

PREDICTION OF THE MAXIMUM TEMPERATURE OF ORGANOGELOATION

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Abstract: Factors are practical guidelines to classify food additives and products, such as ratio, hydrophilic-lipophilic balance (HLB) and sucrose equivalent. Among the main difficulties in the application of organogels is their classification, which would facilitate their use and comparison with other additives. This work evaluated the efficiency of an empirical factor called maximum temperature of organogelation (O_T) to predict the physical state of oil solutions reported in the literature.

Keywords: Organogels; vegetable oils; vegetable waxes; structured vegetable oils; crystallization; fat replacers.

INTRODUCTION

Margarines, spreads, salad dressings, cakes, cookies, ready-to-eat fillings, ice cream and chocolates are examples of food products in which the use of organogel can improve sensory characteristics, accelerate structuration, reduce defects caused by oil migration, replace trans fatty acids and reduce the concentration of saturated fats (Singh et al., 2017; Canizares et al., 2020a).

A gel can be called a hydrogel when the immobilized liquid phase is hydrophilic and organogel, or oleogel, when it is hydrophobic (Blake et al., 2014). Either low molecular weight polymers or molecules can be used to structure hydrophobic liquids (Terech and Weiss, 1997). Some points must be highlighted before approaching the phenomenon of organogelation in a more comprehensive way. First, organogelators are the compounds responsible for structuring the oil and have a lower molecular weight than the polymers commonly used to gel polar liquid media, so organogelators are referred to as low molecular weight compounds (Terech and Weiss, 1997; Dassanayake et al., 2009). Second, the presence of crystals has been shown to be mandatory for the formation of organogels (Canizares et al., 2020a). Finally,

the oil structuration by low molecular weight molecules is the result of crystal-liquid and crystal-crystal interactions, and the possible existence of crystalline nanofibers forming a matrix capable of containing the liquid phase (Terech and Weiss), 1997; Blake et al., 2014).

The oil structuration processes using organogelators are simple. A solution of oil containing organogelator is heated for a time and temperature sufficient to erase the crystalline memory of the previously existing crystals in the organogelator (Canizares et al., 2020a). Then, the solution must be cooled to a temperature below the jelling transition temperature. Since the structuration is conditioned to the presence of crystals, the structuration temperature is expected to be below the crystallization temperature of the solution (Canizares et al., 2020a).

The entire organogelation process is limited by the creation of sufficient crystal-liquid and crystal-crystal interactions capable of structuring the oil. The phenomenon of crystallization of the components solubilized in vegetable oil depends on the generation of a supersaturated state, the nucleation of crystallizable molecules, the crystal growth and, finally, the recrystallization or reorganization of these crystals (Mullin, 1961; Hartel, 2001). The supersaturated state can be induced by increasing the solute concentration, by adding a third component capable of influencing the solubility of crystallizable compounds in the liquid phase, or by reducing the temperature of the solution (Mullin, 1961; Hartel, 2001).

The minimum amount of wax needed to produce an organogel, known as the critical concentration, has been reported to be between 1 and 5 % (Dassanayake et al., 2009; Hwang et al., 2013; Blake et al., 2014; Öğütçü and Yılmaz, 2014; 2014a; Öğütçü and Yılmaz, 2014b; Öğütçü and Yılmaz, 2015), a variation that is understandable due to the significant

differences between the compositions of waxes and oils depending on the source and extraction methods (Barthlott et al., 1998; Racovita et al., 2016).). Knowing that the solubility of the solute is dependent on the temperature of the solution (Mullin, 1961), the amount of wax needed to structure a vegetable oil will depend on the temperature at which the organogel is desired (Dassanayake et al., 2009; Hwang et al., 2013; Blake et al., 2014; Öğütçü and Yılmaz, 2014a; Öğütçü and Yılmaz, 2014b; Öğütçü and Yılmaz, 2015).

Although these basic knowledge are not completely ignored in the articles about organogels and its applications found in the literature, the concentrations of waxes added to products were apparently chosen by the authors in a more subjective or observational way than based on technical data (Moghtadaei et al., 2018; Yılmaz and Öğütçü, 2015). This may be the result of the lack of an indicator or factor that would offer, in a practical way, a guideline on how much and which organogel should be added to a product. This type of factor is commonly used in the food industry, such as the ratio in citrus fruits processing industries, the hydrophilic-lipophilic balance (HLB) for emulsifiers, the sucrose equivalent for water activity, among others. However, little attention has been given to the needs to standardize the methods of physical properties characterization for organogels, which causes a lack of basic information. Consequently, it makes impossible to compare results, directly affecting decision-making and in-depth understanding of the mechanisms and variables involved on organogelation. This problem was already explored in a previous article (Canizares et al. 2020a).

In 2017, an expanded abstract exposed at IFT2017 (Canizares et al., 2017), presented an empirical factor called maximum temperature of organogelation (O_T). This variable is obtained from equation (1) and

would work as an indication of organogel formation. Briefly, it means that solutions with O_T values above the temperature (T) at which structuration is desired would form organogels (Canizares et al., 2017). O_T is obtained from a direct relationship between the onset of crystallization temperature (T_c) and the oil binding capacity (OBC), that could be used as a factor to assist in the choice of an organogel or organogel.

$$O_T = T_c \times V_{OBC} \quad (1)$$

and O_T and T_c are temperatures in °C and V_{OBC} is the fraction obtained from the determination of the oil binding capacity ($V_{OBC} = OBC/100$), dimensionless.

The objective of this work is to evaluate the efficiency of the use of O_T in determining the physical state of the wax-in-oil solutions previously reported in the literature. The articles selected for the work presented the data of onset of crystallization temperature (T_c), the oil holding capacity (OBC) and the physical state of the solution at a specific temperature.

MATERIALS AND METHODS

Articles addressing to vegetable oils structuration were analyzed in the search for the three key information necessities to determine O_T . Thus, the data from the articles were selected exclusively due to analytical methods. As it is an area of recent interest, there are a wide variety of methods carried out to analyze organogels. However, authors have been striving for a standard to be adopted in the analysis of organogels, especially with regard to the binomial of time and temperature necessary to obtain replicable results of structuring the samples (Canizares et al., 2020a).

In the context of O_T determination, protocols must be respected to obtaining relevant data of onset of crystallization

temperature (T_c) and oil binding capacity (OBC). Furthermore, methods to determine the oil structuration must be reliable and replicable. It is expected that the following analyses were present in the articles:

Structuration: Visual assessment of apparent solidification is the simplest and most reported method to demonstrate the organogelation. This method consists in place solutions of wax-in-oil into test tubes with a minimum diameter of 1.5 cm. The sample volume inside the test tube is variable, but a minimum 5 cm height of the sample in the tube is desirable. Then, solutions must be heated at temperatures between 70 °C and 100 °C for at least 20 minutes to erase the crystalline memory (Canizares et al., 2020a). Subsequently, the samples are kept at constant temperature inside, or not, controlled temperature chambers, for 24 h, to reach an equilibrium temperature and to the crystalline structure assume a stable form. Finally, the tubes are fully or partially flipped to allow the analyzer to see which samples are fully structured. Some authors point out that these analyzes must be later confirmed by rheological analyses. On the other hand, numerous articles have reported a strong relation between visual and rheological analyses, demonstrating that the visual method may be capable to determine the physical state of a wax-in-oil solution (Blake et al., 2014; Canizares et al., 2020b).

Onset of crystallization temperature (T_c): T_c must be determined through differential scanning calorimetry with a protocol that meets similar conditions of the sample during cooling for structuration analysis. Typically, the sample is expected to be heated to between 70°C and 100°C and held for a period of at least 20 minutes at this temperature. Subsequently, the sample must be cooled at a constant rate to values below 0 °C. The most common cooling rates are 5°C and 10°C for

samples that the structuration was carried out at room temperature. The different cooling rate can cause alteration on the T_c , which is determined at the starting point of the first exothermic peak, which indicates the beginning of crystallization.

Oil binding capacity (OBC): Oil binding capacity must be determined through a method in a centrifuge. In this analysis, about 1 gram of preheated sample at a temperature between 70 °C and 100 °C for at least 20 minutes is transferred while still hot to Eppendorf tubes (1.5 mL) of known weight. Samples are cooled to induce crystallization and generate possible structuration (Canizares et al., 2020b). Some methodologies reported different cooling processes to obtain the desired temperature and structuring before the centrifugation process (Öğütcü and Yilmaz, 2014a; Öğütcü and Yilmaz, 2014b; Öğütcü and Yilmaz 2015; Fayaz et al., 2017). However, the most coherent methodology is the one used by Fayaz et al. (2017), in which the sample is placed at a known and controlled temperature for 24 h and then centrifuged at 10,000 rpm for 15 min at the same temperature. Then, the supernatant liquid oil is removed, the Eppendorf is weighed again, and the values of the oil binding capacity are obtained gravimetrically, as proposed in the literature (Canizares et al., 2020b).

RESULTS AND DISCUSSION

In Table (1), it was compiled all the T_c and OBC values present in the literature where methodologies similar to those previously cited were carried out. The temperature of structuration (T) was indicated as the room temperature in all articles. The room temperature was reported to be 20 °C in all the articles that mentioned this information. Knowing that temperature is a critical variable for crystallization (Fayaz et al., 2017; Canizares et al., 2017; Canizares et al. al., 2020b) and

laboratories tend to have mild temperatures, in cases where the structuring temperature was only described as being the room temperature, it was considered T as being $20\text{ }^{\circ}\text{C}$ (Öğütçü and Yılmaz, 2014a; Öğütçü and Yılmaz, 2014b; Öğütçü and Yılmaz, 2015). Thus, it was expected that wax-in-oil solutions kept at a structuration temperature (T) lower than O_T values would present structuration, forming organogels (Canizares et al., 2017).

The efficiency of O_T (equation 1) in predicting the physical state of the wax-in-oil solutions from data reported in the literature was initially calculated to be 92.5%. From 40 results reported, 3 of them presented O_T values without adherence to the observed physical state. However, the data presented in Table (1) are literally those presented in the articles and, therefore, it is necessary to make some comments. Starting with the solution carnauba wax (3 % w/w) in olive oil (Öğütçü and Yılmaz, 2014a), it can be noticed that there may have been a slight mistake in compiling the onset of crystallization temperatures (T_C). It was expected an increase of the T_C with the wax concentration, which is not observed when verifying the T_C of carnauba wax in olive oil at concentrations of 7 and 10 % (Öğütçü and Yılmaz, 2014a). Assuming only the accidental inversion of the results, the correct onset of crystallization temperatures would be $53.6 \pm 1.5\text{ }^{\circ}\text{C}$ for 3 %, $56.8 \pm 0.6\text{ }^{\circ}\text{C}$ for 7 % and $63.8 \pm 0\text{ }^{\circ}\text{C}$ for 10 %. Under these conditions, the new O_T values would be $21.9\text{ }^{\circ}\text{C}$, $40.7\text{ }^{\circ}\text{C}$ and $59.6\text{ }^{\circ}\text{C}$, respectively (Öğütçü and Yılmaz, 2014a).

The article by Yi et al. (2017) fails to confirm whether the solutions are solid or not. On the other hands, based on the concentrations used and what was indicated in the text, it is understood that all samples were structured at room temperature. Furthermore, in this article, the authors considered the highest exothermic peak found in the graphs as the

responsible for the onset of crystallization temperature of the solution. In fact, the highest exothermic peak observed in a thermogram may refer to the crystallization of the most abundant compound, but does not necessarily correspond to the beginning of the solution crystallization. Thus, the T_C of the solution must be obtained at the first exothermic peak. Fortunately, the authors presented the thermograms, making it possible to verify the results (Figure 1 of the article by Yi et al., 2017). For solutions of beeswax in grape seed oil, it is possible to observe that the onset of crystallization temperatures reported (Table 1) were different from the T_C of the solution. If the authors had chosen the T_C of the solution, as expected, the T_C values would be approximately 37°C , 46°C and 48°C for 5, 10 and 15% beeswax in grape seed oil, respectively (values manually extracted from Figure 1B of the article by Yi et al., 2017). Considering the new T_C , the O_T values would be $36.8\text{ }^{\circ}\text{C}$, $45.9\text{ }^{\circ}\text{C}$ and $47.8\text{ }^{\circ}\text{C}$ for 5, 10 and 15 % of beeswax in grape seed oil, respectively. The T_C reported by Yi et al. (2017) for solutions of carnauba wax in canola oil did not have a sensitive impact. In these cases, the highest exothermic peak found in the graphs had the onset of crystallization temperature very close to the T_C of the solution.

After observations made above, regarding the results presented in Öğütçü and Yılmaz (2014a) and Yi et al. (2017) articles, it is possible to notice an increase in the prediction efficiency of the physical state of the solutions (equation 1), to 97.5%. The only unexpected result, even after the corrections, was for the solution of carnauba wax in olive oil, 3 % (w/w) which presented a corrected value of O_T equal to $21.9\text{ }^{\circ}\text{C}$ and did not present an apparent structuration (Öğütçü and Yılmaz, 2014a).

One hypothesis for the deviation observed between the calculated O_T value and the result

Organogelator	Oil	% of wax	Structured	T_c (°C)	OBC (%)	O_T
Carnauba wax ^a	Olive	3	No*	63.8 ± 0	40.8 ± 0.2	26
		7	Yes *	53.6 ± 1.5	71.7 ± 1.6	38.4
		10	Yes *	56.8 ± 0.6	93.4 ± 0.4	53
Monoglycerol ^a	Oliva	3	Yes	37 ± 0.5	62.3 ± 0.5	23
		7	Yes	45.2 ± 0.3	80.7 ± 5.3	36.4
		10	Yes	49.0 ± 1.5	99.9 ± 0.1	48.9
Carnauba wax ^b	pomegranate seed	3	No	48.5 ± 0.4	39.2 ± 5.5	19
		7	Yes	55.7 ± 0.5	73.3 ± 3.6	40.8
		10	Yes	56.2 ± 1.7	97.3 ± 2.7	54.6
Monoglycerol ^b	pomegranate seed	3	No	32.3 ± 0.2	37 ± 0.6	11.9
		7	Yes	43.8 ± 1.3	54 ± 1.8	23.6
		10	Yes	48.5 ± 1.1	72.3 ± 0.1	35
Carnauba wax ^c	Canola	15	Yes **	62.9 ± 2.5	99.4 ± 0.2	62.5
		10	Yes **	60.3 ± 0.5	99.0 ± 0.2	59.6
		5	Yes **	54.3 ± 3	85.1 ± 4.2	46.2
Beeswax ^c	Grape seed	15	Yes * **	21.7 ± 0.2	99.5 ± 0.2	21.5
		10	Yes * **	19.8 ± 0.3	99.7 ± 0.5	19.7
		5	Yes * **	15.1 ± 1.9	99.5 ± 0.1	15
Sunflower wax ^d	hazelnut oil	3	Yes	60.8 ± 6.9	99.8 ± 0.1	60.6
		7	Yes	60.1 ± 0.5	99.8 ± 0.1	60
		10	Yes	62.4 ± 0	99.8 ± 0.1	62.2
Carnauba wax ^d	hazelnut oil	3	No	47.9 ± 0.2	39.8 ± 2.3	19
		7	Yes	52.4 ± 0.6	80.6 ± 5.6	42.2
		10	Yes	54.8 ± 1.4	97.7 ± 2.1	53.6
Linseed Straw Wax ^e	Canola	1	No	29.8 ± 1.1	17.4 ± 1.1	5.2
		2	No	31.4 ± 0.7	33.4 ± 2.9	10.6
		3	No	33.3 ± 1	59.8 ± 1.9	20
		4	Yes	37.6 ± 4.5	63.3 ± 3.5	23.8
		5	Yes	40.9 ± 1.3	67.7 ± 3.7	27.8
		6	Yes	41.4 ± 0.2	78.2 ± 1.1	30.2
Wheat Straw Wax ^e	Canola	1	No	23 ± 4.1	5.4 ± 1.1	1.2
		2	No	28.6 ± 1.2	21.3 ± 2.8	6.2
		3	No	31.2 ± 0.5	45.1 ± 1.9	14.2
		4	No	35.5 ± 2.1	49.1 ± 3.5	17.6
Linseed Straw Cuticular Wax ^f	Canola	5	Yes	36.6 ± 0.6	55.5 ± 3.7	20.4
		6	Yes	38.3 ± 4.1	61.1 ± 1.1	23.4
		4	Yes	40.8 ± 0	63.3 ± 3.5	25.8
Wheat Straw Cuticular Wax ^f	Canola	5	Yes	38.2 ± 1.5	56.1 ± 1.3	21.4
Linseed Straw Epicuticular Wax ^f	Canola	2	Yes	38.1 ± 1.2	72.4 ± 12	27.6
Wheat Straw Epicuticular Wax ^f	Canola	4	Yes	32.4 ± 0.8	80.5 ± 10	26.2

* Possible error in data analysis. ** Failure or inconsistency in demonstrating whether or not samples are structured.

^a Öğütçü e Yılmaz, 2014b; ^bÖğütçü e Yılmaz, 2014a; ^c Öğütçü e Yılmaz, 2015; ^dYi et al., 2017; ^eCanizares et al., 2017; ^fCanizares et al., 2020b

Table 1: Presents the physical state, T_c and OBC results found in the literature to support the proposition of using the maximum temperature of organogelation (O_T) to predict the physical state of wax in oil solutions at 20 °C.

of the sample of carnauba wax in olive oil (3 % w/w) could be related to minimal variations of the room temperature, which was not reported. Although usually considered to be 20°C, the room temperature of a laboratory can vary from one season to another in places where there is no air conditioning. For example, if the ambient temperature was about 22 °C and not 20 °C, the O_T would be confirming the non-structuring. For this reason, controlling the temperature of the environment where structuring takes place is very important.

CONCLUSION

The maximum temperature of organogelation (O_T) showed good efficiency to predict the physical state of the wax-in-oil solutions reported in the literature. A relevant fact demonstrated with the use of the equation

proposed in this work is that the O_T helped to identify non-coherent results and, in some cases, was able to find possible inconsistencies.

It is still necessary to increase efforts in obtaining relevant data from standardized methodologies. Furthermore, it will be interesting to relate the physical properties of food products made with organogel and the O_T values of the organogel. In the future, O_T values would be able to indicate the organogel purpose of use and enable to enhance their characterization.

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