Food & Function

c7fo00536a

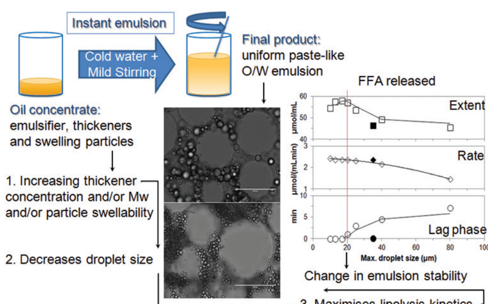
We have presented the Graphical Abstract text and image for your article below. This brief summary of your work will appear in the contents pages of the issue in which your article appears.

1

Instant polysaccharide-based emulsions: impact of microstructure on lipolysis

Amelia Torcello-Gómez* and Timothy J. Foster

The microstructure of instant emulsions is modulated upon mild shearing through specific rheological manipulation of the continuous phase. Finer emulsions display faster lipolysis kinetics under *in vitro* conditions.



Please check this proof carefully. **Our staff will not read it in detail after you have returned it.**

Proof corrections must be returned as a single set of corrections, approved by all co-authors. No further corrections can be made after you have submitted your proof corrections as we will publish your article online as soon as possible after they are received.

Please ensure that:

- The spelling and format of all author names and affiliations are checked carefully. Names will be indexed and cited as shown on the proof, so these must be correct.
- Any funding bodies have been acknowledged appropriately.
- All of the editor's queries are answered.
- Any necessary attachments, such as updated images or ESI files, are provided.

Translation errors between word-processor files and typesetting systems can occur so the whole proof needs to be read. Please pay particular attention to: tables; equations; numerical data; figures and graphics; and references.

Please send your corrections preferably as a copy of the proof PDF with electronic notes attached or alternatively as a list of corrections – do not change the text within the PDF file or send a revised manuscript. Corrections at this stage should be minor and not involve extensive changes.

Please return your **final** corrections, where possible within **48 hours** of receipt, by e-mail to: food@rsc.org. If you require more time, please notify us by email.

Funder information

Providing accurate funding information will enable us to help you comply with your funders' reporting mandates. Clear acknowledgement of funder support is an important consideration in funding evaluation and can increase your chances of securing funding in the future. We work closely with Crossref to make your research discoverable through the Funding Data search tool (http://search.crossref.org/fundref).

Further information on how to acknowledge your funders can be found on our webpage (http://rsc.li/ funding-info).

What is Funding Data?

Funding Data (http://www.crossref.org/fundingdata/) provides a reliable way to track the impact of the work that funders support. We collect funding information from our authors and match this information to funders listed in the Open Funder Registry. Once an article has been matched to its funders, it is discoverable through Crossref's search interface.

PubMed Central

Accurate funder information will also help us identify articles that are mandated to be deposited in PubMed Central (PMC) and deposit these on your behalf.

Providing funder information

We have included the funder information you gave us on submission in the table below. The 'Funder name' shown and their associated 'Funder ID' number is written as listed in the Open Funder Registry. **Please check that the information in the table is correct.** The funder information should match your acknowledgements. This table will not be included in your final PDF but we will share the data with Crossref so that your article can be found via the Funding Data search tool.

Funder name	Funder ID	Award/grant/contract number
FP7 People: Marie-Curie Actions	100011264	326581

If a funding organisation you included on submission of your article is not currently listed in the registry it will not appear in the table above. We can only deposit data if funders are already listed in the Open Funder Registry, but we will pass all funding information on to Crossref so that additional funders can be included in future.

Researcher information

If any authors have ORCID or ResearcherID details that are not listed below, please provide these with your proof corrections. Please check that the ORCID and ResearcherID details listed below have been assigned to the correct author. Please use this space to add your own unique ORCID iDs and not another researcher's, as errors will delay publication.

Please also update your account on our online manuscript submission system to add your ORCID details, which will then be automatically included in all future submissions. See here for step-by-step instructions and more information on author identifiers.

First (given) name(s)	Last (family) name(s)	ResearcherID	ORCID
Amelia	Torcello-Gómez		0000-0003-3276-1296
Timothy J.	Foster		

Queries for the attention of the authors

Journal: **Food & Function** Paper: **c7fo00536a**

Title: **Instant polysaccharide-based emulsions: impact of microstructure on lipolysis**

For your information: You can cite this article before you receive notification of the page numbers by using the following format: (authors), Food Funct., (year), DOI: 10.1039/c7fo00536a.

Editor's queries are marked like this [Q1, Q2, ...], and for your convenience line numbers are indicated like this [5, 10, 15, ...].

Please ensure that all queries are answered when returning your proof corrections so that publication of your article is not delayed.

Query Reference	Query	Remarks
Q1	Please carefully check the spelling of all author names. This is important for the correct indexing and future citation of your article. No late corrections can be made.	
Q2	Ref. 4 and 5: Please provide the name of the patentee(s).	

Food & Function

PAPER

Instant polysaccharide-based emulsions: impact of microstructure on lipolysis

Cite this: DOI: 10.1039/c7fo00536a

Amelia Torcello-Gómez *[a,b] and Timothy J. Foster[b]

The development of emulsion-based products through optimisation of ingredients, reduction in energy-input during manufacture, while fulfilling healthy attributes, are major objectives within the food industry. Instant emulsions can meet these features, but comprehensive studies are necessary to investigate the effect of the initial formulation on the final microstructure and, in turn, on the *in vitro* lipolysis, comprising the double aim of this work. The instant emulsion is formed within 1.5–3 min after pouring the aqueous phase into the oil phase which contains a mixture of emulsifier (Tween 20), swelling particles (Sephadex) and thickeners (hydroxypropylmethylcellulose, HPMC, and guar gum, GG) under mild shearing (180 rpm). The creation of oil-in-water emulsions is monitored *in situ* by viscosity analysis, the final microstructure visualised by microscopy and the release of free fatty acids under simulated intestinal conditions quantified by titration. Increasing the concentration and molecular weight (M_w) of GG leads to smaller emulsion droplets due to increased bulk viscosity upon shearing. This droplet size reduction is magnified when increasing the M_w of HPMC or swelling capacity of viscosifying particles. In addition, in the absence of the emulsifier Tween 20, the sole use of high-Mw HPMC is effective in emulsification due to combined increased bulk viscosity and interfacial activity. Hence, optimisation of the ingredient choice and usage level is possible when designing microstructures. Finally, emulsions with larger droplet size (>20 μm) display a slower rate and lower extent of lipolysis, while finer emulsions (droplet size ≤20 μm) exhibit maximum rate and extent profiles. This correlates with the extent of emulsion destabilisation observed under intestinal conditions.

Received 6th April 2017,
Accepted 7th June 2017

DOI: 10.1039/c7fo00536a

rsc.li/food-function

1. Introduction

From a foods manufacturing perspective, a thorough understanding of the formulation and process design is required to prepare products with defined attributes. Almost all emulsion-based food products are sold in a prepared format, with the consequent need to distribute as such, ensuring both storage and microbiological stability, the latter typically through thermal processing routes (pasteurisation/UHT treatment) and/or chilled/frozen distribution. To overcome the need to distribute water, ensure preservation and stability and offer consumers lower-cost alternatives, dried food products were developed.[1] This opened the field for the development of more functional, end-user friendlier systems such as instant powders for the preparation of dressings, beverages, soups/gravies, *etc*. Additionally, re-hydratable powdered toppings are available where crystallised fat-containing emulsions are spray dried, creating texture upon rehydration.[2] Nonetheless, the control over the final emulsion properties is not warranted for such systems, spray drying can have a negative impact on the quality of the product,[3] and sophisticated processing is required. As a solution to all these issues, the concept of instantly structured emulsions was introduced and patented[4,5] as promising systems for the food industry representing important environmental and financial benefits. In a broad sense, an instant emulsion is formed by addition and mixing of cold water to an oil phase which contains a mixture of thickening biopolymers and emulsifiers. Hence, the final product is not going to be simply obtained by dilution or dissolution of dry material in hot water, like the instant powders, or by hydration of a spray-dried emulsion, ensuring controllable final characteristics. The technological advantage of this approach is that the emulsion is created within a reduced time frame and stabilised in the presence of, and solely by, the constituent ingredients, with a minimum energy input, such as moderate shear generated by hand stirring. Specifically, the time taken for emulsification will depend on the relative rates of hydration of the surface active component and thickening/gelling ingredients, which will also have an effect on the bulk structuring of the water phase to kinetically trap the dispersed oil phase.[6] Efficient viscosifying can be also achieved by swell-

[a]School of Food Science and Nutrition, University of Leeds, Leeds, LS2 9JT, UK. E-mail: M.A.TorcelloGomez@leeds.ac.uk
[b]Division of Food Sciences, School of Biosciences, University of Nottingham, Sutton Bonington Campus, Loughborough, LE12 5RD, UK

This journal is © The Royal Society of Chemistry 2017

ing of particles, such as pregelatinised or cold water swelling starch and citrus fibres. The final product is a stable oil-in-water (O/W) emulsion with a paste-like texture, whose properties depend on: emulsifier and thickener type and concentration, pH and ionic strength of the aqueous phase, oil phase type and concentration, *etc*.

Regarding the emulsifier, Tween 20 (polyoxyethylene sorbitan monolaurate) has been used in this study due to its efficiency in stabilising food emulsions with reduced amount as compared to lecithins.[6] Two different hydrocolloids were used as thickeners: hydroxypropylmethylcellulose (HPMC), which has not previously been used to stabilise instant emulsions as the main thickener, and guar gum (GG) to aid increasing bulk viscosity. Finally, cross-linked dextran particles were used as swelling agents to mimic the swelling behaviour of *e.g.* cold water swelling starch. Starch has been used in the past as swelling particulates of these systems, but does not provide control over the swelling capacity in the presence of hydrocolloids.[7] In fact, previous work was dedicated on the competitive hydration of cold water swelling starch and hydrocolloids but no emphasis was laid on the effect on the final emulsion microstructure.[6] For that reason, this manuscript is particularly focused on the effect of the presence of the emulsifier, to understand the interfacial stabilisation, the molecular weight and concentration of the thickeners, as well as the swelling capacity of particles on the final emulsion characteristics. The systematic study on how these specific parameters affect the final microstructure of the instant emulsion has not been reported yet to the best of our knowledge. The variation in pH and ionic strength in the aqueous phase is not relevant in this case since the considered emulsifier and thickeners are non-ionic and the swelling particles have a wide range of pH stability (pH 2–10).

The thickeners used in this study are polysaccharides and more generally categorised as soluble dietary fibre. The advantage of using such ingredients is two-fold. They are incorporated in food during the processing to improve the nutritional properties,[8,9] such as fat or sugar replacers, as well as the technological and sensory characteristics, such as viscosity, texture and shelf-life. Furthermore, dietary fibre *per se* has beneficial metabolic and physiological effects on its transit within the gastrointestinal tract.[10,11] They are known to lower blood cholesterol[12,13] and prevent lipid absorption.[14–16] It has been recently reported on the potential of polysaccharides to control lipid digestion either as stabilisers added to the continuous phase of the formed O/W emulsions,[17–20] or as emulsifiers themselves.[21,22] Possible mechanisms to explain these features are: interactions between polysaccharides and physiological components in the continuous phase,[19,23,24] droplet flocculation, thus reducing the oil–water interface available for the enzymatic reactions to take place,[20] and/or adsorption of polysaccharides onto the oil–water interface delaying the action of physiological components to hydrolyse the lipids[22] and subsequent absorption in the intestinal mucosa.

Thus, all these properties ascribed to polysaccharides can be applied to develop healthier food emulsions, by regulating the digestion of oils and fats in the body. This strategy is preferred when reducing the total fat content makes difficult food formulation.[25] This study seems appropriate when the market for high quality food products that are viewed as healthy is increasing because of the growing interest in the preventative role of the diet against long-term chronic disease. Precisely, the increased fat intake per individual on daily basis during the past few decades has been linked to rises in obesity, cardiovascular and related diseases.[26] The last section of this manuscript is in fact devoted to evaluate the impact of the initial emulsion microstructure on the rate and extent of lipid digestion in simulated intestinal conditions, and how the manipulation of the microstructure during the processing of the instant product can modulate this process, which will provide an added value not explored yet in these systems.

The knowledge generated by this study will contribute to the rational design of functional instant food emulsions minimising ingredient, process, supply costs, avoiding excessive use of additives currently used for extended shelf life of products, and hence providing healthier, affordable, and ready-to eat options.

2. Materials and methods

2.1. Materials

All the chemicals were used as received, unless stated otherwise. Tween 20 (P1379) from Sigma-Aldrich was used as non-ionic emulsifier ($M_w \sim 1228$ Da). Two different hydrocolloids were used as non-ionic thickeners: HPMC and GG. Two different molecular weight samples were used for each of them. The low-molecular weight (LMw) HPMC, H9262, was purchased from Sigma-Aldrich (M_w range: 22–26 kDa). The high-molecular weight (HMw) HPMC, METHOCEL™ K4M, was kindly provided by the Dow Chemical Company (M_w range: 300–500 kDa). The levels of incorporation of the two substituents are 19–24% methoxyl and 7–12% hydroxypropyl in both HPMC samples. The GG samples were supplied from Danisco: Meyprodor® 30 ($M_w \sim 420$ kDa) and 400 ($M_w \sim 2660$ kDa). Two cross-linked dextran samples from GE Healthcare were used as swelling particles with different wet bead particle size: Sephadex™ G-50 Fine (40–160 μm) and G-100 (100–310 μm).

The small intestine juice was prepared with lipase from porcine pancreas (L3126), Type II (100–400 units per mg protein, using olive oil with 30 min incubation), porcine bile extract (B8631), $CaCl_2$ and NaCl of analytical grade, all of them purchased from Sigma-Aldrich.

The aqueous phase of emulsions and duodenal juice was 2 mM BIS-TRIS (Sigma-Aldrich, ≥99.0% purity) adjusted to pH 7 with HCl. Ultrapure water purified in a Pur1te Select system was used for buffer preparation.

Highly refined olive oil (Sigma-Aldrich) was purified with activated magnesium silicate (Florisil®, Fluka) to eliminate free fatty acids and surface active impurities: a mixture of oil and Florisil® (2 : 1 wt/wt) was shaken mildly for 3 h and centri-

 This journal is © The Royal Society of Chemistry 2017

fuged at 4000 rpm for 30 min. It was then stored away from light.

2.2. Emulsion preparation and viscosity measurements

The emulsifier (0–0.18 g), thickeners (0–0.18 g) and swelling particles (0.9 g) were first homogeneously dispersed in the olive oil (2 g oil). Next, the required amount of aqueous phase (18 g) was added to this slurry and placed immediately into a Rapid Visco Analyser (RVA) Super 4 (Newport Scientific, Australia). The viscosity development was measured at constant temperature (25 °C) and shear rate (180 rpm) for 30 min. The oil concentration was fixed at 10 wt% in the emulsion. The individual concentration of emulsifier/thickeners ranged from 0 to 1 wt% in the aqueous phase, whereas the concentration of the swelling particles was kept constant at 5 wt% (dry) in the aqueous phase.

This emulsion preparation procedure produces lipid droplets within the size range found in many food products (d = 1–100 μm) and that can be observed with optical microscopy.

2.3. Optical microscopy

Emulsions samples were immediately observed at the microscope after preparation in the RVA and after *in vitro* lipolysis. A drop of emulsion was placed on a microscope slide and covered by a cover slip. The microstructure of instant emulsions was visualised with a digital inverted microscope (EVOS fl, AMG) using a 20× and 40× magnification objectives. The images were acquired using an integrated Sony CCD camera and software. Given the apparent and relative uniformity of droplet size distribution, the maximum oil droplet diameter was determined for each sample by multiple-image analysis to evaluate differences in emulsion microstructure.

2.4. Lipolysis of instant emulsions

The release of free fatty acids (FFA) from the instant emulsions due to pancreatic lipase activity was monitored using a titration method which was slightly adapted from a previous study.[22] This static *in vitro* digestion model comprises a simplification of a standardised method,[27] as a preliminary study focused only on intestinal lipolysis. A 20 mL sample of freshly prepared emulsion (pH = 7.0) was placed in a water bath at 37 °C and left to stabilise the temperature. Then, 1 mL of $CaCl_2$ and NaCl solution (containing 27.7 mg $CaCl_2$ and 219.2 mg NaCl) followed by 3 mL of bile extract solution (containing 125 mg bile extract) were added to the emulsion under stirring (400 or 700 rpm for emulsion viscosity values lower or higher than 1200 cP, respectively) and then the system was adjusted back to pH 7 if required. Next, 1 mL of freshly prepared lipase suspension (containing 40 mg lipase powder) was added to the above mixture and the titration started. The final concentration of pancreatic lipase was 1.6 mg mL^{-1}, whereas for bile extract was 5 mg mL^{-1}. $CaCl_2$ and NaCl had a final concentration of 10 mM and 150 mM, respectively. The lipolysis was measured by a pH-stat automatic titration unit (702 SM Titrino, Metrohm) by titrating appropriate amounts of NaOH solution (0.1 M) to maintain the pH at 7.0. The concentration of FFA generated by lipolysis was calculated from the volume of NaOH added, that is the number of moles of NaOH required to neutralize the FFA (assuming 2 FFA produced per 1 triacylglycerol molecule). The rate of lipolysis was determined as the maximum slope of the linear portion of the kinetics profile, the initial lag phase as the intercept of the maximum slope with the x-axis and the extent as the value in the final plateau.

2.5. Data analysis

Results are represented as the mean and standard deviation values of three measurements made on different freshly prepared instant emulsion samples, respectively. The error bars are contained within the size of the symbols of data points given the good reproducibility of the viscosity and lipolysis measurements.

3. Results and discussion

3.1. Development of instant emulsions: final microstructure

3.1.1. Effect of thickener concentration and molecular weight. The first section is focused on the impact of increasing either the concentration and/or the molecular weight of at least one hydrocolloid on the droplet size and viscosity of the instant emulsions. In this set of experiments, the emulsifier (Tween 20) concentration was fixed to 1 wt%, as well as the type and concentration of the swelling particles: 5 wt% (dry) Sephadex G-100.

Fig. 1a and 2a show the viscosity development of the instant emulsions upon formation and their final microstructure reached, respectively. These instant emulsions were formed using 1 wt% LMw HPMC and 0–1 wt% GG as thickeners. As a general trend, Fig. 1a exhibits a steep increase in viscosity, which after approximately 3 min or less becomes fairly constant reaching a plateau. The initial increase in viscosity, just after the addition of the aqueous phase to the oil slurry, is a combination of the competitive hydration of the different components and droplet formation. The hydration of HPMC and GG increases the viscosity of the aqueous phase which along the swelling of the cross-linked dextran particles aids oil droplet break-up under shearing (180 rpm). The role of the emulsifier Tween 20 is to rapidly adsorb onto the oil–water interface reducing the interfacial tension and facilitating the droplet break-up. The instant emulsions seem to be completely developed after 3 min, when appreciable changes are no longer observed in the viscosity over time. The gradual continuous increase in viscosity afterwards may be due to further hydration of the swelling particles and/or the hydrocolloids. Fig. 2a displays the micrographs of the instant emulsions taken after 30 min of shearing in the RVA. As common features, the swelling particles appear as larger (~200 μm on average) and lighter spheres, forming a matrix filled by the smaller (≤40 μm) and darker oil droplets. Increasing the GG concentration gives rise to a reduction of emulsion droplet size, regardless of the GG molecular weight for systems made with 1 wt% LMw HPMC. Focusing first on LMw GG, the

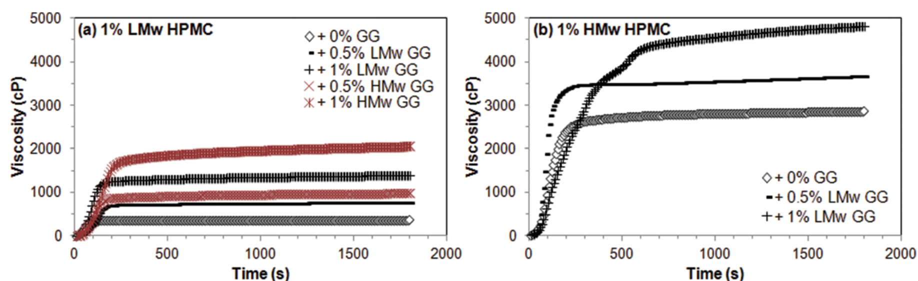


Fig. 1 Viscosity measurements during the formation of 10 wt% olive O/W instant emulsions containing: 1 wt% Tween 20, 5 wt% Sephadex G-100, 1 wt% low- (a) or high- (b) Mw HPMC and 0–1 wt% low- or high-Mw GG.

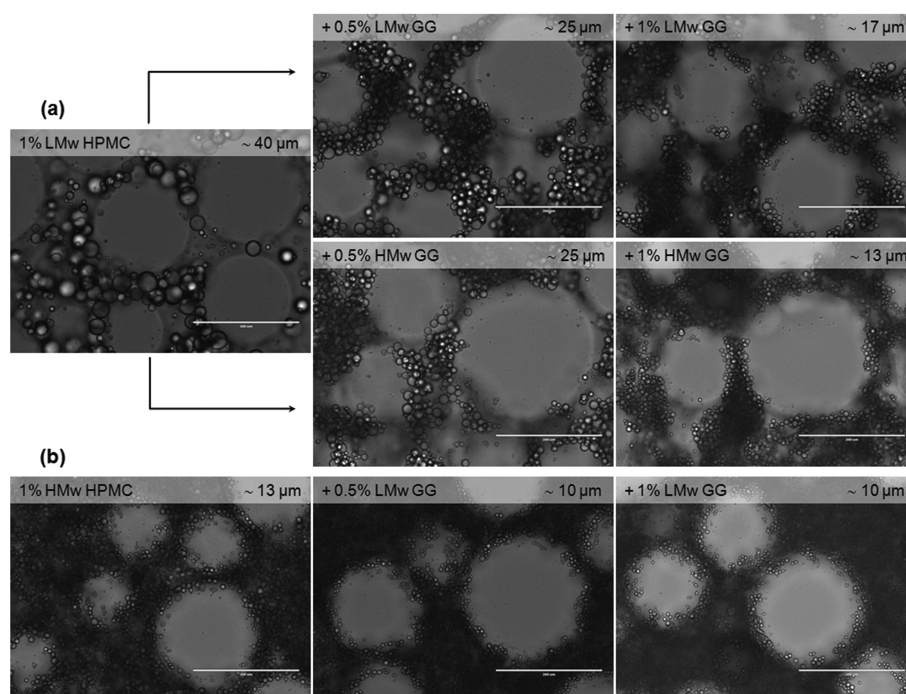


Fig. 2 Microstructure of 10 wt% olive O/W instant emulsions containing: 1 wt% Tween 20, 5 wt% Sephadex G-100, 1 wt% low- (a) or high- (b) Mw HPMC and 0–1 wt% low- or high-Mw GG. Scale bars are 200 μm. Maximum oil droplet size is indicated at the top right corner of each image. Larger brighter and smaller darker particles correspond to Sephadex swollen particles and oil droplets, respectively.

maximum diameter of oil droplets is reduced from 40 μm to 25 μm and 17 μm at 0.5 wt% and 1 wt% LMw GG, respectively. A greater aqueous phase viscosity provided by higher GG concentration enhances the droplet break-up under shearing, reducing the oil droplet size,[6] which is also reflected in higher emulsion viscosity plateau values in Fig. 1a.[28] Next, increasing the molecular weight of GG results in a slight increase in emulsion viscosity as observed in Fig. 1a. This increase in emulsion viscosity is only more appreciable at a GG concentration of 1 wt%. The emulsion microstructure shown in Fig. 2a is very similar in the presence of both high- and low-Mw GG. Only at 1 wt% HMw GG there is an appreciable reduction (1.3 times) of the maximum droplet size (13 μm), as compared to 1 wt% LMw GG (17 μm), which may contribute to the higher emulsion viscosity plateau obtained in Fig. 1a.

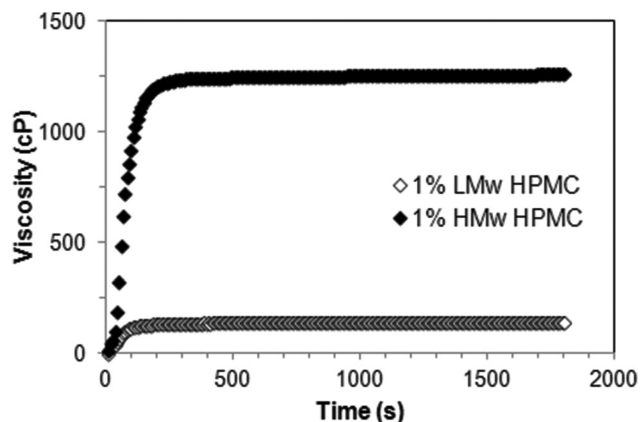
Fig. 1b and 2b show the results of viscosity and microstructure, respectively, for instant emulsions stabilised with 1 wt% HMw HPMC and 0–1 wt% LMw GG. Increasing the molecular weight of HPMC has a dramatic effect on the emulsion viscosity and oil droplet size of instant emulsions. In the absence of GG, the emulsion viscosity raises up to nearly 3000 cP (Fig. 1b), one order of magnitude larger than the viscosity of the emulsion stabilised by LMw HPMC (Fig. 1a). Regarding the oil droplet size, the maximum diameter is 13 μm (Fig. 2b), three times smaller than that found for the emulsion stabilised by LMw HPMC (Fig. 2a). Hence, an emulsion microstructure and viscosity similar to the system stabilised by 1 wt% LMw HPMC and 1 wt% HMw GG (Fig. 1a and 2a) have been achieved by solely using HMw HPMC as the only thickener and half of the total hydrocolloid concentration. The reason

This journal is © The Royal Society of Chemistry 2017

for obtaining slightly higher emulsion viscosity in the latter (~3000 *vs.* 2000 cP) may come from an increased viscosity of the continuous phase due to the higher molecular weight of HPMC or subtly finer droplet size distribution. Indeed, a more viscous aqueous phase would improve the droplet break-up on shearing,[29] in agreement with the microstructure seen in Fig. 2b. In addition, HPMC is known to reduce the interfacial tension, which may act cooperatively with the emulsifier Tween 20. However, it has been reported previously that the interfacial activity of cellulose ethers, in terms of steady values of interfacial tension, does not depend on their molecular weight but on their hydrophobicity.[30] Therefore, similar interfacial activity is expected for both LMw and HMw HPMC.[22] Thus the effect of the continuous phase viscosity is possibly the main reason to explain the great impact of the HPMC molecular weight on the features of the instant emulsions. The fact that increasing the molecular weight of GG (Fig. 1a) does not have such a dramatic effect on the droplet size and emulsion viscosity as compared to HPMC may be due to its lower apparent bulk viscosity related to its molecular structure: short-side branched (GG) *vs.* linear (HPMC).[31,32] Finally, in the presence of GG, the emulsion viscosity further increases (Fig. 1b). However, at the highest GG concentration, the steady state viscosity is attained after 10 min and there is a two-stage process. This delay in viscosity development may be explained by a more complex competition for water during hydration between the hydrocolloids and the swelling dextran particulates.[6] In turn, this may account for the similar final microstructure observed in Fig. 2b, with a maximum droplet diameter of 10 μm, at either 0.5 or 1 wt% LMw GG. It seems that a saturation has been achieved in the initial fast thickening of the aqueous phase since higher GG concentration does not promote droplet size reduction, despite having some interfacial activity.[33] Consequently, the highest emulsion viscosity value observed in Fig. 1b in the presence of 1 wt% LMw GG (nearly 5000 cP) may be due to the increased continuous phase viscosity after prolonged period of hydrocolloids and particles hydration.

3.1.2. Effect of wet size of swelling particles. The influence of the matrix volume on the emulsion characteristics is evaluated by reducing the wet size of the swelling particles, thereby increasing the matrix volume available for the formation of oil droplets. To this end, Sephadex G-50 is used in this set of experiments at the same fixed concentration of 5 wt% (dry) as for G-100 above. According to the supplier, the bed volume ranges, in distilled water, are 9–11 and 15–20 mL g^{-1} for Sephadex G-50 and G-100, respectively. This means that the volume occupied by Sephadex G-50 particles is almost half of the volume occupied by Sephadex G-100 particles, per unit of initial dry weight. The emulsifier Tween 20 and thickener HPMC are used at the fixed concentration of 1 wt%.

Fig. 3 shows the viscosity results and micrographs of instant emulsions stabilised by either low- or high-Mw HPMC as the only thickener and Sephadex G-50. Additionally, Table 1 summarises the formulation and characteristics of the instant emulsions studied in all sections for easy comparison. In order to look at the effect of the wet particle size of the cross-linked dextran, these results are compared with those of the emulsions stabilised by Sephadex G-100 and HPMC (in the absence of GG) in section 3.1.1. (Fig. 1, 2 and Table 1). Increasing the matrix volume, that is decreasing the swelling capacity of Sephadex, leads to an increase in the emulsion droplet size and corresponding decrease in emulsion viscosity. Indeed, on average the wet particle size of Sephadex G-50 is smaller (60–180 μm) than that found for Sephadex G-100 par-



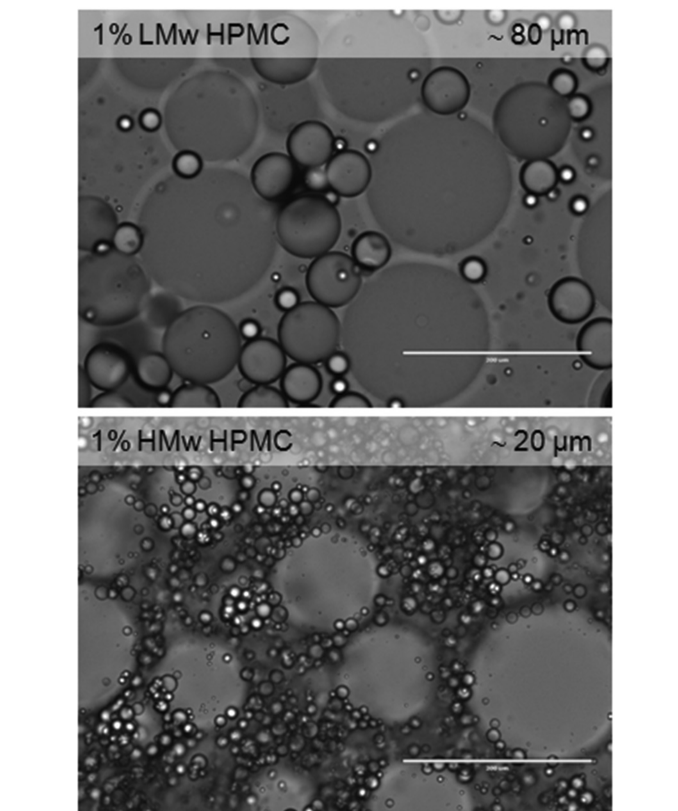


Fig. 3 Viscosity measurements and microstructure of 10 wt% olive O/W instant emulsions containing: 1 wt% Tween 20, 5 wt% Sephadex G-50, 1 wt% low- or high-Mw HPMC. Scale bars are 200 μm. Maximum oil droplet size is indicated at the top right corner of each image. Larger brighter and smaller darker particles correspond to Sephadex swollen particles and oil droplets, respectively.

Table 1 Summary of emulsion formulation (constituents of the continuous phase) and emulsion characteristics: time to reach steady viscosity ($t_{\eta st}$), final emulsion viscosity (η) and maximum droplet diameter (max. d)

Constituents of the aqueous phase (wt%)				Emulsion characteristics		
Tween 20	Cross-linked dextran particles	HPMC	GG	$t_{\eta st}$ (min)	η (cP)	Max. d (μm)
1	Lower swelling capacity (G-50)	LMw	—			
	5	1	0	2	140	80
		HMw	—			
		1	0	2.8	1300	20
	Higher swelling capacity (G-100)	LMw	LMw			
	5	1	0	2	380	40
			0.5	2.7	770	25
			1	2.7	1400	17
			HMw			
			0	2	380	40
			0.5	2.7	980	25
			1	3	2040	13
		HMw	LMw			
		1	0	3	2900	13
			0.5	3	3600	10
			1	10	4800	10
0	Higher swelling capacity (G-100)	HMw	—			
	5	1	0	5.5	3900	35

ticles (100–300 μm), in agreement with the wet size ranges provided by the supplier (see section 2.1.). In the presence of Sephadex G-50 (Fig. 3), the maximum oil droplet size found for the system stabilised by low- and high-Mw HPMC is 80 and 20 μm, respectively, in contrast to diameter values measured in the presence of Sephadex G-100: 40 and 13 μm, respectively. In this case, reducing the volume occupied by the swelling cross-linked dextran particles by half increases approximately twice the maximum droplet size of the emulsion, giving rise to a decrease in the measured plateau viscosity, as reflected in Fig. 3 in comparison with Fig. 1. Specifically, the steady state emulsion viscosity was reduced from *ca.* 380 to 140 cP and from 2900 to 1300 cP, when the systems are stabilised by low- and high-Mw HPMC, respectively (Table 1). It is also worth noting that increasing the M_w of HPMC in the presence of Sephadex G-50 decreases four times the emulsion droplet size upon shearing, which supports the great impact of this thickener on the aqueous phase viscosity to aid in droplet break-up upon shearing, as previously discussed. Therefore, a combined efficient thickening of the aqueous phase through fast hydration of polymers of higher molecular weight and higher swelling capacity of particles leads to a better emulsification during mild shearing due to, on one hand, a higher viscosity gradient between the dispersed and continuous phase and, on the other hand, and reduced gap between swollen particles so that the total hydrodynamic force is enhanced promoting oil droplet break-up.

3.1.3. Effect of the emulsifier. Finally, the role played by the emulsifier Tween 20 is analysed through a set of experiments where Tween 20 is absent in the emulsion formulation. Sephadex G-100 was used (5 wt% dry) as swelling particles and HMw HPMC (1 wt%) as the only thickener due to their better emulsification capacity, that is, ability to produce smaller oil droplet size.

Fig. 4 displays the results of viscosity development upon emulsification and corresponding final microstructure of the produced instant emulsion. The viscosity profile exhibits a lag phase of approximately 3 min after which the viscosity steeply increases up to nearly 4000 cP. The lag phase may suggest that the absence of Tween 20 delays the droplet formation. On the other hand, the relatively high steady state viscosity value might imply that the final instant emulsion contains tiny oil droplets. Nonetheless, the microstructure observed in Fig. 4 rules out this presumption. The maximum droplet size achieved is 35 μm, which provides an emulsion microstructure similar to that obtained for the system stabilised by Tween 20 and LMw HPMC together, in Fig. 2a. To recall, the maximum oil droplet size observed for the latter is 40 μm (Table 1). This means that the sole presence of HMw HPMC in the formulation of instant emulsions, besides the swelling particles, is enough to produce oil droplets even smaller than formulations including Tween 20 and LMw HPMC. Although the timeframe required for developing a steady viscosity, given by the onset of the viscosity plateau, is longer in the former (5.5 min *vs.* 2 min) (Table 1). The ability of HPMC to lower the interfacial

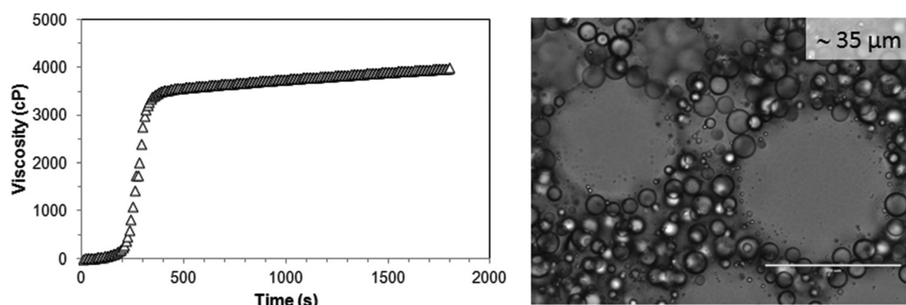


Fig. 4 Viscosity measurements and microstructure of 10 wt% olive O/W instant emulsions containing: 5 wt% Sephadex G-100, 1 wt% high-Mw HPMC. Scale bar is 200 μm. Maximum oil droplet size is indicated at the top right corner of the image. Larger brighter and smaller darker particles correspond to Sephadex swollen particles and oil droplets, respectively.

 This journal is © The Royal Society of Chemistry 2017

tension may contribute to the emulsion formation. It was previously reported interfacial tension values of 16 mN m^{-1} at the olive oil–water interface saturated with the same HMw HPMC, at room temperature.[30] The interfacial tension reported for Tween 20 under equivalent conditions is lower than 4 mN m^{-1}.[34] Therefore, despite the powerful interfacial activity of Tween 20, the increased aqueous phase viscosity provided by HMw HPMC is definitively crucial to reduce the droplet size upon emulsification in combination with its interfacial activity. It seems that the delayed droplet formation in the absence of the emulsifier allows a more complete hydration of both swelling particles and HPMC, which may lead to a higher bulk viscosity, aiding in droplet break-up to form the emulsion. This enhanced continuous phase viscosity would also contribute to the final measured emulsion viscosity in Fig. 4 (~4000 cP), which is slightly higher than that observed in the presence of Tween 20 in Fig. 1b (~3000 cP) for a finer emulsion (Table 1).

3.2. Lipolysis of instant emulsions

Fig. 5 shows the lipolysis profiles of instant emulsions presented in Fig. 1 and 2 after being subjected to duodenal conditions for 2 h. To recall, in this set of experiments the emulsifier (Tween 20) concentration was fixed to 1 wt%, as well as the type and concentration of the swelling particles: 5 wt% (dry) Sephadex G-100, whereas 1 wt% of either low- or high-Mw HPMC and 0–1 wt% GG, were used as thickeners. As a general trend, there is an initial steep increase in the amount of FFA released after which a steady value is reached. The reason for this saturation on the extent of lipolysis, even though all of the lipids have not been digested (14–25%), may be because of the inhibition of lipase activity by the more surface active free fatty acids and monoglycerides present at the droplet surfaces.[35,36] In addition, the presence of emulsifier at the oil–water interface that is resistant to the action of bile salts will definitely affect the rate and extent of lipase activity.[37,38] The presence of an initial lag phase is usually ascribed to the time taken for the surface active components in the bile extract to adsorb and displace the initial emulsifier from the droplet surfaces and hence promote the lipase anchorage to the oil–water interface for the enzymatic reaction to take place.[39] Considering first Fig. 5a, the addition of increasing GG concentration leads to decreasing initial lag phase, faster rate and higher extent of the lipid digestion. The trend of lipolysis curves is similar when using both low- and high-Mw GG, for that reason the results for LMw GG are not shown in Fig. 5a. It was previously reported that increasing the polysaccharide bulk concentration added to the original emulsion reduces the rate and extent of digested lipids[20] in contrast to the observed tendency here. In the current case, increasing GG concentration up to 1 wt% gives rise to smaller emulsion droplets upon emulsification, from 40 to 17 μm or even 13 μm for low- or high-Mw GG, respectively, as seen in section 3.1.1. (Fig. 2a). It is known that increasing the emulsion interfacial area by decreasing the oil droplet size, enhances the digestion of emulsified lipids since larger oil–water interfacial area is available for the enzyme lipase to attach on.[37,40,41] Therefore, the effect of the droplet size on the digestion of emulsified oil prevails over the effect of increasing the hydrocolloid bulk concentration. However, the fact that the lipolysis profiles of emulsions stabilised by 1 wt% low- or high-Mw GG overlap suggests that decreasing the droplet size beyond ~17 μm does not bring further changes in the release of FFA for these systems.

Next, in Fig. 5b, the M_w of HPMC was increased in relation to Fig. 5a. The lipolysis curve in the absence of GG completely overlaps with that of the emulsion stabilised by 1 wt% LMw HPMC and 1 wt% HMw GG in Fig. 5a, which may be attributed to the similar microstructure: maximum diameter 13 μm (Fig. 2). Therefore, it seems that increasing the hydrocolloid bulk concentration does not have an impact on the lipolysis kinetics when also considering similar emulsion droplet size. The addition of GG to the instant emulsion stabilised by HMw HPMC does not affect the release of FFA over time (Fig. 5b), despite the initial oil droplet size being slightly reduced to 10 μm (Fig. 2b). This is consistent with the previous observation: decreasing the initial droplet size beyond ~17 μm does not bring further changes in the lipolysis profile of the emulsion. The *in vitro* digestion of the emulsion stabilised with HMw HPMC and 1 wt% LMw GG was not reliable due to its high emulsion viscosity (nearly 5000 cP), which does not allow homogenising the system under stirring to properly dispersed the NaOH added during the titration. Nonetheless, the emul-

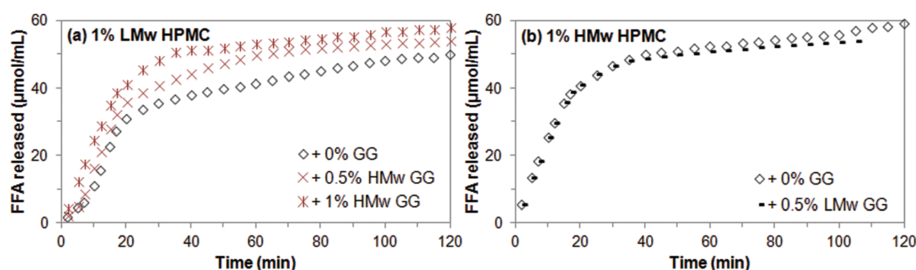


Fig. 5 Fatty acids released from 10 wt% olive O/W instant emulsions containing: 1 wt% Tween 20, 5 wt% Sephadex G-100, 1 wt% low- (a) or high- (b) Mw HPMC and 0–1 wt% low- or high-Mw GG, during 2 h of lipolysis after adding the duodenal juice under physiological conditions (T = 37 °C, pH 7.0).

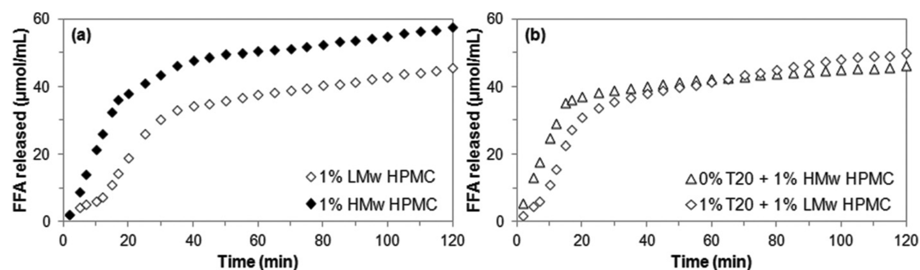


Fig. 6 Fatty acids released from 10 wt% olive O/W instant emulsions containing: 1 wt% Tween 20, 5 wt% Sephadex G-50 (a); 0–1 wt% Tween 20, 5 wt% Sephadex G-100 (b) and 1 wt% low- or high-Mw HPMC, during 2 h of lipolysis after adding the duodenal juice under physiological conditions (T = 37 °C, pH 7.0).

sion microstructure provided by this formulation is similar to that containing 0.5 wt% LMw GG (Fig. 2b), whose lipolysis profile has been just discussed.

Fig. 6a focuses on the lipolysis profiles of instant emulsions presented in Fig. 3, which were stabilised by 1 wt% Tween 20, 5 wt% (dry) Sephadex G-50 and 1 wt% low- or high-Mw HPMC as the only thickener. In the case of the system stabilised by LMw HPMC, the rate and extent of lipid digestion are appreciably lower than those found in Fig. 5a when Sephadex G-100 is present in the emulsion formulation. In addition, the initial lag phase is longer in the former (7 *vs.* 4 min). This can be attributed again to large differences in droplet size for both systems: 80 and 40 µm, respectively. However, for the system stabilised by HMw HPMC, the lipolysis profile shows subtle differences as compared to the system including Sephadex G-100 in Fig. 5b. Namely the rate and extent of released FFA are only slightly lower in the former, which on one hand agrees with the fact that the maximum droplet size is larger (20 *vs.* 13 µm). Nonetheless, the differences in the lipolysis profile due to different emulsion droplet size are considerably small. This feature may be related with the working range of the maximum droplet size for these systems. To recall from Fig. 5, the effect of the emulsion droplet size on the delay of lipolysis was only appreciable up to the range of ~17 µm. Instant emulsions with smaller oil droplets did not show visible changes in the release of FFA.

Fig. 6b corresponds to the *in vitro* digestion experiments of the instant emulsion shown in Fig. 4. The set of experiments were performed in the absence of Tween 20 in the emulsion formulation. Sephadex G-100 was used (5 wt% dry) as swelling particles and HMw HPMC (1 wt%) as the only thickener. For comparison, Fig. 6b also includes the lipolysis curve of the system containing Tween 20 and HPMC in the formulation, with emulsion droplet size within the similar range (35–40 µm). In this case, the differences observed in the initial lipolysis rate could be ascribed to differences in the droplet size. To recall, in the absence of Tween 20, slightly smaller droplets were found (35 µm), hence this would correspond to higher rate of digestion. However, the lipolysis profile in the absence of Tween 20 does not follow the trend found so far in the lag phase and extent *vs.* maximum droplet size, which is illustrated in Fig. 7. This can be explained by the different

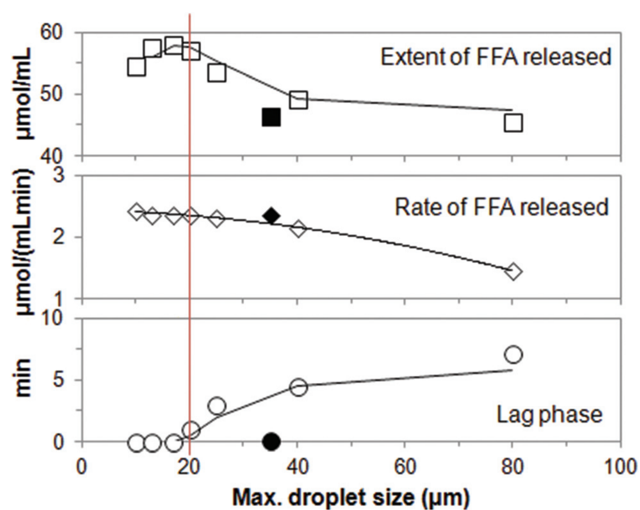


Fig. 7 Lag phase, rate and extent of fatty acids released, determined from lipolysis profiles of Fig. 5 and 6, as a function of the maximum droplet size of the instant emulsions. Closed symbols correspond to instant emulsion from Fig. 6b with no Tween 20 in the formulation. Lines are plotted as a guide for the eye. The vertical line indicates an inflection point.

interfacial composition, which needs to be considered here. The lack of the initial lag phase would lead to think that the sole presence of HPMC at the oil–water interface does not hinder the digestion of the lipid substrate in contrast to an interface mainly stabilised by Tween 20, but interestingly, the final extent seems to be lower than in the presence of Tween 20. This may suggest that eventually the adsorbed interfacial layer of HPMC better resists the displacement by the duodenal components than the emulsifier Tween 20.[22]

The microstructure of the instant emulsions was also evaluated after being subjected to *in vitro* lipolysis. Some representative micrographs are illustrated in Fig. 8, corresponding to all samples discussed above, in decreasing order of the initial maximum droplet size. An interesting pattern has been observed throughout the samples studied here. The emulsion droplet size increased on average after 2 h of lipid digestion for emulsions which originally had oil droplets larger than ~20 µm. This is the case of emulsions stabilised by 1 wt%

 This journal is © The Royal Society of Chemistry 2017

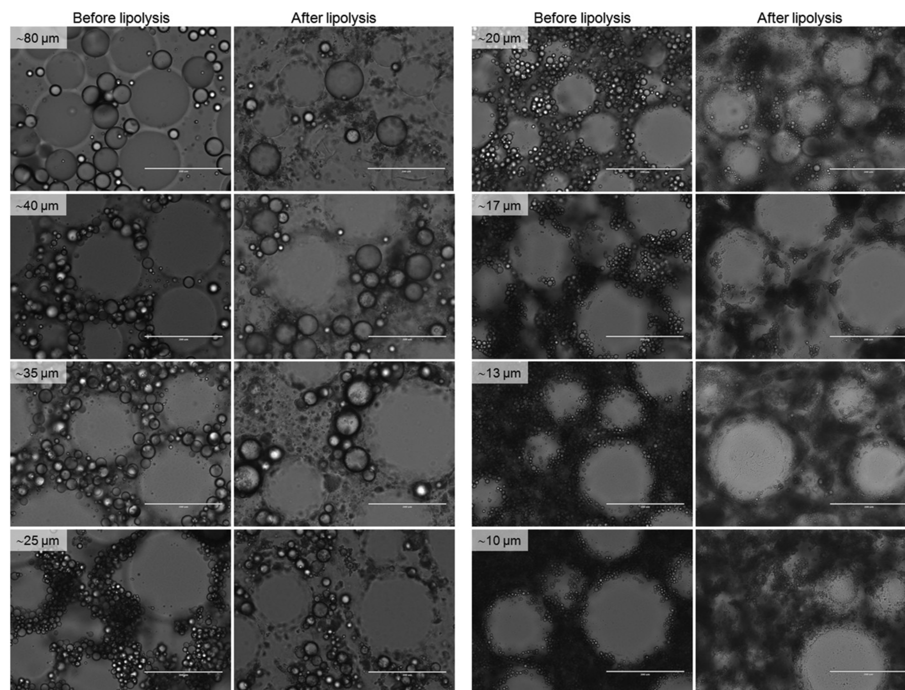


Fig. 8 Evolution of the microstructure of 10 wt% olive O/W instant emulsions after 2 h of lipolysis as a function of the initial maximum oil droplet size (indicated at the top left corner of each image). Scale bars are 200 μm. Larger brighter and smaller darker particles correspond to Sephadex swollen particles and oil droplets, respectively.

LMw HPMC and 0–0.5 wt% low- or high-Mw GG, and emulsions stabilised in the presence of Sephadex G-50 and LMw HPMC or by HMw HPMC in the absence of Tween 20. Conversely, the droplet size decreased on average after the lipolysis of emulsions initially containing droplets smaller than or equal to ∼20 μm. This is observed on emulsions stabilised by 1 wt% LMw HPMC and 1 wt% low- or high-Mw GG or by 1 wt% HMw HPMC either in the absence or presence of GG, and on emulsions stabilised in the presence of Sephadex G-50 and HMw HPMC. This suggests that the systems with relatively larger initial droplet size (>20 μm) destabilise to a larger extent in the presence of the intestinal components than the systems containing relatively smaller initial droplet size (≤20 μm). Duodenal components can cause droplet flocculation/coalescence,[22] which is favoured in the presence of larger oil droplets. Additionally, the aqueous phase viscosity plays an important role during the destabilisation of emulsion lipid droplets under *in vitro* duodenal conditions. In this sense, the emulsions with larger droplets in general had lower aqueous phase viscosity, because they were produced with lower thickener concentration and/or molecular weight, and can contribute to a greater coalescence in the presence of intestinal components.[22] In turn, this emulsion destabilisation greatly decreases the interfacial area available for lipase to anchor and hydrolyse the lipids. This may be a plausible reason to explain the appearance of an initial lag phase in the lipolysis profile, as well as the appreciable reduction in the rate and extent of lipolysis for instant emulsions with droplet size range above ∼20 μm (Fig. 7). In contrast, more stable emulsions with droplet size equal or below 20 μm seem to be readily digested, as supported by a reduction in oil droplet size and maximum lipolysis kinetics (Fig. 7). Therefore, a correlation between these two stability regimes of the emulsions in the presence of intestinal media and the inflection point in the lipolysis kinetics has been found, where the threshold in the maximum droplet diameter coincides at 17–20 μm, as highlighted in Fig. 7.

4. Conclusions

The described emulsification method produces relatively uniform emulsions requiring low-energy input, equivalent to that achieved by hand stirring, within short timeframe. The droplet size can be decreased upon shearing by increasing either the swelling capacity of the viscosifying particles, the molecular weight of the thickeners or their concentration. Also, the efficiency of the emulsifier can be replaced by a thickener providing sufficiently high aqueous phase viscosity and certain interfacial activity. Therefore, the formulation of the instant emulsion can be optimised to achieve different microstructures by reducing emulsifier and hydrocolloids concentration while increasing the molecular weight of the hydrocolloids, and using highly swellable particles. The extent to which these components increase the continuous phase viscosity both individually and collectively, deserves further investigation since they will determine the extent of the reduction of the emulsion droplet size and ultimately the

emulsion viscosity and final texture. This has relevant application to create different types of products.

The *in vitro* lipolysis of the instant emulsions is largely determined by the initial microstructure. A close correlation between the regimes of emulsion destabilisation in the presence of the intestinal components and the trend in lipolysis kinetics is found, which is explained in terms of the interfacial area available for lipolysis. Two stability regimes were found. The first one corresponds to emulsions with droplet size >20 μm, which greatly destabilise under duodenal conditions, while the second one comprises emulsions with droplet size ≤20 μm, which are relatively more stable. Emulsions belonging to the first group show considerable changes in lipolysis kinetics when varying the initial droplet size. Accordingly, the lipid digestion rate and extent are appreciably decreased as the oil droplet size increases in the initial emulsion microstructure. Conversely, emulsions classified in the second group exhibit the highest rate and extent of lipid digestion regardless of the initial droplet size. The interfacial composition has been shown also crucial on modulating the lipolisis kinetics.

Versatile instant emulsions can be developed as promising systems in the control of lipid digestion and targeted released of nutrients through the manipulation of the microstructure during the processing. However, the complex interrelation of the parameters affecting the formation of instant emulsions, above all when multiple stabilising agents are involved, and the subsequent lipolysis assures additional research. A more accurate model of *in vitro* digestion would ultimately help to elucidate the behaviour of instant emulsions within the gastrointestinal tract in the rational design of formulations with healthy attributes.

Conflict of interest

There are no conflicts of interest to declare.

Acknowledgements

Authors thank the financial support from the European Community's Seventh Framework Program (FP7-PEOPLE-2012-IEF) under Grant Agreement No. 326581.

References

1 M. D. Alur and V. Venugopal, in *Encyclopedia of Food Microbiology*, ed. R. K. Robinson, Elsevier, 1999, vol. 1, pp. 530–537.
2 A. Millqvist-Fureby, Characterisation of spray-dried emulsions with mixed fat phases, *Colloids Surf., B*, 2003, **31**, 65–79.
3 C. Ratti, in *Handbook of Food Powders: Processes and Properties*, ed. B. Bhandari, N. Bansal, M. Zhang and P. Schuck, Woodhead Publishing, 2013, pp. 57–84.
Q2 4 *Unilever Pat.*, WO2002000030A1, 2002.
5 *Unilever Pat.*, WO2003053149A1, 2003.
6 T. J. Foster, A. L. Russell, D. B. Farrer, M. Golding, R. M. Finlayson, A. Thomas, D. Jarvis and E. G. Pelan, in *Food Colloids: Self-Assembly and Material Science*, ed. E. Dickinson and M. Leser, RSC, 2007, ch. 28, pp. 413–423.
7 M. D. Lad, S. Samanci, J. R. Mitchell and T. J. Foster, in *Gums and Stabilisers for the Food Industry 15*, ed. P. A. Williams and G. O. Phillips, RSC, 2010, pp. 126–136.
8 M. Elleuch, D. Bedigian, O. Roiseux, S. Besbes, C. Blecker and H. Attia, Dietary fibre and fibre-rich by-products of food processing: Characterisation, technological functionality and commercial applications: A review, *Food Chem.*, 2011, **124**, 411–421.
9 F. Guillon, M. Champ, J. F. Thibault and L. Saulnier, in *Functional Foods: Concept to Product*, ed. M. Saarela, Woodhead Publishing, 2nd edn, 2011, pp. 582–622.
10 I. A. Brownlee, The physiological roles of dietary fibre, *Food Hydrocolloids*, 2011, **25**, 238–250.
11 D. L. Topping, in *Encyclopedia of Human Nutrition*, ed. L. H. Allen and A. Prentice, Elsevier, 3rd edn, 2013, pp. 50–54.
12 J. W. Anderson and A. E. Siesel, in *New Developments in Dietary Fiber: Physiological, and Analytical Aspects*, ed. I. Furda and C. J. Brine, Plenum Press, New York, 1990, vol. 270, pp. 17–36.
13 D. Kritchevsky and J. A. Story, in *CRC Handbook of Dietary Fiber in Human Nutrition*, ed. G. A. Spiller, CRC Press, Boca Raton, 3rd edn, 1993, pp. 163–178.
14 D. Lairon, Dietary fibres: Effects on lipid metabolism and mechanisms of action, *Eur. J. Clin. Nutr.*, 1996, **50**, 125–133.
15 D. J. A. Jenkins, C. W. C. Kendall and T. P. P. Ransom, Dietary fiber, the evolution of the human diet and coronary heart disease, *Nutr. Res.*, 1998, **18**, 633–652.
16 W. Yokoyama, W. H. K. Anderson, D. R. Albers, Y. J. Hong, M. L. Langhorst, S. C. Hung, J. T. Lin and S. A. Young, Dietary Hydroxypropyl Methylcellulose Increases Excretion of Saturated and Trans Fats by Hamsters Fed Fast Food Diets, *J. Agric. Food Chem.*, 2011, **59**, 11249–11254.
17 T. Tokle, U. Lesmes, E. A. Decker and D. J. McClements, Impact of dietary fiber coatings on behavior of protein-stabilized lipid droplets under simulated gastrointestinal conditions, *Food Funct.*, 2012, **3**, 58–66.
18 E. Malinauskyte, J. Ramanauskaite, D. Leskauskaite, T. G. Devold, R. B. Schuller and G. E. Vegarud, Effect of human and simulated gastric juices on the digestion of whey proteins and carboxymethylcellulose-stabilised O/W emulsions, *Food Chem.*, 2014, **165**, 104–112.
19 M. Espinal-Ruiz, F. Parada-Alfonso, L. P. Restrepo-Sánchez, C. E. Narváez-Cuenca and D. J. McClements, Interaction of a Dietary Fiber (Pectin) with Gastrointestinal Components (Bile Salts, Calcium, and Lipase): A Calorimetry, Electrophoresis, and Turbidity Study, *J. Agric. Food Chem.*, 2014, **62**, 12620–12630.
20 M. Espinal-Ruiz, F. Parada-Alfonso, L. P. Restrepo-Sánchez, C. E. Narváez-Cuenca and D. J. McClements, Impact of

This journal is © The Royal Society of Chemistry 2017

dietary fibers [methyl cellulose, chitosan, and pectin] on digestion of lipids under simulated gastrointestinal conditions, *Food Funct.*, 2014, **5**, 3083–3095.

21 F. A. Bellesi, M. J. Martinez, V. M. P. Ruiz-Henestrosa and A. M. R. Pilosof, Comparative behavior of protein or polysaccharide stabilized emulsion under in vitro gastrointestinal conditions, *Food Hydrocolloids*, 2016, **52**, 47–56.

22 A. Torcello-Gómez and T. J. Foster, Influence of interfacial and bulk properties of cellulose ethers on lipolysis of oil-in-water emulsions, *Carbohydr. Polym.*, 2016, **144**, 495–503.

23 M. Thongngam and D. J. McClements, Isothermal titration calorimetry study of the interactions between chitosan and a bile salt (sodium taurocholate), *Food Hydrocolloids*, 2005, **19**, 813–819.

24 A. Torcello-Gómez, C. Fernández Fraguas, M. J. Ridout, N. C. Woodward, P. J. Wilde and T. J. Foster, Effect of substituent pattern and molecular weight of cellulose ethers on interactions with different bile salts, *Food Funct.*, 2015, **6**, 730–739.

25 I. Heertje, Structure and function of food products: A review, *Food Struct.*, 2014, **1**, 3–23.

26 E. Millstone and T. Lang, *The penguin atlas of food*, Penguin Books, New York, 2003.

27 M. Minekus, M. Alminger, P. Alvito, S. Ballance, T. Bohn, C. Bourlieu, F. Carrière, R. Boutrou, M. Corredig, D. Dupont, C. Dufour, L. Egger, M. Golding, S. Karakaya, B. Kirkhus, S. Le Feunteun, U. Lesmes, A. Macierzanka, A. Mackie, S. Marze, D. J. McClements, O. Ménard, I. Recio, C. N. Santos, R. P. Singh, G. E. Vegarud, M. S. J. Wickham, W. Weitschies and A. BrodKorb, A standardised static in vitro digestion method suitable for food – an international consensus, *Food Funct.*, 2014, **5**, 1113–1124.

28 R. Pal, Effect of droplet size on the rheology of emulsions, *AIChE J.*, 1996, **42**, 3181–3190.

29 H. P. Grace, Dispersion phenomena in high viscosity immiscible fluid systems and application of static mixers as dispersion devices in such systems, *Chem. Eng. Commun.*, 1982, **14**, 225–277.

30 A. Torcello-Gómez and T. J. Foster, Interactions between cellulose ethers and a bile salt in the control of lipid digestion of lipid-based systems, *Carbohydr. Polym.*, 2014, **113**, 53–61.

31 C. M. Rosell, E. Santos and C. Collar, Physico-chemical properties of commercial fibres from different sources: A comparative approach, *Food Res. Int.*, 2009, **42**, 176–184.

32 D. Saha and S. Bhattacharya, Hydrocolloids as thickening and gelling agents in food: a critical review, *J. Food Sci. Technol.*, 2010, **47**, 587–597.

33 N. Garti and D. Reichman, Surface properties and emulsification activity of galactomannans, *Food Hydrocolloids*, 1994, **8**, 155–173.

34 O. Kaltsa, P. Paximada, I. Mandala and E. Scholten, Physical characteristics of submicron emulsions upon partial displacement of whey protein by a small molecular weight surfactant and pectin addition, *Food Res. Int.*, 2014, **66**, 401–408.

35 P. Reis, K. Holmberg, R. Miller, J. Kragel, D. O. Grigoriev, M. E. Leser and H. J. Watzke, Competition between lipases and monoglycerides at interfaces, *Langmuir*, 2008, **24**, 7400–7407.

36 P. Reis, R. Miller, M. Leser and H. Watzke, Lipase-catalyzed Reactions at Interfaces of Two-phase Systems and Microemulsions, *Appl. Biochem. Biotechnol.*, 2009, **158**, 706–721.

37 A. Torcello-Gómez, J. Maldonado-Valderrama, A. Martín-Rodríguez and D. J. McClements, Physicochemical properties and digestibility of emulsified lipids in simulated intestinal fluids: influence of interfacial characteristics, *Soft Matter*, 2011, **7**, 6167–6177.

38 S. Mun, E. A. Decker and D. J. McClements, Influence of emulsifier type on in vitro digestibility of lipid droplets by pancreatic lipase, *Food Res. Int.*, 2007, **40**, 770–781.

39 B. S. Chu, G. T. Rich, M. J. Ridout, R. M. Faulks, M. S. J. Wickham and P. J. Wilde, Modulating Pancreatic Lipase Activity with Galactolipids: Effects of Emulsion Interfacial Composition, *Langmuir*, 2009, **25**, 9352–9360.

40 Y. Li and D. J. McClements, New Mathematical Model for Interpreting pH-Stat Digestion Profiles: Impact of Lipid Droplet Characteristics on in Vitro Digestibility, *J. Agric. Food Chem.*, 2010, **58**, 8085–8092.

41 M. Armand, B. Pasquier, M. André, P. Borel, M. Senft, J. Peyrot, J. Salducci, H. Portugal, V. Jaussan and D. Lairon, Digestion and absorption of 2 fat emulsions with different droplet sizes in the human digestive tract, *Am. J. Clin. Nutr.*, 1999, **70**, 1096–1106.