QUALITY OF FROZEN-DEFROSTED MEAT
FROM PROTAMONE-FED AND THIOURACIL-FED HOGS

by

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LIST OF PLATES

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

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CHAPTER I

Introduction

With the recent increase in the use of temperatures
below the freezing point in the preservation of fresh meat
many problems, some of which are related to the quality of
the product, have been brought into the foreground.

One of the major problems of the livestock and meat
industry is to distribute its highly perishable products
evenly throughout the year. Freezing has been used as a
means of preserving the different kinds of meat at the meat
packing companies to be sold when demand justifies.

Another major problem is the increase in the prices
of feeds necessary for meat producing animals. As a result
of much research several new breeds of animals, which uti-
lize their feed more efficiently and require less feed for
every hundred pounds of gain, have been developed. With
the increased knowledge in the field of endocrinology there
is a better understanding of the role of hormones in the
body. Various preparations such as thiouracil, thyroactive
proteins, and iodinated proteins have been studied to
determine their effect on the growth rate, location and

amount of fat deposited, and efficiency of gains in several different breeds. However, these drugs may have either undesirable or beneficial effects on the quality and characteristics of the dressed meat, fresh or stored. From a survey of the literature, as far as we can ascertain, these treatments have not been studied. The purpose of this study is to find out whether the oral administration of thiouracil or protamone to meat producing animals has any effect on the final quality of the meat. For this investigation pork from animals treated with either thiouracil or protamone was kept in freezer storage for a period of six months.

CHAPTER II

Review of Literature

Many chemical, physical, and biological changes take place in fresh meats beginning immediately after slaughter and continuing until the meat is cooked, frozen or processed in some other form. In meat, as in all commodities, all reactions taking place proceed in one direction only, that leading to the disintegration or decay of the product.

Refrigeration may be considered one of the fundamental methods for preserving the original characteristics of fresh meat, but it should be understood that it is not a foolproof procedure. There are many factors, both anti-mortum and post-mortum, which affect the quality of the frozen meat.

The first changes of the animal tissues after death are of a physico-chemical nature such as the appearance of coagulation, change in color, and change in pH value. The appearance of coagulation is most distinctly marked in the stiffening of fat in fat cells and in the coagulation of myosin in striated muscles. The latter is probably brought on by acid formation in the muscle, resulting in rigor mortis (19)[*]. The muscles consequently show an acid reaction. Rigor mortis is probably the result of a

[*]Numbers in parentheses refer to bibliography.

coagulation of myosin through the formation of lactic acid in the muscles. The time of appearance of rigor mortis depends on the muscular activity before death; the stronger the activity of the muscles during life the sooner rigor mortis sets in. High atmospheric temperatures favor the appearance of rigor mortis while cold retards it. Whether the dissolution of rigor mortis results from an increase of acid formation in the muscle which again affects the solution of myosin, or whether it is due to other influences is still the subject of controversy.

Upon chilling, the fatty tissues of a carcass become opaque and firm. The muscles harden before the chilling process is complete because of chemical changes. Various theories have been put forward by several workers to define the nature of changes occurring in the muscle fibers apart from the formation of ice crystals, but the lack of exact knowledge of the structure of the muscle fiber, particularly in regard to the constituent proteins, has tended to restrict the usefulness of such theories. It was generally supposed, however, that the structural alterations within the muscle fibers partly resulting in a diminished capacity for holding water would be greatest when comparatively large ice crystals were formed as in slow freezing and least when an amorphous glass-like structure was produced as in the case of extremely rapid freezing (22). Smorodintsev and

Bystrov (67) found that the modifications in the properties of meat resulting from freezing such as swelling and solubility of myosin and myogin are at a minimum when freezing was carried out at -11° C. The solubilities of both myosin and myogin vary slightly with time of freezing, according to Smorodintsev and Bystrov (68) but follow different laws. Variations in the properties of meat (swelling and distribution of protein N) are minimized with very short (50 minutes) or very long (24 hours) freezing; intermediate values are detrimental to meat quality.

Quality is a broad term and is often used when a specific term would be more appropriate. A number of tests have been applied quantitatively to meat and arbitrary standards have been set up.

In this work a limited number of factors having to do with the quality of meat have been considered. A review of the literature concerning these factors follows.

1. Color of Meat

Good color of meat, while it might not affect its palatability or nutritive value, is generally recognized and demanded by the consumer. The most desirable color for pork meat is grayish-pink or cherry-red and dark-red for older hogs.

The reddish color (hue) of muscle is due to the

presence of a complex and relatively unstable compound,
hemoglobin, which is present as (a) muscle hemoglobin, and
in this case it is found within the muscle fibers; and (b)
in any blood corpuscles remaining in the capillaries (7, 8,
82).

The depth of color (corresponding to brilliance and
saturation) depends on the concentration of hemoglobin and
on the thickness of tissue through which light is reflected
to the eye by optical heterogeneities within the muscle.
The thicker the surface layer (i.e. the less opaque the
tissue) and the greater the concentration of pigment, the
deeper the color will be. The concentration of hemoglobin
depends on the breed, age, and condition of the animal, and
varies in the different muscles of the same animal. The
opacity of the tissue is greatly altered by the loss of
water (8, 82).

Shenk, Hall and King (65) concluded that blood hemo-
globin and muscle hemoglobin are two different compounds.
They report that Gunther suggested that the pigment frac-
tions of muscle and blood hemoglobin are identical but that
the globin fractions are different. Very conclusive proof
of the distinction between the two hemoglobins was presented
by Hektoen and co-workers (31). Shenk and associates (65)
gave added evidence that muscle hemoglobin is quite distinct
biologically from blood hemoglobin particularly in beef
muscle.

Mackintosh and Hall (40) believe that no relationship exists between blood hemoglobin and the color of muscle; their data indicate that seldom more than seven per cent of the total hemoglobin in the tissue is blood hemoglobin. Brooks (7) suggests that the amount of muscle hemoglobin appears to be independent of the degree of blood removal. This point needs further investigation since in the past it has been mentioned that there are some indications that delayed bleeding might influence the color of meat.

Brooks (8) states that there are two main causes, in commercial practice, responsible for color change in meat: Oxidation of Hemoglobin to Methemoglobin: The undesirable properties of hemoglobin from the point of view of preserving color are counterbalanced to a large extent by another factor--the muscle tissue's uptake of oxygen. Muscle, after rigor mortis, retains indefinitely a residue of its respiratory activity. When exposed to air, therefore, a steady state is reached where the depth (d) to which oxygen penetrates is determined by the relative rates of its diffusion and uptake. The relation is given by the equation

$$d = \sqrt{\frac{2 c_0 D}{A}}$$

where (c_0) stands for the pressure of oxygen at the surface of the tissue, (D) stands for coefficient of diffusion through the tissue and (A) stands for coefficient of oxygen consumption. For pork the value of (A) is reported by

Brooks (7) to be roughly 10^{-4} cc/gram/minute at 0° C, the
uptake decreasing slowly with time. Corresponding value of
(d) for pork is roughly 0.2 cm at 0° C. The depth to which
oxygen penetrates decreases with increasing temperature. It
was also found that the depth to which oxygen penetrates in-
creases slowly with time, but that it rarely exceeds 1 cm
even after very long periods of storage (10, 11).

It is now rather well known that, even at freezing
temperatures in the presence of air, hemoglobin is oxidized
to the brown colored methemoglobin. Brooks (10) believes
that, although oxidation does take place very slowly at low
temperatures in frozen meat, the formation of methemoglobin
is much less important (unless the time of storage is unduly
long) than loss of color due to excessive drying. In the
latter case crystals of ice in the superficial layer evapo-
rate and the small bubbles of air left behind scatter the
incident light. Hankins and Hiner (29) are of the opinion
that the change in color of muscle tissue associated with
oxidation of hemoglobin probably occurs more noticeably in
beef than in pork or lamb. In the case of pork, besides the
change in the color of lean, the oxidation of fat is ac-
companied by a change to a yellowish color.

In the lean meat the oxidation of hemoglobin takes
place in the surface layer of tissue containing dissolved
oxygen (7). Brooks (7, 8) considers the color of meat to

be brownish when roughly 60 per cent of the hemoglobin present in the superficial layer has been oxidized to methemoglobin.

From the work of various investigators there are evidently several factors which affect and determine the color of both fresh and stored meat from a given animal. In the fresh lean meat exposed to air the pigment, hemoglobin, is present as oxyhemoglobin and reduced hemoglobin. The penetration of oxygen into the muscle is confined to a well defined surface layer by the oxygen consumption of the tissue. The red oxyhemoglobin is found only in this layer while in the underlying tissue, which contains no dissolved oxygen, the pigment is present as the purplish reduced hemoglobin. The color (hue) depends on the thickness of the oxyhemoglobin region; if this is greater than the effective thickness through which light is reflected the color is a bright red, whereas if the reflected light has traversed tissue containing both oxy- and reduced hemoglobin the color is intermediate between red and purple (8).

a. Oxygen Pressure: The rate of formation of me-themoglobin increases with decreasing pressure of oxygen over a wide range. In tissue, the concentration of oxygen decreases with increasing distance from the surface; hence in air, the rate of oxidation in the oxygen region in-creases with increasing distance from the surface. Also,

the smaller the pressure of oxygen in the gas in which the
tissue is stored the nearer to the surface is the region
where methemoglobin is most rapidly formed since the depth
of the oxygen region is proportional to the square root of
the oxygen pressure (8). The influence of oxygen pressure
on rate of oxidation is interesting from the point of view
of the mechanism of the reaction. The rate increases with
decreasing pressure, as mentioned before, but reaches a
maximum at low pressure (of the order of 4 mm O_2 at 0° C)
(7, 9). Brooks (7) observed that methemoglobin formed
slowly in the oxygen region of tissue exposed to air and
most rapidly at some distance from the tissue-air inter-
face. He also found that freezing and thawing seem to in-
crease the rate of methemoglobin formation.

The relation between oxygen pressure and the rate
of methemoglobin formation is responsible for the rapid
discoloration of tissue stored at 0° C in gases containing
a small amount of oxygen. Methemoglobin formed directly on
or very near to the surface alters the color of reflected
light to a greater extent than the same amount of pigment
produced in the same time but some distance, e.g. 2 mm, or
more below the surface (8).

It is advisable at this point to mention that the
relation of methemoglobin to oxyhemoglobin and reduced
hemoglobin has been the subject of a controversy and still

appears to be in progress to this day (13). Conant and
Scott (13) report that several workers believe that methemo-
globin contains one half the oxygen of oxyhemoglobin while
others believe that methemoglobin contains only one quarter
of the oxygen of oxyhemoglobin.

b. Effect of pH: Brooks (7) found that a decrease
in pH increases the rate of oxidation of hemoglobin in mus-
cle. Winkler (80) agrees that there is a relation between
pH and color of meat from a given muscle in pork but he
believes that there are factors other than pH which are
equal to or more important than pH in determining the color
of meat from a given animal. He adds that within a pH
range of 4.5 to 5.5 meat samples are lighter in color while
above a pH of 5.5 the meat samples become darker. He also
believes that the change in color over a pH range of from
5.5 to 8.5 is mainly in intensity rather than in composition
of the color.

c. Effect of CO_2: It was believed that carbon di-
oxide gas may influence the color of meat but it was found
by Brooks (8) that the oxidation of hemoglobin in muscle was
not affected when the concentration of CO_2 was below 20 per
cent.

Desiccation of Meat: Meat must be packaged very carefully
or excessive desiccation will occur resulting in the ex-
posure of hemoglobin, present in the surface layer, to the

action of air with consequent oxidation of hemoglobin to the brown pigment methemoglobin. Brooks (7) reports that drying in frozen muscle results in a decrease in the depth of color. Hankins and Hiner (29) report that desiccation causes change in the color of meat giving it a pithy appearance, particularly the lean portion. Excessive drying gives an unattractive appearance because of optical changes in the tissue.

2. The pH Value of Meat

The muscle tissue of the live animal is neutral in reaction according to Ziegler (82) but becomes acid after death. Edelmann (19) believes that the reaction of living muscle is slightly alkaline or neutral and under normal conditions is changed to acid within three to six hours following death through the formation of lactic acid, formic acid and potassium acid phosphate. Empey (22) states that the increase in the hydrogen-ion concentration takes place in the muscle fiber during the onset of rigor mortis. Ziegler (82) reports that acid starts to form after rigor mortis sets in and that in the case of beef there is an increase in temperature of from 0.3 to 3.0° F in the muscle which is ascribed to glycogen-lactic acid reaction which changes the pH of muscle from 7.2 to 6.2 in 48 hours.

Ramsbottom and Koonz (59) state that increase in

acidity due to the conversion of glycogen to lactic acid in muscle tissue together with other chemical changes bring about changes in the physical characteristics of the muscle fibers which in rigor shorten and become knotted. Moran (44) adds that living muscle contains less than 0.05 per cent lactic acid but when the animal dies and goes into rigor the lactic acid content increases, reaching a maximum of about 0.56 per cent. One effect of freezing is to promote the formation of this acid. Sair and Cook (63) report that while pH values of beef are relatively constant and close to the value at which maximum drip occurs (5.2 to 5.5), pork and mutton vary in pH from carcass to carcass, and are generally more alkaline and tend to become more so during storage.

Effect of Freezing, Storage, and Thawing on pH: Ramsbottom and Koonz (57) observed that the pH value of beef remained almost constant from the first day after slaughter to the thirty-fifth day. Freezing and thawing did not change the pH value of meat significantly. They also found that no significant change took place in the pH of beef stored for one year at 10 or -30° F (58). Shrewsbury and associates (66) report that no decided change occurred in the pH of the lean meat as the storage period of pork increased when storage temperatures of -6.3 and -8.4° F were used. They

add that there was a slight tendency for the pH to increase
with time of storage but that this was not considered to be
significant. They also concluded that there was no im-
portant difference in the acidity of products from hard or
soft hogs.

Relation of pH Value to Quality of Meat: It has been shown
that the pH of muscle is correlated with color changes (7,
80), drip formation (22,63) and the activity of enzymes,
particularly lipases. A rise in pH is usually associated
with meat decomposition according to Drosdov (17), but the
value associated with true spoilage is variable. He con-
siders the pH of spoiled meat as being 6.3 or higher.

3. Tenderness of Meat

Tenderness is generally recognized as one of the most
important characteristics of meat which commands the great-
est consumer interest. It has a wide inherent variation
among animals of the same species as well as in cuts from
different parts of the same carcass, as reported by Tressler
and associates (72). Fresh meat is usually rather tough
(82) but Mackintosh and co-workers (41) state that in the
case of beef the younger the animal the more tender is the
meat. They add that there is apparently a relationship of
fat or finish to tenderness of the meat and that the

marbling in the muscle and the grade of the carcass have a
definite effect on the value of tenderness. They report
that increased finish apparently renders the meat more
tender.

Mackintosh and co-workers (41) report that Lehman and
associates of the Hygiene Institute of Wursburg concluded
that tenderness of meat is closely related to the connective
tissue content of the flesh. Edelmann (19) states that not
only the amount of connective tissue present in the meat but
also the age, nutritive condition, and breed are associated
to a large extent with the tenderness and toughness of meat.
He adds that following death, lactic acid, formic acid, and
potassium acid phosphate form; the last one causing a swel-
ling and loosening of the connective tissue elements of the
meat thus rendering the meat more tender.

Aging at a temperature above the freezing point is
the oldest and best known method of increasing tenderness
in meat. Moran (44) believes that increasing tenderness
through ripening or conditioning of meat is due to the
action of lactic acid in the muscle, softening the con-
nective tissue elements. Ziegler (82) reports that the
tenderizing process consists of the dissolution of the
connective tissue (collagen) by the action of enzymes.
Ramsbottom and Koonz (59) conclude that it has been shown
that rigor is associated with toughness of beef. Their

tenderness tests with cooked samples of beef showed that beef muscle was more tender immediately after dressing than at any time thereafter for the following few days. They report that glycogen, which is present in amounts ranging from 0.3 to 1.0 per cent in the muscles of the living animal, is converted to lactic acid consequently increasing the acidity which, together with other factors, brings about changes in the physical characteristics of the muscle fibers.

Ramsbottom and Koonz (59) report that the soft tissues of the carcass contain enzymes which are still active in the lean, fatty, and connective tissues following slaughter. Cathepsin acts on the muscle fibers, softening them, thereby gradually improving the tenderness of the lean meat.

Because of the high fat content in pork and the susceptibility of the fat to become rancid, pork is not usually aged.

Considering the limited amount of work related to tenderness of meat, very little has to do with the tenderness of pork meat. For this reason, and the possibility that some of the observations made on beef muscle may apply to pork, the effect of freezing and storage of the various kinds of meat investigated to date will be reviewed.

Effect of Freezing: Tressler and DuBois (70) report that frozen pork is a desirable product provided it is of good quality, carefully frozen and properly stored. They state that freezing tends to make meat more tender but that this does not mean that poor tough meat will become choice, tender meat upon freezing. Poor meat under any circumstances will not make a good frozen product since freezing is primarily a method of preservation; therefore, discretion should be used in selecting animals to be slaughtered, in the handling of meat prior to freezing, in packaging, in wrapping, and in securing temperatures that will preserve and protect quality. Ziegler (82) reports that rapid freezing results in very little water separation and small but very expansive crystals which cause a rupturing of the cell walls and muscular connective tissues in many places resulting in the well known mechanical tenderizing action. Hankins and Hiner (28, 30) agree on the fact that freezing does have a tendering effect on meat and they add that low temperatures such as -10 and -40° F have significantly more tendering effect than high temperatures such as 20 or 45° F. They add that investigators in the Bureau of Animal Industry observed that splitting of the muscle fibers first took place at -10° F and that this physical effect became more and more apparent as the temperatures of freezing were lowered. They explain the increase in tenderness of meat

as being the result of this disintegration of fibers. Hiner
and associates (32) agree on this last point but add that
freezing also breaks or stretches the interstitial con-
nective tissue surrounding the muscle fibers and fiber
bundles. On the other hand, Paul and Child (54) working
with beef found that freezing had no effect on tenderness.
In another paper (12) the same authors reported that freez-
ing did not affect the tenderness of pork. Shrewsbury and
associates (66) agree on the fact that there was no definite
effect on tenderness of pork roasts or chops that could be
related to freezing. Noble and Hardy (49) working with pork
loin roasts report that when the roasts were frozen at 0° F
neither the tenderness nor any of the quality factors of
pork loin roasts were altered.

Effect of Storage: Tressler and co-workers (72) report
that quick freezing and the subsequent storage of the
frozen product effects a marked tendering of beef. They
add that the tendering effect of quick frozen meat con-
tinues during cold storage. Shrewsbury and associates (66)
report that storage had no definite effect on tenderness of
pork roasts or chops. Young and McIntosh (81) found no
significant differences in the shearing value of pork chops
frozen at 0° F and stored for various lengths of time, but
they found that in the case of roasts, those that were

stored for 4.5 months were less tender than the ones stored
for 1.5 months. The difference was highly significant but
no explanation was offered. Noble and Hardy (49, 50) found
no significant change in the tenderness of pork loin roasts
when frozen at 0° F and kept for varying lengths of time at
different storage temperatures.

Effect of Thawing Temperatures: Using different thawing
temperatures Paul and Child (54) state that there was no
significant difference in tenderness of frozen pork thawed
at 175° C, and at $24-25^\circ$ C, and unfrozen pork. Working with
beef and pork, the same authors proved these observations
(12). Vail and associates (73) using different temperatures
of thawing under varying conditions also concluded that
there was no significant difference in tenderness of pork
roasts thawed at different temperatures (room temperature,
refrigerator, or an oven at 350° F).

4. Drip in Meat

Drip denotes the clear, reddish-colored fluid which
exudes from all cut surfaces of frozen meat during thawing.
The presence of drip has been shown to be due to changes
which take place in the meat during freezing and thawing,
but Empey (22) states that even before freezing takes place
several post-mortum changes affect the capacity of the

muscle fibers for holding its fluid content. He reports that increase in hydrogen-ion concentration in the muscle during the onset of rigor mortis is considered one factor of great importance in this respect.

Drip has been shown to be similar both in its chemical and physical constitution to the relatively free fraction of muscle fluid which could be expressed from the cut surfaces of the same unfrozen muscle. Empey (22) proved that susceptibility of a muscle to drip depends on the percentage and ease of expression of this loosely held muscle fluid.

Only a few papers concerned with drip in meat are available in the literature (18, 53, 56, 57, 64). Kaloyercas (33) believes that drip is a property which could be used satisfactorily in almost all cases of quality control of frozen foods. He adds that drip practically corresponds to the result of changes occurring during freezing and its interpretation does not require necessarily any record of the pre-freezing condition of the product as is the case with other methods used for quality control.

The cause of drip is generally attributed to the formation of large ice crystals during slow freezing (71), which exert a mechanical force (22, 71), that may rupture the cells (46, 69, 71), or prevent the cell constituents from reabsorbing the moisture (63) either because the rate

of thawing exceeds the rate of diffusion back into the cells
(69), or because the proteins are irreversibly dehydrated
or denatured by the processes involved in slow freezing
(23, 45).

Whether drip results only from one or a combination
of the above mentioned effects is not yet well known, but
it has been found that rapid freezing reduces both the size
of the ice crystals and the amount of drip obtained when the
product is thawed (69). By analogy it would appear that all
meats should drip when thawed after slow freezing, but this
does not appear to be the case since slowly frozen pork and
mutton were found by Stiles (69), and Cook and co-workers
(14) not to drip to any extent when thawed.

There has been, and there still is some divergence
of opinion among investigators regarding the relative im-
portance of factors which influence the amount of drip that
exudes from frozen meat upon being defrosted.

Effect of Time between Slaughter and Freezing: Sair and
Cook (63) observed that as the period of time between
slaughter and freezing is increased, the amount of drip, in
beef, decreased. Ramsbottom and Koonz (57, 58) made the
same observations and they add that while drip decreases as
the time between slaughter and freezing increases, ice
crystals in the meat become progressively larger as the

time after slaughter increased, although the drip decreased, indicating that large ice crystals do not always accentuate drip. Empey (22), on the other hand, states that the period of time between slaughter and freezing has no definite influence on the susceptibility of a muscle to drip.

Effect of the Size of Meat Cut: Ramsbottom and Koonz (57) report that in large cuts of meat, where the area of cut surface is small in relation to volume of meat there was little drip; in small cuts, where the area of cut surface is large in relation to volume, the amount of drip was dependent to a large extent on the freezing temperature. Their explanation is, that in the large cuts, the muscle tissue has opportunity to reabsorb the "frozen out" water; in small cuts the fluids may be more readily lost by the tissue as drip.

Rate and Temperature of Freezing: Ramsbottom and Koonz (56) report that Kallert pointed out that drip may be lessened by freezing fast enough to prevent the separation of water from the muscle fibers. Cook and co-workers (14) confirmed these observations in that increase in the rate of freezing appeared to be the most important factor in the reduction of drip. Empey (22) reports that Reuter demonstrated that a decrease in the rate of freezing was accompanied by an

increase in size of the ice crystals within the muscle
tissue and showed that extremely rapid freezing produced an
amorphous glass-like structure from which water was not ex-
truded, but frozen wholly within the muscle fibers. Empey
(22) does not agree with the previous workers but states
that rate of freezing does not have any definite influence
on the susceptibility of muscle to drip. As to the tempera-
ture of freezing, Paul and Child (54) report that drip is
unaffected by freezing temperatures. Ramsbottom and Koonz
(58) disagree on this point and state that the amount of
drip is significantly affected by the temperature of freez-
ing. Tressler and Evers (71) explain the advantage of
rapid freezing in that it results in the formation of minute
crystals uniformly distributed throughout the tissue con-
sequently fixing the original spatial distribution of the
colloid in the meat tissue.

Effect of Storage Temperature: Moran and Hale (47) state
that increased drip occurs at high storage temperatures and
fluctuating storage temperatures. They believe that high
temperatures of storage induce changes in rapidly frozen
muscle. Tressler and Evers (71) report that when quick
frozen meat is held under poor storage conditions (low
humidity or fluctuating temperatures) which permit either
the growth of the ice crystals in the tissue or the irre-
versible denaturation of some of the proteins, the product

does not entirely return to its original gel condition.
Some of the liquid resulting from the thawing of the crys-
tals leaks out as drip. At this point one may add that
Empey (22) reported that the denaturation of the muscle
proteins, as the direct result of freezing, may possibly
account also for the reduced capacity of the thawed muscle
fibers to retain muscle fluid. Other factors which may
cause denaturation are increased pressure, desiccation of
the muscle fibers, or the subjection of the fibers to the
action of concentrated salt solutions; all three factors
being possible owing to the change of state of the contained
water. Ramsbottom and Koonz (58) do not agree with previous
workers and state that the temperature of freezer storage
did not appear to be important in regulating the amount of
drip.

Effect of Length of Freezer Storage Period: Ramsbottom
and Koonz (58) report that the amount of drip is signifi-
cantly affected by the length of time meat is held in the
frozen state. They add that part of the drip formed is due
to the act of freezing while part is due to the length of
time meat is held in freezer storage, but they conclude that
it is difficult to evaluate precisely between the amounts in
each case.

Effect of pH: Empey (22) reports that drip production is

primarily a function of hydrogen-ion concentration, that
there is a zone in which drip is at a minimum (about 6.3
for beef), but that there was a specific difference existing
between different muscles in amount of drip even at the same
pH value. The same author suggests that a certain fraction
of the muscle fluid which, in the unfrozen muscle, is held
relatively loosely, probably by mechanical forces alone,
forms the potential drip of that muscle when subsequently
frozen and thawed. Empey (22) also adds that the extent of
drip has been considerably reduced, and in some cases elimi-
nated, by increasing within the muscle fibers prior to
freezing either the osmotic pressure or the pH value or a
combination of the two. Sair and Cook (63) confirmed these
observations when they found that the maximum drip was ob-
tained from beef having a pH value of approximately 5.2,
and as the hydrogen-ion concentration decreased the net drip
decreased to zero at about pH 6.4. They also found that
pork carcasses are generally more alkaline than beef and
tend to become so during storage and they say that this fact
explains the small amount or practical absence of drip from
frozen pork. The same workers (63) report that in unfrozen
material the amount of fluid exuded decreases rapidly during
the first day or two after slaughter, regardless of pH
changes. They add that meat at pH 6.4 or higher does not
drip as a result of freezing while at pH values of 5.2 to

5.5 the amount of drip reaches a maximum and in this region an increased freezing rate reduces the amount of drip obtained. They explain this behavior as being due to a high-water retaining capacity of the tissue proteins at pH 6.4 resulting in complete retention of the water produced on thawing, regardless of the size of the ice crystals found on freezing; at a pH value of 5.2 to 5.5 the water-retaining power of proteins is lower and moisture losses occur, the latter condition possibly being prevented by rapid freezing.

Rate and Temperature of Thawing: As early as 1908, Richardson and Scherubel (61) recognized the importance of thawing frozen meat slowly as a means of reducing the amount of drip. Empey (22) states that variations in the rate of thawing have no definite influence on the susceptibility of a muscle to drip. Child and Paul (12) using different thawing temperatures came to the conclusion that there was no significant difference in drip between pork thawed at 175° C, and at 24-25° C, or unfrozen pork. On the other hand, these workers (54) confirmed Richardson and Scherubel's findings that slow thawing reduced the amount of drip.

Other Factors Affecting Drip: Kaloyereas (33) believes that there appears to be an inverse relationship between the amount of drip and the bound water of the tissues.

Since for products of high bound-water content low freezing
temperatures affect the bound-water free-water equilibrium
to a greater extent than do high freezing temperatures,
quick freezing would not be beneficial for such products as
for those of low bound-water content. Kaloyereas adds that
factors which affect drip are primarily rate of freezing
(rapid freezing causes decreased drip formation) and stor-
age (increases drip). However, he adds that it must be
realized that the effect of quick freezing upon the drip is
not always the same with different products. He considers
this point of importance since there is a general belief
among various investigators that the most rapid freezing
always gives the best product.

5. The Fat in Meat

The firm, white saturated fats play an important
role adding palatability to the lean in meat because of the
flavor and aroma contained in its oils. The unsaturated
fats are soft and oily and tend to lower the grade of the
meat. Ziegler (82) states that meat from highly marbled
carcasses will be far more firm than from thinner, unmarbled
carcasses. He adds that marbling is associated with juicy,
highly flavored pork.

It was only in recent years that the real signifi-
cance of variations in fatness of meat animals and their

carcasses has been given much thought. The main concern was
to develop methods for fattening meat animals through feed-
ing since the opinion at the time was, the greater the fat-
ness of the animal, the more desirable the dressed carcass
and the meat. But it was realized that this opinion was not
founded on adequate facts and as a result research workers
became interested in the role of fat in relation to quality
and quantity factors of meat animal carcasses developed.
Hankins and Ellis (27) report that fatness is an important
factor affecting the proportion of dressed carcass in meat
animals. They add that in a report by H. W. Titus the
latter mentioned that it was evident that changes in pro-
portions of cuts do, at least in part, take place due to
differential fat deposition. Titus showed that as hogs
fatten there are increases in proportions of bacon and cut-
ting fat consisting of back fat, leaf fat, and fat trimmings,
while there are decreases in ham, loin, shoulder and head.
Hankins and Ellis (27) state that increase in fatness con-
tributes little to changes in color of lean meat in cattle
and lamb. They add that, from the general point of view,
it may be reasoned that increase in intramuscular fat con-
tent would normally have the effect of retarding the in-
tensification of the color of lean. In that zero (0) is
used to express the bottom of the brilliance scale of pure
black while ten (10) is used to express pure white, Hankins

and Ellis (27) conclude that it is possible that the percentage of black increases and brilliance values decrease more rapidly with the age of the animal when intramuscular fat content tends to remain constant.

Barbella and co-workers (4) have revealed relationships between the degree of fatness in beef rib cuts and desirability and intensity of flavor of both lean and fat, and quality, or richness and quantity of juice. They also showed that not only fatness but also the breed, age, and sex of animals are relatively important to the mentioned palatability factors.

Hankins and Ellis (27) also studied the relation between per cent of fat in the longissimus dorsi and tenderness of the same muscle in cattle, as well as per cent caul fat and tenderness of leg of lamb. They conclude that evidence is strong that variations in tenderness are caused mainly by factors other than fatness.

6. Rancidity in Pork Fat

The term rancidity is used to designate two entirely different changes which take place in fats and oils; (a) the hydrolysis of the glyserides with the liberation of free fatty acids, and (b) the oxidation of fats and oils containing unsaturated acids resulting in the formation of aldehydes, ketones, and acids having lower molecular weights

than the acids which were naturally present. As a general rule, oxidation and hydrolysis occur simultaneously although from the chemical standpoint the two mechanisms are sharply differentiated. In the process of hydrolysis there is always an increase in titratable acidity (25).

The oxidative process can be divided into two periods; (a) the period of induction where there is a negligible absorption of oxygen, and the susceptibility of a fat to oxidation may be determined by ascertaining under specific conditions the relative length of the induction period, and (b) the period of active oxygen absorption where rapid oxidation sets in and the fat becomes rancid (25, 37). The length of the induction period under a particular set of physical conditions depends on (a) the nature of the constituent glyserides and, (b) the presence or absence of traces of catalysts which either accelerate the reaction (salts of heavy metals) or retard it (certain phenols and aromatic amines) (37). Oxygen is necessary in order to produce the oxidation type of rancidity (25). White (78) found that the quantity of oxygen required to cause rancidity in pork fat is small.

Recent researches have shown that changes in the fats are primarily dependent upon such factors as source and kind of meat, the chemical composition of fat, micro-organism contamination, enzymatic activity, anti-oxidant or

pro-oxidant concentration, and other biochemical properties
(21).

Rancidity development is progressive and results in
a marked diminution in the palatability of the product. Fat
deterioration may be classified as follows;.(a) atmospheric
oxidation of unsaturated fats with subsequent development of
unpleasant aroma, (b) the action of micro-organisms, (c)
the presence of tissue enzymes which lead to the development
of free acidity during storage and may possibly play a part
in oxidation changes, (d) picking up of objectionable
foreign flavors.

Light is among the factors which accelerate oxida-
tive rancidity in fat-containing food stuffs. The reaction
itself is probably of the chain type; the absorption of one
quantum of light energy resulting in the reaction of a con-
siderable number of molecules. Once rapid oxidation has
been started by exposure to light it cannot be stopped by
removal of the exciting source (37).

The presence of minute amounts of oxygen in the fat,
either in solution or in loose chemical combination with
the unsaturated acids can lead to the rancidity of fat
stimulated by light. Light accelerates the oxidation of
fat; irradiation in the complete absence of oxygen is in-
capable of producing rancidity. Ultra-violet light and
visible light both greatly accelerate the oxidation of fats
(37).

Gortner (25) reports that Holm noted that the greatest effect of light occurs when the light has a wave length of approximately 3600 Å.

The fatty tissue and muscle of pork has been shown to contain an enzymic system, lipoxidase, which accelerates oxidation and the development of rancidity in fat. The enzyme is highly active between pH values of four and five (38). Pork tissue apparently shows considerable variation in concentration of the enzyme due possibly to rations and to fattening characteristics of the pigs (21). Fats, as they occur naturally in tissues, are subject to the action of the enzyme lipase. Lipase continues to attack the glyserides at temperatures far below the solidifying point (3, 21). Lea (36) showed that frozen lamb stored for seven months showed free acidity values of 0.5 per cent at approximately -4 and 14° F as compared to nearly 1.0 per cent at 23° F. It seems reasonably certain that lipase activity continues in meats at these very low temperatures at greatly reduced rates. While hydrolysis of the fat in itself does not indicate rancidity, the presence of appreciable amounts of free acids favors the development of rancidity (21).

Indications have been obtained that biological oxidizing systems elaborated by invading micro-organisms can sometimes produce rancid or tallowy odors and flavors (4). Fortunately, bacterial yeast and mold growth which possess

marked lipase and oxidase activity are not especially important factors in low temperature storage provided reasonable precautions have been exercised in the preparation of meat for freezing and storage (21). Lea (39) states that comparatively few micro-organisms grow to an appreciable extent below 19° F, and 14° F is considered as approximately the lower limit for growth.

Ellis and Howe (21) report that Lea considers lipoxidase enzyme, the antioxidants in the tissues, and the degree of unsaturation of the fatty acids forming the glyserides as being the three factors which influence the oxidation and spoilage of the fat in pork. They add that while comparing pigs at different levels of feeding, Lea found that increase in unsaturation as related to decrease in rate of growth and fattening caused a decrease in susceptibility to oxidation. Ellis and Howe (21) report that Lea suggests, in explaining this phenomenon, that the feeding influenced the relative quantities of antioxidants and of body fats deposited. The low plane of feeding retarded the quantity of fat deposited more than that of antioxidants, while on the high plane fat formation from carbohydrates was accelerated to give a wider ratio of fat to antioxidant. At the same time the high plane of feeding produced a somewhat more saturated body fat. The decrease in antioxidant concentration more than offsets the decrease in unsaturation.

They add that Lea suggests feeding meat animals with feeds high in antioxidant properties to increase resistance of the fat tissue to oxidation (21).

<u>Changes in Pork Fat during Cold Storage:</u> Meat commonly stored for long periods of time in the frozen state becomes subject to rancidity development (5). The fats slowly hydrolyze to form free fatty acids; simultaneously they may oxidize and become rancid. The fat of pork is very sus-ceptible to these chemical reactions (71).

When properly packaged pork is stored at 10 to 15° F, the fat begins to show signs of rancidity in about two months. Unpackaged meat and that poorly protected against desiccation may become rancid in shorter periods of time. In desiccation, the fat immediately underneath the desic-cated areas becomes oxidized and more or less rancid (71).

Cook and White (15) state that the storage tempera-ture is the primary factor that has a significant effect on free fatty acid formation and development of rancidity. They agree with Tressler and DuBois (70), and Hankins and Hiner (29) that storage temperatures of 0° F or lower are essential if spoilage of pork fat is to be avoided over storage periods of approximately one year duration. Novi-kova (51) found no change in frozen pork fat stored for over one year at 0° F. At a storage temperature of 18° F the

fat underwent chemical as well as organoleptic changes.
The surface layer became yellow (even in six months) and
acquired a stearic taste; in one year Novikova observed
that the phenomenon had penetrated to approximately 0.25 to
0.40 cm. Novikova (51), and Kiermeier and Heiss (34) found
that frozen pork meat should not be stored for more than
three to four months at 18° F. The latter investigators
state that fat tissues to be stored for long periods should
not be subjected to air currents as the fat becomes rancid
more rapidly. They add that this may be attributed to the
oxygen of the air. Noble and Hardy (50) concluded that
frozen pork loin roasts even though obtained from high grade
animals and carefully handled cannot be stored at 0 to 15° F
for longer than 16 to 22 weeks without danger of having the
flavor and aroma of the fat decrease in desirability.

Gyorgy and associates (26) found that sulfhydryl
compounds, by virtue of their free sulfhydryl radical retard
the development of rancidity in fat but only in the presence
of water and in the absence of copper salts or other in-
hibitors of sulfhydryl radical. They report that thiouracil
and thiourea have this property.

7. Thiouracil

The recent discovery of a series of drugs that will

inhibit the formation of thyroid hormone by the thyroid gland made it possible, by administering such compounds to determine the influence of subnormal levels of thyroidal activity on growth, fattening, and carcass quality in various classes of livestock.

Hypothyroidism, a condition associated with a decrease in the metabolic rate and with obesity would conceivably be desirable in animals that were being fitted for market. Since a hypothyroid condition is usually associated with a tendency to fatten, several investigators have determined the influence of inducing such a condition in meat producing animals. Andrews and Bullard (1) reported rapid fattening and gain in weight following partial thyroidectomy of steers. In this case the rapid gains may have been due partly to compensation for the loss of weight immediately following the operation as well as to the decreased metabolic rate (48).

The goitrogenic effect of a large number of substances related to thiourea has been demonstrated by Astwood (2), Dietrich and Beutner (16), and others. The most active compounds found to inhibit thyroid gland function belong to a group of substances possessing a thiourea grouping. All derivatives of thiourea possess in common the thioureylene radical -NH.CS.NH- (2). Of the numerous compounds showing antithyroid properties, thiouracil seems

to be one of the most suitable to induce hypothyroidism because of its high potency and low toxicity (2).

Properties:

Thiouracil is a white, odorless, bitter powder. (It would appear that the ability to taste thiouracil is in-herited in the same manner as that of phynylthiocarbamide. The ability to taste the latter compound is dominant. This test has been used as a genetic marker in the study of human pedigrees (62).) Thiouracil is insoluble in methyl alco-hol, ethyl alcohol, carbon tetrachloride, acetone, or mineral acids (79).

Thiouracil (NHCSHNCOCHCH) has a type of linkage in which a divalent sulfur is linked to a single non-metallic element (C = S) (79). The entire group -NH.CS.NH- is essential to the thyroid effect. Activity is lost if the S is replaced by another element or group as in uracil (NHCONHCOCHCH). Apparently both imino nitrogen groups are essential, for activity is lost when one of these groups is absent as, for example, in thioacetamide. The sulfur itself is not the active agent obviously as a number of compounds containing sulfur in different forms were inactive. There-fore, it appears that the entire thiourea grouping is essential for response (2).

Due to its acid properties, thiouracil forms very

soluble salts on addition of NaOH or KOH to an aqueous sus-
pension. Dry thiouracil is stable for months at room
temperature. It has been found to remain stable for several
days when kept in an ice-box at 5° C whether in the form of
an aqueous solution, tissue extract or blood (79).

A Method for the Preparation of Thiouracil: (77)

$$
\begin{array}{ccccc}
\text{HNH} & & \text{C}_2\text{H}_5\text{OCO} & & \text{HN}-\text{CO} \\
| & & | & & | \quad\; | \\
\text{S}=\text{C} & + & \text{CH} \longrightarrow & \text{S}=\text{C} \quad \text{CH} & + \text{ NaOH } + \text{ C}_2\text{H}_5\text{OH} \\
| & & | & & | \quad\; \| \\
\text{HNH} & & \text{NaOCH} & & \text{HN}-\text{CH}
\end{array}
$$

| Thiourea | Sodium Salt of Formyl Acetic-ester | Thiouracil |
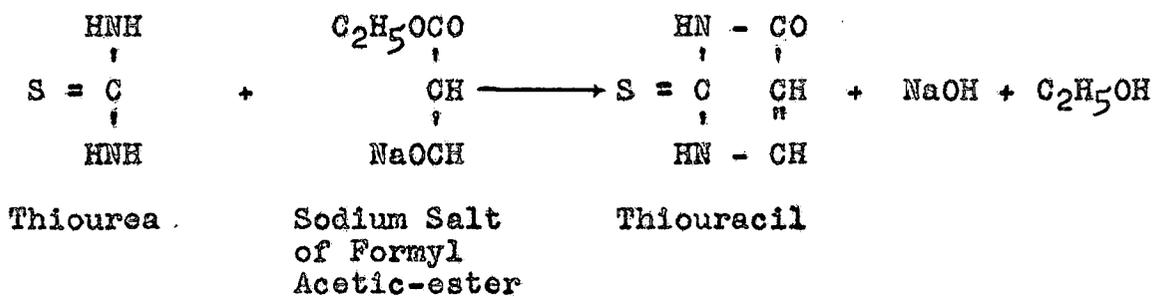
Effect of Thiouracil Fed Orally to Swine:

The most noticeable effect of feeding thiouracil to
swine was the reduction in both the total feed consumption
and the feed required per pound of gain (42). McMillan and
co-workers (42) observed that thiouracil-fed pigs begin to
show a tendency towards laziness by the end of the second
week of treatment. The pigs became sluggish and slow to
get up. Some became fatigued easily when exercised. They
lost some of their bloom and their hair coat became rough.
McMillan and associates (42) believe that they found some
indication that thiouracil treatment may cause a slight

retardation of skeletal growth. They conclude that there was no significant differences in the carcasses attributable to the thiouracil treatment. Van Der Noot and associates (74) obtained economical and rapid gains when they fed 0.25 per cent thiouracil in the feed of pigs. Muhrer and Hogan (48) comparing thiouracil-treated pigs with non-treated pigs found that the former were wider, shorter, and not as tall as the latter. Thiouracil-treated pigs were apparently an example of retarded growth with rapid gains in weight due to deposition of an excessive amount of fat. Thiouracil-treated pigs sleep a greater portion of time and are not active. Muhrer and Hogan (48) believe it is possible that if thiouracil-treated animals were slaughtered too soon, their flesh would be unsuitable for human consumption. Van Der Noot and associates (75) report that on the basis of preliminary tests it is not likely that thiouracil is present in thiouracil-fed pigs in sufficient amounts to influence the consumer. But they agree with Muhrer and Hogan (48) in that investigations concerning the length of time thiouracil is retained in the tissues should be carried out.

Thiouracil is an effective drug for the treatment of thyrotoxicosis but it is an unpredictably toxic drug which may produce serious and uncontrollable effects, especially on bone marrow (24).

Some very recent work by Pipes and Turner (55) showed that no hazard exists in the use of meat from thiouracil-fed animals for human consumption. They add that tissues from animals receiving even 10 to 20 times the amount of thiouracil necessary for optimum fattening contain less than 25 milligrams of thiouracil per pound. At this concentration it would be practically impossible for a human to consume sufficient meat to obtain even the lower medicinal dosages of thiouracil. The work of Pipes and Turner (55) indicated rapid absorption, destruction and excretion of thiouracil by the living tissue. They add that Franklin and associates report that cooking meat for one hour at 130° C had no effect on the thiouracil level, but that most of the compound disappeared from the tissue after storage for one week at 4° C.

8. Protamone

Economical meat production depends upon the rapid growth and fattening of meat animals. Since the thyroidal hormone is intimately concerned in these processes, investigators have attempted by various techniques to alter the amount of circulating thyroidal hormone in a number of species. There is some evidence that a mild hyperthyroid condition, or an active thyroid state (43), is associated with and may be conducive to rapid growth.

The thyrotrophic content of the anterior lobe of the

pituitary gland is highest during the period of rapid growth
in swine. In addition, the pituitaries of slow-growing
strains of swine have been reported to be lower in thyro-
trophic potency than those of more rapidly growing strains
(20).

Within the last few years it has been shown that
thyro-active iodinated proteins in appropriate doses have a
metabolism-stimulating effect on cows (43). Koger, Reineke
and Turner (35) treated immature female mice, both orally
and subcutaneously, and were able to increase, substan-
tially, their rate of growth. The metabolism-stimulating
effect resulted in an increased food consumption of the
animals and a corresponding acceleration in the growth rate.

Different investigators have tried, by using dif-
ferent levels of thyroproteins, to increase the growth rate
of swine; but their results are not consistent. Using pro-
tamone, a synthetic thyroprotein, Reineke and McMillan (60)
found that at low levels (0.005 per cent of the ration) no
noticeable signs of hyperthyroidism were observable during
a twelve-day treatment. At higher levels (0.01 per cent
of the ration) some loss in body weight accompanied by
nervousness occurred. These workers conclude that treated
Berkshire pigs showed slight gains over controls, appeared
more mature, were thicker and meatier, and considerably more
uniform. On the other hand, Van Der Noot and associates

(75) were not able to increase the gains in weight of pigs
even after using different levels of thyroprotein. They
found that while the lower levels had no influence on either
gains in weight or amount of feed required to produce an in-
crease of a hundred pounds in weight, the higher levels de-
creased the gains in body weight and increased the feed con-
sumption per hundred pounds gain in weight. Their conclu-
sion is that in the growing pig nothing is to be gained by
increasing its basal metabolic rate. Braude (6) confirmed
the observations of Van Der Noot and associates. He also
reports that while low levels of thyroprotein were not
sufficiently effected (0.5 g daily per pig) to have any
practical importance, higher doses (1.0 to 1.5 g daily per
pig) did affect the rate of growth. Pigs receiving the high
doses showed signs of retarded growth, heavy breathing,
appeared to be excited and took a long time to finish feed.
Braude's (6) opinion is that high doses of iodinated pro-
tein stimulate the metabolic activity to such an extent as
to cause a rapid loss of condition and weight of the
animals receiving the dose. He believes that, so far as
practical applications are concerned, it seems justifiable
to dismiss the possibility of using iodinated protein for
stimulating growth until further evidence is obtained. He
suggests the possibility that a difference exists between
species in the response to thyroid-activating substances

fed orally.

It is advisable at this point to differentiate between thyroactive proteins and iodinated proteins. A thyroactive iodocasein, for example, belongs to the first group and it is a term used to designate a product formed by iodination of proteins in such a manner as to produce a substance with high thyroidal activity. On the other hand, iodocasein or iodo-albumin are materials which are used merely as carriers of organic iodine, and have little or no thyroidal activity. Protamone, a synthetic thyroprotein, containing approximately 3.16 per cent thyroxine, is a thyroactive iodoprotein.

CHAPTER III

Experimental Procedure

Three groups of hogs, each consisting of five animals were fed a ration containing 77 per cent ground barley, three per cent tankage (60% protein), two per cent oil meal, 10 per cent alfalfa meal, seven per cent soybean meal, and one per cent mineral mix. Feeding was conducted for a period of 47 days starting on May 5, 1948 and ending on June 21 of the same year. The hogs in all three groups were fed all they would consume. They were slaughtered on June 22, 1948 and held in cold storage until they were cut.

Group A: consisted of two purebred Duroc Jersey gilts and three purebred Berkshires (two gilts and one barrow). This group was fed 0.1 per cent thiouracil mixed with their feed.

Group B: consisted of the same number of gilts and barrows from each breed as in group A. This group was fed 0.04 per cent protamone mixed with their feed.

Group C: consisted of two purebred Duroc Jersey gilts, two purebred Berkshires (one gilt and one barrow), and one purebred Hamprace gilt.

The carcasses were cut after one day storage in the cooler and the right loin and ham from six gilts, representing the two breeds, Duroc Jersey and Berkshire, and

the three different treatments, thiouracil, protamone, and control, were chosen. The loins were cut into one-inch thick chops and then every two chops were double-wrapped in an inner wrap of aluminum foil and an outer wrap of ordinary waxed wrapping paper. The drugstore wrap was used with the chops. After removing the outer layer of fat, the ham from each animal was ground, well mixed and then filled into 12-ounce glass jars which were consequently vacuum sealed with very little air space.

The chops and the ground ham were frozen at -20° F for 16 hours and then were stored in a 0° F room.

CHAPTER IV

Analytical Procedure

Color Determination

The Photovolt reflectometer, Plate I, was used for determining the color of pork chops. For every color determination the same location, namely the center of the chop belonging to the longissimus dorsi, was chosen. Separate readings were taken with the three filters, blue, green, and amber. While readings were taken, the photo cell was pressed slightly against the lean part of the chop being tested. Readings were then converted to the Munsell Notation.

pH Value Determination

An industrial model M pH meter, Plate II, was used for determining the pH value of the pork chops. The samples were first thawed at room temperature for five hours and then both electrodes were applied, with a moderate amount of pressure, to the lean portion of both sides of each chop at random. Thawing at room temperature for five hours produced chops with a texture soft enough to insert the electrodes, and short enough so that no changes took place.

Plate I
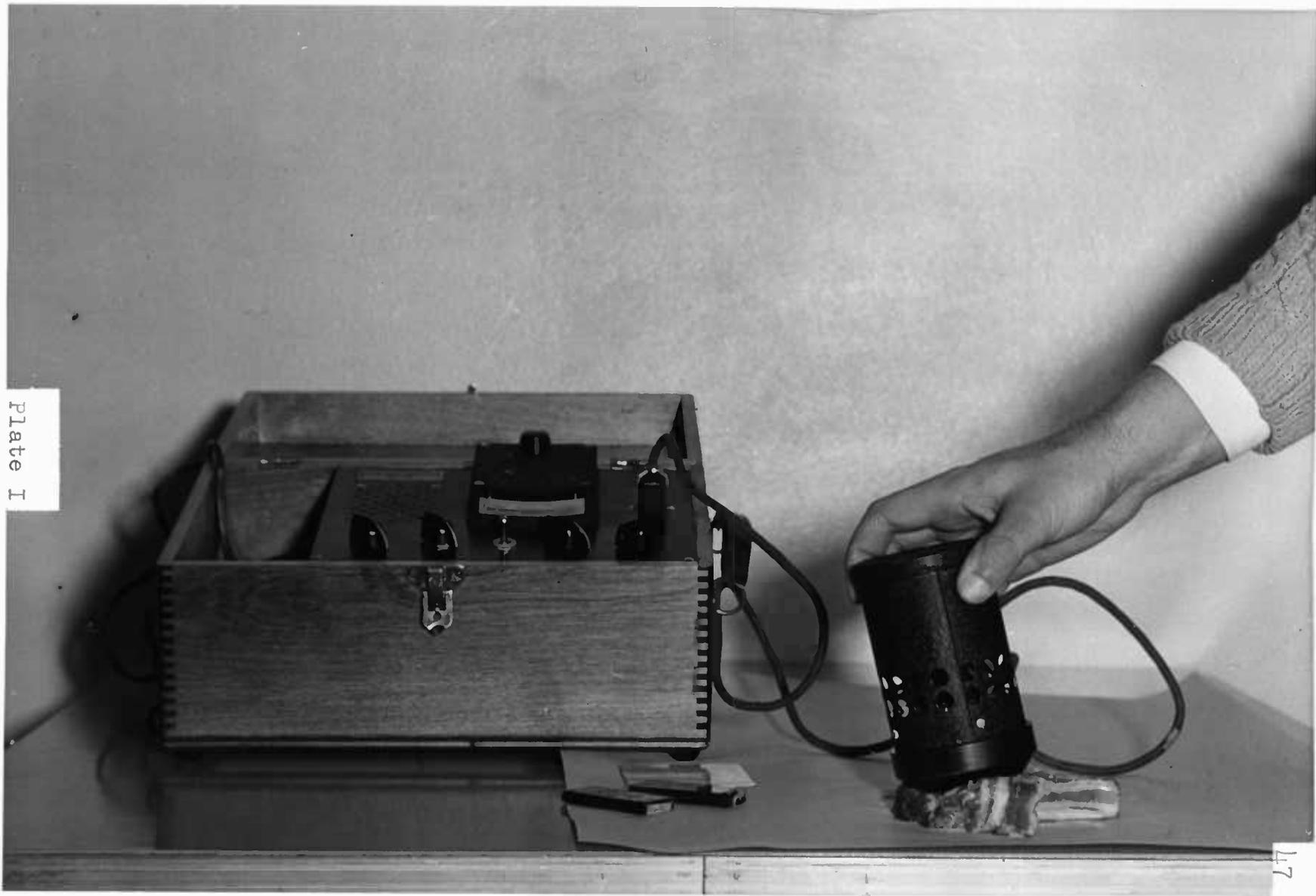
Test for Tenderness

The shearing machine, Plate III, was used for test-
ing the tenderness. It indicates the force in pounds re-
quired to shear a core of meat one inch in diameter and
about one inch long. The cores were cut with a special
coring instrument from the lean part (longissimus dorsi) of
each chop, two cores being taken from each chop. Tenderness
is read as the amount of resistance to shear, in pounds.

Drip Determination

Cylindrical metal cans with covers having a hook in
the center of each, Plate IV, from which the chops were
hung, were used to determine the amount of drip in the
thawed samples. The chops were thawed at room temperature
for 24 hours and the liquid exuding as drip was collected
at the bottom of the cans. The chops were weighed before
and after thawing, the difference being equal to the amount
of drip. The drip was then calculated as a per cent of the
frozen weight of the chops.

Fat Content Determination

The method used was that of Oesting (52) which is a
fast method for fat determination in meat. A sample of
ground meat was run in a mixer with cracked ice and an

Plate II

emulsifying agent, trisodium phosphate. A 10-gram sample of the emulsion was weighed in a Babcock bottle to which glacial acetic acid was added to coagulate the proteins and then followed by enough concentrated sulphuric acid to digest the proteins. Warm water was added to the surface to reduce fat hydrolysis. The bottles were then centrifuged, first for five minutes, water added to the neck of the bottles, and again centrifuged to raise all the fat to the top of the bottle necks. The bottles were placed in warm water, 70° C, for two minutes and the fat content was read on the descending column. Correction was made if the weight of the sample was not exactly 10 grams. The fat content read in the bottle neck was multiplied by 9.2 to give the per cent fat in the meat sample.

Oesting and Kaufman (52) compared their method with that of the A.O.A.C. method and found that their method showed a standard deviation of ± 0.7 per cent with a probable error of ± 0.5 per cent based on an average level of 30 per cent fat.

Peroxide Value Determination

The method used was a modification of the method of Watts and Peng (76). The fat was extracted from the meat sample using carbon tetrachloride while the moisture was removed by adding anhydrous sodium sulphate to the mix in
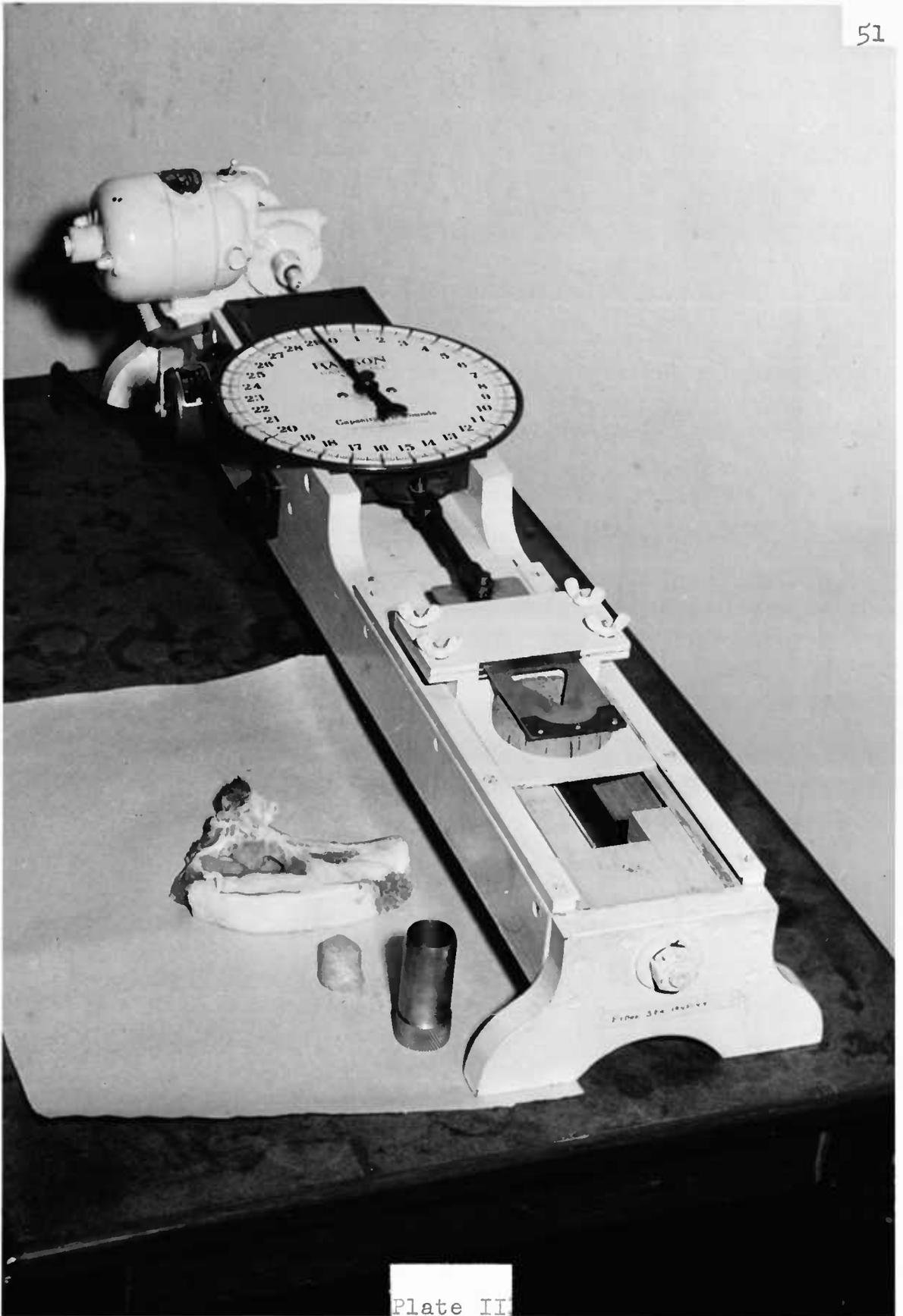
Plate III

the Waring blender. After filtering through a No. 4 Whatman
paper into a graduated cylinder, the filtrate was well
shaken and aliquots were taken for rancidity determination.
Aliquots were placed in dark test tubes to reduce the effect
of light on fat. To each tube one gram of KI and 20 cc of a
mixture of chloroform and glacial acetic acid (1:2) were
added. The tube with its contents was placed in boiling
water for one minute. The contents of each tube were then
transferred into a flask containing 30 cc of water and a
few drops of a freshly prepared starch solution (1.0%) were
added. When a dark blue color appeared (iodine) the solu-
tion was titrated to a colorless end-point with 0.01 N
thiosulphate solution. The following equation was used to
determine and express rancidity:

$$\frac{T \times N \times 500}{\text{weight of fat}} = \text{millimoles of peroxide per Kg fat}$$

where (T) represents the cc of thiosulphate used in the
titration, and (N), its normality.

Plate IV

CHAPTER V

Results

1. Color of Lean

Duroc Jersey

Hue: There was an obvious difference in the hue between the fresh samples representing the three treatments, Table I. During the six months period of storage changes in hue were observed, Figure 1, but whether these were the result of the drug treatment or not is not known. The thiouracil samples changed to a darker red while the protamone as well as the untreated samples changed toward a lighter red. It was noticed that the difference in hue between the three treatments remained almost consistent during the storage period.

Chroma or the degree of color strength changed very little in the three treatments during the whole period of storage. There seems to be some relation between the chroma and hue of a meat sample; when the color of the sample (hue) became deeper red there was a corresponding increase in the chroma reading in most cases.

Value: There was no significant indication that drug-treatment had any effect on the value or brilliance of the color of samples. During the storage period there was

slight apparent change in the value of the three different samples, Figure 3.

Berkshires

Hue: There was a significant difference in hue between the fresh samples representing the three treatments. During storage the hue of the three treatments tended to change from yellow-red to red or from red to a deeper-red, Figure 2.

Chroma: Observations showed that there was little difference in the chroma of colors in the three samples. As in the case of Durocs there were some indications that a relationship exists between the hue and chroma of a color. This fact has to be proved first before coming to any conclusion.

Value: There was some slight difference in the brilliance of the color of the three samples. This difference remained about the same during the whole period with very slight change.

TABLE I

The Color of Meat Based on Munsell Notation

Storage Period in Weeks	Control	Protamone	Thiouracil
	Duroc-Jerseys		
Fresh	10 RP 5.4/4.3	2.5 R 5.5/5.2	0.4 YR 5.7/3.1
4	6.1 R 5.7/4.0	3.8 R 5.6/4.6	8.5 R 5.7/3.6
7	0.3 R 5.5/7.7	3.4 R 5.7/6.6	9.6 R 5.9/3.5
9	9.9 R 5.8/3.9	3.2 YR 5.7/3.4	10 R 5.6/4.1
12	0.2 YR 5.7/3.5	1.2 YR 5.8/4.5	1.7 YR 5.7/3.4
15	7.4 R 5.8/4.1	9.0 R 5.9/4.9	1.9 YR 5.6/3.2
20	9.9 RP 5.4/4.9	5.4 R 5.7/4.9	6.6 R 5.5/3.5
24	0.8 YR 5.8/4.6	5.4 R 5.7/4.9	8.1 R 5.1/4.3
	Berkshires		
Fresh	1.3 YR 6.1/2.8	4.1 R 5.4/5.4	9.6 R 5.7/2.9
4	0.2 YR 5.8/3.9	5.8 R 5.3/5.4	5.9 R 5.5/5.0
7	3.7 R 5.8/5.9	2.9 R 5.2/6.2	7.8 R 5.6/4.3
9	3.0 YR 6.1/3.1	2.1 YR 5.6/3.7	1.2 YR 5.8/3.5
12	7.8 R 5.5/4.8	4.2 R 5.5/5.8	2.3 R 5.1/6.1
15	9.9 R 5.9/3.7	7.6 R 5.5/4.5	5.9 R 5.7/3.9
20	4.0 R 5.8/3.8	1.6 R 5.3/5.8	1.6 R 5.6/5.0
24	----------------	7.8 R 5.2/5.2	5.5 R 5.4/6.0

DUROC-JERSEY
CHANGE IN HUE OF THE COLOR OF CHOPS.

Weeks of Storage
at 0°F.

CHANGE IN CHROMA OF THE COLOR OF CHOPS.

CONTROL
PROTAMONE
THIOURACIL

Weeks Of Storage

Figure 1.

BERKSHIRE.
CHANGE IN HUE OF THE COLOR OF CHOPS

CHANGE IN CHROMA OF THE COLOR OF CHOPS

CONTROL
PROTAMONE
THIOURACIL

Figure 2

BERKSHIRE.
CHANGE IN VALUE OF THE COLOR OF CHOPS

Weeks Of Storage At 0°F.

DUROC-JERSEY
CHANGE IN VALUE OF THE COLOR OF CHOPS

CONTROL
PROTAMONE
THIOURACIL

Weeks Of Storage At 0°F.

Figure 3

2. pH Value

Within each breed the pH value of the fresh samples representing the three different treatments was approximately the same. There was a consistent tendency among all samples for the pH value to drop gradually during the first twelve weeks of storage. During the subsequent twelve weeks the pH value of all samples in both breeds tended to increase, Table II, Figures 4 and 5. During the six months storage period it was observed that the difference between the three treatm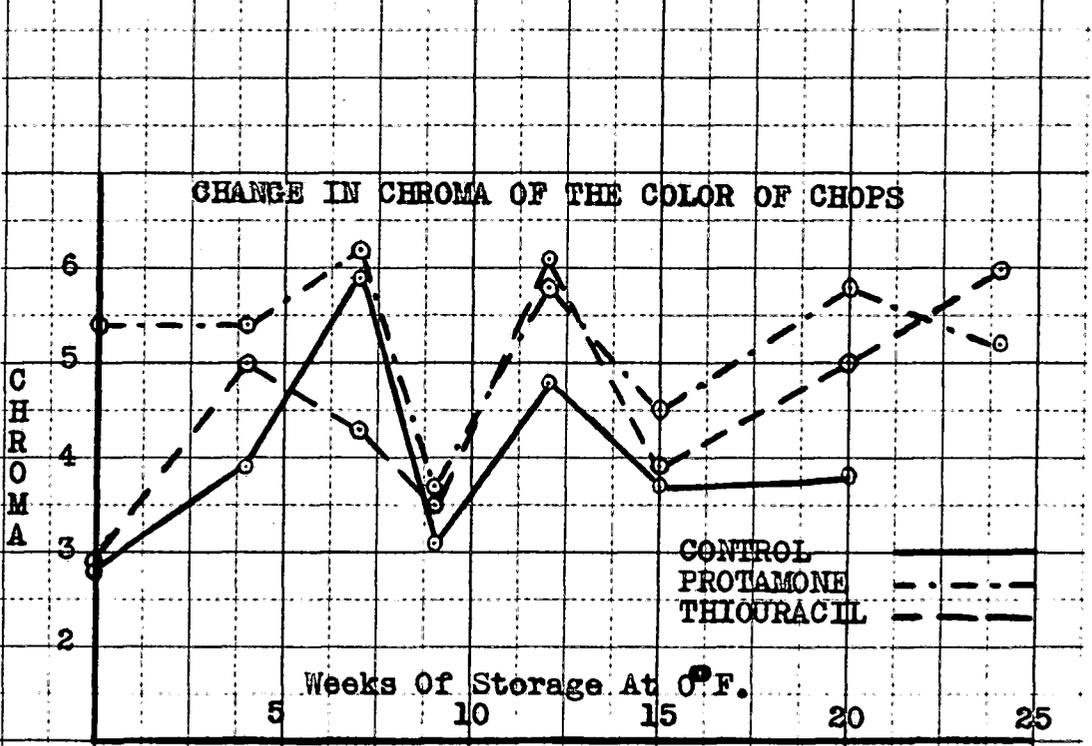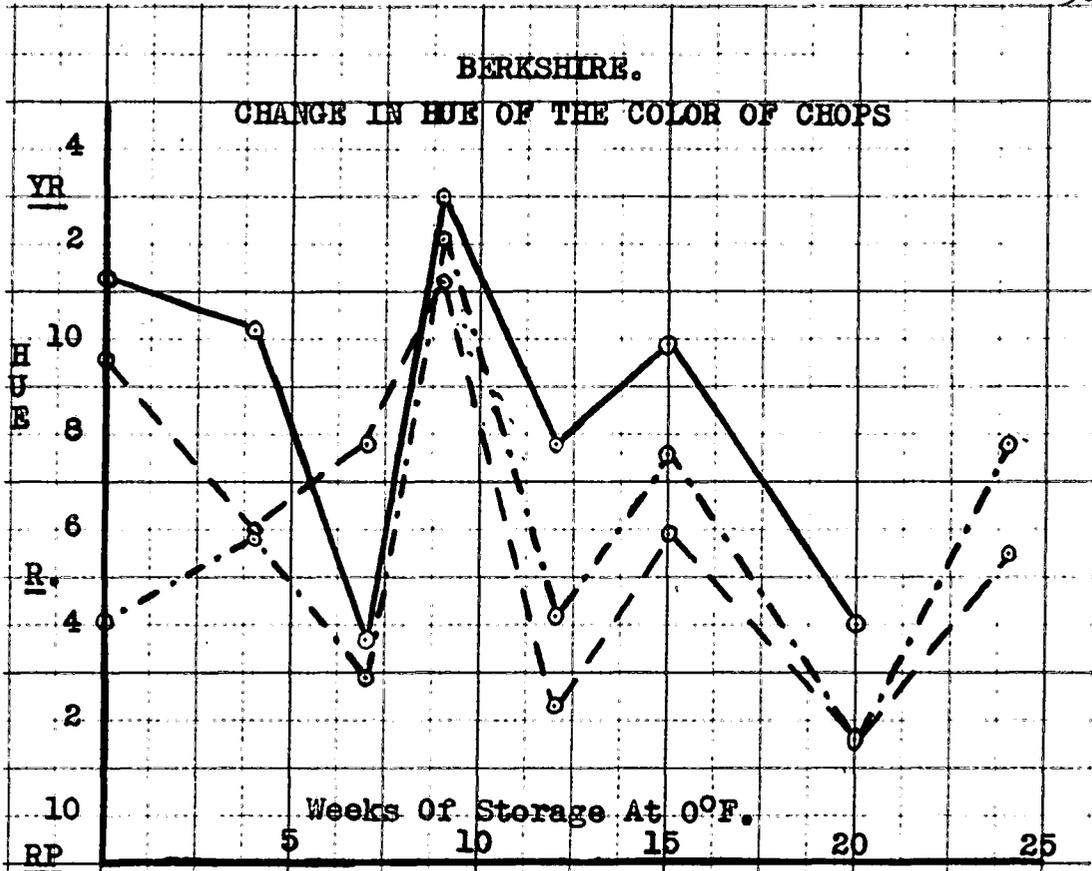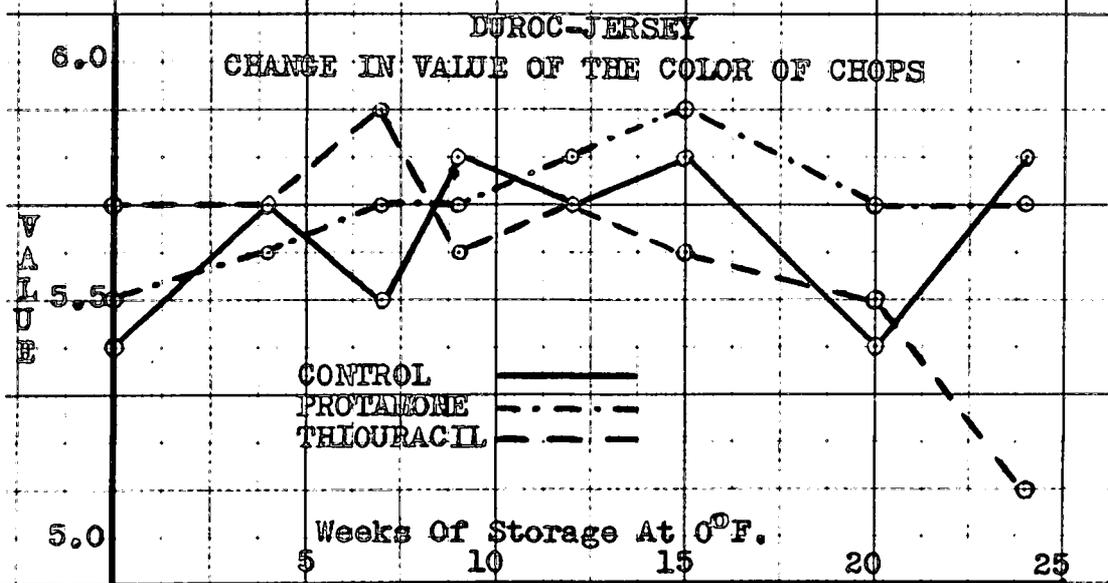ents in pH value was rather small but remained rather consistent throughout the experiment. The protamone samples in both breeds had the highest pH value while the control samples had pH values similar if not lower than the thiouracil samples. In most cases the thiouracil samples had intermediate pH values between the protamone and control samples. Only in one case, at the end of the storage period, the thiouracil sample from the Duroc Jersey breed had the lowest pH value. Due to the lack of a control sample from the Berkshire breed it was not possible to ascertain this observation.

TABLE II

The pH Value of Chops

Storage Period in Weeks	pH Values		
	Control	Protamone	Thiouracil
Duroc-Jerseys			
Fresh	5.5	5.6	5.5
4	5.2	5.5	5.1
7	5.2	5.5	5.2
9	4.9	5.1	5.0
12	4.8	4.9	4.8
15	5.4	5.5	5.4
20	5.4	5.6	5.4
24	5.5	5.6	5.5
Berkshires			
Fresh	5.4	5.4	5.5
4	5.2	5.3	5.3
7	5.1	5.3	5.2
9	4.9	5.2	5.0
12	4.8	5.0	4.9
15	5.4	5.6	5.4
20	5.4	5.6	5.5
24	---	5.7	5.6

THE CHANGE IN PH VALUE OF CHOPS FROM DUROC-
JERSEY PIGS AFTER STORAGE AT 0° F. FOR 24 WEEKS

CONTROL
PROTAMONE
THIOURACIL

Weeks Of Storage
At 0°F
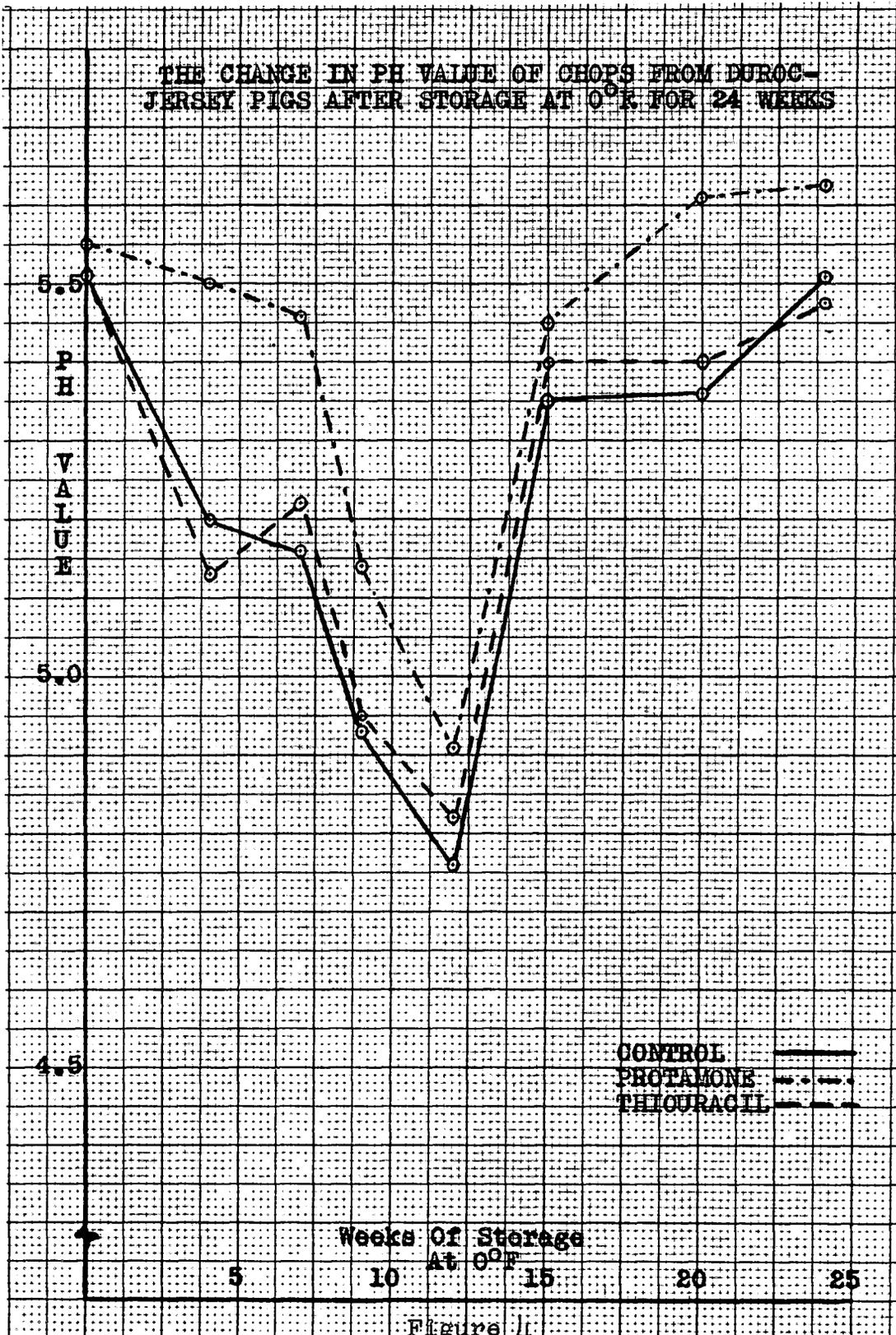
Figure 4

THE CHANGES IN PH VALUE OF CHOPS FROM BERK-
SHIRE PIGS AFTER STORAGE AT 0°F. FOR 24 WEEKS

Figure 5
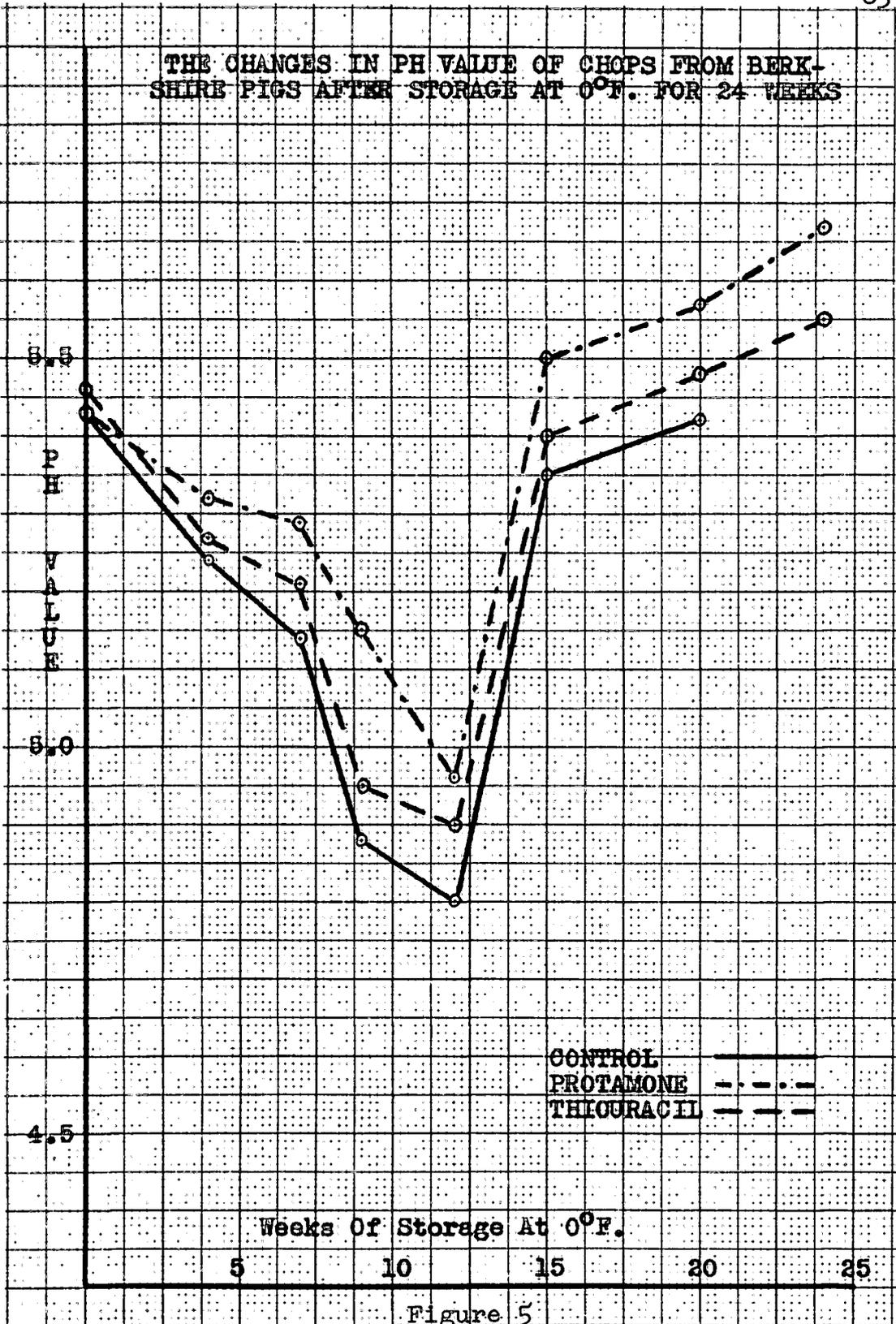
3. Tenderness

Results indicated in Tables III and IV represent
the average shearing values taken from each treatment within
each breed, and for each treatment when results of both
breeds were added. Using chi-square, a highly significant
difference in tenderness was observed between samples of
meat from untreated animals and those from treated animals,
Table V. Both treatments tended to make the muscle tissue
more tender than untreated muscle tissue, Figure 6. But
chi-square value indicates that the difference in tender-
ness between the thiouracil and protamone treated groups
is not significant, Table VI.

TABLE III

Average Shearing Values of Lean Meat from Each Breed

	Control	Protamone	Thiouracil
Duroc-Jerseys			
No. samples	33	26	29
Total shear	302.4	186.8	169.9
Avg shear value	9.16	7.18	5.85
Berkshires			
No. samples	28	24	30
Total shear	235.5	145.4	203.9
Avg shear value	8.41	6.10	6.79

TABLE IV

Average Shearing Values of Lean Meat of Both Breeds Combined

	Control	Protamone	Thiouracil
No. samples	61	50	59
Total shear	537.9	332.2	373.8
Avg shear value	8.82	6.64	6.34

TABLE V

Statistical Analysis for Tenderness
Among the Three Treatments
(Using Chi-square)

	Control	Protamone	Thiouracil	Totals
Observed	537.9	332.2	373.8	1243.9
Expected	446.3	365.8	431.8	1243.9
Deviation	91.6	- 33.6	- 58.0	0.0
$\dfrac{d^2}{exp.}$ =	18.8	3.1	7.8	29.7

TABLE VI

Statistical Analysis of Tenderness for Samples from
Thiouracil and Protamone Treated Groups

	Protamone	Thiouracil	Totals
Observed	332.2	373.8	706.0
Expected	323.8	382.2	706.0
Deviation	8.4	- 8.4	0.0
$\dfrac{d^2}{exp.}$ =	0.21	0.18	0.39

TENDERNESS OF CHOPS.

AVERAGE SHEARING VALUES FOR THE THREE TREATMENTS.

CONTROL

PROTAMONE

THIOURACIL

LBS. RESISTANCE TO SHEAR

10

8

6

4

2

0
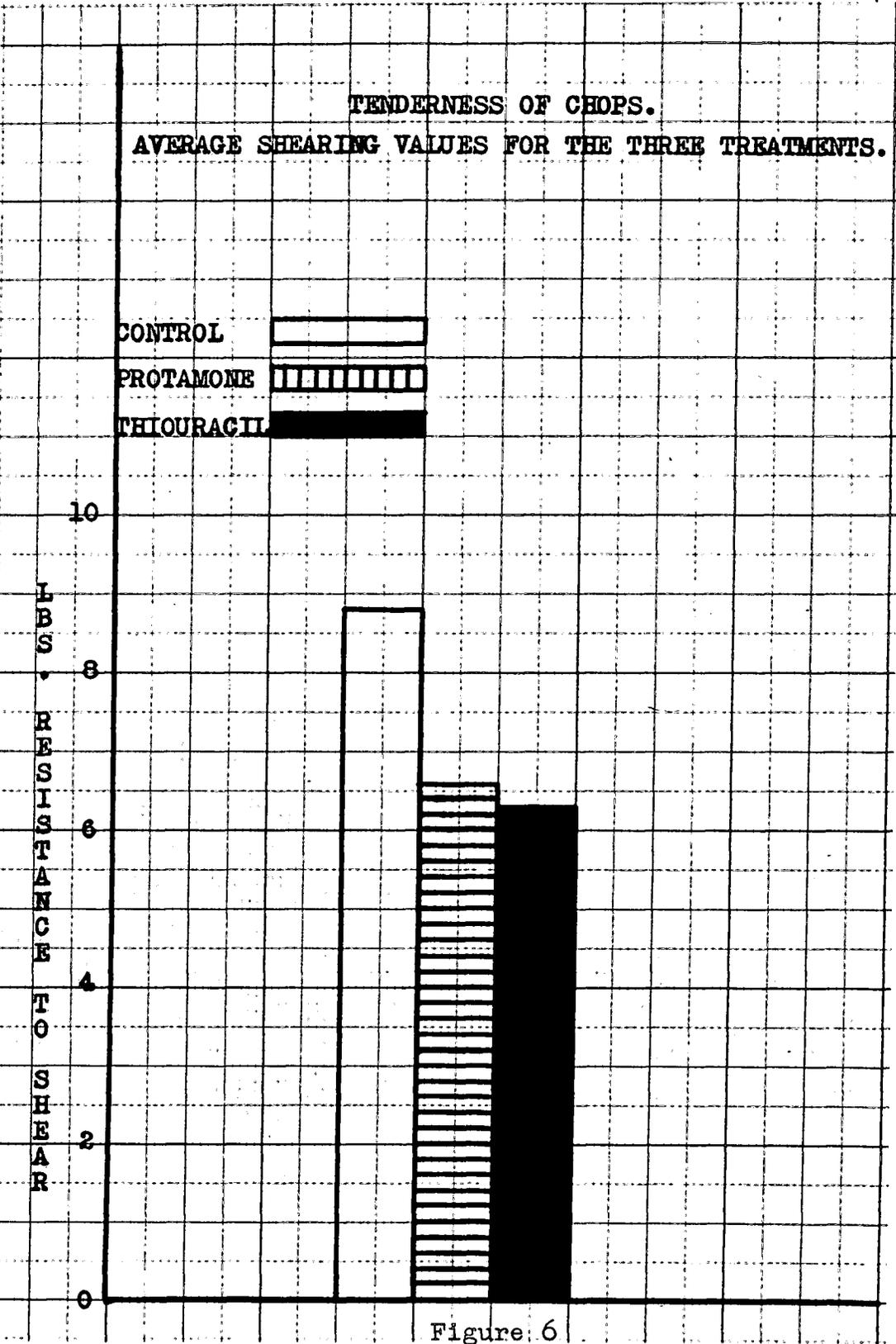
Figure 6

4. Drip

Results indicate that for both breeds the thiouracil samples had the highest percentage of drip upon thawing, the protamone samples had the lowest percentage and the control samples were intermediate, Table VII. The differences were great between the three treatments in both breeds, Figures 7 and 8. Observations made at the time of drip determination indicate that the protamone samples preserved their shape while the thiouracil samples were limp and mis-shaped. The control samples were intermediate, rather on the limp side.

TABLE VII

Percentage Drip from Pork Chops

Storage Period in Weeks	Per Cent Drip		
	Control	Protamone	Thiouracil
	Duroc-Jerseys		
4	1.8	0.2	3.6
7	2.8	1.4	4.4
9	1.6	0.4	5.8
12	3.1	2.3	4.3
15	2.7	1.3	5.8
20	3.4	2.6	4.6
24	2.2	1.2	6.9
	Berkshires		
4	1.2	1.8	2.7
7	3.0	2.0	5.7
9	3.1	1.6	6.2
12	2.5	1.4	6.7
15	5.3	2.6	6.1
20	5.9	2.9	8.6
24	---	1.2	5.4

Figure 7

BERKSHIRES

PERCENT DRIP FROM DRUG-TREATED AND UNTREATED CHOPS

CONTROL ————
PROTAMONE —·—·—·
THIOURACIL —— —— ——
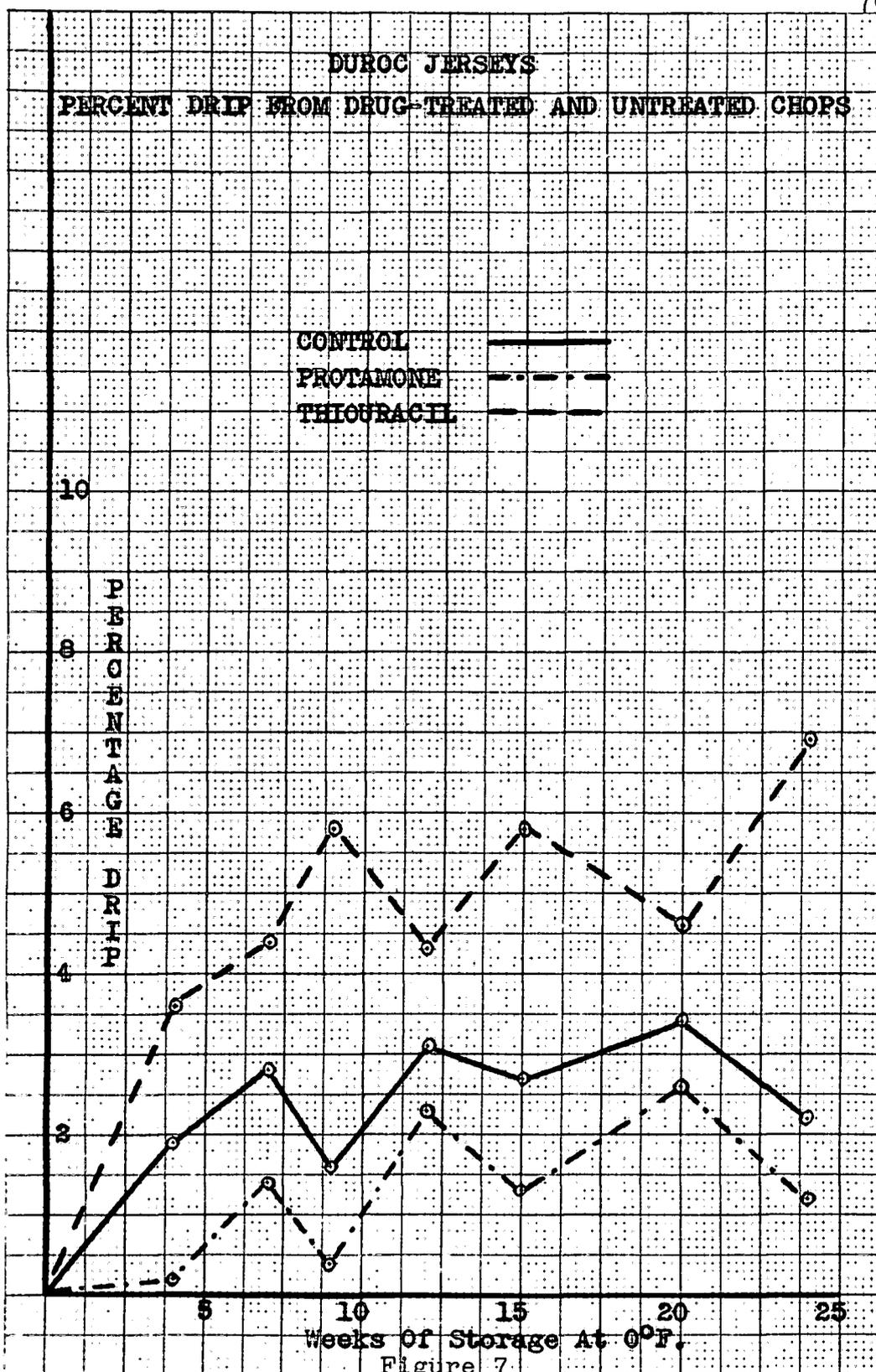
Figure 8

5. Fat Content of Lean

The protamone-fed pigs, in both breeds, had the highest fat content in the lean; the thiouracil-fed pigs had the lowest fat content, Table VIII, Figure 9. Treatment with thiouracil tends to cause very little of the fat formed in the pig to be deposited within the lean; most of the fat is present on the outside of the body and muscles. The untreated pigs showed an amount of fat within the lean intermediate between both treatments.

TABLE VIII

Fat Content of Ground Ham

Treatment	Per Cent Fat (Wet Basis)	Treatment	Per Cent Fat (Wet Basis)
Duroc-Jerseys		Berkshires	
Control	5.52	Control	5.90
Protamone	10.30	Protamone	10.39
Thiouracil	5.20	Thiouracil	4.60

FAT CONTENT OF GROUND HAM,
DUROC-JERSEYS AND BERKSHIRES

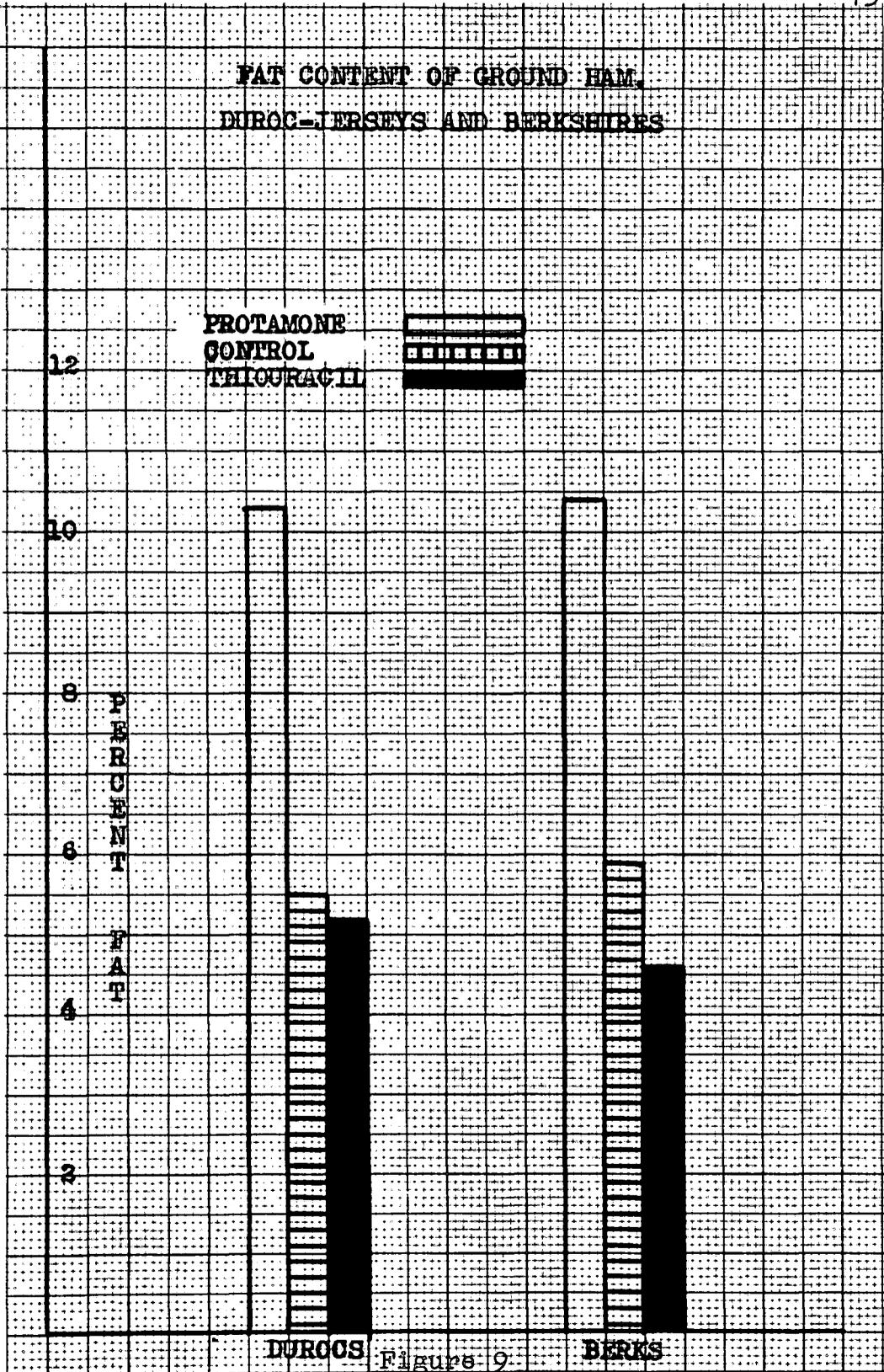Figure 9

6. Rancidity Development in Fat of Ground Ham

When the samples were kept in storage at 0° F for
the period of four months no changes related to fat spoilage
were observed. Following this period the ground samples
were moved to a 15° F cold box.

After five weeks of storage in the 15° F box the fat
of the protamone samples from the Duroc breed showed signs
of oxidation, as indicated by low peroxide numbers. Per-
oxide values increased throughout the remaining storage
period. The fat of the control samples of the Duroc breed
showed some oxidation after ten weeks holding at the higher
storage temperature (15° F). Peroxide values increased
during the following weeks of storage.

The fat of the protamone samples from the Berkshire
breed showed peroxide formation after seven weeks of stor-
age while the control samples of this breed showed peroxide
formation after 13 weeks of storage at the higher tempera-
ture.

There was no sign of oxidative changes in the fat of
any of the thiouracil samples for either breed during the
entire storage period at 0° or at 15° F.

TABLE IX

Rancidity Development in Fat of Ground Ham

Storage Period in Weeks	Millimoles Peroxide/Kg Fat		
	Control	Protamone	Thiouracil
Duroc-Jerseys			
At 0°F			
16	----	----	----
At 15°F			
5	----	1.95	----
6	----	0.89	----
7	----	1.86	----
9	----	2.80	----
10	0.91	5.84	----
12	2.84	7.60	----
13	8.70	10.90	----

TABLE X

Rancidity Development in Fat of Ground Ham

Storage Period in Weeks	Millimoles Peroxide/Kg Fat		
	Control	Protamone	Thiouracil
Berkshires			
At 0°F			
16	----	----	----
At 15°F			
7	----	0.70	----
9	----	0.56	----
10	----	0.87	----
12	----	1.80	----
13	0.89	3.24	----

CHAPTER VI

Discussion

Before going into any discussion based on results obtained, it should be made clear that data obtained represent a small number of samples taken from a very small number of animals.

Further investigation into this work is needed before any conclusive decisions could be made concerning the effects or advantages and disadvantages of such drugs. As a matter of fact all discussion will be based on comparison between the three treatments within one breed and whether observations made in one breed correspond with the second breed.

In an effort to show the effect, if any, of both drugs on the quality factors, in a significant way, all the samples were handled and treated the same from the time the samples were cut from the carcass until they were tested. Results indicate that the effect of one or both drugs has been rather significant even though uncontrollable outside factors came into play.

Color of Lean

As far as measuring the three color attributes it appears that both drugs had no effect. It is difficult to

state such a conclusion since several factors are concerned
with the color of a piece of meat and it is very possible
that either one or both of the drugs do affect the color
but the effect is so obscured or covered up by the effect of
other factors that nothing could be measured to indicate any
effect of the respective drugs. It is also possible that
the amount of each drug used for feeding, as well as the
feeding period, was not sufficient to result in a change in
any of the three color attributes.

Drip

Data indicate that drip was affected by the use of
both drugs. In both breeds, the thiouracil samples had the
highest percentage of drip, the protamone samples had the
lowest, while the untreated samples had intermediate values.
The differences were significant all during the testing
period. All samples showed a rise in percentage of drip
as the storage period increased indicating the possible
effect of such factors as storage temperature, length of
storage period, temperature and length of thawing. Whether
the effect of the drugs upon drip is chemical, nonchemical,
or a result of fat diposition within or outside muscle
tissue is not known. But there is definitely something
that affects the amount of moisture content loosened during

thawing. It is possible, in the case of thiouracil samples, that this drug may affect the bound-water free-water equilibrium.

pH Value

Fresh samples, within each breed, had approximately similar pH values. During storage, pH values followed a certain pattern, within each breed, which corresponded between the two breeds. The differences between the three treatments within each breed, were small but rather consistent. Protamone samples, in both breeds, had the highest pH values, the untreated samples and the thiouracil samples had approximately similar pH values. The change in all samples toward an acid reaction during the first twelve weeks of storage might have been due to lactic acid formation. The following change in pH during the latter twelve weeks of storage toward a less acid reaction has to do with meat spoilage and decomposition. One observation made at the end of the storage period was that after twenty-four weeks the thiouracil sample from the Duroc breed had the lowest pH value while the protamone remained the highest in pH value. It is rather regrettable that this observation could not be made with the thiouracil sample from the Berkshire breed due to the small number of these samples available for the last test. The pH value, as we all know, is

connected with meat spoilage, decomposition and rancidity, but whether the change in pH value is of any significant importance to the packer or consumer remains open for discussion and investigation.

Tenderness

The only important conclusion is based on an overall picture of the difference observed between the raw samples from the three treatments in both breeds. Using chi-square a significant difference in tenderness between untreated samples and both treated samples was found; the treated samples were more tender than the untreated ones. On the other hand there was not a significant difference between both treated samples. This may be due to the amount of drugs used in each case. Just how tenderness is affected through the use of both drugs remains to be solved.

Fat Content

The feeding of thiouracil and thyroprotein (protamone), in the amounts used in this work, did affect the amount and location of fat deposited in the body of the pigs. Thiouracil tends to cause the formation of a large amount of fat except that most of it is formed on the outside and not within the muscle tissue. Protamone which

causes an increase in metabolic activity with more muscle tissue formed than fat still causes most of the fat formed in the body to be deposited within the muscle tissue. It is for this last reason that the amount of fat in the lean of protamone-treated pigs is more than that in the lean of thiouracil-treated pigs.

Rancidity Development

From data thus far obtained it seems evident that protamone treatment tends to favor rancidity development, as compared to the untreated samples, possibly through an increase in fat content of the lean portion of the meat or through an influence on the fatty acids deposited. On the other hand the thiouracil treatment apparently retards oxidation. The fat of these samples gave zero peroxide values throughout the 13 weeks storage period at the higher temperature. In this case it is possible to assume that the retarding effect of thiouracil treatment might have been due to a smaller amount of fat within the lean meat in comparison with either the control or the protamone samples. Although it has been shown that thiouracil is almost lost from the tissues within a few days following slaughter it is possible that some traces of this drug still remain. Since thiouracil is an effective antioxidant, traces remaining in the tissue would probably act to retard fat oxidation.

CHAPTER VII

Conclusion

1. From observations at the time of slaughter as well as
results obtained from the freezer-storage of drug-
treated meat it seems that the oral administration of
0.1 per cent by weight of thiouracil mixed with the
feed of pigs has unsatisfactory effects on the quality
of meat even if the drug is fed for the short period
of 47 days.

    a. Most of the fat in the body of the pig is
    formed largely outside and not within the
    muscle tissue.

    b. There is a large loss in the weight of the
    meat through drip during thawing.

    c. Chops become limp upon thawing and lose their
    shape.

2. The apparent advantages from feeding thiouracil, in
regard to the quality of meat, are:

    a. Meat becomes more tender

    b. Rancidity development in ground ham is re-
    tarded for longer periods of time than usually
    observed with untreated meat, probably because
    of the smaller fat content within the lean.

3. The use of 0.04 per cent thyroprotein, by weight, mixed

with the feed of pigs seems to have more advantages in regard to the final quality of meat over thiouracil treatment.

    a. The meat becomes more tender than untreated meat.

    b. There is less drip from the frozen-defrosted chops.

    c. The chops hold their form well following thawing.

    d. Fat is deposited within the muscle tissue, a desirable characteristic demanded by the consumer.

4. The only objections to the use of thyroprotein lie in the fact that:

    a. The chops become too greasy to the touch.

    b. The ground ham is more susceptible to rancidity development.

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