Peptides (Peptones)[1] and iron absorption

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Introduction - Bioactive peptides in meat

Bioactive peptides are functional components, encrypted in the proteins and can be derived from food of plant and animal origin, including meat. These peptides are specific protein fragments of between 2–30 amino acids which, above and beyond their nutritional capabilities, have a positive impact on the body's function or condition which may ultimately influence health. Although, inactive within the sequence of the parent proteins, these peptides can be released by digestive enzymes in the gastrointestinal tract or by fermentation or ripening (ageing) during food processing and play an important role in human health (Bhat, et al., 2015).

The activity of peptides is based on the inherent amino acid composition and sequence and become active only when they are released from the precursor protein where they are encrypted. Bioactive peptides usually contain 2–30 amino acid residues (Ryan, et al., 2011; Lafarga & Hayes, 2014). Upon ingestion and absorption from the gastrointestinal tract, bioactive peptides, can trigger various physiological effects that may have a positive impact on body functions or conditions and ultimately influence health (Erdmann, et al., 2008; Madureira, et al., 2010; Lafarga & Hayes, 2014).

It has been reported that food derived bioactive peptides have antioxidative, antimicrobial, antihypertensive, antithrombotic, cytomodulatory[2], anticancer, immunomodulatory[3], opioid agonistic[4], mineral binding, hypocholesterolemic[5] and anti-obesity effects which mainly depends on their structure and other properties. In addition, many bioactive peptides possess multifunctional properties i.e., specific peptide sequences may initiate two or more different biological activities (Meisel & FitzGerald, 2003). The activity of bioactive

Figure 1: Health-related functions of biopeptides derived from meat proteins

[1] Peptones are a soluble protein formed in the early stage of protein breakdown during digestion of various protein derivatives that are formed by the partial hydrolysis of proteins (as by enzymes of the gastric and pancreatic juices or by acids or alkalis). Peptones can be used in media in microbiological studies for the production of bacterial toxins from organisms such as *Corynebacterium diphtheriae*, *Staphylococci*, *Clostridiae* and *Salmonellae*. As this literature study will be in a human nutrition context the term "peptides" will be used.

[2] Cytomodulatory refers to the capability of altering cells, especially their growth, immune responsiveness, or reproduction.

[3] Immunomodulatory refers to the capability of a substance that affects the functioning of the immune system

[4] An agonist is a drug that activates certain receptors in the brain. Opioid agonists bind to the opioid receptors and provide pain relief.

[5] Hypocholesterolemic refers to the capability of lowering cholesterol in the blood.

peptides depends on the amino acid composition and sequence. Moreover, compared to conventional small molecules, these peptides have high bioactivity, act on specific targets inside the body, have low levels of toxicity and don't accumulate in small amounts in the human body (Ryan, et al., 2011).

Until now numerous bioactive substances have been studied, but an increasing interest is focused on bioactive peptides of animal origin. Different peptides are assumed to interact with either minerals, vitamins or nutrients by a specific mechanism. These interactions could have an effect on the absorption of these nutrients.

Some of the most well described bioactive peptides in meat are as follows:

- carnosine (β-alanyl-l-histidine) - is a dipeptide molecule, made up of the amino acids beta-alanine and histidine. Carnosine is an endogenous dipeptide consisting of beta-alanine and histidine, present in the milimolar range in skeletal muscle and in the hundred-micromolar range in the vertebrate brain. It is reported that carnosine has antioxidant properties that may be beneficial in reducing the rate of formation of many degenerative diseases, such as diabetes, atherosclerosis, chronic renal failure, and Alzheimer's disease and ageing (Boldyrev, et al., 2013).
- anserine (β-alanyl-l-1-methylhistidine) - is a dipeptide containing β-alanine and 1-methylhistidine, which can be found in the skeletal muscle and brain of mammals and birds. Anserine is a strong antioxidant, previously demonstrated to reduce cognitive decline in the elderly (Szcześniak, et al., 2014).
- glutathione (γ-Glu-Cys-Gly) – is a tripeptide containing γ-L-Glutamic acid, L-cysteine and L-glycine produced naturally by the liver. It is also found in fruits, vegetables, and meats. Glutathione (GSH) is an important antioxidant in plants, animals, fungi, and some bacteria and archaea (single-celled microorganisms). Glutathione is capable of preventing damage to important cellular components caused by reactive oxygen species such as free radicals, peroxides, lipid peroxides, and heavy metals. It is used in metabolic and biochemical reactions such as DNA synthesis and repair, protein synthesis, prostaglandin synthesis, amino acid transport, and enzyme activation (Wu, 2013). Thus, every system in the body can be affected by the state of the glutathione system, especially the immune system, the nervous system, the gastrointestinal system, and the lungs. The content of GSH in meat is determined by many factors, i.e., breed, environmental conditions (including feeding methods), physical activity, age at slaughter, sex, physiological state, castration, and many other factors that are still unidentified (Rakowska, et al., 2017).

- Taurine, Citrulline and Ornithine are non-proteinogenic amino acids and will not be discussed in this literature study.

Table 1. Concentrations of small peptides and free non-proteinogenic amino acids in beef cuts (Wu, et al., 2016)

	Type of beef cut		
	Chuck	Round	Loin
Anserine (mg/g dry weight)	2.79	3.25	3.66
Carnosine (mg/g dry weight)	15.2	21.4	24.2
Glutathione (µg/g dry weight)	675	773	792

Values are means; n = 10. Calculations were based on the molecular weights of intact amino acids or intact small peptides.

The primary criteria used to assess the value of dietary protein have traditionally been amino acid composition and digestibility. Recently, it has been proposed that new parameters be evaluated such as the rate of protein digestion (Bauchart, et al., 2006). Therefore, meat proteins are not only important source of essential amino acids, but of bioactive peptides as well.

ABSORPTION OF IRON IN THE HUMAN BODY

Iron (Fe) deficiency is one of the most widely known nutritional disorders that affect an estimated two billion people worldwide. It occurs when there is a negative balance between iron requirements, absorption and losses. Iron deficiency is caused not only by an iron-deficient diet but also by low bioavailability of the iron in the diet (Zimmermann & Hurrell, 2007). Iron absorption occurs in the duodenum and the initial section of the jejunum. Bioavailability is limited by the physico-chemical and physiological properties of the iron ions (Czerwonka & Tokarz, 2017).

There are two primary forms of iron that are found in food, namely, haem and non-haem iron. Haem iron is derived mainly from haemoglobin and myoglobin in animal tissue, and according to the accepted Monsen model, makes up about 40 % of total iron. Non-haem iron is found mostly in plant-based foods, and makes up the remaining 60 % of iron in animal products (Monsen, et al., 1978).

The diverse mechanisms of absorption of haem iron and non-haem iron make these two forms of iron definitely different in bioavailability. In general, the rate of non-haem iron absorption is in the range of 2–20%, and is related to its solubility in the upper part of the small intestine. The presence of soluble enhancers (ascorbic acid and muscle tissue) and inhibitors (phytates, polyphenols and calcium) consumed during the same meal will have a significant effect on the amount of non-haem iron absorbed (Zimmermann & Hurrell, 2007; Pettit, et al., 2011). According to literature data, haem iron is much less affected by other dietary factors and contributes significantly to absorbable iron. The rate of absorption from the intestinal lumen for haem iron is 15–35% of total iron consumed (Clark, et al., 1997; Zimmermann & Hurrell, 2007; Pettit, et al., 2011).

Ascorbic acid is the most effective enhancer of non-heme iron absorption. Other dietary factors including citric acid and other organic acids, alcohol and carotenes similarly enhance non-heme iron absorption. Ascorbic acid enhances the absorption of non-haem iron by reducing ferric iron to the ferrous state, preventing its reaction with inhibitors such as phytic acid and reversing the inhibiting effect of such substances as tea and calcium/phosphate (Hurrell & Egli, 2010).

It was also demonstrated that non-haem iron absorption, especially from cereal- and legume-based meals, was increased by the simultaneous consumption of meat ("the Meat Factor") (Purchas, et al., 2006; Hurrell & Egli, 2010; Czerwonka & Tokarz, 2017). The addition of chicken, beef, or fish to a maize meal diet increased nonheme iron absorption 2-3-fold (Bjorn-Rasmussen & Hallberg, 1979).

Approximately 90% of dietary iron is consumed in the non-heme form from plant-based food sources. However, due to a low bioavailability it constitutes that only approximately 50% of iron is actually absorbed into the human body (Schönfeldt, et al., 2016). A decrease in meat availability and accessibility due to various reasons (i.e. environmental and other health concerns, poverty) resulting in a reduced intake of meat will make iron deficiency an even more common condition. A low meat intake results in a decreased amount of bioavailable iron in the diet, as well as, a decrease in the effect of meat on absorption of non-haem iron.

The "Meat Factor"

The nature of the enhancing effect of meat on non-haem iron absorption, is uncertain (Purchas, et al., 2006). It was found that other animal proteins with a similar amino acid composition as meat, such as egg albumin, have no enhancing effect on non-haem iron absorption. It therefore seems likely that peptides available after digestion of meat, rather than free amino acids are the origin of the enhancing effect. In a study (Hurrel, et al., 2006) it was demonstrated that haem or its degradation products did not play a role in the enhancing effect. The enhancing effect is mainly protein related. Subsequently, more attention has been centred on the protein digestion products of muscle tissue, particularly the cysteine-containing peptides that could both reduce ferric to ferrous iron and maintain iron in a soluble complex available for absorption (Hurrell & Egli, 2010).

When iron in food becomes more available for absorption, the pro-oxidative effect of iron may be associated with increased oxidative stresses in the human body. Carlsen and colleagues reported in a study (Carlsen, et al., 2003) that the salt soluble protein fraction from meat is the most effective in enhancing absorption of non-haem iron, while the water soluble fraction is the most pro-oxidative. It is important to note that the protein fraction from meat, which promotes iron absorption from non-haem iron when combining meat and vegetables, is not the same protein fraction from meat associated with pro-oxidative activity. Therefore, it should be possible to use meat protein fractionation to promote iron absorption without possibly inducing severe oxidative damage.

To estimate the iron availability and absorption of a combined meal, not only the concentration and type of iron present must be taken into account, but also other factors such as the possible enhancing factors and the presence and type of proteinaceous digestion products.

With the perspective of a reduced availability and accessibility of meat for human consumption due to the various reasons, it will become necessary when determining dietary protein quality to consider the potential effects of latent bioactive peptides that are released during digestion of the protein.

Production of bioactive peptides

The new trend of promoting human health by using bioactive compounds is particularly interesting and presents a great challenge but at the same time opportunity for the meat industry, to improve the quality and image of meat. These peptides can be used for health promotion and disease risk reduction, especially because they have some advantages compared to synthetic drugs (Lafarga & Hayes, 2014).

It was found that the bioactive peptide content was lower in fresh beef muscle and increased during ripening and cooking (Bauchart et al., 2006). Proteolytic degradation of muscle, which occurs post-mortem as part of the meat ageing process, results in the production of protein fragments. Polypeptide fragments may be further degraded by peptidases which will lead to the production of smaller peptides and the release of individual amino acids (Mullen et al., 2000).

There is a large generation of meat by-products, not only from slaughtering, but also in the meat industry from trimming and deboning during further processing. This results in extraordinary volumes of by-products that are primarily used as feeds with low return. The development of functional biopeptides from the diverse meat proteome is a viable method of adding value to meat by-products.

There are a number of methods by which peptides with biological activity can be produced from precursor proteins. The most common ones are (a) enzymatic hydrolysis with digestive enzymes, (b) by means of the microbial activity of fermented foods, (c) through the action of enzymes derived from proteolytic microorganisms (Bhat, et al., 2015). Detection of the low-molecular weight peptides is often hindered by the predominant myofibrillar proteins (larger proteins), but this can be avoided by appropriate fractionation (Erdmann, et al., 2008; Ryan, et al., 2011). Once the structure of bioactive peptides is known, it is also possible to synthesize peptides.

The presence of the structural properties, compared to proteins from other food sources, and diversity of animal-based proteins (in terms of amino acid sequence) provides sustainable bioresources for generating functional biopeptides with diverse human health applications (Udenigwe & Howard, 2013).

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