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Preparation of Nanoemulgels Containing Lemon Essential Oil and Pectin: Physical Stability and Rheological Properties

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Featured Application: The knowledge obtained by this work is a contribution to the field of nanoemulgels and it could be of great interest for the development of new products based on them.

Abstract: Nanoemulgels are novel formulations of great interest for their use as dual-release systems and as fat substitutes in foods. Lemon essential oil, with a large number of benefits due to its antimicrobial, antifungal, and medicinal properties, and low methoxyl pectin, a natural polysaccharide capable of gelling by adding divalent ions such as calcium, are very appropriate ingredients to produce nanoemulgels with potential applications in industries such as cosmetics, agrochemistry, pharmaceuticals, or food. In this work, lemon-essential-oil-in-water nanoemulgels containing low methoxyl pectin derived from citrus peels were prepared following a three-step process that involves the preparation of a nanoemulsion, a pectin gel, and the mixture of both. In the first stage, the stirring time and the rotational rate employed during the mixing step were assessed. Once the preparation protocol was established, the pectin gel/nanoemulsion mass ratio was investigated. Different techniques were combined to evaluate the influence of the processing and the composition variables on the particle size distribution, mean diameters, flow curves, and physical stability of different emulgels obtained. It was found that the processing variables studied, stirring time, and rotational rate, do not influence the mean particle size of the emulgel, with values matching those of the starting nanoemulsion. However, 3 min and 200 rpm were selected for exhibiting the lowest TSI values. Regarding the composition, a higher content of pectin gel caused a higher viscosity, and therefore a higher physical stability, with the 75P/25E emulgel being the most stable. Aggregation of gel particles, because the pectin gel was really a sheared gel, was the main responsible contributor to the results obtained. This work highlights the importance of the preparation and formulation variables to develop stable, innovative formulations based on nanoemulgels.

Keywords: nanoemulgel; microfluidization; lemon essential oil; low methoxyl pectin; sheared gel

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1. Introduction

Emulgels [1,2], emulsion gels [3], emulsion-filled gels [4], gelled emulsions, or creamed gels [5] are some of the different ways to name a system consisting of the dispersion of emulsion droplets in a gel-like matrix or structured phase [6]. Depending on the type of emulsion, they can be classified as oil-in-water or water-in-oil emulgels. At the same time, depending on the size of the droplet, they can be a macroemulsion gel, a nanoemulgel, or a microemulsion gel [5]. However, regardless of their classification, all of them are semisolid emulsions.

In the cosmetic and pharmaceutical industries, emulgels have been employed extensively for some time [7–10] and more recently in the food industry [11,12]. Emulgels, which are formed by an emulsion and a gel-like structure, share the properties of both and can

therefore be used as a dual control release system [13], namely, as delivery systems for hydrophilic and lipophilic ingredients (drugs, carotenoids, vitamins, probiotics, unsaturated fatty acids, etc.). Additionally, as a promising application in foods, they could also be used as fat replacers [14]. Emulgels are capable of overcoming the low solubility and chemical stability, as well as the poor absorption, bioaccessibility, and bioavailability of some active ingredients, while improving the texture, bioadhesivity, and penetration ability into the skin of the formulations [14–16]. These advantages are more relevant in nanoemulgels, that is, when the size of the droplet in the emulsion is less than 500 nm [17].

Three components are essential in the formulation of an emulgel, namely, the dispersed phase, the emulsifier, and the structured phase. As a dispersed phase, essential oils, due to their antifungal, antimicrobial, and therapeutic properties, are a good choice [18]. Among them, lemon essential oil has proven these properties [19–25]. Furthermore, the FDA considers it a safe ingredient [26]; therefore, it could have applications in industries as diverse as food, agrochemicals, cosmetics, flavorings, and household cleaning products. It should be noted that its inclusion in a formulation such as an emulgel is very interesting because this structure protects the lemon essential oil against oxidation, one of the main drawbacks of its use. Several papers on emulgels containing essential oils can be found in the literature. By way of an example, it is worth mentioning the work published by Kharat et al. (2022) [27] who studied emulgels formulated with tea tree oil and thymoquinone to be used to reduce psoriasis disease, or the articles reported by Srivastava et al. (2018) [28] and da Silva et al. (2021) [29] for the treatment of vaginal candidiasis by the formulation and preparation of an emulgel containing mentha essential oil and clove oil, respectively, or Abdallah et al. (2023) [30] who after optimizing the formulation of erythromicynin-loaded transethosomes obtained erytromicynin-loaded transethosomes in cinnamon-oil-based emulgels to treat topical bacterial infections, or Azam et al. (2023) [31] who evaluated anise-based emulgels to also treat skin infections caused by bacteria. Emulsifiers are adsorbed at the oil/water interface, facilitating not only the formation of the emulsion but also its physical stability, thus allowing the shelf life of the emulgel to be prolonged. Tween and Span are nonionic surfactants widely used as emulsifiers in food, cosmetic, or biomedical applications because of their low toxicity and compatibility with the other ingredients of the formulation. Their mixture could help improve the stability of products compared to surfactant alone [32,33]. Regarding the structured phase, it is worth noting the relevance of selecting inexpensive and natural gelling agents that contribute to health to respond to current consumer demands. Pectin satisfies these conditions. It consists of long chains made up of galacturonic acid units, which can be found as such with a free carboxyl group or with a carboxyl esterified by methanol (methoxylated). Units of galacturonic acid are linked by α1-4 bonds. [12]. When the degree of esterification is less than 50%, pectin is termed low methoxyl pectin. These pectins form gels in the presence of calcium ions or other divalent cations. The egg box model has been widely accepted and describes that gelation occurs as a consequence of the interaction between calcium ions and the carboxyl groups of the pectin chains. [34]. Pectin has been incorporated into emulsions or nanoemulsions in numerous works [35–37] to provide structure or stability to the sample, but it should be noted that pectin was added as a powder or in solution, not in the form of a gel. Really, there are a few works that use pectin as a gelled matrix for the preparation of an emulgel, as occurs in this investigation. Among them, it is worth mentioning those carried out by Lupi et al. (2015) [38], Hou et al. (2017) [39], Ren et al. (2020) [40], Jiang et al. (2021) [41], or more recently that of Zhang et al. (2022) [42]. Interestingly, different gelation mechanisms for pectin were used in these works.

There are different preparation processes for the emulgels. One of them commonly used is that which involves three steps: (a) preparation of an emulsion, (b) preparation of a gel, and (c) mixing of both the emulsion and gel [5]. In order to obtain nanoemulgels, nanoemulsions should be obtained. Different high-energy devices could be used for this purpose, such as ultrasonic equipment, high-pressure homogenizers, or microfluidization devices. Another important issue to consider when preparing an emulgel is the control of

the processing variables during the third step, namely, mixing both components because the physical stability of the nanoemulgel could depend on it.

In this work, lemon-essential-oil-in-water nanoemulgels were studied containing low methoxyl pectin derived from citrus peels as the structured phase and a mixture of Tween 80 and Span 20 as the emulsifiers, which were prepared as indicated above. Microfluidization with F12Y and H30Z interaction chambers arranged in series was used to obtain the nanoemulsions. The review of the literature exposed that there were very few works that used low methoxyl pectin as a gel-like matrix and even fewer that used microfluidization to obtain nanodroplets. Another aspect little studied was the influence of long-term aging on this type of sample. We have recently published a work in which, using the same ingredients as in this study, the effect of a change in the $CaCl_2$/pectin mass ratio on the properties of emulgels prepared following a different protocol was assessed [12]. Now, in the first stage, the influence of the rotational rate and stirring time during the mixing step between the nanoemulsion and the gel on the particle size distribution and physical stability of the emulgel obtained was investigated. Next, in the second stage, the influence of the formulation, evaluated through different pectin gel/emulsion mass ratios, on the particle size distribution, rheology, and physical stability of the final emulgel was assessed. For these purposes, laser diffraction, multiple light scattering, and rheology were the techniques used in this investigation to assess the quality of different emulgels obtained, allowing the determination of which processing variables and formulations were the most appropriate to obtain stable emulgels. This work brings to light the important role of both the formulation and preparation in promoting a stabilization process in essential oil emulgels.

2. Materials and Methods

2.1. Materials

Lemon essential oil (batch AELIE028), employed as the dispersed phase, was provided by Bidah-Chaumel (Murcia, Spain). Tween 80 and Span 20 (HLB 15 and 8.6, respectively), used as emulsifiers, were purchased from Sigma Aldrich (Madrid, Spain). As a structured phase, pectin derived from citrus peels (Aglupectin LC S18YP, batch CL1811, Rende, Italy) with a methoxylation degree between 37 and 41% courtesy of JRS Silvateam Ingredients Srl (Rende, Italy) was used. Other ingredients in the formulation were $CaCl_2$ anhydrous (batch 0E012485), citrate buffer solution (citric acid anhydrous (batch 0001923544), tris-sodium citrate 2-hydrate (batch 0001936140), and deionized water) purchased from Sigma-Aldrich (Madrid, Spain), and deionized water. All materials were used as received.

2.2. Emulgel Preparation

Firstly, a nanoemulsion was prepared based on the report of the experimental method of Santos et al. [43] with minor adjustments. For this purpose, a coarse emulsion was obtained by adding to the aqueous phase (Tween 80 and Span 20 (HLB 12) [43] and deionized water) the appropriate amount of lemon essential oil using an Ultraturrax T-50 (Ika Werke GmbH and Co. KG, Breisgau, Germany) rotor-estator device for 40 s at 2000 rpm and then for an additional 90 s at 4000 rpm. The particle size was then reduced by passing the sample through a laboratory-scale microfluidizer (M-110P, Microfluidics International Corporation, Westwood, MA, USA) having a Y-type (F12Y, with 75 µm minimum internal dimension) and Z-type (H30Z, with 200 µm minimum internal dimension) interaction chamber arranged in series, at 20,000 psi and 2 passes.

Secondly, a pectin gel was obtained. Pectin powder (0.3 wt%) was dispersed in a citrate buffer solution (pH = 4.2) and deionized water employing a rotor-estator homogenizer (Ultraturrax T-50) at 2000 rpm for 1 h. Subsequently, gels were obtained by adding $CaCl_2$ in solution (7% w/v stock solution) following the method developed by Lupi et al. (2015) [38] and Lorenzo et al. (2013) [44], that is, stirring at 8000 rpm for 30 s using the same device. The sample was kept at an ambient temperature for 24 h to ensure that the pectin was properly hydrated.

Finally, emulgels were obtained by mixing different proportions of freshly prepared nanoemulsions and pectin gel using an IkaVisc MR-D1 device (Ika Werke GmbH and Co. KG, Breisgau, Germany) and a sawtooth-type impeller. Then, they were kept at 25 °C and allowed to stand until the analyses.

A schematic diagram of the emulgel preparation process can be found in Figure 1.

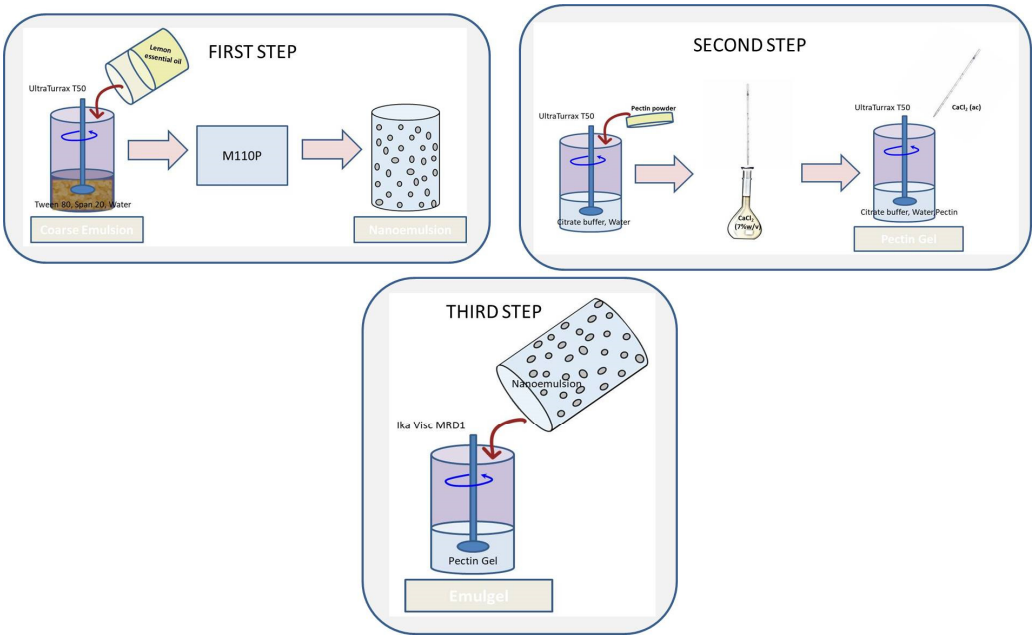

Figure 1. Schematic diagram of emulgel preparation.

Table 1 shows the composition of the emulgels. These compositions have been selected as intermediate values of the two extreme conditions that correspond to the nanoemulsion and the pectin gel. The AP/BE nomenclature will be used to denote the emulgels, with AP being the pectin gel mass proportion and BE being the emulsion mass proportion. It should be noted that the initial emulsion contained 15 wt% lemon essential oil and 15 wt% emulsifiers, while the initial pectin gel contained 0.3 wt% pectin and a $CaCl_2$/pectin mass ratio of 1.

Table 1. Composition of the emulgels studied.

Emulgels	Oil (wt%)	Emulsifier (wt%)	Pectin (wt%)	$CaCl_2$ (wt%)	Buffer + Water (wt%)
75P/25E	3.75	3.75	0.225	0.225	92.05
50P/50E	7.50	7.50	0.150	0.150	84.70
25P/75E	11.25	11.25	0.075	0.075	77.35

2.3. Particle Size Distribution

The particle size and size distribution (PSD) of the emulgels were determined using a Mastersizer 2000 laser particle sizer (Malvern, Worcestershire, UK). For this purpose, the emulgel was dripped into the dispersion chamber containing water until the laser obscuration reached a value around 15%. The refraction indices were 1.474 and 1.330 for lemon essential oil and water, respectively. Each test was carried out at least twice and at an ambient temperature.

The size of the emulgels is expressed as $D_{3,2}$ and $D_{4,3}$ and the polydispersion degree is quantified by the span parameter:

$$D_{3,2} = \frac{\sum_{i=1}^{N} n_i d_i^3}{\sum_{i=1}^{N} n_i d_i^2} \quad (1)$$

$$D_{4,3} = \frac{\sum_{i=1}^{N} n_i d_i^4}{\sum_{i=1}^{N} n_i d_i^3} \quad (2)$$

$$Span = \frac{D(v, 0.9) - D(v, 0.1)}{D(v, 0.5)} \quad (3)$$

where N is the total number of particles, d_i is the diameter of the particle, n_i is the number of particles having diameter di, $D(v, 0.9)$ is the diameter where 90% of the volume distribution is below this value, $D(v, 0.1)$ is the diameter where 10% of the volume distribution is below this value, and $D(v, 0.5)$ is the volume median diameter.

2.4. Apparent Viscosity of the Emulgel

Rheological measurements were carried out employing a controlled stress rheometer Haake MARS II (Thermo Fisher Scientific, Waltham, MA, USA) with a sand-blasted coaxial cylinder geometry (Z-20, Re/Ri = 1.085, Ri = 1 cm). The apparent viscosity of the emulgel was obtained by flow curves that varied the shear rate from 0.05 to 150 s^{-1}. A total of 300 s were used for the structural recovery of the sample after loading it into the measurement system. Each test was carried out at least two times with fresh samples and at 20 °C ± 0.1 °C.

The results obtained were fitted to the power-law equation:

$$\eta_a = K \cdot \dot{\gamma}^{(n-1)} \quad (4)$$

where η_a is the apparent viscosity (Pa·s), $\dot{\gamma}$ the shear rate (s^{-1}), K is the consistency index (Pa·s^n), and n is the flow index of the power law.

2.5. Physical Stability

A multiple light scattering instrument (Turbiscan Lab Expert, Formulaction, Toulouse, France) was used to determine the physical stability of the emulgels at 25 °C. The equipment has an optical head with an infrared light source (850 nm) and two detectors, one for transmittance and the other for backscattering, the latter being ideal for opaque samples like ours. First, the sample prepared in Section 2.2 was immediately transferred to the instrument-matched tube. Next, the tube was placed in the instrument and a moving head inside the equipment moves every 40 mm from the base of the tube to the final height of the sample. The backscattering intensity was recorded for 30 days to calculate the TSI using Equation (5):

$$TSI = \frac{\sum_h |scan_i(h) - scan_{i-1}(h)|}{H} \quad (5)$$

where $scan_i$ is the mean backscattering intensity of the i-th scan, $scan_{i-1}$ is the mean backscattering intensity of the $(i-1)$-th scan, and H is the scan number in the whole measurement. According to this equation, a lower TSI indicates a more stable system.

2.6. Statistical Analysis

The results were analyzed by the analysis of variance (ANOVA) using Origin 8.0 and the data were expressed as mean ± standard deviation. Tukey's test was used to test whether the means were significantly different ($p < 0.05$).

3. Results and Discussion

3.1. Influence of the Stirring Time and the Rotational Rate

Figure 2A shows the particle size distribution (DSP) for the fresh 50P/50E lemon essential oil emulgel as a function of the stirring time, being constant the rotational rate (400 rpm). As can be observed, a monomodal distribution was obtained with a population peak about 0.12 µm and, in the time interval studied (from 1 to 7 min), the DSP was not influenced by this variable. Additionally, in Figure 2B, the mean diameters (Sauter mean diameter, $D_{3,2}$ and mean volume diameter, $D_{4,3}$) and the span as a function of the stirring

time are shown, corroborating that this variable does not influence them. This state was also corroborated by ANOVA analysis.

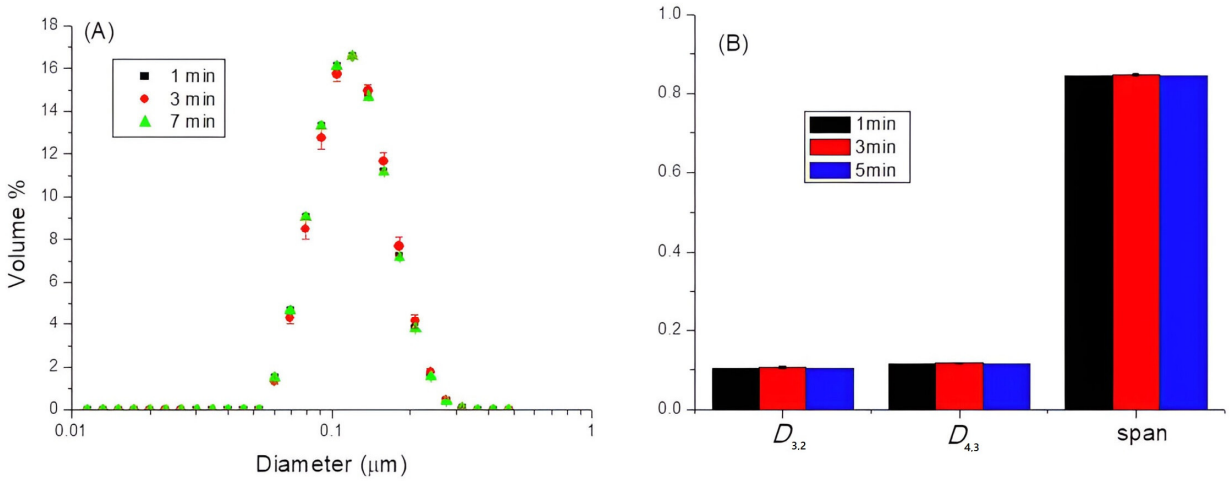

Figure 2. (**A**) Particle size distributions for fresh 50P/50E emulgels at different stirring times. (**B**) Mean diameters and spans as a function of the stirring time. The bars correspond to the standard deviation. The tests were carried out at an ambient temperature.

The physical stability of these emulgels is shown in Figure 3. The stirring time does not influence the stability of the emulgel significantly, although a slightly lower TSI value was found for 3 min. For this reason, this time, 3 min, was selected as the stirring time.

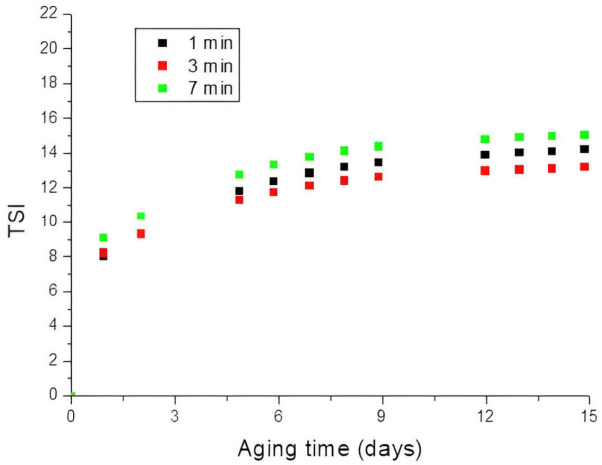

Figure 3. Time evolution of the Turbiscan Stability Index, calculated for the entire height of the measuring cell, with aging time (15 days) for 50P/50E emulgels as a function of the stirring time.

In Figure 4, the DSP (Figure 4A) and the mean diameters and span (Figure 4B) for the fresh 50P/50E lemon essential oil emulgel as a function of rotational rate at the stirring time previously set are shown. All emulgels showed the same particle size distribution and similar to previous samples, they were monomodal with a population peak about 0.11 μm. This fact can also be observed by analyzing the values of mean diameters and span, and in addition, was confirmed by ANOVA analysis. Therefore, it can be concluded that, within the interval of times and rates studied, the mixing protocol used to obtain the emulgels does not influence their final size.

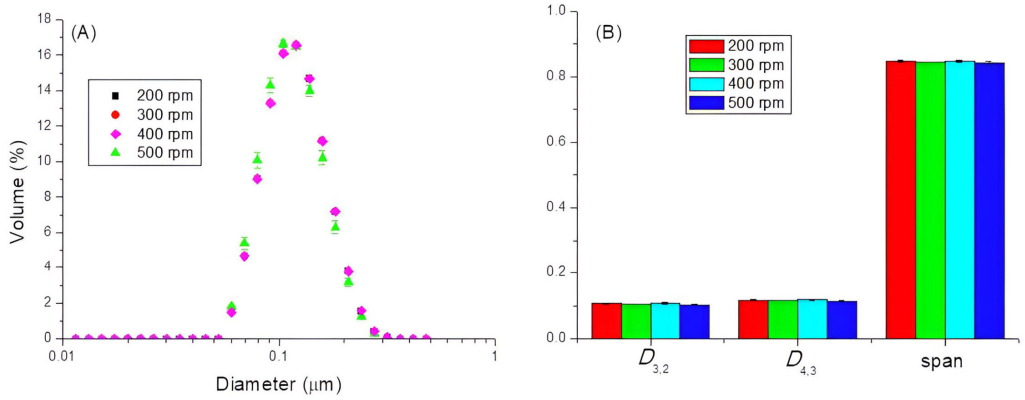

Figure 4. (**A**) Particle size distributions for fresh 50P/50E emulgels at different rotational rates. (**B**) Mean diameters and span as a function of the rotational rates. The bars correspond to standard deviation. The tests were carried out at an ambient temperature.

The evolution of TSI with aging time is shown in Figure 5. All rates exhibited a similar evolution of the TSI and there was no defined trend between the TSI values and the rotational rate. However, 200 rpm showed the lower values of this parameter, namely a higher stability; therefore, this rotational rate was selected for the next stage.

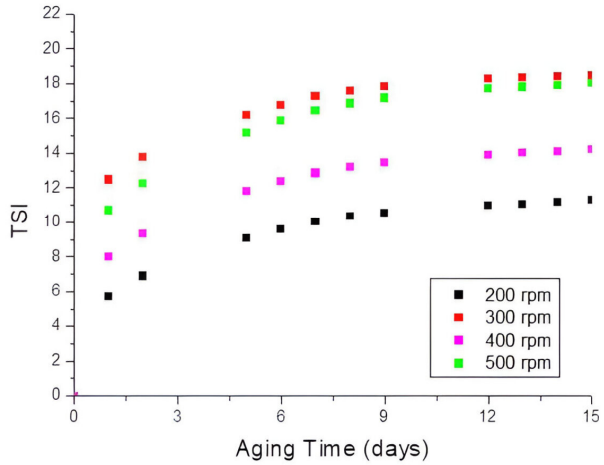

Figure 5. Time evolution of the Turbiscan Stability Index calculated for the entire height of the measuring cell, with aging time (15 days) for 50P/50E emulgels as a function of the rotational rate.

3.2. Influence of the Pectin/Emulsion Mass Ratio

In this section, different pectin gel/emulsion mass ratios were investigated. As occurred with variables previously studied, the particle size distribution was monomodal at the 24 h aging time (Figure 6A). It should be noted that the DSP of the emulsion was the same as that of the emulgel obtained with a higher amount of emulsion. As the pectin content increased to the detriment of that of the emulsion, the intensity of the main peak decreased slightly. No significant differences were found in average particle sizes, as can be seen in Figure 6B and demonstrates the statistical analysis of variance (ANOVA) carried out. It is worth noting that pectin is known not only as a thickener or gelling agent but also as an emulsifier, being the mechanism accepted that pectin is adsorbed on the surface of the droplets through anchor points, resulting in loop and tail adsorption. However, the results previously obtained revealed that the main function of pectin in these samples was not as an emulsifier because its presence does not significantly influence particle size. This result could be expected for several reasons. On the one hand, it might be because similar pectins at the olive oil/water interface are capable of reducing interfacial tension but are not as effective as commercial surfactants [38]. On the other hand, as reported

by Isusi et al. (2020) [45], pectin microgels are less effective as an emulsifier than pectin as a linear polymer. Note that in our research, the pectin gel is really a sheared gel as indicated below. In addition, the emulgel preparation protocol also influences the results. In other works [12,38,42], pectin was added to the continuous phase of the emulsion and once formed, calcium ions were added to proceed to the formation of the emulgel. In those cases, pectin may adsorb at the interface and contribute to the formation and stabilization of the emulsion. However, in this research, the pectin, previously gelled, was added to a nanoemulsion which was also already formed. Under these conditions, in addition to the low interfacial activity as mentioned above, its function as an emulsifier is hindered.

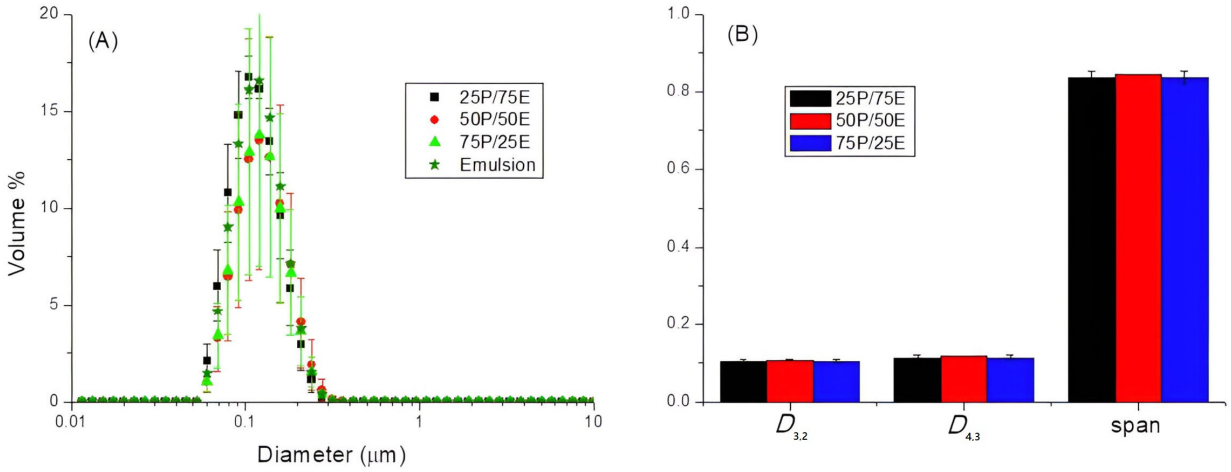

Figure 6. (**A**) Particle size distributions for lemon essential oil emulgels at different pectin gel/emulsion mass ratios. (**B**) Mean diameters and span as a function of the pectin gel/emulsion mass ratio. The bars correspond to standard deviation. The tests were carried out at an ambient temperature.

Note that the mean diameters, as for the previous emulgels and emulsions, were below 150 nm; hence, it is possible to state that they were nanoemulgels.

In Figure 7, the variation of the particle size distribution with the aging time of the assessed emulgels is illustrated. On the one hand, note that these samples are really heterogeneous, as can be deduced from the high value of the standard deviation found. This fact is more relevant when the pectin concentration is higher. To understand this result, it is necessary to remember that the pectin gel used as the structured phase of these emulgels was a sheared gel because its gelation process was interrupted by shear forces by dispersing $CaCl_2$. As a consequence, nanogels and microgels were formed; hence, a sheared gel can be defined as a suspension of gelled particles [46]. This sheared gel is a heterogeneous structure derived from its granular nature at the particle level. As the proportion of pectin gel increased in the formulation of the emulgel, the number of these gelled particles present in the final product also increased, and hence the greater variability observed. The occurrence of these nano and microgels in fluid gels was also stated by other authors [12,45,47]. On the other hand, as can be seen in Figure 7, all emulgels exhibited aggregation of particles with aging and this process was greater with increasing the pectin content. This result may be explained by taking into account not only the possible aggregation of emulsion droplets but also the sheared gel microstructural evolution, both at the intra-particle level as at inter-particle interactions level. It is worth highlighting that the 75P/25E emulgel, which contained the highest proportion of pectin gel, changed its particle size distribution from day 1 to the next day on which the measurement was taken, namely day 6, but afterwards the changes were not so noticeable. This result may be explained by taking into account that this occurs in the formation of a quiescent gel. It is known that the formation of the gel network is not immediate, but requires time, which can vary from hours to days, until those interactions that give rise to the network

stabilize. This process could occur within the particles of a sheared gel [46]. Gabriele et al. (2009) [48] demonstrated that interactions between fluid gel particles not only take place during processing or immediate postprocessing but also that the maturation of the structure can involve a significant time once processing is completed. They also corroborated that fluid gels with shorter processing times, by having less time to form, are more reactive once the processing is finished. This circumstance occurs during the formation of the sheared pectin gel since calcium ions are added for 30 s at 8000 rpm. For this reason, this emulgel (75P/25E) needs a long time to mature its structure.

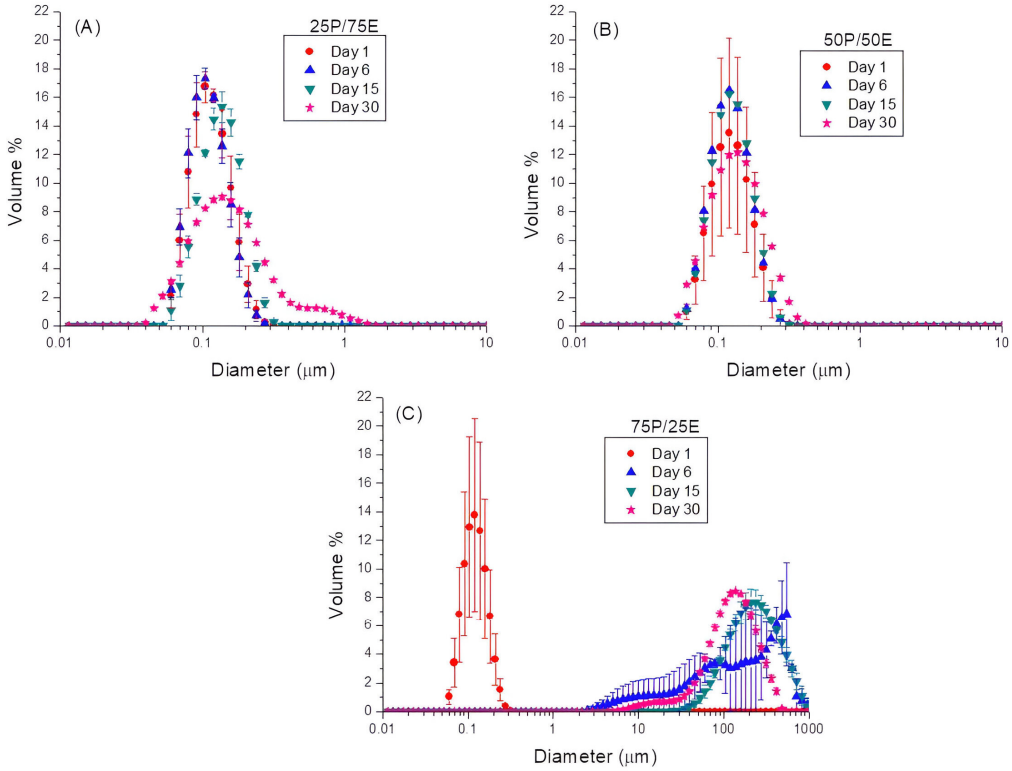

Figure 7. Particle size distributions as a function of aging time for lemon essential oil emulgels at different pectin gel/emulsion mass ratios. (**A**) 25P/75E; (**B**) 50P/50E; (**C**) 75P/25E. The bars correspond to standard deviation. The tests were carried out at an ambient temperature.

The evolution of the flow curves with the aging time of the three emulgels is shown in Figure 8. In these figures, the apparent viscosity is plotted against the shear rate on a double logarithmic scale. Emulgels exhibited a shear thinning behavior which is typical of non-Newtonian fluids. This flow behavior was observed by other authors who also studied emulgels [2,27]. These authors explained the decrease in viscosity with the increase in shear rate by a decrease in contact, and therefore a decrease in interactions, between the polymer chains. In addition, gelled particles may be aligned by shear flow contributing to a decrease in viscosity. On the other hand, all of the emulgels experienced an increase in apparent viscosity with aging time. This result may be due to the existence of the aggregation of droplets and, especially, of gelled particles in the continuous phase of the emulgel. This result is in agreement with laser diffraction results. Note that the viscosity turned out to be higher as the pectin content in the final emulgel increased, pointing out that the rheology of these samples was controlled by the continuous phase and not by the dispersed phase concentration.

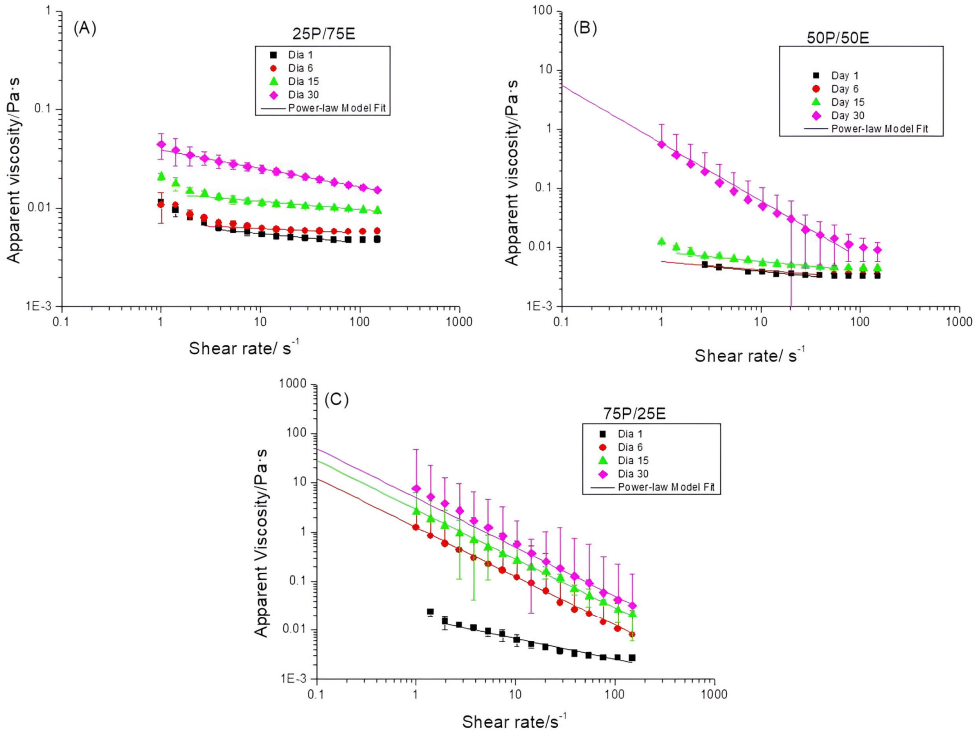

Figure 8. Flow curves for lemon essential oil emulgels as a function of the aging time. (**A**) 25P/75E emulgel, (**B**) 50P/50E emulgel, and (**C**) 75P/25E. The continuous lines exhibit data fitting within the power-law region. The bars correspond to the standard deviation. Temperature: 20 °C.

Table 2 shows the rheological parameters obtained by fitting the experimental data to the power-law model (Equation (4)). All emulgels exhibited n values lower than 1, confirming a shear thinning behavior. Da Silva et al. (2021) [29], with emulgels containing clove essential oil and carboxymethylcellulose as the gelling agent, also found a pseudoplastic behavior that was also fitted to the power-law model. This behavior was explained by the presence of the gelling agent. Overall, a higher pectin content increased the viscosity and shear thinning character of the emulgel (lower n value), with the 25P/75E emulgel being the one with the greatest Newtonian character. This result may be explained by considering that when the pectin content is higher, intermolecular interactions intensify, causing a stronger network. With aging time and regardless of the emulgel considered, there was an increase in the value of the consistency index, k, and a decrease in the flow index, n, indicating a most marked pseudoplastic behavior.

Finally, in order to compare the physical stability in these emulgels, the Turbiscan Stability Index (TSI) was calculated. Figure 9 shows the TSI calculated from the bottom to the top of the tube (from 0 to 30 days), as a function of the mass ratio of the pectin gel/emulsion. It is worth highlighting that when the TSI is calculated as indicated, all the destabilization mechanisms that occur in the sample contribute to its value. As can be observed in Figure 6, the emulgel containing a higher proportion of emulsion, 25P/75E, exhibited both a higher TSI value and a destabilization slope. The 50P/50E emulgel presented a slower destabilization rate for the first two weeks of aging to the point of being considered almost stable. However, from this time, the TSI value and therefore its slope, increased considerably. The TSI values of the 75P/25E emulgel changed during two days after preparation, but then remained practically constant. The multiple light scattering technique brought to light that the emulgel containing a higher proportion of pectin gel exhibited the best physical stability.

Table 2. Mean values and standard deviations of the parameters obtained from the fit of the flow curves of the emulgels studied to the power-law equation. Temperature: 20 °C.

Emulgel	Day	K (Pa·s^n)	n	R2
25P/75E	1	$0.007 \pm 2 \times 10^{-4}$	$0.92 \pm 10 \times 10^{-3}$	0.84
	6	$0.007 \pm 1 \times 10^{-4}$	$0.97 \pm 4 \times 10^{-3}$	0.82
	15	$0.014 \pm 3 \times 10^{-4}$	$0.92 \pm 5 \times 10^{-3}$	0.95
	30	$0.039 \pm 4 \times 10^{-4}$	$0.81 \pm 3 \times 10^{-3}$	1.00
50P/50E	1	$0.006 \pm 2 \times 10^{-4}$	$0.84 \pm 18 \times 10^{-3}$	0.91
	6	$0.006 \pm 3 \times 10^{-4}$	$0,87 \pm 19 \times 10^{-3}$	0.82
	15	$0.008 \pm 3 \times 10^{-4}$	$0.84 \pm 12 \times 10^{-3}$	0.93
	30	$0.579 \pm 324 \times 10^{-4}$	$0.03 \pm 21 \times 10^{-3}$	0.93
75P/25E	1	$0.018 \pm 20 \times 10^{-4}$	$0.580 \pm 2 \times 10^{-4}$	0.91
	6	$1.249 \pm 310 \times 10^{-4}$	$0.002 \pm 2 \times 10^{-4}$	1.00
	15	$2.861 \pm 320 \times 10^{-4}$	$5 \times 10^{-15} \pm 0$	1.00
	30	$4.951 \pm 109 \times 10^{-3}$	$9 \times 10^{-16} \pm 0$	0.95

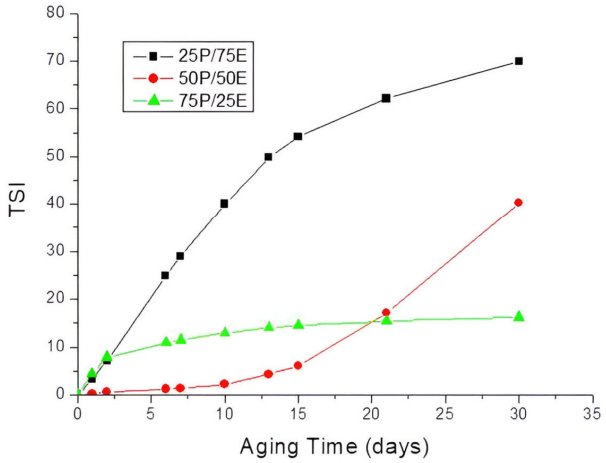

Figure 9. Variation of TSI with aging time as a function of the pectin gel/emulsion mass ratio for emulgels formulated with lemon essential oil.

The results obtained by different techniques are in agreement with each other. Just after preparation, all the emulgels present a similar particle size distribution, hence similar mean diameters. With aging, droplets undergo aggregation, but also, the nanogels and microgels of pectin evolve. As the pectin gel content in the emulgel increases, the inter-particle interaction is higher, the number of the aggregates formed is higher, the friction is higher, and therefore, the viscosity is higher. At the same time, the higher viscosity hinders the movement and destabilization processes. As a result, the most viscous and stable emulgel was 75P/25E, which contained the highest proportion of pectin gel.

4. Conclusions

In this work, oil-in-water nanoemulgels formulated with lemon essential oil and low methoxyl citrus peel pectin have been developed using a preparation method based on the preparation of a nanoemulsion and a pectin gel separately and then mixing them. The stirring time and rotational rate during the mixing step do not influence either the particle size distribution or the values of the mean diameters, pointing out that the mixing process does not affect the droplet properties of the starting nanoemulsion. Note that these processing variables do not significantly influence the global destabilization parameter TSI either. However, from the results obtained, 3 min and 200 rpm were set due to these conditions exhibiting the lowest TSI values. Different pectin gel/nanoemulsion mass ratios were exhibited at 1 day of aging with very similar DTP, according to the fact

that all the emulgels contained the same nanoemulsion. However, with aging time, an aggregation process occurred, especially of the micro or nanoparticles of pectin. These particles were present as the pectin gel was really a sheared pectin gel as a result of its method of preparation. This process was more pronounced with higher pectin content. All samples showed a shear thinning flow behavior and exhibited an increase in viscosity with aging, the greater the higher the initial pectin content. This result was consistent with the aggregation process mentioned above. To conclude, the emulgel containing the higher pectin gel/nanoemulsion ratio presented the longest physical stability. This study brings to light the role played by microgels formed during the preparation of the structured phase in the stability of emulgels formulated with lemon essential oil and low methoxyl pectin. Further tests should be carried out to check if the results obtained here hold for any pectin gel concentration and $CaCl_2$/pectin mass ratio. Furthermore, it is worth exploring different methods of emulgel preparation not only at a lab-scale. Additionally, micrographs obtained by confocal laser and electron microscopy would provide valuable information on the microstructure of these emulgels formed from a matrix of sheared gel.

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