Effect of Permeability on Aqueous Biopolymer Interfaces in Spinning Drop Experiments

Elke Scholten, Leonard M. C. Sagis, and Erik van der Linden*

Food Physics Group, Department of Agrotechnology and Food Sciences, Wageningen University, Bomenweg 2, 6703 HD Wageningen, The Netherlands

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In this paper we show that interfaces in aqueous phase-separated biopolymer mixtures are permeable for all components present in the system. In spinning drop experiments, droplets of the low-density phase decreased up to 90% in volume over a time span of days to weeks, when inserted in a matrix of the high-density phase. We propose an expression for this change of volume in time in terms of diffusion coefficients of the components. From the magnitude of these coefficients, we conclude that the transfer of gelatin from inside the droplet to the outer phase was the rate-determining step in this process. Since the interfaces are permeable to all components, the properties of the system change in time. Therefore, the spinning drop technique is not an accurate method for the measurement of the equilibrium interfacial tension of these aqueous phase-separated systems.

1. Introduction

Interfacial tension plays a role in many interfacial phenomena, and is of importance in a large variety of applications. The interfacial tension is the driving force for phase separation, a process used for the isolation of water-soluble ingredients, such as proteins, nucleic acids, viruses and cells.[1] It also plays a role in the oil tertiary recovery process.[2,3] Since the interfacial tension plays a role in the phase separation process, it is also of importance in the formation of morphologies in aqueous phase-separated mixtures. These aqueous (biopolymer) mixtures are often used to create a large diversity of products and materials with unique properties (e.g., food, pharmaceuticals, and cosmetics). It is therefore of practical interest to gain a better knowledge of the values for the interfacial tension in these mixtures.

There are different techniques that can be used to measure the interfacial tension, which can basically be classified into three groups:

(a) Techniques that use the balance between interfacial forces and gravitational forces, such as the Wilhelmy plate and the pendant drop technique.

(b) Techniques that use the equilibrium shape and size of dispersed droplets, such as the spinning drop method,[4,5] and deformation of droplets in a flow field.[6,7]

(c) Techniques based on the dynamics of dispersed droplets, such as the breaking thread method,[8] and the relaxation behavior of deformed drops after the cessation of an applied flow field.[9]

The interfacial tension of these water/water interfaces are extremely low. Therefore, the measurement of this parameter is not straightforward for these systems and causes many experimental difficulties using classical methods. Since the techniques of class (a) depend on gravitational forces, they cannot be used to measure very low interfacial tensions encountered in aqueous phase-separated systems. For these systems, the interfacial tension can be measured only by techniques of class (b) and (c). These two methods use the same approach; the interfacial forces determined by the interfacial tension, are balanced by some other force, which is obtained by an applied force field.

* To whom correspondence should be addressed. E-mail: erik.vanderlinden@wur.nl.

These techniques are accepted as reliable methods and are therefore widely used to measure interfacial tensions in (aqueous) phase-separated (bio)polymer systems. Although these methods seem to be easy and to give good results, some difficulties and peculiarities were observed in the past. Guido et al. reported measurements[10] of Na-caseinate and Na-alginate mixtures for which they measured the interfacial tension by applying the droplet deformation technique. They found that when one phase was injected into the other phase, the size of the droplets decreased in rest. They attributed this effect to temperature effects and nonequilibrium effects. Van Puyvelde et al.[11] measured the interfacial tension of aqueous gelatin/dextran mixtures. They used light scattering to follow the deformation of the dispersed droplets, which were placed in a shear cell. After subjecting the droplets to a flow field, they observed homogenization of the immersed droplets into the other phase. As a result, they found a shear rate dependent phase diagram of these ternary mixtures. Ding et al.[12] also measured gelatin/dextran mixtures and observed a difference in interfacial tension with different shear rates. Chan et al.[13] observed a similar effect for spinning drop measurements on silicone oil/water and water/air systems. For increasing rotation speeds they found an increase in the interfacial tension for both systems. They investigated several possible causes, such as the effect of capillary width, lack of gyrostatic equilibrium, lagging of the drop diameter behind the rotation speed, pressure effects in the capillary and heat effects of the bearing house, but neither one of these effects seem to be able to explain the observed phenomenon. They concluded that this effect might be attributed to flow patterns in the tube, and that these secondary flows might effect the equilibrium shape of the droplet. Guido et al.[14] studied diffusion effects in a polymer blend, and showed droplet shrinkage from 67 to 51 μm in a few days as a result of the solubility of PIB in PDMS.

Although a lot of these peculiarities have been discussed in the literature for different systems, a good and clear explanation has never been given. In this paper, we want to address the problem of the size and shape change of droplets for aqueous phase-separated biopolymer systems. In these systems, both coexisting phases consist of about 90% of water. Since the water

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does not have any specific preference to stay in either the upper or lower phase, water can diffuse through the interface depending on the forces that act on the system. These interfaces can thus be compared with permeable membranes. In this paper, we want to investigate whether and to what extent this permeability plays a role in the change in size and shape of dispersed droplets.

2. Experimental Section

We performed spinning drop measurements on mixtures with different compositions. The advantage of the spinning drop method is that only one droplet of the low-density phase is inserted in a matrix of the high-density phase, so the droplet is not influenced by interactions of neighboring droplets, as could be the case in techniques based on deformation and relaxation measurements. All effects observed can be attributed to one droplet and the surrounding medium. With this technique it is reasonably easy to measure different properties (such as shape, volume and interfacial tension) of the droplet in a long time frame, so time effects can be tracked easily. To perform these experiments we have used several aqueous fish gelatin/dextran mixtures at different concentrations, and with compositions in the two-phase region. These systems become inhomogeneous upon mixing and phase separate in time. No centrifugation technique was used to speed up the process of phase separation, since a real thermodynamic equilibrium is needed to be able to dismiss possible nonequilibrium effects in the experiments. Centrifugation might slightly change the phase behavior, as in a similar way shear does.[11,15]

2.1. Materials. The high molecular weight fish gelatin was kindly provided by Norland Products Incorporated, Cranbury, US. The molar mass, M_w, of the gelatin is 102 kDa. Fish gelatin is known for its low gelling temperature, and therefore all mixtures remain liquidlike in a large concentration range, which is a prerequisite to be able to use the spinning drop method. The dextran was purchased from Sigma-Aldrich and has a molar mass, M_w, of 511 kDa.

2.2. Methods. 2.2.1. Preparation of the Biopolymer Mixtures. Gelatin and dextran were dissolved simultaneously in a 0.05 M NaI solution. This small amount of salt was used to increase the solubility of the gelatin. Sodiumazide (0.02%) was added as an antimicrobial agent. The mixtures were left overnight to soak the biopolymers, after which they were dissolved easily by heating them at approximately 40 °C for about 30 min and frequent shaking. All mixtures became opaque, and phase separation was allowed to take place to obtain two distinct clear phases. After at least a week (to be sure that equilibrium was obtained) the two phases were removed from each other using a syringe. After the separation, both phases were checked with a microscope in order to confirm that both phases were clear without the presence of small immiscible droplets of the other phase, indicating that both phases were in equilibrium.

2.2.2. Determination of the Concentration of Both Phases. The concentration of the phases was determined by measuring the optical rotation of the phases with a polarimeter (Perkin-Elmer, model 341, Norwalk, CT). The optical rotation was measured at 80 °C since at this temperature, the optical rotation of the mixture was found to be a simple addition of the contribution of each biopolymer. The details about this analysis are described elsewhere.[16]

2.2.3. Determination of Densities of Both Phases. The spinning drop method is a fairly easy technique to measure the interfacial tension. The only disadvantage is that the precise density difference between the two phases is needed in order to calculate the interfacial tension. Since in phase-separated biopolymer mixtures both phases consist mainly of water, the density difference is very small (especially for samples close to the critical point). The viscosities of the mixtures were very high and direct density measurements on the solutions were hampered (no reproducible measurements were obtained using a density meter). Therefore, we calculated the densities of the phases using the densities of the pure components rather than measuring them. The densities of the dry components were determined from calibration curves, for which the densities of solutions with different concentrations were measured with an Anton Paar DMA 5000 density meter.

2.2.4. Spinning Drop Measurements. We have used the SVT20, a spinning drop tensiometer from Dataphysics, Germany. This apparatus consists of a capillary that is mounted on a tilting table. The capillary is surrounded by an oil bath, which regulates the temperature of the capillary. Because the temperature of the capillary can be controlled well, heating effects did not play a role in the measurements that were performed. The capillary was filled with the high-density phase of the phase-separated mixtures with the use of a syringe. As the viscosity of these solutions was substantial, the solutions were rotated overnight at high rotational speed to collect possible air bubbles, which could disturb the shape of the droplets and might induce flow effects. Before a droplet of the low-density phase was inserted with a microsyringe, the high-density solution was checked for air bubbles. The position of the table was adjusted in order to prevent the droplet from wandering to either side of the tube. The apparatus is equipped with a light source, for which the intensity can be varied. A camera is placed in front of the capillary and has a zoom function that allows measuring the size of the droplets at different magnifications. The volume of the droplets is determined with the software from the number of calibrated pixels of the droplet image. The camera is connected to a computer with a frame grabber and software to calculate the interfacial tension with the appropriate method (Vonnegut[4] or Cayias−Schechter−Wade[17]). Using a template that can be placed around the droplet, the shape and size of the droplet was determined, from which the volume and the interfacial tension were calculated.

3. Results and Discussion

3.1. Phase Diagram and Coexisting Phases. For the spinning drop experiments, different samples were prepared that differ in concentration. The samples were allowed to phase separate after which the optical rotation of both phases was measured at two different wavelengths. From these measurements, the concentration of both phases were determined. Details can be found in previous work.[16] From the calculated concentrations, the phase diagram was determined. Figure 1 shows this phase diagram, in which the open circles represent the overall concentration of the mixtures. The overall concentrations do not always fall on the tie-line, which is most significant for sample 5. This deviation is a result of the method of determination with the use of polarimetry, for which we measure at two different wavelengths. This causes small errors in the determination of the compositions. As can be seen, the gelatin concentration was kept constant, while the concentration of dextran was varied.

3.2. Densities of the Phases. The densities of the separate components were determined to be 1.367 g/mL for the dry fish

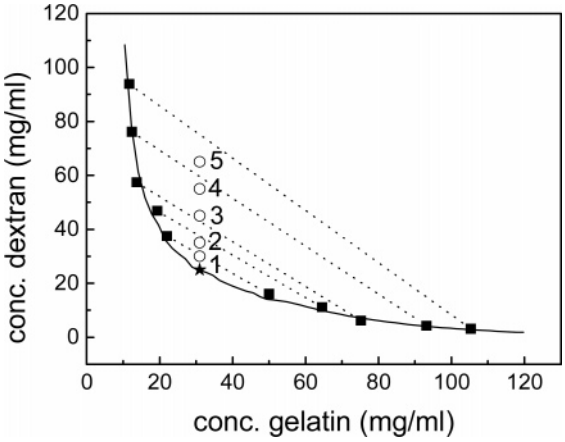

Figure 1. Phase diagram of the gelatin/dextran mixtures. The star denotes the critical point. The open circles refer to the overall concentrations of the mixtures that have been prepared. The squares refer to the compositions of the upper and the lower phase after the mixtures were phase-separated. The solid line is the binodal, which connects the compositions of the phases after phase separation.

gelatin, 1.603 g/mL for the dry dextran, and 1.005 g/mL for the 0.05 M NaI solution. Using these densities, the densities of both phases of all samples were calculated. In addition, the difference in density between the two phases, $\Delta\rho$, was calculated, and was used by the software of the SVT20 to calculate the interfacial tension.

3.3. Spinning Drop Experiments. To compare the results of the spinning drop measurements, we performed all measurements at the same speed, which was 1000 rpm. Higher rotational speeds were not possible, since the elongation of the droplets would be of such extent that the droplets disappear partially out of sight. As a consequence, the volume of the droplets cannot be measured. A lower rotational speed is undesirable, since a certain minimum deformation of the droplets is needed to be able to measure the interfacial tension accurately. Therefore, the range of rotational speeds that could be used was rather narrow.

3.4. Droplet Volume Measurements. To be able to compare the effects between different samples, we tried to control the volume of the droplet that was inserted, which was approximately 2.5 μL. The samples were inserted into the capillary and the rotational speed was set at 1000 rpm. As soon as the droplet was located in the middle of the capillary (which took several seconds only) and did not appear to change shape, the volume of the droplet was determined. This value was taken as the initial volume of the droplet. The time when the droplet reached the middle of the capillary was taken as time zero. Figure 2 shows an example of droplets at different times during the experiment. The left picture shows the volume at the beginning of the experiment ($t = 0$) and the right side shows the volume after a few days.

These results show that the droplets become smaller in time. In our system, we use biopolymers, which are known to be polydisperse by nature. Shi et al.[18] showed that for polydisperse systems the interfacial tension changes in time due to the migration of the smaller polymers to the interface. During their experiments they add a low or high molecular weight polymer to their sample and measure a change in interfacial tension of approximately 15%. Their experiments are clearly performed under nonequilibrium conditions. Since our biopolymers are polydisperse, there might also be some migration of smaller polymers to the interface of the droplet in our experiments. However, we have taken great care that our system is in

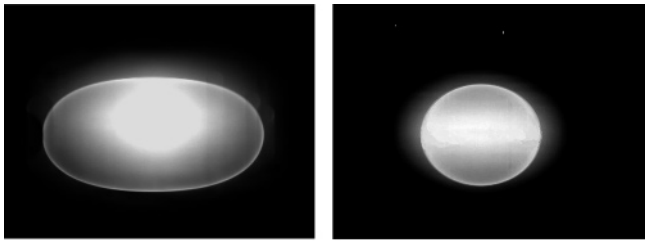

Figure 2. Shape and size evolution of a droplet during the spinning drop experiments. The magnification of the frames is the same. The pictures were taken from sample 3, for which the overall concentration is denoted by the open circle in Figure 1 and the composition of the phases by the squares.

equilibrium prior to the experiments, and we therefore expect this effect to be much smaller in our system. It cannot explain the large change in volume of our droplets. The experiments by Shi et al.[18] show that the redistribution takes place on a time scale of seconds, which is much smaller than the times scales of days or even weeks in our experiments. We therefore conclude that the redistribution of polydisperse polymers cannot explain our results.

The change in volume of the droplets indicates that the interface is permeable to certain components. But to which components is the interface permeable? There are three components present in this system: the biopolymers gelatin and dextran and the solvent water. Since water molecules are much smaller than the biopolymers and smaller than the mesh size of the phases, water should be able to diffuse through the interface with ease. The biopolymers are normally present in the semidilute regime in either one of the phases and due to their entanglements, the biopolymers will exhibit hindered diffusion. So, one might expect that only the water would diffuse through the interface. However, if water is the only component that diffuses through the interface, the concentrations of the biopolymers within the droplets would increase to such extent that the density within the droplet would exceed the density of the outer phase. In that case, the droplet would not have stayed in the middle of the capillary, since the low-density phase is pushed to the middle by the centrifugal forces. This indicates that the density of the droplet is always lower than the surrounding medium, for which the density remains effectively unchanged since its volume is approximately 3 orders of magnitude larger. This indicates that also biopolymers should be expelled from the droplet in order to keep the droplet density lower than the density of the surrounding medium.

We can explain this phenomenon by a shift in the binodal under the influence of the applied force field. Antonov et al.[15] showed that for a gelatin/dextran mixture subjected to high shear rates, the phase behavior is different compared to the phase behavior at rest. They reported the phase diagram, with accompanying binodal, and showed that under shear the binodal shifts as depicted in Figure 3. This means than when dispersed droplet of these mixtures are subjected to a force field, the binodal shifts and therefore the equilibrium composition of the coexisting phases belonging to the overall composition will change. Although the deformation in our experiments is not a simple shear flow, deformation of the droplets will have a similar effect on the phase behavior. When a phase-separated system is in rest, the two phases have the same chemical potential, which is a requirement for the system to be in equilibrium. However, when a force field is applied, such as a centrifugal force field in our case, equilibrium of the system is obtained when for both phases the sum of the chemical potential and the

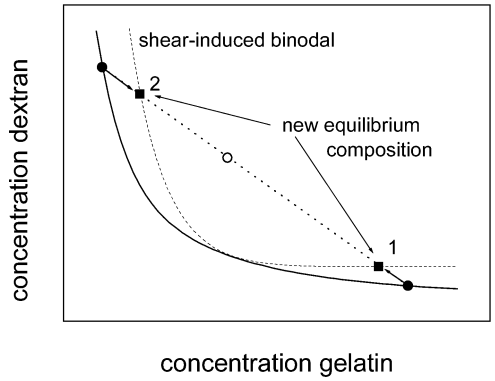

Figure 3. Schematic picture of the composition of the droplet in the capillary during the spinning drop experiment based on the phase diagram by van Antonov et al.[15] The solid line refers to the binodal in rest. The dashed line refers to the shear-induced binodal. The new equilibrium compositions of any overall composition (open circle) on the tie-line (dotted line) are denoted by the squares. The old equilibrium compositions in rest are denoted by the circles. The droplet phase is referred to as number 1 and the surrounding medium as number 2.

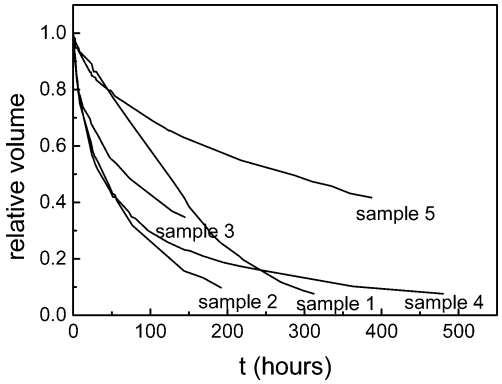

Figure 4. Volume change of all samples during time.

force potential is equal. Since both the droplet and the outer phase experience a different force field (because of the density difference), the total potential will be different, and a new equilibrium will be obtained when a force field is applied. As a result, we will obtain a new deformation-induced phase diagram similar as the shear-induced phase diagram given in Figure 3. As can be seen in Figure 3, the equilibrium composition of the upper phase (number 1) has shifted toward a lower concentration of gelatin, while the equilibrium composition of the lower phase (number 2) has shifted to a higher concentration of gelatin. Therefore, the droplet will expel gelatin in order to lower its concentration, while the outer phase will take up gelatin from the droplet in order to increase the gelatin concentration. However, the concentration of the outer phase remains effectively unchanged since the volume of the outer phase is about 3 orders of magnitude higher than the droplet phase. Therefore, the outer phase continues to take up gelatin from the droplet, which results in a large decrease of its concentration in the droplet. Consequently, the droplet will start to expel water in order to keep the gelatin concentration at the desired value for the composition of the gelatin-rich phase (number 1). Dextran will either enter or leave the droplet depending on its overall concentration. Since the composition of the outer phase will never change sufficiently, the system will never reach a stage with two equilibrium coexisting phases, so the system keeps transferring both biopolymers and water until eventually the droplet will disappear.

Figure 4 shows the volume of these droplets in time, normalized by the initial size of the droplet. From the explanation described above, we know the droplets should disappear. However, in our experiments we do not observe the disappearance of the droplets, since at a certain size its position starts to deviate from the middle of the capillary. At this point, the centrifugal force ($\propto \Delta\rho\omega^2 r$, where $\Delta\rho$ is the density difference between the two phases, ω is the rotation speed and r is the distance from the middle of the capillary) equals the gravitational force ($\propto \Delta\rho g$, where g is the gravitational constant). When the droplets become even smaller, the centrifugal force is not sufficient to exceed the gravitational force and the droplet is pulled up by gravity.

We see from these results that samples 1, 2, and 4 reduce in size by almost 90%. At that size, the droplets started to move from the middle of the capillary. For sample 3 and 5 the measurement was stopped at 60% decrease. During the experiments, we noticed that when the droplet is elongated, the short axis of the droplet does not change significantly; only the long axis of the droplets. When the droplets become smaller and approach a more spherical shape, the width of the droplets starts to reduce as well. This might be explained by the fact that due to the larger curvature in the caps of the elongated droplets, the pressure in the caps is larger than in the middle of the droplet. Therefore, the diffusion of components might be more pronounced in the caps.

The rate at which the components diffuse through the interface depends on several parameters that can enhance or hinder the diffusion, such as the viscosity, the density difference, the size of the biopolymers, and the mesh size of the entangled polymer solutions. Because of the distribution of the components, dextran is concentrated in the lower phase and gelatin is concentrated in the upper phase. Therefore, the dextran concentration inside the droplets and the gelatin concentration outside the droplet are in the dilute regime. In this regime, the biopolymers are present as random coils, and the characteristic length scale is the radius of gyration. The dextran concentration outside the droplet and the gelatin concentration inside the droplet are in the semidilute regime. So inside the droplet, there is an entangled network with a certain mesh size of gelatin, and outside the droplet, there is an entangled network with a mesh size of dextran. The mesh size of the network and the size of the biopolymers diffusing through the network will influence the diffusion rate.

Comparing for example sample 1 and 5, we see that the gelatin concentration inside the droplet differs with a factor of 2. Sample 1 has a concentration of about 50 mg/mL and sample 5 has a concentration of more than 100 mg/mL. Since the concentration is related to the mesh size, we can say that the mesh size of the network of sample 5 is much smaller than in sample 1. Since the network of sample 5 is much more compact, the diffusion of the biopolymers will be hindered more. In a similar way, we also see that the dextran concentration for sample 5 in the surrounding medium is higher than for sample 1. Thus, besides having a much denser gelatin network in the droplet, it also has a network of dextran with a smaller mesh size in the surrounding medium. The denser the network, the more hindered is the diffusion of the components.

Figure 5 shows the viscosity and the density difference between the two phases at the beginning of the experiment. The density difference can be viewed upon as a driving force for diffusion, since this is related to the length of the tie-lines and thus also to the difference between the composition of the original phases and the new equilibrium composition on the shear-sensitive binodal. The viscosity of the solution slows down the diffusion of the biopolymers.

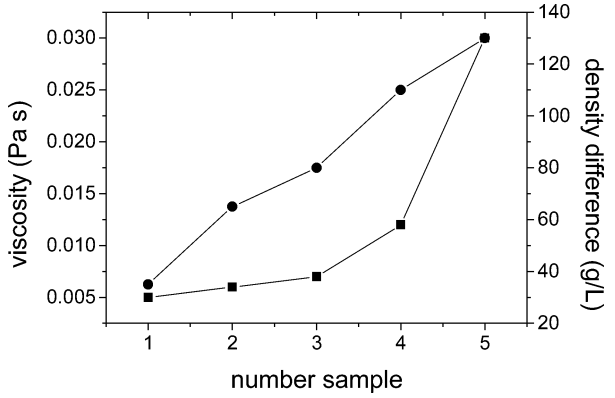

Figure 5. Average viscosity (■) of the system and the difference in density (●) between the two phases at the beginning of the experiments.

All of these variables determine the rate of the diffusion for the components. From Figure 4, we can see that there is no straightforward relation between the change of volume of the droplet and any of the variables, which indicates that apparently all variables are important in the diffusion process. For example, for sample 1, the density difference is very small, so there is a small driving force for the diffusion. On the other hand, the viscosity is small and the mesh size of the network is large, so this will increase the rate of the diffusion process. Looking at the properties of sample 5, we see that the density difference is much higher so there is a larger driving force for diffusion. However, the viscosity is also much higher, which hinders the diffusion. Also, the mesh size of the gelatin network inside the droplet and the dextran network outside the droplet is much smaller, which also hinders the diffusion. The balance between the different variables in the system such as mesh size and concentration difference (which also determines the deviation between the new and the old binodal) eventually determines the total rate of the diffusion process. Since the variables will also change during the diffusion process, there is not a straightforward relation between the involved variables.

The diffusion process is considered to be Fickian and we assume the total diffusion to be a linear addition of contribution from the diffusion of both the biopolymers and the water. The differential equation for the total change in volume in time can be described as[19]

$$\frac{\mathrm{d}V_{\mathrm{total}}}{\mathrm{d}t} = f(V_\infty - V_t) \tag{1}$$

where f is the rate constant for the change in volume, V_∞ is the volume at the end of the process, and V_t is the volume of the droplet at time t. Integration of this equation leads to

$$V_{\mathrm{total},t} = V_\infty + A \cdot \exp(-ft) \tag{2}$$

where A is a constant. Since we assume that the diffusion process is a combination of the diffusion of separate components we describe the total change in volume as

$$V_{\mathrm{total},t} = V_{\mathrm{total},\infty} + \sum_n A_n \cdot \exp(-f_n t) \tag{3}$$

in which n refers to the amount of exponential terms needed to fit the data. The rate constant f_n, is related to a diffusion coefficient, D_n, and the diameter of the droplet, d, as[19]

$$f_n = \frac{4\pi^2 D_n}{d^2} \tag{4}$$

Table 1 shows the results of the fits of eq 3 through the data points from Figure 4. This table gives the number of terms needed to fit the experimental data, and the values for the diffusion coefficients. For the value of the diameter of the droplet, we have taken the initial diameter at the beginning of the experiment.

To determine which process is the rate-determining step that eventually determines the total time for the reduction of the droplets volume, we compare these values to the self-diffusion coefficients of the components. The self-diffusion coefficient of the components can be estimated from the following relation

$$D_{\mathrm{self}} = \frac{k_{\mathrm{b}}T}{6\pi\eta R_h} \tag{5}$$

in which k_{b} is the Boltzmann constant, T is the temperature, η is the viscosity and R_{h} is the hydrodynamic radius of the component. Taking the hydrodynamic radius as 8.4 nm (as determined by viscosity measurements) we find self-diffusion coefficients for gelatin ranging from 2.5×10^{-12} m^2/s for sample 1 to 8.4×10^{-13} m^2/s for sample 5. The values for dextran are the same order of magnitude, since the radius of dextran is comparable to that of gelatin. The self-diffusion coefficient for water is equal to 2.3×10^{-9} m^2/s, 3 orders of magnitude larger than the diffusion coefficient of gelatin. We see that the diffusion coefficients deduced form the fits are comparable to the self-diffusion coefficient of the biopolymers. From this, we can conclude that the rate of the diffusion process is not dominated by the diffusion of water, which can diffuse most easily through the samples due to its small size. As described before, the droplet will try to decrease its concentration in gelatin and to increase its concentration in dextran. Since the droplets decrease in volume, the diffusion of gelatin to the outer phase appears to be more pronounced than the diffusion of the dextran. Therefore, we assume that the diffusion of the gelatin is the rate-determining step. The rate of the other diffusion processes is constrained by the diffusion of the gelatin.

3.5. Interfacial Tension Measurements. Because of the permeability of the interface in these aqueous ternary mixtures, the properties of the interface change during time as all components can transfer through the interface. The change in composition of the droplet phase depends on the shift in the binodal and the rate of the diffusion of the different components. As a result, the density of the droplet will change and subsequently a difference in density between the two phases will change as well. Since the exact density difference is needed in order to calculate the interfacial tension, an error will be present in these calculations. Besides a change in the bulk properties of the samples, the properties of the interface also change. As the composition of the phases changes, the concentration profile in the interfacial region of both gelatin and dextran will differ from its original profile. This results in a change of the thickness of the interfacial region, ξ. This interfacial region is related to the interfacial tension as $\gamma \sim 1/\xi^2$,[20] so a change in composition will result in a change in interfacial thickness and subsequently a change in interfacial tension. So, on one hand, there is an error in the calculations of the interfacial tension since the density of the droplet changes, and on the other hand, the interfacial tension changes due to the change in concentration profile. Thus, during spinning drop experiments, the interfacial tension is not constant but will change in time, and no

Table 1. Diffusion Coefficients for the Three Components

sample	*n*	D_1 (m^2/s)	D_2(m^2/s)	D_3(m^2/s)
1	1	6.7×10^{-14}		
2	2	3.2×10^{-12}	2.2×10^{-13}	
3	2	3.9×10^{-12}	2.2×10^{-13}	
4	3	2.6×10^{-12}	3.9×10^{-13}	7.2×10^{-14}
5	2	1.3×10^{-12}	1.6×10^{-14}	

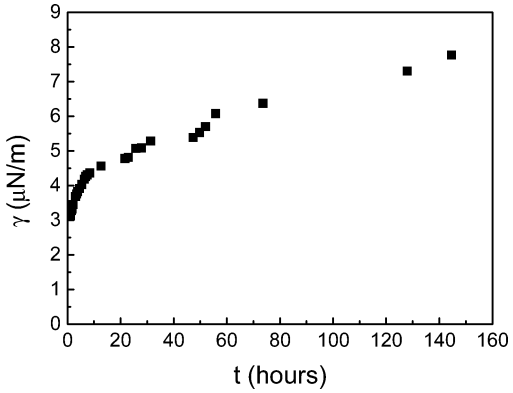

Figure 6. Interfacial tension vs time for sample 3 (31/45 mg/mL gel/dex).

equilibrium interfacial tension can be determined with this method. Figure 6 shows an example of the change in interfacial tension in time. (The measurements for the other samples are not shown, but give similar results.) The interfacial tensions were calculated using the initial value of the density difference between the two phases.

Taking into account the permeability of the interfaces in phase-separated biopolymer mixtures, we can conclude that measuring the interfacial tension at equilibrium for these systems is not possible with the spinning drop technique. At the moment a force is applied on the droplet, the binodal of the system will change, and therefore the coexisting phases in the spinning drop tensiometer are not in equilibrium. Because of this nonequilibrium state, all components in the sample will start diffusing in order to reach a new equilibrium state, which will never be reached. This causes a change in composition and therefore a change in properties of the bulk phase as well as the interface. To what extent the properties will be different than their equilibrium values in rest partly depends on the shift in the binodal for the shear-induced phase diagram. The larger the shift in the binodal is, the larger is the deviation from the properties in the bulk and the interface compared to their properties in rest, and the larger is the error in the interfacial tension. For phase-separated biopolymer mixtures, one should use another method in order to measure the exact value for the interfacial tension. One can deduce the interfacial tension from deformation experiments after cessation of a flow field, for which the interfacial permeability is taken into account in the description of the relaxation process.[21] The spinning drop method is not accurate enough to determine the exact value for the interfacial tension, but can only be used to obtain an estimate of the magnitude of the interfacial tension.

4. Conclusion

We have performed spinning drop experiments to investigate the possible effect of interfacial permeability on the value for the interfacial tension in phase-separated aqueous biopolymer mixtures. These systems consist of two coexisting phases, both with 90% of water. Low-density phase droplets were inserted in a high-density matrix, and the droplets decreased in size by about 90% over a time span of several days when the droplets were rotated around their horizontal axis. The results indicate that the interfaces of these droplets are permeable to all components in the system: water and the biopolymers gelatin and dextran. The rate of volume change depends on several parameters of the system, such as the viscosity, the density difference, and the length scales in the system. We suggest a relation for the change in volume in time, which is related to the separate contributions of the components. From this relation, we obtain different diffusion coefficients, which range from approximately 1.6×10^{-14} m^2/s to 3.9×10^{-12} m^2/s. These values are comparable to the self-diffusion coefficient of gelatin and dextran, from which we conclude that the diffusion of gelatin is the rate-determining step in the process. Because of the diffusion of the components, the composition of the phases changes in time and as a result the properties of the bulk phases and the interface change. This means that for these aqueous phase-separated biopolymer systems, the spinning drop method is not accurate enough to measure the equilibrium interfacial tension.

References and Notes

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