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– Cookable with sustained rheological properties

Effekten av polysackarider och protein på reologiska egenskaper av en baobab baserad sås – kokbar med reologiska bevarade egenskaper

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Swedish University of Agricultural Sciences, SLU
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Abstract

Plant-based sustainable and nutritional food are a recent, growing trend setting out new opportunities for operators in the food value chain to ensure and reduce emissions from the global food production. Baobab (*Adansonia digitata L.*) fruit pulp, approved as a novel food, is rich in dietary fibres, antioxidants, and minerals. Towards the commercialization of baobab fruit pulp, to meet the demand for plant based sustainable food alternatives, Baofraiche, a cookable sauce, based on baobab fruit pulp has been developed and presented. This thesis reviews how the viscosity of the Baofraiche dispersion is affected during heating and by adding external ingredients. A Central Composite Design with three factors, pectin, Agar Agar and protein have been used. Thus, this experiment have been conducted to contribute to knowledge so that an optimal prototype for Baofraiche can be developed. Findings suggest that the viscosity of the Baofraiche decrease at higher temperatures. Pectin and Agar Agar, two different types of polysaccharides, affect the dispersion in different ways. Hence, the conclusion can be drawn that it is not possible to add any polysaccharide and predict that they will have the same effect on the dispersion during heating. Added protein, in combination with the other added factors, have a decreasing effect on the viscosity. Therefore, these factors should not be added together if a higher viscosity is required. Further studies should examine the optimal dispersion of Baofraiche based on the finding in this thesis. The overall conclusion is that Baofraiche is cookable and disperse well in a food system.

Keywords: Baobab, Baofraiche, Central Composite Design, Experimental Design, Rheology, Pectin, Viscosity, Cookable

Sammanfattning

Den växande trenden av växtbaserad miljövänlig och näringsrik mat har öppnat upp för nya möjligheter för livsmedelsoperatörer att bidra till att minska utsläpp från den globala livsmedelsproduktionen. Baobab (*Adansonia digitata L.*) trädets frukter, numera ett godkänt livsmedel innehåller kostfiber, antioxidanter och mineraler. För att möta efterfrågan av växtbaserad och miljövänligt livsmedel har en kokbar sås kallad Baofraiche, baserad på baobabfruktens innehåll tagits fram och presenterats. Denna uppsats undersöker hur viskositeten av såsen förändras under upphettning samt hur adderade ingredienser påverkar Baofraiche lösningen. En Central Composit Design har använts där de tre faktorerna pektin, Agar Agar och protein. Detta experiment har även genomförts för att bidra med kunskap till att en optimal Baofraiche ska kunna framställas. Resultatet av denna studie visar att viskositeten av Baofraiche minskar vid högre temperaturer. De två olika polysackariderna pektin och Agar Agar påverkar lösningen på olika sätt. Därför kan slutsatsen dras att det inte är möjligt att addera en slumpmässig polysackarid och motse ett likvärdigt resultat vid upphettning av lösningen. Det adderade proteinet har i kombination med de andra faktorerna en minskande effekt på viskositeten. Därför ska inte dessa faktorer adderas tillsammans om en hög viskositet efterfrågas. Framtida studier behöver undersöka den optimala blandningen för Baofraiche baserat på resultaten från experimenten i denna uppsats. Den slutliga slutsatsen är att Baofraiche är en kokbar sås som löser sig väl i ett livsmedelssystem.

Nyckelord: Baobab, Baofraiche, Central Composit Design, Experimentell Design, Reologi, Pektin, viskositet, kokbar.

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Abbreviations

BF	Baofraiche
LBB	Liquid Baobab Base
CCD	Central Composite Design
RDI	Recommended Daily Intake
AFT	Arwa Food Tech AB

1. Introduction

1.1. Background

The global food system of today, stands for nearly one third of the world greenhouse gas emission (Tubiello et al. 2021). Therefore, it seems essential to redesign these systems towards a sustainable and environmentally friendly production. The increasing demand and public awareness of sustainable food, in combination with new green technologies give opportunities for operators in the food value chain to ensure and reduce emissions from the global food production (European Commission 2021). Furthermore, the growing market trend on plant-based food, opens up for new business opportunities towards supplementary sustainable and innovative products that can face future adjustments towards a sustainable food system (Aschemann-Witzel et al. 2021). Moreover, it is of importance that the transition towards a sustainable food system aims to ensure nutritional and sufficient sustainable food (European Commission 2021).

In this study, in collaboration with Arwa Food Tech (AFT) AB, member of Aventure AB group, a cookable baobab fruit pulp-based sauce, named Baofraiche has been developed and presented. Baofraiche is aimed to be used as an ingredient in cold or hot food, as an alternative to commercial plant or milk-based crème fraiche. Baofraiche is suggested to be suitable in relation to the necessity of redesigning the global food systems towards being more sustainable, thus meeting the demand for environmentally friendly and nutritious food. Baobab fruit pulp is rich in dietary fibers, antioxidants, and minerals (Gebauer et al. 2002). Because of the high concentration of pectin in the baobab fruit pulp, no thickeners or emulsifiers are needed for the Baofraiche formulation, unlike commercial plant-based crème fraiche. Baobab fruit pulp was approved and authorized under decision 2008/575/EC by the European Parliament and the Council as a novel food under Regulation No 258/97. This has raised an international interest and intensified the acceptance and attentiveness in the baobab leading to an increased demand worldwide (Gebauer et al. 2016). Since then, several studies have been performed to identify opportunities, threats, strengths, or weaknesses with commercialization of baobab (Jäckering et al. 2019). It has been found that it may enhance the income

and give a better integration for farmers into the value chains, especially for women. Likewise, it is shown that commercialization of baobab fruit helps rural people to alleviate poverty because of an increasing cash economy (Venter & Witkowski 2013).
The present study is performed with the intention to develop and present a cookable, baobab fruit pulp-based product with the aim to understand how a combination of added ingredients will effect the dispersion during heating. By standardize processing parameters for Baofraiche and gain knowledge on how the dispersion is effected by adding different ingredients, this study is of importance in the commercialization of baobab fruit pulp.

1.2. Baobab (*Adansonia digitata L.*)

The African baobab tree (*Adansonia digitata L.*), family of Malvaceae, and its related species is a wild, fruit bearing tree that is mainly found in Sub Saharan African and Western Madagascar (Ismail et al. 2019; Sidibe 2002; Fischer et al. 2020). The baobab tree has not been domesticated and is therefore naturally grown in arid and semi-arid lands, particularly in Sub Saharan Africa (Sidibe 2002). The baobab tree is easily recognized due to its bulky appearance, massive height (reaching up to 25 meters) and grey to reddish brown bark (Gebauer et al. 2016). Further, because of the irregular and gnarled crown, which for many months are leafless, the baobab tree is referred to as Africa´s “up-side down tree” because one can argue that it looks like the root system is facing upwards. The baobab fruits are most often globose or oval shaped with a woody pericarp enclosing the dry white mealy pulp that surrounds and protects the seeds. The earliest known mention of the African baobab tree was in the “Book of Roads and Kingdoms”, in the eleventh century (Wickens 2008). Scientists have searched for the oldest baobab tree which has been estimated to have an age close to 1600 years. (Patrut et al. 2015). Moreover, it is known that in the sixteenth century, baobab products were traded in Cairo markets (Sidibe 2002).

Figure 1. Baobab tree. Copyright: Pixabay 2021

As the baobab is a multipurpose tree, almost all parts of the tree are used (Sidibe 2002). The tree itself, consisting of small holes in the stem, can be used by humans or small animals for shelter and storage. From the bark, fibre can be extracted and used to produce rope or for weaving (Gebauer et al. 2016). The baobab leaves are known to contain many amino acids where the highest reported values were found for aspartic acid, leucine, glutamic acid, and arginine (Chadare et al. 2008). Moreover, the leaves are also known to be a significant source of minerals (Sidibe 2002). The leaves are therefore used in cooking, both fresh and dried. Moreover, the fruits pericarp (the shell) is used for handcraft products or for building music instruments whereas the seed kernels are eaten raw or dried for further use in cooking or medicine. Further has baobab seed oil been extracted from the kernels and been consumed for many years due to its nutrient value (Msalilwa et al. 2020).

Figure 2. To the left: Baobab fruit pericarp hanging from a baobab tree. To the right: Dried baobab fruit pericarp with the arrangement of white fruit pulp and fibers shown inside. Copyright: Pixabay 2021

1.2.1. Baobab fruit pulp – Nutritional composition

Baobab fruit pulp has been used for centuries for both hot and cold drinks, sauces and as a fermenting agent (Aluko et al. 2016). Not only is it essential as a food ingredient but also for the centenary use in traditional African medicine (Sidibe 2002). Several studies have been composed to study the nutritional value of baobab fruit pulp. According to Gebauer et al (2002) the baobab fruit pulp contains protein (2.6%), polysaccharides (2.6% pectin where 56.2% of this is galacturonic acid), calcium (655mg/100g dw), phosphorus (50.8mg/100g dw) and sugars (23.2% where 19.9% are reducing sugars). The baobab fruit pulp is also rich in dietary fibers (varying from 17.2–27.9 g/100g dw)(Cissé et al. 2013). Moreover are the levels of fat (0.2%) and starch (0%) low in the baobab fruit pulp (Gebauer et al. 2002). The daily recommended intake (RDI) of ascorbic acid (vitamin C) is 75 mg whereas 100 g dry weight of baobab fruit pulp contains 300 mg, therefore it has been suggested as a good antioxidant and as a good source of ascorbic acid (Aluko et al. 2016; Gebauer et al. 2002; Livsmedelsverket 2021).

1.2.2. Pectin

As described in 1.2.1, baobab fruit pulp contains polysaccharides, no starch, but 2.6% of pectin. Pectin is one of the most structurally complex family of polysaccharides found in nature (Mohnen 200; Wang et al. 2021). Pectin is a polygalacturonan, found in the cell walls of plants. Pectin has an essential role in plant development and defense, cell wall structure, signaling or for the morphology of the plant (Mohnen 2008). The structural appearance of pectin is commonly divided into high- or low-methoxyl pectin depending on the extent of esterification on the carboxyl groups of the galacturonic acid residues (Wang et al. 2021; Mohnen 2008). According to a study made by Dimopoulou et al (2021), baobab fruit pulp is derived to have a low degree of methyl esterification implying that it is in the group of low-methoxyl pectin. One major role for usage of the plant polysaccharide pectin in food industry is for its thickening and gelling properties (Gawkowska et al. 2018). Moreover, general studies made on pectin also suggest that pectin can have an anti-cancer role and lower the cholesterol level in blood (Glinsky & Raz 2009). This makes the pectin attractive because of its prevention and reduction of cardiovascular disease (Soliman 2019).

1.3. Rheology Basics – Concept and Applications

Texture plays an essential role in the influence on consumers perception to food quality (Fellows 2000). Therefore, to meet consumers sensory perceptions, rheology, defined as the study of deformations and flow of matter (Malkin & Isayev 2017) is used for optimizing product development in the food sector (Fellows

2000). Rheological properties are characterized when materials deform during applied stress which contributes to changes and deformation of the properties in the material (Fellows 2000). Rheology, *rheo* in Greek, means “to flow” and it is therefore known that rheology applies to flowing media (Malkin & Isayev 2017). However, the classification between solids and liquid materials is hard to grade, the term rheology is therefore used for any material.

Several terms are applied while discussing rheology. When layers of fluid is sliding over one another with each layer moving faster than the one beneath are described as *shear flow* (Malvern 2016). For shear flow to take place, a shear force, defined as *shear stress* (σ) must act on the fluid. The force (F) acting over a unit area (A) defines shear stress. While applying shear stress to a fluid the bottom layer will remain stationary while the upper layer move a set distance (x) in response to the given force. Hence, *shear strain* (γ) will be defined because there will be a displacement gradient across the sample. For a fluid, the shear strain will continue to increase for the period of applied stress while the shear strain for a solid material will be restricted for an applied stress where no flow will be possible. The rate of change of shear strain with time is termed the *shear rate*.

Viscosity is the measure of the internal friction of a fluid (Fellows 2000). When a layer of fluid is made to move in relation to another layer, this friction will become apparent. The greater amount of force needed to cause movement, the greater the friction will be between the layer of fluid. Fluids that require more force to move are therefore highly viscous whereas less viscous materials will not require as much force to move. In the food processing industry the viscosity of liquid foods is an important characteristic. It is therefore essential to understand the characteristics of the food. When applying external force, for example during heating or cooling the liquid, the viscosity of the liquid will change. As described earlier the forces known to move the liquid are called shear force and shear stress. When the relationship between shear stress and shear rate are linear, liquids are referred to as Newtonian fluids, since that the viscosity will be constant. Fluids that are non-Newtonian will on the other hand have a non-linear relationship between shear stress and shear rate, meaning that they have a variable viscosity. Non-Newtonian liquid food are classified into different types; *Pseudoplastic fluids* (decrease in viscosity with increasing shear rate and recover along the same path when the force is removed), *Dilatant fluids* (viscosity increases as the shear rate increases) and *Thixotropic fluid* (viscosity decreases with continued shear stress).

1.4. Experimental design - Central Composite Design

Experimental design refers to the objectives of the experiment where the chosen design depends on the number of factors to be investigated (NIST/SEMATECH 2012). It is an advanced designed plan for the experiments where input variables are systematically varied within predetermined ranges so that their effects on the responses (variables) can be estimated and checked for significance (Esbensen 2002). Depending on the objective, the way they are built, and the operational constraints of the experiment, the number of experiments will vary. One of the reasons for using experimental designs is to separate the signal from the noise.

Depending on number of factors chosen and on the experiment objectives, different kinds of factorial designs can be selected (Figure 3). A factorial design is used when the effect of multiple independent variables is studied. For example, when studying how a certain parameter or ingredient change the outcome for a developed product. If a 2-level full factorial design is used the experiment factors will only have two levels. This design provides useful information with relatively fewer run per factor, however it is not possible to fully explore a wide region in the factor space. Such experiment are for example useful when exploring where optimal settings may exist within a range of different settings. Further, a full factorial design estimates the main effects of all design variables and all interaction effects. On the other hand, when only half or fractions of the total number of experiments (as in full factorial design) is performed, a fractional factorial design is used. This design is better from the economic/time perspective since less experiments need to be performed. Further, the fractional design can be used while performing a screening design. Screening is performed when several factors need to be explored. The fractional design is therefore used to exclude the least important factors before performing a full factorial design. To find the main, most important effects, it is not necessary to perform all combinations in the experiment at all times, the fractional design can therefore be an option. (MiniTab 2021; Esbensen 2002)

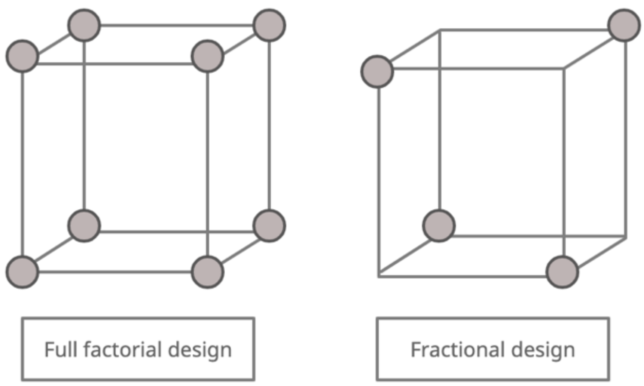


Figure 3. Each point in the cube represents an experiment. Full factorial design with 3 variables at a 2-level gives 2^3 equals 8 experiments (Esbensen 2002).

One example of a design that consists of a full factorial design is the Central Composite Design (CCD) with a quadratic model (Bhattacharya 2021). The CCD is built out of two sets of experiments (Esbensen 2002). First, a cube and centre samples from the full factorial design. Secondly, to add additional levels required to compute a quadratic model, star samples are included in the design, as seen in Figure 4. CCD could be used when it is desired to find the best possible product from several performed experiments (Bhattacharya 2021). Moreover, the CCD is used when optimizing one or several responses in relation to design variables ranging between 2 and 6 (Esbensen 2002). The number of design variables will decide the number of experiments the CCD will consist of. Further, the number of samples for the central samples range from 3—15, also depending on the design variables.

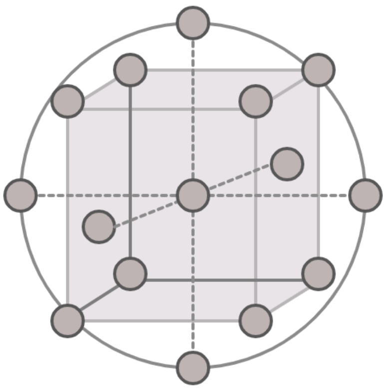


Figure 4. A geometrical representation of a Central Composite Design with 3 variables (Esbensen 2002). Two sets of experiments are represented in the figure: the full factorial design with star samples included.

1.5. Objective

The main aim of this thesis was to develop a cookable sauce based on a baobab fruit pulp-base and to understand the behaviour of the dispersion during heating. The following tasks were included to achieve the aim of this study:

- Develop a baobab fruit pulp-based product, suitable for cooking, inspired by the commercial plant-based crème fraiche.
- Standardise dry matter and oil percentage for the process parameter of the prototype.
- Determine the emulsion stability of the standardized recipe for the prototype of Baofraiche produced.
- Study the rheological parameters of the Baofriache prototype.

- Conduct a Central Composite Design to investigate the effect of adding three factors: pectin, protein, and Agar Agar to the Baofraiche dispersion in terms of rheological behaviours.
- Investigate how rheology properties of the samples in the CCD will behave at different temperatures, ranging from 0 —100 °C.

Because of high pectin levels in baobab fruit pulp, it was of interest to understand the pectin´s role further prior to the development for the prototype for Baofraiche. Pectin, from another source of origin, was therefore included as one of the added ingredients in the CCD. Moreover, it was also interesting to find out how another polysaccharide, besides pectin, would behave and affect the rheological properties of the dispersion. Therefore, based on studies made on gelling properties of polysaccharides in the food industry, Agar Agar was included as the second ingredient in the CCD (Jayarathna et al. 2021; Ashila & Rahmatunnisa 2021). By including plant-based protein as the third factor, there was an opportunity to examine if the rheological properties of the dispersion were affected. It is also known from previous studies that various aspects of protein-polysaccharide interaction can affect the food system, for example the viscosity, which made it even more important to add protein (Sánchez et al. 1995; Schmitt & Turgeon 2010; Benichou et al. 2002). Moreover, it was of interest to understand the effect that protein, pectin and Agar Agar had on each other in order to create the best possible plant-based, clean label and non-starch based dispersion for the Baofraiche. By examine and develop the optimal dispersion with the best possible combination and amount of the three chosen factors, this study could be helpful for future industrial application of Baofraiche.

2. Materials and methods

2.1. Materials

Unprocessed baobab fruit pulp was purchased from Sudan (2021) and used as starting materials for all performed experiments. Rapeseed oil (Zeeta, Sweden 2021) was used for all emulsion systems. As reference products, R1: iMAT FRAICHE (Oatly) and R2: Planti cooking fraiche (Planti) was used. All experiments were performed in lab scale and detailed conditions are confidential.

2.2. Sample preparation

2.2.1. Preparation of Liquid Baobab Base (LBB)

The standard operating procedure revised from the internal document was used in this study when preparing Liquid Baobab Base (LBB) (AFT AB 2021). Briefly, baobab fruit pulp was hydrated with boiling water and stirred using a turbine vortex blade mounted on CAT agitator rotor R18 (LabTeam Scandinavia AB, Sweden) at constant speed for approximately 40—50 minutes in a water bath. After, baobab seeds and fibers were filtered out using a sieve. The filtered dispersion, named LBB was stored at 4 °C—8 °C until further use (Figure 5).

2.2.2. Preparation of Baofraiche (BF)

For each trial, a standardized amount of LBB was heated with constant stirring by hand using a metal whip. The cooked LBB was left to cool down to room temperature before establishing a dry matter using a moisture analyser MJ33 (Mettler Toledo, Switzerland). Rapeseed oil was measured and added to a separation funnel. The cooked LBB was emulsified with Bamix Gastro 350 Pro (Bamix, Switzerland), at a speed of 17000 and 22000 rpm with an oil flow of approximately 15—18g/min from the separation funnel. The BF was stored at 4 °C— 8 °C (Figure 5).

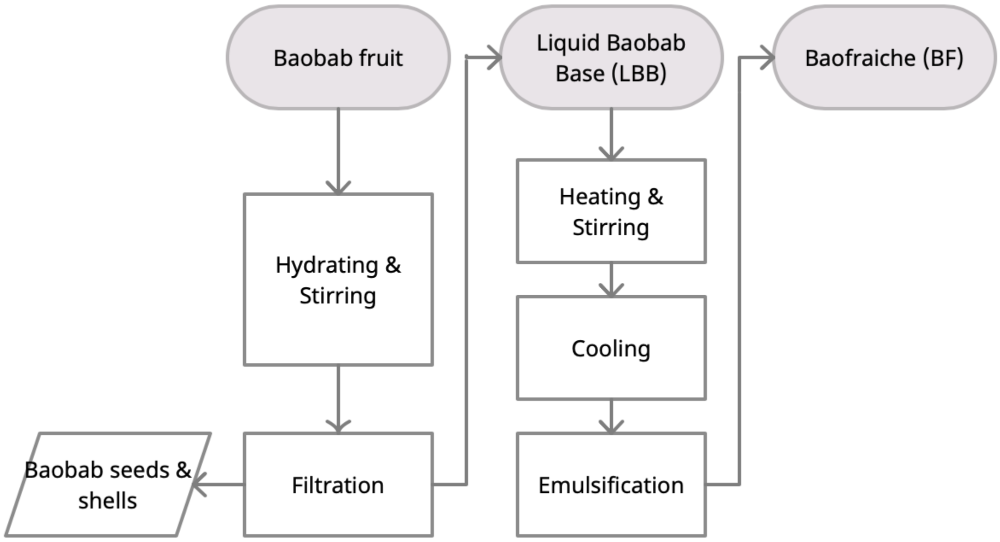


Figure 5. Illustration of the general processing steps of Baofraiche (BF).

2.3. Formulation and processing optimization of Baofraiche

2.3.1. Dry matter

Different dry matters for the BF were evaluated to set a fixed value and thus reduce complexity of the experiment. All steps in these experiments was prepared according to Figure 5 but with alteration in dry matters of the LBB. Furthermore, five different concentrations of dry matter was obtained by adding water to the LBB, after the cooling process. The dry matter of all samples was determined by the moisture analyser MJ33 (Mettler Toledo, Switzerland). Important criteria for an acceptable dry matter for the BF was based on subjective decisions such as mouthfeel, taste, appearance, and thickness. It was also of importance that the Baofraiche solution, after the three added ingredients of pectin, protein and Agar Agar had been added, would be able to be implemented and used for all sets of experiments in the Central Composite Design. Two commercial plant-based crème fraiche, already implemented on the market, R1 and R2 were used as reference products. The oil content was kept constant for all performed experiments. All experiments were performed in triplicates.

2.3.2. Oil percentage

Briefly, to obtain the optimal oil concentration for BF, different concentrations was evaluated after the dry matter of the BF was established. This was implemented by using different oil percentage in the emulsification step (Figure 5). Important criteria for an acceptable oil content for the BF was based on subjective decision such as mouthfeel, taste, appearance, colour and thickness. It was also of importance that the Baofraiche solution, after the three added ingredients of pectin, protein and Agar Agar were added, it would be possible to be implemented and used for all sets of experiments in the Central Composite Design.

2.3.3. Emulsion stability percentage

To investigate the emulsion stability, 10 g of samples were transferred into a 15 ml centrifuge tube and placed in Centrifuge 5804 (Eppendorf, Germany). The samples were centrifuged for 30 minutes, at 5000 rpm at room temperature to accelerate phase separation. The emulsion stability ratio, presented in percentage was the ratio of the height of the stable layer to the height of the total layer. The method was adapted from previous study made on emulsion stability (Zhu & Lam 2021). The same study, made on the emulsion stability showed that added mustard powder has stabilizing effect on food emulsions. Therefore, to investigate the need of mustard for the BF emulsion, one sample, including 1.5% mustard powder was included.

2.3.4. Dispersion of Baofraiche

To study how well the Baofraiche with the set dry matter and oil content would behave and disperse was commercial plain tomato sauce heated to 100 °C under constant stirring. Baofraiche was added to the tomato sauce and left to boil. Observations regarding how well the Baofraiche dispersed in the tomato sauce, how the colour and appearance was affected was obtained. These observations were made from subjective decisions. The experiment was repeated for R1 (Oatly, iMAT FRAICHE) and R2 (Planti, Planti Cooking Fraiche) for the sake of comparison. The experiment was made three times to avoid bias.

2.4. Experimental design

2.4.1. Design ranges – extreme values

Pre-experiments on the BF was performed in order to establish adequate intervals for three chosen factors: Pectin, Cesapectin LM-40 (Tate & Lyle Food systems), Agar Agar, *Rhodophyceae* (Raw food shop) and a food grade protein. Moreover, it was it of importance to establish acceptable working range, so the condition within the design are realistic of a probable prototype. In this study *Rhodophyceae* is referred to as Agar Agar and Cesapectin LM-40 as Pectin. The minimum value was set to 0 (no addition) for all factors. The criteria for establishing the maximum value for each factor was that the dispersion had a viscosity and texture that was possible to work with while using a rheometer. It was also important that the dispersion was somewhat similar to the commercial chosen reference. Thus, the dispersion needed to be homogenous and not too viscous dispersion. This was done by adding each of the factors separately to heated LBB before the emulsification (Figure 5). The extreme values, meaning the minimum and maximum values for each factor, were used for the design of choice.

2.4.2. Central Composite Design

The experiments were design based on a Central Composite Design with three independent factors: Agar Agar, pectin and protein. The design was used because it is an effective tool that helps in the understanding or mapping of a certain region of a response surface. Intervals used for each factor were established through investigating and deciding extreme values. In order to conduct all experiments in the CCD was all combined factors carefully mixed and measured following a given recipe list from the design and added to the heated LBB before the emulsification (Figure 5). A total number of 20 samples with different concentrations of the three factors included were produced (Table 5).

2.4.3. Rheological characterization

All rheological measurements were performed using a dynamic rotational rheometer Kinexus Pro+ with the associated software rSpace. The geometry used was a 21 mm vane type of bob and a 27 mm cup, 4V21:CUP25. Before measuring each sample, a zero gap procedure was performed on the rheometer. The running order for measuring the rheological properties of each sample can be seen in Table 5 as determined from the Central Composite Design. Approximately 30ml of sample was loaded before the vane tool was lowered into the cylinder.

First, an Amplitude Sweep Stress Controlled program was performed in an oscillating measurement, see detailed settings in Table 1. The Amplitude Sweep Stress Controlled program provide information about the linear viscoelastic region (LVER) of the material. This region is important to know because it provides information about the maximum stress or strain that can be applied to the material without destroying its internal network or structure. Normally a stress value from the centre of the LVER is chosen for further investigations.

Table 1. Detailed settings for Amplitude Sweep Stress Controlled program

Amplitude details	**Settings**
Start shear stress	0.1 Pa
End shear stress	300 Pa
Frequency	1.0 Hz
Sample per decade	5

To study the effect of temperature a Single Frequency Stress Controlled Temperature Ramp was applied, see settings in Table 2. For the temperature ramp a shear stress value from the centre of the LVER was applied. The temperature ramp was applied in an oscillating measurement at 1Hz.

Table 2. Detailed settings for the Single Frequency Stress Controlled Temperature Ramp

Ramp details	**Settings**
Start temperature	10°C
End temperature	100°C
Ramp rate	5 °C/minute
Frequency	1.0 Hz
Shear stress	0.1 or 1.0 Pa *
Sampling interval	2 sec

*Each sample had its own shear stress value. Sample 1-5 had 1.0 Pa and sample 6-20 had 0.1 Pa.

2.5. Software

The statistical software MiniTab, version: 19.2020.2.0 was used to create the central composite design, calculate the response surface regression and to create contour plots. The significance level, for all test performed, was set to 0.05 (5%). Microsoft Excel, version 16.54 was used to convert raw data, creating scatterplots and graphs.

3. Results and discussion

3.1. Formulation and processing optimization of Baofraiche

The results after optimizing the formulation for BF was decided to be 20 % for the dry matter and 25 % for the oil content. This was based on subjective decisions. The dispersion with this ratio of ingredients had a good mouthfeel, viscosity and texture. Moreover, the taste of the product was pleasant, with a creamy mouthfeel and uniform consistency and a just about right tart aftertaste. This formulation was therefore used for all experiments performed for the experimental design. The dry matter and oil contents of the emulsion was excluded as factors in this project since the aim was to study the effect of major ingredients.

3.2. Emulsion stability percentage of Baofraiche

The emulsion stability for the BF was shown to be 100% after centrifugation (Table 3). Mustard powder was added to see what stabilizing effect it had on the dispersion. It showed that the emulsion stability was 100 % with added mustard. Due to this finding, it was decided to exclude the mustard powder for the BF because the dispersion without any mustard powder added had as good emulsion stability percentage as the sample with added mustard. Moreover, the mustard powder gave pungent flavour to the product that wasn’t desired. Besides, reference sample 2 (R2) only had an emulsion stability percentage of 55.7% which in comparison to the other samples was inferior (Figure 6). This indicated that the emulsion stability of BF was much better than R2. One could therefore argue that the BF, with the oil content of 25% and a dry matter of 20 are stable enough, without any added mustard powder. Further, commercial plant based crème fraiche usually consist of added starch to increase the stability and to make the dispersion stable (Oatly 2021; Planti 2021). For the BF is this not required since it is stable without additives which provide the product with a desired clean label.

Table 3. Emulsion stability percentage in % for Baofraiche (BF), Baofraiche with mustard added (BF + mustard), reference product 1 and 2 (R1 and R2)

Sample	**Emulsion stability (%)**
BF	100
BF + mustard	100
R1	100
R2	55.7

Figure 6. From left to right, R1, R2, BF, BF + Mustard. Illustrates the results after centrifugation of all samples included in the experiment. The circle illustrates where separation has occurred for R2.

3.2.1. Baofraiche during cooking

The aim for the BF was that it should be able to be used as an ingredient while cooked in a food system. Moreover, it was important to study if the dry matter and oil content set for the prototype for the Baofraiche was convenient to work with. Meaning that it disperses well, without affecting the food system in a negative way such as leaving clumps or give an unpleasant oily surface. Therefore, the BF was dispersed in a plain tomato sauce while cooking to see how it would react and disperse. As illustrated in Figure 7, the BF dispersed very well in comparison to reference sample 1, both during and after cooking. The tomato sauce was homogenous, with no clumps from the BF left in the dispersion. The oil in the Baofraiche dispersed well with only a small, acceptable amount, on the surface of the tomato sauce. This gave the indication that the prototype developed for Baofraiche would work as a food ingredient and that it dispersed well in the food system while, and after being cooked. Further, this concluded that the set dry matter and oil content for the Baofraiche was acceptable to work with while moving forward to the experimental design. Unfortunately, this dispersion has not been tested and analysed before and it is therefore not possible to compare against the literature.

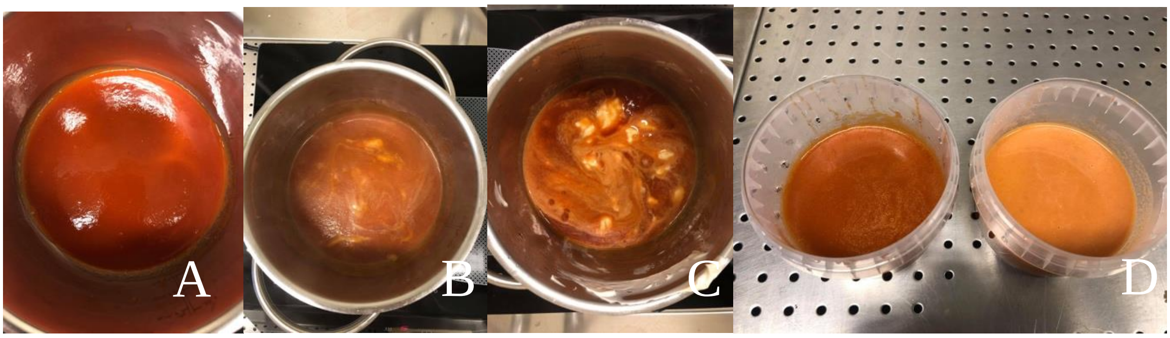


Figure 7. A. Regular tomato soup with no added ingredients. B. Baofraiche added to the tomato sauce during cooking. C. Reference sample 1 added to the tomato sauce during cooking. D. Baofraiche (to the left) and reference sample 1 (to the right) dispersed in tomato sauce after cooking.

3.3. Design ranges - Extreme values

The extreme values for pectin were set from 0 % to 5 %, the Agar Agar from 0 % to 4 % and protein from 0 % to 10 % (Table 4). The texture and viscosity when the different ingredients were added was still good enough to be able to work with. These values were used when creating the central composite design.

Table 4. Extreme values used for the central composite design

Factor	Extreme values: interval (%)
Pectin	0—5
Agar Agar	0—4
Protein	0—10

3.4. Central Composite Design

The central composite design was conducted based on previous findings (section 3.3). The design (Table 5) turned out to have 20 samples. The standard order (StdOrder) indicated that sample 1—8 was cube corners, sample 9—14 was the extreme values and sample 15—20 central points in the design. The amount of added pectin, protein and Agar Agar, are presented in %, are shown in Table 4. Moreover, the Run Order (RunOrder) indicate the production and running order of all samples used in the design.

Table 5. Given amount of the three independent variables added to samples according to the Central Composite Design

StdOrder	RunOrder	Pectin (%)	Protein X (%)	Agar Agar (%)
1	15	1.01	2.03	0.81
2	11	3.99	2.03	0.81
3	8	1.01	7.97	0.81
4	4	3.99	7.97	0.81
5	14	1.01	2.03	3.19
6	13	3.99	2.03	3.19
7	3	1.01	7.97	3.19
8	5	3.99	7.97	3.19
9	16	0.00	5.00	2.00
10	20	5.00	5.00	2.00
11	7	2.50	0.00	2.00
12	9	2.50	10.00	2.00
13	6	2.50	5.00	0.00
14	12	2.50	5.00	4.00
15	18	2.50	5.00	2.00
16	2	2.50	5.00	2.00
17	17	2.50	5.00	2.00
18	10	2.50	5.00	2.00
19	1	2.50	5.00	2.00
20	19	2.50	5.00	2.00

3.5. Amplitude Sweep Stress Controlled

The applied shear stress for the Single Frequency Stress Controlled Ramp differed between two values, 0.1 Pa and 1 Pa. This was not systematically varied in the design, and it can therefore be seen as an error in this experiment. Simply, for the five first samples that were run, a different assessment compared to the rest of the samples was made. However, before analysing the raw data perceived from the Single Frequency Stress Controlled Ramp, it was concluded that this didn’t affect the result in any way that was clearly stated. Because of this judgement, and due to lack of time, the samples were not run again. The applied shear stress for sample 1—5, according to Run Order, was 1 Pa whereas the rest of the samples had a shear stress of 0.1 Pa. According to the standard order these are sample 4, 7, 8, 16 and 19, representing 2 centrum points, 1 cube corner and 2 extreme values in the design. However, because sample 4 and 7 are centrum points it is possible to compare these to the rest of the centrum points in the design. Unfortunately, is it not the same for sample 8, 16 and 19 and this can therefore be considered as a source of error.

3.6. Single Frequency Stress Controlled Temperature Ramp

The process of understanding the results received from the Single Frequency Controlled Temperature Ramp was divided into different phases. To make the data readable and facilitate the process towards analysing all data was the temperature ramp, ranging from 0—100 °C divided into four intervals 15—25, 35—45, 55—65 and 85—95 °C. The reason behind the decision to choose these specific intervals was that they represented the whole curve. Mean values for the raw data for all samples and the intervals chosen are presented in Table 6.

Table 6. Showing the mean value for the raw data of viscosity (Pa) perceived from the Single Frequency Stress Controlled Temperature Ramp with the intervals chosen to represent the whole curve presented in °C.

	Temperature interval °C			
StdOrder	**15-25**	**35-45**	**55-65**	**85-95**
1	181.4	82.9	44.8	74.6
2	500.3	243.5	121.8	97.8
3	228.1	121.1	73.1	93.8
4	322.4	153.6	72.6	55.0
5	289.8	135.6	65.9	89.2
6	410.4	194.9	95.3	87.9
7	284.2	151.2	92.3	119.7
8	331.3	167.5	85.6	65.7
9	188.9	89.4	52.6	91.4
10	467.3	245.6	126.7	78.9
11	361.8	152.9	79.7	95.9
12	259.6	142.4	86.9	98.4
13	266.9	129.3	65.5	58.8
14	394.1	204.1	111.5	113.3
15	333.3	182.3	105.7	104.6
16	378.3	196.8	111.2	93.7
17	321.8	156.9	77.4	82.6
18	259.6	142.4	86.9	98.4
19	370.7	203.3	125.5	123.2
20	369.8	206.6	114.1	101.5
BF	175.8	89.9	47.9	62.9
R1	388.1	148.2	14.3	0.2

To get a better understanding of the raw data were 4 samples chosen to represent all experiments with the aim to present the outcome in a simplified way (Figure 8). The samples selected was 1 cube corner, 2 extreme values and 1 central point.

Moreover, reference product 1 (R1) and Baofraiche (BF) was included in the figure to be able to compare the dispersions at different temperatures. Sample R2 was not run with the rheometer, due to lack of time. Therefore, this sample will not be included in the presented result and discussion. As shown in Figure 8 is it clear that the viscosity of the tested sample is decreasing for all samples with increasing temperatures. Moreover, it is clear that the viscosity of all samples from the experimental design, meaning sample 1—20 (Table 6) and the BF are much higher than the reference product R1 at higher temperatures. R1 has an extreme low viscosity at high temperatures which is not the case for the rest of the samples. Moreover, the samples presented in Figure 8 (sample 1, 10, 14 and 20) have a higher viscosity from the start in comparison to Baofraiche. This agrees with the literature where it is known that polysaccharides, with their water binding capacity, increase viscosity of food matrix and that they are often used as in food dispersions to increase the viscosity (Wang & Steve 2005). The higher viscosity for the design-samples in comparison to the BF could therefore be explained by the added polysaccharides, which is not the case for BF or R1. Yet, as other studies suggest, could the high viscosity for the design samples be explained by the interaction between the polysaccharide and protein (Sánchez et al. 1995; Schmitt & Turgeon 2010; Benichou et al. 2002). However, at higher temperatures the difference between the viscosity of the samples and Baofraiche are not as distinguished. This could be because the added pectin, protein and Agar Agar have larger effects on the dispersion at lower temperature compared to the effects at higher temperatures Also, the tested samples differ in order when looking at low vs high temperatures. For example, sample 10 has the highest viscosity at the temperature interval of 15—25 °C in comparison to the temperature interval of 85—95 °C where it has the second to lowest viscosity out of the design samples. Further, sample 14 had the second to highest viscosity at the temperature interval of 15—25 °C whereas at temperature interval 85—95 °C it was shown to have the highest viscosity. One could therefore argue that the added ingredients will have an effect on the samples in relation to different temperatures because of the change in order of the samples as seen in Figure 8.

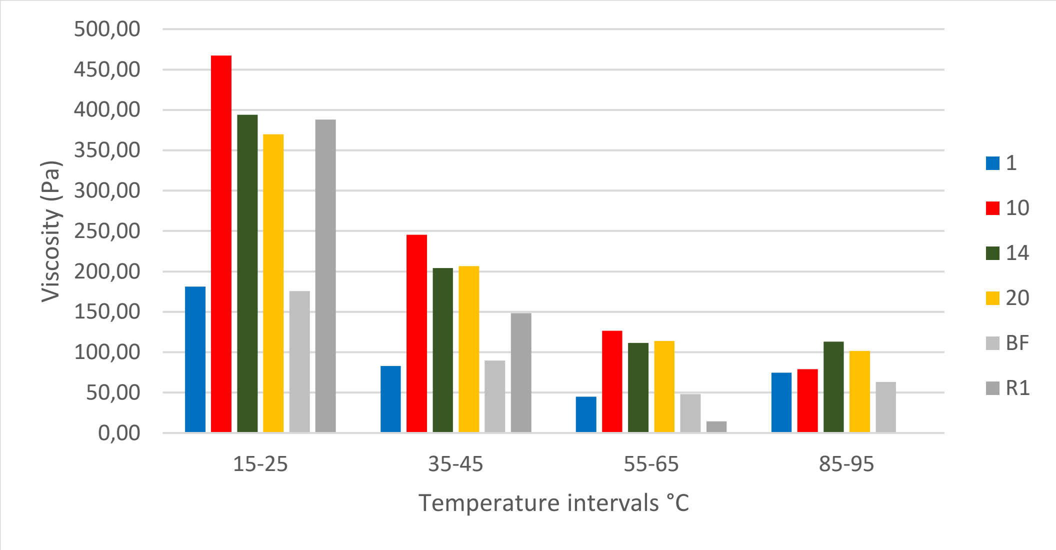
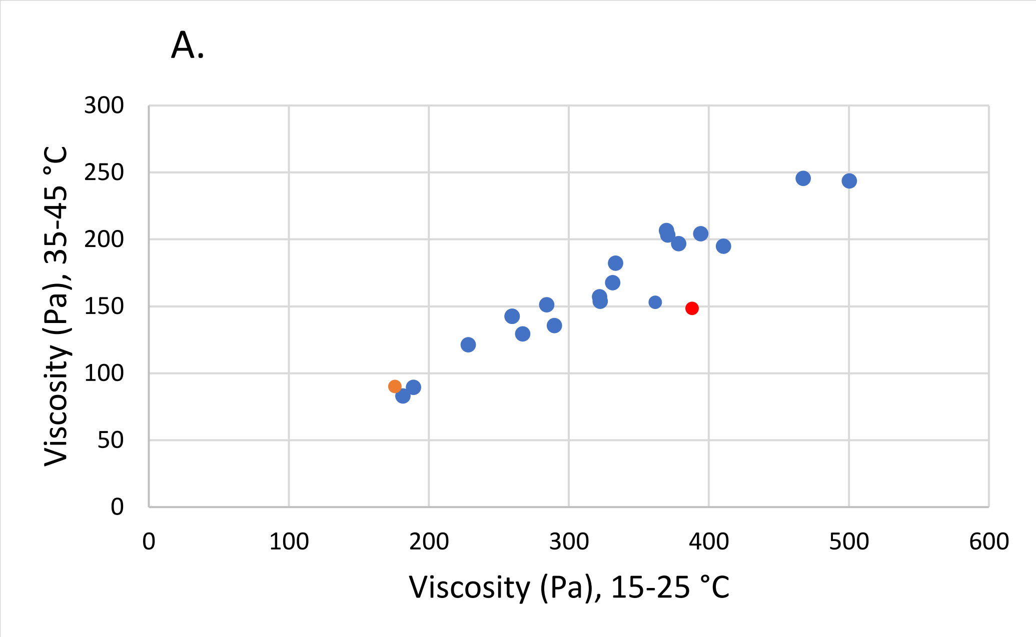


Figure 8. Raw data from sample 1, 10, 14, 20, Reference sample 1 (R1) and Baofraiche (BF) with the presented temperature intervals of 15—25, 35—45, 55—65 and 85—95 °C. The viscosity is shown on the y-axis and the different temperature intervals °C on the x-axis.

Two scatter plots were conducted to compare the intervals to each other (Figure 9) and to further analyse the change in arrangement of the samples. In Figure 9A, are the two temperature intervals of 15—25 °C and 35—45 °C compared against each other. The red circle is the reference sample, and the orange circle is the BF sample. All samples are lined up in a linear arrangement. This is not the case for Figure 9B where the temperature intervals 15—25 °C and 85—95 °C are compared. In Figure 9B, the reference sample has a very low viscosity. These scatter plots strengthen what Figure 8 indicate, meaning that there is a correlation between the different added ingredients and applied temperatures and that the effect of pectin, protein and Agar Agar differ.

When the temperature is increased to 35—45 °C, the viscosity decrease (Figure 9A). The reason behind why there is a linear arrangement could be because the viscosity almost decrease at the same magnitude for all samples, meaning that the viscosity has changed equally for all samples from the temperature interval of 15—25 °C to 35—45 °C. Therefore, a correlation between the samples occurs. However, when increasing the temperature to 85—95 °C (Figure 9B) is it shown that the correlation disappears. This indicates that the changes in viscosity are not parallel at higher temperatures and that the samples do not behave in the same way. Samples that have a high viscosity at low temperatures are not those that are most viscous at high temperatures.



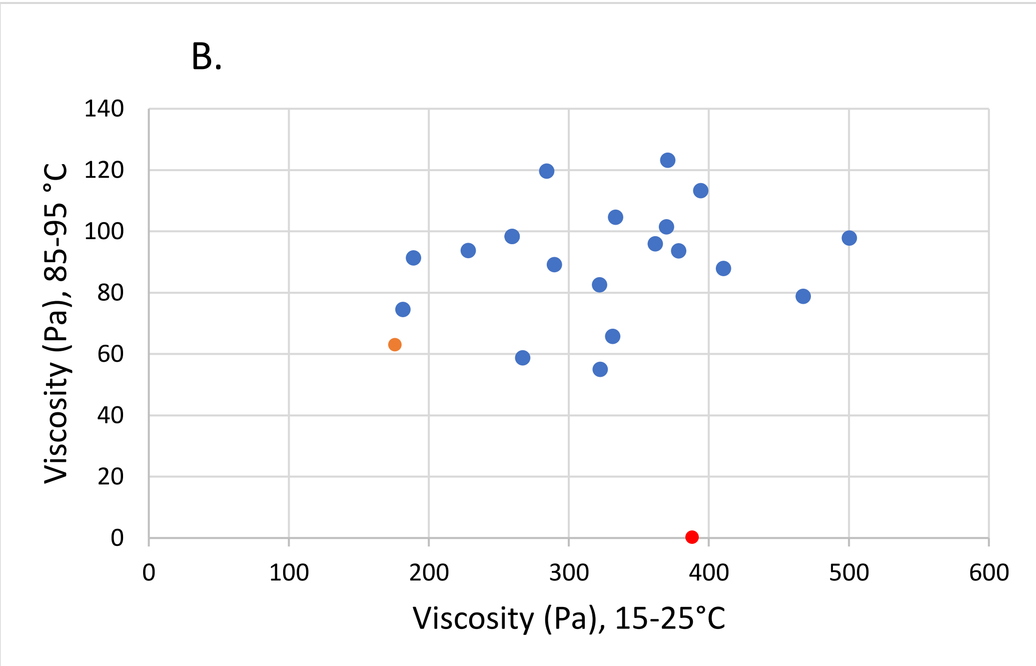


Figure 9. Representation of raw data using scatter plots. Red colour indicates reference sample, orange colour represents Baofraiche. A: temperature interval 15—25°C and 35—45°C compared and scattered against each other. B: temperature interval 15-25°C and 85-95°C compared and scattered against each other.

Due to the fact that the added ingredients are shown to have different effects on the viscosity in the experimental design, response surface regressions were run to find statistically significant effects of pectin, protein and Agar Agar. Moreover, interaction effects between the added ingredients were also investigated. Table 7 illustrates the summary of all regressions for the different temperature intervals.

The significant values are in bold where p≤0.05. The three first lines are the main factors in the design. After the main factors are the quadratic factors shown, meaning the Pectin*Pectin, Protein*Protein and Agar Agar* Agar Agar. These are included to describe the curvatures on the response surface of the contour plots (Figure 10 and Figure 11). The cross terms, meaning the Pectin*Protein, Pectin*Agar Agar and Protein*Agar Agar are included to detect interactions. The R-sq (adj) are included to describe how much of the variance of the response is described in the model. Important to highlight is that the model become less secure with higher temperatures for this design. Meaning that the model in this case is most stable at the temperature interval of 15—25 °C where the R-sq has a number of 81 in comparison to 48 at the temperature interval of 85—95 °C. This could perhaps be because the viscosity is much lower at higher temperatures which means that the measurement of the viscosity will become less secure because of the sensitivity in the instrument used. Another reason for the decreasing number of R-sq could simply be because of error in the measurements performed. However, one could argue that all R-sq values above 0 % are potentially relevant and therefore are 48 % not bad.

Table 7. Summary of results from run response surface regression for all added ingredients (factors) and temperature intervals used.

	Temperature interval °C			
Factors	**15—25**	**35—45**	**55—65**	**85—95**
Pectin	**<0.0005**	**<0.0005**	**0.003**	0.084
Protein	**0.015**	0.367	0.898	0.823
Agar Agar	**0.050**	0.068	0.117	**0.018**
Pectin*Pectin	0.671	0.434	0.252	0.131
Protein*Protein	0.304	0.075	0.112	0.704
Agar Agar*Agar Agar	*a*	*a*	*a*	*a*
Pectin*Protein	**0.015**	**0.024**	**0.033**	**0.010**
Pectin*Agar Agar	0.037	0.102	0.276	0.308
Protein*Agar Agar	*a*	*a*	*a*	*a*
R-sq (adj) (%)	81	72	52	48

a: excluded in model. In order to reduce the noise, lines where no p-value was less than 0.2 was removed. Note: Significant effects of factors (P≤0.05) on individual responses are shown in bold type.

As seen in Table 7, Pectin has a significant effect on viscosity at all temperature intervals except at 85—95 °C. Protein is only shown to have a significant effect at temperature interval 15—25 °C. Agar Agar have a significant effect at the lowest (15—25 °C) and highest (85—95 °C) temperature intervals. There is no significant effect for the quadratic ingredients. However, there is a significant effect for Pectin*Protein. When there is a significant effect on the cross terms this shows that the effect of one factor depends on the settings of the other factor.

3.6.1. Response surface - Contour plots of viscosity

Contour plots provides a two-dimensional view and are used to see how a response variable relates to two predictor variables. All points in a contour plot that have the same response are connected to produce contour lines of constant responses. Moreover, the contour plot contains coloured contour-bands or intervals that represent ranges of the response values that represents different viscosity levels for this thesis. (MiniTab 2021)

To understand the significant effect that the two factors, pectin and protein had on the dispersion, showed in Table 7, contour plots were created (Figure 10). The four temperature intervals were included with the response value that was a mean value in the interval. The contour plots show the viscosity of the combinations of different amounts of combinations of added ingredients for all 20 samples included in the design. It is only possible to plot two factors against each other at the same time in a contour plot. Therefore, a hold value is included for the contour plots, set to 2 for Agar Agar in Figure 10. The amount of added protein, presented in % of the total weight before adding oil to the dispersion, ranged from 0—10 whereas the added pectin ranged from 0—5.

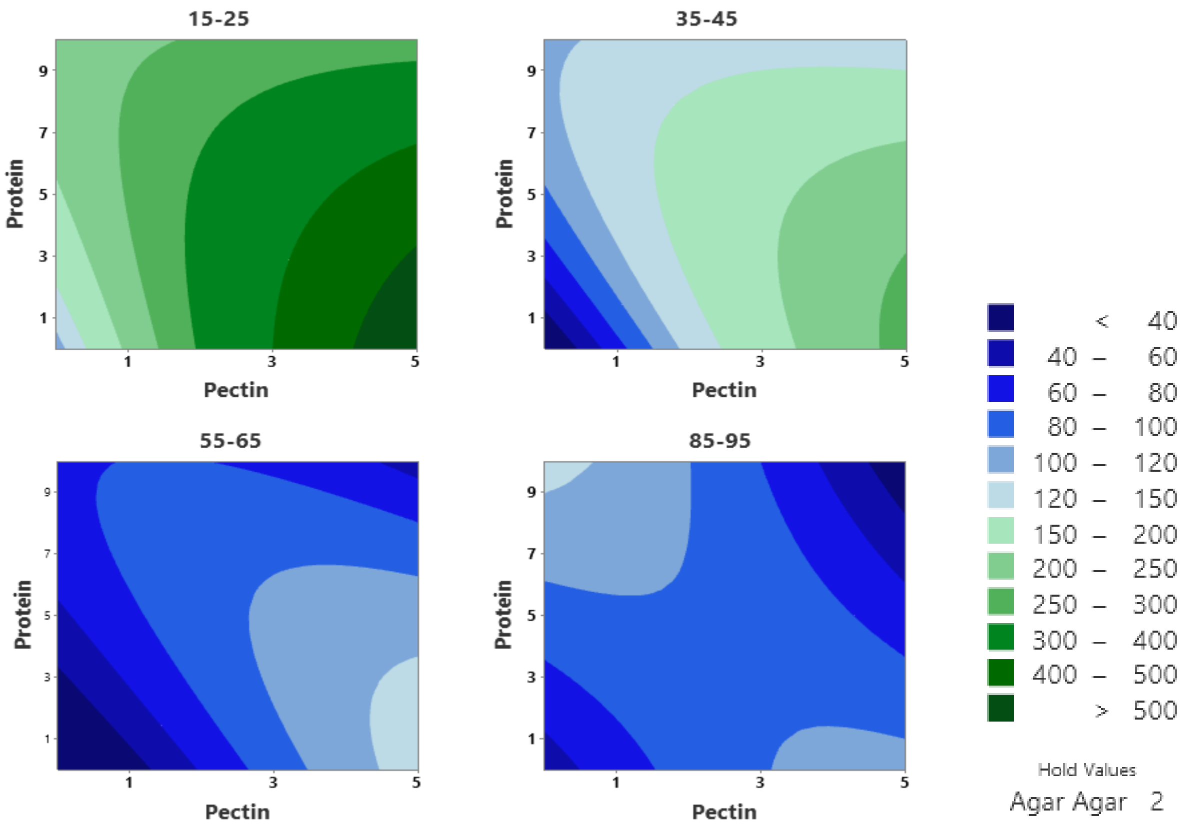


Figure 10. Contour plots of viscosity (15—25, 35—45, 55—65 and 85—95 °C) vs pectin and protein. The colour, seen on the left side in the figure, represents the different viscosity levels (Pa) ranging from 40 Pa too 500 Pa. These values are normalized to fit all contour plots presented in this thesis. The response value is a mean value in the interval. The hold value for Agar Agar was set to 2. The units for the x and y axis are the % of the total weight before adding oil to the dispersion. The added protein ranging from 0—10 and the pectin from 0—5.

The results in Figure 10 indicate that at lower temperatures, if protein is added, pectin will have less of an effect on the viscosity. Meaning that when pectin is added, the viscosity will increase, thus, if at the same time protein is added will the effect of the pectin will have less effect on the dispersion. This indicates that added protein have a negative effect on the viscosity. Moreover, if a high viscosity is desired at high temperatures pectin, or, protein need to be added, but not a combination of both. Also, the contour plot with the response surface for 85-95 C° in Figure 10 have a different surface with different shapes of lines, in comparison to the other contour plots. This means that when the added protein is high and the temperature is high as well, is the effect of protein very good. The interaction between protein and pectin are therefore much more efficient at higher temperatures.

Moreover, because the cross terms of Agar Agar and pectin was close to have an significant effect and because Agar Agar on itself had a significant effect, contour plots were created for this combination (Figure 11). Equivalent temperature intervals, viscosity levels and samples were included as in Figure 10. The hold value for protein was set to 5. The amount of added pectin, presented in % of the total weight before adding oil to the dispersion, ranged between 0—5 and from 0—4 for Agar Agar. The contour plot show that the Agar Agar have a larger effect at higher temperatures in comparison to lower temperatures. Moreover, when the added amount of pectin is somewhere in between the added extreme values will the Agar Agar have the best effect. Pectin seems to have a bigger effect on the viscosity when the temperatures are high in comparison to lower temperatures.

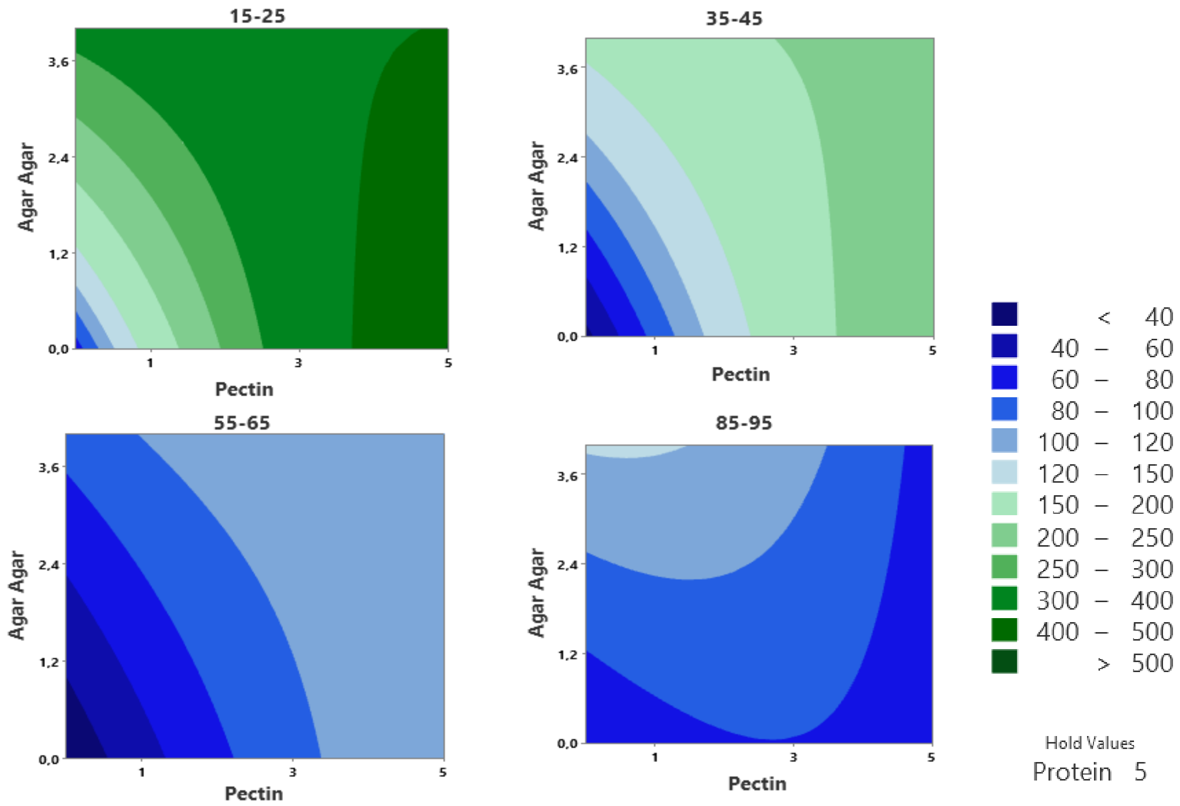


Figure 11. Contour plots of viscosity (15—25, 35—45, 55—65 and 85—95 °C) vs pectin and Agar Agar. The colour, seen on the left side in the figure, represents the different viscosity levels (Pa) ranging from 40 Pa too 500 Pa. These values are normalized to fit all contour plots presented in this thesis. The response value are a mean value in the interval. The hold value for protein was set to 5. The units for the x and y axis are the % of the total weight before adding oil to the dispersion. The added Agar Agar ranging from 0—4 and the pectin from 0—5.

Unfortunately, this complex dispersion of Baofraiche have not been studied before and it is therefore hard to make comparisons to the literature. Thus, the experimental design performed for this thesis helped in understanding what effects the added ingredients had on the dispersion of BF. However, no further experiments have been performed to develop the best possible recipe for BF. Therefore, based on the results presented in this thesis, the next step towards commercialization of Baofraiche is to develop an optimal recipe. Sensory evaluation of the BF should be taken into consideration. Most desirable is that Baofraiche, in the future, meet the demand for sustainable, nutritional and sufficient food.

4. Conclusion

In conclusion, a prototype for a cookable baobab fruit pulp-based sauce, named Baofraiche (BF), have been developed. BF have desired characteristics, meaning that it is cookable, has a good emulsion stability and it disperse well in a food system. Furthermore, BF have a pleasant taste, appearance and mouthfeel that meet the requirements set for this study.

The rheological parameters such as the viscosity of the prototype and how they are affected by different temperatures, was studied using a rheometer. An experimental design, Central Composite Design (CCD), was used in order to understand and study the effect of adding ingredients to the Baofraiche dispersion. Three factors were included in the design; pectin, protein and Agar Agar. The viscosity was shown to decrease at higher temperatures. The order of the samples changed with higher temperatures meaning that the added factors had influence on the properties of the dispersion. The results from the CCD disclosed that pectin and Agar Agar, two different polysaccharides, affect the dispersions in the design in different ways. Thus, the conclusion can be drawn that it is not possible to add any polysaccharide and predict that they will have the same effect on the dispersion. Further, the results also indicate that added protein has a negative effect on the viscosity, meaning that the viscosity decrease with added protein, in combination with the other added factors. Therefore, these factors should not be added together if a higher viscosity is required.

Important to highlight is that the results presented in this thesis are carefully selected because they represent and answer the main objectives of this thesis. However, the CCD provides more information than presented in this thesis. Based on these findings is it clear that the CCD gives lots of data, but it is beyond the scope of this thesis to analyse in detail all the result received from the CCD. Rather, to mention that the choice of experimental design depending on the aim of the experiment is of great importance.

To conclude, baobab fruit pulp is nutritional with a dispersion useful for producing healthy, clean labeled sauce products. The developed sauce, Baofraiche, is cookable and disperse well in a food system.

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6. Appendix – Popular Scientific Summary

The global food system of today, stands for nearly one third of the world greenhouse gas emission. Because of this, it seems essential to redesign the world food systems towards being more sustainable and environmentally friendly. Moreover, the rising popularity and demand of plant-based food has caused a shift in the food industry. This has opened up for new business opportunities towards sustainable and innovative products that can face future adjustments towards a sustainable food system. Baofraiche, a plant-based sauce, have been developed and suggested in relation to the necessity of redesigning the global food system towards being more sustainable. This is one of the conclusions from the master thesis: *Effects of polysaccharides and protein on rheological properties of a baobab based sauce - cookable with sustained rheological properties*

The plant-based sauce, Baofraiche, developed in this master project was a collaboration between the Swedish University of Agricultural Sciences and Arwa Food Tech AB, member of Aventure AB group. Baofraiche consist of baobab fruit pulp. Baobab is a wild, fruit bearing tree that is mainly found in Africa. Baobab fruit pulp, found inside hard pods hanging from the tree are usually dried and ground to a fine white powder that are used for several food products. The baobab fruit is known to be very nutritional and it contains a lot of pectin. Pectin is used in the food industry as a gelling agent or stabilizer, particularly in jams, for dessert fillings or in sauces and drinks. The high amount of pectin makes the baobab fruit pulp very interesting while developing a plant-based sauce. This is because one could argue that no thickeners or gelling additives will be needed to reach the right consistency of the sauce.

Baofraiche is aimed to be used as an ingredient in cold or hot food, as an alternative to commercial plant or milk-based crème fraiche. Moreover, it was of importance that the sauce could be heated without effecting a food system in a negative way. To find the best possible recipe for Baofraiche, three ingredients were added to the sauce. The effect of the added ingredients was carefully studied and analysed. The conclusion from this master thesis showed that Baofraiche was cookable and it dispersed well in a food system. The added ingredients affected the sauce in different ways. Thus, further experiments can be conducted to find the best possible recipe for Baofraiche based on the findings in this thesis.