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Evaluation of 4-Methyloctanoic Acid Compound in Goat Meat

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Abstract

In this experiment, 4-methyloctanoic acid concentrations were measured in kidney and body fat from goats harvested at 9 months of age. For instrumental analysis, samples were extracted by using the saponification technique to obtain free fatty acids. Then, Gas Chromatography-Mass Spectrometry (GC-MS) was used after direct saponification with KOH/methanol and derivatization with N, O-Bis Trimethylsilyltri Fluoroacet Amide (BSTFA). In addition to the chemical analyses, 4-methyloctanoic acid was added at ratios of 0, 0.02, 0.05, 0.07, 0.10, 0.20, and 0.5 ml to goat meat and evaluated through sensory testing, by 13 trained-panelists using the descriptive method with 8 scales (8 = desirable, 7 = moderately desirable, 6 = slightly desirable, 5 = neither desirable nor dislike, 4= dislike slightly, 3 = dislike moderately, 2 = dislike very much, 1= undesirable).

Results showed that the 4-methyloctanoic acid concentration was found at 0.0005 mg/ml of kidney fat and 0.0003 mg/ml in body fat of goats harvested at 9 months. For sensory testing, most panelists can detect the 4-methyloctanoic acid concentration at level of 0.05 ml or higher levels. Therefore, the result of this project was provided value to improve the goat flavor in meat industry.

Keywords: Goat meat; 4-methyloctanoic Acid; Descriptive method

Introduction

Flavor is an important characteristic that influences consumer acceptability of goat meat [1,2]. Goat meat is being gradually introduced to consumers in the U.S. due to its lower fat and higher unsaturated fatty acid content than other red meats [3,4]. The 4-methyloctanoic acid is a medium chain-length (C_6-C_9) unsaturated fatty acid and is speculated to provide a highly characteristic odor in goat meat [5-7]. Previous studies demonstrated that breed, gender, age, type of tissue and diet influence the 4-methyloctanoic acid concentration in the fat tissue [8,9]. During chilled meat storage, oxygen is a direct catalyst in the reaction that causes the lipid oxidation to occur. Lipid oxidation can produce undesirable rancidity causing rejection by the consumer [10]. Deterioration of meat odor and taste is directly related to changes in fatty acids [11]. Therefore, the aim of this study was to determine the 4-methyloctanoic acid compound of goat meat. This approach was contributed to acquiring a deeper knowledge of goat meat in terms of their flavor characteristics.

Materials and Methods

Physicochemical analysis

Sample Preparation: Subcutaneous fat samples (30 g) were collected from shoulder area at 24 h post slaughter and frozen at -20°C until needed. Prior to analysis, the surface layer of the fat was removed and the remainder cut into small portions. Adipose tissues were minced (particle diameter of 6 mm) with an electrical grinder, and manually homogenized. Each treatment was analyzed in duplicate and was stored at approximately 4°C until used.

Chemical and gases: 4-methyloctanoic acid was purchased from Sigma-Aldrich (WI) and was of > 98% purity. Nitrogen and helium were ultra-high purity grade (Linde, Altona). All other reagents were of analytical reagent grade.

Extraction procedure: Duplicate 5 g quantities from each sample were accurately weighed out into 50 ml conical centrifuge tubes and 5 mg of the internal standard (IS: 4-methyloctanoic acid) was added before saponification. Then, 6 ml of saponification solution (5M KOH in methanol: water (50:50, v/v)) was added. Tubes were flushed with N_2, sealed and shaken for 10 min prior to placing in a 60°C water bath for 60 min for direct saponification. After that, the reaction mixture was diluted with 12 ml of 0.5% NaCl and 5 ml of petroleum ether. Samples were vortexed for 5

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Table 1: The concentration of 4-methyloctanoic acid from goat kidney and body fats harvested at 9 months.

Goat sample	4-methyloctanoic acid compound (mg/ml)
Kidney fat	0.0005
Body fat	0.0003
SEM	0.00048

Results showed that the 4-methyloctanoic acid concentration was found at 0.0005 mg/ml of kidney fat and 0.0003 mg/ml in body fat of goats harvested at 9 months.

min, 5 drops of absolute ethanol were added and then centrifuged at 1000 rpm for 5 min for layer separation. The top layer (containing the non-saponifiable extract, i.e. cholesterol) was removed and discarded. Then, 3 ml of glacial acetic acid was added to neutralize the KOH fraction and 5 ml of petroleum ether was added and vortexed for 10 min. This step also resulted in protonation of the Fatty Acid (FA) carboxyl groups. Samples were centrifuged again at 1000 rpm for 5 min. The top layer of each test tube was transferred to a clean screw-cap glass tube to which 100 µl of 2, 2-dimethoxypropane was added and then vortexed for 2 min.

Derivatization procedure: Samples were reduced to dryness under N_2 at 40°C and re-dissolved in 1 ml of methanol: toluene (2:1 vol.) solution and vortexed for 5 min. Methanol is a catalyst for the BSTFA reaction and drives the reaction in favor of methyl ester formation. At this stage, 100 µl of methylation reagent BSTFA was added. The solvents were removed from the samples by using a gentle stream of N_2 at 60°C for 20 min to obtain methyl ester. For gas chromatography, 1 µl of methyl ester sample was injected onto the column and run under an optimized temperature program with optimized gas flow rate.

Gas chromatography-mass spectrometry analysis: The fatty acid-TMS esters were separated by injecting (1 µl) onto a HP-5MS 5% Phenyl Methyl Siloxane column (30 m x 0.25 mm i.d. x 250 µm film thickness) in a Varian 3400 Gas Chromatograph (GC) and detected by a Saturn 2000 ion trap mass spectrometer operating in full scan mode. The septumless programmable GC-MS was programmed starting at 70°C and increased to 280°C. The GC oven was held at 70°C for 1 min then increased to 280°C at 15C min-1 and held at this temperature for 2 min. Helium was used as the carrier gas at a constant pressure of 105 kPa. The mass spectrometer transfer line was at 280°C. Mass spectra were acquired using an ion source temperature of 220°C and an electron multiplier voltage of 2400 V. The mass spectrometer was calibrated using FC43 (Varian, Inc., Springvale).

Descriptive sensory analysis

The sensory analysis was performed by 13 trained panelists, with previous experience in the sensory evaluation of 4-methyloctanoic acid compound in goat meat. Samples were minced and served in disposable, odor-free, plastic plates, covered with a plastic wrap and allowed to reach room temperature before serving. Each panelist was given two samples per session, chosen at random. Goat meat samples were identified by 3-character codes (587, 710, 309, 972, 625, 427, and 168). All the samples (25 g/sample) were tested twice in two different sessions.

Eight flavor attributes 8 scales (8 = desirable, 7 = moderately desirable, 6 = slightly desirable, 5 = neither desirable nor dislike, 4= dislike slightly, 3 = dislike moderately, 2 = dislike very much, 1= undesirable) were evaluated by thirteen trained panelists from McNeese State University. The Post Hoc and Dunnett t-tests of SPSS

Table 2: Trained panel scores for goat samples with or without added 4-methyloctanoic acid.

Descriptive testing (N = 13 trained panelists)			
4-methyloctanoic acid concentration (ml)	Sample code	Mean score	SEM
0.00 (control)	587	5.2308	0.6218
0.02	710	3.9231	0.8123
0.05	309	3.6923	0.6241
0.07	972	3.6154	0.6154
0.1	625	4.1538	0.5412
0.2	427	3.3077	0.5925
0.5	168	3.2308	0.5679

window were used to evaluate the significance of differences of the obtained dat. All data are presented as means with Standard Error of Mean (SEM) and a significance level of $P<0.05$ was used of statistical analysis of means from treatments.

Results and Discussion

Physicochemical analysis

The results of this study showed that the concentration of 4-methyloctanoic acid compound was found at 0.0005 mg/ml of kidney fat and 0.0003 mg/ml in body fat of goats harvested at 9 months. Another conclusion was that kidney fat contained the highest concentration of hircinoic acid, as compared to back and side-fat tissue. This conclusion is consistent with prior research suggesting that OBCFA synthesis carried out by ruminal bacteria starts when the ruminant function first develops [12,13], which would lead to the logical conclusion that the organs/tissue developed first would contain the most hircinoic acid. Further research is appropriate. Demonstrated that breed, gender and age influence the hircinoic acid concentration in the subcutaneous fat tissue. Another conclusion of this experiment is that no significant differences based on gender were detected in the types of fat tissue studied, suggesting that conclusions in prior research cannot be generalized to all fat tissue. Further research is appropriate.

Descriptive sensory analysis

For descriptive testing, the 0.5 ml of 4-methyloctanoic acid was rated the least desirable flavor at score of 3.2308. Most panelists preferred the control (without 4-methyloctanoic acid concentration) treatment at score of 5.2308 (Table 2).

Conclusions

The results of this experiment were provided value to improve the goat flavor in meat industry. Specifically, participants rated six treatments similarly with 0.02-0.5 ml of 4-methyloctanoic acid concentration in minced goat samples. Therefore, these data can help the meat producer to find the proper time to harvest goat which decrease the goaty flavors (4-methyloctanoic acid).

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