Investigation of the effect of trisodium-citrate on blood coagulation by viscometric approach

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ABSTRACT

Blood coagulation is a process, which is initiated by certain physico-chemical effects. This process results in a change in the blood from the sol state,that is well suited for further processing, to gel state. 13 blood clotting factors take part in the cascade system of blood coagulation. Trisodium-citrate affects factor IV, the calcium and prevents the change in blood texture. The effect of different amounts of trisodium-citrate (0 g, 0,1 g, 0,5 g, 1 g, 2 g, 3 g, 4 g, 5 g) on the texture of blood is investigated. Porcine blood was collected in 20 cm^3 test tubes in a slaughterhouse directly before trisodium-citrate addition and was stored for one day under refrigerated conditions. The samples without trisodium-citrate coagulated and the samples with high trisodium-citrate (4-5 g) became solid as well because of the protein salting-out. The viscosity of measurable samples and the shear stress was measured with a rotational viscometer (Physica MCR 51, Anton-Paar) with concentric cylinders and Couette type method. The flow behavior of all samples could be described by the Herschel-Bulkley model. The yield point, the consistency index and the power of law index, which are determined by the equation of the model, showed that the samples with lower trisodium-citrate content coagulated "better" and the sample with high trisodium-citrate were most similar to Newtonian fluid. The yield point of the sample, which contained 3 g trisodium-citrate, was by 37.3% less than the sample's containing 0.1 g trisodium-citrate, and the consistency index of the sample with 3 g trisodium-citrate was by 20.5% higher than that of the sample with 0.1 g trisodium-citrate.

KEY WORDS: blood processing, blood coagulation, trisodium-citrate, viscosimetry

INTRODUCTION

The utilization of byproducts, especially byproducts of animal origin, will be more important than ever before in the near future because of the increasing world population and need of food especially meat products. The animal blood produced in slaughterhouses is the most problematic from animal byproducts because it has very high polluting power (Lynch et al, 2017). Besides blood is produced in a very high amount. The weight of blood is 3-4% of the live animal weight, which is 3-4 kg per an average pig. (Meat Technology Update, 2003)

The blood may be a perfect additive or main component for functional foods because it is a rich source of iron and proteins of high nutritional and functional quality. (Putnam, 1975) If we would like to utilize the animal blood for human consumption in high amount, the blood must be separated to plasma and RBC fraction or other blood fractions because the whole blood has dark color and irony taste, which most consumers doesn't like (Ofori & Hsieh, 2012). Whole blood or other blood products can be spray dried or freeze dried for storage or transportation because the protein rich liquids have very unfavorable shelf-life. (Salvador et al, 2007)

Blood coagulation is a process, which is initiated by certain physico-chemical effects. This process results in a change in the blood from the sol state, that is well suited for further processing, to gel state. 13 blood clotting factors take part in the cascade system of blood coagulation (Schmaier & Hillard, 2011). But the blood coagulation must be inhibited directly following blood collection before any other preservation technologies would be used. The coagulated blood cannot be handled in a closed pipe system and it's important technological attributes are lost. 10 weight% sodium-citrate solution is added in 0,24 weight% to the fresh whole blood for inhibiting blood coagulation in most places according to the old industrial recommendation (Gárgyán, 1991). But if water is added into the blood, the concentration and drying process will be slower and more expensive.

The effect of dry trisodium-citrate powder on the coagulation of whole animal blood was investigated in this study. The change in blood texture is measured by viscometric approach. If small amount of dry trisodium-citrate powder can inhibit the blood coagulation in industrial environment, the water content of blood may be lower before the following processes.

MATERIAL AND METHODS

Materials

Porcine blood was collected manually in a slaughterhouse directly from the slaughtered pork in South Transdanubia during the bloodletting. 20 cm^3 test tubes were used as sample holder. The trisodium-citrate was added to the blood directly after collection then the substances were mixed well. The samples were mixed with different amounts of trisodium-citrate: 0 g, 0,1 g, 0,5 g, 1 g, 2 g, 3 g, 4 g, 5 g. If we calculated with the density of porcine blood, which is 1040 kg/m^3 (Kowalsky et al, 2011), the trisodium-citrate content of the samples were the followings in weight percent: 0%, 0.48%, 2.4%, 4.8%, 9.6%, 14.4%, 19.2%, 24%. The prepared samples were stored in the test tubes for one day under refrigerated condition before the measurements.

Methods

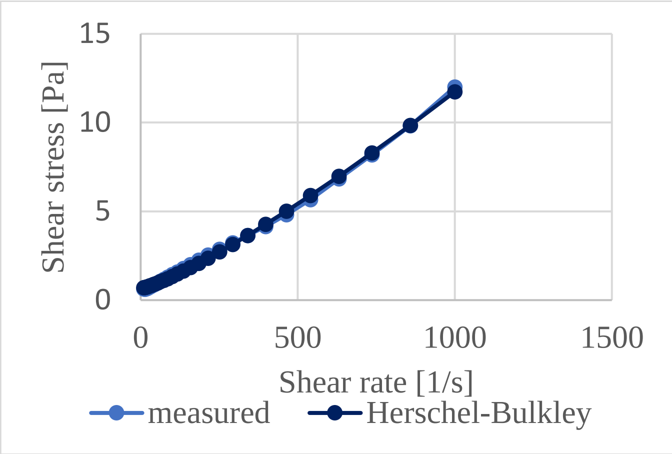
An Anton-Paar Physica MCR 51 viscosimeter device was used for the investigation. The sample temperature was 20 °C during the measurement. The viscosity of measurable samples and the shear stress was measured with concentric cylinders and Couette type method. Data were collected between 10 and 1000 1/min shear rate and the flow behavior of all samples could be described by the Herschel-Bulkley model, which considers the following parameters: shear stress (τ), theoretical yield point (τ_0), deformation speed (γ), consistency index (C) and power of law index (p). The parameters were calculated from the shear rate and shear stress. A new shear rate was calculated from the calculated parameters calculated and this new shear rate validates the compliance of the model. The correlation coefficient (R^2) shows how appropriate/correct a model is.

RESULTS

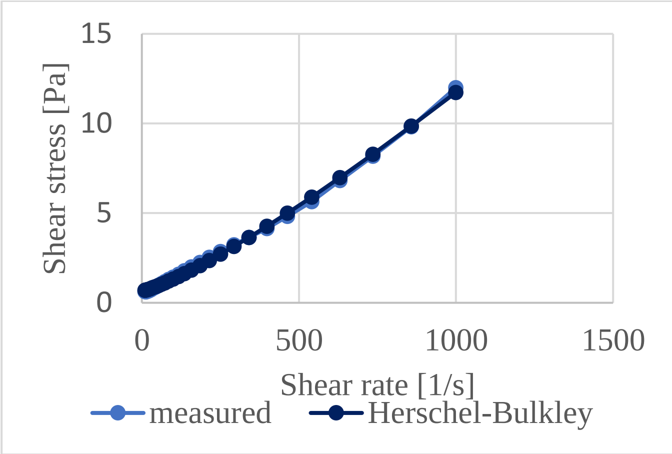
The yield point, the consistency index and the power of law index, which are determined by the equation of the model, showed that the samples with lower trisodium-citrate content coagulated “better” and the sample with high trisodium-citrate were most similar to Newtonian fluid. The yield point of the sample, which contained 3 g trisodium-citrate, was by 37,3% less than the sample’s containing 0.1 g trisodium-citrate, and the consistency index of the sample with 3 g trisodium-citrate was by 20.5% higher than that of the sample with 0.1 g trisodium-citrate. It can be described globally that the sample, which contains 0.48% trisodium-citrate, was least similar to Newtonian fluid and the other samples were similar to each other in terms of yield point, consistency index and power of law index. The correlation coefficients of the samples were higher than 0.995 so the model, that was used for calculating the parameters, was appropriate.

Table 1. Calculated constants of Herschel-Bukley model of measurable porcine blood samples with different trisodium-citrate

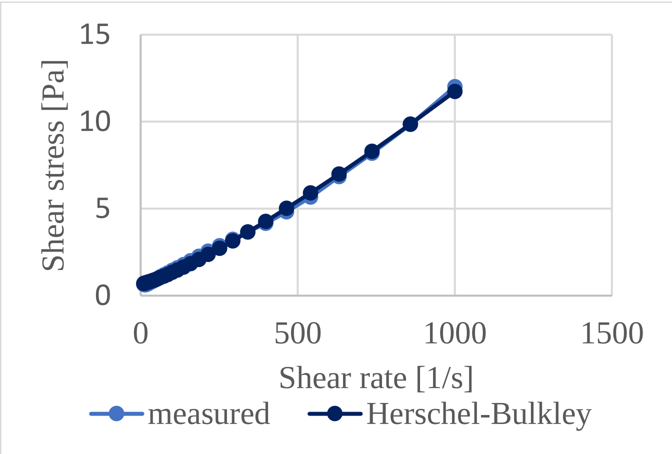
Na_3-citrate (weight%)	τ_0 (Pa)	C	p	R_2
0.48	0.658	2.49E-03	1.22	0.999
2.4	0.346	8.95E-04	1.34	0.995
4.8	0.268	3.33E-03	1.17	0.997
14.4	0.279	8.19E-04	1.36	0.997
19.2	0.246	1.21E-02	1.03	0.999



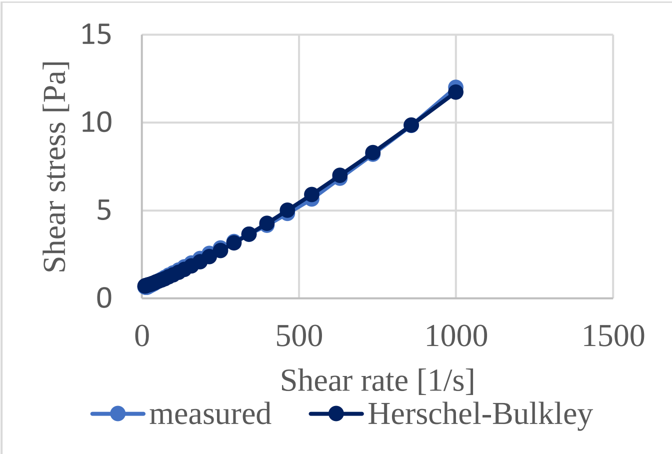
a) 0.48% trisodium-citrate



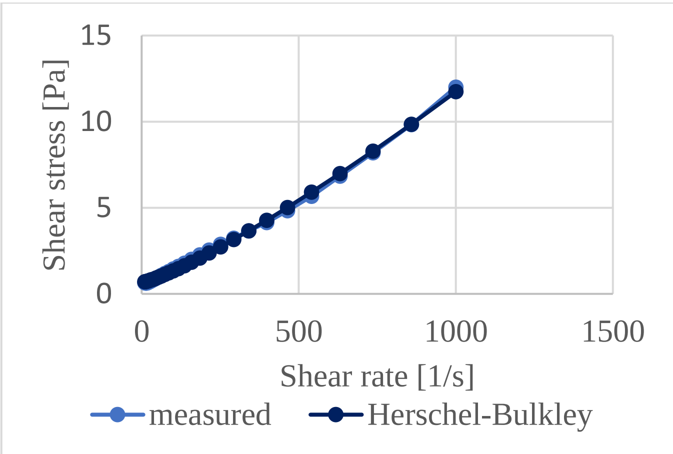
b) 2.4% trisodium-citrate



c) 4.8% trisodium-citrate



d) 14.4% trisodium-citrate



e) 19.2% trisodium-citrate

1. figure: Flow charts of measurable porcine blood samples with different trisodium-citrate

The samples without trisodium-citrate coagulated and the samples with high trisodium-citrate content (4-5 g) became solid as well. The blood coagulated in the control samples. The proteins of samples with high trisodium-citrate were probably salting-out. The pH cannot reach the isoelectric point of proteins because the trisodium-citrate is alkaline and the isoelectric point of the main blood proteins are in the acidic region. Besides there was a solid layer on the surface of the sample with 0.48% trisodium-citrate.

CONCLUSION

The least sodium-citrate content, which is able to inhibit the blood coagulation for one day, is 2.4% according to the results of this study. If a firm collects animal blood for drying or producing blood products with low water content, it is advantageous to use dry sodium-citrate powder instead of a liquid solution.

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