Measurement of a Milk Gelation Time Constant Using Laser-Scanning Fluorescence Confocal Microscopy and Image Processing Techniques

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The gelation kinetics of milk can dictate how nutrients are absorbed after ingestion and are therefore important when determining the nutritional benefit of a dairy product. Current methods to measure gelation kinetics, such as near-infrared spectroscopy and rheology, are destructive and only provide one-dimensional data, while other methods, such as the Berridge clotting time method, are subjective because they depend on an operator's skill. A two-dimensional, non-destructive, objective measurement technique is needed to accurately quantify the gelation kinetics of milk. The purpose of the present study was to investigate the ability of laser-scanning fluorescence confocal microscopy (LSFCM) to measure gelation kinetics.

In this study, a mixture of raw milk and chymosin was imaged using LSFCM. The milk was stained with the fluorescent markers Nile red, which stains lipids, and fast green FCF, which stains proteins. Once chymosin was added to the raw milk, images were captured every 5 seconds for 30 minutes. Because gelation causes the milk to change from a liquid to a solid, the instantaneous gelation rate could be estimated by calculating the mean difference between successive images (R). As the milk begins to gel, the movement of the lipids and proteins eventually ceases, and the mean difference between successive frames eventually reaches zero. R was plotted versus time and fit to the curve where B is the initial value of R, T is the temperature of the milk at the time the images were acquired, [Ch] is the concentration of chymosin, t is the time, and k is the gelation time constant of the milk. The gelation time constant, k, was then used to characterize the gelation kinetics. Because this method is able to account for the initial rate of the gelation process, the chymosin concentration, and the temperature when calculating the gelation time constant, it shows promise as a technique to measure and compare the intrinsic gelation characteristics for different milk varieties.