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DEVELOPMENT OF PAPRIKA OLEORESIN DISPERSIONS FOR IMPROVING THE BIOACCESSIBILITY OF CAROTENOIDS

DESARROLLO DE DISPERSIONES DE OLEORRESINA DE PAPRIKA PARA MEJORAR LA BIOACCESIBILIDAD DE CAROTENOIDES

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Abstract

Red pepper (*Capsicum annuum L.*) oleoresin contain a diversity of carotenoids, which has been associated with lower risk for different chronic diseases like various types of cancer, cardiovascular disease and age-related macular degeneration. However, they are very sensitive to pro-oxidant conditions, heat and light. Previous attempts have been made to improve bioavailability and stability of carotenoids, of which emulsions have proven to be a feasible method. Conventional (CE) and nano (NE) emulsions loaded with paprika oleoresin carotenoids (POC; 1% wt/wt) were fabricated using surfactants blend (Tween 40 and Span 20) with a hydrophilic-lipophilic balance (HLB), ranging from 12 to 15.6, and surfactant: POC ratio of 1:1 (wt/wt). POC bioaccessibility was studied using an in vitro model to simulate oral, gastric and small intestine phases of the gastrointestinal tract (GIT). In general, higher HLB values produced CE's and NE's with smaller particle size and negative value of zeta potential. The smaller the droplet size, the higher was POC bioaccessibility. NE prepared with a HLB of 15.6 had a particle size of 38.93 nm and a bioaccessibility of 74%. Bioaccessibility of unemulsified POC was practically nil. Conventional and nanoemulsions protected paprika oleoresin carotenoids deterioration during simulated gastrointestinal tract.

Keywords: nanoemulsions, paprika oleoresin, gastrointestinal tract, bioaccessibility.

Resumen

La oleorresina de paprika (*Capsicum annuum L.*) contiene una diversidad de carotenoides que se han relacionado con la disminución de enfermedades crónico degenerativas, cáncer, enfermedades cardiovasculares y degeneración macular. Los carotenoides son muy sensibles a condiciones pro-oxidantes, calor y luz, por lo que algunas investigaciones se han enfocado en mejorar su biodisponibilidad y estabilidad, de las cuales las emulsiones han demostrado ser un buen método. En este trabajo, se realizaron emulsiones convencionales (CE) y nanoemulsiones (NE) de carotenoides de oleorresina de paprika (POC, 1% p/p) utilizando surfactantes (Tween 40 y Span 20) con un balance hidrófilo-lipófilo (HLB) de 12 y 15.6, a una relación surfactante: POC de 1:1 (p/p). La bioaccesibilidad se estudió utilizando un modelo gastrointestinal (GIT) para simular las fases oral, estómago e intestino delgado. Los valores más altos de HLB produjeron CE y NE con menores tamaños de partícula y potencial zeta negativo. A menor tamaño de gota, mayor fue la bioaccesibilidad, mientras que para POC no emulsificada fue prácticamente nula. La NE con HLB de 15.6 mostró un tamaño de partícula de 38.93 nm y bioaccesibilidad del 74%. Las NE y CE protegieron los carotenoides de la oleorresina de paprika al pasar el GIT simulado.

Palabras clave: nanoemulsiones, oleorresina de paprika, tracto gastrointestinal, bioaccesibilidad.

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1 Introduction

Paprika oleoresin is a solution of carotenoid pigments obtained from the ripe fruit of *Capsicum annuum L.* It is an oil commonly used in the food industry as a colorant for sauces, soups and meat-based meals. Carotenoids consumption has been associated with beneficial effects on human health, such as reduced risk of certain cancers, cardiovascular diseases, and age-related macular degeneration (Meléndez-Martínez, *et al.*, 2007; Wildman *et al.*, 2016). For these reasons, there is a strong interest in using carotenoids as functional ingredients in food products. Carotenoids are insoluble in water and only slightly soluble in oil at room temperature. Its crystalline forms are considered to have poor bioavailability and are difficult to incorporate in food formulations.

Modern methods of encapsulation technology allow the production of a physically/chemically stable and bioavailable structure after their preparation, storage, and application in food systems. These include emulsion-based delivery systems (Ribeiro *et al.*, 2008), liposomes (Socaciu *et al.*, 2000), the entrapment of carotenoids in microspheres (Laos *et al.*, 2007) and production of microcapsules by drying processes (Rascón *et al.*, 2011) as well as carotenoids entrapment within food nanoemulsions stabilized with non-ionic surfactants (Pascual-Pineda *et al.*, 2015)

Within the human body carotenoids first undergo solubilization from the food matrix followed by micellization. This refers to their incorporation into micelles, which are molecular aggregates that transport fat-soluble material, making it more accessible by the intestinal epithelium. It has been shown that the percentage of carotenoid ingested versus the amount assimilated, reaches up to 10% when a raw food is consumed (Fernández-García *et al.*, 2012). Previous attempts have been made to improve bioavailability and stability of carotenoids, of which nanoemulsions have proven to be a feasible method. Variables affecting the bioaccessibility or bioavailability of carotenoids incorporated within nanoemulsion-based delivery systems have been investigated, that include the impact of carrier oil composition (medium chain triglyceride to long chain triglyceride), total oil concentration, droplet size, lipid phase physical state and interfacial structure (Liu *et al.*, 2012; Qian *et al.*, 2012; Xia *et al.*, 2015). However, the underlying mechanisms through which absorption is improved have not been fully understood.

Selection of the emulsifier is one of the most important factors because the oil-water interface composition often determines the stability and bioavailability of lipophilic molecules (Golding and Wooster, 2010). Emulsifiers are adsorbed at the oil-water interface, forming a stabilizing layer at the droplet surface, which depends on the hydrophilic-lipophilic balance (HLB). According to McClements (2013) the HLB of interfacial coating surrounding a lipidic bioactive molecule may influence the way of the particle responses to changes in gastrointestinal tract, such as temperature, pH, ionic composition and enzyme activity.

Interactions of the adsorbed layer and lipase are critical in determining the bioavailability of lipid bioactive molecules. The interface composition and initial interfacial area of the emulsion would be expected to have an impact on lipase binding and carotenoids bioaccessibility. Properties of the interface strongly affect the behavior of emulsion and these are in continuous changes during gastrointestinal transit. To the best of our knowledge no reports have been published aimed to understand, how the initial composition of interface can modulate the bioactivity and bioaccessibility of lipid bioactive molecules and what may the influence of the HLB on the bioaccessibility of carotenoids. The aim of this work was to fabricate conventional (CE) and nanoemulsions (NE) loaded with POC and to study the effect of in vitro bioaccessibility of carotenoids.

2 Materials and methods

2.1 Materials

Polyoxyethylene sorbitan monopalmitate (Tween 40), Sorbitan monolaurate (Span 20), porcine mucin, lipase from porcine pancreas and bile porcine extract were purchased from the Sigma-Aldrich Chemical Co. (St. Louis, MO, USA). NaCl, HCl, NaOH, $CaCl_2$, KCl, KH_2PO_4 and Na_2HPO_4 were purchased from Merck. Paprika oleoresin was used as carotenoids source (POC) and was obtained from AMCO (Mexico City). The water used during the study was of ultrapure grade from a Milli-Q Plus system (Millipore, Bedford, MA, USA).

2.2 Conventional emulsions and nanoemulsions formation

Conventional emulsions and nanoemulsions were prepared according our previous work using the maximum stability criteria (Pascual-Pineda *et al.*, 2015). All emulsions consisted of paprika oleoresin, mixed surfactant and Milli-Q water. Concentration of paprika oleoresin in the emulsion systems was fixed at 1% wt/wt at ratio of emulsifier: paprika oleoresin of 1:1 (wt/wt). A preliminary work was carried out in order to study the effect of HLB and surfactant concentration on stability and particle size of the emulsions. The mixture of emulsifiers was in the HLB range of 8.0 to 15.6, being the more suitable to prepare stable emulsions HLB values of 12.0, 14.0 and 15.6. These values were adjusted according to Griffin (1954).To obtain conventional emulsions (CE), paprika oleoresin was dispersed in aqueous surfactant solution by using a high speed homogenizer (Wiggen Hauser D-500; Berlin, Germany) at 4900 g, 40 ºC for 5 min. Nano emulsions (NE) were obtained by exposing the conventional emulsion through a high-power ultrasonicator (Branson Digital Sonifier 250 W, 24 kHz; Danbury, CT, USA). Preliminary experiments were conducted with varying volume and power input to get the smallest particle size, which it was obtained when the emulsions were prepared with amplitude of 65% and 20 mL of sample. The sonifier tip horn was adjusted to 2 cm below the sample surface. A water bath was used to maintain the temperature of the mixture at 30±2 ºC. Paprika oleoresin carotenoids (POC) was dispersed at 1% (wt/wt) in aqueous solution at 4900 g and 40 ºC for 5 min by using a high speed homogenizer.

2.3 Droplet size, polydispersity index and zeta potential measurements

Average particle size, polydispersity index (PDI) and zeta potential of the samples were evaluated using a dynamic light scattering instrument (Nano ZS 2000; Malvern Instruments, Malvern, UK). Samples were diluted 1:100 using Milli-Q water. These measurements were immediately determined after each phase. The results were reported as an average of three individual injections.

2.4 Simulated gastrointestinal tract model

1. Mouth phase: Simulated saliva fluid, containing porcine mucin at 3% wt and various salts, was prepared according to Sarkar *et al.* (2009). The emulsions were mixed with saliva fluid at a 50:50 ratio and the mixture was then adjusted to pH 6.8. The mixture was incubated at 37 ºC for 10 min using continuous agitation at 100 rpm.

2. Gastric phase: Simulated gastric fluid was prepared using the method reported by the same research group (Sarkar *et al.*, 2009) by dissolving 2 g of NaCl and 7 mL of HCl (37%) in 1 L of water and the pH was adjusted to 1.2 using 1.0 M HCl. The "bolus" sample from the mouth phase was mixed with simulated gastric fluid at a 50:50 volume ratio. The pH of the sample was adjusted to 2.5 using 1 M NaOH and incubated at 37 ºC for 2 h with continuous agitation at 100 rpm.

3. Small intestinal phase: A pH-stat (Metrohm USA Inc., Riverview, FL) device was used to simulate the conditions in the small intestinal phase of the gastrointestinal tract (McClements and Li, 2010). An aliquot of 30 mL of the "bolus" sample was placed in a temperature-controlled chamber at 37 ºC and the pH was set at 7.0 using 1 M NaOH solution. Then, 4 mL of bile extract (46.87 mg mL^{-1}) and 1 mL of calcium chloride (110 mg mL^{-1}) solutions dissolved in phosphate buffer were added to the sample and the pH was adjusted to 7.0 if necessary. Then, 2.5 mL of freshly prepared lipase suspension (24 mg mL^{-1}) dissolved in phosphate buffer was incorporated into the mixture. During 2 h of the intestinal digestion process, the pH of the solution was maintained at 7.0 by adding 1 M NaOH manually. The amount of NaOH added over time was recorded throughout the digestion.

2.5 Carotenoids determination

Carotenoids content was determined through a spectrophotometric method proposed by Hornero-Méndez and Minguez-Mosquera (2001). Approximately 10 μL of sample for each phase were dissolved in a volumetric flask containing 25 mL of acetone, and then, absorbance measurements were made using a diode array spectrophotometer (model 8453; Agilent Technologies; Waldbronn, Germany) at 472 and 508 nm.

Absorbance values were introduced in the following equations to obtain the isochromic carotenoid fractions:

$$C^R = \frac{A_{508} \times 2144.0 - A_{472} \times 403.3}{270.9} \quad (1)$$

$$C^Y = \frac{A_{472} \times 1724.3 - A_{508} \times 2450.1}{270.9} \quad (2)$$

$$C^T = C^R + C^Y \quad (3)$$

where C^R, C^Y and C^T are the red, yellow and total fractions (mg mL^{-1}), respectively, and A is the absorbance of the sample. Carotenoid determinations were carried out in triplicate after the mouth, gastric and small intestine phases.

2.6 *Bioaccessibility determination*

Bioaccessibility was determined with the method proposed by Qian *et al.* (2012). After in vitro digestion, 10 mL of sample were collected and centrifuged (8160 g) at 25 °C for 40 min using a HERMLE centrifuge model Z160M (Wehingen, Germany). The middle phase was assumed to consist of mixed micelles that solubilized the bioactive component. Aliquots (5 mL) were collected directly from the middle phase of centrifuged samples. A syringe filter (0.45 μm PP, WHATMAN) was used to filter the middle phase to remove any residual large particles. Aliquots were vortexed with 5 mL acetone and centrifuged at 8161.4 g at 25 °C for 30 min. The concentration of carotenoids extracted from the sample was determined as described above (section 2.5) and taking into account the dilution factor. Bioaccessibility was calculated using the following equation:

$$Bioaccessibility = 100 \times \frac{C_{Micelle}}{C_{Emulsion}} \quad (4)$$

where $C_{Micelle}$ and $C_{Emulsion}$ are the concentration of carotenoids in the micelle fraction and in the emulsion sample before the pH-stat experiment, respectively.

2.7 *Statistical analysis*

The mean values and standard deviations were calculated for all the experiments carried out in triplicate. Two-way Analysis of Variance (ANOVA) of independent variables followed by a Tukey HSD test was carried out on the measurements to establish significant differences ($p < 0.05$) using the StatPlus Mac LE software.

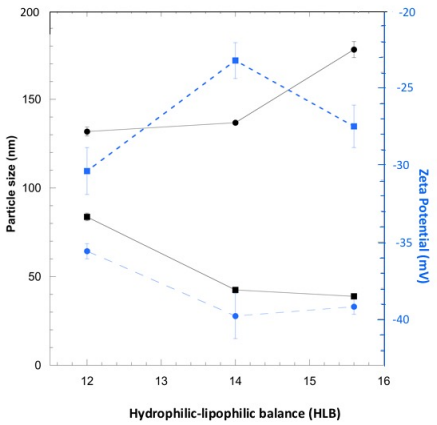

Fig. 1. Effect of HLB value on the particle size and zeta potential of conventional emulsions (circles) and nanoemulsions (squares). Bars represent the standard deviation of two replicated experiments made in triplicate.

3 Results and Discussion

Fig. 1 depicts the variation of particle size and zeta potential as a function of hydrophilic-lipophilic balance for conventional and nano-emulsions, respectively. In all experiments the particle size for nanoemulsions was significantly smaller than that of the conventional emulsions. It can be observed that NE and CE had mean particle sizes smaller than 100 nm and 200 nm, respectively. NE samples showed that the particle size decreased as the HLB value increased, conversely than CE. The required HLB to produce a stable emulsion is that, which provides the lowest interfacial tension (Malcolmson *et al.*, 1998). When this interfacial tension is achieved, the surfactant concentration is the lowest, which was 1% for all HLB values used in our previous work (Pascual-Pineda *et al.*, 2015). Therefore, the surfactant concentration employed in this work was 1%, with mean particle sizes below 200 nm in all the treatments. Malcolmson *et al.* (1998) and Warisnoicharoen *et al.* (2000) also reported that particle size depends on the surfactant structure and chain length. Wang *et al.* (2009) found that structure of Tween had a greater effect on the decrease of droplet size than that of Span. These authors also found that the formation of nanoemulsions with smaller droplet size depends on the CH chain length of the surfactant. The surfactants used in our work have similar CH hydrophobic chain length, where this is of C15

and C11 for Tween 40 and Span 20, respectively, which favors the formation of small particle sizes. Associative adsorption is produced when two different surfactants are mixed (Bergenståhl, 2008). Therefore, the properties displayed by the interfacial layer are an average of both emulsifiers, and the HLB value should describe the properties of the emulsifier blend (Davies, 1957).

A PDI ranging from 0.301 to 0.268 and 0.228 to 0.170 for CE and NE, respectively, was observed in this study for the three HLB values, which indicated that ultrasonic process produce narrower particle size distribution. However, as shown in Fig. 1, all emulsion formulations exhibited a zeta potential negatively charged, ranging from −35.60 to −39.20 and from −23.53 to −30.40 mV, for CE and NE, respectively. These results suggest that the input energy did not improved the physical stability significantly on the emulsion systems during the sonication step. Similar results were obtained by Teo *et al.* (2016) for the lutein of whey protein isolate nanoemulsions and emulsions prepared by the emulsification and solvent evaporation method. Usually, particle aggregation is less likely to occur for charged particles with pronounced zeta potential (> |20|) due to the electrostatic repulsion between particles with the same electrical charge (Gonzalez-Mira *et al.*, 2010).

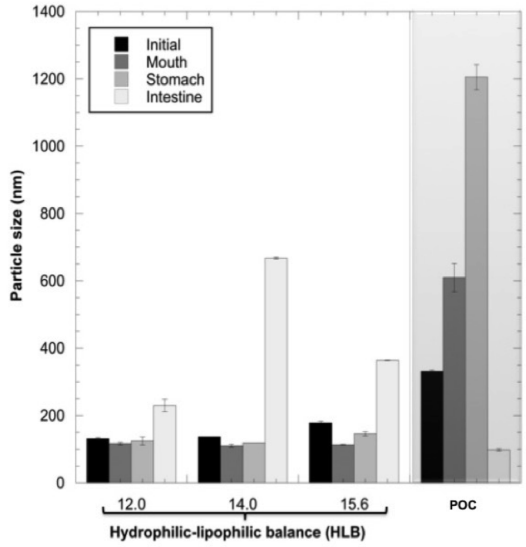

Fig. 2(A). Effect of HLB value on the mean diameter of the particles during in vitro digestion of conventional emulsions in comparison to paprika oleoresin. Bars represent the standard deviation of two replicated experiments made in triplicate.

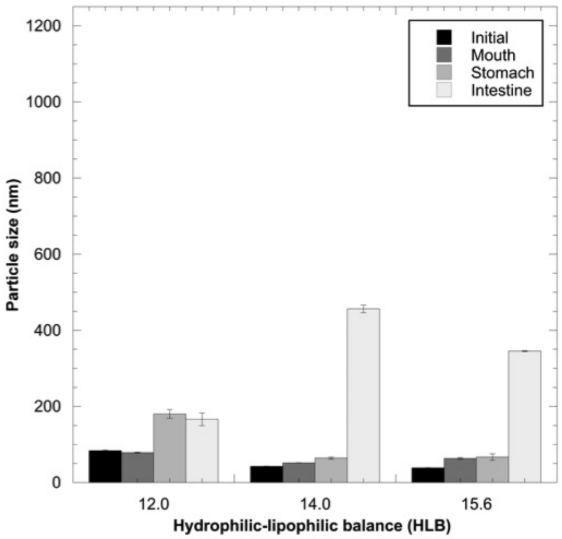

Fig. 2(B). Effect of HLB value on the mean diameter of the particles during in vitro digestion of nanoemulsions. Bars represent the standard deviation of two replicated experiments made in triplicate.

Therefore, in this work the electrostatic repulsive forces might contribute to the stabilization of droplet. This behavior has been observed previously for paraffin oil in water nanoemulsions (Liu *et al.*, 2006). The origin of the negative surface charges on the nonpolar oil droplets has been discussed previously. It has been suggested that the existence of EO groups of Tween 40 surfactant create hydrogen bonds with the hydroxyl ions of the water molecules in the boundary layer, which promote negative surface charges (Liu *et al.*, 2006). Hsu and Nacu (2003) reported similar results for soybean oil in water emulsion stabilized by Tween surfactant series.

The mean particle size (Z-average diameters) at different HLB and 1% of surfactant concentration of the initial emulsions and of the samples collected after each stage of the *in vitro* digestion are shown for conventional emulsions in Fig. 2(A). This figure also shows the mean particle size for POC. Fig. 2(B) depicts the behavior of particle size for NE. It can be observed that after incubation in artificial saliva (pH 6.8, 10 min) and in the simulated gastric fluid (pH 2.5, 2h) the mean particle size of all emulsions remained relatively small with particle sizes of less than 180 nm, for the three HLB used in both processes. The initial particle size for NE was less than 100 nm and this size had a slightly increase at the mouth and gastric phases. The POC showed a great increase in the particle size, with 331.7,

609.9 and 1205.0 nm at initial, mouth and gastric phases, respectively. After incubation in simulated small intestinal fluids, all the emulsions exhibited a large increase in the mean particle size, being higher for a HLB of 14.0 (Fig. 2A and B). These results suggest an increase in the particle size in the intestinal phase, when the composition of the interfacial layer around the emulsified droplets has more hydrophilic moiety of the surfactant molecule, which could affect the bioavailability and stability of encapsulated carotenoids. Singh *et al.* (2009) reported that the concentration and the physicochemical properties of the emulsion-water interface determined the extent of the enzyme binding on the emulsion surface and consequent lipolysis. An increase in particle size in the mouth phase has been previously attributed to depletion and/or bridging flocculation caused by droplet interactions with mucin (Sarkar *et al.*, 2009). When mucin simultaneously binds to the surfaces of two or more droplets bridging flocculation occurs (Salvia-Trujillo *et al.*, 2013; Singh *et al.*, 2009). On the other hand, depletion flocculation occurs by the presence of sufficiently high levels of non-adsorbed polymer (mucin) molecules in aqueous phase surrounding the droplets (Silletti *et al.*, 2007).

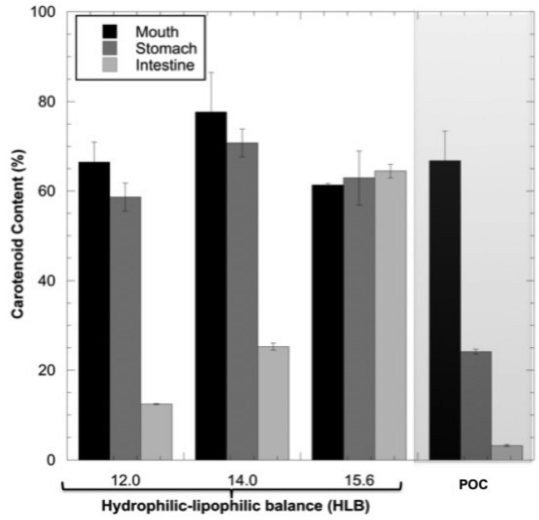

Fig. 3(A). Influence of HLB value on the carotenoids stability during in vitro digestion of conventional emulsions in comparison to paprika oleoresin. Bars represent the standard deviation of two replicated experiments made in triplicate.

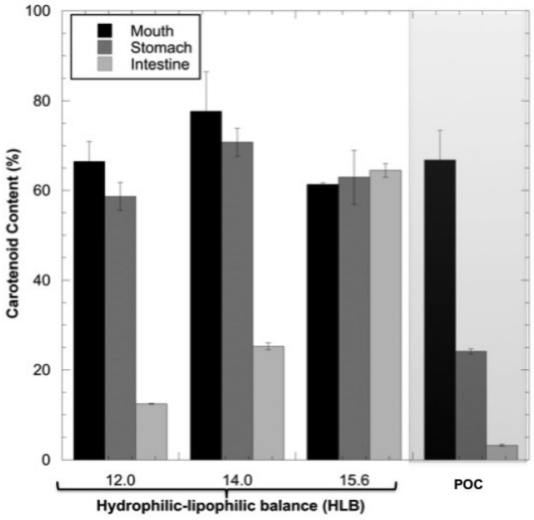

Fig. 3(B). Influence of HLB value on carotenoids stability during in vitro digestion of nanoemulsions. Bars represent the standard deviation of two replicated experiments made in triplicate.

We found that in the stomach phase droplets remain stable to aggregation caused by the strong steric repulsion generated by the hydrophilic head groups of the non-ionic surfactant coating (Xia *et al.*, 2015). Qian *et al.* (2012) reported similar results for β-carotene nanoemulsions using Tween 20 as emulsifier. Previous studies have also shown that lipid droplets coated by non-ionic surfactants are relatively stable to exposure in simulated oral and gastric fluids (Golding and Wooster, 2010; Nik *et al.*, 2012).

The drastic increase in particle size of the emulsions in simulated small intestinal system may be because the nonionic surfactant molecules originally adsorbed on the surface of the lipid droplets can be displaced by other molecules with active surface, such as, bile and lipase (present in the simulated gastrointestinal system) salts, phospholipids and free fatty acids and monoacylglycerols. The changes in the composition, structure and properties of the interfacial layer surrounding lipid droplets, may lead to decrease of stability (Qian *et al.*, 2012). This suggests that ingested nanoparticles may not be nanoparticles when they reach the small intestine. In addition, for non-emulsified paprika a drastic decrease of particle size from the stomach (1205.0 nm) to the intestinal phase (97.8 nm) is shown in Fig. 2(A). Sample dilution, mechanical agitation and/or changes in the solution composition can lead to the dissociation of flocculated

droplets within the intestinal phase (Dickinson, 2003). Sample dilution weakens any depletion attraction between droplets and may also weaken bridging interactions (Salvia-Trujillo *et al.*, 2013). Liu *et al.* (2012) showed that interfacial components of β-carotene emulsion are altered as they pass through different regions of the GIT by changing the ability of digestion components to absorb for absorbing the droplets or altering the droplet breakup and coalescence.

During carotenoids digestion, these must be released from the food matrix to be emulsified in the lipid phase of chyme, and solubilized in mixed micelles, to be accessible for uptake by absorptive epithelial cells (Failla and Chitchumronchokchai, 2005). Fig. 3(A) and 3(B) show the chemical stability of carotenoids of conventional and nanoemulsions, respectively, in comparison with the behavior of POC at the three phases of the simulated gastrointestinal tract. The best carotenoid stability was found at the highest HLB for the three gastrointestinal phases, where the maximum degradation of carotenoids for CE and NE occurred in the mouth phase being 23 and 19% respectively, while in the stomach and intestinal phase this degradation was negligible. The highest carotenoid degradation was at the intestinal phase when the interface was more lipophilic.

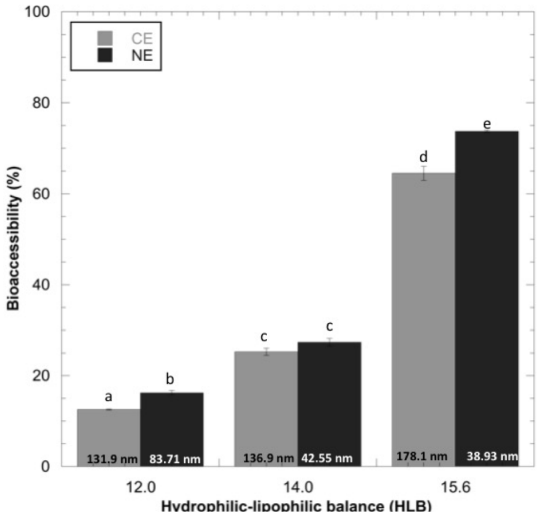

Fig. 4. Influence of HLB value on the bioaccessibility of paprika carotenoids in conventional and nanoemulsions. Means with different letters are significantly difference ($p < 0.05$). Bars represent the standard deviation of two replicated experiments made in triplicate.

Carotenoids final concentration for POC was 3% in the simulated gastrointestinal tract. Similar results were obtained by Tyssandier *et al.* (2002) who obtained a final concentration of 5.6, 4.6 and 2.0% for β-carotene, lutein and lycopene, respectively, in the duodenum.

It was observed that after two incubation hours in simulated gastric phase the concentration of carotenoids in the emulsified systems was almost the same. Qian *et al.* (2012) found a small degradation of β-carotene in nanoemulsions stabilized with non-ionic surfactant (Tween 40) in the mouth and stomach phases. They reported that the degradation of β-carotene was not appreciable under highly acidic gastric conditions, because the digestion time was of two hours. For both processes the best barrier against carotenoids degradation was obtained as the HLB value was increased. Recently Pascual-Pineda *et al.* (2015) reported that the rate of chemical degradation of paprika carotenoids encapsulated within nanoemulsions was less as the HLB value was increased. The droplet interface consists of a narrow region, which surrounds the emulsion droplet and contains a mixture of oleoresin, water and emulsifier molecules. We observed less carotenoid degradation for NE in comparison to CE at the same HLB. This was attributed to the smaller particle size generated by the cavitation process. At a HLB of 15.6, initial particle sizes of 178 and 38.9 nm were obtained for CE and NE, respectively. The higher stability of carotenoids was found at the smaller particle size, possibly because the interface at smaller size reduces the oxygen diffusion, due the curvature interface was similar to the curvature of surfactant monolayer, which provides a barrier against degradation of carotenoids (Pascual-Pineda *et al.*, 2015).

Fig. 4 depicts the effect of the interface composition on the bioaccessibility of carotenoids emulsions for coarse emulsions and nanoemulsions. The bioaccessibility was within the ranges of 12.5 to 64.6 and 16.2 to 74%, for CE and NE, respectively. Overall, the bioaccessibility of carotenoids increased as the HLB increased and particle size of emulsions decreased. The highest bioaccessibility (74%) was obtained at the highest HLB, when the interface was more hydrophilic at the smallest particle size. It can be observed that the available carotenoids to be absorbed by the epithelial cells are strongly influenced by emulsifier composition, caused by the protection provided by the molecular interactions of emulsifier at the interface. The higher surface area of the smaller droplet size makes that the lipase performs better

on the surface of lipid droplets. This behavior was reported by Armand *et al.* (1992) and Liu *et al.* (2012). The β-carotene content in the micellar phase increased from 5 to 10% as the droplet diameter decreased from 18 to 0.7 μm (Wang *et al.*, 2009). We found that bioaccessibility can be influenced by a synergistic effect of energy interactions and droplet size. When the bioaccessibility is driving by energy interactions, the affinity of lipase molecules for various chemical components depends on the polar or non-polar interface. In this work, the surface with more lipophilic moieties decrease of the bioaccessibility, which possibly was promote by the binding decrease of lipase to the interfacial layer, which caused a decrease of the digestion of carotenoids. Therefore, an increase of contact surface area at the nanometric scale, promotes a better performance of the lipase at the interface when this has more hydrophilic moieties.

Conclusions

Simulated gastrointestinal tract is a model that provides important information about the bioaccessibility of carotenoids in emulsified systems. The HLB had a pronounced effect on the bioaccessibility of paprika carotenoids. Carotenoids degradation can be effectively avoided within the emulsions during the gastrointestinal transit by controlling the polarity of the interface. Therefore, bioaccessibility depends on the structure and interfacial composition, which can be improved by controlling particle size and energy interactions. This knowledge can be used to design food delivery systems to improve the bioaccessibility of lipophilic bioactive components.

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Nomenclature

CE	Conventional emulsions
NE	Nanoemulsions
POC	Paprika oleoresin carotenoids
HLB	Hydrophilic-lipophilic balance
GIT	Gastrointestinal tract
PDI	Polydispersity index
C^R	Red carotenoids fraction (mg mL^{-1})
C^Y	Yellow carotenoids fraction (mg mL^{-1})
C^T	Total carotenoids content (mg mL^{-1})
$C_{Micelle}$	Total carotenoids in the micelle fraction (mg mL^{-1})
$C_{Emulsion}$	Total carotenoids in the emulsion (mg mL^{-1})

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