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## Comparaison of dynamic in vitro digestion of human milk vs standard infant formula to better understand their digestive kinetics

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INTRODUCTION and OBJECTIVE

Human milk (HM) is an optimal bioactive fluid, which meets infant requirement and is frequently substituted by infant formula (IF).

These two infant diets are assumed to have different digestion kinetics although they are rarely directly compared.

The present study aimed to evaluate the digestion kinetics and the structure evolution using the DIDGI® dynamic digestion system at the infant stage

METHODOLOGY

In vitro digestion



DIDGI® system



**Human Milk:** Pool of 50 raw milk samples  
Lactation time: 1.8 - 2 months post-delivery  
**1.0% true proteins, 2.8% lipids**



**Infant formula:** NativIF basic IF powder (Yu et al. 2021)  
Rehydrated at **1.4% true proteins, 3.2% lipids**

**Parameters based on literature** (Roman et al. 2007; Bourlieu et al 2014):

- Gradual decrease of gastric pH →  $pH = 8 \times 10^{-5} \times time^2 - 0,031 \times time + pH_{meal}$
- Enzymes: Rabbit Gastric Extract + Porcine pancreatin. Bovine bile
- Gastric emptying by Elashoff fitting (half-time emptying –  $T_{1/2 HM} = 47$  min ;  $T_{1/2 IF} = 78$  min).

**Sampling times** (min of digestion):

- Diet (G0)
- Gastric phase: G20, G40, G80, G120, G180\*
- Intestinal phase: I20, I40, I80, I120, I180

\*only for IF sampling

MACROSCOPY Scale

Evolution of the matrix structure

- Laser light scattering
- Confocal microscopy(Confocal Zeiss)

MOLECULAR scale

Lipolysis & Proteolysis

- GC : Gas chromatography
- SDS-Page
- OPA

RESULTS

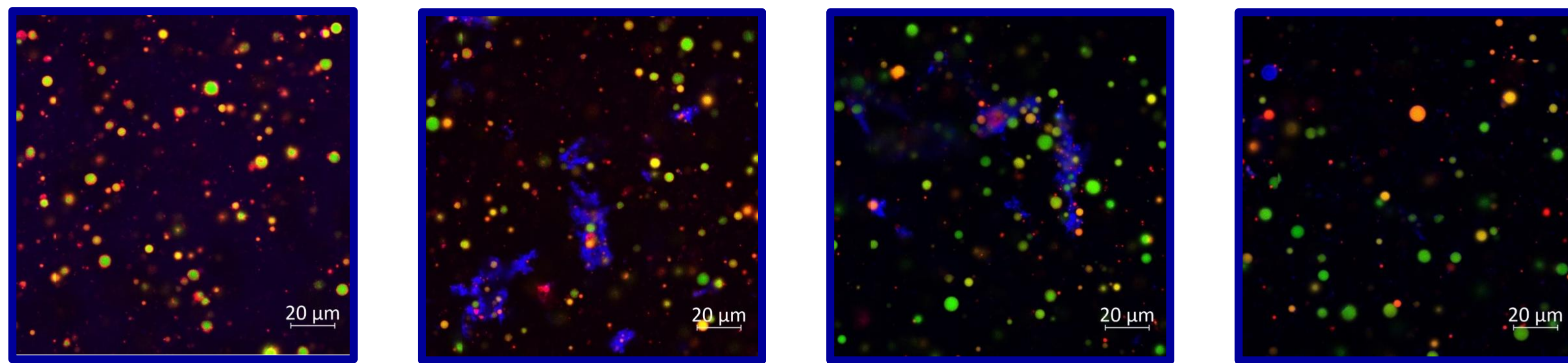
— Human milk — Infant formula

Structure:

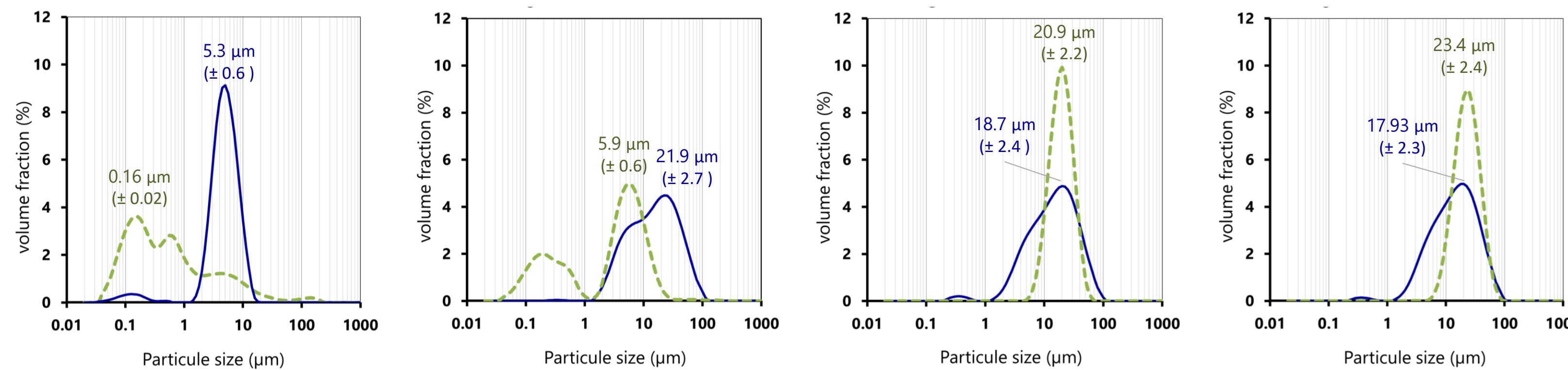
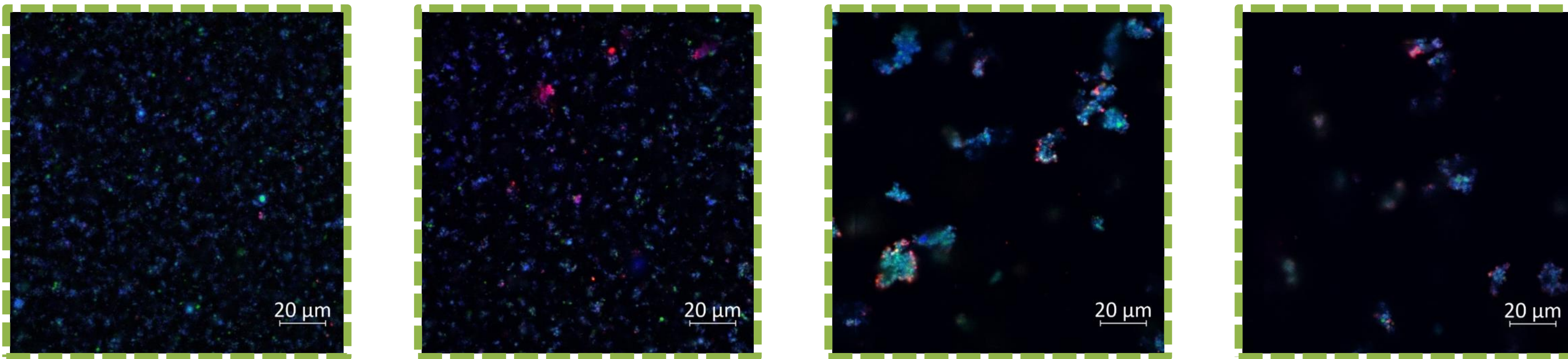
G0 G40 G80 G120 Time (min)



Human Milk



Infant formula

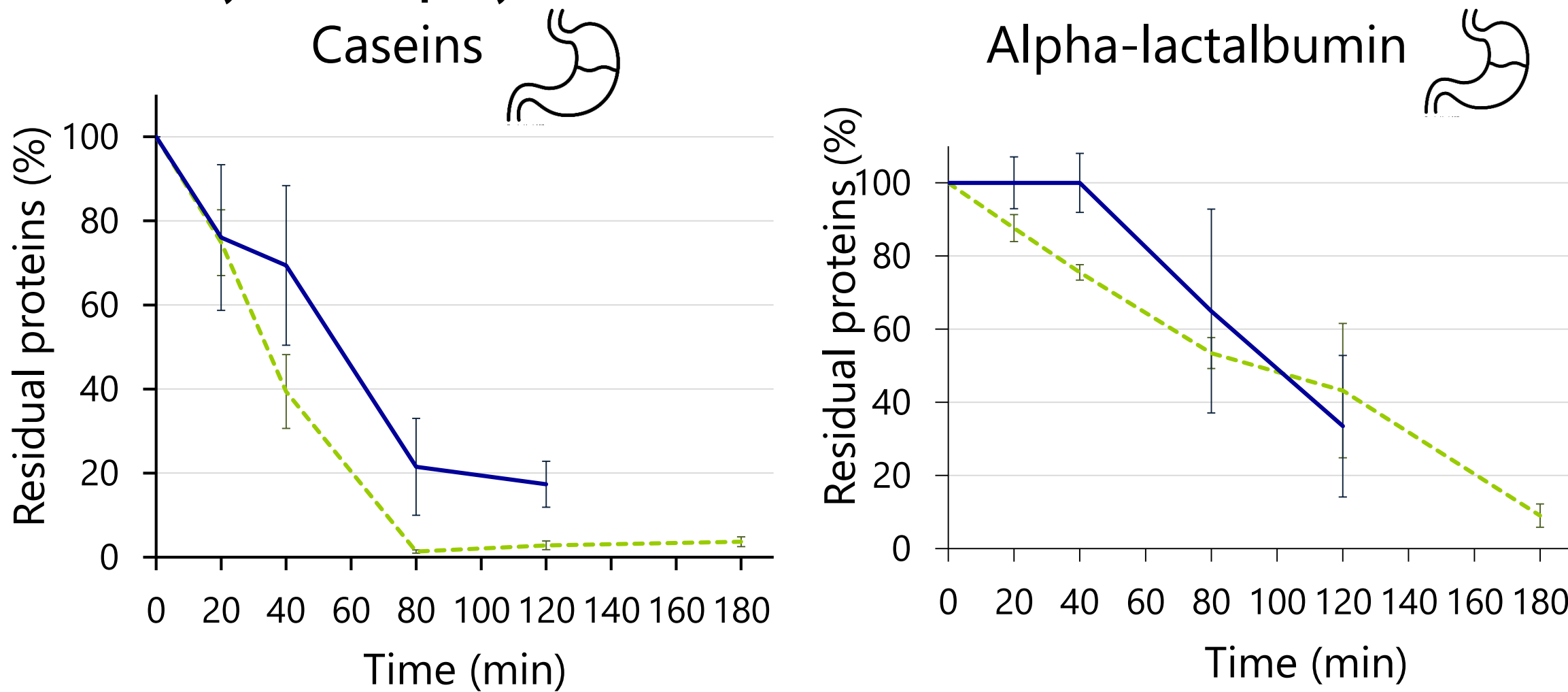


- Apolar lipid
- Proteins
- Amphiphilic molecules

Highlights :

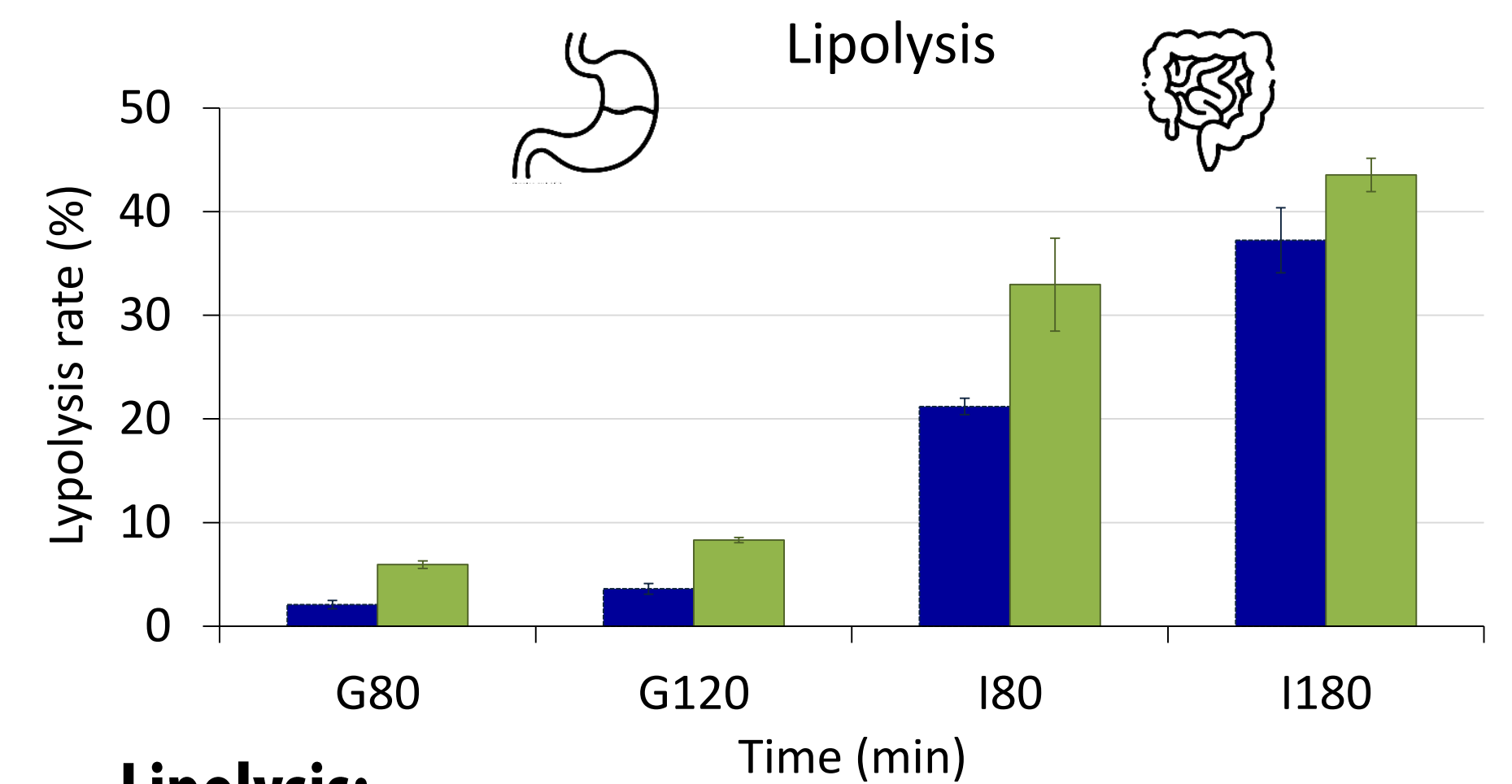
- **HM fat globules** were sized around **5 µm** while **IF fat droplets** were sized under 1 µm. HM fat globules remained present across time.
- **Particle aggregation** specifically protein one was **faster in stomach** during **HM** digestion (40 min) than in IF (80 min).
- **Final aggregate sizes** were more **heterogenous** for **HM**.
  - For **HM**, particle size was due to protein aggregation and **remaining native fat globules**.
  - For **IF**, **high particle size** observed **after 80 min** was due to **protein aggregation**.

Proteolysis & Lipolysis: Representation corrected by meal dilution and emptying (Mean ± SEM)



Proteolysis:

- **No significant difference** between **caseins** and **alpha-lactalbumin** release between diets, although HM proteins tended to be more resistant in the gastric phase.
- **Proteolysis** was significantly **lower** in **HM** at **I40** and **I120**. Faster proteolysis for IF during the first digestion times.



Lipolysis:

- **High lipolysis** rate in **raw HM** prior to **digestion** due to endogenous lipase activity (10 %) → subtracted here for lipolysis rate during digestion
- **Lipolysis** was **not** significantly **different** although it tended to be faster for IF during the early intestinal digestion phase.

CONCLUSION

Despite nutritional similarity, this study highlights that the **influence** of the **matrix** on the **structure of the digesta** and on the **digestion kinetics** and gives some **further understanding** to the **global value of digestibility**, such as determined *in vivo*.