Validation of capillary electrophoresis for the ultra-rapid determination of inorganic phosphate and citrate in milk. Jesus M. Izco*, Monica Torma, Phil S. Tong, and Rafael Jimenez-Flores, Dairy Products Technology Center, Cal Poly University.

The aim of this work is to optimize a Capillary Electrophoresis method for the ultra-rapid determination of citrate and inorganic phosphate in milk. The quantification of these compounds is very important because their distribution between soluble and colloidal phases of milk, and their interactions with milk proteins influence the stability and some functional properties of dairy products. Various parameters affecting analysis have been optimized, including capillary length, type, composition and pH of the electrolyte, and sample extraction. The separation was carried out on an uncoated capillary (50 cm, 75 μm I.D.) at -25kV for 2.5 min. According to pKa values for citric and phosphoric acid, pH of the running buffer between 9.5 and 3.0 were tested in order to obtain all the possible ionized forms for both acids and to select the pH yielding the best separation. Ethanol, acetonitrile, sulfuric acid, water at 50°C and at room temperature were tested as sample buffers (SB). Water at room temperature gave the best overall results and was chosen for further validation. The extraction time was checked and could be shortened to less than 1 min. Also, sample preparation was simplified to pipette 12 μl of milk into 1 ml of water containing tartaric acid as an Internal Standard, not being necessary further treatment. The linearity of the method was excellent (R² >0.999) with CV values of response factors <3%. The detection limits for phosphate and citrate were 5.1 and 2.4 nM respectively. The accuracy of the method was calculated for each compound (103.2 and 100.3%). In addition, several commercial samples were analyzed and the results showed a deviation less than 5% from values obtained when analyzing the samples by official methods. Also, to study the versatility of the technique, other dairy products such as cream cheese, yogurt or Cheddar cheese were analyzed. Accuracy was similar to milk in all products tested. Because of the speed and accuracy of this method, it is promising as an analytical quantitative quality testing technique.

Key Words: Capillary Electrophoresis, Milk, Phosphate and Citrate