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Static vs dynamic *in vitro* digestions of an innovative *Citrus* concentrate: Bioaccessibility of its phytomicronutrients

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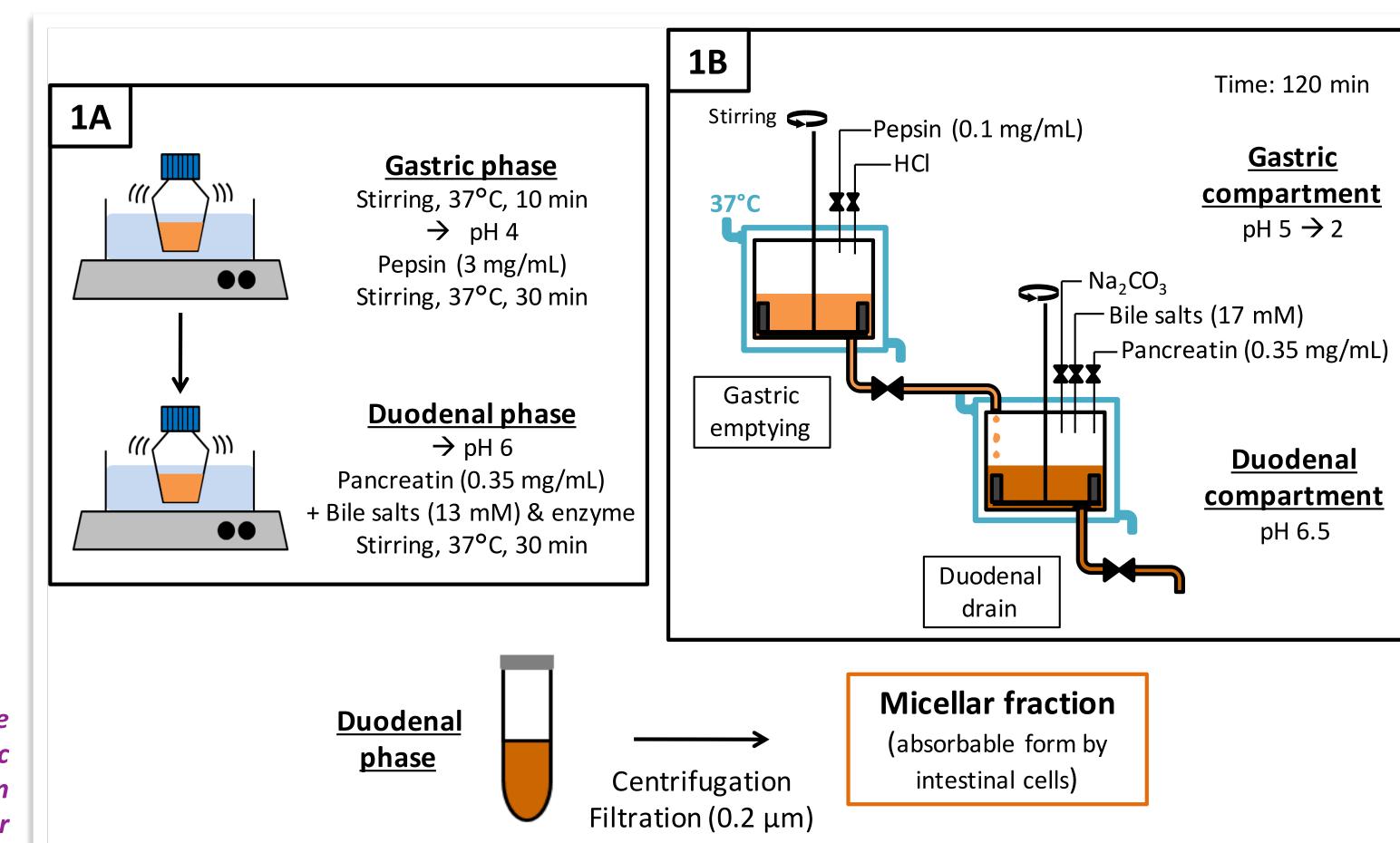
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Citrus juices and fruits, highly consumed worldwide, represent a significant dietary source of pro-vitamin A carotenoids and flavonoids which could contribute to the beneficial health effects of Citrus products. The aim of the present work was to assess the bioaccessibility of the main carotenoids and flavanones of a new Citrus clementina concentrate specially enriched in β -cryptoxanthin and hesperidin. Dynamic vs Static in vitro digestion systems were used to compare carotenoid micellarization and flavanone solubility.

Methodology

The innovative citrus concentrate was obtained by cross-flow microfiltration. The *in vitro* static model used (**Figure 1A**) has been validated against human studies and considered as a reliable model for carotenoid behavior [1]. The dynamic gastrointestinal model DIDGI® used (**Figure 1B**) was controlled by the STORM software. Pro-vitamin A carotenoids (β -cryptoxanthin, β -carotene) and flavanones glycosides (hesperidin, narirutin) were analyzed by HPLC-DAD.

Figure 1. Diagram of the static (1A) and dynamic (1B) in vitro digestion processes used for clementine concentrate



Results

Carotenoid micellarization was slightly higher in the dynamic *in vitro* digestion compared to the static one (**Table I**). This can be accounted by varying bile contents which are higher until 38 min and then lower than in the static model. In both digestion systems, β -cryptoxanthin bioaccessibility tended to be greater (15 to 21%) than that of β -carotene (12 to 17%) due to molecule polarity and in agreement with literature [2].

Finally, the dynamic system led to bioaccessibility kinetics which showed an optimal carotenoid micellarization at 90 min of digestion (Figure 2).

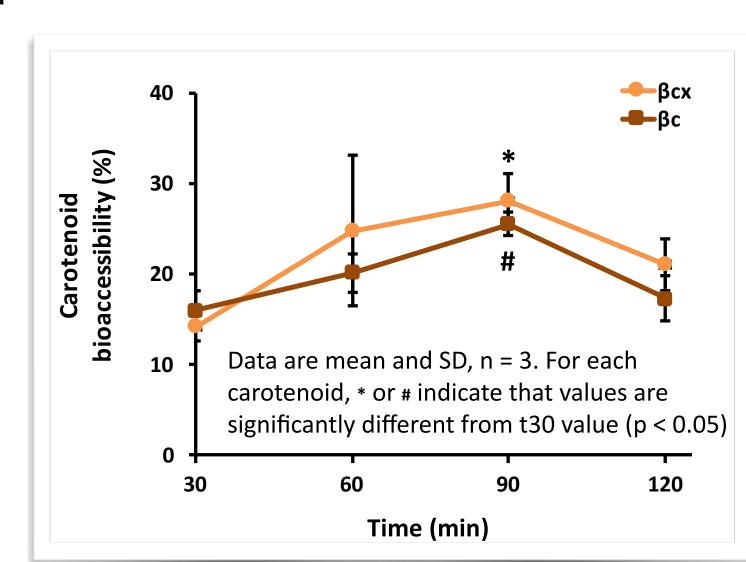


Figure 2. Kinetic bioaccessibility of carotenoids during dynamic in vitro digestion of the concentrate

Table I. Bioaccessibility of pro-vitamin A carotenoids and solubility of flavanone glycosides from Clementine concentrate

In vitro digestion model		Carotenoid bioaccessibility (%)		Flavanone glycoside solubility (%)	
		β-cryptoxanthin	β-carotene	Hesperidin	Narirutin
Static	Mean	14.9	12.3	2.9	16.1*
	SD	0.8	2.3	0.1	0.1
Dynamic	Mean	21.1 [‡]	17.3*‡	2.8	16.7*
	SD	2.8	2.4	0.2	0.3

Data are mean and SD, n = 3. ‡ indicates significant difference between digestion models and * indicates that values in the same case are significantly different (p < 0.05)

✓ Additionally, hesperidin in the duodenal aqueous phase appeared very low (3%) in both models as limited by solubility while that of narirutin reached 16% (**Table I**). Total flavanone glycoside contents decreased around 35% during duodenal dynamic digestion as did their soluble contents (**Figure 3**).

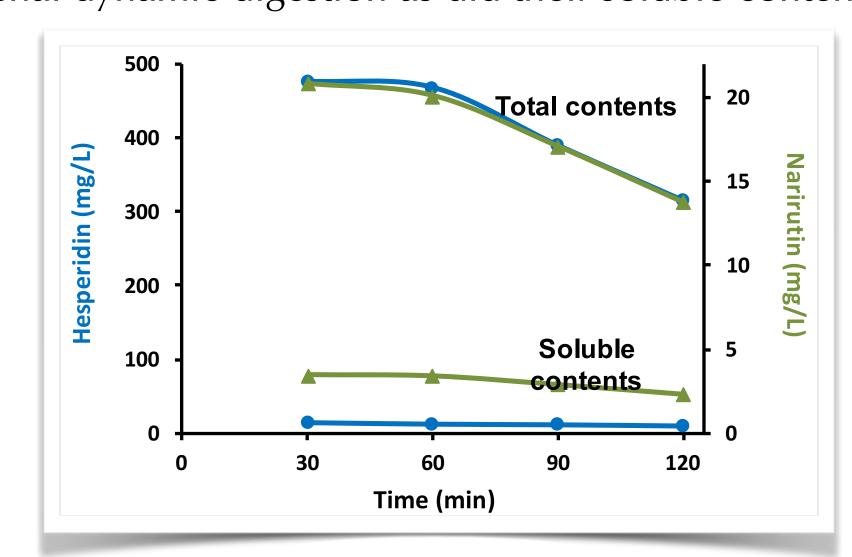


Figure 3. Duodenal flavanone glycoside contents during dynamic in vitro digestion of the concentrate

Conclusion

[1] ETCHEVERRY, P., GRUSAK, M. A., & FLEIGE, L. E. (2012). Application of *in vitro* bioaccessibility and bioavailability methods for calcium, carotenoids, folate, iron, magnesium, polyphenols, zinc, and vitamins B(6), B(12), D, and E. Front Physiol, 3, 317.

[2] DHUIQUE-MAYER, C., BOREL, P., REBOUL, E., CAPORICCIO, B., BESANCON, P., & AMIOT, M. J. (2007). Betacryptoxanthin from citrus juices: assessment of bioaccessibility using an *in vitro* digestion/Caco-2 cell culture model. Br J Nutr, 97(5), 883-890.

References

✓ Overall, these results indicated that approximately 5 – 7 mg/L of pro-vitamin A carotenoids were bioaccessible and 85 mg/L of soluble hesperidin after both *in vitro* digestion models, suggesting interesting contents for intestinal or colon absorption. The dynamic digestion confirmed the efficiency of the static model which estimates bioaccessibility and solubility in a faster, simpler and less expensive manner. On the other hand, the dynamic digestion model can be used to study kinetics of food digestion and reactivity allowing to uncover associated mechanisms (matrix effect or oxidative degradation).



