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Ca-NutriChip: Gastrointestinal tract on a chip for monitoring the bio-availability of calcium (Concept)

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Introduction

Calcium is essential to human life and health, and it plays a major role in health and disruption of calcium homeostasis is associated with osteoporosis, obesity, diabetes, cancer, hypertension, cardiovascular diseases, kidney stones and so on. The importance of pharmacokinetics has long been recognized in medicine, but in nutrition research, the concept of nutrikinetics still is in its infancy. To trace fate of nutrients in human gut, the new concept ***Ca-NutriChip*** innovated from Nutrichip is proposed. This concept will be able to quantitatively monitor the adsorption and transport of calcium through epithelial cell layer as well as it's uptake by target cells using a fluorescence-based imaging technique cells.

Concept

To investigate the fate of a nutrient after its processing journey, in vitro models have been developed that mimic the digestion of nutrients in the gastrointestinal tract as well as their transport though the intestinal barrier [1]. However, these in vitro systems do not readily permit dynamic real time analyses. The Ca-Nutrichip possessing a new functionality to measure the bioavailability of Ca timely would strengthen the ability of NutriChip platform and widen its application. In this concept, the confluent Caco-2 cell monolayer cultured both in transwell and chip serves as a model for the intestinal barrier through which calcium transport will take place [2], intracellular (see Figure 2) and free calcium (see Figure 3) will be measured by fluorescence resonance energy transfer (FRET) and Fura-2.

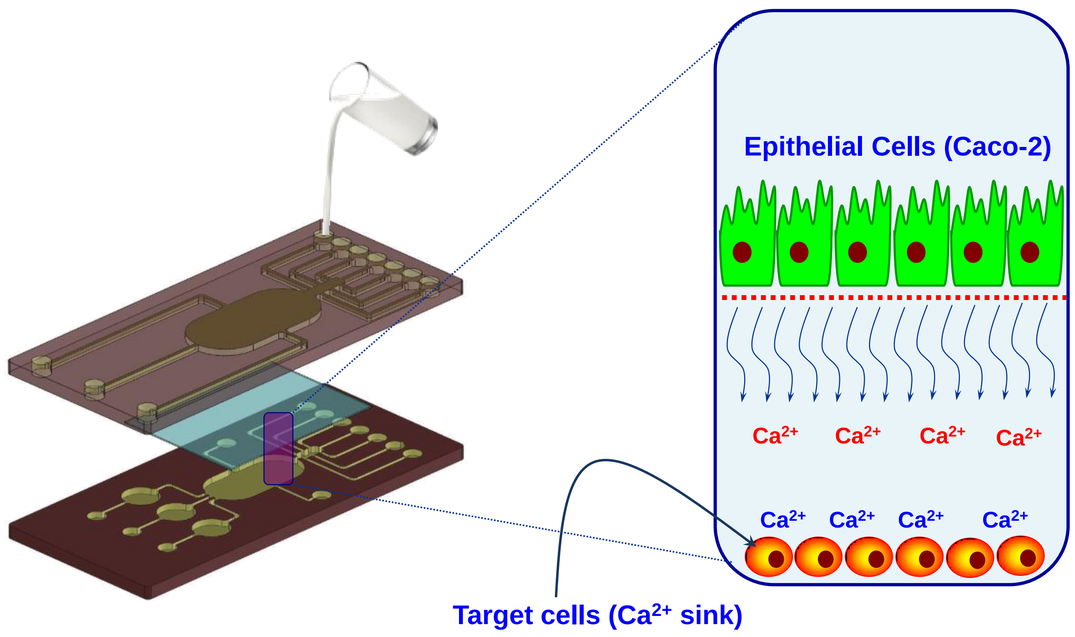

Figure 1. Schematic representation of Ca-Nutrichip

Calcium Image Techniques

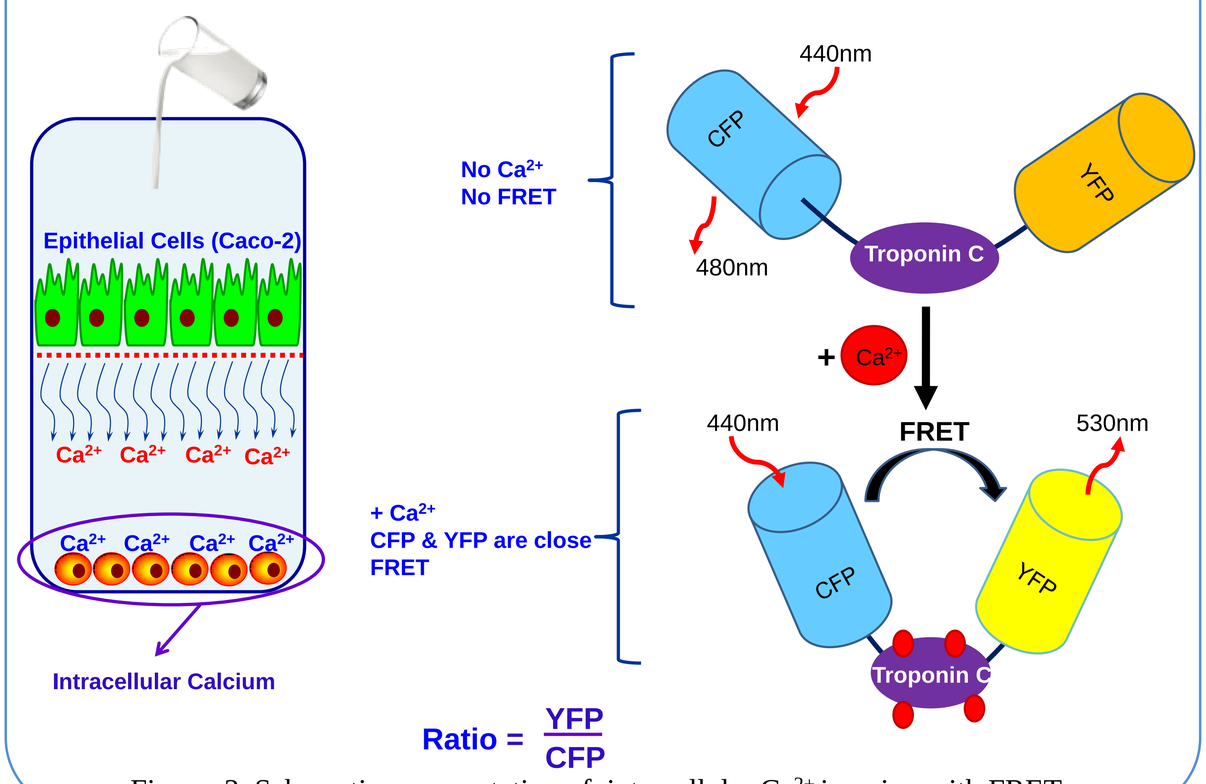

Figure 2. Schematic representation of intracellular Ca^{2+} imaging with FRET

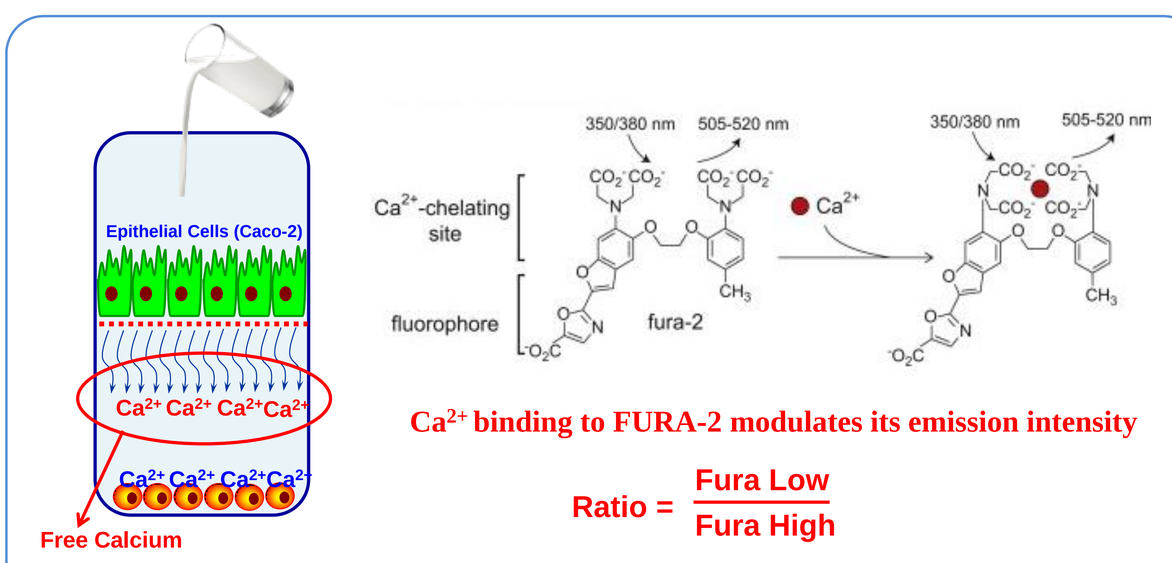

Figure 3. Schematic representation of free Ca^{2+} imaging with Fura-2

Results

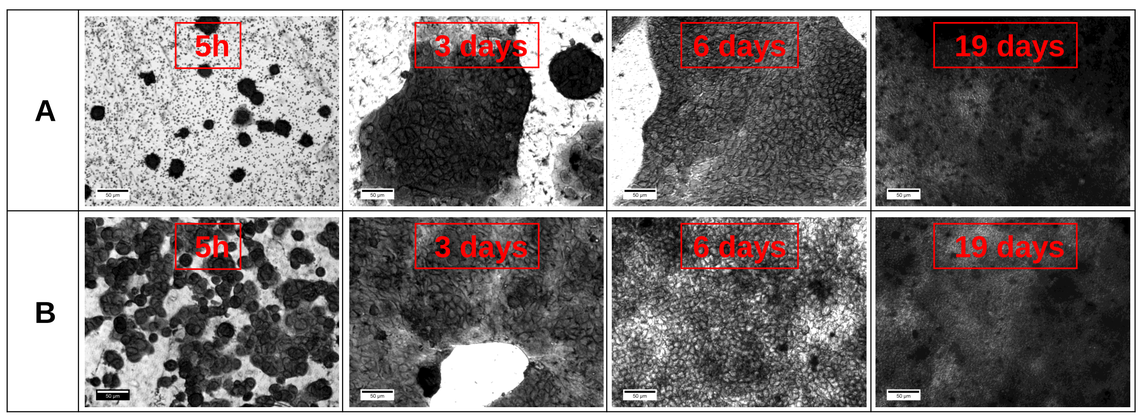

Figure 3. Transmission images of Caco-2 cells in 6 well transwell plate (4.67 cm^2 per insert) in different culture times (5h, 3 days, 11 days, 19 days). **A)** Seeding concentration of Caco-2 was 1.1 $\times 10^4$ cells/cm^2. **B)** Seeding concentration of Caco-2 was 5.5 $\times 10^4$ cells/cm^2. Scale bar is 50 µm. All samples were stained by crystal violet.

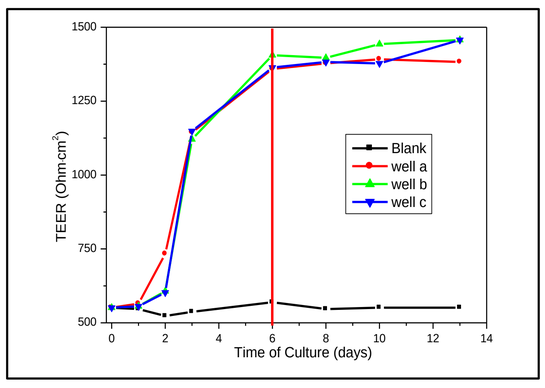

Figure 4. Transeptihelial electrical resistance (TEER) values of Caco-2 cell monolayer cultured in 6 well Transwell plate (4.67 cm^2 per insert). Seeding concentration of Caco-2 was 5.5 $\times 10^4$ cells/cm^2.

Summary & Outlook

1. Through the Ca-NutriChip project we are proposing a microfluidic system for cell mono- and co-culture which mimics the epithelial layer of the gastrointestinal tract (GIT) as well as interaction with target cells. The in vitro GIT model will be used to quantify bio-availability of calcium in dairy products.
2. The microfluidic chip consists of two polydimethylsiloxane (PDMS) layers sandwiching a thin, porous, and optically clear polyester membrane.
3. The transport of Ca^{2+} through the epithelial cells as well as the uptake of Ca^{2+} by target cells will be measured using ratio imaging techniques employing FURA-2 and YFP/CFP (FRET) calcium indicators.
4. Preliminary study of Caco-2 culture has been conducted and cell confluent monolayer has been achieved after one week of culture.

References

[1] Au, A.P. et al. *Journal of Nutrition.2010*, 130:1329-1334.

[2] Cosentino, S et al. *The Journal of Nutritional Biochemistry.* 2010, 21:247-254.

[3] Kim E et al. Angewandte Chemie International Edition.2006, 45,4562-4588