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The influence of non-ionic surfactant on lipid digestion of gum Arabic stabilized oil-in-water emulsion

Xiaolin Yao [a, b, *], Ke Nie [a], Yu Chen [a], Fatang Jiang [a], Ying Kuang [a], Heng Yan [c], Yapeng Fang [a, **], Hao Yang [d], Katsuyoshi Nishinari [a], Glyn O. Phillips [a]

[a] School of Bioengineering and Food Science, Hubei University of Technology, Wuhan 430068, China
[b] School of Public Health, Tongji Medical College, Huazhong University of Science and Technology, Wuhan 430030, China
[c] Hubei Provincial Institute for Food Supervision and Test, Wuhan 430070, China
[d] Key Laboratory for Green Chemical Process of Ministry of Education and School of Chemical Engineering and Pharmacy, Wuhan Institute of Technology, Xiongchu Street, Wuhan 430073, China

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ABSTRACT

Lipid digestion is the process of enzymatic hydrolysis at the interface, and the interfacial structure play an important role in the lipid digestion. Non-ionic surfactant Tween80 (T80) presented a significant impact on the lipid digestion of gum Arabic (GA) stabilized oil-in-water emulsion. The interfacial structure of GA-T80 emulsion droplets has been investigated by using gel permeation chromatography-multiangle laser light scattering, interfacial measurement and atomic force microscope. With addition of T80, the surface loading of GA decreased until a plateau value reached. About 15% of GA still remained on the interface when T80 concentration increased to ~4%. Addition of T80 contributed to a stiffening of the network formed at the droplets interface. The kinetics of lipid digestion exhibited a biphasic rate under this certain interface, shown as an induction period followed by a speeding up process. It probably resulted from the adsorption behavior of lipase on the emulsion interface during the lipolysis process. In was found that the accumulation of lipase on the emulsion droplets interface contributed to the lag period of the free fatty acids release. The biphasic rate of lipid digestion was mainly caused by the rate of lipase adsorption, which was highly dependent on the interface structure of emulsion droplets.

1. Introduction

Lipids are the most energy dense components of the three major macronutrients in foods (lipids, proteins, and carbohydrates), however, they may be the least effective at promoting satiety and satiation (Lissner & Heitmann, 1995). There is considerable interest in reducing the overall lipid content of foods, while increasing their ability to promote satiety and satiation, and therefore combat overweight and obesity (Beglinger & Degen, 2004; Karra & Batterham, 2009; Li, Hu, Du, Xiao, & Mcclements, 2011; Li, Hu, & Mcclements, 2011). A possible approach to achieving this goal is to design functional foods that delay the digestion of lipids in the gastrointestinal tract (GIT), which may promote satiety (Mei, Lindqvist, Krabisch, Rehfeld, & Erlanson-Albertsson, 2006).

Previous studies have shown that emulsion-based delivery systems can be designed to control the rate and extent of lipid digestion within the GIT (Mcclements & Li, 2010; Singh, Ye, & Horne, 2009; Wilde & Chu, 2011). In the small intestine, the emulsified lipids are mixed with digestive juices that contain pancreatic lipase, colipase, bile salts, and phospholipids (Golding & Wooster, 2010). The bile salts and phospholipids compete and displace any surface active material present at the oil-water interface, and the lipase/colipase complex binds to the lipid droplet surfaces and initiates the lipid digestion process (Reis, Holmberg, Watzke, Leser, & Miller, 2009). The pancreatic lipase converts triacylglycerols (TAGs) into monoacylglycerols (MAGs) and free fatty acids (FFA), which leave the lipid droplet surfaces and are incorporated into mixed micelle structures consisting of phospholipids and bile salts, which then transport them to the epithelial cells, where they are adsorbed (Yao, Xiao, & Mcclements, 2014; Wilde & Chu, 2011). Therefore, understanding the fundamental aspects of

* Corresponding author. School of Bioengineering and Food Science, Hubei University of Technology, Wuhan 430068, China.
** Corresponding author.
E-mail addresses: yaoxiaolin1113@163.com (X. Yao), fangypphrc@163.com (Y. Fang).

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the digestion of emulsified lipids under conditions that simulate the human GIT is of paramount importance to gain insights into the physicochemical and biochemical processes in the physiological milieu that further bioengineers the initial food structure (Golding et al., 2011; Mackie & Macierzanka, 2010; Sarkar et al., 2016; Singh & Sarkar, 2011; Singh et al., 2009).

Lipid digestion is an interfacial phenomenon that involves adsorption of bile salts and lipase molecules to lipid droplet surfaces so that the enzyme can come into close proximity with its substrate (usually TAGs). The initial emulsion form of lipids may impact its subsequent absorption and digestion (Porter, Trevaskis, & Charman, 2007; Singh & Sarkar, 2011). A number of studies found that the droplets size played an important role in determining the lipid digestion rate, which was correlated with the surface area of lipid exposed to the lipase (Golding et al., 2011; Malaki, Nik, Wright, & Corredig, 2011). However, it was reported that the digestion rate of lipid digestion was actually slower in the nanoemulsion (d = 60 nm) prepared using the homogenization/solvent displacement method than in the β-lactoglobulin (BLG)-stabilized conventional emulsion (d = 200 nm), which was attributed to differences in the interfacial structure. The lipid droplets in conventional emulsion would be coated by a thin monolayer of globular protein molecules, while the droplets in nanoemulsion would be coated by a much thicker layer of aggregated globular protein molecules due to the shrinkage of the droplets during their preparation by solvent evaporation (Mcclements & Li, 2010). Sarkar et al. (2016) revealed that the heat-induced microgel particle could achieve an increased coverage-fused network at the oil-water interface, which was more efficient in delaying the rate of lipid digestion compared to the intact whey protein microgel particles. Thus, the nature and thickness of the interfacial layer surrounding the lipid droplets played an important role in determining the rate and extent of lipid hydrolysis (Torcellogómez, Jódarreyes, Maldonado-Valderrama, & Martínrodríguez, 2012; Yao et al., 2013; Fillery-Travis, Robins, & Foster, 1995).

Moreover, it has become evident that the activity of lipase and other lipolytic enzymes is markedly affected by the physical properties of the oil-water interface upon which catalysis occurs. For hydrolysis of liquid crystalline phosphatides, the reaction consisted of a short initial burst of hydrolysis, a lag period of very slow reaction, followed by a dramatic increase in the reaction rate. Addition of lysolecithin or fatty acid abolished the lag period. It was postulated that the enzyme adsorbed irreversibly to the surface of the liquid crystalline phase, but the reaction products would stimulate desorption of enzyme from the surface. Thus, changes of hydrolysis rate for dispersed phosphatidylcholines were attributed to the desorption of enzyme from the lipid surface (Tinker, Purdon, Wei, & Mason, 1978). The absorption of liposomally entrapped [^{14}C] inulin into the venous effluent is biphasic, first a lag period of 30 min, followed by a rapid linear increase. The possible explanation of the lag period based on the evidence presented is that it is the time required to degrade liposomes in the lumen and/or the time required for liposomes to be taken up by mucosal cells and digested intracellularly (Patel, Tuzedotl, & Stevenson, 1985). Similarly, the rate of lipid digestion was also biphasic as observed in the protein or polysaccharide stabilized emulsions with addition of non-ionic surfactants (Li & Mcclements, 2011). To our knowledge, there has been a few studies to analyze the reason contributing to the biphasic rate of lipid digestion. The aim of this study was to understand the possible mechanism from the interfacial perspective to explain why the lipid digestion represents biphasic rate in polysaccharides stabilized emulsions in the presence of non-ionic surfactant. This knowledge will facilitate the development of interfacial structures that affect lipid digestion and hence improve the nutritional impact of foods through reduction of postprandial hyperlipidemia (Lopezmiranda, Williams, & Lairon, 2007) and promotion of satiety (Ritter, 2004).

2. Materials and methods

2.1. Materials

Gum Arabic (GA) (SD LOT 110512) provided by San-Ei Gen F.F.I., Inc. (Osaka, Japan) has a weight average molecular weight of MW 9.88−105 Da and a moisture content of 5.56 wt%. The non-ionic surfactant Tween80 (T80) was purchased from Xilong Chemical Co., Ltd (Shanghai, China). Medium Chain Triglyceride (MCT) was obtained from KLK Oleo, Ltd. (Malaysia), which contains 58 wt% C8 fatty acid and 42 wt% C10 fatty acid, and has a relative density of 0.95 g/mL. Lipase from porcine pancreas, Type II (L3126, triacylglycerol hydrolase E.C. 3.1.1.3, PPL), was obtained from Sigma Aldrich (St. Louis, MO, USA). It has a reported lipase activity of 100−400 units/mg protein toward olive oil during incubation at 37 °C for 30min. The lipase activity measured following the supplier's protocol with titrimetry was around 320 U/mg toward olive oil and 500 U/mg toward MCT, due to the lipase activity highly dependent on the substrate. Bile salts No. 3 was purchased from Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China). It has a bile acid juice content of >90 wt%. Bile salts is from bovine bile acid juice and its main components comprised of cholic acid (63.92%) and deoxycholic acid sodium salt (18.89%), conjugated with either glycine (75%) or taurine (25%), as measured by chromatographic and spectral methods. The lipase inhibitor 4-bromophenylboronic acid (4-BPB) was purchased from Sigma Aldrich. The other chemicals used in the study were from Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China) and were of analytical grade.

2.2. Emulsion preparation

GA solutions were prepared by dispersing gum powders in Millipore water without adjusting pH, then put on a roller mixer at room temperature overnight to ensure complete hydration. T80 was dissolved in a buffer solution (5 mM phosphate, pH 7.0) and stirring overnight. MCT was added to the emulsifier solutions to achieve the final concentration of 20 wt% in the emulsion, the systems were prehomogenized using a high-speed blender (Polytron PT 2100) at 26,000 rpm/min for 3 min, followed by a high-pressure homogenizer (USA MFIC M-110L) at 75 MPa with one pass. The emulsion containing 10 wt% GA was then mixed with T80 solution and stirred for at least 4 h at 37 °C. The mixtures were adjusted to pH 7.0 using NaOH (0.15 M) with continuous stirring for 30 min to promote dispersion and adsorption of T80 molecules onto the GA-coated droplets surfaces, which was called GA - T80 emulsion. For T80 - GA emulsion, the primary emulsion was prepared with T80 solution and then GA powder was dispersed in it and stirred at room temperature for at least 4 h to ensure complete hydration and adsorption of GA molecular onto the T80-coated droplets interface. For GA + T80 emulsion, GA and T80 solutions were mixed together and prepared the emulsion according to the above procedure, contributing a competitive adsorption onto the droplets interface for GA and T80 moleculars.

2.3. Effect of surfactants on the physical properties of emulsion

The particle size distribution (PSD) of emulsions was measured using a laser diffraction technique (MasterSizer 2000, Malvern Instruments, UK). The emulsions were dropwise added into the dispersing unit (Millipore water) until a laser obscuration of 10%