# INHIBITION OF HETEROCYCLIC AMINE FORMATION IN GROUND BEEF

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### **Summary**

The natural antioxidant carnosine, moisture retention by covered cooking, and low temperature cooking were evaluated as ways to inhibit heterocyclic amine (HCA) formation in fried ground beef. Samples were fried at 375EF for 5 min/side, 300EF for 5 min/side, or 250EF for 8 min/side, with surface browning enhanced by applying a caramel solution (Maillose®) near the end of cooking times. Analysis for HCAs was performed on both the crust and the whole patties. Carnosine reduced 4,8-DiMelQx, a major HCA, to below its detection limit (.31 ng/g). HCAs were reduced when the cooking temperature was lowered from 375 to 300 or 250EF even with caramel applied on the surface. Cooking in a covered pan reduced levels of most HCAs but less than carnosine addition.

(Key Words: Ground Beef, Heterocyclic Amine, Inhibition.)

#### Introduction

Meat cooked at high temperature may contain mutagens and animal carcinogens called heterocyclic amines (HCAs). To inhibit the formation of these compounds, naturally occurring antioxidants can be added to the meat before cooking. Carnosine, a Beta-alanine-histidine-containing dipeptide present in skeletal muscle, may be effective through a combination of free radical scavenging and metal chelation. Thus, we added carnosine to meat samples to detect its effect in reducing HCA formation. Water movement during cooking may carry HCA precursors from inner portions of the meat patties to outer surfaces. Consequently, we studied

minimizing water loss and consequent movement by covered cooking. Lower cooking temperatures may reduce levels of HCAs formed, so we also studied effects of longer cooking at lower temperatures.

## **Experimental Procedures**

Raw ground beef from eye round steaks (2.9% fat) was formed into 100 g patties, 1.5 cm (.59 in.) thick and 10 cm (3.9 in.) in diameter. Meat patties for control (no additive). carnosine added (1.5%), and moisture retention (covered cooking) treatments were fried in a thermostat-controlled Teflon-coated frying pan at 375EF for 5 min/side. For low cooking temperatures, meat samples were fried at 300EF for 5 min/side or 250EF for 8 min/side. Surface browning was enhanced by applying caramel solution (Maillose®) near the end of cooking times. The internal temperature was recorded by inserting a probe thermocouple into the center of the patty at a 45E angle. Final internal temperatures were 160EF for all treatments.

Solid phase extraction was followed by high pressure liquid chromatography (HPLC) with a Hewlett-Packard 1090 A, series II HPLC system. A photodiode array ultraviolet detector and a fluorescence detector were used to monitor the separations. A TSKgel ODS80 (TosoHaas, Montgomeryville, PA), 25 cm × 4.6 mm I.D. (5 Fm particle size) column protected by a Supelguard ODS-80<sup>TM</sup> (TosoHaas) precolumn was used for separation of HCAs.

The mobile phase consisted of three solvents: solvent A, .01 M triethylamine (pH 3.2); solvent B, .01 M triethylamine (pH

3.6); solvent C, acetonitrile. The gradient profile was linear, and the program was 0-10 min, 5-15% C in A; 10-10.1 min, exchange of A with B; 10.1-20 min, 15-25% C in B; 20-30 min, 25-55% C in B, followed by 15 min for column equilibration.

Abbreviations for reporting HCAs are as follows:

IQ=2-amino-3-methylimidazo [4,5-f]quinoline;

MelQ=2-amino-3,4-dimethylimidazo [4,5-f]quinoline;

MelQx=2-amino-3,8-dimethylimidazo [4,5-*f*]quinoxaline;

4,8-DiMelQx= 2-amino-3,4,8-trimethyl-imidaxo[4,5-*f*]quinoxaline;

PhIP=2-amino-methyl-6-phenyl-imidazo [4,5-*b*]pyridine;

harman=1-methyl-9H-pyrido[3,4-*b*];

norharman=9H-pyrido[3,4-b]-indol.

#### **Results and Discussion**

Adding carnosine lowered levels of HCAs formed on the meat patty crust. Both MelQ and 4,8 DiMelQx (specific HCAs) were reduced below their corresponding detection limits (.28 ng for MelQ and .31 ng

for 4,8-DiMelQx). The highest reduction in MelQx (43.00%) was with added carnosine as contrasted to the controls. However, carnosine increased PhIP 60% compared to the controls. This increase probably was due to alanine in the carnosine molecule, which may contribute to PhIP formation.

When moisture was retained in the meat patties by covering the cooking pan, the effect on HCAs was variable, some were reduced, MelQ (to nondetectable), DiMelQx (57%), harman(12%), and norharman (30%), whereas some were increased; MelQx (7.0%) and PhIP (9.0%). The final internal temperature of the moisture retained patties (181EF) was higher than the control (173EF), which might account for the increase in MelQx and PhIP.

Caramel (Maillose) added near the end of cooking increased HCAs in patties cooked at 300 or 250EF, but levels were still lower than those in patties cooked at 375EF.