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Impact of excipients on buffer viscosity

Key Words: viscosity, buffer solutions, arginine hydrochloride, histidine hydrochloride, sucrose, D-mannitol, trehalose, maltose, D-galactose, sugar alcohols, amino acids, monosaccharide, disaccharide

Goal: Buffer solutions containing a variety of common additives, including sugars and individual amino acids, were prepared. The viscosity of each was measured to illustrate the ability of the fully automated VROC® initium to differentiate low viscosity fluids.

Introduction

It is often desirable to distinguish low viscosity fluids such as buffer solutions prepared to solubilize proteins or antibodies. Buffers are formulated with a variety of excipients or inactive ingredients to ensure protein stability or to reduce the viscosity without lowering the concentration of the active. Common additives include mono or disaccharides, sugar alcohols, and individual amino acids. Since the buffer solution is the foundation of the fully formulated protein, it is important to accurately measure its viscosity. This will ensure consistency of the raw materials as well as the formulation process. Most viscometers are unable to make such distinctions, and those that are capable tend to be very tedious and time consuming. This application note demonstrates the advantages of measuring with the VROC® initium, which incorporates microfluidic and MEMS technology into a fully automated system.

Experiment

Buffer solutions were prepared by adding a variety of common excipients to a phosphate buffered saline (PBS, pH = 7.4). Arginine hydrochloride (ArgHCl), histidine hydrochloride (HisHCl), and sucrose solutions were made at multiple concentrations up to a maximum of 500 mM. Mannitol, trehalose, maltose, and galactose solutions were formulated at 250 mM. The viscosity of the buffer solutions was measured on the VROC® initium with a B05 chip (depth = 50 μm, P_{max} = 40K Pa) at 25°C using a flow rate of 1000 μL/min (shear rate = 19400 sec^{-1}). The loaded volume for each sample was 80 μL and twelve measurements were made for each with the retrieval feature activated.

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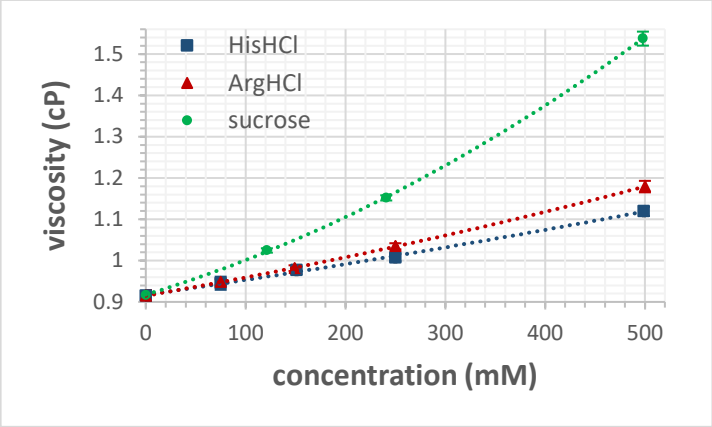

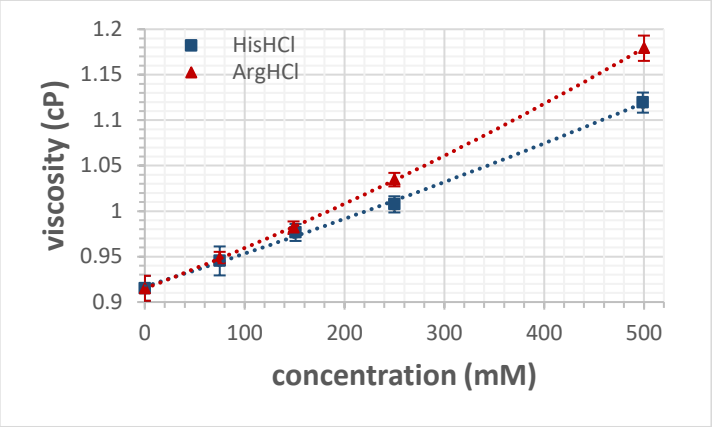

Figure 1: Viscosity of PBS including various concentrations of HistHCl, ArgHCl, and sucrose. Error bars represent ± three standard deviations.

Viscosity Data and Discussion

Viscosity as a function of concentration is presented in Figure 1 (*left*) for the PBS with added HistHCl, ArgHCl, and sucrose. The curves for the buffers containing the ArgHCl and HisHCl are also shown on an expanded scale to emphasize the ability to distinguish the two samples (Figure 1, *right*). Even at the lowest concentrations (75 &150 mM) where the viscosity is less than 1 cP, the samples can be differentiated from the PBS. Also, as the concentration is further increased, the difference between the two amino acids can be measured. Although the two are nearly equivalent in molecular weight (Table 1), it is expected that a significantly higher fraction of ArgHCl molecules will have a positive charge at the solution pH. Therefore, the more rapid increase in viscosity of the buffer containing ArgHCl may result from stronger electrostatic pair interactions.

buffer	η (cP)	Std dev	RSD %	MW (g/mole)	Wt %
PBS	0.92	0.0033	0.36	N/A	N/A
+trehalose dihydrate	1.17	0.0030	0.26	378.33	9.13
+maltose hydrate	1.17	0.0023	0.19	360.31	8.72
+D-galactose	1.03	0.0029	0.28	180.16	4.42
+sucrose	1.16	0.0033	0.28	342.3	8.28
+D-mannitol	1.04	0.0027	0.26	182.17	4.47
+Arginine monoHCl	1.03	0.0025	0.24	210.66	5.14
+Histidine monoHCl	1.01	0.0030	0.29	209.63	5.18

Table 1: Viscosity (η) of PBS solutions containing various additives at a concentration of 250 mM. (Std dev = standard deviation, RSD = relative standard deviation, MW = molecular weight, Wt % = weight percent)

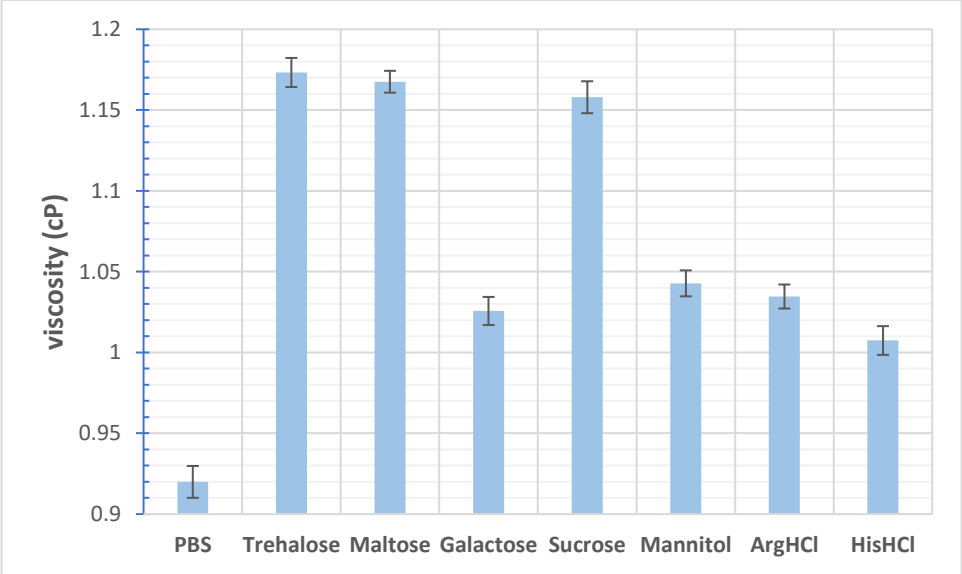

Figure 2: Viscosity of PBS with various additives at a concentration of 250 mM. Error bars represent ± three standard deviations.

Figure 2 illustrates the impact on viscosity of all additives used at a concentration of 250 mM, with the individual values and measurement statistics also included in Table 1. Relative standard deviation values based on twelve measurements are consistently less than 0.5 % for each formulation and clearly demonstrates the precision of the technique.

Concluding Remarks

Buffers are the foundation of therapeutic formulations. Excipients are almost always included to ensure long term stability and a viscosity acceptable for the application. It is crucial that the buffer is properly and consistently formulated. Monitoring the viscosity is an effective quality control process. The VROC® initium is a fully automated system capable of distinguishing viscosity increments on a sufficiently small scale appropriate for this purpose.

If this note is helpful, please let us know! If you have questions or need more information about this product or other applications, please contact us:

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