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# THE EFFECT OF POST-MORTEM CONDITIONING ON BEEF FLAVOUR QUALITY

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Please refer to Folio 19.

#### INTRODUCTION

Meat flavour perception is dependent upon several key pre-mortem factors such as the age, sex and nutritional status of the animal as well as the final end-point cooking temperature, manner of cooking, and manner of storage. However, the most important factors in the perception of meat flavour are the flavour and texture changes that occur during the postmortem aging period (PAP) and during cooking and storage. Meat shows a significant alteration in the level of numerous chemical components such as sugar, organic acids, peptide and free amino acids, and metabolites of adenine nucleotide metabolism such as ATP during the conditioning period. Many of these changes are due to hydrolytic activity (Spanier *et al.*, 1990). These compounds serve as a pool of reactive flavour compounds and intermediates which interact to form additional flavour characteristics during cooking. It is apparent, therefore, that the development of meat flavour quality is a highly dynamic process. It is the objective of this investigation to examine the effect of post-mortem aging on beef sensory characteristics as a function of conditioning-dependent development of flavour precursor compounds and other flavour producing components.

### MATERIALS AND METHOD

### Animals

Two animals were used, #827 and #828. Both were Brangus breed types at 5/8 Angus and 3/8 Brahman. Animals were maintained for 166 days on corn-soybean meal concentrate diet consisting of 79% TDN (total digestible nutrients), 12% CP (crude protein), and 10% F (fibre). Final attributes at slaughter are tabulated below.

# Sample handling

Animals were slaughtered by traditional means, hung and the inside top round (*semimembranosus* muscle) removed within an hour. The "round" from the right hind quarter was used from each animal. After trimming of all gross fat and connective tissue the muscle was longitudinally sliced into two strips per animal muscle (Rep) and then each Rep cut crosswise into six potions (samples). Each sample was identified by appropriate notation. All samples were immediate vacuumed packaged in oxygen impermeable bags and stored in a cold room at 4°C. A sample of each Rep was removed for analysis at 0h (+45 minutes), 4-hours, 2-days, 7-days, and 14-days. Sample collection within each Rep was randomly determined prior to sampling. Temperature and pH of each sample were determined at the times indicated.

# Burger preparation

Each sample of each Rep was prepared by grinding of the beef with two passes through a 1.0cm hole grinding disc followed by two additional passes through a disk with 0.75cm holes (General Slicer/grinder, Model MC-100). The ground meat was portioned into patties weighing 85.0g± 0.02. The samples of each Rep were frozen until presentation to sample. Cooking of each sample was on a Farberware grill for seven minutes on each side yielding burgers having an appearance of medium to medium/well done.

### Thiobarbituric acid reactive substances (TBARS)

TBARS were used to indicate the degree of lipid oxidation and rancidity and were measured by the distillation procedure of Tarladgis *et al.* (1960). The reddish/pinkish chromogen formed by the assay was measured at 532nm and served as an indication of lipid oxidation and thus indirectly the food flavour quality.

# Gas chromatographic (GC) analysis of flavour volatiles

A packed-column GC procedure (Dupuy *et al.*, 1987) was utilized and consisted of a modification of the procedure originally developed for foods other than meat by Dupuy *et al.* (1978). The column was packed with a Tenax Thermostable polymer (2,6-diphenyl-p-phenylene oxide, 60-80 mesh coated with 7% poly-m-phenoxylene). Analysis of the food flavour volatiles of the beef patties was performed using an MT-220 gas chromatograph (Tracor, Austin, Texas) with dual independent hydrogen flame detectors; data were collected using an MT22 Westronics recorder and an Hewlett-Packard 3357 automated data system.

### Electrophoresis

Sodium dodecyl sulfate (SDS) polyacrylamide gel electrophoresis PAGE) was performed using a Pharmacia<sup>tm</sup> PhastSystem electrophoresis unit with precast gradient gels (8-25% with a 4% stacking gel). Electrophoresis of solubilized (Spanier and Bird, 1982) low-speed beef extracts (30.000xg) were performed following the manufacturer's directions. Capillary Electrophoresis was performed on a BioRad HPE-100<sup>tm</sup> high performance capillary electrophoresis unit. Gels were monitored at 200nm with an eight second pre-load at 8kV and a 10 minute run at 8kV (constant voltage) with amperage averaging 13.4 $\mu$ A. The running buffer was 0.1mM phosphate buffer at pH2.5 and all samples were diluted with the same buffer but at 0.01mM. The chart set at 0.05AUFS and run at 1cm/minute. The column was 20cm x 25 $\mu$ m coated capillary.

#### Sensory analysis

Descriptive sensory profiles of beef patties were generated by the Spectrum methods described by Johnsen and Civille (1986). Sensory attributes used included the following (modified from Johnsen and Civille, 1986):

salty (STY): taste on the tongue associated with sodium ions.

cooked beef (BEF): the aromatics commonly found associated with matured cooked beef muscle products and found in the broth of boiled beef.

brothy (BRO): the aromatics associated with the drippings from roasted meat which is characteristic of all meat, i.e., poultry, beef, pork, etc.

painty (PTY): the aromatic associated with rancid oil and fat (distinctly like linseed oil).

serum (SER): the aromatic associated with raw beef lean.

browned/caramel (BRC): the aromatic associated with the outside of grilled or broiled beef (seared but not blackened/burnt).

cooked liver (CKL): the aromatic associated with the cooked organ meat liver.

cardboard (CBD): the aromatic associated with slightly stale beef (refrigerated for a few days only) and associated with wet cardboard and stale oils and fats.

sour (SOU): taste on the tongue associated with acids.

sweet (SWT): taste on the tongue associated with sugars, and

bitter (BTR): the taste on the tongue associated with bitter agents such as caffeine, quinine, etc.

The panel consisted of 12 staff members from the Southern Regional Research Center who were trained in descriptive sensory analysis using a 15 point universal scale as that described by Meilgaard *et al.* (1987). All panellist were fully trained and their proficiency statistically tested by Love (1988) before their data were used in experiments; the majority of the panellist had served on the meat sensory panel for over two years.

#### Statistical analysis

The PC version of SAS (1985) was used to perform all statistical analysis. Panel averages per REP were analyzed after statistical removal of outliers from the raw data. Principal components solutions were obtained by factor analysis to

examine the relationship among sensory, chemical and instrumental attributes. Unrotated factor scores from this analysis were then averaged for each experimental combination (defined by the design matrix) and bivariate plots were produced (see Figure 4).

# RESULTS AND DISCUSSION

Data presented in figure 1A demonstrate that during the post-mortem aging period there is a gradual decline in the four desirable flavour descriptors. Beefy (BEF), brothy (BRO), browned/caramel (BRC) and sweet (SWT), show a 2<sup>nd</sup> order rate of decline in intensity of -0.13, -0.05, -0.10, and -0.05 intensity units/day respectively while the undesirable flavours, painty (PTY) and cardboard (CBD), bitter (BTR) and sour (SOU) show a moderate rate of increase in their flavour intensity of 0.04, 0.07, 0.06, and 0.08 respectively. Correlation coefficients in all cases were >0.9.

TBARS (thibarbituric acid reactive substances), hexanal and total volatiles were used as indicators of lipid oxidation and, therefore, rancidity development in the meat. No significant change in the products of lipid oxidation was seen (Figure 1B) based on the overlap of the standard error bars; the lack of appreciable change in these markers of lipid oxidation and rancidity development is not surprising since the model used was one of post-mortem aging and not one of warmed-over flavour.

The typical drop in intramuscular pH from 6.5 to 5.4 at two days post-mortem was seen (Figure 1C). Between two and 14 days post-mortem the pH remained stable averaging pH5.4 during this time period. Carcass (sample) temperature, like pH, dropped to its lowest level by two days of post-mortem conditioning where it remained until the completion of the experiment (Figure 1C).

A correlation matrix (not shown) compares the sensory attributes over the range of treatments (days of post-mortem aging). The statistical evaluation indicated a very strong negative correlation between the bitter taste (BTR) and the more desirable flavours of beefy (BEF), brothy (BRO), browned/caramel (BRC) and sweet (SWT). On the other hand, the taste of sour (SOU) only gave a strong negative correlation to brown/caramel (BRC). The undesirable aromatic flavour cardboard (CBD) had a strong negative correlation with both beefy (BEF) and brown/caramel (BRC). All four desirable flavour descriptors, i.e., BEF, BRO, BRC and SWT, had a strong positive correlation to each other.

TBARS and hexanal and total volatiles which are normally strongly correlated to each other in experimental models examining warmed-over flavour development (St. Angelo *et al.*, 1988), clearly do not show as strong correlation to each other in this post-mortem aging model. Furthermore, these off flavour/rancidity markers, which normally show strong correlation to the undesirable flavours of PTY and CBD in WOF model systems, show no correlation with any sensory attribute in this non-WOF model.

Analysis of the changes in protein composition in aging beef (*semimembranosus* muscle) show several peaks that change during the post-mortem aging process. Sodium dodecyl sulfate (SDS) polyacrylamide gel electrophoresis (PAGE) of low speed (30,000xg) extracts from beef indicate that a protein of approximately 10,000 molecular weight reaches its peak concentration by four days post-mortem (Figure 2). Different protein peaks, as determined by capillary electrophoresis (CE) are distinguished by their distinct electrophoretic mobility. Quantisation of the area under each electronic peak shows that CE-3 (peak #3) is depleted during the post-mortem aging process. A major change in protein profile is seen between four and seven days. Proteolysis of large peptides or proteins would result in compositional/conformational change, i.e., a protease could make a series of `nicks' in CE-3 creating other fragments such as Ce-4, CE-6, CE-7 and CE-11 (Figure 3).

Application of multivariate analysis of the data by principal components solutions yield a bivariate plot of variable factors (Figure 4). In this empirical summary of the pattern of intercorrelations among variables, we can see that the data for the flavour attributes have positioned themselves into different regions of the grid. For example, the desirable flavours SWT, BRC, BEF and BRO all cluster in the upper left of the plot, while the undesirable flavours, such as CBD, PTY, BTR and SOU cluster in the lower right of the grid. Chemical attributes, such as the TBARS, the hexanal and total volatiles, which seem to have no effect or correlation with the sensory attributes, all cluster near the centre of the

grid, or at the X and Y intercept. Further examination of these data reveals that freshly slaughtered and meat aged four hours cluster in the region of the desirable flavours, while meat aged seven or 14 days clusters with the undesirable flavours. The two and four day aged meat clustered in a central position of the grid somewhere midway between the desirable and undesirable flavours. While these data indicate that a trained sensory panel can accurately assess the flavour of meat, we feel that it would be unlikely that the consumer would be able to clearly distinguish these subtle, yet significant, changes. On the other hand, these changes in flavour, no matter how subtle, are important to recognize and examine from the point of view of understanding the potential flavour impact of the different flavour precursor compounds generated during the post-mortem conditioning period.

### CONCLUSIONS

The data seem to suggest that the best overall flavour quality of aged meat, i.e., meat with optimum flavour (and possibly texture), is found in meat aged for up to four days post-mortem. Lipid oxidation, which can cause the production of free radicals which in turn can alter flavour through secondary reactions with flavour constituents (e.g., fragmentation of proteins, cross-linking of proteins, peptides, lipids, etc.), appears to be of little importance in the post-mortem aging process. On the other hand, the activity of various hydrolases, such as calcium-dependent proteinases implicated in meat tenderization (Koomaharie *et al.*, 1988) and the cathepins implicated in the production of flavour peptides (Spanier *et al.*, 1990; Spanier and Miller, 1993), may play an important temporal role in the generation of the flavour of meat. Methods, such as multivariate analysis of chemical attributes, flavour attributes and experimental treatments, is a valuable tool permitting us to map the flavour of foods (Spanier *et al.*, 1992).

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Table 1. Attributes at slaughter of study animals.

	Animal #827	Animal #828
Hot carcass wt, kg	381.4	367.3
Marbling score, ABI	slight <sup>55</sup>	slight <sup>20</sup>
USDA quality grade	Select +	Choice -
Backfat, mm	8.9	14.0
Ribeye area, cm <sup>2</sup>	76.13	77.42
Yield grade	3.1	3.7