Optimisation of the pH and boiling conditions needed to obtain improved foaming and emulsifying properties of chickpea *AQUAFABA* using a response surface methodology

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Abbreviations:

WHO: World Health Organisation; FAO: Food and Agriculture Organisation of the United Nations; UN: United Nations; CCW: Chickpea cooking water; RSM: Response surface methodology; CWR: Chickpea:water ratio; FC: Foaming capacity; EC: Emulsifying capacity; ES: Emulsion stability; FS: Foam stability; *Ch*: Chroma; *ΔE*: Colour difference from the control; S.D: Standard deviations; C.V: Coefficient of variation.

Abstract

Chickpea cooking water, also known as aquafaba, generated in our homes is generally discarded as waste. However, this valuable resource contains high quantities of proteins with excellent techno functional properties. The current paper aimed at optimising the pH and the cooking conditions needed to improve the foaming and emulsifying capacity of aquafaba, as well as the stability of the generated foams and emulsions using a response surface methodology. In general, lowering the pH of the aquafaba using lemon juice and decreasing the chickpea:water ratio during boiling resulted in increased functional properties. The validation tests carried out confirmed the overall adequacy of the response surface models in predicting the functionality of the aquafaba. Moreover, the chickpea cooking water obtained using the optimised conditions was used to develop meringues and mayonnaises, which were compared to those obtained using egg proteins.

Keywords: Chickpea cooking water, emulsifying capacity, foaming capacity, vegan meringue, vegan mayonnaise, response surface methodology.

Introduction

Agricultural practices in the near future will need to adapt and contribute to the mitigation of climate change, help preserve natural habitats, protect endangered species, and maintain high levels of bio- diversity. Protein production and consumption are pivotal to sustainability. Animal-derived proteins have been traditionally used to create edible foams, such as meringue, and emulsions like mayonnaise. However, producing 1 kg of animal protein requires approximately 6 kg of plant protein, rendering the large-scale production of animal protein one of the major drivers of diversity loss, climate change, freshwater depletion, and antibiotics resistance (Aiking, 2014). For this reason, together with an increase in the proportion of individuals choosing to follow a vegan diet (Radnitz et al., 2015), big efforts are being made in the development of novel foods which are suitable for those consumers who choose not to consume animal proteins for either cultural, traditional, or religious factors. Indeed, several products containing plant- derived proteins, utilised as meat or egg replacers, have been launched to the market over the last years and these include VeganEgg™ (Earth Island, USA), Økologisk Plantefars™ (Naturli' Foods, Denmark), and Just Mayo (Hampton Creek Foods, USA) among others. Beyond their nutritional and bioactive attributes, proteins are especially interesting ingredients because of their functionality, which allows them to be used as ingredients in the development of a wide range of products. Industrially relevant functional properties of proteins include foaming and emulsifying capacities. Emulsions consist of two immiscible liquids, generally water and oil, with one of them being dispersed as small droplets in the other. Mayonnaise belongs to the oil-in-water emulsions group and consists of oil droplets dispersed in an aqueous phase. It is possible to form an emulsion using pure oil and pure water together, but emulsions are thermodynamically unstable systems and water and oil rapidly separate into two phases. For this reason, in order to obtain emulsions that are stable for a reasonable period of time it is necessary to use stabilisers such as emulsifiers (or texture modifiers, weighting agents, or ripening inhibitors) which usually are amphiphilic molecules such as proteins that are capable of adsorbing to an oil-water interface and avoid aggregation (McClements, 2015). Moreover, liquid and solid foams are encountered extensively not only in the food industry (i.e. meringues, cakes, beer) but also in the cosmetic and textile industries. Liquid foams can be considered as an "emulsion" of, for example, air in water and there are many similarities between the mechanisms underlying stability of emulsions and foams (Hunter et al., 2008). The key building component of most common aerated structures are protein foams. The inherent instability of foams, which can be improved by modifying the pH or adding co-solutes such as modified starch, make their use in industrial applications very difficult (Asghari et al., 2016). Other strategies such as sonication also showed to improve foaming capacity (Meurer et al., 2020). However, for some applications, egg white proteins remain the only industrially available alternative.

Most common plant proteins used at industrial levels include those derived from pulses such as soy, chickpeas, and peas. Chickpeas (*Cicer arietinum* L.) are annual grain legumes native to the Mediterranean region and the major ingredient of many Mediterranean, Indian, and Middle Eastern dishes such as hummus or falafel. Chickpeas are generally soaked overnight and boiled to obtain both, a texture that is acceptable to consumers, and improve nutritional quality (Chenoll et al., 2009). Chickpea boiling water or aquafaba generated at domestic level or obtained from canned chickpeas is generally discarded. However, this resource is valuable not only because of its high content of health-promoting compounds such as polyphenols (Lafarga et al., 2019) but also because of its high protein content. Recently, Buhl et al. (2019) calculated the protein content of canned *aquafaba* as approximately 13 g/L. Over the last few years, a large number of recipes and online tutorials showed how chickpea boiling water could mimic functional properties of egg whites (Meurer et al., 2020). However, only a limited number of scientific studies assessed and measured the functionality of chickpea cooking water (CCW) or *aquafaba* (Damian et al., 2018; Mustafa et al., 2018; Stantiall et al., 2018; Meurer et al., 2020;

Buhl et al., 2019). Functional properties of proteins in solution depend on different factors including protein concentration and pH. The effect of the pH on the functionality of proteins is related to the modification of their net charge (Drago and González, 2001). For this reason, the current paper aimed at optimising the pH and domestic boiling conditions (chickpea:water ratio) required to improve the foaming and emulsifying properties of CCW using response surface methodology (RSM). The CCW obtained using the optimised conditions was used to develop an edible foam and an emulsion that were compared against ones made using egg white proteins.

Materials and methods

Plant material and chemical reagents

Chickpeas var. *Blanco lechoso* (Legumbres Luengo S.A., León, Spain), Azucarera® caster sugar (Azucarera Iberia S.L., Madrid, Spain), pasteurised egg whites (Guillén S.L., Valencia, Spain), Solimón® lemon juice (Derivados Citricos S.A., Murcia, Spain), and Solnatur™ sunflower oil (Borges Branded Foods S.L., Lleida, Spain) were purchased locally.

Experimental design and sample processing

Chickpeas were soaked in tap water for 24 h at a chickpea:water ratio (CWR) of 1:3 (w/v). After this period, the soaking water was discarded and the soaked chickpeas were boiled at different CWRs (Table 1) in a 3.9 L stainless steel cooking pot with lid. The total volume (chickpea plus boiling water) was kept constant by adding boiling water when needed. Boiling time was 190 min as this was the minimum time needed to reach tenderness for an adequate palatability and taste, according to Spanish eating habits. Once the chickpeas were cooked, the CCW or aquafaba was recovered and filtered using glass wool (SigmaAldrich Química S.L., Madrid, Spain). The pH of the CCW was adjusted after filtration using lemon juice added drop by drop (only a few drops were needed).

Protein determination was performed using the Bio-Rad Protein Assay Kit, which is based on the Bradford method (Bradford, 1976).

A response surface methodology, or more explicit a central composite face-centred design, was used to observe the effect of CWR and pH on the foaming capacity (FC) and emulsifying capacity (EC) of the CCW and on the stability of the generated foams and emulsions. To give a good estimation of the replicate error, five replicates at the centre point were conducted. Coded values for each parameter are shown in Table 1. Independent variables were CWR, which varied from 1:1.5 to 1:5.0, and pH, which varied from 3.5 to 6.5. The levels of each independent parameter were chosen considering conventional chickpea domestic cooking conditions and pH values of foods. The experimental runs were performed twice resulting in two blocks that were randomised and analysed in triplicate. Experimental data were fitted to a polynomial response surface, which was predicted by the following Equation (1):

$$Y = \beta_0 + \sum_{i=1}^{3} \beta_i X_i + \sum_{i=1}^{3} \beta_{ii} X_i^2 + \sum_{i=1}^{2} \sum_{j=i+1}^{3} \beta_{ij} X_i X_j \quad (1)$$

where Y is the dependent variable, β_0 is the centre point of the system, β_i , β_{ii} , and β_{ij} are the coefficients of the linear, quadratic, and interactive effect, and X_i X_i^2, and X_iX_j are the linear, quadratic, and interactive effect of the independent variables. The non-significant terms ($p < 0.05$) were deleted from the second order polynomial model after ANOVA analysis and a new ANOVA was performed to obtain the coefficients of the final equation for better accuracy. The optimisation was done as described by Odriozola-Serrano et al. (2009). Design Expert 7.0 software (Stat Ease Inc., MN, US) was used to generate the models that fit the experimental data and to obtain the response surface plots.

Foaming capacity and foam stability

To create foams, samples were homogenised using a T-25 digital ULTRA-TURRAX® homogeniser (IKA, Saufen, Germany) operating at 14,000 rpm for 1 min and the volume of foam generated was measured using a graduate cylinder. The volume of the graduated cylinder was 250 mL and the initial volume of liquid was 30 mL. FC was calculated as volume of foam generated as a percentage of the initial volume of solution and foam stability (FS) was expressed as the percentage of decrease of foam after 10 min described by Garcia-Vaquero et al. (2017). FC and FS were calculated using the equations:

$$FC\ (\%) = \frac{V_F - V_0}{V_F} \cdot 100$$

where V_0 is the volume of CCW before homogenisation and V_F is the volume of foam generated after homogenisation, and

$$FS\ (\%) = \frac{V_{10}}{V_F} \cdot 100$$

where V_{10} is the volume of foam remaining after 10 min and V_F is the initial volume of foam generated after homogenisation.

Emulsifying capacity and emulsion stability

Emulsions were generated by adding sunflower oil to the CCW at an oil:aqueous phase ratio of 3:2 (v/v) and homogenising the mixture using a T-25 digital ULTRA-TURRAX® homogeniser (IKA, Saufen, Germany) operating at 14,000 rpm for 2 min. Emulsions were then centrifuged at 14,000 rpm for 5 min using a Sigma 3–18 KS centrifuge (Sigma Laborzentrifugen GmbG, Osterode am Harz, Germany). EC was expressed as the percentage of emulsion generated and was measured as described by Garcia-Vaquero et al. (2017). To determine the emulsion stability (ES), the previously generated emulsions were heated at 85 °C for 10 min, cooled at room temperature for 5 min, and further centrifuged at 14,000 rpm for 2 min. ES was expressed as the percentage of emulsion remaining after centrifugation as determined by Garcia-Vaquero et al. (2017). EC and ES were calculated using the equations:

$$EC\ (\%) = \frac{V_E}{V_T} \cdot 100$$

where V_E is the volume of the emulsion layer after centrifugation and V_T is total volume inside the tube and

$$ES\ (\%) = \frac{V_H}{V_i} \cdot 100$$

where V_H is the volume of the emulsion remaining after heating and V_i is volume of emulsion subjected to heating.

Development of vegan edible foams and emulsions

Meringues were made as described by Stantiall et al. (2018). Each batch consisted of 160 g of either CCW (pH adjusted using lemon juice) or commercial pasteurised egg whites and 230 g of caster sugar. Foaming agents were whipped for 3 min at a medium speed (level 4) using an AM-7000 mixer (Orbegozo, Murcia, Spain) equipped with a 15-wire whip whisk. The caster sugar was then incorporated into the foam and whipped for a further 13 min period at maximum speed (level 6). Doses of 15 ± 1 g were weighed, placed on a baking tray, and each batch was baked at

100 °C for 75 min in a Rational SCC WE-101 oven (Rational AG, Landsberg am Lech, Germany). Meringues were allowed to cool at room temperature for 1 h before being placed in sealed polyethylene bags at room temperature until analysis took place. Mayonnaises were made following the recipe described by Di Mattia et al. (2015) with some modifications. Mayonnaise made using egg whites contained: 500 g of sunflower oil, 120 g of egg whites, 30 g of vinegar, and 1 g of table salt. Mayonnaise made using CCW contained 500 g of sunflower oil, 150 g of CCW (pH adjusted using lemon juice), and 1 g of table salt. Mayonnaises were prepared using a T-25 digital ULTRA-TURRAX® homogeniser (IKA, Saufen, Germany) in a two-step process. Eggs or CCW, vinegar or tap water, and salt were preliminary mixed at 100 rpm for 3 min. Once these ingredients were mixed, oil was added under homogenisation at 14,000 rpm for 5 min. Both emulsions were prepared in triplicate and stored at 4 °C until further analysis.

Colour

Colour readings of the meringues and mayonnaises were taken in triplicate using a Minolta CR-200 chroma meter (Minolta INC, Tokyo, Japan). Calibration was performed using a standard white tile provided by the manufacturer and the D65 illuminant, which approximates to daylight. CIE values were recorded in terms of L* (lightness), a* (redness/greenness), and b* (yellowness/blueness). Chroma (Ch) and difference from the control (ΔE), which compares foams or emulsions made using CCW and those made using egg white, were calculated using the equations:

$$Ch = \sqrt{a^{*2} + b^{*2}}$$

$$\Delta E = \sqrt{(L^*_{CCW} - L^*_{EW})^2 + (a^*_{CCW} - a^*_{EW})^2 + (b^*_{CCW} - b^*_{EW})^2}$$

where L^*_{CCW}, a^*_{CCW}, and b^*_{CCW} are the colour parameters of the foam or emulsion made using CCW and L^*_{EW}, a^*_{EW}, and b^*_{EW} the colour parameters of foams or emulsions made using egg whites.

Sensorial analysis

Sensory evaluation was undertaken approximately 24 h after meringues and mayonnaises were made with 40 untrained panellists recruited from IRTA Fruitcentre. Sensory evaluation was conducted in triplicate in a sensory laboratory with separate booths as described by Lafarga et al. (2018b). Briefly, samples were place on white polystyrene plates labelled with random codes and presented to consumers in a randomised order (mayonnaises were served spread on a slice of soft white bread). Each panellist assessed all the samples and was asked to indicate his or her opinion on the overall appearance, overall acceptance, flavour, and texture of the products using a 9-point hedonic scale (from 1: dislike extremely to 9: like extremely). The acceptability index was calculated using the following equation:

$$Acceptability\ index\ (\%) = \frac{X}{9} \cdot 100$$

where X is the mean of the scores obtained for overall acceptance.

Statistical analysis

Results are expressed as mean ± standard deviation (S.D.). Difference between samples were analysed using analysis of variance with JMP 13 (SAS Institute Inc., Cary, USA). Where significant differences were present, a Tukey pairwise comparison of the means was conducted to identify where the sample differences occurred. The criterion for statistical significance was $p < 0.05$.

Results and discussion

Effect of chickpea:boiling water ratio and pH on foaming capacity and foam stability

Table 1 shows the effect of the different conditions studied on the FC and FS of the CCW obtained varying CWR and pH values. Although the capacity of CCW to generate foams is now well known, and CCW is used for developing vegan foams such a meringues in many homes, scientific data assessing the FC and FS of CCW is scarce. This makes it difficult to compare the results obtained herein with those previously reported by other research groups. Mustafa et al. (2018) recently evaluated the foaming capacity and stability of aquafaba and obtained foaming capacity values ranging from 400 to 500%. However, the aquafaba assessed in that study was obtained from commercial chickpea cans and the method used for evaluating the foaming capacity was different to the one reported herein: whipping time varied from 1 min in the current study to 2–15 min in the paper authored by Mustafa et al. (2018). In addition, Stantiall et al. (2018) assessed the foaming ability of the cooking water of haricot beans, chickpeas, green lentils, and split yellow peas, obtained after boiling at a pulse:water ratio of 1:1.75 (w/ w), and reported foaming capacities ranging from 39 to 97%. The foaming ability of the chickpea cooking water in that study was 58%. Results obtained by Stantiall et al. (2018) are lower to the ones obtained herein. However, in that study the pH of the cooking water was not adjusted and lowering the pH of the solution is of key importance to obtain increased foaming abilities (Lafarga et al., 2018a). The effect of the pH on the FC of proteins is related to the modification of their net charge, which affects foam formation as well as film viscoelastic properties (Drago and González, 2001).

The FC could be fitted by a polynomial quadratic equation in terms of the studied conditions as described in Equation (2).

$$FC = 499.14986 - 14.60263 \cdot CWR - 44.75161 \cdot pH + 9.92381 \cdot CWR \cdot pH - 9.20985 \cdot CWR^2 - 1.60230 \cdot pH^2 \quad (2)$$

The statistical analysis indicates that the proposed quadratic model for FC was adequate ($p < 0.001$) with good determination coefficient ($R^2 = 0.9224$; Table 2). Both CWR and pH had a significant effect on FC ($p < 0.001$) as well as the combined effect of both parameters ($p < 0.05$). The linear term of both CWR and pH was negative, which indicates that an increase in these parameters will cause a decrease on FC. This is clear as a higher CWR means a lower protein content in the cooking water and both, protein content and pH are of key importance for good foaming abilities. Previous studies suggested that FC is higher at low pH values because of increased net charges on the proteins, which weaken the hydrophobic interactions but increase the flexibility of the proteins (Ragab et al., 2004). Kaur and Singh (2007) also linked good foaming abilities with flexible molecules that can reduce surface tension. Although foams are thermodynamically unstable as their decay results in decrease of the free energy, the kinetic mechanisms involved in their breakdown can be slow enough so that they can be considered as metastable for their application (Hunter et al., 2008).

In the present study, the ANOVA also indicated that the proposed two-factor interaction model for FS was suitable and showed a good determination coefficient ($p < 0.0001$; $R^2 = 0.9583$; Table 3). CWR and pH also affected FS ($p < 0.001$ and $p < 0.0001$, respectively). FS could be modelled by Equation (3).

$$FS = 139.70061 + 3.92381 \cdot CWR - 11.10397 \cdot pH - 2.50476 \cdot CWR \cdot pH \tag{3}$$

No significant lacks of fit were obtained in both cases, suggesting that they both fit properly for prediction across the design space. Both, the pH and the CWR were indirectly related to FC and FS values (Fig. 1).

Effect of chickpea:boiling water ratio and pH on emulsifying capacity and emulsion stability

Table 1 also shows the effect of the different conditions studied on the EC and ES of the CCW obtained varying CWR and pH values. Results obtained for EC and ES were comparable to those obtained for FC and FS, as lower pH and CWR values resulted in increased EC and ES (Fig. 1). Results also correlate well to those reported for other legume-derived proteins where both FC and EC were higher at lower pH values (Lafarga et al., 2018b). This is caused because both the EC of proteins and the ES depend on the hydrophilic-lipophilic balance, which is pH-dependant (Ragab et al., 2004). In the current study, a negative correlation was observed between CWR and protein concentration ($R^2 = 0.9866$; 0.05) indicating that the protein content of the *aquafaba* obtained boiling the chickpeas at a lower CWR had a higher protein concentration. The protein concentration of the aquafaba obtained at CWR of 1.50, 3.25, and 5.00 was measured as 0.48 ± 0.01, 0.23 ± 0.04, and 0.08 ± 0.00 mg/100 mL, respectively.

Proteins are surfactants which contain a hydrophilic “head” group that interacts with water and a lipophilic “tail” group which has affinity for oil and improve emulsion formation and stability (McClements, 2015). Gharsallaoui et al. (2009) reported higher EC and ES of pea proteins at pH 2.4, when compared to alkaline conditions, attributed to a lower adsorption of proteins combined with interfacial film reorganisation, which prevented film rupture increasing ES. Moreover, addition of sodium chloride, generally used at home level during the preparation of mayonnaise, could also affect the EC of the proteins as Khalid et al. (2003) observed that incorporation of salt increased EC by increasing the solubility of the proteins. Again, only a limited number of studies evaluated the technofunctional capacity of *aquafaba*. Damian et al. (2018) recently reported EC values for the cooking water of haricot beans, chickpeas, whole green lentils, and split yellow peas, cooked at a legume:water ratio of 1:1.75 (w/w), as 22.8, 38.6, 47.1, and 16% respectively. Although these values were within the range measured in the current study (3.9–72.3%), these cannot be compared as the pH in that study was not adjusted and ranged between 6.07 and 6.47 for haricot beans and lentils, respectively. In the current study, the EC could be fitted by a polynomial quadratic equation in terms of the studied conditions as described in Equation (4).

$$EC = 95.41264 + 20.09228 \cdot CWR - 15.38922 \cdot pH + 0.22857 \cdot CWR \cdot pH - 5.71823 \cdot CWR^2 + 1.01686 \cdot pH^2 \qquad (4)$$

The statistical analysis indicates that the proposed quadratic model for EC was adequate ($p < 0.0001$) with good determination coefficient ($R^2 = 0.9659$; Table 2). Both CWR ($p < 0.0001$) and pH ($p < 0.05$) had a significant effect on FC. Moreover, ES could be modelled by Equation (5).

$$ES = 196.10952 - 37.42857 \cdot CWR - 21.92857 \cdot pH + 4.15238 \cdot CWR \cdot pH \qquad (5)$$

The ANOVA also indicated that the proposed two-factor interaction model for ES was accurate and showed a good determination coefficient ($p < 0.0001$; $R^2 = 0.9234$). CWR and pH influenced FS ($p < 0.0001$ and $p < 0.005$, respectively). Again, as it happened with FC and FS, no significant lacks of fit were obtained, suggesting that both EC and ES fit properly for prediction within the ranges studied herein.

Optimisation and validation of the boiling conditions

The combined CWR and pH conditions that led to, on the one side, higher FC and FS, and on the other, higher EC and ES, were determined. Overall, higher foaming and emulsifying abilities were obtained when working at lower pH and CWR (higher protein content) values, which correlates well with the results shown in Fig. 1. To obtain the highest FS and FC, the optimum CWR and pH conditions were predicted to be 1.50 and 3.50, respectively (Desirability = 1.000). In addition, in order to obtain the optimum (highest) EC and ES values, the predicted CWR and pH values were 1.72 and 3.50, respectively (Desirability = 1.000) – The nearest the Desirability is to the unit, the more adequate the system is (Odriozola-Serrano et al., 2009). To complete the study, a set of experiments was conducted in order to validate the predictive models. The data, shown in Fig. 2, demonstrated that the predicted FC, FS, EC, and ES values were accurate enough to fit the experimental results, as correlation coefficients were 0.9261, 0.9609, 0.9706, and 0.9274 for FC, FS, EC, and ES, respectively. In addition, the optimum conditions were also validated against experimental results. The optimised FC, FS, EC, and ES values were predicted 332.4, 93.6, 73.0, and 80.0%. These values were validated in vitro and were calculated as 325.6 ± 8.3, 91.6 ± 5.5, 76.2 ± 4.9, and 80.9 ± 4.6%, respectively.

Comparison between vegan and egg-based meringues and mayonnaises

Colour attributes of the egg-based and vegan meringues and mayonnaises are listed in Table 4. Overall, no differences were observed between the colour parameters of meringues made using egg of aquafaba – except for a small difference in a* values ($p < 0.05$). The ΔE value between both samples was calculated as 1.7. Those samples with $\Delta E > 3$ display a colour deviation which is visible to the human eye (Wibowo et al., 2015), suggesting that no colour differences could be seen between both samples (Fig. 3B). Results contrast with those recently reported by Stantiall et al. (2018), who made meringues using either egg proteins or *aquafaba* obtained after boiling different legumes and observed that the visual appearance as well as the volume of the legume-derived meringues was lower than that of the control. In that study, the cooking conditions were not optimised to obtain higher FC and FS values. Moreover, the authors of that study used fresh egg white (in the current paper we utilised pasteurised eggs) and that can partially explain the observed differences. Mayonnaises made using egg or aquafaba did show colour differences ($p < 0.05$). Egg-based mayonnaises showed higher L* and lower *Ch* values ($p < 0.05$), which suggest a lighter appearance and a lower colour intensity when compared to mayonnaises made using aquafaba or CCW. In addition, ΔE was higher than three suggesting a visible colour difference between both mayonnaises.

No differences were detected between the sensorial (flavour, texture, and overall acceptance) scores of meringues made using pasteurised egg whites or *aquafaba* (Table 4). Results on sensorial analysis are preliminary and must be taken with caution, especially those on overall acceptance as the ideal would have been to assess acceptability using ~100 consumers. Similar results were observed for mayonnaises, although the overall acceptance score was lower for the mayonnaises formulated using CCW ($p < 0.05$). Results were comparable to those recently published by Damian et al. (2018), who observed no significant differences between the appearance of mousse made using chickpea proteins and the control made using egg-derived proteins.

Mustafa et al. (2018) also obtained good quality sponge cakes after substituting egg proteins by aquafaba, although in that case the *aquafaba* was obtained from commercial chickpea cans and not simulating domestic cooking conditions. Because of its high foaming ability, chickpea cooking water also showed potential for being used as texture improvers in gluten-free breads. Indeed, Bird et al. (2017) observed reduced crumb hardness after incorporating chickpea boiling water into a bread formulation and when compared to the breads obtained by using xanthan gum. Ma et al. (2016) also suggested that pulse-derived proteins were promising and valuable replacements of egg proteins in emulsion-type food products. The authors of that study optimised a salad dressing formulation using RSM and obtained products with textural properties that were comparable to those of the control. Although products made using egg whites obtained higher acceptability indexes, all of the samples had an acceptability of over 80%. For a product to be accepted in terms of sensorial characteristics, it is necessary to achieve an acceptability index greater than 70% (Lucas et al., 2018). Therefore, based on the sensorial analysis we can expect that the manufactured vegan meringues and mayonnaises would have a good acceptance. However, as highlighted previously, results on sensorial analysis are preliminary and further studies using a larger group of panellists are required. In addition, panellists were not told which sample was made using egg whites and which ones not.

Conclusions

When prepared at home level, both the boiling conditions and the adjustment of the pH resulted to be of key importance in order to maximise the foaming and emulsifying abilities of chickpea *aquafaba* as well as the stability of the developed foams and emulsions. Once the pH and the boiling conditions were optimised, the obtained aquafaba was used to develop meringues and mayonnaises that showed quality and sensorial properties which were comparable to those of the controls, made using egg proteins. Overall, chickpea cooking water or *aquafaba* may be potential replacers of egg whites in the manufacture of high quality meringues and mayonnaises. Future studies will evaluate the effect of processing (heat and sonication) as well as the effect of NaCl and other common food ingredients on the functional properties of *aquafaba*.

Conflicts of interest

None.

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Figure Captions

Fig. 1. Model graphs obtained for (A) foaming capacity, (B) foam stability, (C) emulsifying capacity, and (D) emulsion stability.

Fig. 2. Scatter plots of the predicted and experimental data of (A) foaming capacity, (B) foam stability, (C) emulsifying capacity, and (D) emulsion stability.

Fig. 3. Comparison between (A) foaming/emulsifying agents, (B) meringues and (C) mayonnaises made using: (*i*) egg whites or (*ii*) chickpea cooking water.

Table 1. Central composite response surface design and calculated responses under different conditions

Parameters			Variables [b]			
Run [a]	CWR (w/v)	pH	FC (%)	FS (%)	EC (%)	ES (%)
1	1:3.25	5.0	264 ± 14	59.2 ± 5.0	56.1 ± 1.0	43.6 ± 2.8
2	1:3.25	5.0	264 ± 6	56.8 ± 3.2	55.6 ± 0.6	40.2 ± 0.9
3	1:5.00	3.5	294 ± 1	78.3 ± 1.7	15.7 ± 0.0	0.0 ± 0.0
4	1:5.00	6.5	175 ± 7	3.4 ± 3.1	3.9 ± 0.0	0.0 ± 0.0
5	1:3.25	6.5	185 ± 1	31.8 ± 5.5	47.2 ± 2.0	12.5 ± 1.8
6	1:1.50	5.0	270 ± 3	70.2 ± 3.4	62.8 ± 1.5	66.0 ± 2.7
7	1:3.25	5.0	260 ± 9	64.8 ± 1.1	40.1 ± 1.4	25.6 ± 1.8
8	1:5.00	5.0	162 ± 1	36.9 ± 0.6	6.3 ± 1.2	0.0 ± 0.0
9	1:1.50	6.5	199 ± 3	44.8 ± 4.0	58.1 ± 0.5	32.7 ± 2.3
10	1:3.25	3.5	296 ± 7	81.5 ± 4.6	61.5 ± 0.9	44.8 ± 3.4
11	1:3.25	5.0	223 ± 2	47.4 ± 1.3	57.6 ± 0.4	40.6 ± 1.5
12	1:3.25	5.0	260 ± 3	60.0 ± 5.4	54.0 ± 0.9	37.6 ± 1.0
13	1:1.50	3.5	324 ± 15	93.4 ± 1.9	72.3 ± 1.0	76.3 ± 2.3

Abbreviations: CWR, chickpea:boiling water ratio; FC, foaming capacity; FS, foam stability; EC, emulsifying capacity; and ES, emulsion stability.

[a] Run number does not correspond to the order of processing.

[b] Data shown are the average of three independent experiments ± S.D.

Table 2. ANOVA calculated for the response surface quadratic models of foaming and emulsifying capacities at different conditions.

	FC (%)			EC (%)		
	Mean square	F value	Prob > F	Mean square	F value	Prob > F
Model	5580.83	16.64	0.0009 [a]	1169.22	39.63	< 0.0001 [a]
CWR	11344.80	33.82	0.0007 [a]	4664.88	158.11	< 0.0001 [a]
pH	10982.48	32.74	0.0007 [a]	270.68	9.17	0.0191 [a]
CWR × pH	2714.41	8.09	0.0249 [a]	1.44	0.049	0.8315
CWR^2	2197.18	6.55	0.0376 [a]	847.00	28.71	0.0011 [a]
pH^2	35.90	0.11	0.7531	14.46	0.49	0.5065
Lack of fit	1107.15	1.19	0.4195 [b]	0.70	0.014	0.9975 [b]
Pure error	369.05			51.11		
Corrected total	310.22			6052.60		
S.D.	18.31			5.43		
Mean	237.18			45.48		
C.V. (%)	7.72			11.94		
R^2	0.9224			0.9659		
Adjusted R^2	0.8669			0.9415		

Abbreviations: FC, foaming capacity; EC, emulsifying capacity; CWR, chickpea:boiling water ratio; S.D., standard deviation; and C.V., coefficient of variation.

[a] Model terms are significant ($p < 0.05$).

[b] Lack of fit is not significant relative to the pure error ($p < 0.05$).

Table 3. ANOVA calculated for the response surface 2FI models of foam and emulsion stability at different conditions.

	FS (%)			ES (%)		
	Mean square	F value	Prob > F	Mean square	F value	Prob > F
Model	2177.21	68.98	< 0.0001 [a]	2179.85	36.17	< 0.0001 [a]
CWR	1359.02	43.05	0.0001 [a]	5104.17	84.69	< 0.0001 [a]
pH	4999.71	158.39	< 0.0001 [a]	960.14	15.93	0.0032 [a]
CWR × pH	172.92	5.48	0.0440 [a]	475.24	7.89	0.0204 [a]
Lack of fit	20.54	0.45	0.7960 [b]	69.34	1.42	0.3787 [b]
Pure error	45.35			48.93		
Corrected total	6815.73			7081.98		
S.D.	5.62			7.76		
Mean	56.23			32.30		
C.V. (%)	9.99			24.04		
R^2	0.9583			0.9234		
Adjusted R^2	0.9444			0.8979		

Abbreviations: FC, foaming capacity; EC, emulsifying capacity; CWR, chickpea:boiling water ratio; S.D., standard deviation; C.V., coefficient of variation.

[a] Model terms are significant ($p < 0.05$).

[b] Lack of fit is not significant relative to the pure error ($p < 0.05$).

Table 4. Comparison between the colour and sensorial attributes of the formulated vegan and egg-based meringues.

	Meringue made using		Mayonnaise made using	
	chickpea cooking water	pasteurised egg whites	chickpea cooking water	pasteurised egg whites
$L*$	94.9 ± 0.4 ^{A}	95.0 ± 0.4 ^{A}	84.6 ± 0.2 ^{A}	88.5 ± 0.30^{B}
$A*$	−0.6 ± 0.0 ^{A}	−0.8 ± 0.0 ^{B}	−3.4 ± 0.1 ^{A}	−3.2 ± 0.0 ^{A}
$b*$	3.5 ± 0.3 ^{A}	3.5 ± 0.2 ^{A}	13.0 ± 0.1 ^{A}	9.9 ± 0.1 ^{B}
Ch	3.5 ± 0.3 ^{A}	3.6 ± 0.2 ^{A}	13.5 ± 0.1 ^{A}	10.5 ± 0.1 ^{B}
ΔE	1.69 ± 0.22		4.95 ± 0.46	
Visual acceptance score	7.7 ± 1.0	7.6 ± 0.9	7.8 ± 0.8	8.5 ± 0.5
Flavour score	7.2 ± 1.0	7.5 ± 1.2	7.3 ± 1.0	7.6 ± 1.2
Texture score	8.0 ± 0.9	7.5 ± 1.1	8.0 ± 0.7	8.4 ± 0.5
Overall acceptance score	7.3 ± 1.3	7.8 ± 1.1	7.2 ± 0.6	8.3 ± 0.7
Acceptability index (%)	81.7	86.1	80.0	92.2

Values represent the mean of three independent experiments ± S.D. Different letters indicate significant differences between meringues or emulsions made using aquafaba or egg whites. Sensorial scores were assessed using a 9-point hedonic scale – panellists were not told which sample was made using egg whites and which one was made using *AQUAFABA*. The criterion for statistical significance was $p < 0.05$.

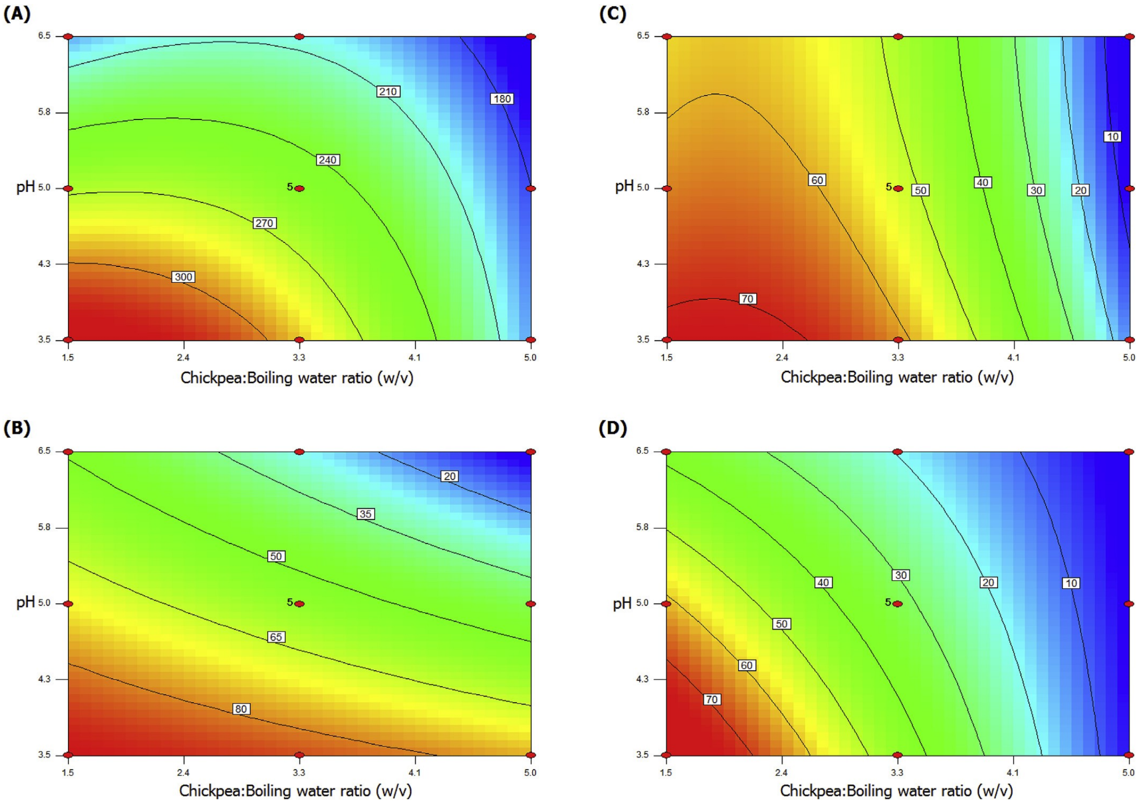

Figure 1.

(A)
pH
6.5
5.8
5.0
4.3
3.5
1.5
2.4
3.3
4.1
5.0
Chickpea:Boiling water ratio (w/v)
180
210
240
270
300
5
(C)
pH
6.5
5.8
5.0
4.3
3.5
1.5
2.4
3.3
4.1
5.0
Chickpea:Boiling water ratio (w/v)
10
20
30
40
50
60
70
5
(B)
pH
6.5
5.8
5.0
4.3
3.5
1.5
2.4
3.3
4.1
5.0
Chickpea:Boiling water ratio (w/v)
20
35
50
65
80
5
(D)
pH
6.5
5.8
5.0
4.3
3.5
1.5
2.4
3.3
4.1
5.0
Chickpea:Boiling water ratio (w/v)
10
20
30
40
50
60
70
5

Figure 2.

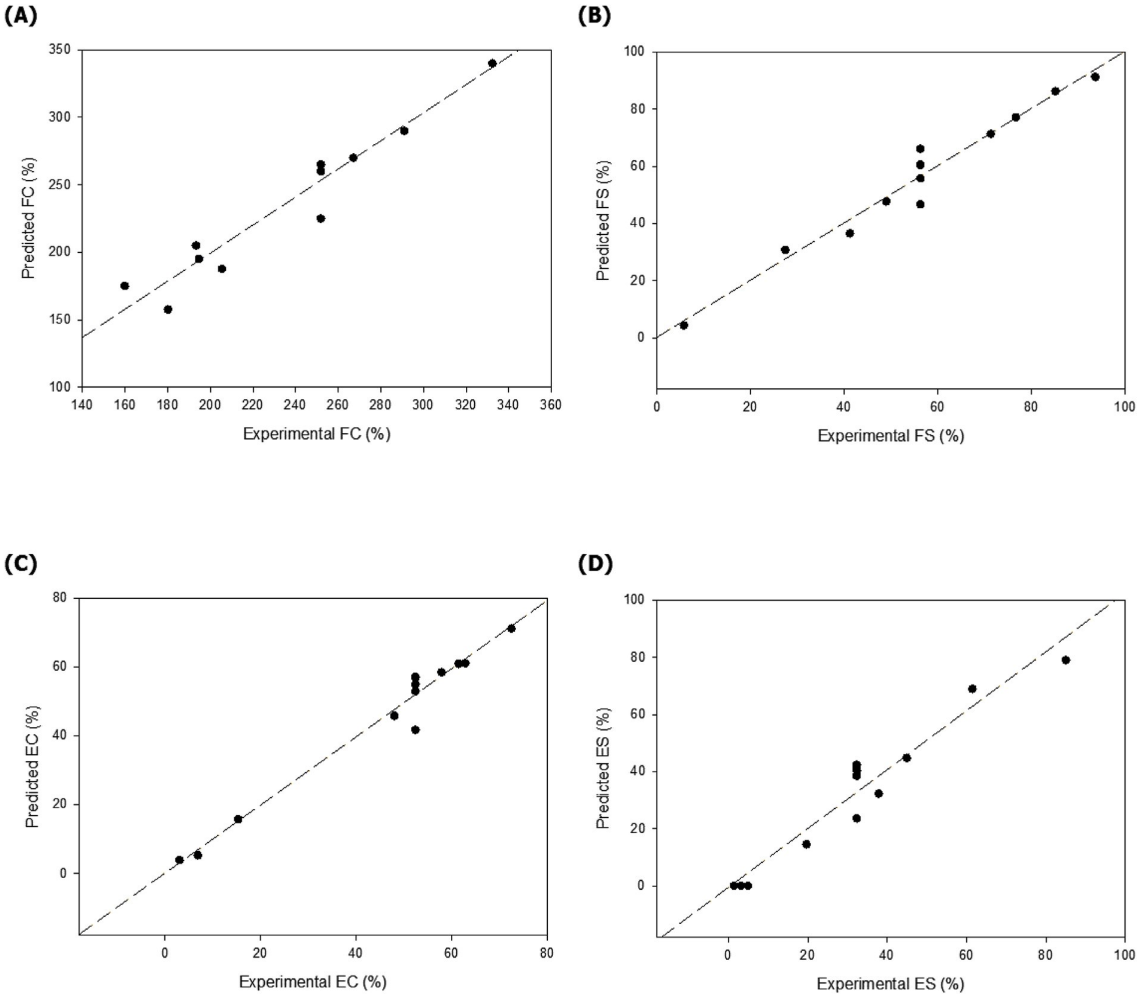
(A)
Predicted FC (%)
Experimental FC (%)
100
150
200
250
300
350
140
160
180
200
220
240
260
280
300
320
340
360
(B)
Predicted FS (%)
Experimental FS (%)
0
20
40
60
80
100
(C)
Predicted EC (%)
Experimental EC (%)
0
20
40
60
80
(D)
Predicted ES (%)
Experimental ES (%)
0
20
40
60
80
100

Figure 3.