

INFOGEST 2.0 digestion method: Characterization of the sterol content in the digestion reagents



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INTRODUCTION

The INFOGEST digestion method was adapted for the evaluation of sterol bioaccessibility in a plant sterol (PS)-enriched beverage¹. Recent modifications of the method such as the addition of gastric lipase and cholesterol esterase, showed that the presence of sterols in the digestion reagents negatively affects to the sterol bioaccessibility². However, the sterol content in these reagents as well as their contribution during gastrointestinal digestion remains unknown.

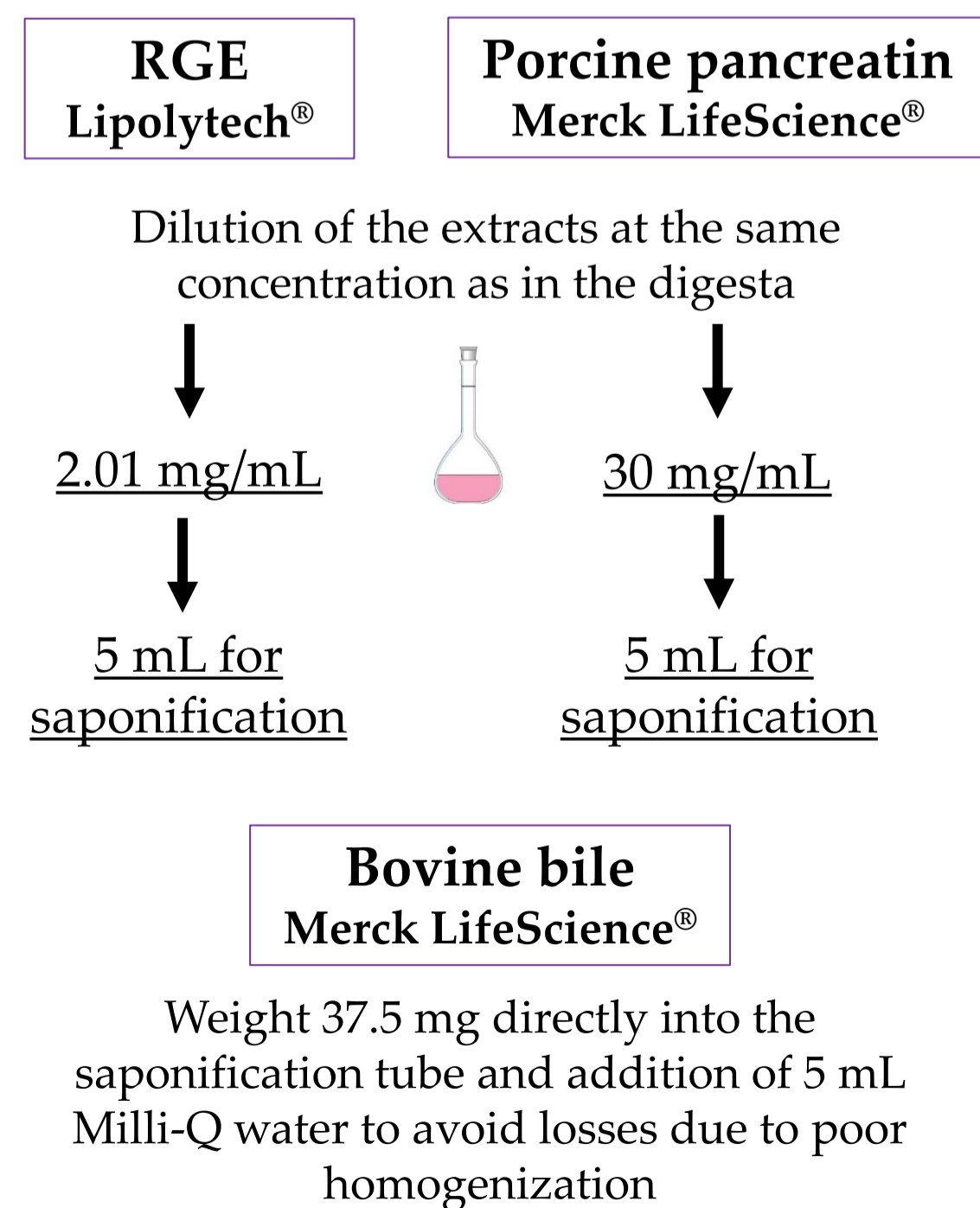
OBJECTIVES

To determine the sterol content of the extracts used in the INFOGEST 2.0 digestion method (rabbit gastric extract – RGE –, porcine pancreatin and bovine bile) and to evaluate their sterol contribution during the digestion.

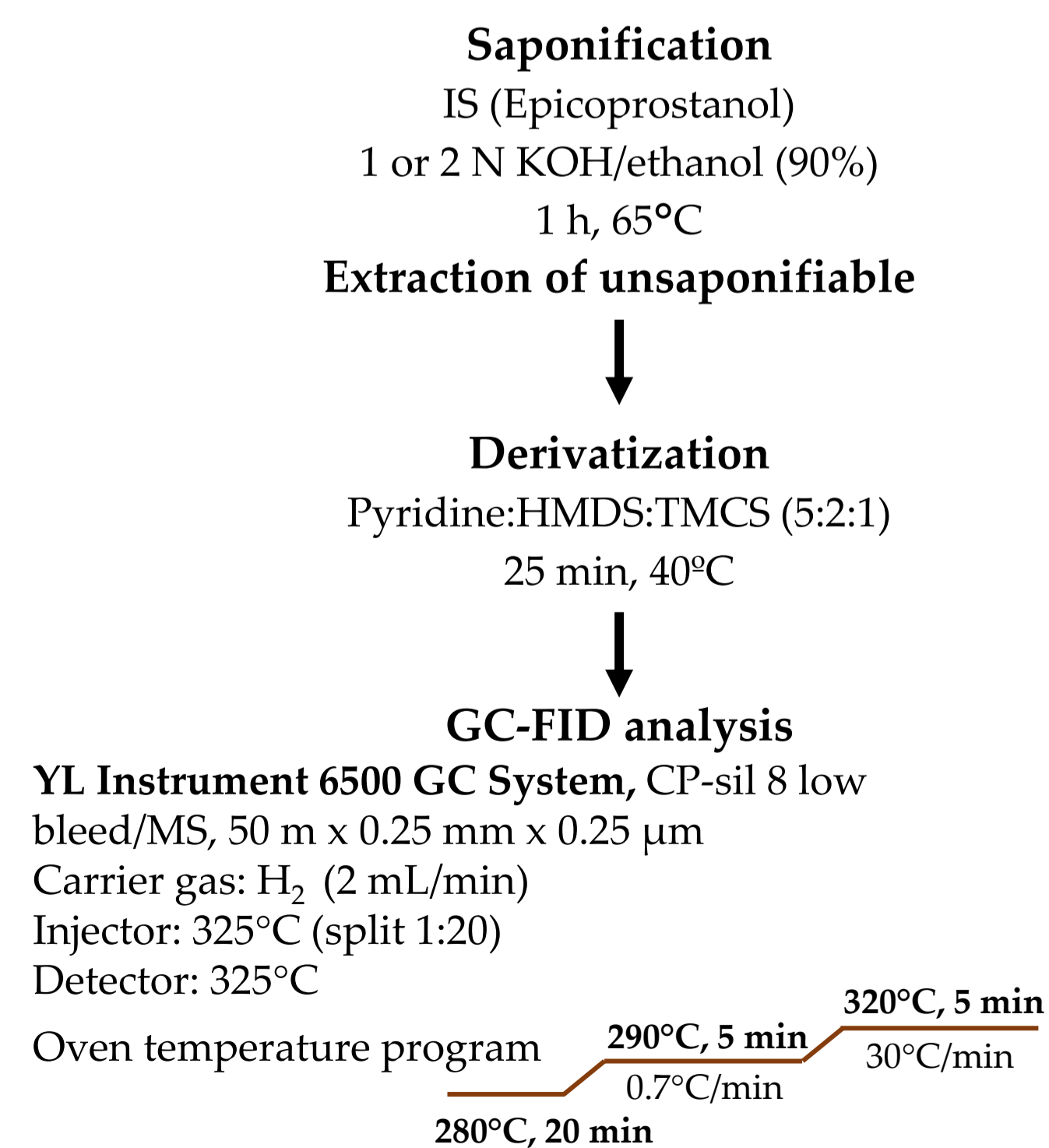
METHODOLOGY

The preparation of the digestive extracts is carried out according to their homogenization capacity, while determination is performed following the methodology indicated by Blanco-Morales et al.¹

Preparation of digestive extracts



Sterol determination



RESULTS

Sterol content

Results show that total sterol content of the extracts is as follows: bile > pancreatin > RGE. Cholesterol is present in all three extracts and PS are found in pancreatin and bile. On the other hand, cholesterol is the major sterol in bile, while β-sitosterol is the main sterol in pancreatin (Table 1).

Table 1. Sterol content in the extracts analyzed.

| | Cholesterol | Campesterol | Stigmasterol | β-Sitosterol | Sitostanol | Total sterols |
|------------|--------------|---------------|--------------|--------------|-------------|---------------|
| | mg/g extract | | | | | |
| RGE | 0.57 ± 0.05 | - | - | - | - | 0.57 ± 0.05 |
| Pancreatin | 1.41 ± 0.07 | 0.103 ± 0.004 | 0.25 ± 0.02 | 2.17 ± 0.42 | 0.32 ± 0.01 | 4.25 ± 0.54 |
| Bile | 4.07 ± 0.18 | - | 0.14 ± 0.01 | 0.93 ± 0.07 | 0.18 ± 0.01 | 5.26 ± 0.25 |

Data are represented as mean ± standard deviation (n=3). RGE: rabbit gastric extract.

Amount of sterols provided by the extracts in the digesta

Pancreatin and bile are the extracts contributing with larger amounts of sterols to digestion, being pancreatin the major contributor of both cholesterol and PS. However, cholesterol in bile is found in pre-formed micelles that increase its solubility and could thus compete for the micellarization of cholesterol provided by the digested food³ (Table 2).

Table 2. Amount of sterols provided by the extracts in the digesta.

| | Cholesterol | Campesterol | Stigmasterol | β-Sitosterol | Sitostanol | Total sterols |
|------------|---------------|-------------|---------------|--------------|---------------|---------------|
| | mg/digesta | | | | | |
| RGE | 0.046 ± 0.004 | - | - | - | - | 0.046 ± 0.004 |
| Pancreatin | 1.72 ± 0.08 | 0.13 ± 0.01 | 0.31 ± 0.03 | 2.64 ± 0.52 | 0.39 ± 0.02 | 5.18 ± 0.66 |
| Bile | 1.00 ± 0.04 | - | 0.035 ± 0.003 | 0.23 ± 0.02 | 0.044 ± 0.003 | 1.30 ± 0.06 |

Data are represented as mean ± standard deviation (n=3). RGE: rabbit gastric extract

CONCLUSION

The characterization of the sterol content in the digestion reagents provides valuable information for the evaluation of sterol bioaccessibility, since changes in the amount of extract added to the digestion could condition the results obtained. Therefore, the sterol content of these digestion reagents must be determined for an accurate estimate of the sterol bioaccessibility.

REFERENCES

¹ Blanco-Morales et al. Food Funct, 2018, 9, 2080-2089; ² López-García et al. 2021, Book of abstracts "Virtual International Conference on Food Digestion", pp 44; ³ Wilson and Rudel. J Lipids Res, 1994, 35, 943-955.

ACKNOWLEDGEMENTS

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