Lab Report 4: Proteins

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Introduction:

Proteins are an important factor in life as they are located all over the human body, contribute to oxygen being transported throughout the bloodstream, and makeup enzymes needed for different chemical processes (Harvard 2017). Proteins are made up of varying amounts of amino acids that are linked together through peptide bonds. Every amino acid contains two functional groups, the amino group (NH₂) and a carboxylic acid, that are attached to differing organic compounds that are often referred to as "R" groups (McWilliams 2012). The order and number of amino acids linked together dictate the purpose of the protein as well as its shape. There are four main structures of protein: primary, secondary, tertiary, and quaternary structures. The primary structure is straightforward, and simply a chain of amino acids. The secondary structure interacts with the backbone structure through hydrogen bonding, and can form either an *a*-helix structure or a β -pleated sheet (Rice University). In the tertiary structures amino acids interact through hydrogen bonds, salt bridges, and covalent bonds, further promoting folding of the protein (McWilliams 2012). Lastly, the quaternary structure is formed with two or more amino acid chains, and all of the interacting bonds create a bundling structure (Rice University). When strain is applied to proteins in the form of heat, pH changes, or mechanically through actions such as pounding or mastication, the structure of the secondary, tertiary, quaternary proteins can begin to unwind, and this change in form also changes the function of the protein. These changes are referred to as denaturing (McWilliams 2012).

The different forms of proteins contribute to different functional properties. Some functions of proteins in food are shaping foams, thickening agents, and providing structure support. In foams, proteins that are whipped, such as egg whites, begin to denature, and provide a firmness and stability to the walls of the foam. Familiar foods that use this process are angel food and sponge cakes, as well meringues (McWilliams 2012). In thickening, heat is used to denature the proteins from a tertiary to secondary structure, and as the mixture begins to solidify, water is bound between the hydrogen and other bonds, creating a gel. This method is often used in hollandaise sauce and custards (McWilliams 2012). Any baked goods involving wheat flour contain gluten, which is a protein. Gluten provides structure to the bread, as well as the chewy texture. The gluten extends and stretches during the baking process, expanding further with the build up of

the gas within the mix. As the heat during baking solidifies the mix, the gluten become stiff enough to support the structure of the bread (McWilliams 2012). Each of these are ways that protein contributes to different food components, with the characteristics of gelatin being explored in this experiment.

The purpose of this experiment was to be able to recognize and detail the impact a change in pH has on the viscosity of a dairy based dessert, as well as understand the impact of proteolytic enzymes on, and recount the process of gelation of gelatin. During this experiment texture analysis, viscosity measurements, and pH readings would be taken using the TA.XT Plus Texture Analyzer, the Fungilab viscometer, and the Oakton pH Meter respectively.

Materials & Methods:

Two procedures must be finished in order to complete this experiment. The first step involved the measuring of pH and viscosity of the dairy sample that was made during the lab. In a bowl, two different types of canned milk were mixed: 354 ml of evaporated milk and 397g of condensed milk. On the side, 125 ml of fresh squeezed lime juice was obtained. Next, 15 ml of lime juice was added to the dairy mixture, the pH was checked directly from the bowl using the Oakton pH Meter. After the pH of the sample was recorded, about 140 ml of the sample was transferred to a 150 ml beaker to measure the viscosity using the Fungilab viscometer. The action of adding the lime juice and recording pH and viscosity measurements was repeated five times until the sample reached a pH of 4.5-4.7. The second part of the experiment involved making a gelatin. The fresh pineapple juice was provided by the instructor. One packet of Jello was dissolved in 250 ml of boiling water, and cooled down to 50°C. The temperature was recorded using a battery operated thermometer. A dissolved gelatin was poured into six sample cups. Two cups contained 40 ml water, two more cups contained 40 ml of processed pineapple juice, and the final two cups contained 40 ml of fresh pineapple juice. The liquid was cooled to 30°C and refrigerated. The following week the firmness of the samples was measured using the TA.XT Plus Texture Analyzer.

Results and Discussion

Amount of lime juice (ml)	рН	Viscosity (cP)	%
15	5.90	278.0	2.7
30	5.59	313.3	3.1
45	5.27	832.2	8.3
60	4.95	2007.2	20.1
75	4.65	1988.3	19.9

Table 1. MI of lime juice and pH measurements of dairy mixture

Graph 1. Viscosity vs. pH of dairy mixture



Sample	Kitchen 1	Kitchen	Kitchen 3	Kitchen	Mean	SD
water	0.0224	0.0187	0.0041	0.0040	0.0123	0.009645
water	0.0214	0.0167	0.0032	0.0037	0.0113	0.009211
Processed pineapple juice	0.0148	0.0102	0.0027	0.0027	0.0076	0.005962
Processed pineapple juice	0.0153	0.0123	N/A	0.0026	0.0101	0.006638
Fresh	N/A	N/A	N/A	N/A	N/A	N/A
pineapple juice						
Fresh pineapple juice	N/A	N/A	N/A	N/A	N/A	N/A

Table 2. Table of firmness of each gelatin treatment.

The viscosity of the dairy mixture increased as more lime juice was added because the decrease in pH made it more acidic. When the pH increases or decreases the electrical charge of the amino acids from the protein changes. In milk products the milk protein, casein, is negatively charged at a pH of 6.5-6.7 (Herbstreith). When this change in pH occurred, the protein was modified to a net charge of zero. This pH value is called the Isoelectric point (Ip), and it is noticeable at pH 4.9-5.1 as shown on Graph 1. This value was where proteins began to lose their solubility, thus a thickening of the solution or an increase in viscosity was observable. Pectin which is naturally occurring in fruits is often used as a stabilizer in dairy products. The interaction between pectin and casein are mutually repellent causing the stabilization of the milk proteins (Herbstreith). This

was why the change in the electrical charge caused a thickening effect due to the disruption of this balance. When comparing the gelatinization of jello the differences were significant in the use of water, fresh pineapple juice and processed pineapple juice as a solvent. The samples that were made by the use of fresh pineapple juice were watery, their color was cloudy red, and they did not become firm. The samples made with a processed pineapple juice were a little bit cloudy, and firm with no flow. The samples made with plain water were transparent red color and they were the most firm. The process of forming a gelatin involve proteins that make links between the chains of collagen. However, a fresh pineapple juice contains the enzyme called bromelain which breaks down the proteins. This prevented the formation of the gelatins. Pineapple, as well as other fruits such as kiwi, mango, papaya, and guava, contain enzymes that cleave the bonds between the amino acids. These enzymes work on the gelatin, separating the bonds, so that the gelatin cannot solidify and bind the water. This results in a liquified state. If the fruit is heated, such as in the canning process, then the enzymes are destroyed, and the gelatin would be able to solidify (Scientific American 2013). Evaporated and condensed milk are very similar but have one key difference. Evaporated milk is made by removal of water only from milk by evaporation. It contains no less than 6.5 percent by weight of milkfat and no less than 23 percent by weight of total milk solids. Evaporated milk contains added vitamin D and it is homogenized. It is sealed in a container that is processed by heat to prevent spoilage. Condensed milk is made by the removal of water through evaporation. The milk fat and total milk solids contents of the food are not less than 7.5 and 25.5 percent. It is pasteurized but is not processed by heat to prevent spoilage (2006).

Conclusion:

During the protein lab, the effects of pH changes on viscosity of a dairy dessert were observed through the creation of a dairy dessert via lime juice, evaporated milk, and condensed milk. As noted in Table 1 above, when the pH increased with the amount of lime juice added, the viscosity began to increase as well, thus causing a direct correlation between lime juice acidity and viscosity of the dairy dessert. During this portion of the lab, the viscosity was measured using the Fungilab Viscometer, and the pH was measured using the Oakton pH Meter. In

addition to measuring viscosity, gelation of gelatin was also noted through creating a Jell-O mixture using three different additions to the gelatin: processed pineapple juice, fresh pineapple juice, and water. Each method created a different sample with the fresh pineapple juice not producing a gelatin mixture when data collection was noted. This was a result of the enzymes in the fresh pineapple juice that prevented the gelatin mixture from solidifying thus making the final product liquidity rather than the expected gelatin. The firmness of the water and processed pineapple juice gelatins were measured using TA.XT Plus Texture Analyzer after a week of refrigeration. The firmness of the fresh pineapple juice mixture was not measured simply because it had not solidified enough for the machine to take a measurement.

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