Kodama et al., 2005
Gastrointestinal system	Increased appetite/obesity	<i>In vivo</i>	β-Conglycinin fragment	Soy	Nishi et al., 2003 in Erdmann et al., 2008

Beside mentioned wheat gliadin, another protein isolate that had antioxidative effect is rapeseed protein isolate. Yoshie-Stark et al. (2008) reported that precipitated rapeseed protein isolate had a greater radical scavenging capacity than ultrafiltered protein isolate.

Another complication related to CVD is the developing of thrombosis due to disorders in blood coagulation (Erdmann et al., 2008). Peptides with antithrombotic effect were reported by Silva-Sánchez et al., 2008. They were identified in a tryptic digest of amaranth glutelin fraction and their sequences were Pro-Pro-Gly, Pro-Gly, and Gly-Pro.

An unfavorable profile of blood lipids is another major risk factor for the development of CVD. Many studies have found a positive correlation between hypercholesterolemia and/or hypertriglyceridemia and the probability for developing CVD. Peptides with hypocholesterolemic effect have high amounts of hydrophobic amino acids, which enable them to bind bile acids and thereby enhance the excretion of steroids through feces (Erdmann et al., 2008).

Because cholesterol is a water-insoluble molecule, its intestinal absorption has a mechanism similar to that of triglycerides. Cholesterol, as well as triglycerides, requires micellar solubilization with bile salts. It has been reported that some peptides can decrease cholesterol solubility by replacing it in micellar structure. Similar hypocholesterolemic peptides were present in *B. carinata* protein hydrolysates (Pedroche et al., 2007).

Duranti et al. (2004) demonstrated that α -subunit from soybean 7S globulin exerts both lipid lowering effect in plasma and upregulation of the β -VLDL receptors in liver cells from hypercholesterolemic rats. This polypeptide was given to rats orally. Another study of soybean globulins showed that one peptide, isolated by HPLC from the pepsin hydrolysate of 11S-globulin, also possessed hypocholesterolemic activity. Its molecular weight was 755.2 Da and amino acid sequence was Ile-Ala-Val-Pro-Gly-Glu-Val-Ala. The hypocholesterolemic effect of this peptide was determined by two tests. The first test was the analysis of bile acid binding and

the second was percent inhibition of 3-hydroxy-3-methylglutaryl coenzyme A reductase *in vitro*. The lowering of the cholesterol level was explained by the binding of this peptide to the bile acids, which prevented their reabsorption in the gastrointestinal tract (Pak et al., 2005).

Wheat proteins exhibited hypolipidemic effects in several studies. Tani et al. (1994) isolated proteinous lipase inhibitor (LI) from wheat flour. Molecular masses of the LIs were determined by SDS-PAGE and their values were approximately 28 and 25 kDa. Lipase inhibitory activity was determined by the inhibition of porcine pancreatic lipase. Proteins isolated from wheat germ, whose molecular masses were 24.4 and 27.5 kDa also showed inhibitory effect on lipolysis (Borel et al., 1989 in Möller et al., 2008).

Yoshie-Stark et al. (2008) examined bile acid-binding capacity of rapeseed protein isolates. They found that precipitated protein isolate had higher binding capacity than ultrafiltered protein isolate. It was explained by the higher concentration of fiber in precipitate.

Peptides that have an effect on high level of glucose in blood are usually inhibitors of digestive enzymes that are involved in the degradation of starch.

Objective of research conducted by Kodama et al. (2005) was to examine the effects of single dose and long-term administration of wheat albumin (WA) on levels of glucose in blood. WA showed 150 times higher *in vitro* α -amylase inhibitory activity than that of wheat flour. *In vivo* test was also conducted with 12 healthy adult male volunteers for the single administration and with 24 patients with mild type II diabetes for the long-term administration. It was concluded that WA suppressed postprandial hyperglycemia without adverse effects such as hypoglycemia, gastrointestinal symptoms, or hepatotoxicity.

In many developed countries obesity is one of the risk factors for cardiovascular diseases. Hyperinsulinemia, insulin resistance and dyslipidemia have been linked to obesity. Lipoprotein profile of obese subjects shows higher levels of triglycerides, high LDL-cholesterol and low HDL-cholesterol (Erdmann et al., 2008).

Table 5.

Examples of peptides derived from plant proteins with effect on the immune system

Examples of peptides derived from plant proteins with effect on the immune system					
Disease/condition		Biological test	Encoding protein/protein hydrolysate	Source	References
Nervous system	Opioid agonist	Database	Glutelin tryptic digest	Amaranth	Silva-Sánchez et al., 2008
Nervous system	Opioid agonist	<i>In vivo</i>	Gluten exorphins	Wheat	Takahashi et al., 2000; Fukudome et al., 1993 in Hartmann and Meisel, 2007

Nishi et al. (2003) identified the peptide Val-Arg-Ile-Arg-Leu-Leu-Gln-Arg-Phe-Asn-Lys-Arg-Ser as the bioactive appetite suppressant in soy protein. The sequence of this peptide corresponds to the residues 51–63 of β -conglycinin. This fragment interacts directly with the intestinal mucosal cells to stimulate cholecystokinin (CKK) release. CCK is an important factor in appetite regulation. The stimulation of CCK release contributes to appetite suppression both in the central and periphery nervous system (Erdmann et al., 2008).

Regulation of the nervous system

There isn't much research about peptides that regulate function of the nervous system. It can be seen that peptides that have an opioid effect are derived from glutelin fraction of proteins.

Food-derived peptides with opioid activity were termed "exorphins" on the basis of their structural similarity to endogenous opioid peptides endorphins. The amino acid sequence common to both endogenous and exogenous peptides is an N-terminal tyrosine residue and the presence of another aromatic residue in the third or fourth position from the N terminus (Phe or Tyr). Fukudome et al. (1993) discovered novel opioid peptide exorphin C (Tyr-Pro-Ile-Ser-Leu) in wheat gluten. Takahashi et al. (2000) reported that newly isolated bioactive food protein fragment, gluten exorphin A5 (Gly-Tyr-Tyr-Pro-Thr), exhibited opioid activity in mice (Hartmann and Meisel, 2007). Silva-Sánchez et al. (2008) identified several peptides in a tryptic digest of amaranth glutelin fraction that can act as potent analgesics.

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ПОТЕНЦИЈАЛ БИОАКТИВНИХ ПРОТЕИНА И ПЕПТИДА ЗА ПРЕВЕНЦИЈУ И ЛЕЧЕЊЕ МАСОВНИХ НЕЗАРАЗНИХ БОЛЕСТИ

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Сажетак: Хронична незаразна стања су далеко водећи узрок смртности у свету. Она се често повезују са факторима исхране. Функционална храна се може дефинисати уопштено као било која храна која може да обезбеди здравствену корист једној или више телесних функција, поред основне исхране. Бројни пептиди са различитим функционалним својствима могу се наћи кодирани у примарној структури многих прехранбених протеина. Ови пептиди се могу ослободити током варења протеолитичким ензимима у гастроинтестиналном тракту или током ферментације и обраде хране. Биоактивни пептиди имају физиолошко дејство на главне телесне системе - кардиоваскуларни, дигестивни, ендокрини, имуни и нервни систем. Извори биоактивних протеина и пептида се могу поделити на животињске и биљне изворе. Културе богате протеинима, као што су житарице, псеудожитарице и махунарке су потенцијални извори биоактивних протеина и пептида са различитим дејствима, укључујући антипролиферативно, имуномодулаторно, антиоксидативно, антиромботично, антихипертензивно, хипохолестеролемично и опиоидно дејство.

Кључне речи: биоактивни пептиди, канцер, кардиоваскуларне болести, житарице, псеудожитарице, махунарке

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