SODIUM CHLORIDE AFFECTS ACTIVITY OF ACID PHOSPHATASE AND PYRUVATE KINASE, WATER-EXTRACTABLE PROTEINS OF DUCK MEAT HEATED TO SPECIFIC ENDPOINT TEMPERATURES

James Chun-Chin Kuo and Chun-Lin Huang
Department of Food Science, Tunghai University, Taichung, Taiwan.
Key words: endpoint temperature, sodium chloride, duck meat, acid phosphatase, pyruvate kinase, water-extractable proteins

Background

Inadequately cooked meat products are most commonly in food-borne disease outbreaks.Wang et al. (1996) reported that acid phosphatase might be useful in assays to verify processing temperatures of ground beef. Davis et al. (1988) found pyruvate kinase activity could be used as an indicator of temperature attained during cooking of cured pork. Huang and Kuo (2000) reported that pork loin added with 1.8% sodium chloride and /or 0.012% sodium nitrite had lower ($p<0.05$) water-extractable biuret-positive ratio values than the control. Little information has been published concerning these methods used in duck muscle and the influence of sodium chloride on the residual enzyme activity and the water-extractable proteins in heat processed duck breast meat product.

Objective

Our objective was to determine the influence of sodium chloride on residual acid phosphatase and pyruvate kinase activity and water-extractable biuret-positive ratio values of duck breast meat after heat treatments to various endpoint temperatures.

Methods

Acid phosphatase (ACP) activity of duck breast meat extracts was measured using a phosphatase diagnostic kit (No. 104, Sigma) at 37°C. The method reported by Davis et al. (1988) was used to extract pyruvate kinase in meat samples, however, Tris-HCl buffer (0.02 M, pH 7.4) suggested by Bogin et al. (1992) was used to replace deionized distilled water. And the method described by Bernofsky (1980) was used to determine pyruvate kinase activity in meat extracts. The ratio of water-extractable, biuret-positive proteins from heated duck muscle to the base value at 70°C for 15 min was determined according to Davis et al. (1985).

Results and discussion

Acid phosphatase was reported to be a potential end-point temperature indicator in pork (USDA-FSIS, 1986; Kormendy et al., 1992) and in poultry meat (Davis and Townsend, 1994). Acid phosphatase activity in raw meat (0, 1.6, 2.0 and 2.4% sodium chloride) was 625-650 U/Kg meat (Sigma Units) and decreased to 10 U/Kg meat or less (1.1-1.6% of original activity) in duck meat heated to 69°C (Table 1). Activity decreased as endpoint temperature of duck meat increased. Differences in activity between meat cooked to 65 and 67°C, as well as 67 and 69°C were significant ($p<0.05$). Differences in activity between the heat treatments of 69 and 73°C were not different ($p>0.05$). Wang et al. (1996) reported that acid phosphatase activity in raw meat was 603 U/Kg and decreased to 7.9 U/Kg meat in beef patties heated to 71°C. Acid phosphatase activity in duck meat added sodium chloride (1.6, 2.0 and 2.4%) was lower ($p<0.05$) than that in meats added no sodium chloride at 65°C; however, activity in duck meat heated to 67 to 73°C was not affected ($p>0.05$) by levels of sodium chloride. The cooking time to reach 65°C probably increased in the treatments added sodium chloride, and the residual acid phosphatase activity decreased. However, the differences in activity were diminished as the endpoint temperatures reached to 67°C or higher.

Pyruvate kinase was reported to be an indicator of temperature attained during cooking cured pork (Davis et al., 1988). Pyruvate kinase activity was 172-189 U/Kg meat (Sigma Units) in raw duck breast meat with the addition of 0, 1.6, 2.0 and 2.4% sodium chloride, which decreased to 86-119 U/Kg meat in samples heated to 65°C (Table 2). Enzyme activity decreased as endpoint temperature of duck meat increased from 65 to 73°C. Enzyme activity in duck meat was less than 16 U/Kg meat (7.9-8.7% of original activity) at 69-73°C. Significant differences in activity between the control (0%) and the treatments (1.6, 2.0 and 2.4%) were observed at 65°C. But at higher endpoint temperatures (67-73°C), pyruvate kinase activity was not affected ($p>0.05$) by the addition of sodium chloride. This phenomenon was also found in acid phospatase activity (Table 1). We found that pyruvate kinase was still active when duck meat heated to 73°C; however, Davis et al. (1988) reported that no pyruvate kinase activity was detected in cured pork product heated to 69.9°C.

Table 3 shows water-extractable, biuret-positive ratio value of duck meat. A rapid decrease ($p<0.05$) of water-extractable protein ratios was observed between the raw meat and the cooked meat. Water-extractable biuret-positive ratio values decreased as endpoint temperatures of duck meat increased from 65 to 73°C was observed in duck meat (with the addition of 0, 1.6, 2.0 and 2.4% sodium chloride). It indicated that the endpoint temperatures increased, more proteins would be denatured or coagulated, and therefore less water-extractable, biuret-positive compounds. The ratios in duck meat heated to 65°C (1.53) was higher ($p<0.05$) than that in meats added with 1.6, 2.0 or 2.4% sodium chloride (1.35, 1.27 and 1.23, respectively). This probably due to that the addition of sodium chloride to duck meat would result in loss of solubility of sarcoplasmic proteins extracted with water. Water-extractable biuret-positive ratio values in meat with no added sodium chloride were slightly higher than those in treatments with sodium chloride (1.6, 2.0 or 2.4%) in all five heat treatments (except the differences in the heat treatment of 65°C were significant). This is due to that sodium chloride would increase salt soluble proteins and increase the cohesiveness of meat, therefore more heating time was needed to reach specific endpoint temperature, and more proteins were denatured. The cooking time to reach a given endpoint temperature probably increased with increasing sodium chloride content. Results suggested that water-soluble biuret-positive ratio might be useful in assays to verify processing temperature of cooked duck breast meat.

Conclusions

Acid phosphatase and pyruvate kinase activity decreased as end internal temperature increased. None of the enzymes was completely inactivated at 73°C. Water-extractable biuret-positive ratio and the activities of acid phosphatase and pyruvate kinase might be useful in assays to verify processing temperature of duck meat (such as peking duck and spicy water duck). In our results, we found that levels of sodium chloride content significantly affected the water-extractable biuret-positive ratio values and the activity of acid phosphatase and pyruvate kinase of duck meat heated to 65°C or less. The water-extractable biuret-positive ratios and the enzyme activities were not affected by sodium chloride when duck meat heated to 67-73°C.

References

Bernofsky, C. (1980). Affinity chromatography of NAD. In D. B. McCormick, and L. D.Wright. *Methods in Enzymology* (Vol. 66, pp. 40-45). New York: Academic Press, Inc.

Bogin, E., Isreali, B. A., & Klinger, I. (1992). Evaluation of heat treatment of turkey breast meat by biochemical methods. *Journal of Food Protection*, 55, 787-791.

Cohen, E. H. (1969). Determination of acid phosphatase activity in canned hams as an indicator of temperature attained during cooking. *Food Technology*, 23, 961-964.

Davis, C. E., & Anderson, J. B. (1983). Effect of heat on biuret-positive water-extractable porcine muscle proteins. *Journal of Food Science*, 46, 947-949.

Davis, C. E., Bracewell, A. J. Anderson, J. B. & Reagan, J. O. (1985). Time temperature heating effect on biuret-positive water-extractable porcine and bovine muscle proteins. *Journal of Food protection*, 48, 215-220.

Davis, C. E., Searcy, G. K., Blankenship, L. C., & Townsend, W. E. (1988). Pyruvate kinase activity as an indicator of temperature attained during cooking of cured pork. *Journal of Food Protection*, 51, 773-777.

Davis, C. E., & Townsend, W. E. (1994). Rapid Fluorometric Analysis of Acid Phosphatase Activity in Cooked Poultry Meat. *Journal of Food Protection*, 37, 1094-1097.

Huang A., & Kuo, C. C. (2000). Effect of end internal temperature on color and water-extractable biuret-positive proteins ratio of cured and uncured pork. *Tunghai Journal*, 41, 15-23.Kormendy, L., Zsarnoczay, G., &Mihalyi, V. (1992). A new, modified acid phosphatase assay for determining the extent of heat treatment in canned hams. *Food Chemistry*, 44, 367-375.

Wang, S. F., Abouzied, M. M., & Smith, D. M. (1996). Proteins as potential endpoint temperature indicators for ground beef patties. *Journal of Food Science*, 61, 5-7.

Table 1. Acid phosphase activity of duck breast meat as affected by sodium chloride and endpoint temperature

	Activity(U/ Kg sample)					
	Endpoint temperature(℃)					
Sodium chloride(%)	Uncooked	65	67	69	71	73
0	650^{aX}(100 %)	189^{bX}(29.1 %)*	34^{cX}(5.2 %)	9^{dX}(1.4 %)	8^{dX}(1.2 %)	8^{dX}(1.2 %)
1.6	628^{aX}(100 %)	157^{bY}(25.0 %)	31^{cX}(4.9 %)	10^{dX}(1.6 %)	9^{dX}(1.4 %)	7^{dX}(1.1 %)
2.0	629^{aX}(100 %)	159^{bY}(25.3 %)	30^{cX}(4.8 %)	10^{dX}(1.6 %)	8^{dX}(1.3 %)	7^{dX}(1.1 %)
2.4	625^{aX}(100 %)	153^{bY}(24.5 %)	30^{cX}(4.8 %)	8^{dX}(1.3 %)	7^{dX}(1.1 %)	7^{dX}(1.1 %)

$^{a-d}$Means in the same row with different letters are different ($P<0.05$).
^{X,Y}Means in the same column with different letters are different ($P<0.05$).
*Number in brackets is % residual enzymatic activity (a,b,c,d) caculated from original activity (100%, uncooked).

Table 2. Pyruvate kinase activity of duck breast meat as affected by sodium chloride and endpoint temperature

	Activity (U/ Kg sample)					
	Endpoint temperature					
Sodium chloride(%)	Uncooked	65	67	69	71	73
0	189^{aX}(100 %)	119^{bX}(62.9 %)*	30^{cX}(15.9 %)	16^{dX}(8.5 %)	16^{dX}(8.5 %)	16^{dX}(8.5 %)
1.6	172^{aX}(100 %)	94^{bY}(54.7 %)	22^{cX}(12.8 %)	15^{dX}(8.7 %)	15^{dX}(8.7 %)	14^{dX}(8.1 %)
2.0	177^{aX}(100 %)	88^{bY}(49.7 %)	23^{cX}(13.0 %)	15^{dX}(8.5 %)	15^{dX}(8.5 %)	14^{dX}(7.9 %)
2.4	175^{aX}(100 %)	86^{bY}(49.1 %)	23^{cX}(13.1 %)	14^{dX}(8.0 %)	14^{dX}(8.0 %)	14^{dX}(8.0 %)

$^{a-d}$Means with different letters within the same row are significantly different ($P<0.05$).
^{X,Y}Means with different letters (X,Y) within the same column are significantly different ($P<0.05$).
*Number in brackets is % residual enzymatic activity (a,b,c,d) caculated from original activity (100%, uncooked).

Table 3. Water-extractable biuret-positive ratio of duck breast meat as affected by sodium chloride and endpoint temperature

	Water-extractable biuret-positive ratio					
	Endpoint temperature (℃)					
Sodium chloride (%)	Uncooked	65	67	69	71	73
0	4.72^{aX}	1.53^{bX}	1.17^{cX}	1.13^{cX}	1.05^{dX}	1.03^{dX}
1.6	4.24^{aY}	1.35^{bY}	1.11^{cX}	1.09^{cX}	1.03^{cX}	1.03^{cX}
2.0	4.26^{aY}	1.27^{bZ}	1.12^{cX}	1.08^{cX}	1.02^{cX}	1.02^{cX}
2.4	4.26^{aY}	1.23^{bZ}	1.11^{cX}	1.08^{cX}	1.03^{cX}	1.02^{cX}

$^{a-d}$Means with different letters within the same row are significantly different ($P<0.05$).
^{X,Y}Means with different letters within the same column are significantly different ($P<0.05$).
*Number in brackets is % residual enzymatic activity (a,b,c,d) caculated from original activity (100%, uncooked).