

Rheological Properties of Citrus Pectin Dispersions and Its Blends with Polyquaternium-7 and Colloidal Particles

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ABSTRACT: This article reports on the rheological behavior of a 5% (w/w) citrus pectin (CP) dispersion, its blends with polyquaternium-7 (PQ) and also with 200 nm nanoparticles (NP) and 1.8 μm microparticles (MP) of CP. The viscous (G'') and elastic (G') moduli of the CP dispersion were similar, whereas G' was higher than G'' for PQ. The 1:20 (w/w) blend of CP and PQ enhanced the viscoelastic profile of CP and also decreased its cross-over frequency. NP and MP were equally effective in enhancing the viscoelastic properties of CP. The best viscoelastic behavior was obtained with 1:20:0.4 (w/w) CP:PQ:(NP or MP) composition. The association of CP with PQ and/or MP or NP tended to change the behavior of CP and PQ dispersions from pseudoplastic to Newtonian. Cox-Merz superposition was observed for CP:PQ and CP:(MP or NP). These findings contribute for modulation of the rheological properties of CP dispersions for specific applications. © 2014 Wiley Periodicals, Inc. *J. Appl. Polym. Sci.* **2014**, *131*, 40583.

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INTRODUCTION

Pectin is an anionic heteropolysaccharide that is formed by residues of D-galacturonic acid linked by α -[1,4] glycosidic linkages in the backbone. The side chains are linked by α -[1,2] linkages with neutral sugars, such as rhamnose.¹ The number of carboxyl groups methyl-esterified in the backbone determines the degree of esterification (DE) of pectins; a DE over 50% characterizes highly esterified pectins and values below 50% describe low esterified pectins. The DE can be modified by chemical or enzymatic reactions.² For pectins, the biopolymer conformation plays an important role in the rheological behavior and is affected by intramolecular and intermolecular interactions among the polysaccharide chains. The DE, pH, ionic strength, temperature, and cosolutes are the main factors used to modulate this behavior into the desired rheological properties.³ Pectins are among the most versatile commercially available stabilizers. The gelling, thickening, and stabilizing attributes make pectin an essential additive in the production of many food products. In the pharmaceutical industry, pectin is used to reduce blood cholesterol levels and treat gastrointestinal disorders. Other applications of pectin include use in edible films, as a paper substitute, in foams, and plasticizers, for which viscoelastic properties are important technologically.

Polyquaterniums are polycationic polymers that are widely used in cosmetic products, particularly products for hair care, such

as conditioners, shampoo, hair mousse, hair spray, and hair dye. Some polyquaterniums have antimicrobial properties and are used in contact lens solutions. Different polyquaterniums are distinguished by a numerical value, which is not associated with their chemical structure but with the order they were registered. Polyquaternium-7 (PQ), which was used in this work, is a copolymer of acrylamide and diallyldimethylammonium chloride. Figure 1 shows the molecular structures of an esterified pectin and PQ.

Complexes of polyelectrolytes exhibit a broad spectrum of properties that are governed by the characteristics of the individual components (e.g., properties and positions of ionic sites, charge density, and rigidity of macromolecular chains) and the chemical environment (e.g., solvent, ionic strength, pH, and temperature).^{4,5} In addition to foods, these properties have generated various other applications, mainly in the cosmetic and pharmaceutical fields, such as for the development of rheologically smart fluids as well as for carriers for the release of pharmacologically active compounds.^{6,7} Polyelectrolyte complexes have considerable potential for many practical applications, such as thickener additives, to form membranes with selective retention ability or as a model to study complex living systems in which biomacromolecules are ionically associated.^{4,5}

The functional properties of biopolymers depend on the presence of ordered structures capable of stably associating into

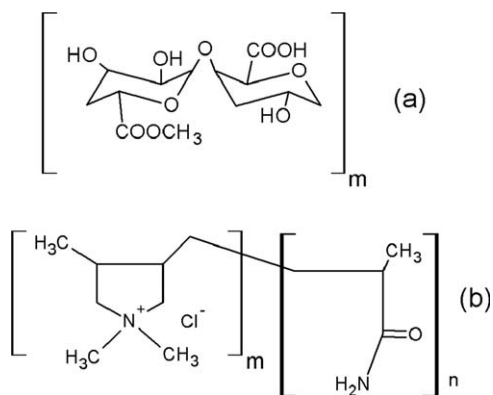


Figure 1. Molecular structures of an esterified pectin (a) and polyquaternium-7 (b).

long-range networks or packed aggregates, which determine their rheological behavior.⁸ These interactions can be monitored by oscillatory measurements of solid-like and liquid-like responses, which characterize the storage (or viscoelastic) and the loss (or viscous) moduli, G' and G'' , respectively, in addition to the dynamic (η^*) and steady-shear viscosity (η).

Viscoelastic behavior has previously been reported for pectin solutions and is influenced by the temperature and cosolutes. Generally, a decrease in the viscosity is observed when the temperature is increased due to pectin degradation.^{9–12} Cosolutes, usually sucrose, may be used to promote an increase in the viscosity of a pectin solution. The polysaccharides entangled with pectin chains strengthen the gel network, increasing the viscosity of the system.¹³ Dilute pectin solutions exhibit Newtonian behavior with minimal rearrangement of the polymer chains. The transition to pseudoplastic (shear-thinning behavior) is observed under high shear rates when the polymer chains do not undergo considerable rearrangement. Pectin branching plays an important role in the viscoelastic behavior; at a zero-shear rate, the solution shows a higher shear rate dependence on viscosity and a higher storage modulus (G'). Therefore, associating compounds that bind with pectin forming networks intensifies the viscoelastic character.^{13,14}

Although the rheological behavior of polyquaterniums has been studied, the combination of polyquaterniums with natural polymers is scarce and specific. Previous studies with acacia gum have shown that the polymer enhanced the viscoelastic properties by forming polyelectrolyte complexes.¹⁵ To the best of our knowledge, there are no studies reported in the literature on the rheological properties of the polyelectrolyte complex of citrus pectin (CP) and PQ are on the inclusion of colloidal particles to the dispersions of those complexes.

This study aims to show the rheological behavior of CP dispersions its blends with PQ and the inclusion of nanoparticles (NP) or microparticles (MP) of CP. Oscillatory measurements were used for the characterization of the rheological behavior of the CP systems. The rheological properties are important because reflect the microstructure of the system and drive to its functionality. Initially, the isolated dispersions of CP (5% w/w) and PQ (10% w/w), as well as the blend CP:PQ, were character-

ized. Afterward, the effects of the inclusion of NP or MP of CP to fluid CP, PQ or to CP–PQ were also characterized and compared. The results could provide a deeper insight into the potential of CP, its polyelectrolyte complexes, and the association of NP and MP to CP dispersions or their complexes for specific applications.

MATERIALS AND METHODS

Materials

Pectin extracted from citrus peel fruits (DE = 56%, MW = 54,229 g/mol) was purchased from CPKelco (Limeira, SP, Brazil). PQ (Mirapol 550 P[®]) and quaternized guar gum (QGG, Jaguar C13[®]) were obtained from Rhodia (Campinas, SP, Brazil). All other reagents were purchased from Merck unless otherwise specified.

Preparation of Pectin Particles

Initially, CP and QGG were separately dispersed in ultrapure water under mechanical stirring for 6 h. The pH of the dispersions was adjusted to 7.0 with 0.5 M NaOH to guarantee that all carboxyl groups of the pectin were ionized and available for ionic interactions. Pectin was used in two concentrations: 10.0 g/L and 20.0 g/L. The particles were produced through polyelectrolyte complexation by ionic crosslinking, for which QGG was gradually added to the solutions of pectin at 15 min intervals under mechanical stirring (Rushton turbine at 300 rpm). QGG was 1.0% (w/w) in relation to pectin.

Physicochemical Characterization of Pectin Particles

The pectin particles were characterized by their hydrodynamic mean diameter, polydispersity and zeta potential, which were measured using a Nano Series ZS Zetasizer (Malvern Instruments, UK) at a fixed angle of 173° and a temperature of 25°C. The measurements were performed in mili-Q water.

Rheological Characterization

The rheological measurements were performed using a Rheo-Stress 1 Haake Rheometer (Haake, Germany). Table I shows the

Table I. Citrus Pectin Systems Used for Rheological Characterization

System ^a	Composition
CP	Fluid pectin 5% w/w
PQ	Fluid polyquaternium-7 (10% w/w)
CP:PQ	1:20 (w/w) fluid pectin:polyquaternium complexes
CP:NP	5:1 (w/w) fluid pectin:nanoparticles
CP:MP	5:2 (w/w) fluid pectin:microparticles
PQ:MP	50:1 (w/w) fluid polyquaternium-7:pectin microparticles
CP:PQ:NP	1:20:0.4 (w/w) fluid pectin:polyquaternium-7: pectin nanoparticles
CP: PQ:MP (A)	1:20:0.4 (w/w) fluid pectin:polyquaternium-7: pectin microparticles
CP:PQ:MP (B)	1:50:3.6 (w/w) fluid pectin:polyquaternium-7: pectin microparticles

^a CP, citrus pectin; PQ, polyquaternium-7; MP, pectin microparticles; and NP, pectin nanoparticles.

Table II. Cumulative Mean Hydrodynamic Diameter (Z-average), Polydispersity (Pi), and Zeta Potential (Zp) of the NP and MP of Citrus Pectin

Particle	Pectin Concentration (g/L)	Z-average (nm)	PI	ZP (mV)
Nanoparticles (NP)	10.0	216.5 (± 69.43)	0.365 (± 0.05)	-59.0 (± 0.95)
Microparticles (MP)	20.0	1833 (± 90.74)	0.203 (± 0.02)	-56.7 (± 2.05)

analyzed pectin systems. The measurements were performed in the steady and oscillatory regimes at $25 \pm 0.1^\circ\text{C}$ using a parallel plate geometry of 20 mm. The oscillatory measurements were performed in the linear region at a stress of 1.188 Pa and in the frequency range of 0.1–100 Hz. The steady shear measurements were obtained at shear rates of 0.1–50 s^{-1} . The CP systems used for rheological characterization are shown in Table II.

RESULTS AND DISCUSSION

Physicochemical Properties of the Particles

The CP particles were characterized by their mean hydrodynamic diameter, polydispersity, and zeta potential. Table II shows the results for NP and MP. Although the polydispersity of NP was approximately twice that of MP, both were less than 0.5. The produced particles were electrostatically stable (high absolute zeta potential value) with a negative zeta potential for interaction with the medium.

Rheological Characterization

The viscous and viscoelastic responses of the systems, which were quantified by dynamic oscillatory measurements, represent the storage (G') and loss (G'') moduli as functions of the frequency. The moduli are defined in terms of the amplitude ratio and phase shift relative to the strain, as defined by eqs. (1) and (2).¹⁶

$$G' = (\sigma_o/\gamma_o) \cos(\delta) \quad (1)$$

$$G'' = (\sigma_o/\gamma_o) \sin(\delta) \quad (2)$$

where (σ_o/γ_o) is the amplitude ratio, δ is the phase shift, σ_o is the amplitude of the shear stress, and γ_o is the amplitude of the strain equal to L/h . When the motion of the upper (oscillating) plate is $L \sin(\omega t)$, ω is the frequency expressed in rad s^{-1} , which is equivalent to $\omega/(2\pi)$ Hz, and h is the distance between the plates.

In this work, both the G' and G'' moduli increased with frequency. Because the storage modulus usually represents the elastic character and the loss modulus describes the viscous behavior, this result suggests that enhancement in the structural entanglement increases the G' modulus, whereas structural breakdown increases the G'' modulus.

Figure 2 shows the dynamic characterization of the systems as controls: CP, PQ, and CP:PQ. The curves are straight lines, characteristic of diluted dispersions. For both the CP and PQ systems and their complexes, the G' and G'' moduli were similar at low frequencies, with G'' slightly higher than G' . In all cases, there is a tendency for cross-over at the approximate frequencies ($\log \omega_o$) 3.89 rad s^{-1} (CP), 1.54 rad s^{-1} (PQ), and 2.00 rad s^{-1} (CP:PQ).

The influences of NP and MP on the dynamic behavior of the CP dispersion are comparatively shown in Figure 3. The viscous response G'' dominates G' at low frequencies, but at high

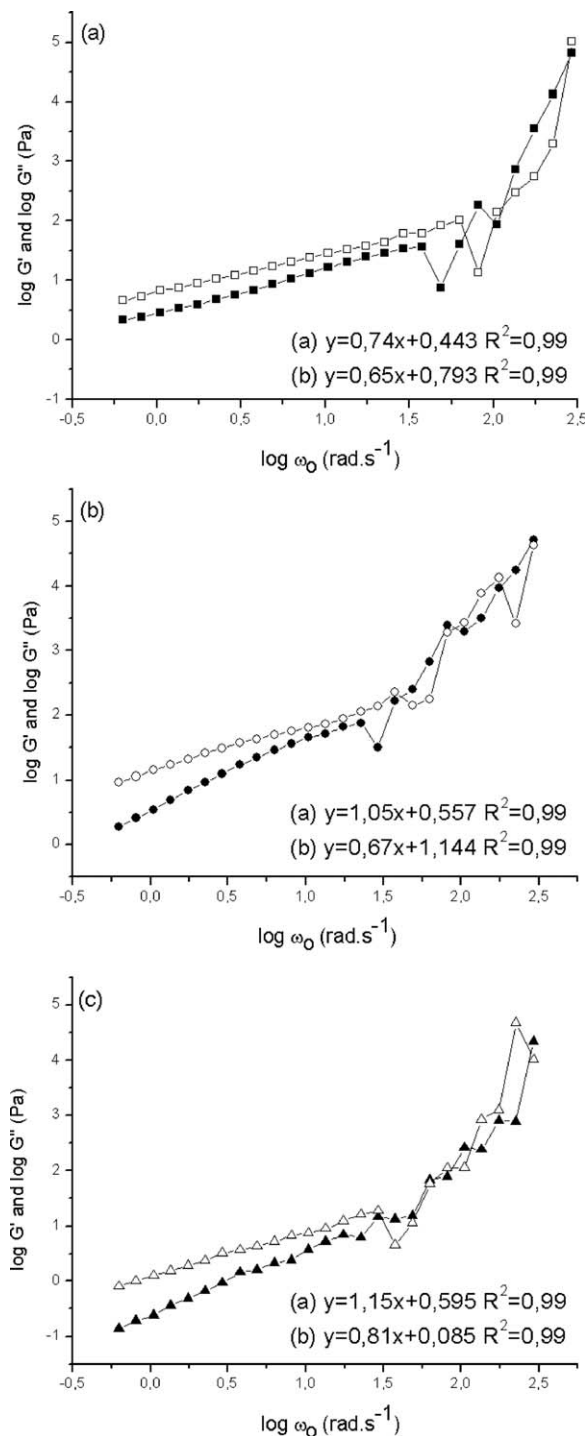


Figure 2. Storage G' (closed symbol) and loss G'' (open symbol) shear moduli of hydrogels at 25°C as a function of the angular frequency: (■) and (□) CP; (•) and (○) PQ; (▲) and (Δ) CP + PQ. The model fits and their equations are shown: (a) G' and (b) G'' .

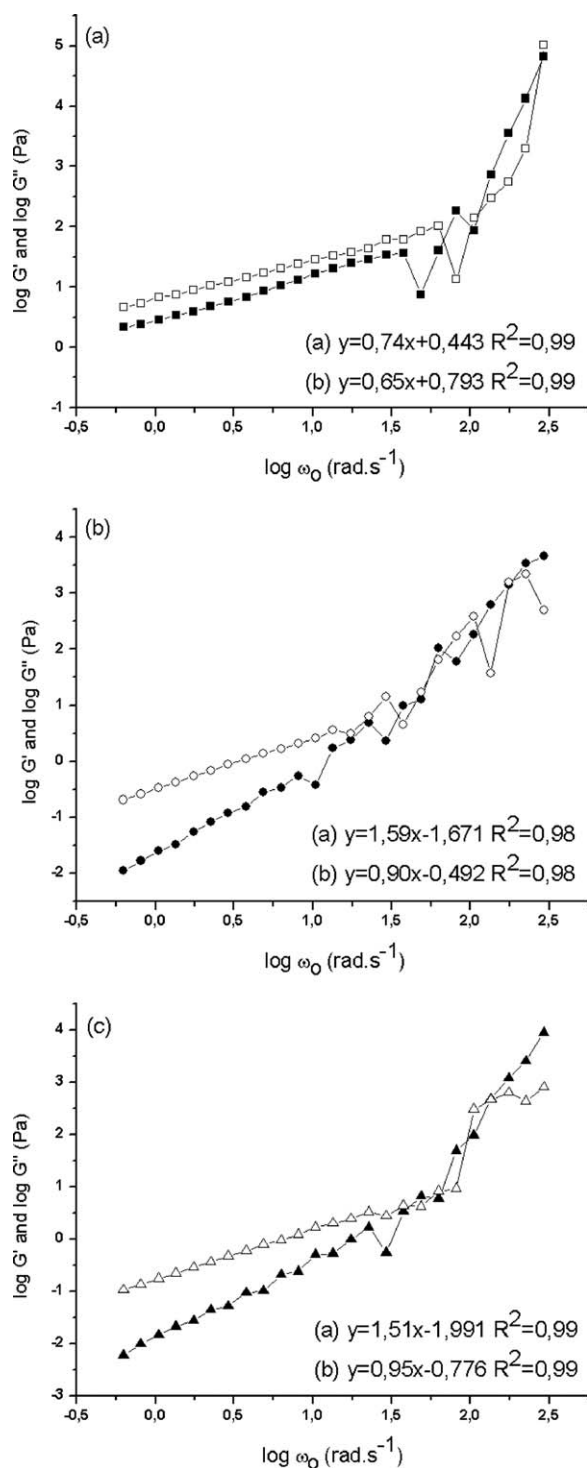


Figure 3. Storage G' (closed symbol) and loss G'' (open symbol) shear moduli of hydrogels at 25°C as a function of the angular frequency: (■) and (□) CP; (•) and (○) CP + NP; (▲) and (Δ) CP + MP. The model fits and their equations are shown: (a) G' and (b) G'' .

frequencies, G' approaches G'' as a result of the progressive storage of energy by the contortion of the chains into strained conformations.

The addition of NP or MP enhanced the viscoelastic behavior of CP, increasing G' from 0.74 Pa (CP) to 1.51 Pa (CP + NP)

and 1.59 Pa (CP + MP) when twice the amount of MP was added. In contrast, the cross-over frequency ($\log \omega_c$) was lower for the CP + MP system (1.71 rad s⁻¹) than for CP + NP (2.03 rad s⁻¹) or CP (3.89 rad s⁻¹). The negative zeta potential measured for the NP may cause rearrangements in the CP chains due to repulsion, and the particles may fill spaces among the chains, compressing the polymer. This behavior has been observed for other polymers, such as hyaluronic acid, in which NP and MP were efficient at increasing the viscoelasticity of the fluid polymer.¹⁷

Figure 4 shows the dynamic behavior of PQ and the effect of its associations with MP, as well as for the systems CP:PQ:(MP or NP). The profiles were similar to CP, with G'' higher than G' at low frequencies.

Table III summarizes the values of the G' and G'' moduli, as well as the cross-over frequencies, in terms of $\log \omega_c$, for the studied systems. MP increased G' (1.91 Pa) compared to pure PQ (1.05 Pa) and slightly decreased the cross-over frequency (1.54–1.41 Pa) compared with the reduction of the cross-over to CP:MP (1.71 Pa) and CP (3.89 Pa). The proportion of CP:PQ also influenced the parameters, as shown for CP:PQ:MP (A) and (B).

The increase in the G' in mixtures of CP with PQ and/or NP/MP indicates greater structural entanglement tending to gel-like behavior. The pectin gels exhibit junction zones stabilized by hydrogen bonds and hydrophobic interactions between chains.¹⁸ In pectin dispersions systems where the hydrogen bonds outweigh the hydrophobic interactions the behavior tend to be pseudoplastic.¹⁹

The overall resistance of the sample to the applied deformation may be characterized by the unresolved complex modulus, G^* [eq. (3)].

$$G^* = (G'^2 + G''^2)^{1/2} \quad (3)$$

The frequency of oscillation ω (rad s⁻¹) can be considered as the oscillatory analogue of the shear rate and the complex dynamic viscosity η^* (Pa s), which describes the total resistance to dynamic shear and is defined as:

$$\eta^* = G^* / \omega = (G'^2 + G''^2)^{1/2} / \omega \quad (4)$$

For dilute solutions, η^* is virtually independent of the frequency, analogous to the behavior of Newtonian liquids under steady shear, where the viscosity remains independent of the shear rate.

Figure 5 shows the behavior of the shear viscosity, η , and the complex viscosity, η^* , as functions of γ and ω , respectively. The curves show pseudoplastic behavior for CP with a strong tendency toward Newtonian behavior for the complex CP:PQ. The complex also lowered the viscosities compared to the pure polyelectrolytes.

The superimpositions of η and η^* at equal frequencies (ω) and shear rates (γ), known as the Cox-Merz rule and defined by $\eta^*(\omega) = \eta(\gamma = \omega)$, is applicable to fluids with a homogeneous structure, such as random-coil polysaccharide solutions. Good superposition of $\eta(\gamma)$ and $\eta^*(\omega)$ was observed for the pure polyelectrolytes and their complexes. The addition of the particles to

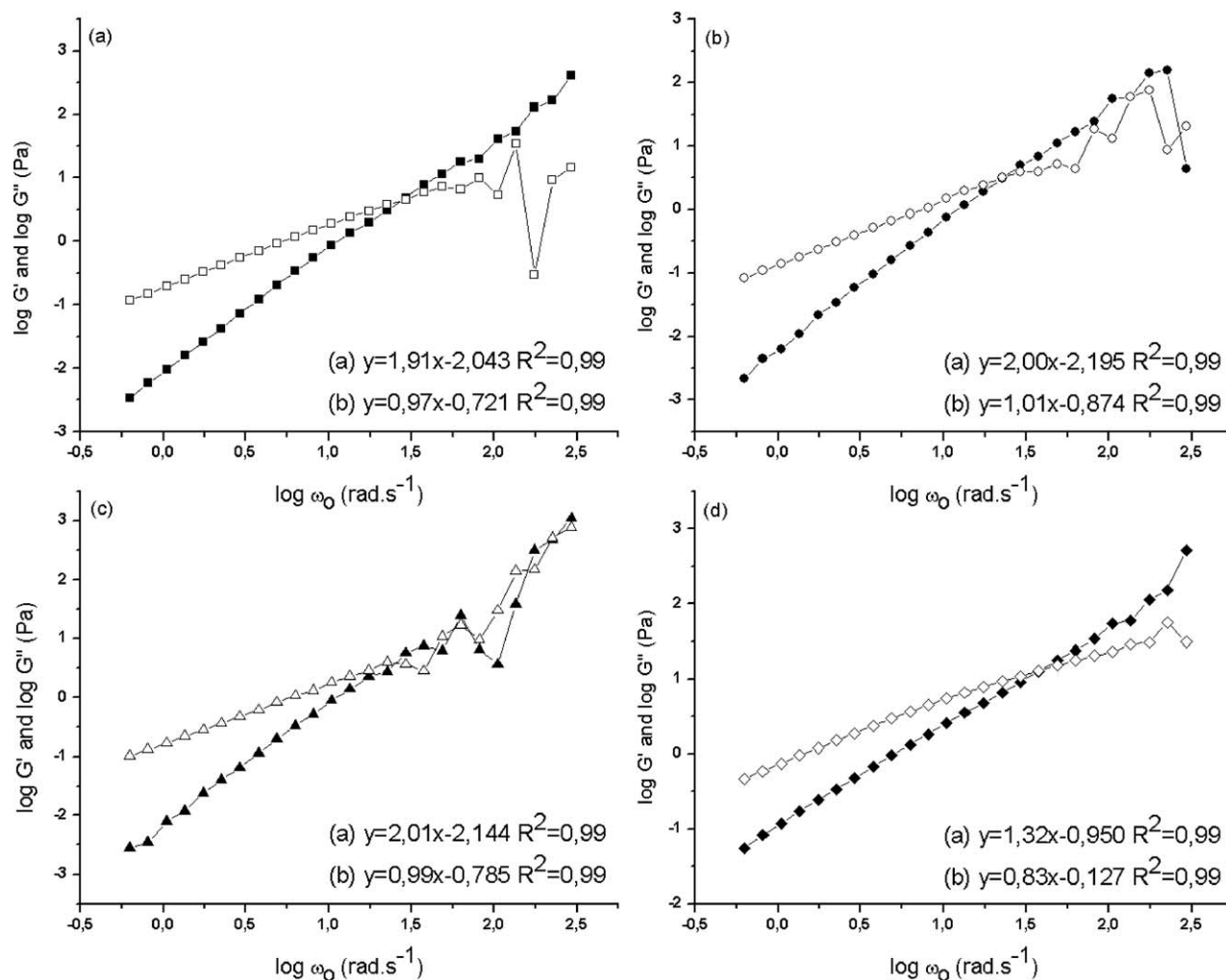


Figure 4. Storage G' (closed symbol) and loss G'' (open symbol) shear moduli of hydrogels at 25°C as a function of the angular frequency: (■) and (□) PQ + MP; (●) and (○) CP + PQ + NP; (▲) and (△) CP + PQ + MP (A) 1:20:0.4 CP:PQ:MP; (◆) and (◇) CP + PQ + MP (B) 1:50:3.6 CP:PQ:MP. The model fits and their equations are shown: (a) G' and (b) G'' .

CP further lowered both viscosities and increased the tendency for Newtonian behavior, as shown in Figure 5 (a,b).

However, the systems PQ:MP and CP:PQ:(MP or NP) (A) [Figure 5(c)] showed a strong deviation between the viscosities: at

low frequencies, the shear-viscosity had a pseudoplastic shear-thinning behavior, whereas the complex viscosity had a Newtonian behavior. At high frequencies, the complex viscosity increases with the frequency because of the high G' values, as in

Table III. G' and G'' Slopes and Intercepts Determining the Cross-Over Frequency (Ω_c) From Pectin Samples and its Derivatives

System	Composition w/w	G' (Pa)		G'' (Pa)		Intercept $\log \omega_c$
		Slope	R^2	Slope	R^2	
CP	5%	0.74	0.99	0.65	0.99	3.89
PQ	10%	1.05	0.99	0.67	0.99	1.54
CP:PQ	1:20	1.15	0.99	0.81	0.99	2.00
CP:NP	5:1	1.51	0.99	0.95	0.99	2.03
CP:MP	5:2	1.59	0.98	0.90	0.98	1.71
PQ:MP	50:1	1.91	0.99	0.97	0.99	1.41
CP:PQ:NP	1:20:0.4	2.00	0.99	1.01	0.99	1.33
CP:PQ:MP(A)	1:20:0.4	2.01	0.99	0.99	0.99	1.33
CP:PQ:MP(B)	1:50:3.6	1.32	0.99	0.83	0.99	1.68

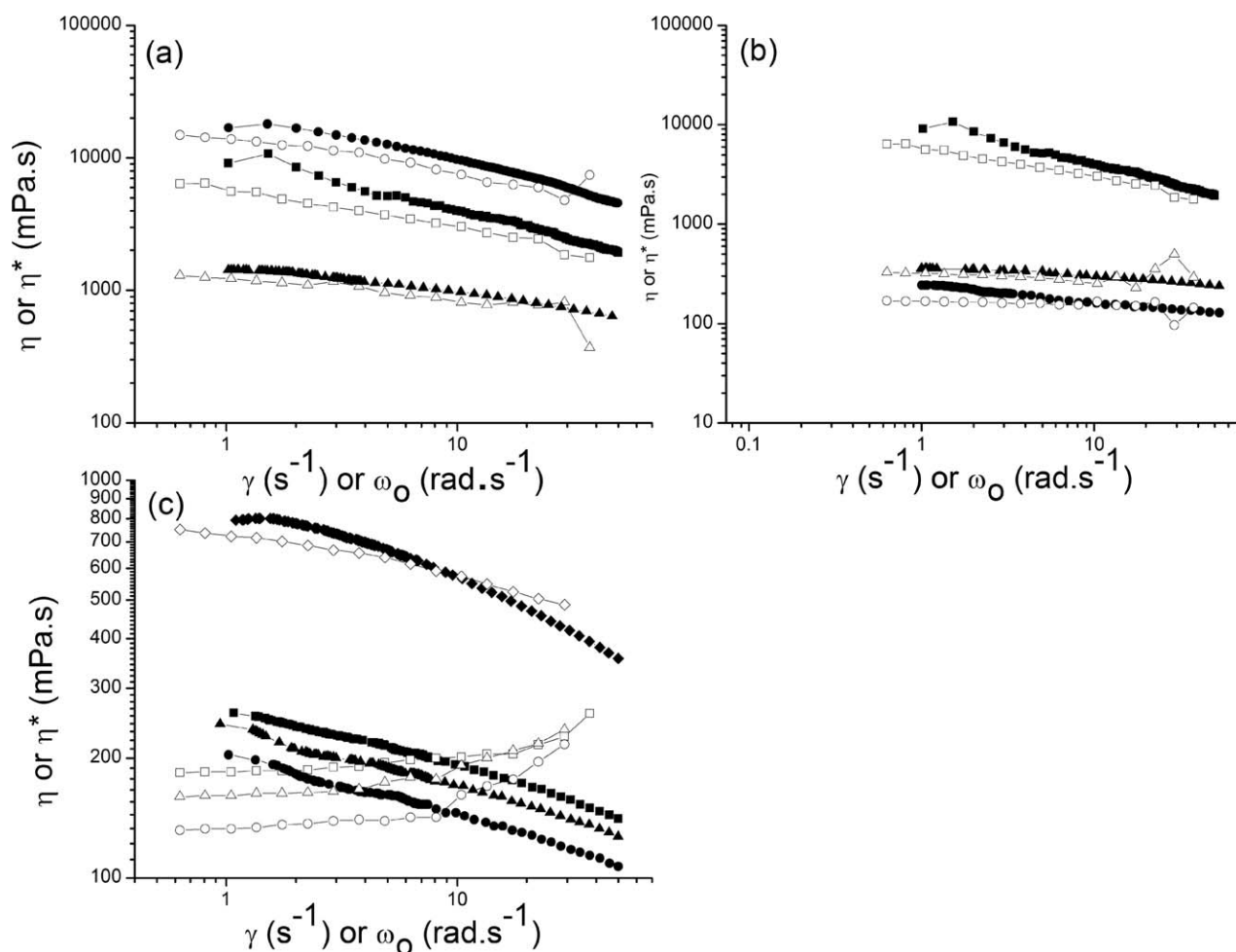


Figure 5. Cox-Merz plot of pectin and its derivatives at 25°C. Closed symbol (η) and open symbol (η^*). (a): (\blacktriangle) and (\triangle) CP + PQ; (\bullet) and (\circ) PQ; (\blacksquare) and (\square) CP. (b): (\blacksquare) and (\square) CP; (\blacktriangle) and (\triangle) CP + MP; (\bullet) and (\circ) CP + NP (c): (\blacksquare) and (\square) PQ + MP; (\bullet) and (\circ) CP + PQ + NP; (\blacktriangle) and (\triangle) CP + PQ + MP (A) 1:20:0.4 CP:PQ:MP; (\blacklozenge) and (\diamond) CP + PQ + MP (B) 1:50:3.6 CP:PQ:MP.

eq. (4). This is uncommon behavior because, in most cases of deviations, the complex viscosity is higher than the shear viscosity, which is caused by hyperentanglement or by the presence of aggregates. The proportion of the components also had an important role in the rheological behavior, such as for the system CP:PQ:MP (B), which showed pseudoplastic shear-thinning behavior for both viscosities compared to the uncommon behavior described for CP:PQ:MP (A).

Therefore, in the case of the PQ systems, the presence of the particles led to different behavior compared to CP due to the interactions among the particles with the opposite charge of PQ, which must be further investigated. The behavior was also dependent on the proportion of CP:PQ:MP.

Table IV shows that the Ostwald de Waele power law ($\eta = k \gamma^{n-1}$) parameters confirm the tendency for changing the behavior from pseudoplastic to Newtonian for the various systems containing particles.

For the polymers dispersions, the more pronounced pseudoplastic behavior reflects the low residual stress due to high hydration. However, the inclusion of MP and/or NP tends to disrupt

the water solvation for the formation of new electrostatic interactions, hydrophobic and hydrogen bonds, so that the systems tend to the Newtonian behavior.

Table IV. K and n Values in the Pseudoplastic Domain of η Versus γ Determined from the Flow Curves of Pectin and its Blends with Polyquaternium-7 and the NP and MP

Samples ^a	K	n
PQ	23988	0.59
CP + PQ	1531	0.79
CP	10914	0.56
CP + MP	373	0.90
CP + MP + NP	312	0.89
CP + MP + NP	245	0.83
PQ + MP	271	0.85
CP + PQ + NP	205	0.85
CP + PQ + MP (A)	239	0.85
CP + PQ + MP (B)	914	0.79

^aThe compositions of the systems are presented in Table I.

CONCLUSIONS

The rheological properties of CP dispersions may be modulated by the blending with PQ and also with MP or NP with negative zeta potential. In both cases, the particles increased the viscoelasticity of the systems and reduced the cross-over frequency. The particles also reduced the shear-viscosity and the complex viscosity and changed the systems to Newtonian behavior. The CP systems followed the Cox-Merz rule, but strong deviations were observed for the PQ systems, even when including CP, which was also dependent on the proportion of CP:PQ:MP. Therefore, the blending of CP dispersions with PQ and with the colloidal particles modulates its rheological properties and opens opportunities for applications when higher viscoelasticities are required.

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REFERENCES

1. Liu, L.; Cao, J.; Huang, J.; Cai, Y.; Yao, Y., *Bioresour. Technol.* **2010**, *101*, 3268.
2. Diaz, J. V.; Anthon, G. E.; Barret, D. M., *J. Agri. Food Chem.* **2007**, *55*, 5131.
3. Stephen, A. M.; Phillips, G. O.; Williams, P. A. *Food Polysaccharides and their Applications*, 2nd ed.; CRC Press Taylor & Francis Group: Boca Raton, FL, USA, **2006**; Chapter 11, pp 353.
4. Racapé, E.; Thibault, J. F.; Reitsma, J. C. E.; Pilnik, W., *Biopolymers* **1989**, *28*, 1435.
5. Ralet, M. C.; Dronnet, V.; Buchholt, H. C.; Thibault, J.F., *Carbohydrate Res.* **2001**, *336*, 117.
6. Thibault, J. F.; Rinaudo, M., *Biopolymers* **1985**, *24*, 2131.
7. Ralet, M.-C.; Crépeau, M.-J.; Buchholt, H.-C.; Thibault, J.-F. *Biochem. Eng. J.* **2003**, *16*, 191.
8. May, C. D., *Carbohydrate Polym.* **1990**, *12*, 79.
9. Bosio, V. E.; Machain, V.; López, A. G.; De Berti, I. O.; Marchetti, S. G.; Mechetti, M.; Castro, GR., *Appl. Biochem. Biotechnol.* **2012**, *167*, 1365.
10. Brandão, E. M.; Andrade, C. T., *Polímeros: Ciência e Tecnologia.* **1999**, *9*, 38.
11. Kasapis, S.; Al-Alawi, A.; Guizani, N.; Khan, A. J.; Mitchell, J. R., *Carbohydrate Res.* **2000**, *329*, 399.
12. Erçelebi, E. A.; Ibanoglu, E., *J. Food Sci.* **2009**, *74*, C506.
13. Masmoudi, M.; Besbes, S.; Ben Thabet, I.; Blecker, C.; Attia, H., *Food Sci. Technol. Int.* **2010**, *16*, 105.
14. Gharst, G.; Clare, D. A.; Davis, J. P.; Sanders, T. H., *J. Food Sci.* **2007**, *72*, C369.
15. Sanchez, C.; Renard, D.; Robert, P.; Schmitt, C.; Lefebvre, J., *Food Hydrocolloids* **2002**, *16*, 257.
16. Steffe, J. F., *Rheological Methods in Food Process Engineering*, 2nd ed.; Freeman Press: Michigan, **1996**; Chapter 1, p 1.
17. Shimojo, A. A. M.; Pires, A. M. B.; de la Torre, L. G.; Santana, M. H. A., *J. Appl. Polym. Sci.* **2012**, *128*, 2180.
18. Axelos, M. A. V., Thibault, J. F. *The Chemistry and Technology of Pectin*; Academic Press: New York, **1991**; p 109.
19. Sato, A. C. K.; Oliveira, P. R.; Cunha, R. L., *Food Biophysics.* **2008**, *3*, 100.