The milk-coagulating enzymes (MCEs) from traditional Chinese Jiuqu microbes are the key enzymes that causes milk coagulation during processing of traditional Chinese Royal Cheese – soft unripened cheese, produced by combination of enzymatic-thermal coagulation. Chinese Jiuqu that is traditionally used as microbial starter to make rice wine contains microorganisms producing MCEs. However, very little is known about the enzymatic properties of MCEs and their effect on the cheese quality. Therefore, it is of significance to carry out the research focused on the application of MCE in cheese production, and the results achieved will provide new knowledge about the innovative microbiological MCE and the possibility of its application in innovative cheese manufacture.

The main objective of this project is to evaluate the implicational potential of MCE-coagulating enzymes from traditional Chinese Jiuqu microbes (MCEj) in added-value cheese manufacturing. The specific aims include: determination of factors affecting coagulation process of milk induced by MCE-coagulating enzymes from traditional Chinese Jiuqu microbes; understanding the molecular basis of coagulation mechanism, and determination of influence of use of innovative enzymes on molecular basis of coagulation, composition and ripening (development of flavor and texture) of Dutch-type cheeses as well as the characterization of selected biological properties of digests and peptides derived from caseins in cheese matrix.

In the present project, the mechanism of milk coagulation by the MCEj will be studied. The capability of the MCEj to hydrolyze casein will be studied in terms of formation of bioactive peptides by peptidomics method with computer simulation analysis. Bioactivities of the peptides will be evaluated by *in vitro* methods. The effects of the MCEj on cheese ripening properties including cheese composition, texture, microstructure, flavor, protein degradation and bioactivities will be studied. The prophylactic potential of the new cheese in Metabolic Syndrome will be demonstrated. In particular, the studies will concern peptides, released during digestion, which show the following types of activity: antihypertensive (inhibition of angiotensin converting enzyme), antidiabetic and anti-obesity (inhibition of dipeptidyl peptidase IV, α-glucosidase, α-amylase, lipase) and antioxidant. Peptidomics and spectral methods will be used to further explore the biological potential of cheese during ripening as affected by the MCEj. Proteomics methods will be used to further explore the biochemical changes of cheese during ripening as affected by the MCEs. Cheese-making parameters in relation to the quality of cheese made with the MCEj will be studied thoroughly, combining the results described above, to elucidate the developing mechanism of cheese quality.

The results of this study will provide new knowledge about the enzymatic coagulation and casein hydrolysis/digestion properties as affected by the novel microbial MCEj obtained from traditional Jiuqu. Further understanding of the biochemical changes that take place during ripening process of cheese made with the MCEj resulting in formation of bioactive peptides and their precursors released during digestion, as well as cheese-making parameters and cheese quality development will provide scientific basis for establishing processing operation specifications of cheese with novel functionalities. Use of innovative coagulating enzymes would alter cheese making characteristics but can also contribute to production of new cheese varieties in the future.