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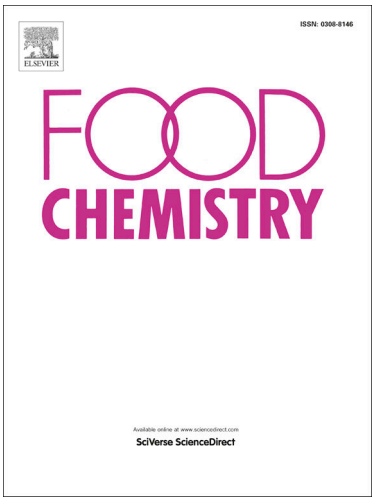


Review

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# Are quinoa proteins a promising alternative to be applied in plant-based emulsion gel formulation?

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**Abstract**

Emulsion gels are structured emulsion systems that behave as soft solid-like materials. Emulsion gels are commonly used in food-product design both as fat replacers and as delivery carriers of bioactive compounds. Different plant-derived proteins like soy, chia, and oat have been used in emulsion gel formulation to substitute fat in meat products and to deliver some vegetable dyes or extracts. Quinoa protein isolates have been scarcely applied in emulsion gel formulation although they seem to be a promising alternative as emulsion stabilizers. Quinoa protein isolates have a high protein content with a well-balanced amino acid profile and show good emulsifying and gelling capabilities. Unlike quinoa starch, quinoa protein isolates do not require any chemical modification before being used.

The present article reviews the state of the art in food emulsion gels stabilized with vegetable proteins and highlights the potential uses of quinoa proteins in emulsion gel formulation.

**Keywords:** vegetable proteins, gelling properties, emulsifying properties, quinoa.

## 1. Introduction

Emulsion gels could be described as colloidal soft-solid materials that behave both as emulsion and as gel. These systems can be categorized either as emulsion-filled gels, when the oil droplets are embedded into a continuous cross-linked biopolymer network, or as emulsion particulate gels, when aggregated droplets form a gel network of their own (Dickinson, 2012, 2014). Due to the difficulty in identifying these two idealized models in food systems, emulsion gels are considered a hybrid between them (Dickinson, 2012, 2014). Besides, emulsion droplets can be divided into active and inactive fillers according to their interactions with the gel matrix. Active fillers are connected by noncovalent and/or covalent bonds to the gel network through emulsifiers. On the other hand, inactive fillers have little affinity to the molecules of the gel matrix (Dickinson & Chen, 1999). Emulsion gels could also be classified into three types according to their morphological properties: bulk emulsion gels, emulsion gel particles, and fluid emulsion gels. In turn, fluid emulsion gels are subdivided into gel-like Pickering emulsions and disrupted emulsion gel systems (Lin, Kelly, & Miao, 2020).

The initial stage in emulsion gel formulation involves mixing the gelling agent, the emulsifier and oil. The second step may be either aggregation of the emulsion droplets or gelation of the continuous phase by different gelling methods (Dickinson, 2012; Lin, Kelly, Maidannyk & Miao, 2020). The choice of the appropriate gelling method (heating, acidifying, or using enzymes) depends on the matrix components and the applications of the prepared emulsion gels

The emulsifying or gelling agent may be a polysaccharide and/or a protein. When a polysaccharide is used, the emulsion gel formulation generally involves a mechanical or ultrasonic homogenization and some ions, such as calcium and magnesium, should be added to induce the gelling process. The most widely used neutral polysaccharides are agar and inulin (Paradiso et al., 2015), while the most frequent anionic polysaccharides are k-carrageenan (Koç et al., 2019), gellan gum (Lorenzo, Zaritzky, & Califano, 2013), and pectin (Soltani & Madadlou, 2015). Regarding proteins, the most commonly used as stabilizers are animal-derived proteins such as whey protein isolate (WPI) (Balakrishnan, Nguyen, Schmitt, Nicolai, & Chassenieux, 2017; Leon, Medina, Park, & Aguilera, 2018; Liang et al., 2020), and bovine gelatin (Satapathy et al., 2015).

Following the current trend in food emulsion design, more sustainable ingredients should be used (McClements, 2020) and plant-derived proteins may be considered as an option to emulsify or stabilize emulsion gels (Berton-Carabin & Schroën, 2019; McClements, Bai, & Chung, 2017). Legume proteins such as soybean, pea, chickpea, lentil, bean (Ben-Harb et al., 2018; Burger & Zhang, 2019; Sharif et al., 2018), and some species of the family *Amaranthaceae* and *Polygonaceae* such as amaranth, buckwheat, and quinoa derived proteins (Dakhili, Abdolalizadeh,

Hosseini, Shojaee-Aliabadi, & Mirmoghtadaie, 2019; Gurbuz, Kauntola, Ramos Diaz, Jouppila, & Heinonen, 2018; Janssen et al., 2017; López-Castejón, Bengoechea, Díaz-Franco, & Carrera, 2020) have been used as emulsifiers.

The role of quinoa proteins in stabilizing emulsions has been studied, but the application of quinoa proteins in emulsion gels has been scarcely investigated (Ingrassia, Busti, & Boeris, 2022) and has not been systematically described. Quinoa proteins seem to be a promising alternative due to their well-balanced essential amino acid composition. Over the last few years, quinoa (*Chenopodium quinoa* Willd.) has been revalued as a result of its interesting nutritional features (Ceyhun Sezgin & Sanlier, 2019). The chemical composition of quinoa is shown in **Table 1**. Regardless of the origin or variety, quinoa seeds represent a good protein source, having on average a 14% protein with high content in lysine and histidine (Elsohaimy, Refaay, & Zaytoun, 2015; Mota et al., 2016; Pereira et al., 2019; Wang, Zhao, & Yuan, 2020). Moreover, quinoa provides dietary fiber, unsaturated fatty acids, such as α-linolenic, and polyphenolic compounds, which have been shown to play an important role in disease control and to have an attractive antioxidant activity (Pellegrini et al., 2018).

**Table 1**

Quinoa proteins have been shown to possess not only nutritional value but also some functional and physicochemical properties that facilitate the processing and development of food products. Several studies deal with the emulsifying ability and emulsion stability of quinoa proteins (Mir, Riar, & Singh, 2019a; Shen, Tang & Li, 2021; Shi, Fidelis, Ren, Stone, Ai, & Nickerson, 2020), as well as their gelling capabilities (Kaspchak et al., 2017; Ruiz, Xiao, Van Boekel, Minor, & Stieger, 2016) since these properties are key parameters which directly influence the product behavior (Dakhili et al., 2019; Elsohaimy et al., 2015).

This work aims to summarize the state of the knowledge in food emulsion gels, focusing on vegetable protein emulsion gels as well as to review quinoa protein properties as a future target for the study of gelled emulsions. Quinoa proteins are characterized by analyzing their composition and emulsifying and gelling capacities compared to other vegetable sources. Furthermore, food applications of emulsion gels with the addition of vegetable derivatives are also discussed.

## 2. Emulsion gels stabilized with vegetable proteins

Destabilization phenomena like Ostwald ripening, droplet flocculation, and coalescence can occur in food emulsion gels (Mc Clements et al., 2017). In order to avoid these processes, proteins, polysaccharides, and surfactants are used. Some vegetable proteins seem to be useful in the formulation and stabilization of emulsion gels. Depending on the vegetable source, proteins can

improve the nutritional profiles of the products to which they are incorporated as well as their physicochemical and technological properties (Jiménez-Munoz, Tavares, & Corredig, 2021; Muñoz-González et al., 2019).

Soy protein isolates (SPI) are the most widely used protein components in emulsion gel design, because of their well-known emulsifying and gelling capacity. The gelation process of soy protein-stabilized emulsion gels is generally induced by acid or enzyme treatment, glucono-δ-lactone and transglutaminase being the most common agents (Li, Kong, Zhang, & Hua, 2012; Luo et al., 2019; Qin et al., 2017; Tang, Chen, & Foegeding, 2011, Tang, Luo, Liu, & Chen, 2013; Wang, Luo, et al., 2018; Yang, Liu, & Tang, 2013).

The nature of the interactions between the emulsifiers/stabilizers and the biopolymeric network, the droplet volume fraction, and the matrix characteristics are important factors that determine the structure and rheological properties of emulsion gels (Dickinson, 2013, 2014; Lorenzo, Zaritzky, & Califano, 2018).

As mentioned above, droplets can behave as active or inactive fillers. As the concentration of the droplets with active behavior increases, the gel strength increases; thus the dispersed oil phase not only behaves as space filler but also contributes to building up the structure of the gel matrix by interactions between proteins at the droplet surface and proteins in the gel matrix (Dickinson & Chen, 1999). In transglutaminase-set soy emulsion gels, a change in the oil volume fraction from 0.2 to 0.6 resulted in an increase in the storage modulus from about 200 to 8000 Pa (Yang et al., 2013). In acid-induced SPI emulsion gels, an increase in the oil volume fraction from 0.2 to 0.4, led to an in increase in the storage modulus from 100 to 1600 Pa and to a decrease in the gelation time by about 80 min (Li et al., 2012). Pre-aggregation of SPI emulsion gels with the addition of $Ca^{2+}$ concentrations between 2.5 and 7.5 mM also promoted stronger and denser gel structures. However, the gel became weaker (Wang, Zeng et al., 2018) over 10 mM. This probably happens since $Ca^{2+}$ ions make droplets behave as an inactive filler, interacting weakly with the protein gel matrix and thus causing a decrease in storage modulus values (Chen, Dickinson, Langton, & Hermansson, 2000).

Concerning the size and content of protein aggregates, Wang et al. (2017) demonstrated that both size and content of proteins improved the storage modulus in $CaSO_4$-induced SPI emulsion gels. An enhancement in G′ of about 1.5-fold resulted from an increase in soy protein aggregate size from 875 to 1455 nm, while an enhancement of around 1.8-fold was observed when the soluble aggregate content was increased from 0% to 100%. Water holding capacity was also enhanced by approximately 12% and 20% by increasing the size and content of soy protein aggregates, respectively. Besides, both the size and content of the protein aggregates decreased oil droplet size significantly, from 2.1

to 1.2 μm. At constant volume fraction, mean droplet diameter reduction increases the droplet surface-area-to-volume ratio and the particle amount; thus, the droplet-droplet interactions become more important as they are closely packed together (Tatar et al., 2017). A significant reduction in oil droplet size in emulsion gels formulated with sonicated SPI dispersions was also observed (Paglarini, Martini, & Pollonio, 2019).

Another vegetable source, chia (*Salvia hispanica* L.), could also be used to develop and stabilize emulsion gels. Emulsion gels with chia flour showed thermal stability and water holding capacity without any release of exudate after heating. As regards textural properties, chia gelled emulsions had a higher puncture force (1.43 ± 0.07 N) than those prepared with soy proteins (0.34 ± 0.02 N) and higher gel strength value (3.12 ± 0.40 J) than those with soy protein samples (0.79 ± 0.07 J). In all cases, 2% alginate was used as a gelling agent (Muñoz-González et al., 2019). The same conclusions were reached by Pintado, Ruiz-Capillas, Jiménez-Colmenero, Carmona, & Herrero (2015). Their results showed negligible syneresis and better thermal stability in emulsion gels prepared with chia flour and the addition of casein, alginate, or gelatin as gelling agents. The highest puncture force was ~1 N and the highest gel strength ~1.6 J for those emulsion gels prepared with chia seed and alginate (0.75%). These results may be due to the presence of proteins and mucilage with water-binding capacity and emulsifying and gelling ability that contribute to high emulsion gel stability, puncture force, and gel strength value in chia emulsion gels (Muñoz-González et al., 2019).

Pea proteins have also been studied in emulsion gels formulation by thermal, acid, and enzyme treatments. Ben-Harb et al. (2018) designed emulsion gels with either pure pea proteins or mixed pea-milk proteins. After thermal treatment, pure pea emulsion gels had a higher G´ value than mixed ones; around 3500 Pa for an emulsion gel of pea proteins and about 2200 Pa for mixed emulsion gels, both with 14.8% of protein content. For acid and enzyme treatments, G´ values were even higher, around 12400 Pa for pure pea emulsion gels and about 5800 Pa for mixed systems. These authors also demonstrated that fat addition to protein suspensions contributed to strengthening the mechanical properties of the resulting emulsion gels. This strengthening was more pronounced under acid and enzyme treatments than under thermal treatment.

## 3. Quinoa as a stabilizer of emulsion gels

### 3.1. Quinoa proteins

Quinoa seeds have a high protein content ranging from 12 to 17% depending on their origin and variety (Table 1). More than 50% of the quinoa storage proteins are globulins (~50-60%), 11S

and 7S, and albumins (~13%); with low content of glutelins (~3-6%) and prolamins (López, Galante, Robson, Boeris, & Spelzini, 2018; Wang, Zhao et al., 2020).

The isolation procedure of quinoa proteins from defatted flour consists mainly of the solubilization of proteins in dilute alkali followed by isoelectric precipitation at acidic pH (Abugoch, Romero, Tapia, Silva, & Rivera, 2008; Elsohaimy et al., 2015). As reviewed by López et al., (2018) and Dakhili et al., (2019) the higher the pH, the higher the protein extractability, while the highest yield of precipitated protein was obtained at pH 4.5. These findings confirm that the isoelectric point of quinoa proteins seems to be similar to other vegetable sources (~4.5) (Alsohaimy, Sitouhy, & El-Masry, 2007). Thus, the composition of quinoa protein isolates (QPI) and their protein content are highly influenced by pH. Wang, Zhao et al. (2020) reported a protein content of 69 to 73% in QPI, similar to that informed by Abugoch et al. (2008), and slightly lower than the 84 and 88% previously reported by Mir et al. (2019a) and Elsohaimy et al. (2015), respectively. The starch content was around 20% for the QPI obtained by Wang, Zhao et al. (2020), and between 12 and 18% for the samples analyzed by Abugoch et al. (2008). These authors reported a mean ash content of around 2%. Besides, Aluko & Monu (2003) found an oil content of 9.25%, this value being higher than that reported by Wang, Zhao et al. (2020) (~2%).

Regarding the amino acid profile, quinoa protein has a well-balanced configuration with high levels of essential amino acids lysine and histidine, which makes it represents a valuable source to supplement cereals (Elsohaimy et al., 2015; Shi et al., 2020; Wang, Zhao et al., 2020) (*See* Supplementary Table). Apart from the proportion of essential amino acids, protein quality also depends on its digestibility. *In vitro* digestibility, which could be a limiting factor in protein utilization as a food ingredient, has also been analyzed for QPI. Sánchez-Résendiz et al. (2019) found digestibility above 95% for Black and Yellow QPI, these values being higher than those previously reported by Nasir, Pasha, Butt, & Nawaz (2015), which ranged from around 76 to 78% for different quinoa genotypes. The high protein bioavailability in quinoa seeds is partially due to the relatively low content of trypsin inhibitors which reduce protein enzymatic digestion and absorption (Hernández-Ledesma, 2019).

Quinoa protein presents functional properties such as pH-dependent solubility (Elsohaimy et al., 2015), foaming capacity comparable to soy protein (Abugoch et al., 2008), good emulsifying ability and stability, and gelling properties similar to amaranth protein (Kim, Renkema, & Van Vliet, 2001; Bejarano-Luján et al., 2010).

### 3.1.1. Emulsifying properties of quinoa proteins

Quinoa proteins have been shown to have techno-functional properties such as emulsifying and emulsion stabilizing capacities, which are key characteristics in the development and processing of food products (Dakhili et al., 2019). According to the method proposed by Pearce and Kinsella (1978), the emulsion activity index (EAI) for 1% w/v QPI at pH 10.0 and 8.0 was 2.37 and around 50 $m^2/g$, respectively (Aluko & Monu, 2003; Elsohaimy et al., 2015). These values were similar to those observed for 1% w/v SPI at pH 7.5, where an EAI value of 30 $m^2/g$ was observed (Ren et al., 2020). However, in other reports, higher EAI values were obtained for 0.1% w/v SPI, about 72 $m^2/g$ at pH 6.5 (Zhao, Dong, Li, Kong, & Liu, 2015), and about 116 $m^2/g$ for 0.1% pea protein concentrate (Zhao, Shen, Wu, Zhang, & Xu, 2020). Fig. 1A shows QPI emulsifying ability, expressed as the volume of the emulsion layer relative to the total volume of the mixture in the tube, as a function of the emulsification pH. An increase in the emulsification pH led to an increase in the emulsifying ability of QPI. Shen et al. (2021) studied the emulsifying ability of 7% w/v QPI dispersions at pH 5.0, 6.0, 7.0, and 8.0 and found the highest emulsifying ability, around 60%, at pH 8.0. Emulsifying ability values of about 60% and 65% were also found for 2% QPI dispersions at pH 9.0 and 11, respectively. However, emulsifying properties above 55% were found for the same QPI concentration at pH 3.0, 5.0, and even 7.0. QPI was also found to have an emulsifying ability significantly higher than that for protein isolates from album, which is another variety of *Chenopodium*. For album, the maximum emulsifying ability percentage was around 57% at pH 11 while for QPI, it reached 65% at the same pH (Mir et al., 2019a). Values of the emulsifying ability of 19.5% at pH 9 and 55% at pH 8 were observed for a protein-rich fraction of chia, obtained by dry fractionation (López, Galante, Raimundo, Spelzini, & Boeris, 2019).

The emulsion stability index (ESI), expressed as a ratio between EAI at the initial time and EAI at a certain time, of 1% QPI-stabilized emulsion was about 27%, 84%, and 76% at pH 6.0, 7.0, and 8.0, respectively at 30 min (Aluko & Monu, 2003). These values were similar to those determined for SPI-stabilized emulsions (about 85% at pH 7.0 and 47% at pH 6.5 at 30 min) (Zhao et al., 2015) and higher than those for pea protein concentrate emulsions (about 15% at pH 7.0 at 10 min) (Ren et al., 2020). Fig. 1B shows the effect of the emulsification pH on QPI emulsion stability, determined as the percentage ratio between the height or volume of the emulsified layer and the height or volume of the total content in the tube after a certain time or after thermal treatment at 80°C for 30 min. The values range from 81% to 99% for 1% w/v QPI at pH values from 3.0 to 7.0 at 30 min (Shi et al., 2020) and about 50% for 2% w/v QPI at a pH range from 3.0 to 11.0 after thermal treatment (Mir et al., 2019a). It can be seen that an increase in emulsification pH led to an increase in emulsion stability. QPI showed the highest emulsion stability at pH 8.0 and 7%. The minimum emulsion stability

percentage reported for QPI was 30%, with 7% of protein concentration and at pH 5.0 (Shen et al., 2021). For a protein-rich fraction of chia, values of ESI of about 14% at pH 9.0 at 24 h and 98.2% at pH 8.0 after heat treatment were observed (Lopez et al., 2019).

**Figure 1**

For high-intensity ultrasound treatment, emulsifying properties were improved as compared to native QPI. Mir, Riar, & Singh (2019b) observed that after 25 min sonication treatment, emulsifying ability and stability increased from ~60% and 50% to 95% and 90% respectively. This improvement may be related to an increase in solubility, surface hydrophobicity, and conformational changes which could make more proteins available to be adsorbed at the oil-water interface and to form more stable films.

In addition, Shen et al. (2021) investigated the effect of drying methods on QPI´s emulsifying properties in the pH range from 5.0 to 8.0. These authors found that freeze-dried proteins showed the highest emulsifying ability and stability at pH 8, compared with spray and vacuum drying methods. Emulsifying ability and stability decreased by around 17% and 34%, respectively for vacuum-dried proteins. Lower emulsifying properties could be related to lower solubility, which may be attributed to more aggregation of denatured proteins during the drying process. Cerdán-Leal, López-Alarcón, Ortiz-Basurto, Luna-Solano, & Jiménez-Fernández (2020) found that freezing–lyophilization favors the formation and stabilization of emulsions with QPI, preventing their coalescence.

Besides the ultrasonic and drying methods, chemical modification techniques have been recently used to modify the functional properties of quinoa proteins. Teng et al. (2021) showed that glycosylation through Maillard reactions improved the EAI of QPI from 0.236 to 0.499 $m^2/g$ after mannose glycosylation and from 0.192 to 0.447 $m^2/g$ for xylose glycosylation.

The presence of an enzymatic hydrolysate can improve not only the biological properties but also the functional properties of food systems. In particular, emulsifying properties can be improved by the addition of hydrophobic amino acid residues. Shi, Hao, Teng, Yao, & Ren (2019) studied the EAI and ESI of the quinoa protein hydrolysates obtained using papain, pepsin, and pancreatin (maximum degree of hydrolysis: 16%, 37%, and 20% respectively) for 0, 30, 60, 90, and 120 min in the pH range from 3.0 to 8.0. EAI and ESI values significantly decreased after hydrolysis treatment, except in the case of the treatment with pancreatin at pH 5.0 and 6.0. Aluko & Monu (2003) found the same behavior in EAI values for QPI treated with alcalase for 240 min (48% hydrolysis degree). However, the emulsion stability was higher than that for QPI without treatment. Moreover, these researchers studied the emulsion stability in 10 kDa and 5 kDA permeate fractions of QPI treated with alcalase; the fractions of 10 kDa permeate at pH 4.0 and 5.0 and 5 kDa at pH 5.0 and 6.0 showed

higher emulsion stability than QPI without treatment. This improvement in emulsion stability is in agreement with the findings by Li, Da, Li, Xue, & Zang (2018), who reported that after QPI was treated with alcalase, the surface hydrophobicity and oil absorption capacity increased whereas water holding capacity decreased.

More recently, Daliri et al. (2021) showed that pancreatin-prepared quinoa protein hydrolysates (degree of hydrolysis: 19.17% after 180 min) had better emulsifying properties than quinoa concentrates at all pH values evaluated (from 3 to 8), whereas ESI was lower.

### 3.1.2. Gelling properties of quinoa proteins

Food texture and sensorial perception depend on the gel-forming ability of proteins. The gelling process could be induced by pH adjustments, heating treatments, and some enzymatic processes. The storage modulus (G′) of a QPI heat-set gel system as a function of the gelation pH is shown in Fig 2A. The lower the gelation pH, the higher the storage modulus, about 30000 Pa. An increase in protein concentration did not significantly modify the G′ value at acidic pH. However, at a neutral pH, a protein concentration of 15% led to enhanced G′ (Kaspchak et al., 2017). Ruiz et al. (2016) studied the influence of the extraction pH on the heat-induced aggregation, gelation, and microstructure of QPI. Quinoa seed protein extraction was performed by adjusting pH at 8.0, 9.0, 10.0, and 11.0, followed by acid precipitation at pH 4.5. Protein yield and denaturation were found to be lower as pH decreased; however, heating 10% w/v QPI extracted at 8.0 and 9.0 pH resulted in more aggregation and denser structured semi-solid gels. Suspensions of quinoa protein extracted at pH 10 and 11 failed to form gels. This behavior was correlated with a higher particle size for QPI obtained at pH 9 and pH 8 compared to that obtained at pH 10 and 11. Gelation temperature was around 70ºC for pH 8 and 9, while at 10 and 11 the G′ values started to increase after cooling. Quinoa showed to be able to form stronger gels, compared to other vegetable sources such as 11% w/v SPI, with G′ values up to 830 Pa at pH 7.0 (Kim, Renkema, & Van Vliet, 2001), or 12% w/v amaranth protein concentrates with G′ values of 877 Pa at pH 9.0 (Bejarano-Luján et al., 2010).

In addition to the influence of the extraction pH, the effect of divalent ions on the heat-set gelation of QPI at 10% and 15% (w/v) was studied. At pH 3.5, the addition of $CaCl_2$ and $MgCl_2$ led to significantly lower G′ values; for example, $Ca^{2+}$ showed G′ at about 10000 Pa and 6000 Pa at a protein concentration of 10% and 15% respectively. The addition of $MgCl_2$ resulted in lower G′ values (less than 3000 Pa at 10%), but increasing the concentration of proteins to 15%, G′ values reached around 20000 Pa. Similar results were obtained with samples without divalent ions at the same protein concentration. However, the highest G′ values were obtained for samples with 10 % of protein content in the absence of salt (around 30000 Pa). At pH 7, G′ values were considerably lower,

both at a protein concentrations of 10% and 15% without salt, compared with the values at acidic pH. Moreover, the addition of $CaCl_2$ and $MgCl_2$ was not suitable, resulting in gels with less strength than the samples without ions. G′ values were less than 50 Pa at 10% for both salts. Kaspchak et al. (2017) reported G′ values less than 900 Pa for CaCl2 at 15%, and less than 500 Pa for MgCl2 at the same concentration. These authors also found a gelation temperature variation for a QPI extract from around 60 °C to 90 °C, in agreement with the results reported by Ruiz et al. (2016).

Several authors have studied the influence of the drying methods on heat-induced quinoa protein gels. Compared to freeze-drying, spray and vacuum treatments resulted in weaker gels with low G′ and G′′ values, which may be related to poor protein solubility (Shen et al., 2021). Following the same trends, Wang, Dong et al. (2020), investigated the effect of different heat treatments on the gel-forming ability of QPI. These authors found that at least 12% concentration of protein isolates was needed to form firm gels with QPI when subjected to no thermal treatment or to microwave heating. When subjected to boiling or baking, the concentration of isolates had to reach 16% and 20% respectively to be able to gel. Steaming negatively affected the gelation properties and there was no gel formation, even at 20% concentration.

Mäkinen, Zannini, & Arendt (2015) studied the structural characteristic of 5% QPI cold-set gels obtained from quinoa protein suspension heat-treated (100°C for 15 min) at pH 8.5, 9.5, and 10.5. The G′ values obtained by acidification using glucono-δ-lactone are shown in Fig. 2B. When gels were observed in a confocal microscope, the gel obtained by acidification of the solution heat-treated at pH 8.5 was coarse with irregular clusters of particles. In contrast, images of gels formed from samples heat-treated at pH 10.5 showed a regular and fine structure. These findings were complemented with solubility and particle size distribution determinations of the heat-treated solutions. The solubility of samples heat-treated at pH 10.5 was higher than for those heat-treated at pH 8.5. Particle size distribution at pH values of high protein solubility (pH 3 and 8) showed similar behavior among the samples heat-treated at pH 8.5 and non-heat-treated. Nevertheless, the heating treatment produced a dissociation into small particles (diameters about 0.2–0.8 μm) and the presence of large aggregates (30-200 μm) bound by hydrophobic interactions. In a later work, Mäkinen, Zannini, Koehler, & Arendt (2016) studied the role of thiol groups and surface hydrophobicity in heat treatments. Disulfide bridges were disrupted at both pH 8.5 and pH 10.5; however, surface hydrophobicity at 15 min of heat-treatments at pH 10.5 was higher than that at 8.5, which shows a change in the structure, particle size distribution, and intermolecular interaction of QPI. Therefore, it is clear that the condition of denaturation influences the acid gelation properties.

It is important to note that quinoa protein partial hydrolysates also showed gelling capabilities. Galante, De Flaviis, Boeris, & Spelzini (2020) demonstrated that quinoa proteins hydrolysed by an *Aspergillus niger* serin peptidase at a low degree could be used for semi-solid system development.

**Figure 2**

### 3.2. Emulsion gels with quinoa

The role of quinoa protein in stabilizing emulsion gels has been scarcely studied. Ingrassia et al. (2022) developed an acid-induced emulsion gel fortified with quinoa protein concentrate and flour. These authors obtained soft and self-sustainable systems with a fluid retention capacity above 99% and good stability over a period of 28 days. Quinoa protein fortification promoted an increase in gel strength of about 3.5 N and improved the nutritional profile of emulsion gels, positioning them as possible saturated fat replacers in meat-based or non-meat-based products.

Only a few works have focused on quinoa starch granules. Starch is the major component of quinoa seeds, making up approximately 70% of the dry matter (Li & Zhu, 2018). Quinoa starch presents a low amylose amount (~6-8%), compared with other starches, and high amylopectin content with a considerable number of short chains and super-long chains (13-19%) (Li & Zhu, 2018). These structural features play an important role in many physicochemical properties of this seed and its derived food products. Li, Wang, & Zhu (2016) observed a gelatinization temperature range from 52.4 to 69.5 ºC, which is lower than that of buckwheat, sorghum (Wolter, Hager, Zannini, & Arendt, 2013), and amaranth starches (Singh et al., 2014). The degree of retrogradation for quinoa starch was higher than that for amaranth and lower than that for maize and wheat (Li & Zhu, 2018). As regards enzyme susceptibility, Li et al. (2016) found a value of around 70% in quinoa starch, indicating a hydrolysis rate much faster than for potato, yam, lentil, wheat, and oat (Hoover & Vasanthan, 1994). This suggests a significant amount of rapidly digestible starch fraction in quinoa, probably related to granule size and more surface area in contact with enzymes (Li et al., 2016).

Both native and modified starches have been used for several food applications, such as film manufacturing (Pagno et al., 2015) and ingredient encapsulation (Zhu, 2017). As regards chemically modified quinoa starch, octenyl and dodecenyl succinic anhydride have been used to change some physicochemical properties in order to improve Pickering emulsions and Pickering emulsion gel stabilization (Rayner, Timgren, Sjöö, & Dejmek, 2012; Matos, Timgren, Sjöö, Dejmek, & Rayner, 2013; Li, Xu, & Zhu, 2019; Lin et al., 2020; Kierulf et al., 2020). Li et al. (2019) studied Pickering emulsion gels stabilized with octenylsuccinate quinoa starch at different degrees of substitution and oil volume fraction. These researchers found that the higher the degree of substitution and oil volume

fraction, the higher the elastic modulus. G´ values increased from less than 100 Pa to more than 400 Pa when oil volume fraction increased from 40 to 70% respectively, at a 0.0286 degree of substitution. Systems with 50 to 70% of oil volume fraction showed gel-like behavior. Li, Zhang, Li, Fu, & Huang (2020) also developed Pickering emulsion gels to deliver lutein. These authors showed that at an oil volume fraction of 30% to 60% and a 0.045-degree of substitution, G´ values ranged from around 300 Pa to more than 700 Pa and presented gel-like features due to interfacial arrangement and packaging effect of particles. However, the same degree of substitution and 70% of an oil volume fraction of 70% resulted in an emulsion phase inversion.

In addition to quinoa starch, QPI nanoparticles have been used as an effective food-grade stabilizer for Pickering emulsions. Qin, Luo & Peng (2018) studied the effect of ultrasound on quinoa protein nanoparticle-stabilized Pickering emulsions. The ultrasound process improved the wettability and surface hydrophobicity of the nanoparticles and modified the type of intraparticle interaction. Compared to natural QPI-stabilized emulsions, sonicated particles exhibited a hexagonal array model arrangement with higher packing and adsorption of protein particles at the droplet interface. Sonicated quinoa protein particles exhibited better emulsifying properties. Cen, Yu, Gao, Feng, & Tang (2021) formulated high internal Pickering emulsions stabilized with ultrasonicated QPI nanoparticles with oil volume up to 75%, which were then gelled. Their results showed that ultrasound treatment increased QPI nanoparticles' hydrophobicity, making these particles effective in improving freeze-thaw stability of such high internal Pickering emulsion gels. Moreover, Kierulf et al. (2020) also developed an alkaline isolation method to extract starch from quinoa flour while retaining a high protein content. These authors demonstrated that this high-protein starch sample, with 2.7% protein content, had an emulsifying ability comparable to that of modified quinoa starches; thus, it could be used as a food-grade stabilizer in Pickering emulsion formulation.

## 4. Food applications of emulsion gels with the addition of vegetable derivatives

According to published results, emulsion gels are widely used in food products as they provide desired textural properties, improve shelf life, and can also effectively deliver some functional ingredients, such as vitamins and phenolic compounds among others (Brito-Oliveira, Bispo, Moraes, Campanella & Pinho, 2017; Lu et al., 2019). Moreover, emulsion gels have an interesting application in fat-reduced food design (Herrero & Ruiz-Capillas, 2021), due to their adjustable textural properties and lower fat digestibility rate (Guo, Ye, Bellissimo, Singh, & Rousseau, 2017; Mao, Miao, Yuan, & Gao, 2018). However, the development of this kind of products represents a huge challenge as they should look, feel, sound, and taste like the original versions so that consumers accept them

(McClements, 2020; Sha & Xiong, 2020). **Table 2** shows examples of food applications of emulsion gels prepared with vegetable sources.

**Table 2**

The use of different plant-derived ingredients as animal fat replacers in the formulation of emulsion gels represents an interesting alternative to improve fatty acid quality and to increase soluble and insoluble fiber, micronutrients, and antioxidants intake (Pintado et al., 2018).

Pintado, Herrero, Jiménez-Colmenero, & Ruiz-Capillas (2016) and Pintado, Herrero, Ruiz-Capillas, et al. (2016) found that chia flour has a great potential as saturated fat replacer in different meat products. These researchers examined the incorporation of emulsion gels with chia flour, olive oil and different gelling agents in reduced-fat frankfurters. Reformulated frankfurters had high levels of polyunsaturated fatty acids, mainly linolenic acid, and exhibited good stability to oxidation. Similar results were obtained by Botella-Martínez, Viuda-Martos, Pérez-Álvarez, & Fernández-López (2021) who developed reduced-fat frankfurters based on emulsion gels with buckwheat flour and hemp oil. Pintado et al. (2018) also studied emulsion gels with chia and oat as fat replacers in fresh sausages like *longaniza* type. In addition to fat quality improvement, these products increased calcium, magnesium, iron, and manganese contents as well as some amino acid content depending on the vegetable source. Chia mucilage was also found to be an effective fat replacer with the same satiating capacity as pork fat (Câmara et al., 2020). Emulsion gels with SPI, inulin and carrageenan were also used as pork fat replacers in meat products (Paglarini et al., 2018). Lucas-González et al. (2020) formulated emulsion gels with chestnut flour, chia oil, and gellan gum to be used as fat substitutes in reduced-fat pork burgers. Their results showed an increase in linoleic and linolenic acids, good stability to oxidation, as well as high sensory scores when compared to control samples (Lucas-González et al., 2020).

Quinoa flour was also used in similar product designs such as beef burgers (Baioumy et al., 2018), dry-cured sausages (Fernández-Diez et al., 2016), reduced-fat sausages (Öztürk-Kerimoğlu, Kavuşan, Tabak, & Serdaroğlu, 2020), frankfurters (Zapata & De La Pava, 2018) and other meat (Fernández-López, Viuda-Martos, & Pérez-Alvarez, 2021; Vargas Zambrano, Riera González, & Cruz Viera, 2019) and vegan food products (Felix, Camacho-Ocaña, López-Castejón, & Ruiz-Domínguez, 2021). However, quinoa was scarcely studied in emulsion gel formulation. Botella-Martinez, Pérez-Álvarez, Sayas-Barberá, Fernández-López, & Viuda-Martos, (2021) developed gelled emulsions elaborated with either chia or hemp oils and pseudocereal flours. The samples based on quinoa flour and chia oil along with those with buckwheat flour showed high emulsion stability,

even after freezing, positioning these systems as possible saturated fat substitutes for the formulation of novel food products.

Emulsion gels have been particularly studied in recent years regarding their role as delivery systems. Being emulsions, they are able to deliver lipophilic compounds, while their gel behavior confers good mechanical properties and stability (Torres, Murray & Sarkar, 2016). Moreover, due to the gel structure in the continuous phase, emulsion gels can deliver hydrophilic substances in food matrices, improving their dispersibility, stability, and bioavailability (Lu, Mao, Hou Miao, & Gao, 2019; McClements, 2020). For example, a possible alternative for artificial dye replacement in food products using plant-derived ingredients could be curcumin encapsulation and its incorporation in soy protein emulsion gels (Brito-Oliveira et al., 2017). Du et al. (2022) developed emulsion gels with SPI and corn oil as delivery systems for quercetin and showed an encapsulation efficiency above 80% and high bioaccessibility.

In addition, gelled double emulsions based on chia mucilage and different biopolymers were studied and showed to be effective in preserving total phenols and antioxidant activity of green tea extract over a storage period of more than a month (Guzmán-Díaz et al., 2019). Polyphenol extracts from grape, like gallic acid, catechin and epicatechin, were also incorporated in emulsion gels with a desirable content, (Muñoz-González, Ruiz-Capillas, Salvador, & Herrero, 2021) and these systems were then applied as animal fat replacers in frankfurter formulation (Pintado, Muñoz-González, Salvador, Ruiz-Capillas,& Herrero, 2021).

Curcumin and some flavor compounds, like vanillin, were also encapsulated in emulsions stabilized by quinoa starch modified with octenyl succinic anhydride, which shows quinoa starch potential for effective delivery of bioactive compounds (Li & Zhu, 2018; Marefati et al., 2017). Although most quinoa research has focused on starch granules as an option to encapsulate functional ingredients, quinoa proteins also seem to be suitable to develop emulsion gels with different food applications, as well as pharmaceutical and cosmetic usages (Dakhili et al., 2019).

## 5. Conclusions

The research results presented in this comprehensive review indicate strong evidence that emulsion gels have been attracting increasing interest in the food industry due to their potential to improve texture, expand shelf life, and deliver functional components in foodstuff.

The most relevant issues concerning the emulsion gels reviewed in this article are shown in Fig 3.

The data presented throughout this work confirm that the use of vegetable protein isolates as components of emulsion gels appears to be a good alternative to animal protein sources. This study particularly focused on the potential of quinoa protein as a sustainable ingredient of emulsion gels. The reviewed features of quinoa protein isolates (nutritional value and emulsifying and gelling capabilities) are comparable to those of other vegetable sources and support the use of quinoa protein as a promising option for the formulation of emulsion gels with suitable technological properties.

Thus, further research should be conducted to gain a deeper understanding of the behavior of this novel alternative source in emulsion gels.

**Figure 3**

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**Figure captions**

**Figure 1.** Effect of emulsification pH and protein concentration on the emulsifying ability (A) and emulsion stability (B) of quinoa protein isolates (QPI)"

**Figure 2.** Effect of gelation pH on storage modulus (G´) of heat-set gels (A) and cold-set gels (B) prepared with quinoa protein isolates (QPI)

**Figure 3.** Summary of the most relevant issues on the emulsion gels revised.

**Table 1**

Proximate chemical composition of quinoa (g per 100g, dry weight basis)

| Origin | Peru and North Canada[1] | Colombia[2] | Peru[3] | Spain, Bolivia and Peru[4] | Egypt[5] | Argentina[6] |
|---|---|---|---|---|---|---|
| Ash | 2.20 ± 0.01 | n.d. | 2.85 ± 0.01 | 2.25 ± 0.27 | 2.97 ± 0.02 | 2.01 ± 0.02 |
| Fat | 6.40 ± 0.15 | 5.47 ± 0.02 | 6.07 ± 0.05 | 5.50 ± 0.29 | 6.79 ± 0.19 | 6.31 ± 0.11 |
| Moisture | 8.60 ± 0.01 | 11.64 ± 0.01 | 9.02 ± 0.09 | 6.87 ± 0.18 | 9.68 ± 0.33 | 11.30 ± 0.05 |
| Proteins | 13.20 ± 0.30 | 13.46 ± 0.25 | 17.45 ± 0.69 | 12.63 ± 0.19 | 14.03 ± 0.25 | 12.10 ± 0.30 |
| Starch | 62.95 ± 0.30 | 68.30 ± 0.71 | 55.62 ± 1.35 | n.d | 72.15 ± 0.28 | 57.20 ± 0.60 |
| TDF | n.d. | n.d. | n.d. | 15.65 ± n.d. | n.d. | 10.40 ± 0.60 |

Abbreviations: Total Dietary Fiber (TDF); Not determined (n.d)

References:

[1] (Shi et al., 2020)

[2] Real variety (Contreras-Jiménez et al., 2019)

[3] Black and Yellow varieties (Sánchez-Reséndiz, Escalante-Aburto, Andía-Ayme, & Chuck-Hernández, 2019)

[4] White, White Real, Red Real and Black Real varieties (Pellegrini et al., 2018)

[5] (Elsohaimy et al., 2015)

[6] (Nascimento et al., 2014)

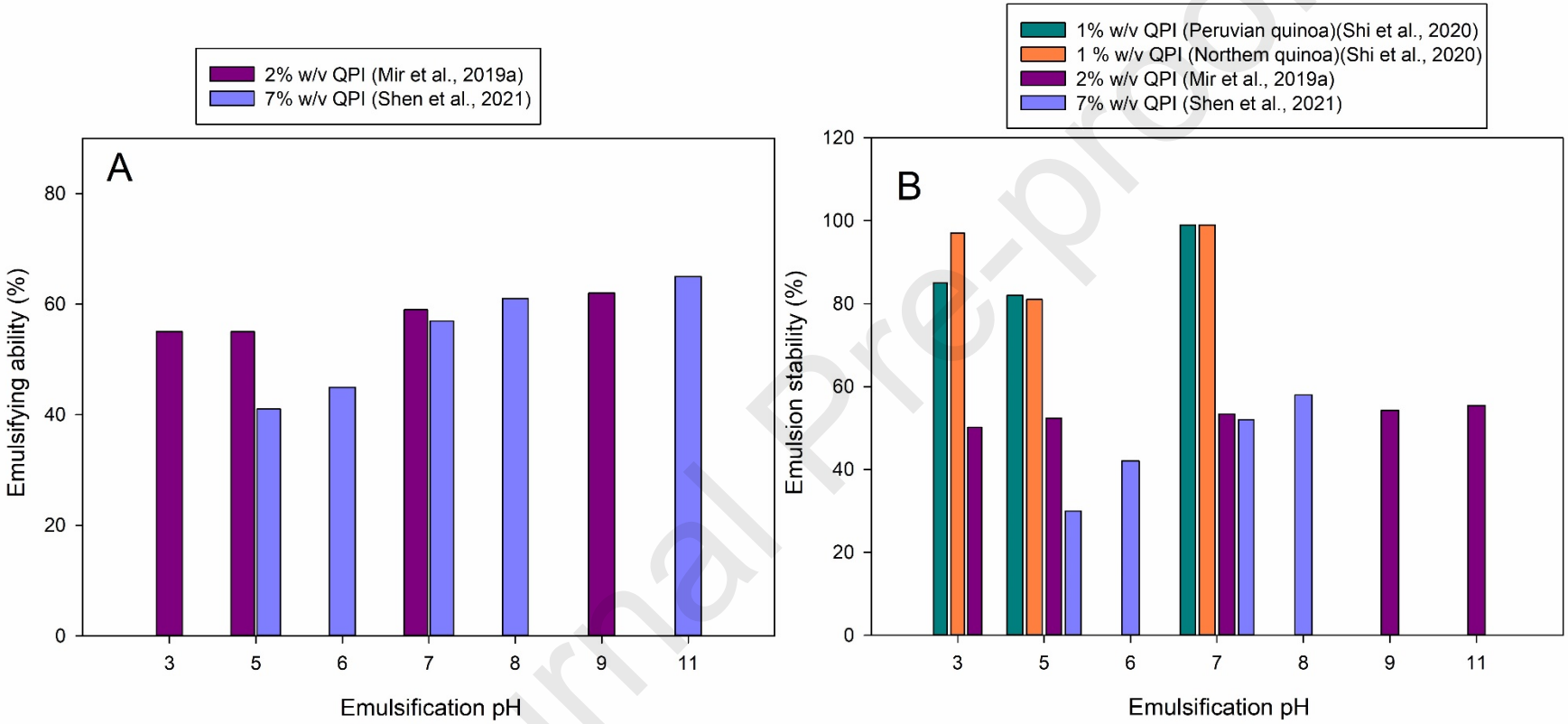
**Table 2**

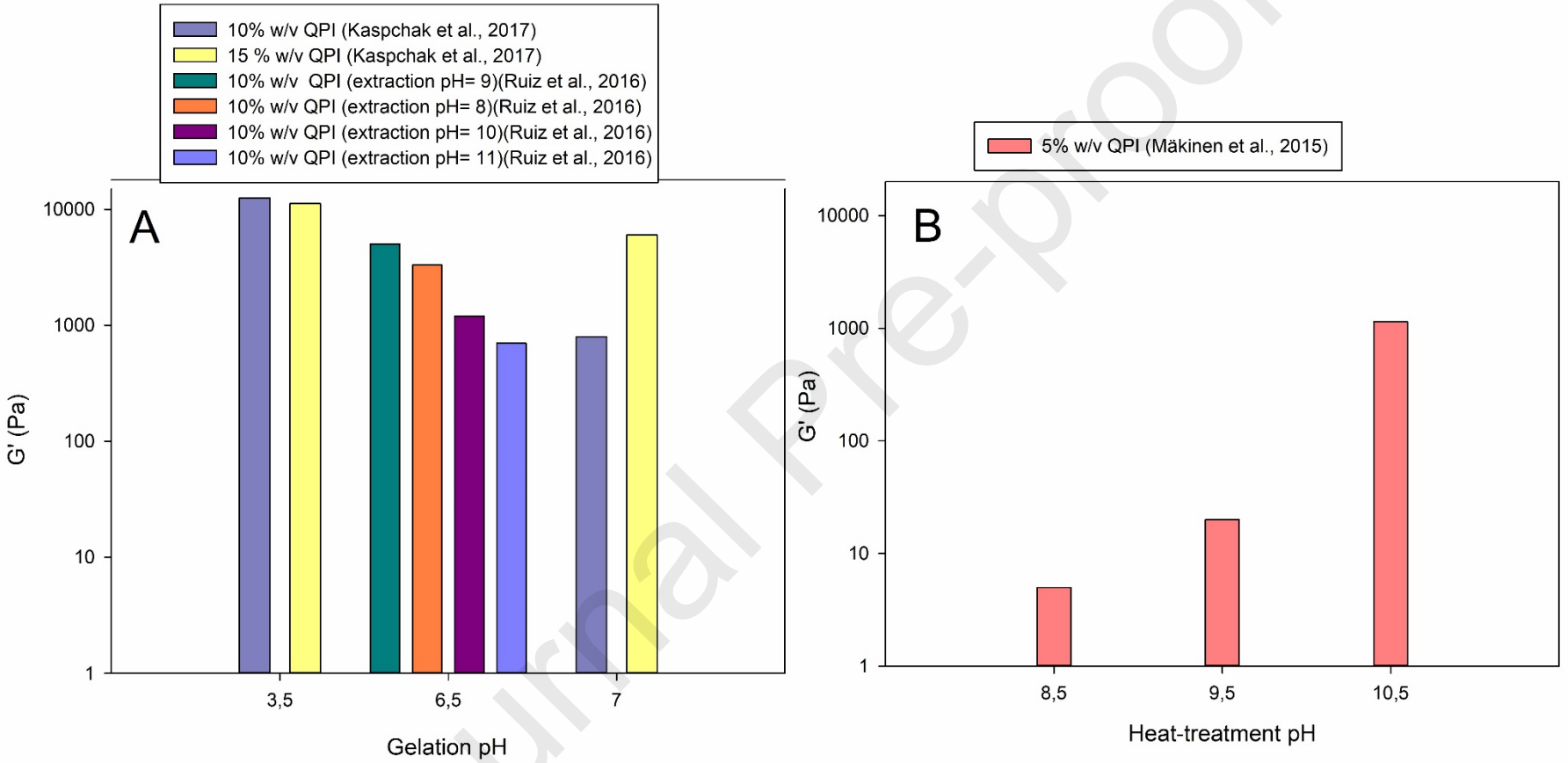
Food applications of emulsion gels with different vegetable sources

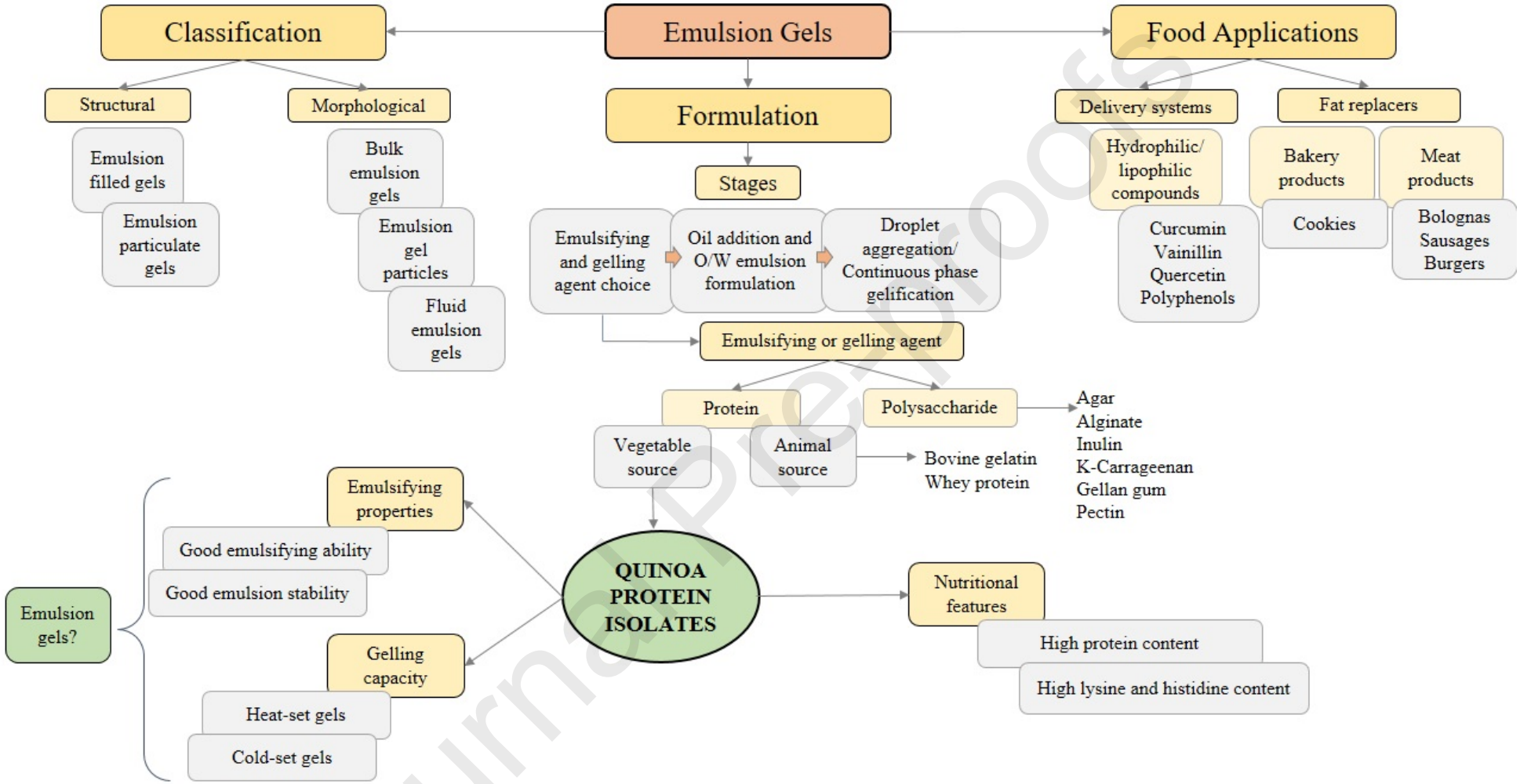
| Food application | Vegetal source | Ingredients | Products | Compounds | References |
|---|---|---|---|---|---|
| FAT REPLACERS | Chia | Flour<br>Olive oil<br>Alginate/ transglutaminase/ gelatin<br>Mucilage | Frankfurters<br>Sausages (*longaniza* type) | - | (Pintado, Herrero, Jiménez-Colmenero, et al., 2016; Pintado, Herrero, Ruiz-Capillas, et al., 2016)<br>(Pintado et al., 2018) |
| | | Olive oil<br>Alginate | Bolognas | | (Câmara et al., 2020) |
| | Oat | Oat bran<br>Olive oil<br>Alginate | Sausages (*longaniza* type) | - | (Pintado et al., 2018) |
| | Soy | SPI<br>Soybean oil<br>Sodium caseinate<br>Inulin<br>Carrageenan<br>Pectin<br>Soy lecithin | Meat products | - | (Paglarini et al., 2018) |
| | Buckwheat | Buckwheat flour<br>Hemp oil<br>Gellan gum<br>Pork gelatin | Frankfurters | - | (Botella-Martínez, Viuda-Martos et al., 2021) |
| | Chestnut | Chestnut flour<br>Chia oil<br>Gellan gum | Burgers | - | (Lucas-González et al., 2020) |
| DELIVERY SYSTEMS | Chia | Mucilage<br>WPI<br>Canola oil<br>κ-Carrageenan<br>Locust bean gum<br>Thixogum | - | Green tea extract | (Guzmán-Díaz et al., 2019) |

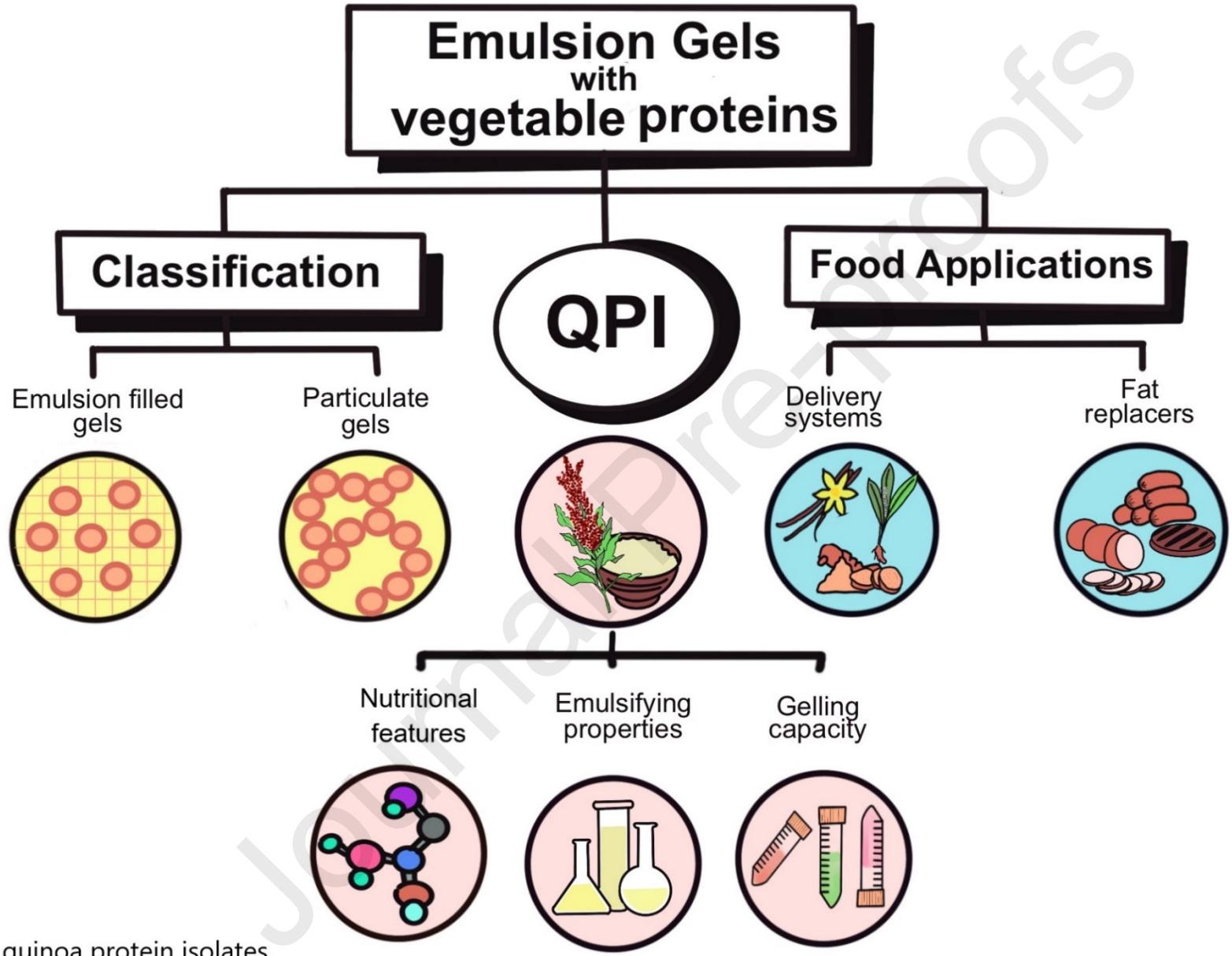
| | | | | |
|---|---|---|---|---|
| Soy | SPI<br>Solid lipid particles<br>Xanthan gum | - | Curcumin | (Brito-Oliveira et al., 2017) |
| | SPI<br>Corn oil<br>Dextran | - | Quercetin | (Du et al., 2022) |
| | SPI<br>Olive oil<br>Alginate | - | Polyphenolic extracts | (Muñoz-González et al., 2021) |
| | SPI<br>Olive oil<br>Alginate | Frankfurter | Polyphenolic extracts | (Pintado et al., 2021) |

Abbreviations: Soy Protein Isolate (SPI); Whey Protein Isolate (WPI)










QPI: quinoa protein isolates

**Highlights**

1) Quinoa proteins are encouraged due to their nutritional features and functional properties

2) Quinoa proteins have a potential for stabilizing emulsion gel systems

3) The usage of quinoa proteins is increasing in novel functional food development