In operando-SAXS analysis of multiscale structural changes in food dispersions during in vitro digestion processes

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The multiscale structures in food play a critical role in determining the physicochemical properties such as stability, texture before oral processing, and digestibility and absorption during gastrointestinal bioprocessing. Therefore, a series of methods has been widely developed to analyze the structure of foods at various scales, ranging from the molecular level to the macroscale. Among these techniques, small- and wide-angle X-ray scattering (SAXS and WAXS) have been gaining increasing attention because of their potential to characterize the internal structures of materials non-destructively and at the nanoscale. Moreover, the techniques provide nanometer-resolution images in their native state, without the need for freezing the sample.

The structural hierarchy of foods and beverages is partially or mostly broken down and simultaneously reorganized during digestion processes in the human gastrointestinal tract. To achieve efficient and controlled delivery and absorption of major and minor nutrients, including bioactive compounds, it is essential to understand the multiscale structural changes that occur during digestion processes. Several studies have previously reported analyses of in vitro simulated gastric processes using SAXS to understand structural changes in lipid-based systems. Here, we report the development of an INFOGEST simulated digestion model integration into the SAXS system. The new INFOGEST-SAXS tool enables the study of nanostructure-digestion correlations *in operando* and for a broad range of food materials, including proteins, lipids, and fibres. Our new *in vitro* digestion-SAXS tool deepens the understanding of multi-structural changes throughout the gastrointestinal tract.

In this contribution, we will focus on structural changes during the digestion of a) model plant protein in solution, b) plant protein microgel dispersion, and c) plant-based milks (e.g., soymilk). The samples will undergo bioprocessing in a temperature-controlled digestion chamber, where they are step-wise mixed with gastric and intestinal juices, including amylases, proteinases, and lipases, according to the well-established INFOGEST protocol. An aliquot of the digesta in the chamber is constantly circulated through a pump system composed of a pipe and a flow-through glass capillary, where the digesta is exposed to X-ray. The time-dependent structural changes in the average scattering patterns obtained within a fixed irradiation time will be understood in comparison with conventional methods for digestion analysis.

Keywords:
Small-angle X-ray scattering (SAXS); in vitro digestion; multi-scale structure