한국농화학회지 Vol. 14, No. 2, Sept. 1971
J. Korean Agricultural Chemical Society

Changes in Characteristic Proteins during Chilling of Dressed broilers.

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(Received Sept. 10, 1971)

Dressed broilers의 冷藏中 特殊蛋白質 變化

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(1971. 9.10, 수리)

Summary

From both breast and leg muscle of 12 week-old broiler chicken held for aging in slushed ice and dry chilling at 33—35°F., myosin, actomyosin and other nitrogenous fractions were extracted with KCl-phosphate buffer for various periods from 1 hr. to 25 hr. post-mortem.

The changes in extractable nitrogen occurred mainly as a result of decrease in extractability of myosin and to some extent, increase in extractability of actomyosin.

Changes in stroma, sarcoplasmic and NPN fractions were small. Myosin extractability decreased rapidly during the first 3 hr. post-mortem and then reduced Continuously in both leg muscle and breast muscle during wet chilling.

The decrease of myosin extractability in leg muscle was much more than that in breast muscle, and then the extractability increased after 17 hr. post-mortem in dry chilling.

Actomyosin was extracted at low consistent level in wet chilling, while it increased considerably after 17 hr. post-mortem in dry chilling. The tendency was similar in both breast and leg muscle.

I. Introduction

Extractability of muscle proteins has been studied as an indication of changes taking place in muscle after death. In the post-mortem conversion of muscle to meat, the retention of water binding properties and the development of a pliable muscle which will be tender when corked are of prime importance. Both of these factors are dependent to a large extent on the condition of the myofibrillar proteins.

During the study of the effect of fresh and frozen storage on chicken muscle proteins (Khan et al. 1963), a wide variation in the results on protein extractability was noted in leg muscle from old birds which were aged for 24 hr. in drained crushed ice, whereas the breast muscle from the same birds gave consistent results. Since more recent work using test panel and shear press tests has shown that tenderization occurs more slowly in leg muscle than in breast

muscle (Van den Berg et al. 1964), the consistency in the results on leg muscle may be attributed to the degree of completion of post-rigor tenderization changes in leg muscle at 24 hr. post-mortem.

Khan et al. (1964) reported that nitrogen extractability from chicken breast muscle decreased to a minimum at 4—5 hr. post-mortem, then increased to a maximum at 1—1½ days. They also reported that the changes in extractability were due mostly to changes in the myofibrillar proteins. but the nature of the extracted myofibrillar protein was not examined. Weinberg et al. (1960) extracted chicken pectoralis at 30 minutes and 24 hr. post-mortem and found no difference in sarcoplasmic protein or nonprotein nitrogen extractability.

Rapid decrease of myosin and an increase of actomyosin were also noted by them. Van den Berg et al. (1964) found that the tenderness of chicken breast muscle incresed rapidly during the first 24 hr. of aging and did not change after 1—2 days.

This study was designed to investigate the effect of chilling time and chilling methods on the extractability of actomyosin, myosin and other nitrogenous fractions of chicken breast muscle and leg muscle during chilling period.

II. Experimental

1. Materials and preparation:

Tests were made with meat from 12 week-old broiler type chickens (female of Indian river strain) whose average carcass weight was 1.09 kg.

Two batches of 14 birds were obtained from two flocks. They were killed in the laboratory by cutting the jugular vein and carotid arteries, bled for 2—3 minutes, scalded for 2 minutes at 53—54°C., plucked by hand, eviscerated and washed.

7 birds of 1 hour post-mortem were kept in slushed ice tank for wet chilling. Other 7 birds of 1 hr post-mortem were placed in plastic bags and kept in chill room (33—35°F) for dry chilling.

White muscle and dark muscle were taken separately from breasts and legs of two birds in chilling tank at 1,3,5 and 6 hr. post-mortem. In case of dry chilling, breast muscle and leg muscle from breasts and legs of two birds were taken at 1,9,17 and 25 hr. post-mortem.

To reduce the effect of bird variability, samples were taken from two birds and minced after cutting thorough mixing.

The whole experiment was carried out in duplication.

2. Nitrogen extraction and fraction

(a) Nitrogen soluble in 0.55 ionic strength-KCl phosphate buffer: The extraction and fractionation was a modification of the type reported by Khan (1962).

10 g. of minced sample was blended with 490 ml of KCl phosphate whose T/2=0.55 (pH 7.5), using baffle plate for 2 minutes. The blended material was centrifuged at 2,800 Rpm for 15 minutes, and then filtered through glass wool. The filterate is No.1 solution.

(b) Nitrogen soluble in 0.225 ionic strength-KCl-phosphate buffers:~Dilute a portion of No. 1 solution with water to T/2=0.225. It was centrifuged at 2,800 Rpm for 15 minutes and then filtered. The protein soluble at T/2=0.55 and insoluble at T/2=0.225 is actomyosin.

(c) Nitrogen soluble in 0.05 ionic strength-KCl-phosphate buffer:- A part of No.1 solution was diluted 11 times with water and then centrifuged at 2,800 Rpm for 15 minutes. The solution was filtered through glass wool and filtrate collected for nitrogen estimation.

Protein soluble at T/2=0.225 and insoluble at T/2=0.225 and insoluble at T/2=0.05 is myosin.

The protein soluble at T/2=0.05 is sarcoplasmic protein.

(d) Nitrogen soluble in 0.1N—NaOH : ~10g. of sample was blended with 490 ml of 0.1N—NaOH with baffle plate for 2 minutes.

The solution was centrifuged at 2,800 Rpm

for 15 minutes and then filtered.

Total nitrogen minus nitrogen in. 01N—NaOH is corresponding to Stroma nitrogen.

(e) Non-protein nitrogen:~ protein soluble in 0.1 N—NaOH was precipitated in a 5% solution of trichloroacetic acid, and then filtered. Nitrogen in filterate is NPN.

(f) Total nitrogen: 2 g. of wet muscle (minced) was digested and then nitrogen was estimated.

3. Nitrogen estimation:

Various nitrogens were estimated by the Kjeldahl method in duplication.

III. Results;

(1) Stroma nitrogen:

Then stroma nitrogen contents in breast muscle and leg muscle remained at an essentially constant level throughout the chilling period. The trend was same in wet chilling as well as dry chilling. The average stroma nitrogens were 8.9% in breast muscle and 16.8% in leg muscle based on the total nitrogen. The stroma nitrogen contents in both breast muscle and leg muscle was slightly less than other researcher' data, such as Khan et al. (1964) and Khan (1962). This might be due to the removal of excess connective tissue before mincing and that left in mincer after mincing.

(2) Sarcoplasmic nitrogen

Sarcoplasmic protein extractability remained essentially constant at average 17.3% of total nitrogen in leg muscle and at average 25.5% of total nitrogen in breast muscle throughout chilling period.

Although the data indicate a slight trend for decreased solubility in case of dry chilling, the difference was not considerable.

(3) Non-protein nitrogen(NPN)

NPN averaged 12.9% of TN in leg muscle and 16.2% of TN in breast muscle, and did not change significantly throughout chilling period. Although these were small fluctuations of NPN extractability, it was not meaningful.

(4) Actomyosin nitrogen

From (Fig. 1) actomyosin solubility showed minor change throughout 5hr. post-mortem. The increase of its extractability was slightly more beyond 5hr. post-mortem in wet chilling.

Actomyosin was extracted at a low level from both leg muscle and breast muscle, but the increase of its extractability in wet chilling till 6hr. post-mortem was slightly more in leg muscle (23.1%), than in breast muscle (14.7%). There was not much difference in actomyosin extractability between wet chilling and dry chilling, except considerable increase of it beyond 17 hr.

Table 1 : Non-myofibrillar nitrogen containing fractions of chicken breast muscle and leg muscle(% N of TN)

Chilling method	Hr. post mortem	Stroma		Sarcoplasmic		NPN	
		leg muscle	breast muscle	leg muscle	breast muscle	leg muscle	Breast muscle
Wet chilling	1	16.1	8.7	16.4	26.4	12.9	15.9
〃	3	16.5	9.1	16.4	25.2	13.0	16.4
〃	5	16.0	8.7	16.7	26.1	12.8	16.9
〃	6	16.7	9.0	16.4	25.4	12.9	15.9
Dry chilling	1	17.0	8.6	18.2	26.9	13.0	16.6
〃	9	17.5	9.2	18.1	25.1	12.8	15.4
〃	17.5	16.9	9.4	18.8	24.6	13.2	15.7
〃	25	17.7	8.7	17.7	24.4	13.1	16.4

Table 2: Myofibrillar nitrogen fractions of chicken breast and leg muscle(% nitrogen of TN)

Method of chilling	Hr. post-mortem	Actomyosin		Myosin	
		leg muscle	Breast muscle	leg muscle	Breast muscle
Wet chilling	1	6.5	8.2	11.8	12.2
〃	3	7.0	8.8	8.9	8.8
〃	5	7.4	8.9	7.7	7.75
〃	6	8.0	9.4	6.7	7.4
Dry chilling	1	6.5	8.9	11.7	12.0
〃	9	8.1	9.7	8.1	9.7
〃	17.5	7.7	9.7	4.8	8.8
〃	25	10.1	11.3	5.8	9.0

post-mortem in case of dry chilling.

Here also the increase of actomyosin extractability for 24 hours dry chilling was more in leg muscle (35.6% of initial) than in breast muscle (27% of initial). Since the extractability of actomyosin increased, even though it was very slow and minor during chilling period, the decrease of extractable nitrogen during chilling was not due to the extractability of actomyosin.

(5) Myosin nitrogen:

The myosin extractability was rapidly decreased for first 3hr. post-mortem in wet chilling and then reduced continuously till 6hr. post-mortem (Fig. 1). There was similar tendency of myosin extractability in both breast muscle and leg muscle during wet chilling. From(Fig. 2), myosin extractability in leg muscle drastically decreased till 17hr. post-mortem and then increased.

In breast muscle, myosin extractability rapidly decreased till 9hr. post-mortem and slowly decreased, and then slowly increased after 17 hr. post-mortem in dry chilling.

The tendency of decreasing myosin extractability was much higher in leg muscle in breast muscle in dry chilling.

The differences of myosin extractability between wet chilling and dry chilling were that myosin extractability in breast and leg muscle decreased in more or less similar pattern, but it was quite different in dry chilling and it increased slowly after 17 hr. post-mortem in dry chilling, whereas

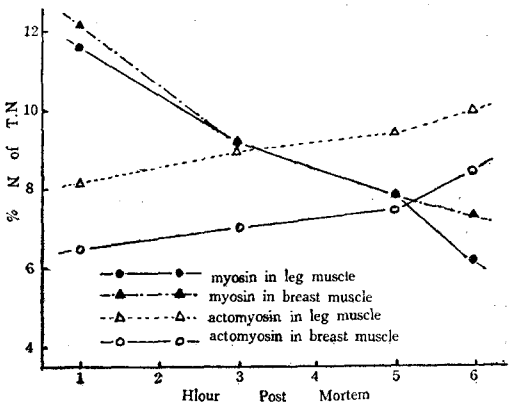

Fig. 1. Myosin and Actomyosin Extractability During Wet Chilling

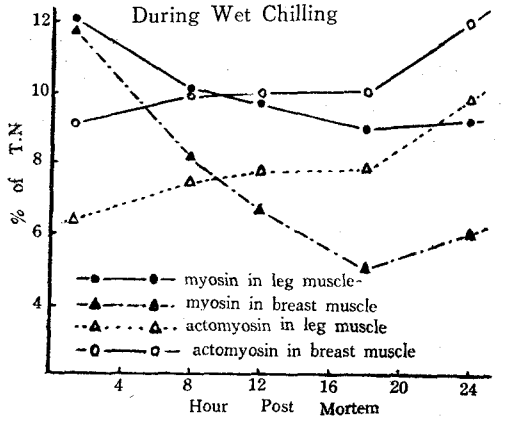

Fig. 2. Myosin and Actomyosin Extractability During Dry Chilling

it decreased till 6hr. post-mortem in wet chilling. Therefore, the decrease of extractable nitrogen must be due to the rapid decrease of myosin extractability during the chilling period.

IV. Discussion

The buffer extractable nitrogen decreases con-

tinuously till 6hr. post-mortem in wet chilling. This agrees with the finding by Khan et al(1964 b). In case of dry chilling, extractable nitrogen tends to increase after 17hr. post-mortem. This may be due to the increase of extractable actomyosin plus myosin after 17hr. post-mortem.

The lack of change in stroma and sarcoplasmic nitrogen during chilling confirms to the results by Khan et al(1964 b). Fairly consistent level of NPN has been observed in both muscle during dry chilling and wet chilling. This was also reported by earlier investigators. Weinberg et al (1960) did not observe changes of NPN in chicken muscle between 30 min. and 24hr. post-mortem. A more detailed investigation of the NPN fraction of the chicken muscle by Khan et al (1964) revealed only small changes in the amount of NPN, but changes were noted in the composition. The rapid initial decline in extractable myosin indicates that buffer insoluble actomyosin is formed extensively during rigor mortis development. Subsequent to the development of rigor mortis, actomyosin becomes increasingly extractable although myosin extractability continues to decline. Weinberg et al (1960) observed the decreased myosin and increased actomyosin extracted from muscle after aging. 24hr. and suggested that a specific cleavage of actin, allowing its extraction and binding to myosin in solution, might account for post-mortem tenderization.

Van den Berg et al (1964) reported that tenderness of thigh increased more slowly than for the breast.

During dry chilling, myosin extractability in leg muscle is far less than that in breast muscle.

The slow increase of tenderness in leg muscle is possibly due to the less myosin extractability at the first stage of tenderization.

ATP being broken down, post-mortem, and myosin being bound to actin, the buffer soluble proteins decreased due to the non-extractability of actin (Sayre, 1968). After 6hr. of aging in wet chilling and 16 hrs in dry chilling, the actin slowly starts to be extracted, resulting in the appearance of increasing amount of actomyosin in solution.

요 약

Slushed ice 와 dry chilling chamber 에 각각 도계 직후 냉장한 12주생 영계의 가슴 및 다리 근육으로부터 KCl-phosphate buffer 를 사용하여 도계후 1~25시간 사이에 함질소물을 추출했다.

Extractable nitrogen 의 변화는 주로 myosin 의 extractability 감소, 그리고 어느정도 actomyosin 의 extractability 증가 결과로 일어남이 밝혀졌으며, 한편 stroma. sarcoplasmic protein 및 비단백함질소물(none protein nitrogen)의 변화는 적었다.

Myosin extractability 는 도계후 처음 3시간 동안에 급격히 감소하여 그후 계속적으로 wet chilling 동안 감소했으나 dry chilling 의 경우 myosin extractability 의 감소는 가슴근육보다 다리근육에서 더 많았으며 extractability 는 도계 17시간 후부터 증가했다.

Actomyosin 은 wet chilling 의 경우 계속적으로 소량밖에 추출되지 않았으나 dry chilling 의 경우 도계후 17시간 이후부터 증가했는데, 이런 경향은 breast muscle 과 leg muscle 이 동일했다.

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