Some Properties of Abnormal Porcine Muscles (PFE, PFD) Differred from PSE and DFD

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<u>SUMMARY</u> Abnormal porcine muscles (PFE, PFD) differred from well known PSE and DFD were recently found at an abattoir. By visual subjective evaluation, PFE showed the intermediate properties between PSE and normal pork, while PFD showed the intermediate between DFD and normal.

Some properties of PFE and PFD were examined according to the methods used for the detection of PSE and DFD, which were the measurements of (1) pH₁ and pH₂₄, (2) R-value (250/260nm), (3) transmission-value (T-value). Results obtained from these measurements ^{Suggested} that the properties of PFE (pH₁=5.5, T=62%, R=1.3) were similar to that of PSE $(pH_1=5.35, T=88\%, R=1.3)$, whereas general features of PFD $(pH_{24}=6.38, T=15\%, R=1.3)$ were ^{Similar} to that of DFD $(pH_{24}=6.43, T=8\%, R=1.3)$.

Then the following three methods were applied to differentiate PFE and PFD from PSE and DFD, respectively: (4) contractility of myofibrils, (5) detection of phosphorylase band on SDS-polyacrylamide gels of myofibrils, (6) Mg²⁺-ATP induced morphological changes of the precipitates formed in the pH 4.5 solution for the T-value test. These results indicated that PFE had the intermediate properties between PSE and normal pork, while PFD had the intermediate between DFD and normal. As a result of this research, it was found that the method(6) was a useful means to differentiate abnormal porcine muscles, that is ^{PSE}, PFE, PFD and DFD, in each other.

INTRODUCTION Abnormal porcine muscle types differred from well known PSE and DFD were recently found in a commercial abattoir in Japan. One of these abnormal muscles with rapid rigor-mortis showed the properties that is pale in colour, firm in texture and soft and exudative; then we called this muscle as PFE. Another abnormal muscles showed pale apparence and firm texture as same as PFE, but dry to the touch; then we called this as PFD.

In order to clarify the biochemical properties of PFE and PFD, measurements of pH, R-value(250/260nm) and transmission-value(T-value) of muscles were applied as an easy and rapid detecting method of abnormal porcine muscles in this study. Measurements of pH and R-value were proposed to be an easy and rapid method for the detection of PSE and DFD in an abattoir by Honikel and Fisher (1977). T-value was proved to express the degree of degeneration on porcine muscle (Hart et al., 1963) and subsequently to evaluate the degree of PSE (Eikelenboom et al., 1974, Koishikawa et al., 1979).

Then the following three methods were carried out to differentiate PFE from PSE and PFD from DFD. (1) Contractility of myofibrils; Sung et al (1976) showed that contraction did not occure in PSE muscles. (2) Detection of phosphorylase band on SDS-polyacrylamide Sel electrophoregrams of myofibrils. This band was found in the myofibrils prepared from PSE muscles (Yamamoto et al., 1979, Yuasa et al., 1981). (3) Morphological changes of the precipitates formed in the course of T-value test were observed under the phase-contrast microscope. This application of method (3) to the differentiation of PFE from PSE, and of PFD from DFD was the first trial in the field of meat research.

The aim of this study is to clarify the biochemical properties of abnormal porcine muscles (PFE, PFD, PSE, and DFD) according to the methods used for the detection of PSE and DFD.

<u>MATERIALS AND METHODS</u> Meat samples were excised from porcine M. semimembranosus muscles between 45 and 60 min post-mortem. Measurements of pH were carried out 1 hr (pH₁) and 24 hr (pH₂₄) post-mortem. The other measurements were carried out between 1 hr and 2 hr post-mortem. Criteria of PSE and DFD in this study were as follows: Muscles with a pH₁ \leq 5.4, a R-value \geq 1.05, and a T-value \geq 80% were classified as being PSE, while muscles with a pH₂₄ \geq 6.2 and a R-value \geq 1.05 were classified as being DFD. Figure 1 shows the typical porcine carcass in which abnormal muscles were found shortly after slaughter.



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Fig.1. The typical porcine carcass with abnormal muscles (center)

 \underline{pH} The pH of muscles was measured by insertion of the glass electrode directly into the samples.

<u>R-value</u> The procedure of Honikel and Fisher(1977) was used. Muscle (2g) was homogenized in 15 ml of 1M perchloric acid with a high speed homogenizer (Mitamura Riken Kogyo Co, LTD) for 60 sec. The homogenate is centrifuged and then 0.1 ml of the supernatant is diluted into 4.9 ml of 0.1M phosphate buffer pH7.0. The absorption at 250 nm and 260 nm is measured with phosphate buffer as reference and R-value is calculated as the 250/260 nm ratio of absorbances.

<u>T-value</u> T-value was determined according to the procedure of Hart(1962) with a slight modification. Muscle (5g) was homogenized with a high speed homogenizer for 60 sec. The homogenate was centrifuged (2000rpm, 10 min) and filtered. One ml of the filtrate was added to one test tube containing 5 ml citrate phosphate buffer adjusted to pH 4.⁵ and to one tube containing 5 ml distilled water as a reference. After 10 minutes, turbidity was measured at a wavelenght of 600 nm with double-beam spectrophotometer.

<u>Contractility of myofibrils</u> Contractility of myofibrils was determined according ^{to} the procedure of Sung et al (1976) with a slight modification. A drop of myofibrillar suspension was placed on a slide glass and the appearance of myofibrils in the suspension before and after the addition of the Mg²⁺-ATP solution was observed with a microscope. Myofibrils were prepared from porcine muscles by the procedue of Briskey and Fukazawa (1971) with a slight modification.

<u>Morphological changes of the precipitates</u> A drop of precipitate suspension, which formed in the pH 4.5 solution for the T-value test, was placed on a slide glass and used for microscopic examination. The appearance of precipitates in the suspension before and after the addition of the Mg^{2+} -ATP solution was observed with an Olympus phase contrast microscope and photographed with an Olympus camera attached to the microscope.

RESULT and DISCUSSION

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(1) Visual subjective evaluation

Visual characteristics of abnormal Porcine muscles (PFE, PFD, PSE and DFD) between 45-60 min post-mortem were shown in Table 1.

(2) Physiochemical examination

Table	1. Visua	al Chara	acteristics of Abnormal
	Porc	cles at 1 hour p.m.	
PSE	pale	soft	exudative
PFE	pale	firm	exudative
PFD	pale	firm	dry
DFD	dark	firm	dry

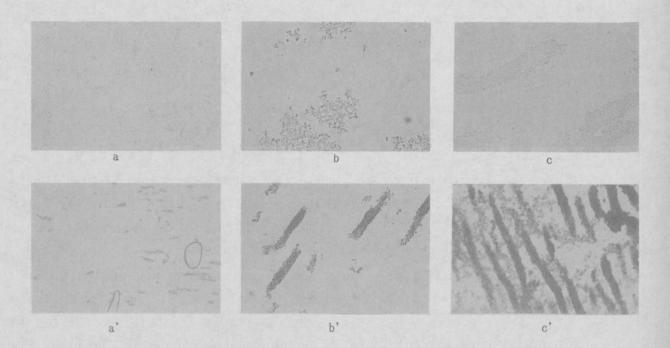
Results of physiochemical examination in normal and abnormal porcine muscles (PFE, PFD, PSE and DFD) were shown in Table 2. PH_1 -value of PFE showed a relatively rapid decrease to 5.5, resembling that of PSE (5.35). PFD had a high pH_{24} value (6.38) similar to that of DFD (6.43). Then both PFE and PFD showed a much higher R-value (1.31, 1.29, respectively) than normal muscle (0.89). These high R-values of PFE and PFD muscles were similar to that of PSE (1.33) and DFD (1.24). These results suggested a rapid post-mortem breakdown of ATP and a rapid rigor mortis in not only PSE and DFD in but PFE and PFD. T-value of PFE was high (62%) and close to that of PSE (88%), while T -value of PFD was low (15%) and close to that of DFD (8%). These T-values observed in abnormal porcine muscles implicated that denaturations of PFE muscle proteins occured slightly , while denaturations of PFD did not occured. Results from these measurements suggested that PFE was similar to PSE in characteristic properties, while PFD was similar to DFD.

Then the following three examinations were carried out to differentiate PFE from PSE and PFD from DFD. (1) Experiments on contractility of myofibrils revealed the results that PFE contracted in part, while myofibrils prepared from PSE contracted very slowly or did not contracted after the addition of Mg²⁺-ATP solution. (2) Detection of Phosphorylase in myofibril was carried out on the muscles by means of SDS polyacrylamide Sel electrophoresis (Yamamoto et al. 1979). The phosphorylase bands were observed on the ^{electrophore}grams of myofibril ·prepared from PSE and PFE, but did not observed from normal, PFD and DFD. (3) Phase-contrast microscopic observations of precipitates Produced in the course of T-value test indicated the distinct differences on the morphological changes among normal, PSE and DFD, as shown in Fig. 2. This microscopic method was considered as a useful means to differentiate normal, PSE and DFD, in each other. The results obtained from experiments (1) and (2) suggested that PFE was

	Muscles						
	<u> </u>	pH ₁	pH ₂₄	R-value	T-value(%)		
PSE	4	5.35±0.03	*	1.33±0.04	88±1.4		
PFE	3	5.55±0.15	5.50±0.16	1.31±0.05	62±16.0		
PFD	3	6.16±0.04	6.38±0.04	1.29±0.01	15±8.0		
DFD	4	6.64±0.24	6.43±0.14	1.24±0.09	8±3.0		
Normal	20	6.44±0.35	5.57±0.06	0.89±0.05	29±19.1		

Table 2. Physical and Chemical Properties of Normal and Abnormal

* pH₂₄ in PSE muscle was not measured, because pH₁ in PSE muscle already reached to the ultimate pH.



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Fig. 2. Phase contrast micrographs of the precipitates formed in the pH 4.5 solution before(a,b,c) and after(a',b',c') the addition of 1mM Mg²⁺⁻ ATP solution (a,a',normal; b,b',PSE; c,c',DFD; Magnification: x400.)

classified as a slight and \checkmark or mild PSE, but that PFD could not be distinguished from DFD. Results from experiments (3) suggested that PFD was classified as a slight and or mild DFD.

In conclusion, we considered that PFE are as an intermediate muscle type between normal and PSE, and PFD are as an intermediate between normal and DFD.

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