SOME OBSERVATIONS ON THE WATER-HOLDING-CAPACITY OF GROUND LEAN MEAT

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Introduction

In previous report, it was suggested that the binding-quality of ground lean meat, as was seen in products of chopped style like sausage's, was concerned with denaturation of myosin B especially. Recently, from studies on denaturation of myosin, it was defined that myosin played an important role to the binding-quality, and we found it was certain from its practical studies. On the otherhand, several reports on the water-holding-capacity (W-H-C) of lean meat have been contributed, which is the same important problem parallel with binding-quality. However, detailed mechanism of it is unsolved, and we are called upon to resolve these now.

It is indisputable there is a close connection between the both, binding-quality and W-H-C. For example, the solubility was increased by the hydration and the increasing promoted the binding of ground lean meat. But it can be possible to think of the both separatly for convenience. In recently published paper, some authers indicated that increasing the alkalinity of meat increase its ability to retain moisture. The investigation of the effects of phosphate on meat, especially emphasizing factors effecting water retentions, is reported lately. R. HAMM attributed the phosphate effects on meat swelling partly to the increasing of meat pH and to the salting action, J. BENDALL concluded that, as the result of the investigation of the swelling produced by polyphosphate, the increased ionic strength produced the swelling obtained by the use of orthophosphates and polyphosphates. Now, before the study concerning the increase of W-H-C resulted from the increasing of pH by the addition of phosphate, it will be significant to study the effect of pH on water-holding-properties in different ways, from the view that the pH probably greater influence upon the water-holding-properties. In this respect, I can understand only its rough tendency in the level of ground lean meat in which many proteins coexist. Therefore I intend to carry on the work with the level of isolated protein, but

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in this paper I will report on the results of practical experiments obtained up to now. From the result of practical experiment so far, I can understand that water is indispensible for good binding-quality and this is made sure also by experience, but good water-holding-retention dose not always coincide with good binding-quality.

Method and Material

The determination of W-H-C was estimated by some method and it is as follows; The method explained by BENDALL, its modification by DETHERAGE et al, Filter-method reported by GRAU, HAMM, and as to raw meat, the method of SURI or the mechanical one of WIERWICKI, et al. In this paper, the following methods, heating-method which was previously used for the purpose of binding-quality and centrifuging method, were employed.

Method A: Ten gr. ($\pm$0.2 gr.) of ground lean meat trimmed as far as possible fat and connective tissue were placed in 50 ml.-centrifuge tubes and 20 ml. dist. water was added to each one. The dist. water and the ground lean meat were mixed and the mixture were allowed to stand for approximately 30 minutes in water-bath at 25°C, being stirred at times. After 30 minutes, centrifuged at 3,000 r.p.m., the upper layer was at once poured off. And the precipitation-part was used for the calculation of the swelling-rate of ground lean meat according to the following formula:

The swelling-rate of sample

$$= \frac{\text{sample weight after centrifuge}}{\text{sample weight before centrifuge}} \times 100$$

Method B: About 2-3 gr. of the twice ground sample meat were weighed on a porcelain dish. It was dried as far as the constant value in the vacum dryer of 55-60°C., under depressor of 60 inches/cm^2 and its moisture content was estimated. Then, 50 gr. portions of sample was stuffed into casing of gum hydrochloride and was boiled for 30 minutes at 70°C. After the complete cooling, it was cut into 0.2 mm. thick and 2$\pm$0.3 gr. weight. And the slice was placed between two filter papers, which was laid between two glass plates (each 100 gr. weight). Then a 200 gr. poise was set on all of these for one minute in order to be absorbed the surface moisture of the sample in aid of filter paper. By the method above mentioned, the moisture content in the sample was determined. The W-H-C was defined as follows:

Water-Holding-Capacity

$$= \frac{\%\ \text{moisture content of after cooking at 70°C.}}{\%\ \text{moisture content of before cooking at 70°C.}} \times 100$$

The pH values were determined by the use of a glass electrode pH meter. (About 20 ml. of dist. water was added to 10 gr. of ground meat and were prepared by means of a waring blender which made by Nihon Seiki Kogyo Co). The pH values of meat was controlled by injection of insulin before slaughter, according to the method of BATE-SMITH and BENDALL. Moreover, Lactic Acid and Sodium Hydroxide were used for controlling of it's pH values.

Material: Rabbit meat, *M. Semitendineus* of mutton, veal, pork and horse removed as far as possible fat and connective tissue were used.

Result and Discussion

Exp. 1. Change in the swelling-rate of fresh ground lean meat during storage at 2±2°C.

The swelling rate of ground lean meat was estimated, and carried out under

TABLE 1. The swelling-rate of the ground meat during storage at 2±2°C.
(in the absence of NaCl)

Storage time in days	increase %			
	Mutton	Veal	Pork	Horse Meat
0	7.51	3.35	1.62	6.01
2	10.08	4.59	9.16	4.38
4	10.81	5.05	14.39	0.92
6	10.29	—	30.39	—
8	0.64	—	20.64	—
10	20.04	—	14.98	—

TABLE 2. The swelling-rate of the ground lean meat during storage at 2±2°C.
(in the presence of NaCl)

Storage time in days	increase %			
	Mutton	Veal	Pork	Horse Meat
0	5.94	25.04	4.42	—
2	8.30	14.59	25.61	—
4	15.09	35.06	23.89	30.04
6	30.04	36.52	25.42	25.03
8	25.82	20.10	21.19	23.91
10	16.61	18.09	10.08	—

storage at 2±2°C., and the used sample developed for 24 hours after slaughter. Moreover, the 2.5% (W/W) of NaCl was presented to the sample on which salt was added every definit times. The result was shown in Table 1 and Table 2. From these, it was recognized that there seemed to be some difference among species of animals and in spite of the presence of salt under such storage condition, the highest swelling-rate was seen from on the fourth day to the sixth. At the same time, the effects of NaCl on swelling-rate of meat could be observed, distinctly, but it is impossible to explain about the relation between the swelling-rate and the pH, because the change of the pH was not determined.

Exp. 2. Change in the pH value and the W-H-C of rabbit meat during storage at 20°C.

Rabbit meat immediately after slaughter was incubated at 2.0°C, and its pH and W-H-C were estimated every definit times. The result was provided in Fig. 1. The pH was controlled by aid of injection of insulin (2.5 units/Kg. body weight). It was as shown in previous report that the insulin-treated meat could maintained comparatively higher pH. In this case, the experiment was carried out under the condition of no addition of NaCl and water. The change of W-H-C during storage progressed in the similar course with that of pH. Particurally, the insulin-treated sample (pH 7.0) passed for 48 hrs. had rather higher W-H-C than that at 0 hr. The unheated sample in Exp. 1 increased much moisture in it than that the initial sample contained, while the heated

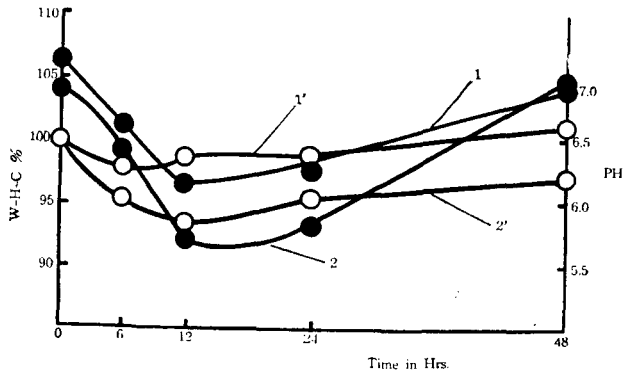

Fig. 1. Change in the PH value and W-H-C of rabbit meat during storage at 20°C.

- ● 1 : The curve of the PH value of insulintreated rabbit.
- ● 2 : The curve of the PH value of well fed immobilized rabbit.
- ○ 1′: The curve of the W-H-C of insulintreated rabbit.
- ○ 2′: The curve of the W-H-C of well fed immobilized rabbit.

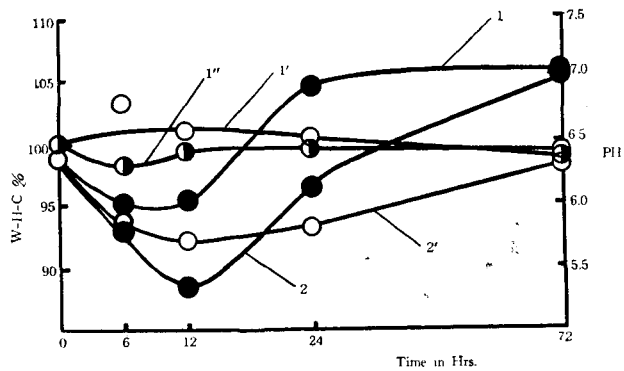

Fig. 2. Change in the PH value and the W-H-C of rabbit meat during storage at 20°C. and effect in addition of NaCl.

- ● 1 : The curve of the PH value of insulintreated rabbit.
- ● 2 : The curve of the PH value of well fed immobilized rabbit.
- ◑ 1″: The curve of the W-H-C of insulintreated rabbit (in the absence of NaCl).
- ○ 1′ : The curve of the W-H-C of insulintreated rabbit (in the presence of NaCl).
- ○ 2′ : The curve of the W-H-C of well fed immobilized rabbit (in the presence of NaCl).

sample in Method B had an tendency of decreasing moisture than that in the initial sample contained. But a test, close to practical condition, was tried next.

The change of pH and that of W-H-C was experimented by the addition of 2.5% (W/W) NaCl every definit times under the same condition as above. Fig. 2 was shown as the result. The difference of the degree between the change of pH and that of W-H-C in both the insulin-treated sample and the unheated one shown in Fig. 1 became more clear by added NaCl. Namely, as to the insulin-treated sample, the W-H-C seemed hardly to change through the storage for 48 hrs. under the presence of NaCl. The sample containning no salt with the insulin-treatment showed a good W-H-C, compared with the contrast sample, but the sample on which the salt was added with the insulin-treatment is the same one with that the salt was not added. From these, it was observed that the addition of salt was very available on the W-H-C. It was also recognized that, when the pH of raw meat was controlled by insulin before death, it had an important effect on the change of the W-H-C.

Exp. 3. The change of the pH value and the change of the W-H-C that take place by addition with lactic acid in the various degree of concentration on rabbit meat immediately after death.

Rabbit meat of pH 6.2 immediately after slaughter was used here, and the pH was controlled by lactic acid. In this case, I calculated respectively the one having controlled (that is, the one not adding lactic acid) per each sample's lost moisture as 0 and the one having released (the one adding 1 M.) the greatest of moisture as 100. And then, the sample of rabbit meat was ground twice. It's 50 gr. were weighed and to it I served, adding 1.25 gr. NaCl and 10 ml. lactic acid in the various degree of concentration, and on the controlling one 10 ml. dist. water, respectively. When the varied lactic acid in concentration was added, the following states were observed. With its 5×10^{-1} M. a little coagulation was occured, for the pH of the sample meat was lowered than the pH required, and with its 1 M., the coagulation became perfect. The result of having measured the pH change by the addition of lactic acid in the various degree of concentration and the W-H-C is shown in Fig. 3. Consequently, it was evidently admited that there appeared little difference on both the pH and the W-H-C within the 0 M. and 1×10^{-1} M., and the pH sought for was between 1×10^{-1} M and 5×10^{-1} M., moreover, it was able to release the moisture even with the meat used immediately after slaughter, by moving the pH to the acidic region.

From Fig. 3, it was clarified that the concentration from on 1×10^{-1} M. to 5×10^{-1} M. was necessary to get the neighborhood of isoelectric point of

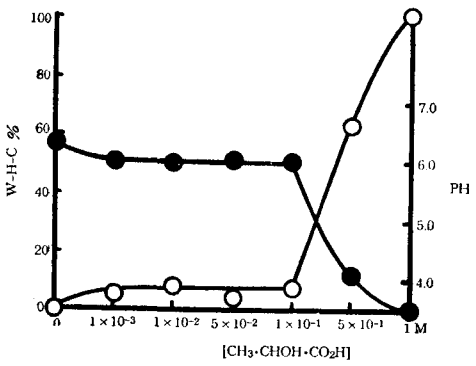

Fig. 3. Change in the PH value and the W-H-C of rabbit meat in addition with the lactic acid in the various degree of concentration immediately after death.
● : The curve of the PH value of rabbit meat.
○ : The curve of the W-H-C of rabbit meat.

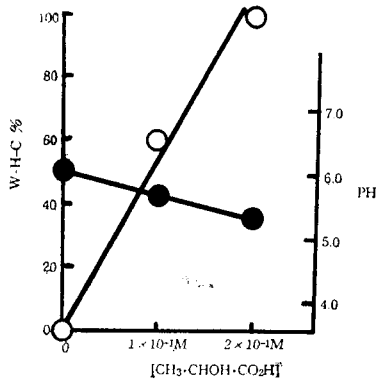

Fig. 4. Change in the PH value and the W-H-C of rabbit meat in addition with lactic acid on from 0 M. to 2×10^{-1} M. immediately after death.
● : The curve of the PH value of rabbit meat.
○ : The curve of the W-H-C of rabbit meat.

myosin, where, it is said, the W-H-C of meat was the worst. So, supposing the rabbit meat of pH 6.0 immediately after slaughter as the sample shown in Fig. 3, we added on it lactic acid in the various degree of concentration. The result is shown in Fig. 4. In this result, the pH became 5.39 at 2×10^{-1} M. and it being below 5.0 at 4×10^{-1} M. there occurred coagulation. At this pH was pretty lower than the isoelectric point, we took it away from data. Now, from this Fig. 4, it was found that the pH value and the W-H-C made a linea roughly between the 0 M. and 2×10^{-1} M. concentration added.

Exp. 4. The change of the pH and that of the W-H-C that take place by addition with lactic acid in the various degree of concentration on rabbit meat which incubated at 20°C. for 70 hrs. after death.

In Fig. 3 and Fig. 4, I tried, using the one immediately after slaughter as the sample. Here I carried out an experiment in the same method as above mentioned, using as the sample the meat which was pretty spoiled on account of the incubation at 20°C. for 70 hrs. after slaughter. In this case, there happenning the coagulation on the concentration above 4×10^{-1} M., the change of the state was observed. The effect made experiments from the result in Fig. 3

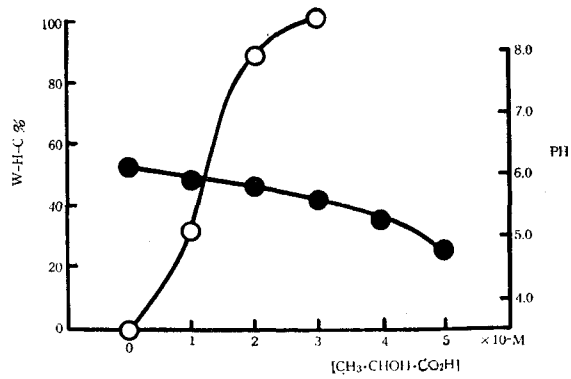

Fig. 5. Change in the PH value and the W-H-C in addition with lactic acid in the various degree of concentration on rabbit meat which incubated at 20°C. for 70 hrs. after death.

● : The curve of the PH value of rabbit meat.
○ : The curve of the W-H-C of rabbit meat.

and 4, about the range from the 0 M. to 3×10^{-1} M. is just shown in Fig. 5. The pH (that is, the one not adding lactic acid) of the 0 M. in this case, was 6. 1. Under normal autolytic condition at 20°C., the pH of meat may increase from 5.4-5.8 to 6.5-7.0, however, the increase of the pH of sample meat was not observed through the process to the putrefaction.

As is seen in Fig. 5, the decrease of the W-H-C of meat was found to be proportional to that of the pH value.

Exp. 5. The change of the pH and the change of W-H-C that take place by addition with sodium hydroxide in the various degree of concentration on rabbit meat which incubated at 20°C. for 20 hrs. after death.

It can be considered that the opposite effect to the one observed with the use of lactic acid ought to occur by using NaOH. Now, the following experiment was carried out. The rabbit meat immediately after slaughter was incubated at 20°C. for 20 hrs. And its pH shifting to the isoelectric point (pH 5.5), NaOH in the various degree of concentration was added on it, and thus the pH was controlled.

The change in the W-H-C of such meat was tested in the way before done. The result is shown in Fig. 6. However, in this case, the W-H-C was increasing in contrast with the control. So, it was expressed as increase %, i.e., the W-H-C of 0 M. NaOH was zero and that of 1 M. NaOH 100, respectively. The pH of the 0 M. here was 5.6. It increased to the alkaline side in gentle slope up to 1×10^{-1}M., but in the concentration above 5×10^{-1} M., the curve showed sudden rise and in 1 M., the remarkable change of the state of the sample were seen, it became less viscous.

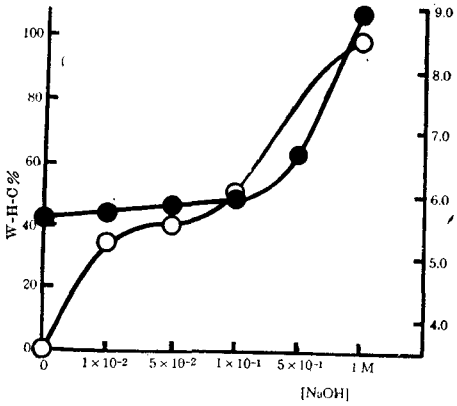

Fig. 6. Change in the PH value and the W-H-C in addition with NaOH in the various degree of concentration on rabbit meat which incubated at 20°C. for 20 hrs. after death.

● : The curve of the PH value of rabbit meat.

○ : The curve of the W-H-C of rabbit meat.

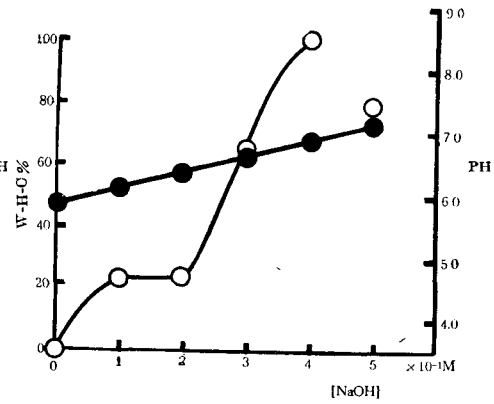

Fig. 7. Change in the PH value and the W-H-C in addition with NaOH in the various degree of concentration on rabbit meat which incubated at 20°C. for 20 hrs. after death.

● : The curve of the PH of rabbit meat.

○ : The curve of the W-H-C of rabbit meat.

From this result, the W-H-C was found to be controlled by the change of the pH, almost having nothing to do with the state of the freshness of meat.

From the result of Fig. 6, it was clarified that the sample meat incubated at 20°C. for 20 hrs. immediately after slaughter was found in the neighborhood of the isoelectric point. When it was alkalized by NaOH of the various concentration, the concentration required was between 1×10^{-1} M. and 5×10^{-1} M. From this fact, the experiment using NaOH (instead of lactic acid) was carried out in the same sample, and the result is shown in Fig. 7. In consequence, it was observed that the pH curve was nearly linear between 2×10^{-1} M. and 4×10^{-1} M., that is in the neighborhood between pH 6.3 and pH 6.8, the increase of W-H-C brought about a great change. The change of the pH between the 0 M. and 2×10^{-1} M. had little difference from that between 2×10^{-1} M. and 4×10^{-1} M., but its increase of W-H-C showed a great difference. It can be supported the effect is due that the neighborhood of pH 6 is close to the isoelectric point of muscle structural protein.

Summary

(1) The swelling-rate of fresh meat progressed to the maximum on the fourth or sixth day under the storage condition at 2 ± 2°C. And the addition of NaCl increased the swelling-rate of meat remarkably.

(2) The water-holding-capacity of meat after death progressed in the same degree as the change of the pH value through the time of storage at 20°C. On the other hand, the rabbit meat treated with insulin kept its pH value in the neutral region and its W-H-C, being settled, dose not change. Moreover, the addition of NaCl increased the W-H-C of meat, and beside, it made the difference of changing degree of W-H-C between the two sample above mentioned more definite.

(3) It was clarified that, there W-H-C could be controlled having no concerned with the time of storage of meat, by controlling the pH of meat with the use of lactic acid or sodium hydroxide. Namely, when lactic acid being added, the pH of meat was adjusted to the neighborhood of isoelectric point (5.5-6.0), the W-H-C remarkably decreased in opposition, when the pH of meat was ajusted to the neutral region by using sodium hydroxide, the W-H-C astonishingly increased.

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