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## Detection of elastin, collagen, and cartilage particles in ground beef by enzyme digestion and sensory analysis

### Abstract

An enzyme digestion technique was developed using a proteolytic enzyme concentrate to quantitate connective tissue particles in ground beef samples, which had been formulated to contain various amounts of connective tissue. Replicate samples were also evaluated by a taste panel to quantify detectable connective tissue particles. Results for the laboratory enzyme technique and the taste panel were highly correlated. Therefore, the enzyme digestion technique can be used to estimate total connective tissue in ground beef and those particles that are detected upon chewing.

### Keywords

Cattlemen's Day, 1987; Kansas Agricultural Experiment Station contribution; no. 87-309-S; Report of progress (Kansas State University. Agricultural Experiment Station and Cooperative Extension Service); 514; Beef; Elastin; Collagen; Cartilage; Ground Beef

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Detection of Elastin, Collagen, and Cartilage Particles  
in Ground Beef by Enzyme Digestion and  
Sensory Analysis

G. Whipple, J.B. Axe, S. Goll,  
and C. Kastner

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### Summary

An enzyme digestion technique was developed using a proteolytic enzyme concentrate to quantitate connective tissue particles in ground beef samples, which had been formulated to contain various amounts of connective tissue. Replicate samples were also evaluated by a taste panel to quantify detectable connective tissue particles. Results for the laboratory enzyme technique and the taste panel were highly correlated. Therefore, the enzyme digestion technique can be used to estimate total connective tissue in ground beef and those particles that are detected upon chewing.

### Introduction

The meat industry uses many processing techniques to reduce the amount of perceivable connective tissue in ground beef. However, the industry needs a rapid, inexpensive, simplified laboratory method for quantitating connective tissue in ground beef. Several enzymes from plant and microbial sources digest both muscle fiber proteins and connective tissue proteins. Some of those enzymes are more selective against one type of protein than another. We wanted to find an enzyme that would digest the muscle fiber (myofibrillar protein) and leave connective tissue structures intact, allowing measurement of elastic and collagenous connective tissue particles, cartilage, and bone. Such a method could be used to help prevent ground beef containing detectable connective tissue from reaching the consumer.

### Experimental Procedures

In Phase 1, we found the most effective enzyme system to be the HT proteolytic enzyme.<sup>1</sup> The remainder of the assays were carried out as follows: Stock solution was prepared in 128 F water to contain 163 Northrup units per ml. Twenty-five gm of ground beef were placed in a 250 ml flask with 100 ml of enzyme solution and incubated with agitation for 30 min in a 128 F water bath. Flask contents were filtered on screen cloth (250  $\mu$ m mesh openings) and the residue was rinsed with 250 ml of 5% NaCl, followed by 250 ml of distilled water. The residue was scraped from the screen cloth, weighed (Phases 1 and 2 only) and spread on plexiglass. Connective tissue particles 3mm or larger were removed, counted, and weighed.

In Phase 2, the enzyme digestion procedure was implemented on ground beef samples containing 0, 6, 8, and 10% added connective tissue. Data were treated statistically to estimate repeatability and sensitivity of the technique.

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<sup>1</sup>Miles Laboratories Inc., Biotech Products Division, Elkhart, IN 46514.

In Phase 3, we determined what type of connective tissue was actually perceivable upon chewing. Only samples to which cartilage was added contained hard particles that were detectable upon chewing. Based on those results, coarsely ground (0.5 inch plate) cartilage with collagen attached was added at levels of 0, 3, 6, and 9% to coarsely ground (0.5 inch plate) beef. All treatment batches were formulated to 22% fat. Finally, all treatment samples were reground through a 0.125 inch plate and samples were analyzed by both the enzyme digestion technique and the taste panel. Hard particles were detected by pressing the index finger through the residue from enzymatic digestion. When detected, the hard particles were removed, counted, and weighed. Using forceps, soft connective tissue particles were also removed, counted, and weighed.

For the taste panel analysis, thawed patties (one 4 oz. patty per treatment) were cooked at 250 F for a total of 3.0 minutes. The taste panel consisted of six trained members. The technique involved chewing each treatment sample normally and recording the number of hard particles detected. Panelists were also asked to pass another sample through their incisors and count the number of hard particles.

### Results and Discussion

From preliminary study results, we concluded that digesting samples with HT proteolytic concentrate (128 F for 30 min) was the most effective way to digest muscle fiber protein and leave connective tissue intact. That process allowed for differentiation, separation, and quantitation (both by count and weight) of connective tissue particles.

Total residue weight, connective tissue particle count, and particle weight for various levels of added connective tissue in Phase 2 were statistically analyzed. For total residue weight, no differences ( $P > .05$ ) were observed among the four treatments; however, there were differences ( $P < .05$ ) for particle count and weight. High correlations were seen between percent added connective tissue and the connective tissue particle weight (0.888) and count (0.924), and also between particle weight and particle count (0.906). These high values indicate that the enzyme digestion technique is highly repeatable and that either particle count or weight could be used to indicate the amount of particulate connective tissue in a sample.

Among the treatments (0, 3, 6 and 9% added connective tissue) in Phase 3, there were no differences ( $P < .05$ ) for hard particle count, soft particle count, total particle count, and soft particle weight. However, weights of hard particles and total particles were different ( $P > .05$ ).

Emphasis was placed on the hard particle count in the taste panel analysis because hard particles detected upon enzyme digestion were the only ones which affected taste panel perception. Figure 14.1 illustrates the mean hard particle counts for normal chewing, incisor detection, and enzyme digestion. For each detection method, differences were significant ( $P < .05$ ) among the means for each treatment.

The correlation coefficient between normal chewing hard particle count and incisor hard particle count was 0.998. The correlation of enzyme hard particle count with normal chewing and incisor hard particle counts were 0.985 and 0.980, respectively. Thus, the enzyme digestion technique of ground beef can be used successfully as a quality control method for estimating the total amount of connective tissue and hard connective tissue particles detected upon chewing.

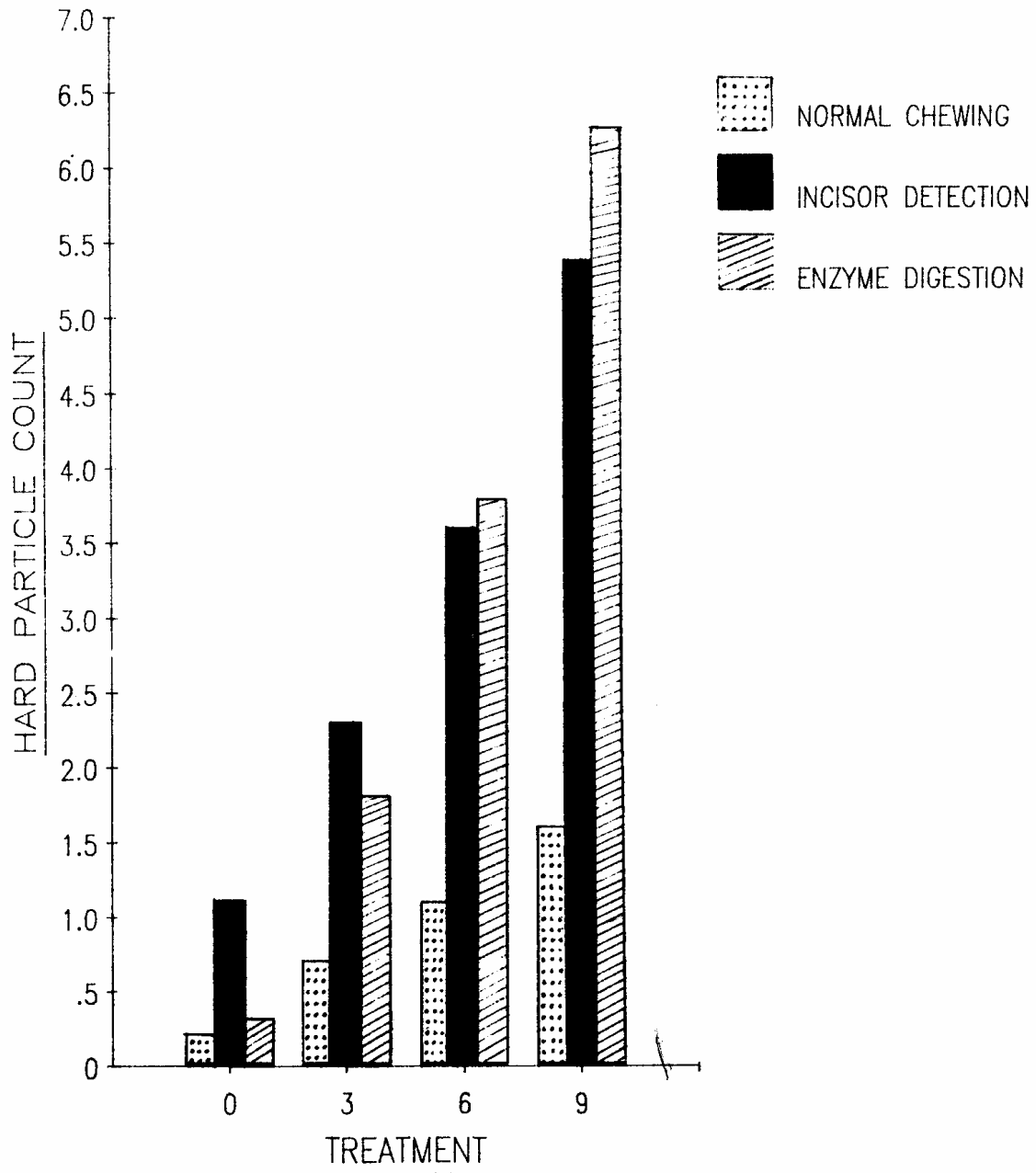


Figure 14.1. Mean values of hard particle count for normal chewing, incisor detection and enzyme digestion. Treatments are added percentage levels of connective tissue. All means for each detection method differ significantly ( $P < .05$ ).