Supporting Material

for

Achieving realistic gastric emptying curve in an advanced dynamic in vitro human digestion system: experiences with cheese - a difficult to empty material

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Preparation of simulated digestive fluids

In brief, the stock solutions of various salts including KCl (0.5 mol/L), KH$_2$PO$_4$ (0.5 mol/L), NaHCO$_3$ (1 mol/L), NaCl (2 mol/L), MgCl$_2$(H$_2$O)$_6$ (0.15 mol/L) and (NH$_4$)$_2$CO$_3$ (0.5 mol/L) were prepared in advance. The SSF was prepared by dissolving \( \alpha \)-amylase in the SSF stock electrolyte solution, followed by adding 0.3 M CaCl$_2$ and deionized water to achieve 150 U/mL \( \alpha \)-amylase. The pH of SSF was adjusted to 7.0 using 1 mol/L NaOH. Similarly, pepsin was dissolved in the SGF stock solution, and 0.3 M CaCl$_2$ and deionized water were added to achieve 4000 U/mL pepsin in the final SGF with pH of 1.6 adjusted using 6 mol/L HCl.

Discussions on the physiological relevance of the inclination of the stomach system

In terms of the tilting of the stomach system, it may physiological relevant to the modification of gastric emptying rate induced by posture \(^1\), which provides an in vitro way to investigate the effect of posture on gastric emptying. Moreover, it is likely that the relative position of the stomach in humans would change in relation to the volume of gastric contents remaining in the stomach during digestion. It has been reported that the normal capacity of the human stomach varies from less than 0.25 L in the fasting state, and it can expand up to 1.7 L after intake of a big meal \(^2\). Although the human stomach is generally characterized as a J-shaped organ, its geometry and size can be significantly influenced by the position of the body, the condition of surrounding viscera and organs, the amount and type of meal ingested, and the digestion time \(^2\). Due to the effect of gravity and the gastric accommodation to food intake, the fasting stomach with a fist-like morphology seems to be almost at a vertical position (\( T = t_1 \)); however, the stomach may tilt towards a more horizontal position when it is full of meal (\( T = t_3 \)). With the continuous reduction of gastric volume due to gastric emptying, the expanded stomach may recover back to the approximately vertical position (\( T = t_5 \)). The evolutions of the position, size and morphology of the human stomach during food intake and gastric emptying are schematically illustrated in Fig. S1 below. We think
this is a reasonable speculation in terms of the unique morphology and anatomy of the human stomach, although direct in vivo evidences have not been found. On this basis, it is reasonable to claim that the inclination of stomach system as shown in this study is physiologically relevant to that occurring in vivo.

**Fig. S1.** Schematic diagram illustrating the evolutions of the position, size and morphology of the human stomach during food intake and gastric emptying.

**Fig. S2.** Changes in the gastric pH as a function of gastric emptying ratio (%).