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Summary

Characteristics and identification of biologically active products from oat (*Avena sativa* L.) proteins hydrolysis in the aspect of prevention of the metabolic syndrome

The aim of the research presented in this thesis was the molecular characteristics of hydrolysates and peptides from oat kernels (*Avena sativa* L.) and their biological activities important from the point of view of the prevention of metabolic syndrome (MSyn), determined using *ex vivo* method in preliminary research and the hybrid system - *in silico* and *in vitro* methods. The biological activity of digests and peptides derived from oat proteins was analyzed under conditions of simulating digestion in the human gastrointestinal tract (INFOGEST international protocol). The new, integrated research strategy to identify and characterize the properties of oat kernels proteins that go beyond their nutritional functions in terms of preventive measures in the MSyn was used in this research.

At the preliminary studies extraction of oat kernels proteins was conducted and then *ex vivo* digestion using human digestive juices, isolated from volunteers was applied. Hydrolystaes of oat kernels proteins showed angiotensin I converting enzyme inhibitory activity. As the result of *in silico* experiment it was established that all analyzed oat kernels proteins had in their sequences peptides characterized by dipeptidyl peptidase IV (DPP-IV) and angiotensin I-converting (ACE) enzymes inhibitory activity, antioxidant activity. It was possible to release them during hydrolysis with pepsin, trypsin and chymotrypsin. Based on the obtained bioinformatics analysis results, it was found that selected oat kernel proteins can be a potential source of peptides with DPP-IV and ACE inhibitory activity, antioxidant activity that are released during *in silico* digestion.

The next step of the studies was *in vitro* digestion using commercial enzymes preparations - pepsin and Corolase PP. The simulated *in vitro* digestion of oat kernels proteins in conditions imitating the human digestive tract were carried out according to the INFOGEST protocol. It includes the digestion phases in the oral cavity – 3 minutes, stomach - 2 hours at pH 3.0, and stomach-small intestine - 2 hours at pH 7.0. Hydrolysates from oat kernels were also examined for the amount of non-hydrolyzed protein content. Hydrolysates were separated and analysed using SDS-PAGE, 2D-PAGE, RP-HPLC methods. Then the hydrolysates were analysed in the aspect of their ability to inhibit the activity of the following enzymes: DPP-IV, α-glucosidase and ACE as well as of their antioxidant activity during determined ability of the

reduction free radicals DPPH and ABTS, the reducing power of Fe (III), the ferric reducing antioxidant power and the ability to chelate ferrous of Fe (II). The peptide's identification was carried out with mass spectrometry (LC-MS/MS). Hydrolysates of oat kernels proteins showed enzymes inhibitory activity i.e.: DPP-IV, α-glucosidase, ACE and antioxidant activity. Increased inhibitory activity against DPP-IV, α-glucosidase and ACE. Antioxidant activity was found during all steps of *in vitro* digestion. The LC-MS/MS method used to identify peptides were effective and it was possible to identify 21 peptides with DPP-IV and ACE inhibitory activity and antioxidant activity.

The novel hybrid approach combined with *ex vivo* analysis enabled identification and characteristics of proteolysis products from oat kernels proteins in the aspect of the potential possibility of their use in the prevention of MSyn. The research strategy involving the *in silico* and *in vitro* methodologies may be useful in studies of proteins from oat kernels supporting the treatment of MSyn dysfunctions.