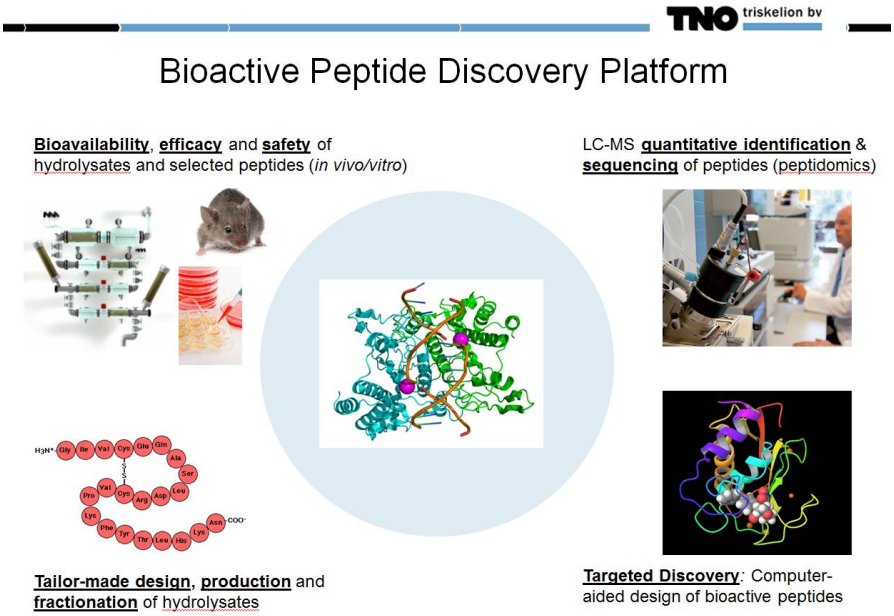
# Bioactive Peptide Discovery Platform

Bioactive peptides are short amino acid chains that are inactive within the sequence of the parent protein and can be activated through gastrointestinal digestion, food processing, storage or in vitro hydrolysis by proteolytic enzymes. Since bioactive peptides have the ability to impart a biological effect resulting in a positive impact on body functions or conditions, their application is of relevance for food industry. Together with TNO, TNO Triskelion offers a discovery platform covering relevant know-how and expertise needed to assess the hidden bioactive potentials of food proteins. An overview of this platform is presented in the figure below.




The digestion of proteins and the intestinal stability of resulting peptides can be investigated with the dynamic gastrointestinal model (TIM). This validated *in vitro* model allows to measure the true ileal digestibility of proteins and to monitor the luminal formation of breakdown products, i.e. peptides. Samples can be taken at various sites to identify specific bioactive peptides whether or not in relation with other ingested food compounds. The TIM system can be adjusted to mimic different species or ages, as for example infants or elderly, that have an altered digestive physiology. Additionally, *in vitro* and *in vivo* efficacy testing of peptides and hydrolysates can be performed in relevant pharmacology models.

We are fully equipped and experienced to perform hydrolysate screening and sequence analysis of proteins using Liquid Chromatography – tandem Mass Spectrometry (LC-MS/MS). When needed, we develop a suitable protein purification method to enrich the protein of interest from the (biological) matrix. After application of our optimized in-house digestion method, chromatographic separation is performed using UPLC or nano-LC after which the eluting peptides are determined with our Thermo Q Exactive system, that provides very accurate m/z determinations. Sequence information or protein identifications can be obtained through MS/MS analysis and subsequent database searching and/or manual interpretation. When high sensitivity is required, we perform targeted protein analysis using signature peptides on one of our Waters Xevo triple quad systems. Although we are continuously trying to further increase the sensitivity, we are now able to quantitatively and selectively determine several individual proteins with an LLOQ of 10-20 ng/ml in biological matrices using only 25 µl sample volume.

Design of peptides based on relevant structure-function parameters can lead to the discovery of 'new' bioactive peptides. Computational chemistry and molecular modeling tools provide methodologies to identify peptide sequences present in the parent protein and their 3D structure. Using *in silico* modeling, these can be related to biological activity and targets of interest. The *in silico* targeted discovery approach is well known and frequently used in drug discovery in pharmaceutical industry. Computational chemistry at TNO is based on a broad palette of experience in pharmaceutical drug discovery, and application of this knowledge towards food industry may lead to new insights through elucidation of mechanistic knowledge on ingredients such as bioactive peptides.

For more information please contact us: info@tno.triskelion.nl