

Project Title:	Using metabolomics to predict and guarantee beef flavor to the consumer
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Background

Consumers make decisions based on visual appraisal, package labeling, historical experience, and branding. Package labeling can include cut name and carcass grade. The addition of new, objective package information is the only opportunity to provide non-biased, factual descriptors of flavor prior to purchase. Several researchers have investigated the relationships between trained sensory panels, consumer sensory panels, and volatile beef flavor compounds but the results have not been consistent. With no two studies leading to a consensus of traits that can be used to predict positive beef flavors we propose using metabolomics to develop signatures 48 hours postmortem, simulating the time of grading, for predicting flavor in aged, cooked beef, and profiles at 26 days postmortem for guarantees of these flavors in cooked beef. The objective of this study was to compare metabolic signatures of beef steaks from different USDA quality grades to predict desirable beef flavor.

Methodology

Ninety, A maturity beef carcasses were selected at the time of grading at a commercial facility. Carcasses will be chosen by marbling score, Slight (n = 30), Small (n = 30), Modest and Moderate (n = 30), to product a Quality Grade Selection Category. Carcasses were graded by a USDA-AMS Grading Supervisor. Carcass data means by Quality Grade Selection Category are shown in Table 1.

One striploin from each carcass was purchased and transported to the University of Missouri for sample cutting and aging. The boneless striploins were fabricated into one-inch steaks for the following samples: one steak at the time of fabrication for metabolomics, seven to eight steaks (consideration will be given to the size of the longissimus muscle and when additional muscles appear in the subprimal) after 26 days of aging for metabolomics, volatiles, trained and consumer panels. Individual steaks were aged in vacuum packaged bags. The steak removed at fabrication was treated as a raw steak to simulate the way all meat is handled prior to the consumer and generate metabolomics signatures for predicting meat flavor when beef is consumed. After aging, steaks were cooked on a clamshell grill.

Metabolomics beef samples were incubated with methanol in glass vials. The vials were then placed on an orbital shaker for 24 hours at room temperature with constant shaking. After centrifugation, the supernatant was recovered and an internal standard, ribitol, is added. The samples were then dried and derivatized with methoxyamine and MSTFA+1% TMCS and subjected to GC-MS analysis using an Agilent 6890 GC coupled to a 5973 MSD. Metabolites were separated on a 60 m DB-5MS column and analyzed using MSD with a scan range of m/z 30 to 650. Data were deconvoluted using AMDIS (Automated Mass spectral Deconvolution and Identification System). Volatile analyses was performed using solid-phase microextraction (SPME) and gas chromatography-mass spectrometry (GC-MS) as previously described by Watanabe et al. (2015). Samples were first placed into headspace sample vials containing an internal standard (4-methyl-2-pentanone) and sealed tightly. Volatiles were then extracted from the headspace using a divinyl benzene/carboxen/polydimethylsiloxane (DVB/CAR/PDMS) fiber 50/30 μm (Supelco,



Bellefonte, PA, USA) using a GERSTEL MPS autosampler. The volatiles were then de-absorbed from the SPME fiber and analyzed using Agilent 7200B GC-TOF MS. A descriptive attribute panel for flavor comprising of 6-8 trained panelists was oriented with the samples before the blind evaluations. A 0 to 15-point scale with 0.5-point increments was used to score and measure the responses. A panel of 120 beef consumers were recruited to determine the acceptability/palatability of the samples. Liking of appearance, flavor, texture and overall liking was determined using a 9-point Hedonic scale (1 – dislike extremely to 9- like extremely).

Findings

The carcass characteristics for this study are not surprising since the carcasses were selected to a fill specific categories. It should be noted that of the carcasses in the Upper Choice category only one carcass has a Moderate marbling score with the rest being Modest. In general, volatiles in raw samples increased with aging from 2 to 26 days (Figures 1 and 2). More interesting is the fact that specific polar metabolites, sugars and amino acids, increased with aging (data not presented in tabular form). This trend should be explored further focusing on the polar metabolites on day 2 or at the time of grading. Partial least squares discriminant analysis of metabolites was able to differentiate between quality grade categories between Choice and Select in cooked meat. However, there was overlap between the two categories of Choice which could be due lack of carcasses with Moderate marbling score in the population.

Trained sensory panel rated Upper Choice slightly superior, numerically, for many of the desirable flavor attributes but these differences were not statistically difference. In addition, the same trend was found with consumer sensory panel. Another trend that was noted was greater variation in both the flavor attributes and consumer liking scores for Select steaks.

Implications

The goal of this project to use metabolomics as a tool to segregate beef carcasses. Specific metabolites from sugars and amino acids increased with aging time from 2 to 26 days postmortem in raw beef samples. Metabolites from cooked beef samples were able to segregate based on Choice and Select grades. Further research is needed to determine if the metabolites from sugars and amino acids could be used as markers for segregation.

Tables/Figures

Table 1. Carcass data mean (SD) by Quality Grade Selection Category ^a.

Carcass Characteristic	Select	Lower Choice	Upper Choice
Marbling Score ^a	352.7 (21.6)	451.7 (18.8)	538.0 (29.2)
USDA Quality Grade ^b	252.7 (21.6)	317.1 (6.2)	345.5 (9.6)
Adjusted Fat Thickness (in)	0.53 (0.17)	0.56 (0.13)	0.71 (0.11)
Hot Carcass Weight (lbs)	858.6 (88.6)	805.4 (62.3)	863.4 (63.7)
Ribeye Area (in ²)	14.2 (1.2)	13.4 (1.1)	13.3 (1.3)
USDA Yield Grade ^c	2.5 (0.7)	2.6 (0.7)	3.2 (0.5)

^a300 = Slight, 400 = Small, 500 = Moderate, 600 = Modest

^b200 = Select, 300 = Choice; All carcasses were A maturity

^c2.0% was used at the kidney, pelvic, and heart fat based on the 2016 National Beef Quality Audit averages

Figure 1. Two-dimensional score plots of Principal Component Analysis (PCA) (A) and Partial Least Squares-Discriminant Analysis (PLSDA) (B) with all 6 groups on raw beef analyzed together.

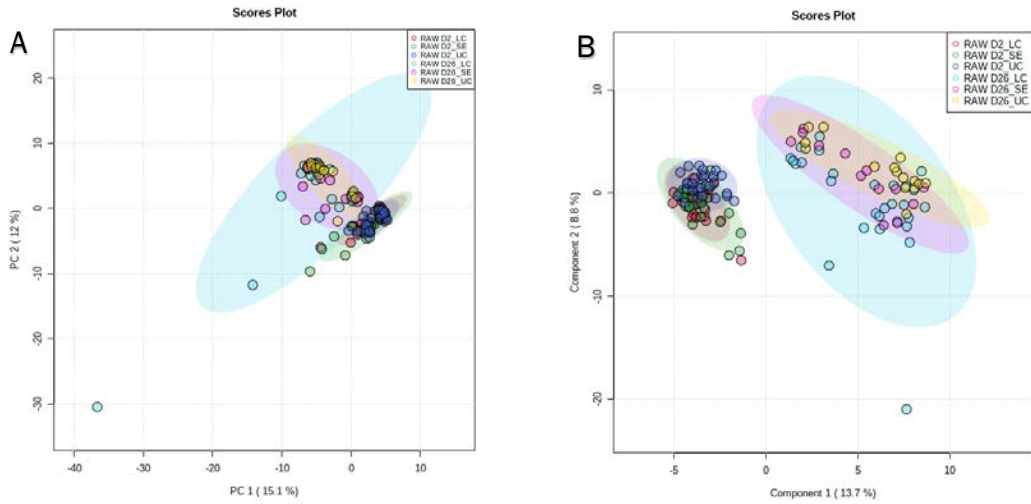


Figure 2. Box plot presentation of selected significant metabolites represented by red dots in ANOVA, displaying their fold change of instrument response across all 6 groups of raw beef samples.

