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PRE-SLAUGHTER TREATMENTS AS RELATED TO POST-MORTEM
CHANGES IN PORK MUSCLE

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Many of the present-day pork processing and marketing problems in the United States are seemingly attributable to differences in pork muscle appearance and character. These differences are manifest in a wide variety of pork muscle qualities ranging from pale, soft, watery pork to extremely dark, dry product. These different qualities vary in consumer appeal, presumably differ in nutritional value and definitely reflect major yield differences in the processing line.

The objectives of the Wisconsin research program which encompasses this problem are (1) to characterize the physical and chemical properties of pork exhibiting various qualities (2) to develop pre-slaughter treatments to experimentally alter post-mortem changes (3) to study the fundamental mechanisms involved in the creation of the various qualities and (4) to develop control measures for producing more uniform pork.

In the limited time which I have available for this report, I wish to (1) present a summary of our initial experiments with this problem and (2) to discuss pre-slaughter treatments which the Wisconsin station has administered to effectively alter post-mortem changes. I would like to mention at the outset that much of this work is in direct support of the work of Dr. Bate-Smith, Dr. Bendall, Dr. Callow and Dr. Lawrie of England; Dr. Ludvigsen and Dr. J. Wismer-Pedersen of Denmark and Dr. R. Hamann of Germany.

Preliminary experiments were conducted to characterize the various qualities. The first slide presents the word descriptions which were used to clarify the photographic standards used to classify the hams.

HAM CLASSIFICATION

I. (Pale two-toned) muscles were pale in color, soft and watery in structure and possessed loose inter-muscular binding.

II. (Two-toned) muscles adjacent to the bone were dark in color, while the large outer muscles were pale in color, lacking in firmness, moderately soft in structure and moderately watery immediately after cutting.

III. (Normal) muscles were uniform in color (grayish-pink) and moderately firm in structure.

IV. (Dark) muscles were firm in structure and moderately to extremely dark in color. Muscles were dry immediately after cutting.

The next slide (Slide II - Photographic Standards) photographically illustrates these muscle "classes". The dark inner muscles are the gluteus accessorius and gluteus profundus, whereas the light (outer) muscle is the gluteus medius.

Slide III - (Characterization, pH values):

pH Differences (Gluteus Medius) between Muscle Classes.

pH Values	Ham muscle classes			
	I (Pale two-toned)	II (Two- toned)	III (Normal)	IV (Dark)
pH at slaughter	6.19	6.36	6.29	6.35
pH, 40 minutes	<u>5.73</u>	<u>5.90</u>	6.09*	6.07*
pH, 24 hours	<u>5.42</u>	<u>5.53</u>	<u>5.61</u>	5.93*

Part of the initial characterization study of these muscle classes, as shown in the next slide (Slide III - Characterization), revealed that the pH values at the time of slaughter were similar for all muscles regardless of the

final (24 hours) classification. The pH values of the gluteus medius muscles from hams in Class I (Pale two-toned) and Class II (Two-toned), 40 minutes after slaughter, were significantly ($P < 0.05$) lower than those of muscles from Classes III (Normal) and Class IV (Dark) hams. The increase in acidity appeared to continue in Classes I and II during the 24 hour chilling process. At the conclusion of the chilling period (24 hours), the pH values of the gluteus medius muscles of Classes I and II were significantly ($P < 0.01$) lower than those in Classes III and IV.

Conversely, the glycogen concentration of the gluteus medius muscles were significantly higher in Classes I and II than in Classes III and IV, at the time of slaughter.

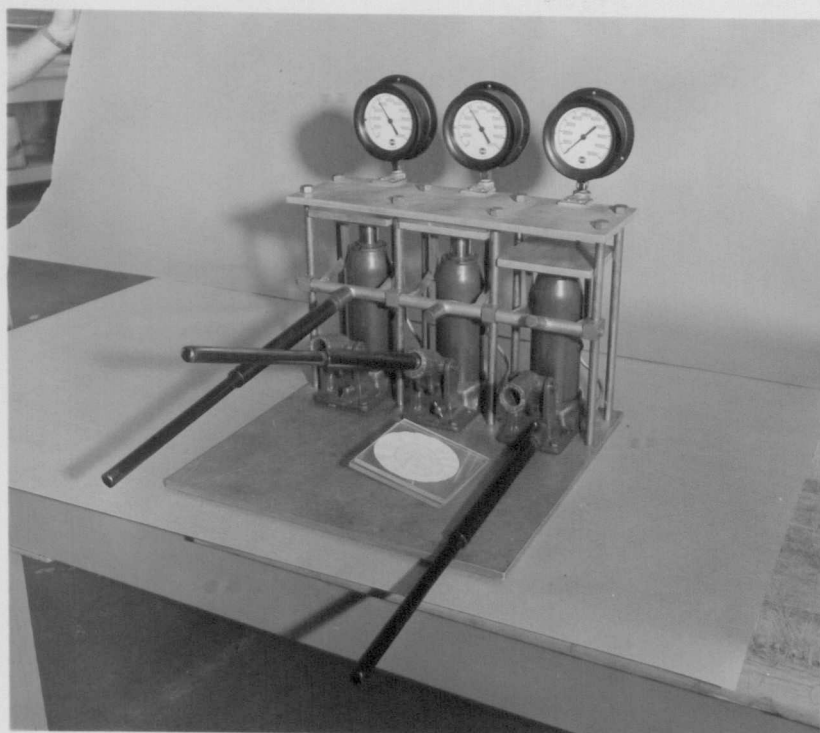
The next slide (Slide IV) shows the differences in myoglobin concentrations between muscle classes. (Characterization, Myoglobin):

Myoglobin Comparison of the Gluteus medius Between "Muscle Classes".

	Ham muscle classes			
	I	II	III	IV
	(Pale	(Two-	(Normal)	(Dark)
Myoglobin	two-toned	toned)		
mg/gm	.91	.99	1.11	1.20

Since the main visual criterion for muscle class distinction was that of color, special attention was directed to the absence of a significant increase in pigment concentration with an increase in the intensity of the color. It should be recognized, however, that there seemed to be a trend toward increased myoglobin concentration in Class IV. The significance of this trend will be discussed later in this report.

Slide V - (Apparatus - modification of methods of Grau and Hamm, 1953)



In an effort to measure the change in "water-retention" the above apparatus was developed as a modification of the "Filter Paper Moisture Absorption Technique" presented by Grau and Hamm. A large number of samples were herein, checked simultaneously with increased efficiency.

It was found that there was a gradual decrease in expressible H_2O from Class I (Pale two-toned) to Class IV (Dark).

This study suggested a definite relationship between color, pH, expressible H_2O and glycogen.

With these characteristics in mind, the next step in this problem was to find means of experimentally producing certain muscle classes. In this way tissues of known origin and nutrition could be studied. Since the two most striking differences in these muscles were the glycogen concentration at slaughter and the subsequent acid accumulation, it appeared logical that the depletion

of glycogen before slaughter would be a means to this end.

Slide VI - (Side view of the animal exerciser): This slide shows the side view of the animal exerciser that was designed and constructed for this purpose.

Slide VII - (End view of the animal exerciser): This apparatus effectively exercised from 2 to 6 hogs simultaneously under controlled conditions. The speed and duration were herein accurately controlled.



Animal exerciser

The following slide, (Slide VIII - Representative hams from each treatment): shows the results of an experiment designed to study the effectiveness of various levels of exercise in altering muscle appearances.

Lot I - Control

Lot II - 1 mile/day - 4 days

Lot III - 2-3 miles/day - 4 days

Lot IV - 3 miles prior to slaughter

The differences due to treatment were very striking. Eleven of the 12 hogs in the first three lots were either pale or two-toned, whereas, the hogs in Lot IV were either normal or dark. Seemingly the animals which received repeated exercise adjusted and restored their glycogen stores. This was supported by the fact that in Lot IV the muscles were apparently depleted of glycogen and were dark and dry with a significantly higher pH.

In another experiment exhaustion was used as the end point rather than any specific degree of stress. This exhaustive exercise was administered under normal and high sucrose regimens. Representative hams from this experiment are shown in the following slide. (Slide IX -- Representative hams from sucrose and exercise treatments).

Data from this experiment implied that exhaustive exercise, with normal rations, created dark, dry tissue. In contrast, the high sucrose rations seemed

to produce muscles which were pale in color, soft in structure and extremely watery in appearance.

Since exhaustive exercise and high sucrose rations were effective in producing dark and light hams, respectively, it seemed desirable to compare the effects of exhaustive exercise on low, medium and high carbohydrate rations. The following experiment was conducted to accomplish this goal.

Forty Poland China barrows ranging in weight from 190 lb. to 240 lb. were secured from purebred breeders, fed a 14% protein basal ration for a two-week period, stratified according to weight, and then randomly allotted into four lots each of ten pigs. The pigs were individually fed one of the following experimental rations (as shown on the next slide, Slide X - Rations) for a two-week period:

Table 1. EXPERIMENTAL RATIONS

<u>Ration I</u>	<u>Ration II</u>
14% Protein	30% Protein
4.2% Fat	30% Fat
<u>Ration III</u>	<u>Ration IV</u>
14% Protein	14% Protein
15% Fat	50% Sucrose
25% Sucrose	1.7% Fat

Lot I received a 14% protein basal ration and served as the control for this experiment. All the rations contained corn, oats, tankage, alfalfa meal, dried skim milk and soybean oil meal. Lot II received a low carbohydrate ration, as tankage, soybean oil meal, alfalfa meal and fat were increased at the expense of corn and oats. Lot III received a 14% protein ration that was considered medium in fat ($\frac{1}{2}$ of the fat content of ration II) and medium in sucrose ($\frac{1}{2}$ of the sucrose content of ration IV); Lot IV received a 14% protein - 50% sucrose ration.

At the conclusion of the feeding period, five from each lot were exercised to exhaustion immediately prior to slaughter. As the animals were bled, samples

from the gluteus medius, biceps femoris and rectus femoris muscles were removed from the right ham. After the carcasses were chilled for a 24-hour period at 34° - 38° F., the left hams were removed and the exposed ham butt muscles were classified. Chilled samples (24 hr.) were subsequently removed from the previously mentioned muscles as well as the pectoralis profundus.

The next slide (Slide XI) shows the positions of the muscles sampled. The two ham muscles, (Biceps femoris and Gluteus medius) and the Pectoralis profundus belly muscles are shown in the external musculature of this pork carcass.

It can be seen from the next slide (Slide XII - Effect of various rations and exercise on the properties of the Biceps femoris) that exhaustive exercise raised the muscle pH regardless of ration, however, it was most effective for both the basal and low carbohydrate lots.

Properties of the Biceps femoris

Lot and Ration	pH (24 hr.)	Glycogen mg/gm (Fat-free dry)	% of total
Lot I (Basal)			
Unexercised	5.70	19.72	59.07
Exercised	6.15**	7.37**	47.50*
Lot II (High protein, High Fat)			
Unexercised	5.93**	10.19*	57.65
Exercised	6.10**	5.77**	51.96*
Lot III (Med. Fat, Med. Sucrose)			
Unexercised	5.56	22.37	60.29
Exercised	5.80	11.56*	58.87
Lot IV (High Sucrose)			
Unexercised	5.64	27.17	59.27
Exercised	5.75	12.93	59.47

This was supported by sizable reductions in the glycogen concentrations due to exercise (Lot I and Lot II) and slight reductions in the percentages of expressible H₂O. The gluteus medius responded similarly to the biceps femoris in response to ration and exercise. The rectus femoris and the pectoralis profundus.

although they responded to exercise, did not show significant differences due to ration.

The next slide (Slide XIII - Representative Hams - Ration Experiment) shows representative hams from each treatment and ration. It can be seen that the hams from the unexercised hogs of Lots III and IV (medium and high sucrose) were definitely lighter in color and softer in structure than those from the basal and low carbohydrate rations. Similarly, it can be seen that hams from the exercised hogs were significantly darker and firmer than the unexercised, regardless of ration, however, the darkest hams were from the low carbohydrate lot.

Another experiment was conducted to check the effect of methyl-thiouracil in creating the "soft-watery" condition. An additional lot was added to study the effectiveness of exercise in correcting the condition brought about by the use of this agent. This slide (Slide XIV - Representative hams from methyl-thiouracil experiment) shows representative hams from each treatment. It can be seen that hams from hogs receiving 1 mg. of methyl-thiouracil per day for 10 days prior to slaughter were pale, soft and watery when compared to their controls.

This confirms the results of Ludvigsen, Denmark). Similarly it can be seen that exercise apparently depleted the glycogen (accumulated due to the reduced metabolic rate) and the hams were darker and drier in appearance than the unexercised hogs from the same lot.

Conversely the next slide (Slide XV - Hams from insulin experiment) shows the effectiveness of 250 and 300 units of protamine Zn insulin in apparently reducing the glycogen, raising the pH and increasing the intensity of the color.

I might also add that the Wisconsin station (Robert Sayre, graduate student) has recently demonstrated a similar type of response, although not as pronounced, by suspending pigs in an ice cold water bath prior to slaughter.

The last experiment which I wish to discuss is concerned with the time

course of these post-mortem changes. The experiment was designed to study the effect of exhaustive exercise and sucrose feeding on pH, glycogen and ATP at $\frac{1}{2}$ hr., 8 hrs., 16 hrs. and 24 hrs. after slaughter. (This experiment has not been completely analyzed, so only preliminary results will be given).

The next slide (Slide XVI) shows changes in pH with time after slaughter.

Post-Mortem Changes in the (24 hr.) pH of the Biceps Femoris).

Time after slaughter (hours)	T R E A T M E N T		
	Control	Sucrose-fed	Exercised
$\frac{1}{2}$	6.61	6.09	6.18
8	5.88	5.62	5.87
16	5.61	5.42	5.90
24	5.45	5.35	5.86

The first samples were taken 20-30 minutes after slaughter. The ultimate pH of samples from the sucrose-fed and control lots were definitely lower than those of the exercised lot. I wish to point out that these values are averages and that individual muscles which became especially pale and watery dropped to a pH of around 5 at either the 8 or 16 hr. period. These muscles subsequently increased to 5.2 or 5.3 at the 24 hour period. It also seemed that the color change was simultaneous with this drastic reduction in pH. Since at this point there was a definite increase in percent "free" H_2O , further studies are being conducted to check the possibility of myoglobin extraction into extracellular water as an explanation of the loss of muscle color.

The next slide (Slide XII) shows the changes in glycogen during post-mortem chilling.

Post-Mortem Changes in the Glycogen Concentration of the Biceps Femoris.

Time after slaughter (hours)	T R E A T M E N T		
	Control	Sucrose-fed	Exercised
$\frac{1}{2}$	37.60	29.05	8.30
8	25.83	15.48	1.33
16	15.56	11.87	1.17
24	8.22	5.02	.26
	(Mg/gm (Fat-free, dry tissue))		

One can see that the major part of the glycogen was depleted in muscles from the exercised hogs. Although there was a continuous decrease in muscle glycogen concentration, it seemed that the first eight hours were the most important. It also appeared that anaerobic glycolysis was slightly more rapid in the muscles from the sucrose fed hogs. It is further interesting to point out that the muscles in the exercised hogs possessed greater myoglobin concentrations. The presence of the increased pigment quantity, although not believed to be due to the single exercise treatment, may have an effect on the reduced level of glycogen at slaughter by permitting increased aerobic and anaerobic metabolism prior to and after slaughter.

The next slide (Slide XVIII) shows the decrease in % ATP - P of T.S.P. during post-mortem chilling.

Post-Mortem Changes in the ATP - P as % of T.S.P. in the Biceps Femoris.

Time after slaughter (hours)	T R E A T M E N T		
	Control	Sucrose-fed	Exercised
$\frac{1}{2}$	18.69	18.43	6.72
8	5.72	8.19	6.27
16	3.53	6.31	7.50
24	3.53	2.97	7.50

Since glycogen was severely depleted in the musculature of the exercised hogs, it was expected that there would be only a negligible change in ATP during post-mortem chilling. It is also interesting to point out, however, that during exercise there was also a major reduction in ATP. The fact that there was a slower post-mortem reduction in ATP and a faster reduction in glycogen in the muscles from the sucrose-fed pigs suggests that perhaps glycolysis was proceeding at a faster rate which accounts for a greater resynthesis of ATP.

In Summary:

The muscles from pigs fed sucrose or methyl-thiouracil contained moderate to high initial glycogen concentrations and were ultimately soft, pale and "watery" in appearance. During post-mortem chilling the muscles from the sucrose fed pigs decreased rapidly in pH. These muscles decreased slowly and then rapidly in ATP and increased consistently in expressible water.

The pigs which were exhaustively exercised or fed high levels of insulin possessed muscles with a small amount of glycogen at the time of slaughter. These muscles were darker and drier in appearance, higher in pH, and lower in expressible water after post-mortem chilling. Similarly there were negligible changes in the ATP content of these muscles (exercised animals) during post-mortem chilling. Although pH and related water-binding characteristics appear to be the dominant factors in determining color appearance, concentration and distribution of myoglobin may influence directly or indirectly the resulting color.

In closing, I wish to thank you for inviting me to participate in this conference. This has been an honor and privilege which I will forever cherish.

(RESUME)

Traitements avant l'abattage relatifs au processus
de la rigidité cadavérique des muscles du porc.

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Ce rapport examine les effets des différents traitements avant l'abattage sur le processus de la rigidité cadavérique dans plusieurs muscles du porc. Les traitements avant abattage comprennent plusieurs exercices menés à différents niveaux: exercice physique unique et intense de l'animal; rations de sucrose (à dose fortes ou modérées); rations faibles d'hydrocarbures; méthyl-thiouracil; choc par l'insuline ou par l'eau froide. Des analyses physiques et chimiques de l'élément aqueux agglutinateur et de ses propriétés associées ont été faites sur différents muscles du porc à différents moments du refroidissement qui suit la mort.

Les muscles des porcs nourris de sucrose ou de méthyl-thiouracil contenaient des concentrations initiales de glycogène moyennes ou élevées. Les muscles finissaient par devenir mous, pâles et d'aspect aqueux. Pendant le refroidissement qui suit la mort, les muscles des porcs nourris de sucrose diminuaient de volume lentement d'abord, puis avec rapidité dans l'ATP pendant que la quantité d'eau exprimée croissait.

Les porcs qui avaient été soumis à des exercices intenses ou qui avaient reçu des injections à fortes doses d'insuline possédaient des muscles contenant peu de glycogène au moment de l'abattage. Ces muscles avaient un aspect plus sombre et plus sec, leur contenu de pH était plus élevé et la quantité d'eau exprimée moindre après le refroidissement qui fait suite à la mort.

(SUMMARY)

Pre-Slaughter Treatments as Related to Post-Mortem Changes in Pork Muscle.

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The effects of various pre-slaughter treatments on the post-mortem changes in several pork muscles were discussed in this report. The pre-slaughter treatments included various levels of exercise; single, severe exercise; sucrose rations (high and medium levels); low carbohydrate rations; methyl-thiouracil; insulin and cold water stress. Physical and chemical analysis of the water-binding and associated properties were conducted on several different pork muscles at various stages during post-mortem chilling.

The muscles from pigs fed sucrose or methyl-thiouracil contained moderate to high initial glycogen concentrations and were ultimately soft, pale and watery in appearance. During post-mortem chilling, the muscles from the sucrose fed pigs decreased slowly and then rapidly in ATP and increased consistently in expressible water.

The pigs which were exhaustively exercised or injected with high levels of insulin possessed muscles with a small amount of glycogen at the time of slaughter. These muscles were darker and drier in appearance, higher in pH and lower in expressible water after post-mortem chilling.

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In dieser Arbeit wurde der Einfluss verschiedener Bedingungen vor der Schlachtung auf post-mortem Veränderungen in einigen Muskeln von Schlachtschweinen diskutiert. Die Bedingungen erstreckten sich auf verschiedene Grade von erzwungener körperlicher Bewegung (mässige bis harte Bewegung); Zucker in der Futterration (grosse und mittlere Mengen Sucrose); Futterrationen mit niedrigem Gehalt an Kohlenwasserstoffverbindungen sowie mit Methyl-Thiouracil; Insulin und Kaltwasserschok. Zu verschiedenen Zeitpunkten während der post-mortem Reifung der Schlachtkörper wurden physikalische und chemische Untersuchungen über das Wasserbindungsvermögen und den damit verbundenen Eigenschaften an mehreren, verschiedenen Muskeln durchgeführt.

Die Muskeln der mit Sukrose oder Methyl-Thiouracil gefütterten Tiere enthielten zu Anfang mässige bis hohe Konzentrationen von Glykogen und waren weich, blass und wasserig in ihrer äusseren Erscheinung. Während der post-mortem Reifung verloren die Muskeln der mit Sukrose gefütterten Tiere zuerst langsam und dann rapide an ATP-Gehalt. Zu der gleichen Zeit nahm der Gehalt an auspressbarem Wasser stetig zu.

Die Tiere, die bis zur Erschöpfung getrieben waren oder die grössere Mengen Insulin erhalten hatten, hatten Muskeln, die nur geringe Mengen Glykogen zur Zeit der Schlachtung enthielten. Diese Muskeln waren auch dunkler und trockener in ihrer äusseren Erscheinung. Sie hatten ebenfalls höhere pH Werte und einen geringeren Gehalt an auspressbarem Wasser nach der post-mortem Reifung.