

# EFFECTS OF FREEZING TEMPERATURE ON THE PHYSICOCHEMICAL AND PROCESSING QUALITY OF PORK

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## SUMMARY

*Longissimus dorsi* muscle from 6 pigs (24hr postmortem) was cut into portions of similar size and shape (ca.700g) and vacuum packed in polyfilm. The muscle specimens were divided in three samples, one frozen at  $-20^{\circ}\text{C}$ , another at  $-80^{\circ}\text{C}$  and one to serve as the control (not frozen). The meat sample frozen at  $-80^{\circ}\text{C}$  was transferred to the  $-20^{\circ}\text{C}$  freezer. After one month, both frozen pork samples were thawed at  $2^{\circ}\text{C}$  and drip loss (%) was measured. Hunter color, metmyoglobin formation (MetMb, %), water holding capacity (WHC), TBA, transmission value (TM) and myofibril fragmentation were also determined. There was no significant difference in drip loss for the two frozen samples. No MetMb formation could be detected and Hunter values were also basically the same for all three samples. WHC, TBA and TM were essentially the same for all three samples. TBA was quite low for each frozen sample, clearly indicating that lipid oxidation did not occur during freezing. Histological examination of both frozen samples indicated inter- and intracellular ice crystal formation at  $-20^{\circ}\text{C}$ , and intracellular ice at  $-80^{\circ}\text{C}$ , the extent being less than at  $-20^{\circ}\text{C}$ . At  $-20^{\circ}\text{C}$ , ice crystals were larger and fiber diameter smaller than for the control  $-80^{\circ}\text{C}$  sample. Myofibril fragmentation in both frozen samples was significantly higher than in the control. Pork sausage was prepared from all three samples by adding 2% NaCl and 100ppm NaNO<sub>2</sub>. Cooking loss and color forming ratios were essentially the same. The sausage sample made from the  $-20^{\circ}\text{C}$  frozen meat was harder than that of the other two samples according to rheological measurement.

## INTRODUCTION

As the condition for storage and marketing of pork in Japan, domestic pork is refrigerated in most cases. About 20% of the pork for this country is imported, generally in frozen form. Freezing has many advantages for the preservation of meat and facilitates its marketing, but there is some destruction of muscle fiber due to the formation of ice crystals. This may lead to problems such as drip loss at meat thawing and oxidation of muscle pigment (myoglobin), and the reduction in gel forming ability of myofibrillar proteins. A preliminary study was made of pork stored at  $-19 \pm 1^{\circ}\text{C}$  and drip loss during thawing was noted to be greater for longer freezing periods, and deterioration of meat quality to occur after 9 month of storage based on color and TBA data (shown in Fig.1, SAKATA et al., 1989).

This study was conducted to examine the effects of freezing temperature at  $-20$  and  $-80^{\circ}\text{C}$  on pork quality.

## MATERIALS and METHODS

**Meat sample:** *Longissimus dorsi* muscle from left side of 6 pigs (24hr postmortem) was cut into 6 portions of similar weight (ca.700g) and divided in three samples, one frozen at  $-20^{\circ}\text{C}$ , another at  $-80^{\circ}\text{C}$  and one to serve as the control (not frozen), by the method of Latin square design. Meat samples subjected to freezing were vacuum packed in barrier multilayer film (Diamiron M, Mitsubishi Ind. Ltd.) and frozen either at  $-20^{\circ}\text{C}$  or  $-80^{\circ}\text{C}$  in freezer with monitoring the meat temperature by inserting thermocouple recorder. Frozen samples were stored at  $-20^{\circ}\text{C}$  for one month. The control sample was swiftly analyzed.

**Analysis:** After one month, both frozen pork samples were thawed at  $2^{\circ}\text{C}$ , the packages were opened, and drip loss (%) was measured. The meat surface of the loin eye was exposed to air for 30 min (blooming) to determine MetMb content from the K/S value of reflectance at 572nm and 525nm using a Beckman model 25 spectrophotometer. Hunter values were determined by a color difference meter (Nippon Denshoku Kogyo Co. Ltd. Model ND-1001 DP). Histological examination of muscle fiber was conducted using the hematoxylin/eosin staining method. Myofibrils and sarcoplasm fraction were also prepared by the method of GOLL et al. (1974) for measuring myofibril fragmentation and contractility, and sarcoplasmic protein extractability. The appearance of myofibrils in the suspension was observed with a phase-contrast microscope/camera (Olympus BH-2/C-35DA-2). WHC was determined by a slight modification of the filter-paper press method (HOFMANN, 1982; SAKATA et al., 1991). The meat sample was minced and heme pigment content (OKAYAMA and NAGATA, 1979) and TBA value were determined. Pork sausage was prepared from

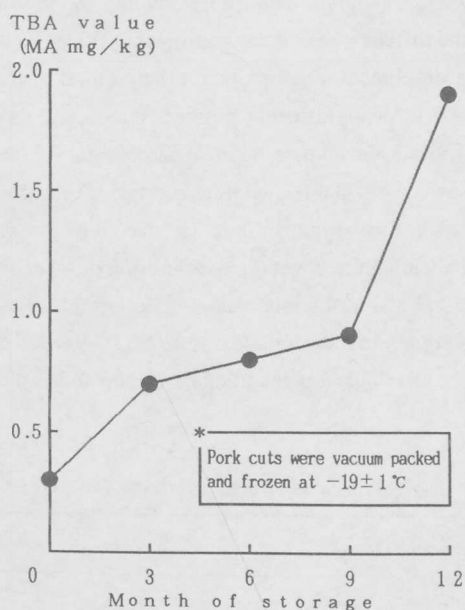


Fig.1. Effect of freezing period on TBA value of pork\*

all three minced samples by adding 2% NaCl and 100ppm NaNO<sub>2</sub>. Texture (Taketomo Co. Ltd., Tensipresser TTP-50BX), cooking loss, color forming ratio (CFR) and residual NO<sub>2</sub><sup>-</sup> were measured.

RESULTS and DISCUSSION

Fig.2 shows changes in meat temperature during freezing, storage and the thawing period. At -20° C freezing, about 6hr were required to pass through the maximal ice crystal forming zone (-1~-6° C). For the -80° C sample, only about half as much time was required. Though the -80° C freezer used the convection style, the temperature decreased in the same manner as in the industrial air-blast system. The meat sample was analyzed after a 15hr thawing period, since about 12hr were required for the internal temperature of the frozen sample to reach

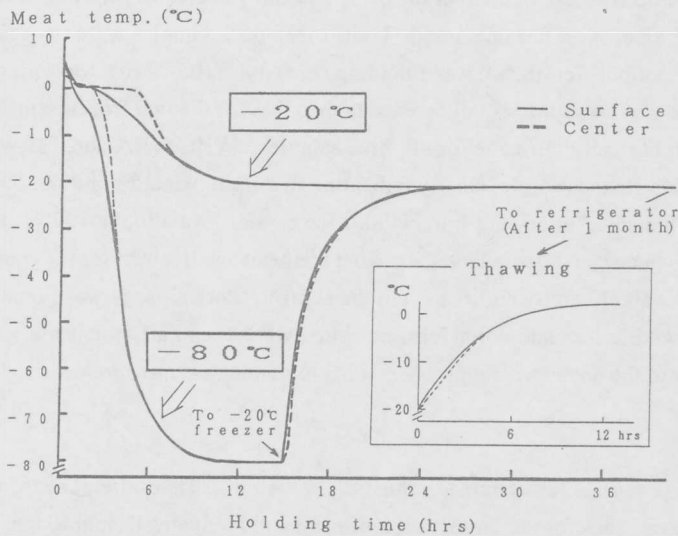


Fig.2. Changes in meat temperature during freezing, storage and thawing

2° C in a refrigerator. Moisture, WHC and TBA values are indicated in Table 1. The three measured items were virtually the same for the control and frozen samples. TBA values showed only slight variation, indicating that lipid oxidation did not occur during freezing. Determination was made of the extractability of sarcoplasmic proteins in terms of TM as

Table 1. Physicochemical characteristics of meat (I)

Items	Control	- 2 0 °C	- 8 0 °C
Moisture(%)	72.0±1.5	72.3±1.5	72.1±1.8
WHC (%)	72.0±2.1	73.1±3.0	71.1±1.8
TBA value	.08±.04	.09±.06	.24±.27

estimated by HART (1962) and extracted soluble protein by the biuret method. Higher TM is considered indicative of greater denaturation of sarcoplasmic proteins. Extractability of the sarcoplasmic proteins of the two samples was ca.97% and 93%, at -20° C and -80° C respectively, the difference not being significant (Table 2). TM of sarcoplasm from all three meat samples was below 30% (data not shown), indicating the normal meat range based on the method of HART (1962). Drip loss from the -80° C sample was slightly higher, but between -20° C and -80° C, no significant difference was noted, as shown in Table 2.

Table 3 shows data on meat sample color. The Hunter a-value, indicating redness, was higher at -20° C. Light scattering from the meat surface may be related to redness, since the porous state of muscle fiber has been observed by microstructure analysis (Fig.3). But this point has yet to be confirmed. Hunter color values were basically the same for all three samples. No MetMb formation could be detected, indicating there was no discoloration of meat under the present experimental conditions.

Fig.3-a shows a microscopic photograph of a cross section of the control meat sample. Muscle fiber structure was highly retained in this sample. In muscle frozen at -20° C, larger ice crystals formed inter- and intracellularly, causing great damage (Fig.3-b). In Fig.3-c is shown the histological preparation of the -80° C sample; it can be seen that ice crystals formed intercellularly and were smaller than those of the -20° C

Table 2. Physicochemical characteristics of meat (II)

Items	- 2 0 °C	- 8 0 °C
Extractability of sarcoplasmic proteins <sup>1</sup>	97.3±9.1	92.7±2.7
Drip loss <sup>2</sup>	3.7±1.5	5.2±2.5

<sup>1</sup> Relative content in each sample, calculated from that of the control as 100.

<sup>2</sup> Values are weight percentages of drip to meat after thawing.

Table 3. Physicochemical characteristics of meat (III)

Items	Control	- 2 0 °C	- 8 0 °C
Hunter values			
L	45.6±2.6	43.1±2.9	43.7±2.7
a <sup>1</sup>	18.7±1.8 <sup>a</sup>	21.4±1.5 <sup>b</sup>	20.1±2.0 <sup>a</sup>
b	8.5±0.9	9.5±0.9	9.6±1.0
Heme pigment content <sup>2</sup>	103.9±10.9	110.5±9.6	116.0±36.5

<sup>1</sup> Means with different superscripts significantly differ (p<0.05).

<sup>2</sup> Determined by the acetone/HCl extraction method; expressed as mg% of Mb.

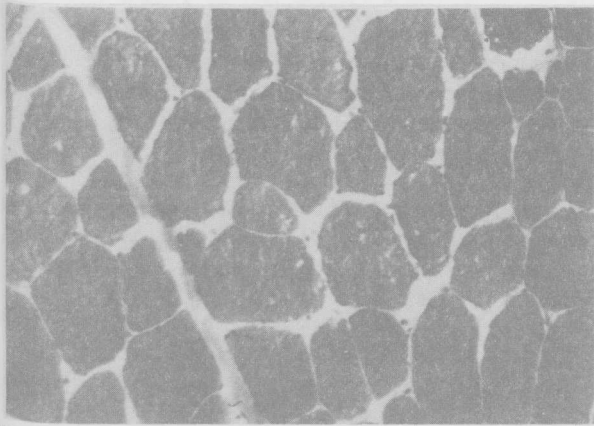


Fig.3-a. Cross-section of control porcine muscle (not frozen,  $\times 33$ )

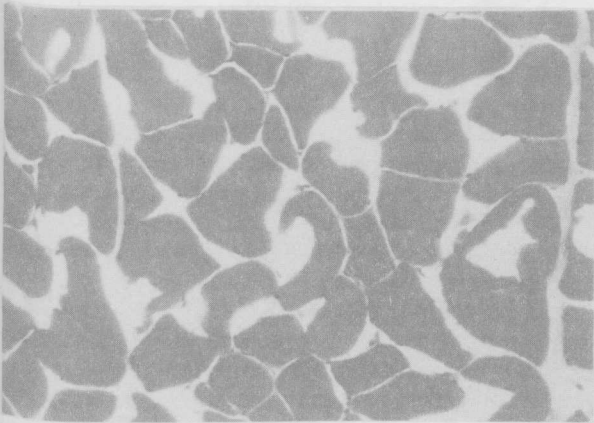


Fig.3-b. Cross-section of porcine muscle frozen at  $-20^{\circ}\text{C}$  ( $\times 33$ )

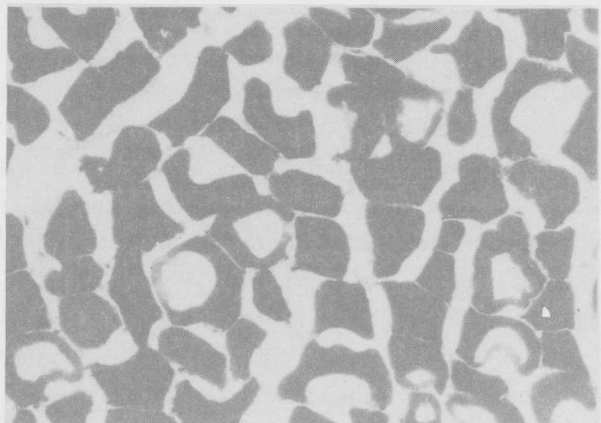


Fig.3-c. Cross-section of porcine muscle frozen at  $-80^{\circ}\text{C}$  ( $\times 33$ )

Table 4. Diameter of a muscle fiber ( $\mu\text{m}$ )\*

Control	$-20^{\circ}\text{C}$	$-80^{\circ}\text{C}$
$66.4 \pm 8.6^a$	$56.1 \pm 5.6^b$	$60.5 \pm 2.6^a$

\* As determined using one hundred fibers for a cross-section of each meat sample.

muscle. Muscle fiber diameter is indicated in Table 4. At  $-20^{\circ}\text{C}$ , it was less than that of the control or  $-80^{\circ}\text{C}$  sample. The  $-80^{\circ}\text{C}$  sample and control showed basically the same value for diameter. RAHELIC et al.(1985) made a study of histological changes in beef *longissimus dorsi*



Fig.4. Phase-contrast microphotograph of the myofibrils from  $-20^{\circ}\text{C}$  frozen muscle ( $\times 333$ )

Table 5. Fragmentation of myofibrils (%)\*

Control	$-20^{\circ}\text{C}$	$-80^{\circ}\text{C}$
$2.1 \pm 0.5^a$	$24.2 \pm 7.9^b$	$18.1 \pm 6.0^b$

\* Five hundred myofibrils were observed for each sample; Expressed as % of myofibrils whose sarcomere number was below 4

Table 6. Processing quality of sausage prepared from meat<sup>1</sup>

Items	Control	$-20^{\circ}\text{C}$	$-80^{\circ}\text{C}$
Cooking loss (%) <sup>2</sup>	$15.8 \pm 4.3$	$12.3 \pm 5.3$	$12.0 \pm 3.1$
CFR (%) <sup>3</sup>	$85.6 \pm 2.8^a$	$81.7 \pm 2.9^b$	$81.1 \pm 2.8^b$
Residual $\text{NO}_2^-$ (ppm) <sup>4</sup>	$48.6 \pm 5.1$	$50.2 \pm 9.4$	$51.0 \pm 3.9$
Texture			
Hardness (kg/cm <sup>2</sup> )	$2.2 \pm 0.2^a$	$2.5 \pm 0.3^b$	$2.3 \pm 0.3^{ab}$
Cohensiveness	$.35 \pm .02$	$.37 \pm .03$	$.41 \pm .09$
Elasticity (%)	$.30 \pm .05$	$.31 \pm .10$	$.31 \pm .09$

<sup>1</sup> Contained 100ppm  $\text{NaNO}_2$ , 2% NaCl and 10% added water; Stuffed into polyvinylidene chloride casing and cooked at  $75^{\circ}\text{C}$  for 40min.

<sup>2</sup> Values are weight percentages of drip to meat after cooking.

<sup>3</sup> Percentages of nitroso heme pigments to the total heme pigments (SAKATA and NAGATA, 1991).

<sup>4</sup> Determined by the method of MIRNA and SCHÜTZ (1972).



muscle frozen at several different temperatures between -10 and -196°C and found damage to be greatest at -22°C due to intra- and intercellularly formed ice. In muscle frozen at -78°C, ice crystals have been observed intracellularly and gaps to be present in all fibers. The present data are essentially in agreement with these findings in spite of the double freezing treatment for preparing cross section cut (isopentane/dry ice, -80°C).

Fig. 4 shows phase-contact microphotograph of myofibrils from the -20°C frozen meat before contraction. Fragmentation of the myofibrils increased in the frozen sample, and in both frozen samples, this parameter was greater than that of the control (Table 5). Fragmentation is used as an index of meat aging. However, this increase may not be due to aging, but to physicochemical changes that result from freezing. Contraction of myofibrils by adding  $Mg^{2+}$ -ATP solution (SUNG et al., 1976) was observed in both the control and frozen sample, indicating biochemical activity not to be lost in frozen pork. No significant changes in myofibrillar or sarcoplasmic proteins prepared from the three experimental meat samples could be detected by SDS-polyacrylamide gel electrophoresis (using 12.5% gel, data not shown).

In Table 6 are shown the results for sausage prepared from experimental meat. In a preliminary study (SAKATA et al., 1989), sausage from -20°C frozen pork had significantly lower CFR when stored over 9 months. The freezing period in this study was shorter (1 month) and changes in CFR differed from those noted in our previous research. CFR of both frozen samples decreased to less than that of the control but exceeded 80%, so that the normal color of cooked cured meat products was evident (SAKATA and NAGATA, 1991). The texture of sausage from the -20°C frozen meat was harder. The relationship between hardness and shrinkage of muscle fiber in Fig.3-b is not clear at the present.

Generally, fast and deep freezing is desirable for maintaining the excellent quality of fresh raw meat. In this study, no deteriorative effect of freezing temperature on meat quality could be found. Therefore, -20°C freezing for a shorter period may be of advantage for the marketing of frozen pork. Even at -80°C, whether there is any actual advantage must be based on consideration of velocity of chilling wind and meat freezing rate. The effects of frozen storage time on meat quality should be studied and sensory evaluation made.

## CONCLUSIONS:

1) Freezing at -20°C and -80°C has no effect on WHC of meat, and lipid oxidation does not occur during one month of frozen storage; 2) -20°C frozen storage produces a slight increase in redness on the surface; 3) Ice crystals are formed even in -80°C frozen meat; 4) Myofibril fragmentation significantly increases with frozen storage, regardless of the freezing temperature, and 5) Freezing of meat decreases CFR of sausage.

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