A Simple Centrifugal Method for Measuring Expressible Moisture, A Water-Binding Property of Muscle Foods

C. A. JAUREGUI, J. M. REGENSTEIN, and R. C. BAKER

ABSTRACT

With the development of more refined methods of measuring water-binding properties of meats, the term "water-holding capacity" needs to be replaced with more specific and carefully defined terms such as expressible moisture, water-binding potential, and free drip.

An improved method of measuring expressible moisture is described which is simple and reproducible. It basically measures the amount of liquid squeezed out of a protein system with centrifugal force, by measuring the weight gain of a filter paper surrounding the sample. This method seems to be highly sensitive to factors that affect the water-binding properties of muscle foods.

INTRODUCTION

WATER IS BOUND to the protein in muscle with many different binding constants, but there is no clearcut demarcation between the various types of water-protein interactions. According to Hamm (1960), the majority of water in muscle is in the form of "free immobilized water." If a force is applied to a meat system, part of this water is released as "loose water." Measurements of loose water released under application of force have been used as indicators of water-binding properties of proteins, and are commonly referred to as "water-holding capacity" (Chou and Morr, 1979; Regenstein et al., 1979; Vadehra et al., 1973).

To mitigate the confusion caused by improper use of the term water-holding capacity, we propose the use of three terms: (1) water-binding potential, (2) expressible moisture, and (3) free drip.

Water-binding potential (WBP) refers to the ability of a protein system to hold water present in excess and under the influence of an external force. It therefore represents potential maximum water retention of a protein system under the measurement conditions (Regenstein et al., 1979).

Expressible moisture refers to the amount of liquid squeezed from a protein system by the application of force, and measures the amount of loose water released under the measurement conditions. No absolute figures exist since the amount of loose water depends upon the type and amount of force applied.

Free drip refers to the amount of liquid lost by a protein system without application of external force other than gravity.

A simple method of measuring expressible moisture is described which combines a gravimetric adaptation of the filter paper press method (Karmas and Turk, 1976) with application of a centrifugal force (Miller et al., 1968). This method, which is similar to that of Dagbjartsson and Solberg (1972), is simple and reproducible, and allows simultaneous multiple sample treatments; it avoids previously used centrifugal methods calling for special equipment (Wiebiciki et al., 1957).

EXPERIMENTAL

FISH, CHICKEN AND BEEF muscle were used in this study. Fresh rainbow trout (Salmo gairdneri) obtained from Tunison Laboratory of Fish Nutrition, Cortland, NY, were killed by removal of the head, placed in ice until resolution of rigor (2-3 days), filleted, and divested of all visible fat and dark muscle. A ready-to-cook broiler and a beef sirloin tip steak from a local supermarket were prepared as follows. The chicken was deboned and skinned by hand; dark and light meat were obtained from legs and breast. The beef quadsiceps femoris muscles were removed and surrounding fat and connective tissue eliminated. The various muscles were cut into pieces and ground through a precooled grinder using a 1.6 mm plate (Hobart model N50 mixer with Kitchen Aid grinder attachment). The ground muscles were stored in ice.

To determine expressible moisture, three pieces of Whatman #3 filter paper, 5.5 cm in diameter, and one piece of Whatman #50, 7.0 cm in diameter, were folded into a thimble shape over the outside of an inverted 16 x 150 mm test tube with the #50 filter paper as the internal surface of the thimble. The filter paper was weighed on a Mettler H100 balance before and after addition of 1.5 ± 0.3 g sample of ground muscle. The sample in the thimble was then centrifuged in a 50 ml polycarbonate centrifuge tube at various speeds and times in a refrigerated centrifuge (Sorvall RC2B; SS34 rotor) at 2°C. The filter paper and sample were then removed from the tube with tweezers, the meat "cake" removed from the filter paper, and the paper reweighed. All samples were run in quadruplicate and the expressible moisture reported as percent weight lost from original sample.

---Continued on page 1273---

Table 1—Effect of centrifugation time and rotor speed on an expressible moisture a,b of ground trout muscle

<table>
<thead>
<tr>
<th>Time (min)</th>
<th>Rotor speed (rpm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>8,000 (7,100 xG)</td>
<td>12,000 (17,300 xG)</td>
</tr>
<tr>
<td>10</td>
<td>29.6 ± 1.1 e</td>
</tr>
<tr>
<td>15</td>
<td>29.6 ± 1.6 e</td>
</tr>
<tr>
<td>30</td>
<td>32.2 ± 1.7 e</td>
</tr>
</tbody>
</table>

a Expressed as the percent of the original weight lost after centrifugation.
b Mean ± standard deviation, n = 4.
c All means followed by the same superscript letters were not significantly different (p < 0.05).
d Measured at the bottom of the centrifuge tube.

Table 2—Comparison of the expressible moisture values obtained for quadsiceps femoris muscle of beef, light and dark meat of chicken, and white muscle of rainbow trout

<table>
<thead>
<tr>
<th>Species</th>
<th>Expressible moisture a,b (%)</th>
<th>Initial moisture (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Beef</td>
<td>38.2 ± 1.7</td>
<td>76.8</td>
</tr>
<tr>
<td>Chicken</td>
<td>Light meat</td>
<td>22.9 ± 1.2</td>
</tr>
<tr>
<td></td>
<td>Dark meat</td>
<td>21.2 ± 1.7</td>
</tr>
<tr>
<td></td>
<td>Rainbow trout</td>
<td>39.0 ± 1.9</td>
</tr>
</tbody>
</table>

a Expressed as the percent of the original weight lost after centrifugation for 15 min at 16,000 rpm.
b Mean ± standard deviation, n = 4.
A Research Note
The Use of Mechanically Separated Spleen in Meat Patties

R. J. BITTEL and P. P. GRAHAM

ABSTRACT
Ground meat (beef) patties containing 0, 5, and 10% mechanically separated spleen (MSS) yielded 69% after oven broiling. Control patties contained more fat and less copper and zinc than the 5 and 10% MSS products. Products made with 5 and 10% MSS contained 2.6 and 3.8 times more iron, respectively, than the control. Taste panel data indicated no significant differences between the patties with respect to juiciness, flavor, mouthfeel, or overall acceptability. Results indicated that up to 10% MSS can be added to ground beef without adversely affecting consumer acceptance of the cooked product.

INTRODUCTION
THE BOVINE SPLEEN holds great potential as a source of animal protein and iron for the natural fortification of comminuted meat products. The removal of connective tissue from beef spleen using a mechanical deboner fitted with a desinewing head (Bittel et al., 1981) produced splenic pulp for use in comminuted meat products.

The use of MSS, a nonskeletal tissue, in products labeled "ground beef" or "hamburger" is prohibited by Federal Regulation, United States Department of Agriculture (USDA, 1973). However, its use in patty products with unique names or in those intended for institutional serving, such as school lunches, may be permissible.

This study was conducted to evaluate the effects of adding mechanically separated spleen (MSS) to meat (beef) patties on the organoleptic and nutritional attributes of the cooked product.

MATERIALS & METHODS
Patty formulation and preparation
Ground meat patties were produced from ground beef and MSS to contain 0, 5, and 10% spleen. Approximately 9.1 kg of boneless beef chuck containing an estimated 20% fat was ground through a 1.27 cm plate. Units of ground chuck weighing 2.27, 2.15, and 2.04 kg were thoroughly mixed with 0, 113.4, and 226.8 g of MSS, respectively. One percent salt (22.7 g) was included in each product. The hand-mixed products were ground through a 0.32 cm plate, remixed and chilled. Patties were formed with the aid of a 10.5 cm diameter x 1.5 cm deep plastic mold, weighed and oven broiled for 7 min per side. Final weights were taken and shrink losses were calculated.

Proximate analysis
Four cooked patties from each spleen addition level were ground through a 0.5 cm plate. Triplicate 100g samples were weighed into 150 x 25 mm, plastic, tissue culture dishes and frozen.

Moisture was determined by lyophilization (Bittel and Graham, 1977). Each freeze-dried sample was then finely broken in a Waring Blender and distributed into three 118-ml glass jars. The samples were freeze-dried again and held in desiccators until needed for proximate and elemental analysis. Crude protein, ash and other extract determinations were made on the dried samples by AOAC (1975) methods.

Elemental analysis
Iron, copper, and zinc determinations were made on freeze-dried samples of the cooked patties. Triplicate samples were hydrolyzed in duplicate by the method of Bittel et al. (1981). Elemental concentrations were determined using a Perkin-Elmer 403 atomic absorption spectrophotometer in accordance with the manufacturer's specifications (Perkin-Elmer, 1976). Values were expressed as µg element/g cooked patty.

Taste panel
Immediately after cooking, the meat patties were cut into quarters, coded and served to an untrained 18-member taste panel. Patties were evaluated and scored on 5-point rating scales for juiciness (5 = very juicy, 1 = very dry), flavor and mouthfeel (5 = very desirable, 1 = very undesirable) and overall acceptability (5 = very acceptable, 1 = very unacceptable).

Statistical analysis
Data collected from the experiments were subjected to analysis of variance and Duncan's multiple range test according to the procedures of Bar et al. (1976).

RESULTS & DISCUSSION
THE UNCOOKED MEAT MIXES WITH 5 and 10% MSS were darker in appearance than the control due to the abundant quantities of hemoglobin contributed by the spleen. After cooking, all products were similar in appearance. As shown in Table 1, level of MSS did not affect cooking yield. There were no significant differences among the cooked meat patties in proximate composition, except that the control patties contained more fat than those with 5 or 10% MSS.

The elemental content of the cooked meat patties is presented in Table 1. Patties made with 5 and 10% MSS contained more copper than the control. This result may be explained by the lower fat content of the treated patties and the close similarity between spleen and muscle in copper concentration. At the levels determined (0.7 - 0.9 ppm), 100g of cooked patty would provide 3.5 - 4.5% of the 2 mg RDA for copper (Schmidt, 1976). Level of added spleen did not consistently affect the zinc content of the

Table 1—Cooked meat patty characteristics

<table>
<thead>
<tr>
<th>Level of spleen (%)</th>
<th>0</th>
<th>5</th>
<th>10</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cooking yield (%)</td>
<td>69.2</td>
<td>68.1</td>
<td>68.0</td>
</tr>
<tr>
<td>Moisture,µg/g</td>
<td>53.9</td>
<td>53.6</td>
<td>54.3</td>
</tr>
<tr>
<td>Fat, µg/g</td>
<td>19.1</td>
<td>17.6</td>
<td>17.1</td>
</tr>
<tr>
<td>Protein, µg/g</td>
<td>25.4</td>
<td>26.3</td>
<td>26.0</td>
</tr>
<tr>
<td>Ash, µg/g</td>
<td>2.1</td>
<td>2.0</td>
<td>2.1</td>
</tr>
<tr>
<td>Fe,µg/g</td>
<td>30.8</td>
<td>79.1</td>
<td>118.2</td>
</tr>
<tr>
<td>Cu,µg/g</td>
<td>0.7</td>
<td>0.9</td>
<td>0.9</td>
</tr>
<tr>
<td>Zn,µg/g</td>
<td>46.5</td>
<td>49.1</td>
<td>46.3</td>
</tr>
</tbody>
</table>

a. Oven broiled 7 min/side.

b. Means of triplicate analyses, Expressed as percent on a wet basis.

c. Means of six hydrolysates, Expressed as µg/g or ppm cooked patty.

d. Means in the same row with different letters are significantly different (P < 0.05).

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RESULTS & DISCUSSION

EXPRESSIBLE MOISTURE VALUES for different combinations of centrifugation times and speeds are shown in Table 1. The low standard deviations are an indication of reproducibility of the method. Higher values were observed with longer times and higher rotor speeds, but expressible moisture was more affected by the latter than the former, indicating that most of the moisture is removed during the first 15 min. All subsequent measurements used 15 min and 16,000 rpm to parallel conditions of Regenstein et al. (1979) for WBP.

Expressible moisture values obtained for the three species studied are shown in Table 2, to illustrate the possibility of using the method for different species without losing reproducibility. For even greater accuracy, we recommend, when applicable, the use of an appropriate untreated control in the centrifuge run as the experimental samples (Regenstein et al., 1979). Composition of the fluid in the filter paper may also be analyzed, to distinguish water from fat, iron for the fortification of ground meat products.

USE OF SPLEEN IN MEAT PATTIES...

REFERENCES


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REFERENCES


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Effects of Salt and Phosphates on Some Functional Characteristics of Hand and Mechanically Deboned Turkey Meat

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ABSTRACT
Mechanically deboned turkey meat (MDTM) and hand deboned, ground turkey meat were evaluated for proximate composition and percentage salt soluble proteins, as well as for water binding (WBC), water holding (WHC), and emulsifying capacities (EC), using several salt solutions. MDTM was found to have less protein and more fat than hand deboned, ground turkey meat, as expected. Addition of 0.3% phosphate to a 3% NaCl solution increased the amount of extractable protein obtained from both meat systems; however, a significant interaction between meat types and solutions occurred. The MDTM was affected to a lesser degree by the phosphates than the hand deboned meat. WBC, WBC, and EC in hand deboned meats followed expected trends with improved function when salt was added, and a combination of NaCl and phosphate salts had a synergistic effect. However, MDTM showed unexpected results, with a 0.3% phosphate solution improving WBC, and EC to a greater extent than either 3% NaCl alone or a 3% NaCl-0.5% phosphate combination.

INTRODUCTION

Due to the fine consistency of mechanically deboned turkey meat (MDTM), it is used frequently in comminuted meat products. The binding of these products has been shown to be due to the salt-solubilized proteins (Fukazawa et al., 1961). MDTM is less stable than hand deboned meat in comminuted products, a fact attributed to its higher fat and collagen levels and to its initially lower protein content which is rendered partially insoluble by the heat and friction of processing (Froning, 1970).

Several workers have improved the functionality of MDTM for use in sausage products. Froning and Jankay (1971) examined the effects of pH adjustment and salt preblending, Froning and Johnson (1973) evaluated centrifugation, while Acton (1973) used salt preblending, extrusion, and heat processing to form texturized strands of deboned meat, all of which improved the functional properties of MDTM. Froning (1973) reported that 6% polyphosphates added to chill solution for fowl significantly increased emulsification capacity and stability of mechanically deboned meat from these birds. Sodium chloride and the sodium salts of condensed phosphates have been shown to improve the binding and water-holding capacities of meat, and have influenced the pH, swelling, and structure of muscle proteins (Shuils and Wierwicki, 1973; Swift and Ellis, 1956, 1957). Maesso et al. (1970) and Froning (1965) found that the addition of phosphates to chicken rolls and ground turkey patties improved binding and decreased cooking losses. The exact mechanism is not fully understood, but the action of phosphates has been related to the change in pH, to the ionic strength of the solution, and to the physical action of the phosphate on the protein, resulting in smaller molecules with improved water binding characteristics (Fukazawa et al., 1961; Sherman, 1961a, 1962).

Several workers have reported on the synergistic effect of NaCl and phosphate salts in improving the functional characteristics of meat systems (Swift and Ellis, 1957; Sherman, 1962; Shuils and Wierwicki, 1973). A
combination of these salts resulted in greater improvement than either one alone.

This study attempted to examine the effect of several NaCl/phosphate salt solutions on some functional characteristics of hand deboned, ground turkey meat and mechanically deboned turkey meat.

MATERIALS AND METHODS

Meat Source. Commercially obtained tom turkey breasts and thighs were ground through 4 mm. plates of an electric grinder and mixed 50:50 by weight, sealed in Cryovac® bags (Cryovac Division, W. R. Grace Co.) in experimental proportions, and held at -18°C for a maximum of 1 month until used. MDTM from the rack and necks (Beehive AV-968 deboner, setting E-10) was obtained from the same commercial source, bagged in experimental proportions, and held at -18°C within 3 hrs. of deboning. The meat was thawed overnight at room temperature and evaluated for proximate composition, % soluble proteins, water binding, water holding, and emulsification capacities, using several salt solutions, as described later. The salt solutions used in the evaluations are listed in Table 1. Duplicate or triplicate analyses were performed, as indicated for each analysis, on replicated samples of product.

Proximate Composition. Fat, moisture, and protein values were determined for both hand deboned, ground turkey meat and for mechanically deboned meat samples, before the salts were added, using A.O.A.C. (1965) methods: 23.005, 23.003, 23.009 respectively.

% Soluble Proteins. The method for determining salt soluble protein was similar to that of Saffle and Galbreath (1964). Duplicate 5 gm. samples of meat were ground in a mortar with sand and salt solution (see Table 1). The mixture was then transferred quantitatively to a 400 ml. beaker, using a total of 250 ml. solution, and stirred for 15 min. with a magnetic stirring bar; the slurry was then centrifuged for 10 min. at room temperature at 3020 x g. Micro-Kjeldahl protein determinations were done on triplicate 25 ml. aliquots of the supernatant. The remaining sample was treated with 50% TCA to precipitate the protein, and triplicate 25 ml. aliquots of the remaining liquid were digested by the micro-Kjeldahl method to determine nonprotein nitrogen. Protein was determined by multiplying the nitrogen value by a factor of 6.25: actual soluble protein was calculated by subtracting the nonprotein nitrogen from the total soluble nitrogen value. The soluble protein was expressed as both a % of the total meat sample, and as a % of the total protein present.

Water Binding Capacity (WBC). WBC was expressed as the percentage swell due to absorbed water, and was determined by a modification of the method of Shults et al. (1972). A high value indicates high binding potential. Triplicate 50 gm. meat samples were mixed with 150 ml of salt solution (see Table 1) for 90 sec. at medium speed in a 400 ml beaker, using a magnetic stirring bar. The slurry was then transferred to a 25°C room for 1 hr. The supernatant fluid was weighed, and the percentage water (Wt. %) was calculated.

Table 1: Salt mixtures used to evaluate certain objective characteristics of MDTM and hand deboned, ground turkey meat systems

<table>
<thead>
<tr>
<th>Objective evaluation</th>
<th>Salt mixtures used</th>
</tr>
</thead>
<tbody>
<tr>
<td>Water binding capacity</td>
<td>Deionized distilled (dd) water</td>
</tr>
<tr>
<td>Emulsification capacity</td>
<td>6 M NaCl with dd water 5% phosphate (w/v.) with dd water</td>
</tr>
<tr>
<td>Salt soluble protein</td>
<td>6 M NaCl + 5% phosphate with dd water</td>
</tr>
<tr>
<td>Water holding capacity</td>
<td>Control (no additive) 3% NaCl, dry (by weight) 5% phosphate, dry (by weight) 3% NaCl + 5% phosphate, dry (by weight)</td>
</tr>
</tbody>
</table>

*Phosphate refers to Kena, a commercial food grade sodium phosphate produced by Calgon Corp., Pittsburg, PA.*

Water H defined last dual: A low water samples weighed sealed, water binding was reweighed, and the supernatant supernate (1965) was by the f % water.

Where % water in water was:

Emulsifies it fat glob and...
ned, as indicated for each analyzed samples of product.

Composition. Fat, moisture, and HAC were determined for both ground turkey meat and for deboned meat samples, before they were added, using A.O.A.C. (1960) 005, 23.003, 23.009 respectively.

Protein. The method for determinable protein was similar to that described by Galbreath (1964). Duplicate samples of meat were ground under a mortar and pestle and added to a 250 ml. centrifuge bottle and spun at room temperature for 15 min. at 1000 r.p.m.

The supernatant was decanted into a graduated cylinder and measured. The percentage swell, or water bound by the system was calculated by this formula:

\[ S = \frac{\text{ml. supernatant}}{50 \text{ gm.}} \times 100 \]

Water Holding Capacity (WHC). WHC was defined as the % shrinkage, or the moisture lost during heating (Wierbicki et al., 1967). A low value is desirable. Triplicate 50 gm. samples of blended meat (Table 1) were weighed into 250 ml. centrifuge bottles, sealed, and heated for 30 min. in a 70°C water bath. The bottles were cooled in running water (2°C) for another 30 min., then weighed to determine there was no evaporation loss, and then centrifuged at 170 x g for 15 min. at room temperature. The supernatant was collected and measured, and the moisture content of both the raw sample and the supernatant was determined by A.O.A.C. (1963) methods. The WHC was calculated by the following formula:

\[ \% \text{ water lost (WHC)} = \frac{\text{ml. supernatant} \times F}{G} \times 100 \]

Where F = % water in supernatant. G = % water in sample. x sample wt. = gms. water in sample.

Emulsification Capacity (EC). EC measures the ability of the system to stabilize fat globules dispersed in an aqueous media, and was determined by the method of Kuehler and Steele (1974), which is a recent modification of the method originally used by Swift et al. (1961). Duplicate 50 gm. meat samples were blended with 200 ml. salt solution (Table 1) for 2 min. in a Waring microblender cup at 4°C. The slurry was poured into a 250 ml. centrifuge bottle and spun at 10,000 r.p.m. for 20 min. at 4°C. Triplicate five ml. aliquots of the supernatant were mixed with 45 ml. of solution in a 400 ml. beaker. Corn oil was added at a constant rate of about 1 ml./sec. while the mixture was continuously stirred with a Hamilton Beach single rotor blender. The emulsion was considered broken when electrical resistance across the mixture, as measured by a volt meter, reached infinity. The EC was expressed as both ml. of oil emulsified/gm. meat and as ml. oil emulsified/gm. protein.

Statistical Analysis. All data were subjected to a two-way analysis of variance test for significance, using Multivariate, a computer program designed for use by the IBM 6500 computer, Finn (1967).

RESULTS AND DISCUSSION

Composition and Soluble Proteins. The percentage of fat, water, and protein found are reported in Table 2. These values show slightly higher protein values, but generally correspond to values reported in the literature (Froning and Janya, 1971; Grunden et al., 1972). MDTM has a higher level of fat and a lower proportion of protein than hand deboned meat because of the source of some of the components in the product from bone marrow.

<table>
<thead>
<tr>
<th>Table 2. — Proximate composition of raw, hand deboned turkey meat, mixed breasts/thighs, 50:50, and of raw mechanically deboned turkey racks and backs</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fat</td>
</tr>
<tr>
<td>-----</td>
</tr>
<tr>
<td>Hand deboned</td>
</tr>
<tr>
<td>Mechanically deboned</td>
</tr>
</tbody>
</table>

1 Values reported are means of 3 samples.
Salt soluble proteins are responsible for binding (Fukazawa et al., 1961). MDTM is subjected to heat and frictional stress during processing, with a potential decrease in protein solubility due to denaturation. MDTM contained only 16% total protein, compared to 22% in hand deboned meat, thus less protein was available initially. Table 3 reports the percentages of protein extracts by salts from MDTM and hand deboned, ground turkey meat. Only about 4% (total weight) or about 25% (total protein) was extracted from MDTM with a 3% saline solution. Almost 8% (total weight) or 36% (total protein) was extracted from hand deboned, ground meat. Addition of 0.5% phosphate caused only 2% more total weight to be extracted from MDTM, (reflecting a 2% increase in total protein), as compared to 2.4% increase in total weight (an 11% increase in total protein) when the hand deboned system was treated with the phosphate.

This suggests that the effect of phosphates in enhancing the extraction of salt soluble proteins from MDTM is not as great as their effect on hand deboned meat. Perhaps this is due to the smaller amounts of protein initially present. The types of meat reacted differently when treated with phosphate salt solutions.

**Water Binding, Water Holding, and Emulsification Capacities.** Data obtained in this study relative to water binding, water holding, and emulsification capacities for MDTM and hand deboned, ground turkey meat are reported in Table 4 and Figure 1. A 3% NaCl solution increased the water binding and emulsification capacities of hand deboned meat, while decreasing the water lost during heating. A combination of sodium chloride and phosphate salts has a synergistic effect on these functional properties, resulting in more favorable functioning with the combined salts than with either one singly. These data follow trends reported by Shultz and Wierbicki (1973).

A significant interaction occurred between solutions and meat types. The effect of each salt type extracted the c

![Image](https://source.unsplash.com/random/800x600)

**Fig. 1.** Some functional characteristics of turkey meat with salt and phosphate added. C—control; S—3% salt; K—0.3% phosphate.

**Table 4.** Two-way multi-variate analysis of variance with salt solutions and meat types as independent variables and water holding, water binding, emulsification capacities as dependent variables

<table>
<thead>
<tr>
<th>F—Statistic</th>
<th>d.f.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Effect—Solutions</td>
<td>29.7694*</td>
</tr>
<tr>
<td>Effect—Meat types</td>
<td>33.1026*</td>
</tr>
<tr>
<td>Interactions</td>
<td>545.2014*</td>
</tr>
</tbody>
</table>

*indicates significance at P < .01.
Deboned Turkey Meat

Water binding, water holding capacities for MDTM and ground turkey meat are and Figure 1. A 3% NaCl, the water binding and capacities of hand deboned the water lost during addition of sodium chloride has a synergistic effect properties, resulting in functioning with the combination either one singly. These reported by Shults and action occurred between types. The effect of each

| Multi-variate analysis of solutions and meat types as and water holding, water | capacities as dependent variables |
|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|
| 29.709*         | 1 & 8           | 0.026*          | 1 & 8           | 0.014*          | 1 & 8           | 0.014*          | 1 & 8           | 0.014*          | 1 & 8           |
| t < .01         |                  | t < .01         |                  | t < .01         |                  | t < .01         |                  | t < .01         |                  |


The solution is dependent on the particular meat type in question; thus it is impossible to extricate and determine the significance of the effect of the solution.

The data from the MDTM system shows an unexpected trend in results, consistent throughout all three tests. (See Table 4 and Figure 1) It was anticipated that the sodium chloride and phosphate salt additions would improve functional properties, with an even greater improvement being observed when a combination of both salts was used. However, the phosphate treated meat showed the most improvement in functionality, while the phosphate-sodium chloride combination showed only a slight increase in functionality over the NaCl alone. No explanation for this was found in the literature. It is difficult to understand why products treated with 0.5% phosphates alone showed improved functionality while a combination of phosphates and NaCl did not.

Sherman (1961b) discussed emulsion formation with alkaline phosphates and the free fatty acids of meat fats. These emulsions were of mere academic interests, he felt, as they were destroyed by the addition of NaCl, a vital ingredient in sausages, and the free fatty acids required for their formation are limited in normal sausage meats (i.e., beef and pork).

It is possible that the water binding, water holding, and emulsification capacities of MDTM were maintained by a phosphate stabilized emulsion as well as by extracted protein. Addition of NaCl in this case would destroy the emulsion and thus decrease functionality, the observed result.

Mechanically deboned and hand deboned meat systems differ both in composition and in the nature and source of their constituents. A large proportion of the proteins in MDTM were denatured, a phenomenon not found in hand deboned meat; the lipids in the MDTM included those from bone marrow and interstitial tissue, while those in hand deboned meat were almost exclusively from muscle sources; the lipids in MDTM were subjected to the heat and friction of processing, which might have altered their characteristics. The observed differences in reaction to added NaCl and phosphate salts might well be due to these compositional differences between the meat systems.

Sodium chloride is considered essential in sausage formulations. Besides dissolving the salt-soluble proteins needed for binding, salt inhibits bacterial growth and produces a characteristic flavor. While some work has shown that phosphates alone can extract sufficient proteins for binding (Hellerdorn, 1962), the salt is essential for shelf life and flavor. Since NaCl apparently destroys the effect of phosphate salts on MDTM, yet is vital in sausage formulations, it appears that the full potential of phosphates in improving the functionality of MDTM will be realized. Further research is needed to identify the reactions involved, and to investigate practical applications of this knowledge.

MDTM was found to have less emulsification capacity than hand deboned meat, both on a meat and on a protein basis. The reasons for this have already been discussed, and related to the amounts and nature of the proteins available. However, the data for water holding and water binding capacities show higher values for MDTM than for hand deboned meat.

Emulsification capacity was determined on the system as a whole; the meat and solution were blended to a slurry. However, the other two tests were designed to measure the water holding and water binding capacities that would be exhibited in actual production of a semi-dry sausage. Therefore, the meats were evaluated in the form in which they would be incorporated into the sausages: hand deboned meat was coarsely ground, and MDTM was in puree form. Despite the lower actual total protein content of MDTM, more protein would likely be immediately available.
for extraction due to the increased surface area of the meat. A longer extraction time for the ground meat would have increased functionality and given a more valid representation of an actual production situation, since there is a 36 hour time period prior to the cook cycle during which extraction of proteins from meat particles occurs. For this reason, attention was focused primarily on trends within the meat system, rather than on a comparative basis for a given solution.

REFERENCES


Effect of Auger- and Press-Type Mechanical Deboning Machines on Selected Characteristics of Mechanically Deboned Poultry

M. G. MAST, Th. G. UITTENBOGAART, A. R. GERRITS, and A. W. DEVRIES

ABSTRACT

Selected chemical, functional, textural, and sensory properties were evaluated for mechanically deboned poultry (MDP) prepared from three auger-type (Paoli, Beehive, and Yieldmaster) and one press-type (Procon) mechanical deboners. Protecon MDP, prepared without preliminary grinding of the incoming raw material, contained slightly larger intact muscle fibers than MDP from the auger-type machines. Although variation occurred among the four meats, no significant differences (P > 0.05) were observed in their composition. Differences existed in the emulsifying capacity, emulsion stability, and waterholding capacity of the meats; however, all were successfully used in the preparation of acceptable frankfurters. Frankfurters prepared from Protecon or Beehive MDP were generally firmer, as determined by shear tests and sensory evaluations.

INTRODUCTION

MECHANICALLY DEBONED POULTRY (MDP) has been used in food products in the United States since about 1965 (Murphy et al., 1979). It is primarily used in the manufacture of emulsion-type products such as frankfurters.

In Western Europe, where the Codex Alimentarius term "separated meat" is used, MDP is primarily used in soup broth, luncheon meat, and sausages, but not in the American-style frankfurter.

Poultry processors in the U.S. and Europe predominantly use auger-type deboning machines which work on a two-stage continuous process, i.e., the raw material is first chopped into smaller portions, after which the edible meat or soft material is separated from the bone or hard material via screens or filter plates.

A one-step press-type process is employed in a Dutch-built deboner currently used in Europe. Bones with adhering meat are placed directly into the chamber of the machine without preliminary grinding or breaking. Residual meat is then separated from the bones by the application of high pressure (2-3 tons/in² or 315-473 kg/cm²) forcing the meat through a set of filters.

Several studies have been reported in which more than one type of MDP was utilized, usually for the preparation of emulsion-type products. Baker et al. (1974) found a wide range of efficiency in the MDP from three deboners (two commercial and a laboratory model), when used in the production of frankfurters. Since this efficiency was unrelated to the fat content of the MDP, they concluded that other factors, such as the design of the machine, pressures involved in forcing the material out of the machine, etc., may be involved.

Krol et al. (1975) used varying amounts of MDP from Paoli, Bihun, or Beehive machines to prepare three sausage-type products. They reported that no differences existed in the chemical composition of the MDP from these machines. Products made from MDP from one of the machines were scored lower by sensory panels. However, these authors concluded that variations in handling practices may have accounted for these differences. In a study comparing MDP from two different deboners (Septromatic and Beehive) and raw materials (poultry back and necks) from three different plants, Orr and Wogar (1979) reported that the source of raw material had a greater influence on emulsification characteristics and composition than the type of deboning machine.

Although the type of deboning machine was not always indicated in the above studies, it is assumed that all were auger-type machines, since this is the type of machine which has generally been used for poultry meat, and not press-type machines. Due to the different processes involved with these two types of machines, the characteristics of the final MDP may also be different. Therefore, the objective of this study was to evaluate selected chemical, physical, functional, and sensory properties of MDP prepared from both auger-type and press-type mechanical deboning machines.

MATERIALS & METHODS

Procurement of MDP

MDP used in this study was produced from broiler backs and necks (three parts backs plus one part necks by weight). These backs and necks were obtained from a common source, a poultry processing plant in the Netherlands using dry slaughtering and chilling practices, and then transported to locations (Netherlands and Belgium) with different types of deboners. Three auger-type deboners, Beehive, Paoli, and Yieldmaster, and one press-type deboner, Protecon, were used. The small quantity of raw material (30-40 kg) processed through each deboner did not permit accurate yield determinations; however, all machines yielded about 65 ± 5% MDP. The temperature of the incoming backs and necks was 28 ± 1°C. The temperature of the final MDP produced by Paoli and Protecon deboners was 15 ± 2°C, and 42 ± 2°C from Yieldmaster and Beehive machines. Following production, one of these machines, all MDP taken to Spelderholt Institute for Poultry Research where it was frozen (−30°C) and stored (−20°C) until evaluated.

Experimental design

A randomized block design, with three blocks (trials) and four treatments (meat types), was used in this experiment. In the first trial only three types of meat were used, since it was impossible to obtain material from the Beehive machine at that time. Therefore, there were 11 "batches" of meat analyzed in this experiment. Samples were taken from these batches and analyzed according to the following procedures. Depending upon the test performed, a minimum of three and a maximum of 15 measurements were made on each batch.

Preparation of frankfurters and meat patties

The procedures described by Mast et al. (1981) were used to prepare frankfurters and meat patties in this study. The formulation for the frankfurters is shown in Table 1. Emulsions were prepared with an Alexanderwerk cutter (bowl revolutions, 17/min; blade speed, 300 revolutions/min) by chopping to an endpoint temperature of 12.5°C, which required approximately 12 min. After emulsification, emulsions were extruded into Nojax 22 mm casings, hand tied into 14 cm links, and heat processed in a smoke-coker following the schedule in Table 2, which is a modification of a process recommended by Lenninck (1978).

---Continued on next page---
Table 1—Frankfurter formulation

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>Grams</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mechanically deboned poultry</td>
<td>4,000</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>100</td>
</tr>
<tr>
<td>Corn syrup solids(^a)</td>
<td>80</td>
</tr>
<tr>
<td>Dextrose</td>
<td>40</td>
</tr>
<tr>
<td>Phosphate(^b)</td>
<td>20</td>
</tr>
<tr>
<td>White pepper</td>
<td>8</td>
</tr>
<tr>
<td>Nutmeg</td>
<td>2.8</td>
</tr>
<tr>
<td>Ascorbic acid</td>
<td>2</td>
</tr>
<tr>
<td>Sodium nitrite</td>
<td>0.62</td>
</tr>
<tr>
<td>Water</td>
<td>400</td>
</tr>
</tbody>
</table>

\(^a\) Mor-Sweet 01934; CPC Nederland.  
\(^b\) Sodium pyrophosphate plus sodium acid pyrophosphate; Vass- sen-Schoemaker Chemische Industrie B.V.

Table 2—Processing conditions for frankfurters

<table>
<thead>
<tr>
<th>Process</th>
<th>Time (min)</th>
<th>Temp (°C)</th>
<th>RH (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pre-heating</td>
<td>30</td>
<td>46</td>
<td>80–90</td>
</tr>
<tr>
<td>Smoking</td>
<td>30</td>
<td>45</td>
<td>80–90</td>
</tr>
<tr>
<td>Drying</td>
<td>20</td>
<td>45</td>
<td>35–45</td>
</tr>
<tr>
<td>Pasteurizing</td>
<td>15</td>
<td>80</td>
<td>100</td>
</tr>
</tbody>
</table>

The internal temperature of the frankfurters after pasteurization was 73–75°C. Immediately after the heating process, frankfurters were rinsed in cool (8°C) water for about 30 min. Casings were then removed and frankfurters were held in plastic bags overnight at 4°C before vacuum packaging (6 pack).

Hamburger-like patties were prepared in order to determine cooking losses and evaluate the texture of the cooked MDP when used in a nonemulsified product, i.e., in contrast with frankfurters. Patties, with 2% salt added or without salt, were prepared.

Two kg of MDP, previously frozen (−20°C) and then thawed, were placed into the bowl of a Hobart Mixer (Model N-50) and mixed for 5 min at low speed. This ‘tumbling’ was done to thoroughly incorporate the salt into the meat. The meat was then stuffed into cellulose casings (85 mm diameter), which were sealed and hung in a −30°C air-blast freezer for 24–48 h. Frozen rolls were then cut on a band saw to obtain 1.5 ± 0.2 cm thick slices, each weighing approximately 80 g. Slices were accurately weighed, vacuum-packed in plastic bags (1.0 kg/cm²), and stored at −20°C until analyzed for cooking loss and texture.

Proximate analyses and calcium

NEN (1968) procedures were used to determine moisture (NEN 3440), protein (NEN 3442), fat (NEN 3443), and ash (NEN 3441) in samples of MDP and frankfurters. Calcium content was determined by a Corning Calcium Analyzer (Corning Ltd., Halstead Essex, England).

Emulsifying capacity (EC)

A modified system of Swift et al. (1961) was used to determine EC. One hundred ml of cold 1 M NaCl were added to 25 g of MDP in a blender jar and mixed for 2 min at 13,000 rpm with a Sorvall Omni-mixer. Twelve and one-half g of this slurry were placed into a second blender jar with an additional 37.5 ml of 1 M NaCl and mixed for 5 sec. Fifty ml of corn oil were added to this material and mixed at 13,000 rpm. Additional oil was then added at 0.8 ml/sec. The initial portion of this oil (approx. 75 ml) was added in buret; the final 20–50 ml were pumped in with a Dosimat (Metrosim Herisan, Switzerland). The Dosimat automatically stopped pumping at the moment that the mixture changed from an oil-in-water emulsion to water-in-oil. EC was reported as ml of oil emulsified per 2.5 g of tissue.

Emulsion stability

A modification of the method reported by Townsend et al. (1968) was used to evaluate emulsion stability. Samples for this test were obtained at the same time frankfurter emulsions were stirred in casings. Thirty-four grams of emulsion were placed into 10.8 × 2.5 cm centrifuge tubes, covered with Paraffin and held at 4°C for 1–2 hr. Emulsions with a constant temperature of 5°C were then placed into a 45°C water bath. The temperature of the water was then increased at 0.5°C/min until the internal temperature of the emulsion was 75°C. [These temperatures are slightly different from those used by Townsend et al. (1968) i.e., 48.8°C to 68.8°C; they were chosen since they approximate temperatures used in the heat process for frankfurters.] After heating, the contents of the tubes were placed into a funnel and the released liquids were collected in graduated centrifuge tubes. Total volume, fat volume, gel-water volume, and solids were determined and reported as ml/100 g.

Water holding capacity (WHC).

Although the term “water holding” is used by different authors to indicate different concepts, it generally refers to the loss of moisture from a meat system during heating or centrifugation or both. A modification of the method outlined by McMahon and Dawson (1976) was used to determine WHC in this study. Thirty grams of MDP were placed into six 10.8 × 2.5 cm centrifuge tubes. These tubes were sealed with Parafilm and placed into a 70°C water bath for 30 min. Immediately following heating, these tubes were removed from the water bath and cooled in running water (10–12°C) for 30 min. The contents of the tubes were then placed into funnels leading into graduated centrifuge tubes. After 5 min of draining, these tubes were sealed with Parafilm and centrifuged for 15 min at 3000 rpm. The ml of total liquid, fat, water and solids were then recorded.

The remaining three tubes were not placed into cool water; instead the contents were removed and the released material was measured in the same manner as described above. The determination of WHC from hot MDP is an addition to the method reported by McMahon and Dawson (1976). This was added to permit comparison of WHC of various types of MDP when evaluated hot or cold.

The moisture content of the raw MDP was determined; this value was used in the following formula to determine WHC:

\[
\text{WHC or % water retained} = \left(1 - \frac{\text{ml H}_{2}O \text{ in supernatant}}{\text{g H}_{2}O \text{ in raw sample}}\right) \times 100
\]

This formula has been modified from Wierzbicki et al. (1957) to obtain the percent of water retained by a sample; this more accurately reflects the name of the test, i.e., water holding capacity.

Cooking loss

Meat patties (see Preparation of frankfurters and meat patties) were used to determine cooking losses. Packaged meat patties were removed from the freezer (−20°C) and placed into a 90°C water bath for 20 min. Immediately after heating, the plastic pouch was cut open and the contents, released juices and meat patty, were placed into a funnel leading into a graduated centrifuge tube. The released material was centrifuged for 15 min at 3000 rpm. The total volume, fat, gel-water, and solids were then recorded as ml/100g MDP.

Shear measurements of frankfurters and meat patties

An Instron Testing Instrument, model TM-M, was used for all texture measurements in this study. Two different cells were employed to evaluate texture of frankfurters. The first was the standard Warner-Brazler cell, which was used to measure force required to make a cross-sectional cut through the frankfurter. Five frankfurters (each 14 × 1 cm long) from each group were sheared at three points, i.e., in the center and approximately 1.5 cm from each end. Shear tests were performed on frankfurters 6–8 days after production and subsequent storage at 4°C; the temperature of samples during evaluation was about 20°C. Intron settings for these tests were: 2 kg full scale deflection; chart speed, 10 cm/min; cross head speed, 5 cm/min.

The above test is useful for determining the total force required to "bite" through a frankfurter. However, much of the force is required to penetrate the skin of the frankfurter and not to shear through the core of the product. Therefore, strips of meat, 10 × 10 mm, were cut from the center of the frankfurter so that the variable of skin toughness could be eliminated. For this test, a Volodkovskiy-type cell was used. Remaining pieces from the Warner-Brazler test were used for these tests; 15 measurements were obtained for
were stuffed into 10.8 mm diameter at 4°C for 1 day. The meat was leached of fat for 8°C, then at 4°C, and the tubes were packed with obtained 100 g. 

Histology
Histological preparations, using Masson and Kossa stains (Romels, 1968), were prepared for each type of MDP and frankfurter evaluated in this study.

TBA tests
MDP and frankfurters were tested for oxidative rancidity using the thiobarbituric acid (TBA) test, as described by Tarladgis et al. (1960).

Statistical analyses
For each of the tests, other than sensory tests, the means of the repeated measurements on a batch of meat were computed; these means were then subjected to an analysis of variance. The estimated value of the missing unit, i.e., Beehive meat in trial 1, was calculated using the formula of Cochran and Cox (1957).

Sensory evaluation
Three different tests were used to evaluate the sensory attributes of frankfurters prepared in this study: triangle tests, to determine differences among treatments; paired comparison tests, to investigate which attributes caused differences, and finally a scaling method, in which the magnitude of these differences was determined for each attribute.

The sensory panel consisted of 12 employees of the Spelderholt Institute for Poultry Research. All were familiar with the tests used and the previously served as panelists to evaluate poultry products. However, since the American-style frankfurter is not a popular product in Europe, this was the first time most of the panelists evaluated frankfurters.

Frankfurters were evaluated after 5-7 days storage at 5°C. The frankfurters were tempered to approximately 20°C, cut into pieces 4 cm long, and served to panelists seated in light-controlled booths.

Scores for a particular frankfurter could possibly be influenced by the presence of other frankfurters evaluated in the study. Therefore, data from Trial 1 were not used, since unlike Trials 2 and 3, Beehive frankfurters were unavailable for comparison. For all three tests, presentation order and permutations were completely balanced to reduce potential secondary effects.

Tables prepared by Roessler et al. (1948) were used for analysis of triangle test data. Scores of the paired comparison tests were analyzed with techniques detailed by David (1969), and scaling scores were subjected to analysis of variance tests.

RESULTS & DISCUSSION
SEVERAL IMPORTANT FACTS must be considered regarding the limitations of this study when examining the results. Only one machine of each of the four types was used. Therefore, it may not be valid to assume that all machines of a certain type will give results comparable to those presented here. Since mechanical deboners can be adjusted for yield, etc., the nature of MDP may also vary within machine-type. Processors producing MDP for this study were requested to operate their mechanical deboners under conditions which they use in routine practice. All machines were located in food processing plants and were used to prepare MDP, except for the Protecon machine, which was located in the products laboratory of the Protecon equipment manufacturing factory.

Proximate analyses
The proximate analyses for MDP and frankfurters are presented in Table 3. Although considerable variation occurs among meat types, the composition of the various meats does not differ significantly (P>0.05). The variation in meat composition among the three trials was relatively high. For example, the amount of fat was generally lower in Trial 1 than in Trials 2 and 3. In addition to variation in fat composition, considerable variation occurred in calcium content of the four meat types. Beehive and Protecon meats were below 1% bone equivalent in all trials, Paoli meat was slightly about 1%, and Yieldmaster meat was about 2%. These results indicate that perhaps the Paoli, and particularly the Yieldmaster machine, may have been improperly adjusted, thereby permitting high bone equivalents (as determined from calcium content).

Emulsion capacity and stability
Results of the emulsifying capacity and emulsion stability tests are shown in Table 4. Meat systems with high emulsifying capacities, i.e., the ability to stabilize larger quantities of oil in an aqueous medium, also produced the most stable emulsions, i.e., the fat did not separate from the emulsion during heating. In our studies, meat from Protecon and Beehive machines had the highest emulsifying capacities and produced the most stable emulsions. These values differed significantly (P<0.05) from Yieldmaster meat but not from Paoli meat.

---Continued on next page---

Table 3—Proximate analyses of mechanically deboned poultry and frankfurters

<table>
<thead>
<tr>
<th></th>
<th>Protecon</th>
<th>MDP</th>
<th>Frankfurter</th>
<th>Paoli</th>
<th>Frankfurter</th>
<th>Yieldmaster</th>
<th>MDP</th>
<th>Frankfurter</th>
<th>MDP</th>
<th>Frankfurter</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein, %</td>
<td>15.0 ± 1.1a</td>
<td>13.7 ± 1.1</td>
<td>13.3 ± 1.2</td>
<td>12.8 ± 0.8</td>
<td>13.8 ± 0.6</td>
<td>13.0 ± 0.6</td>
<td>13.8 ± 1.2</td>
<td>12.6 ± 0.1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fat, %</td>
<td>19.7 ± 5.9</td>
<td>18.4 ± 4.9</td>
<td>23.2 ± 4.8</td>
<td>20.3 ± 3.3</td>
<td>19.5 ± 1.6</td>
<td>18.3 ± 1.2</td>
<td>24.8 ± 1.9</td>
<td>20.9 ± 1.6</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Moisture, %</td>
<td>64.3 ± 4.6</td>
<td>62.6 ± 3.6</td>
<td>62.3 ± 3.9</td>
<td>61.4 ± 2.3</td>
<td>64.7 ± 1.3</td>
<td>62.9 ± 1.1</td>
<td>60.7 ± 1.1</td>
<td>60.9 ± 1.2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ash, %</td>
<td>1.0 ± 0.1</td>
<td>3.0 ± 0.1</td>
<td>1.3 ± 0.1</td>
<td>3.2 ± 0.2</td>
<td>1.6 ± 0.6</td>
<td>3.5 ± 0.3</td>
<td>1.0 ± 0.0</td>
<td>3.0 ± 0.1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Calcium, %</td>
<td>0.12 ± 0.04</td>
<td>0.12 ± 0.02</td>
<td>0.20 ± 0.03</td>
<td>0.20 ± 0.02</td>
<td>0.35 ± 0.18</td>
<td>0.30 ± 0.11</td>
<td>0.11 ± 0.01</td>
<td>0.14 ± 0.01</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bone equivalent, %b</td>
<td>0.66</td>
<td>0.66</td>
<td>1.16</td>
<td>1.16</td>
<td>2.09</td>
<td>1.78</td>
<td>0.59</td>
<td>0.78</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Moisture/protein</td>
<td>4.3</td>
<td>4.6</td>
<td>4.7</td>
<td>4.8</td>
<td>4.7</td>
<td>4.8</td>
<td>4.4</td>
<td>4.8</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Moisture/fat</td>
<td>3.3</td>
<td>3.4</td>
<td>2.7</td>
<td>3.0</td>
<td>3.3</td>
<td>3.4</td>
<td>2.4</td>
<td>2.9</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

a Mean of three trials ± standard deviation. Exception, Beehive only has two trials.
b Bone equivalent = (% Ca - 0.015) x 6.25.
The emulsifying capacity results reported here are, in general, comparable to emulsifying capacities of various types of MDP as reported by Froning and Neelakantan (1971) and Mast and MacNeil (1976). They are, however, higher than results of Orr and Wogar (1979); those authors used the method of Swift et al. (1961), but added oil at 0.5 ml/sec instead of 0.8 ml/sec. According to Swift et al. (1961), the amount of fat emulsified increases as the rate of fat addition increases.

Although significant differences did exist among the emulsion stabilities of meat types, all emulsions prepared in this study proved to be stable and released only small quantities of fluid upon heating, i.e., generally less than 0.5%. In addition, no problems were encountered from emulsion breakdown or "fat caps" in the frankfurters. The addition of polyphosphates and corn syrup solids (see frankfurter formulation, Table 1) undoubtedly enhanced the stability of these emulsions.

**Water holding capacity (WHC)**

As indicated earlier, WHC was determined in two ways; first by the method of McMahon and Dawson (1976) whereby the heated meat was first chilled before collecting the released material, and second, by collecting the released material immediately without chilling. As would be expected, more water was retained in chilled samples than in nonchilled samples (Table 4). With both methods, Paoli meat retained the highest percent of the original moisture; Beehive meat had the second highest WHC. Protecon and Yieldmaster meats exhibited the lowest WHC for both methods, with Protecon the lowest using the chilled meat method, and Yieldmaster meat the lowest using the nonchilled method. Although considerable variation occurred among meats with each method, the only significant (p < 0.05) difference was found using the nonchilled method, in which Paoli meat was higher than Yieldmaster meat.

According to Miller et al. (1968), moisture:protein ratios of skeletal meat (nonpoultry) are inversely related to moisture lost through centrifugation or cookout. The four types of comminuted meat used in this study did not follow this pattern. For example, Paoli meat lost the least moisture in WHC tests but had the highest moisture:protein ratio (4.7). Moisture:protein ratios are presented in Table 3.

Acton (1973) reported that a significant (p < 0.01) negative correlation exists between emulsion stability tests and WHC for extruded and texturized poultry meat. This relationship was also not seen in our study.

Although these results seem contrary to the above discussion and do not follow the pattern of other functional property tests conducted in this study (Fig. 1), they do closely parallel cooking losses observed in unsalted meat patties (Fig. 2). Both of these methods, WHC and cooking loss, involve the measurement of released material upon heating.

**Cooking loss**

Cooking losses for the four meat types, with and without 2% NaCl added, are shown in Fig. 2. The addition of salt was useful in dramatically reducing cooking losses in all meat. This was expected, since salt-soluble proteins are responsible for binding properties in meat (Fukazawa et al., 1961). In a study on the WHC of fresh chicken meat, Shults and Wierbicki (1973) reported that the addition of NaCl dramatically decreased meat shrinkage when heated at 70°C. For example, meat shrinkage when no NaCl was added was approximately 30%; the addition of 2% NaCl (the amount used in the present study) reduced shrinkage to about 10%.

Upon examination of the ratio of losses occurring in unsalted and salted MDP in the present study, it can be seen that not all types of MDP were affected equally when NaCl...
was added. The unsalted:salted ratio for Protecon meat was 3.3 (28.9:8.9, see Fig. 2); Beehive, 3.0; Paoli and Yieldmaster, 2.1. These ratios closely parallel the moisture:protein ratios shown in Table 3.

No significant differences (P>0.05) were found for the various components in the released material of the four meats, with the exception that significantly (P<0.05) more gelatin was obtained in salted Yieldmaster meat than in

sated Protecon meat.

Lyon et al. (1978) evaluated cooking losses of frozen MDP patties, which contained structured vegetable protein, which were cooked in a 177°F oven for 1 hr. Using this system, they obtained total cooking losses of 19-25%, which are lower than losses obtained in unsalted patties in our study. Total cooking losses found in the salted meats, also follow the pattern observed in other functional property tests (Fig. 1).

Shear values for frankfurters and meat patties

The two cells used to evaluate the frankfurters, Warner-Brazet and Volodkowitch, gave similar patterns of results, i.e., frankfurters prepared from Beehive or Protecon MDP were consistently the most firm, and those prepared from Yieldmaster were the least firm (Table 5). When the frankfurters were scored with the Warner-Brazet cell, in which the skin was still intact, no significant differences (P>0.05) were observed. However, using the Volodkowitch cell, in which only the core of the frankfurter was evaluated, frankfurters prepared with Protecon meat were significantly (P<0.05) more firm than those made from Yieldmaster meat.

The frankfurters prepared in this study were generally less firm in comparison to those prepared by Young and Lyon (1973); they reported maximum shear resistance with a Warner-Brazet cell of 593 g/cm², whereas in our study, Beehive frankfurters had 411 g/cm² or 4.03 Newtons of force. The nature of MDP and the fat used in the frankfurters differed in the two studies; Young and Lyon (1973) used skinless backs and necks (low in fat) to prepare the MDP and then added lard to the frankfurter formulation, whereas in our study no supplemental fat was added to the MDP containing only the softer poultry fat.

Patties prepared from Beehive MDP were generally the most firm; however, no significant differences (P>0.05) occurred among any of the measurements for the different patties (see Table 5). The texture of Paoli and Yieldmaster patties was similar for both salted and unsalted MDP. The total energy required to shear these patties with the circle blade cell was always less than that required to shear Protecon and Beehive patties.

Adding 2% NaCl had a toughening effect on the meat, with the exception of Beehive meat, where less maximum force was required to shear the unsalted sample.

Although less total bone was present in Protecon MDP than in two of the other meat types (see Table 3) the bone particles which were present were frequently larger in size. Of the three auger-type machines, Beehive produced a MDP which appeared to have more structure than the more finely comminuted meat from Paoli and Yieldmaster machines; subsequently, the bone particles which were present were also smaller in size.

TBA values

TBA values of MDP and frankfurters are presented in Tables 6 and 7. During the first three months of storage at -20°C, Paoli MDP exhibited the largest increase in TBA numbers (Table 6). However corresponding increases were not observed in frankfurters, prepared from this MDP, when stored at either 4°C or -20°C (Table 7).

It was also observed that Paoli frankfurters were downgraded in flavor by the sensory panel. Although the Paoli MDP used to prepare these frankfurters had a TBA of only 0.5 (Table 6, 0-time), and the frankfurters were evaluated within 1 wk by the sensory panel, the condition which eventually led to the higher TBA values in the MDP may also have contributed to the poorer flavor score in the frankfurters.

The lower TBA values in the frankfurters than in the MDP may be the result of the heat-processing during manufacture, as well as the potential antioxident effect of sodium nitrite and ascorbic acid, which were included in the formulation. Mast and MacNeil (1976) also reported that TBA values of MDP subjected to heat pasteurization were much lower than in nonpasteurized meat.

---Continued on next page

<table>
<thead>
<tr>
<th>Cell type</th>
<th>Protecon</th>
<th>Paoli</th>
<th>Yieldmaster</th>
<th>Beehive</th>
</tr>
</thead>
<tbody>
<tr>
<td>Warner-Brazet Cellα</td>
<td>Max. shear force, N</td>
<td>4.0a</td>
<td>3.8a</td>
<td>3.6a</td>
</tr>
<tr>
<td>Energy for max. force, mJ</td>
<td>14.9a</td>
<td>16.1a</td>
<td>14.8a</td>
<td>16.3a</td>
</tr>
<tr>
<td>Total energy to shear, mJ</td>
<td>40.0a</td>
<td>37.2a</td>
<td>37.5a</td>
<td>38.3a</td>
</tr>
<tr>
<td>Volodkowitch Cellα</td>
<td>Max. shear force, N</td>
<td>3.0a</td>
<td>2.4ab</td>
<td>2.1b</td>
</tr>
<tr>
<td>Energy for max. force, mJ</td>
<td>6.4a</td>
<td>5.6a</td>
<td>4.5a</td>
<td>6.2a</td>
</tr>
<tr>
<td>Total energy to shear, mJ</td>
<td>11.4a</td>
<td>9.4a</td>
<td>7.8b</td>
<td>10.9ab</td>
</tr>
</tbody>
</table>

Meat Patties - Unsalted

| Circle Blade Cellβ | Max. shear force, N | 14.6a | 13.9a | 15.5a | 19.0a |
| Energy for max. force, mJ | 64.0a | 67.9a | 61.7a | 70.5a |
| Total energy to shear, mJ | 133.2a | 117.4a | 117.1a | 152.0a |

Meat Patties - 2% salt added

| Circle Blade Cellβ | Max. shear force, N | 14.9a | 15.1a | 16.0a | 17.9a |
| Energy for max. force, mJ | 78.0a | 65.3a | 67.0a | 74.5a |
| Total energy to shear, mJ | 162.3a | 145.8a | 147.8a | 165.5a |

α Each value represents the mean of three trials, each with 15 determinations. Means in a row with different subscripts are significantly different (P<0.05). N = Newtons; mJ = milliJoules.
β Each value represents the mean of 3 trials, each with 8 determinations. N = Newtons; mJ = milliJoules.
Sensory evaluation

Results of the triangle tests are shown in Table 8. The dominant trend was that panelists consistently detected differences between Paoli's frankfurters and other frankfurters. Similar results were observed in both trials.

The results of paired-comparison tests were also similar for both trials; therefore, scores were combined and are presented in Table 9. The maximum score obtainable is 72, since each frankfurter was compared with three other types by 12 panelists in two trials.

The scores for Paoli and Protecon frankfurters differed significantly (p<0.05) for each attribute. On the other hand, scores of Yieldmaster and Beehive frankfurters were never significantly different (p>0.05). These two frankfurters generally received scores similar to Protecon frankfurters but different from Paoli frankfurters.

When requested to indicate preferences, panelists liked the color of Paoli frankfurters best, but the texture, taste, and overall quality of Paoli frankfurters were scored the lowest. No differences (p>0.05) in preference were present among the other three types of frankfurters.

Results of the scaling test are shown in Table 10. In this test, panelists placed a mark on a line serving as a continuum between two contrasting terms, such as dry and juicy. After panelists marked their scoresheets, distances were measured and recorded; theoretical limits for scores are 0 (extreme dry) and 100 (extreme, juicy). Results of the scaling test were similar to and further confirmed findings of the paired-comparison test.

Most of the attributes dealt with various aspects of frankfurter texture. Protecon frankfurters consistently received highest scores for firmness, coarseness, etc. For the "fine-coarse" scale, scores for all frankfurters were significantly different (p<0.01). Protecon frankfurters had the coarsest texture, followed in order by Yieldmaster, Beehive, and Paoli frankfurters.

Although some similar trends in the texture measurement were observed with sensory scores and shear tests, the differences among frankfurters were more pronounced in panel evaluations than in the objective tests. Panelists also perceived differences which were not always substantiated by objective tests; for example, Paoli frankfurters were scored by panelists as fattier than all others, whereas fat analyses indicated that Beehive frankfurters actually contained slightly more fat.

---Continued on page 1766---

Table 9—Sensory evaluation of frankfurters prepared from four sources of MDP: choices registered in paired-comparison tests

<table>
<thead>
<tr>
<th>Meat source</th>
<th>Attributes</th>
<th>Protecon</th>
<th>Paoli</th>
<th>Yieldmaster</th>
<th>Beehive</th>
</tr>
</thead>
<tbody>
<tr>
<td>Color</td>
<td>redness</td>
<td>22a</td>
<td>56b</td>
<td>40a</td>
<td>26a</td>
</tr>
<tr>
<td></td>
<td>uniformity</td>
<td>14a</td>
<td>63c</td>
<td>31b</td>
<td>36b</td>
</tr>
<tr>
<td></td>
<td>preference</td>
<td>28a</td>
<td>56b</td>
<td>33a</td>
<td>33a</td>
</tr>
<tr>
<td>Texture</td>
<td>firmness</td>
<td>58c</td>
<td>3a</td>
<td>41b</td>
<td>42b</td>
</tr>
<tr>
<td></td>
<td>juiciness</td>
<td>28a</td>
<td>50b</td>
<td>36ab</td>
<td>30a</td>
</tr>
<tr>
<td></td>
<td>fineness</td>
<td>11a</td>
<td>66c</td>
<td>28b</td>
<td>39b</td>
</tr>
<tr>
<td></td>
<td>fattiness</td>
<td>31a</td>
<td>52b</td>
<td>24a</td>
<td>37b</td>
</tr>
<tr>
<td></td>
<td>preference</td>
<td>46b</td>
<td>18a</td>
<td>42b</td>
<td>37b</td>
</tr>
<tr>
<td>Taste</td>
<td>spiciness</td>
<td>48b</td>
<td>15a</td>
<td>43b</td>
<td>38b</td>
</tr>
<tr>
<td></td>
<td>flavor</td>
<td>47b</td>
<td>22a</td>
<td>36b</td>
<td>36b</td>
</tr>
<tr>
<td></td>
<td>preference</td>
<td>44b</td>
<td>20a</td>
<td>39b</td>
<td>41b</td>
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<tr>
<td>General</td>
<td>preference</td>
<td>44b</td>
<td>18a</td>
<td>38b</td>
<td>43b</td>
</tr>
</tbody>
</table>

Table 8—Sensory evaluation of frankfurters prepared from four sources of MDP: correct responses registered in triangle tests

<table>
<thead>
<tr>
<th>Trial</th>
<th>n</th>
<th>Pr x Y</th>
<th>Pr x B</th>
<th>Pr x Pa</th>
<th>Y x B</th>
<th>Y x Pa</th>
<th>B x Pa</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>24</td>
<td>12 n.s.</td>
<td>14 **</td>
<td>24 ***</td>
<td>11 n.s.</td>
<td>19 ***</td>
<td>16 ***</td>
</tr>
<tr>
<td>3</td>
<td>24</td>
<td>8 n.s.</td>
<td>12 n.s.</td>
<td>19 ***</td>
<td>11 n.s.</td>
<td>20 ***</td>
<td>16 ***</td>
</tr>
<tr>
<td>2 + 3</td>
<td>48</td>
<td>20 n.s.</td>
<td>26 ***</td>
<td>43 ***</td>
<td>22n.s.</td>
<td>39 ***</td>
<td>32 ***</td>
</tr>
</tbody>
</table>

2 Pr = Protecon; Y = Yieldmaster; B = Beehive; Pa = Paoli.
3 n.s. = not significant (P > 0.05); ** = P < 0.01; *** = P < 0.001.
EFFECT OF POSTMORTEM MUSCLE CHANGES ON POULTRY MEAT LOAF PROPERTIES

INTRODUCTION

INCREASED MARKETING usage of pound poultry meat products such as loaves, roasts, rolls and similar items has created a need to evaluate the effect of processing techniques on finished product quality. Chicken muscle rapidly undergoes rigor mortis, starting within 3 hr and reaching completion between 12–24 hr (deFremer and Pool, 1960). Many chemical and physicochemical changes occur within this time interval.

Most postmortem changes in nitrogen extractability from breast muscle are due to changes in the myofibrillar proteins (Khan and van den Berg, 1964; Sayre, 1968; Weinberg and Rose, 1960). Myosin extractability rapidly decreases during the first 3 hr postmortem while actomyosin extractability maximizes within 24–36 hr of aging (Sayre, 1968; Khan and van den Berg, 1964). Postrigor tenderness occurs rapidly during the first 24 hr of aging with little tenderness on further aging (Khan and van den Berg, 1964; Koonz et al., 1954).

No significant changes of physicochemical properties have been observed in actomyosin extracted from prerigor and postrigor muscle (Hay et al., 1972), although greater hydration of myofibrillar proteins occurs with an increase of aging time (Sayre, 1968). Froning and Norman (1966) found that light muscle tissue from poultry possesses a significantly larger water-holding capacity than dark tissue.

Perrigor poultry or beef muscle tissue has a significantly higher emulsifying capacity and possesses greater emulsion stabilizing ability than postrigor muscle (Acton and Saffle, 1969). Froning and Neelakantan, 1971). Muscle pH and emulsifying characteristics are highly correlated (Froning and Neelakantan, 1971) and adjustment of meat pH from 5.0 to 8.0 significantly increases the tensile strength of meat loaf slices (Masso et al., 1970a). No study has reported the influence of muscle postmortem state on poultry meat loaf properties.

Vadehra and Baker (1970) stated that the binding mechanism important to fabricated meat products is a heat mediated reaction. Acton (1972) found that binding strength of poultry meat loaves significantly increased as the internal temperature increased from 35°C to 82°C. The loss of water-holding capacity of meat during heating (Hamn and Deatherage, 1960) and its possible role in binding was reviewed by Vadehra and Baker (1970). Schnell et al. (1970) reported that muscle tissue disruption and addition of salt, polyphosphates or ribonucleic acid which reduce cooking loss of poultry loaves also result in an increase of binding strength, irrespective of the chemical or mechanical actions involved.

This study was conducted to determine the effect of postmortem aging of broiler meat on the pH, water-holding capacity and extractability of myofibrillar proteins and to relate these tissue parameters to the cooking loss, binding strength and shearing force of poultry meat loaves. The effect of freezing 24 hr postrigor meat was included as an additional treatment.

EXPERIMENTAL

Source of meat

Broilers of 8-wk and 10-wk age groups were obtained from a Clemson University flock. The broilers were slaughtered, bled and scaled at 55°C for 1 min. For a pre rigor muscle treatment (0 hr postmortem), the pectoralis muscles were immediately excised, coarse ground through an 8 mm plate and regrind through a 5 mm plate with an Osterizer Model 480 Food Grinder. The pre rigor meat was used for all analyses within 30 min after death.

Other treatments of meat included placing the carcasses in an air tank for 3, 6, 12 and 24 hr beforeexcising the pectoralis muscles, grinding and evaluation. An additional treatment involved freezing the pectoralis muscles from 24 hr postmortem carcasses in Cryovac bags for 2 wk at −20°C. Thawing in the bags was conducted at room temperature (22°C) for 10–12 hr followed by grinding. Drip or fluid exudate from the thawed muscles was blended into the ground meat prior to analysis.

Extraction of muscle tissue for nitrogen fractions

The extraction and fractionation procedure was modified from the method of Sayre (1968). 20g of finely ground muscle from each postmortem interval were mixed with 10 vol of phosphate buffer (pH 7.0, ionic strength 1.0) and blended for 45 min at 4°C. The slurry was centrifuged at 30,000 × G for 30 min and the supernatant filtered through Whatman #1 paper. The residue was resuspended in 10 vol of the phosphate buffer. The extraction was repeated and the second supernatant combined with the first. Actomyosin in the supernatant was fractionated at an ionic strength of 0.25 by diluting with 3 vol of distilled water and centrifuging for 30 min at 30,900 × G. Myosin was fractionated from the supernatant by further dilution to attain an ionic strength of 0.04 and centrifuged as described above. Proteins remaining soluble at an ionic strength of 0.04 were designated the sarcoplasmic fraction.

Muscle tissue and extract fractions were analyzed for protein nitrogen by the Kjeldahl method (AOAC, 1970). Total nitrogen in the fractions was corrected for nonprotein nitrogen by preparing and analyzing dichloroacetic acid filtrates. Protein concentrations were expressed as mg protein nitrogen per g wet tissue.

pH and water-holding capacity

The pH of meat samples was measured using tissue extract 10g of meat were placed in 2.8 × 11 cm centrifuge tubes, 22.5 ml of 0.6 M NaCl solution added and the contents stirred for 1 min with a glass rod. After holding for 15 min at 4°C, the meat slurry was again stirred for 1 min and immediately centrifuged at 12,000 × G for 15 min. The supernatant layer was decanted and the volume recorded. The amount of added solution retained by the meat is reported as the water-holding capacity in ml per 100 g meat.

Loaf preparation and evaluation

Loaves of 150–160g of the ground meat containing 1.5% NaCl (w/w) were prepared by blending the meat with the salt for 1 min in a Kitchen Aid 3-C Mixer, weighing and pressing into loaf form. Aluminum pans 11 cm × 8.5 cm × 3.5 cm with aluminum lined board tops were used. The loaves were cooked from an initial internal temperature of 4°C to a final internal temperature of 82–84°C in a 176°C oven. After removal from the oven and cooling to approximately 50°C, one end of the pan lid was opened. The weight of fluid exudate (condensate and juice) drained from the loaf is reported as the percent cooking loss based on the initial meat weight.

Binding strength of loaf slices was determined as previously reported by Acton (1972). A trained panel of nine members evaluated the strength of the loaf using a 9-point hedonic scale (1 = extremely poor; 9 = extremely good). Meat slices approximately 3 cm × 3 cm × 1.2 cm were used for evaluation.

Two samples measuring 7 cm × 3 cm × 1.5
cm were cut from each loaf for shearing force determinations. An Allo-Kramer Shear Press equipped with a 3000 lb ring was used with a 30 sec downstroke at a range of 300. Shearing force was calculated as kg per g meat.

Statistical analysis

Results were subjected to analysis of variance and the significance of means tested by Duncan's method (Steel and Torrie, 1960). Two replications were conducted (with duplicate or triplicate samples) for each parameter with broilers of each age group for an overall four replications within the study. Linear correlation analysis was conducted to test for significant relationships between variables.

RESULTS & DISCUSSION

EXTRACTABILITY OF THE nitrogenc fractions from muscle tissue as a function of postmortem aging time is given in Table 1. Myosin extractability significantly (P < 0.05) decreased from the initial 0-3 hr aging through 12 hr of postmortem aging. The extractable quantity of the actomyosin fraction was initially (0-3 hr) at a low level, 1.5-2.1 mg protein N/g tissue, but significantly increased between 3 hr and 12 hr of aging to 6.6 mg protein N/g tissue. Myosin and actomyosin extractability remained constant from 12 hr to 24 hr postmortem. The sarcoplasmic protein fraction did not significantly change in extractable quantity during the 24 hr aging period or in the frozen 24 hr tissue sample. These data are consistent with the findings reported by Sayre (1968) and Weinberg and Rose (1960). No significant changes were observed for nonprotein nitrogen content (Table 1) during the aging period. Wierbicki et al. (1956) also reported no increase of non-protein nitrogen in bovine muscle during an aging period of several days.

The myosin fraction from 24 hr tissue frozen for 2 wk at -20°C was considerably lower than the 24 hr nonfrozen tissue (Table 1). However, there was a significant (P < 0.01) increase in the quantity of extractable actomyosin from the frozen tissue. Actomyosin nitrogen increased approximately 83% to 12.6 mg N/g tissue whereas the myosin content decreased approximately 86% to 1.4 mg N/g tissue. It is evident that the freezing and thawing of the muscle tissue promoted a greater ease of extraction of actomyosin while extraction of the remaining myosin decreased. The formation of high molecular weight aggregates which can be centrifuged out of solution at low centrifugal fields has been reported by Burikut (1970) to occur on freezing of rabbit and trout myosins.

During the first 6 hr postmortem, muscle pH significantly dropped from pH 6.3 to pH 5.9 (Table 1). Little change in pH occurred during the remainder of the aging period. A decreasing pH during the time course of rigor mortis has been used previously as a criterion for distinguishing the prerigor state from the postrigor state (Bate-Smith and Bendall, 1956; Aston and Saffle, 1969; Froning and Neelakantan, 1971).

The water-holding capacity of the ground meat significantly (P < 0.05) increased at each interval of aging from 3 hr to 24 hr postmortem (Table 1). There appeared to be no direct correlation between muscle pH and the water-holding capacity in this study (r = 0.44). Hydration accompanied the formation of actomyosin as noted by Payre (1968). Hamm (1960) stated that the minimum water-holding capacity coincides with the maximum of muscle rigidity. The water-holding capacity and pH values of the frozen 24 hr aged muscle tissue were not significantly (P < 0.05) different from those found for the 24 hr unfrozen muscle.

The percent cooking loss, tissue binding strength and shear force for loaves prepared with meat during postmortem aging are given in Table 2. No significant (P < 0.05) changes in cooking loss or binding strength were observed when loaves were prepared with tissue excised at any interval during the 24 hr aging period. However, when loaves were prepared with the 24 hr frozen tissue, there was a significant decrease of cooking loss when compared to the cooking loss of loaves of meat from the 0 to 6 hr periods. Binding strength of the loaves of the 24 hr frozen tissue was also significantly higher when compared to some of the earlier intervals. Maeso et al. (1970b) found no significant difference in average tensile strength of fresh loaves and once frozen loaves, but they reported a significant decrease in tensile strength in loaves frozen and thawed twice versus fresh and once frozen. The effect of freezing as a physical and physicochemical treatment warrants further study due to the current marketing of poultry loaves in the frozen uncooked and precooked state.

There was a significant (P < 0.05) correlation between water-holding capacity of the meat tissue and the cooking loss observed from the loaves (r = -0.71). Schnell et al. (1970) reported a significant reduction in the amount of coagulant when NaCl, polyphosphate or ribonuclease acid were used in loaf preparation. The interaction of NaCl with the soluble muscle proteins is known to increase water-holding capacity (Hamm, 1960). No significant correlation was observed between

### Table 1—Extractable nitrogen fractions, pH and water-holding capacity of muscle tissue as a function of time postmortem

<table>
<thead>
<tr>
<th>Hours postmortem</th>
<th>Nitrogen fraction (mg N/g muscle)</th>
<th>Water-holding capacity</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Myosin</td>
<td>Actomyosin</td>
</tr>
<tr>
<td>0h</td>
<td>9.33a</td>
<td>2.07ab</td>
</tr>
<tr>
<td>3</td>
<td>10.80a</td>
<td>1.50a</td>
</tr>
<tr>
<td>6</td>
<td>4.42b</td>
<td>4.38be</td>
</tr>
<tr>
<td>12</td>
<td>2.08c</td>
<td>6.6de</td>
</tr>
<tr>
<td>24</td>
<td>2.61ce</td>
<td>6.87d</td>
</tr>
<tr>
<td>24°F</td>
<td>1.40e</td>
<td>12.60e</td>
</tr>
</tbody>
</table>

* Any two means within a column having the same or one of the same letters are not significantly different at P < 0.05.

### Table 2—Percent cooking loss, tissue binding strength and shear force values for poultry loaves prepared with meat to 0 to 24 hr postmortem

<table>
<thead>
<tr>
<th>Hours postmortem</th>
<th>% Cooking loss</th>
<th>Binding strength</th>
<th>Shear force, kg/g</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>8 wk</td>
<td>10 wk (Mean)</td>
<td></td>
</tr>
<tr>
<td>0h</td>
<td>14.2a</td>
<td>7.5ab</td>
<td>2.58a</td>
</tr>
<tr>
<td>3</td>
<td>15.2a</td>
<td>7.4ab</td>
<td>2.18b</td>
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<td>6</td>
<td>15.3a</td>
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</tr>
<tr>
<td>12</td>
<td>13.5ab</td>
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<td>1.79e</td>
</tr>
<tr>
<td>24</td>
<td>11.8ab</td>
<td>7.6ab</td>
<td>1.63e</td>
</tr>
<tr>
<td>24°F</td>
<td>9.7b</td>
<td>8.1b</td>
<td>1.78e</td>
</tr>
</tbody>
</table>

* Any two means within a column having the same or one of the same letters are not significantly different at P < 0.05.

* Tissue excised and processing began within 30 min postmortem.

* Tissue excised at 24 hr postmortem, frozen at -20°C for 2 wk, thawed and further processed.
water-holding capacity and binding strength, \( r = 0.34 \). However, there was a significant relationship (\( P < 0.01 \)) between binding strength and cooking loss (\( r = 0.67 \)) which is in agreement with the reports of Schnell et al. (1970) and Vadehra and Baker (1970).

No significant broiler age effect (8 wk versus 10 wk) was observed for any response except shear force values for meat loaf slices. In general, muscle tissue of the 10 wk old group of broilers produced loaves of higher shear resistance when compared to loaves from meat of the 8 wk old broilers. The overall means of shear resistance compared by time postmortem (Table 2) shows that loaves prepared with prestigor tissue, 0–3 hr postmortem are significantly (\( P < 0.05 \)) less tender than loaves prepared with prestigor tissue, 6–24 hr postmortem. Loaves from frozen 24 hr meat gave shear responses similar to loaves of the poststrog intervals.

Although prestigor meat of broilers and that of beef and pork, in comparison to poststrog meat, has been shown to possess significantly better emulsification properties for comminuted meat products (Froning and Neelakantan, 1971; Acton and Saffle, 1969; Trautman, 1964), no significant industrial advantage appears to exist for the use of prestigor meat in poultry loaf production. The rapid onset of rigor in poultry muscle is the most obvious disadvantage to normal processing procedures involved in the usage of pre-

**REFERENCES**


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INTRODUCTION
THE EFFECTS of different methods of chilling poultry carcasses on microbiological quality, particularly the total bacterial count on the freshly processed poultry carcass, as well as the incidence and numbers of certain microorganisms of public health significance have been studied. Brewer et al. (1961), Kotula et al. (1962), Farrell and Barnes (1964), Skerkiewicz et al. (1969), Keel and Farmlee (1968), Knoop et al. (1971), Veerkamp et al. (1972), Simonsen (1973) and Brant (1973) found that total bacterial counts on poultry carcasses were reduced during continuous immersion chilling. Thomson et al. (1965, 1966) reported no significant difference in the total bacterial count among poultry carcasses sampled at five locations in commercial immersion chillers. However, Clark and Lentz (1969), Lillard (1971) and Peric et al. (1971) reported an increase in total bacterial counts of carcasses during continuous immersion chilling.

Excessive numbers of Enterobacteriaceae may indicate greater food poisoning potential. Builing and Pietzsch (1966) and Tamura et al. (1971) stated that the occurrence of Salmonella and enteropathogenic Escherichia coli in broilers is less likely if the Enterobacteriaceae count is low.

The objectives of this study were (a) to determine the changes in total plate count and the numbers and types of Enterobacteriaceae on carcasses chilled by continuous immersion or by a combination of immersion and airblast chilling and (b) to determine numbers and types of Enterobacteriaceae on eviscerated carcasses during refrigerated storage.

EXPERIMENTAL
Locations of sampling
In-plant. Ten broiler carcasses were sampled at each of three locations in a commercial processing plant. Carcasses were sampled (1) before chilling, (2) after about 25 min in-line slush-ice immersion chilling and (3) after a combination of slush-ice followed by 45 min at −7°C airblast chilling. Sampling was by swabbing a 12.3 cm² area of breast skin for 30 sec with a calcium alginate swab. Also water from the chilling well was sampled for bacteriological analysis.

Storage. Ten freshly processed broiler carcasses were obtained from a local processing plant immediately after slush-ice immersion chilling. A 12.3 cm² area of skin on the left breast was swabbed and the Enterobacteriaceae count was determined. This was the storage day 0 sample. Each of the 10 birds was placed in a polyethylene bag and stored at 4°C until spoilage occurred (10 days). A 12.3 cm² area of skin on the right breast was swabbed to determine storage day 10 Enterobacteriaceae count. A preliminary study in our laboratory indicated that there was no significant difference in numbers or types of Enterobacteriaceae between the left and right breast of poultry carcasses. Enterobacteriaceae cultures isolated from carcasses stored at 4°C for 10 days were tested for their ability to grow on brain heart infusion agar (Difco) at −2°C, 1°C, 4°C, 25°C and 35°C incubation for up to 21 days.

Microbiological methods
Total aerobic plate count with standard methods agar (BBL) was made for in-plant studies. Plates were incubated at 20°C for 72 hr.

To estimate the Enterobacteriaceae count, violet red bile agar (Difco) with 1% glucose was used (Mossel et al., 1962). The double-poured plates were incubated for 18–24 hr at 35°C. The counts were reported as logarithmic averages and expressed as microorganisms per cm² or per ml.

The experiment was replicated three times and for replications two and three of the in-plant study, and replication three of the storage study, Enterobacteriaceae were isolated from the highest dilution plated during microbiological analysis and identified to determine which genera of the Enterobacteriaceae predominated at each of the various chilling stages.

RESULTS & DISCUSSION
In-plant study
Total plate count (TPC) and Enterobacteriaceae count (ENT) of carcasses sampled before chilling and after immersion and dry chilling declined significantly after chilling regardless of whether the birds were slush-ice chilled only or slush-ice plus airblast chilled (Table 1). The average Enterobacteriaceae count after

Table 1—Total plate counts (TPC) and Enterobacteriaceae (ENT) counts of broiler carcass breast skin and chiller water during commercial chilling

<table>
<thead>
<tr>
<th>Sampling location</th>
<th>TPC</th>
<th>ENT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Carcass before chill</td>
<td>3.17a</td>
<td>2.27a</td>
</tr>
<tr>
<td>Carcass after slush-ice chill</td>
<td>2.57b</td>
<td>1.48b</td>
</tr>
<tr>
<td>Carcass after combined slush-ice and air blast chill</td>
<td>2.64b</td>
<td>1.02b</td>
</tr>
<tr>
<td>Slush-ice chiller water</td>
<td>3.26</td>
<td>2.87</td>
</tr>
</tbody>
</table>

*Carcass counts expressed as logarithms of number per cm²; water counts per ml. Each value is the average of 30 samples. Means within a column followed by the same letter are not significantly different (P = 0.05).
Table 2—Genera of Enterobacteriaceae isolated from broiler carcass breast skin and chiller water during commercial chilling (average percentages—replications two and three only)

<table>
<thead>
<tr>
<th>Locations sampled</th>
<th>Genus</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Carcass before chill</td>
<td>Escherichia</td>
<td>90.7a</td>
</tr>
<tr>
<td></td>
<td>Enterobacter</td>
<td>5.8</td>
</tr>
<tr>
<td></td>
<td>Klebsiella</td>
<td>1.2</td>
</tr>
<tr>
<td></td>
<td>Unclassified</td>
<td>2.3</td>
</tr>
<tr>
<td>Carcass after slush-ice chill</td>
<td>Escherichia</td>
<td>96.7</td>
</tr>
<tr>
<td></td>
<td>Enterobacter</td>
<td>3.3</td>
</tr>
<tr>
<td>Carcass after slush-ice and air blast chill</td>
<td>Escherichia</td>
<td>87.2</td>
</tr>
<tr>
<td></td>
<td>Enterobacter</td>
<td>9.5</td>
</tr>
<tr>
<td></td>
<td>Providencia</td>
<td>1.2</td>
</tr>
<tr>
<td></td>
<td>Unclassified</td>
<td>2.1</td>
</tr>
<tr>
<td>Chiller Water</td>
<td>Escherichia</td>
<td>96.4</td>
</tr>
<tr>
<td></td>
<td>Enterobacter</td>
<td>2.4</td>
</tr>
<tr>
<td></td>
<td>Klebsiella</td>
<td>1.2</td>
</tr>
</tbody>
</table>

a The number of isolates for each location sampled ranged from 81–94.

Enterobacteriae on Poultry—45

slush-ice chilling was log 1.48/cm². This is in agreement with results reported by Mulder and Veerkamp (1973) who found that the Enterobacteriaceae count was reduced by one log after immersion chilling, when the immersion chilling was preceded by spray washing. A spray washer also preceded the in-line slush-ice chiller at the plant in which our study was made.

The Enterobacteriaceae in the chiller water and on the poultry carcass at the various stages of chilling (Table 2) were mainly of the genus Escherichia (87.2–96.7%) of the isolates. Other genera found less frequently were Enterobacter, Klebsiella and Providencia. These findings agree with those of Berner et al. (1969) who also reported that Escherichia was the predominant genus of the Enterobacteriaceae isolated from carcasses immediately after chilling. The distribution of these species in the chill water and on carcasses at the various chilling stages were similar.

Salmoneia, Shigella, Proteus and other genera of the Enterobacteriaceae group were not found. This does not indicate that they were not present either on the birds sampled or in the chiller water. If present in low numbers, it is unlikely that they would have been isolated from the higher dilutions. Secondly, their absence from the 12.3 cm² area of breast skin sampled does not preclude their presence elsewhere on the carcass.

Storage study

The number of Enterobacteriaceae on poultry carcasses stored for 10 days at 4°C increased significantly (Table 3). Carcasses were spoiled by this time and the Enterobacteriaceae count had increased by a factor of $10^2$–$10^3$. To determine if the predominant genus or genera had changed during storage, the types of organisms isolated and identified from the samples on storage day 0 were compared with those from storage day 10 (Table 4). On storage day 0, Escherichia predominated, but after 10 days of storage, Enterobacter had become the predominant genus. These results agree with Berner et al. (1969) who reported that of the Enterobacteriaceae, the Klebsiella-Enterobacter-Serratia group predominated on spoiled poultry. All 98 Enterobacteriaceae cultures isolated from carcasses stored 10 days were identified as either Escherichia or Enterobacter, except one, Klebsiella. After incubation of the isolated cultures for 7 days at 1°C, none of the 14 Escherichia cultures grew, while 13 of the 83 Enterobacter cultures grew. After 21 days of incubation at 1°C, two Escherichia cultures had yet failed to show growth, while all 83 Enterobacter showed growth. At 4° and 2°C incubation, Enterobacter showed growth more frequently than Escherichia. All isolates were able to grow at 25° and 35°C incubation.

Our results indicate that immersion slush-ice continuous chilling, either alone or in combination with air blast chilling, results in a significant reduction of both the total and Enterobacteriaceae count of broiler carcasses. The proportions of the different Enterobacteriaceae genera on carcasses chilled by the two procedures were similar. Escherichia was the predominant genus in both instances. Thus, air chilling to supplement immersion chilling of broiler carcasses appears to have little microbiological significance. Leistner et al. (1972) reported that air chilling reduces Enterobacteriaceae counts between a water spray chilling system and a combination water spray and air blast system.

Enterobacter, which constituted only a small percentage of the Enterobacteriaceae on freshly processed carcasses, was the predominant genus of this family at the time of spoilage. Enterobacter isolates from the spoiled carcasses were shown to be more psychrophilic than Escherichia. Schultz and Olson (1960) reported that, in dairy products, psychrophilic strains of coliforms were more commonly found in the genus Aerobacter (Enterobacter) than in the genus Escherichia.

Additional information regarding the source, incidence and types of Enterobacteriaceae on poultry carcasses is needed to assess the significance of this group in poultry processing.

REFERENCES


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The authors are indebted to Mr. R.L. Richardson, Ms. F. Gabie and Mr. R.H. Dean for their technical assistance.

Mention of specific brand names does not imply endorsement by the authors or institutions at which they are employed to the exclusion of others not mentioned.
Esters Present in Pre-ferments.
The only esters detected in the Fleischmann pre-ferment were those of acetic acid. The ethyl ester was presumably produced because of the high concentration of ethyl alcohol in the fermentation mixture. Wiseblatt (26) found only ethyl esters in bread crumb.
The change in ethyl acetate concentration in the pre-ferment with time is shown in Figure 2. The ester reached its maximum concentration after 6 to 8 hours of fermentation and decreased to zero after 48 hours. The concentration of ethyl acetate in American Dry Milk Institute pre-ferments was not determined because it was necessary to adjust the pH of that pre-ferment to 10.0 to precipitate a part of the nonfat dry milk. The ester was hydrolyzed at this pH.

Acknowledgment
Financial assistance from the Corn Products Sales Co. is gratefully acknowledged.

Literature Cited
(1) Baker, J. C., Food Engineering 25, 60, 183 (1933).
(3) Baker, J. C., Mize, M. D., Ibid., 18, 19–34 (1941).
(12) Food Processing 16 (10), 23–5, 32 (1953).

WATER CONTENT OF MEATS

Determination of Water-Holding Capacity of Fresh Meats

The water-holding capacity of nondisintegrated muscles and ground and comminuted fresh lean meats was determined on fresh and heated meat. By pressing a 400- to 600-mg. fresh muscle sample on No. 1 Whatman filter paper of constant humidity in a specially made press operating under 500 p.s.i., the area of the paper wetted in 1 minute by the expressed juice is directly proportional to the weight of water in the press juice. The method gives the reproducible results within 2 to 3%. The amount of free water in beef, pork, veal, and lamb varies from 30 to 50% of the total moisture content, depending on the kind of meat and period of aging.

 Recent studies on consumer qualities of meats, such as tenderness, texture, drip on freezing and thawing, and shrinkage on cooking indicate that these qualities depend on the degree of hydration of muscle proteins (1, 17, 36–40). The highly polar water molecules are attracted to the muscle proteins by ionizable basic and acidic groups as in arginine, histidine, lysine, glutamic acid, and aspartic acid or by polar nonionic groups such as in cystine, cysteine, serine, methionine, threonine, tyrosine, and tryptophan. The mechanism of the protein hydration is not well understood. Some pioneering work on hydration of various proteins, other than muscle proteins, and polypeptides has been done by Bull (2), Pauling (33), Mellon and Hoover (29), and others (4, 5, 26).

Lean meat contains about 3.5 grams
of water per gram of protein, about 10 times as much as the water of hydration of commonly known proteins (26). Consequently, muscle like other biological material contains water of hydration, or electrostatically bound water, and physically absorbed water, bound on the proteins by the secondary forces, such as water dipole-dipole induction, hydrogen bonds, and capillary and surface attractions.

In this paper, free water is considered to be that portion of the total moisture which has been released by pressing or heating the meat under specified laboratory conditions which are presented here. The remaining portion of the total mixture of meat is the bound water, consisting of the water of hydration and that portion of the physically absorbed water which has not been so released. The exact amount of bound or free water cannot be determined in meat, for it contains different protein components and the water of hydration of each is not known. Furthermore, the amount of the physically absorbed water is changed by various laboratory and processing techniques. However, by considering the various muscle proteins as a single protein component and by using the same method under the same experimental conditions, relative changes in the water-holding properties of meat can be measured.

A simple centrifugal method for measuring the water-holding capacity during heating, freezing, and thawing of meat has been recently published by Wierbicki, Kunkle, and Deatherage (40). The method is very useful for measuring relative shrinkage of meat under different experimental conditions (38). However, when this is applied to the meat samples heated below 100°F, or not heated at all (fresh meats), the amount of juice, if any, collected in the centrifuge tubes was within the range of the experimental error (0.1 to 0.3 ml).

In 1953 Grau and Hamm, at the Bundesforschungsanstalt für Fleischwirtschaft in Kulmbach, reported a simple filter paper method for the determination of water-holding capacity of fresh meats (12). Since that time, while using this method, Grau and Hamm and their associates have published several papers dealing with the theory of protein of meat (27, 28, 29) and the effects of pH (17, 18, 20), adenosine triphosphate (ATP) (16, 20), meat aging (10, 20), sodium chloride (7, 9, 12, 17, 19), calcium, magnesium, zinc, and potassium ions (15, 18, 19, 21, 22), various phosphates (5, 13, 15, 17, 18, 25), and various organic and inorganic anions (22) on the hydration of meat proteins. The usefulness of this method for meat research has been reported also from Finland (30-32, 34), Poland (23, 25), and Hungary (27, 28), but it has not been used in the study of cooked meats.

This paper presents a modification of the Grau and Hamm's original method for the determination of the water-holding capacity of fresh meats. The pressing by hand has been replaced by a pressing device which controls the pressure and assures a greater accuracy of the determination. Inasmuch as the centrifugal method (40) is useful in studying heated, frozen, and defrosted meats, and the German method (12) is particularly adapted to fresh meats, the two methods have been applied to the same meat. The limitations of each method appear to be complemented by the usefulness of the other method.

**Experimental**

**Apparatus.** This method of Grau and Hamm involves pressing a freshly cut 400- to 600-mg sample of muscle or well-minced lean meat onto filter paper under constant pressure and for a fixed time and measuring the area occupied by the water, which diffused from the meat sample onto the filter paper. The amount of the free or "loose" (12) water expressed is then calculated from the area by using an appropriate conversion factor. In the original version of the method (12), two Plexiglas plates screwed together as firmly as possible by hand were used. The pressure thus developed is about 60 to 70 kg per sq. cm. (853 to 996 p.s.i.) and the variation in pressure, 30 to 70 kg per sq. cm. (427 to 996 p.s.i.), has no effect on the wetted area. However, Figure 2 indicates that this is not strictly so. Pohja and Numi-vaara (34) in their modification of the method (pressing by the balance weights) showed also that the area varies with the pressure, particularly at the acid side of the meat, pH 5.0.

An apparatus has been built for pressing the meat samples under constant pressure. The apparatus, shown by Figure 1, consists of a 5-ton capacity hydraulic jack (Model CG-9, Blackhawk Manufacturing Co., Milwaukee, Wis.), which is built into the 1/4-inch steel frame with the side walls 15 inches high and 8 inches wide, welded on the bottom and the top with 8 x 8 inch steel plates; a movable 8 x 8 inch steel plate is located inside the frame, just over the hydraulic jack, which can be pressed to the top plate of the frame during the operation. A pressure gage reading from 0 to 1000 p.s.i. with increments of 10 p.s.i. is tapped to the base of the jack. Two 8 x 8 x 1/4 inch Plexiglas

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**Figure 1.** Hydraulic meat press with pressure gage

**Figure 2.** Effect of increasing pressure on free water area

X Semimembranosus pork muscle
× Semimembranosus beef muscle

---
plates are placed between the movable steel plate and the top plate of the frame; these are secured from the steel plates by rubber cushions to protect the Flexiglas plates and assure an equal distribution of the pressure through the plates.

A 400- to 600-mg meat sample is weighed on a 9-cm No. 1 Whatman filter paper of constant moisture content, 10.18 ± 0.10%, obtained by holding the filter paper in a desiccator over saturated potassium chloride solution as suggested by Grau and Hann (17, 12). The filter paper and meat are then placed between the Flexiglas plates and pressed immediately at a constant pressure for a fixed period of time. A pressure of 500 p.s.i. and a pressing time of 1 minute were most suitable. By pressing, the muscle material is squeezed to an almost circular film (meat film), while the expelled water is absorbed by the filter paper forming a circular brown-red color area (free moisture area).

Immediately after the pressing has been accomplished, the Flexiglas plates with the meat sample are removed and the meat film area is marked with a colored pencil on the other side of the filter paper before removing the Flexiglas plate from the meat side of the paper, as the meat film usually adheres to the plate. The filter paper is then removed from the upper plate and the meat film taken off. The filter paper can now be stored for the surface measurement, if necessary, for a long period of time.

For the calculation, the surface of the free moisture area (juice ring) around the pressed muscle is determined by subtraction of the surface of the meat film from the entire surface. For the surface measurement, an Ott compensating planimeter with vernier range of 0.01 square inch (Type 16, Charles Brucning Co.) was satisfactory.

The centrifugal method was used as previously reported (40).

Pressure. Samples, 500 mg, of lean beef semimembranosus and pork longissimus dorsi, 2 days post-mortem, were accurately weighed on an analytical balance and then pressed for 1 minute at varying pressures. Free moisture area was calculated and plotted as a function of the pressure from 100 to 1000 p.s.i.

The results (Figure 2) indicate that the free moisture area varies with the pressure, usually the amount of the pressed-out juice increased with the increased pressure. However, the increase of the free moisture area is not directly related to the magnitude of the pressure, but rather a steepwise increase of the pressed-out juice occurred within the pressure range investigated. Presumably, the water present in meat is bound or fixed by the muscle proteins with different forces forming several water layers around the protein molecules, which are held by different water-binding energies, each water layer requiring a different pressure for being released by the protein molecules.

The data given in Figure 2 do not represent the characteristic pattern for the water binding of all pork and/or beef muscles. The absolute amounts of juice were found to vary from muscle to muscle, however, all the samples investigated showed more or less definite stepwise binding of the water by the muscle substance. At 500 p.s.i. pressure a plateau always resulted and, therefore, this pressure was selected as the constant pressure for the determination of the water-binding capacities of fresh meats.

Pressing Time. Table I shows the effect of the pressing time on the meat film and the free moisture area from pork semimembranous muscle. The free moisture area increases with the pressing time, up until about 4 minutes. However, as the spreading of the meat film reached its maximum after 1 minute of pressing, pressing time was standardized at 1 minute.

Pressing Delay. During the preliminary experimentation, the first sample out of the four samples weighed for the simultaneous pressing gave a somewhat smaller free moisture area. Evaporation appeared to be responsible for this error. Consequently, the effect of the time of the exposure of the samples to the air was studied with the results presented in Table II.

Air exposure for longer than 5 minutes causes significant evaporation of moisture from the sample and probably from the filter paper. A change in temperature of the samples is another factor affecting the protein-water relationship, as the same beef samples taken from the refrigerator (70 to 105 C) gave somewhat smaller free moisture areas than after bringing them to room temperature (25 C) before pressing. Therefore, in the course of further experimentation, meat samples taken from a cooler were placed first in a 70 to 25 C. refrigerator for a few hours, and then promptly

| Table I. Effect of Pressing Time on Free Moisture Area* |
|-------------------------|-------------------------|-------------------------|-------------------------|
| Time, Min. | Total | Meat film | Free moisture |
| 0.5 | 4.38 | 1.18 | 3.20 |
| 1 | 4.50 | 1.26 | 3.24 |
| 2 | 4.85 | 1.28 | 3.58 |
| 3 | 5.24 | 1.31 | 3.93 |
| 4 | 5.38 | 1.30 | 4.08 |
| 5 | 5.40 | 1.28 | 4.12 |
| 6 | 5.38 | 1.27 | 4.11 |

* 500-mg. samples of pork (semimembranosus) at 500 p.s.i. pressure.

| Table II. Effect of Pressing Delay on Free Moisture Area |
|-------------------------|-------------------------|-------------------------|-------------------------|
| Min. between Weighing and Pressing | Total | Meat film | Free moisture |
| Preparing | Area, Square Inches | 10 | 3.32 | 1.40 | 1.92 |
| 7.5 | 3.59 | 1.65 | 1.94 |
| 5 | 3.81 | 1.79 | 2.11 |
| 2.5 | 3.95 | 1.85 | 2.10 |

* 500-mg. samples at 500 p.s.i. pressure.

| Table III. Reproducibility of Duplicate Determinations |
|-------------------------|-------------------------|-------------------------|-------------------------|
| Pair No. | Sample Wt., Mg. | Area, Square Inches | Free H2O Area per 500 Mg. | Relative Error, % |
| Bees* | 1 | 535 | 4.50 | 3.22 | 2.28 | 2.13 | 2.665 | 3.1 |
| 495 | 4.51 | 2.53 | 1.98 | 2.00 |
| 2 | 534 | 4.49 | 2.40 | 2.09 | 1.96 | 2.075 | 5.5 |
| 533 | 4.78 | 2.45 | 2.33 | 2.19 |
| 3 | 514 | 4.73 | 2.55 | 2.18 | 2.12 | 2.065 | 1.7 |
| 492 | 4.79 | 2.77 | 2.02 | 2.05 |
| Pork* | 1 | 467 | 3.74 | 1.26 | 2.48 | 2.68 | 2.725 | 1.3 |
| 464 | 4.03 | 1.49 | 2.54 | 2.77 |
| 2 | 513 | 4.30 | 1.14 | 3.16 | 3.07 | 3.010 | 2.0 |
| 472 | 4.10 | 1.32 | 2.78 | 2.95 |
| 3 | 501 | 4.63 | 1.21 | 3.42 | 3.41 | 3.343 | 1.6 |
| 584 | 5.05 | 1.23 | 3.82 | 3.28 |

* Same beef sample used for all three pairs. Pressure, 500 p.s.i. per min.

Each pair represents another hog. Pressure, 500 p.s.i. per min.
weighed (within the time interval, not longer than 5 minutes), and pressed.

Filter papers were weighed in advance and covered with dry beakers; the samples weighed first were also covered before weighing the remaining samples. All samples were taken from freshly cut meat. With filter papers weighed in advance and the apparatus made ready for pressing, four samples can be easily weighed and pressed by an experienced operator within 5 minutes. By doing so, the reproducibility of the method was maintained within the desired limits.

The area of the Plexiglas plates is great enough to press four 9-cm., No. 1 Whatman filter papers at a time. However, the apparatus was made to hold five Plexiglas plates, or 16 samples of meat, if all 16 samples can be weighed within the time interval of 5 minutes, say by three or four operators simultaneously.

Reproducibility of the Method. After the operating conditions were standardized at 500 p.s.i. pressure with a pressing time of 1 minute, and weighing time not longer than 5 minutes, the reproducibility of the method was checked on different kinds of meat and different muscles of the same kind of meat. The reproducibility of the method was within ±3%. For the series of analyses, the number of samples taken from the same muscle was reduced to two. Table III represents the accuracy of the duplicate determinations. The sample size of 400 to 600 mg. was required to get this accuracy. Smaller samples (200 to 300 mg.) gave usually a relatively greater free moisture area than the larger ones (700 to 800 mg.). The accuracy of the determination requires also that the juice from does not reach the edge of the filter paper, as found originally by Grau and Hamm (77, 72).

Loss of Free Moisture under Meat Film. For the calculation of the free moisture only the free moisture area is considered. Yet, the filter paper under the meat film may absorb some of the free moisture. Grau and Hamm (77-79) state that the amount under the meat film is negligible, because of a greater pressure exercised on the meat sample than on the filter paper outside the meat film. Meat samples, 500 mg., were pressed on the filter paper before and after waxing both sides of the filter paper area occupied by the resulting meat film. Paraffin wax was used. The results given in Table IV indicate that the total moisture area increased by 1.4 to 3.4% as the result of the waxing of the meat film area of the filter paper.

This increase is within the experimental error of the method (Table III) and confirms the statement of Grau and Hamm.

Standard Curve. Free Moisture Area vs. Free Moisture. For the calculation of the free and bound water in a meat sample, the conversion factor of the free moisture area in the amount of free moisture must be known. For this purpose, a sample of chilled beef semimembranous muscle 2 days post-mortem was ground. Several 20-gram samples of the meat were placed in the centrifuge tubes used for shrinkage determination (49), warmed at 27°C, for 30 minutes, and then centrifuged. The juice which collected at the bottom of the centrifuge tubes was removed, filtered through a coarse filter to remove suspended solid particles, and the clear juice obtained in this way was stored for a few hours in a

---

**Table IV. Effect of Waxing Meat Film Area on Total Area**

<table>
<thead>
<tr>
<th>Total Area, Square Inches</th>
<th>Effect of Waxing</th>
<th>Effect of Waxing</th>
<th>Effect of Waxing</th>
</tr>
</thead>
<tbody>
<tr>
<td>Before Waxing</td>
<td>After Waxing</td>
<td>+ Rel. %</td>
<td>Before Waxing</td>
</tr>
<tr>
<td>Round</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>4.43</td>
<td>4.67</td>
<td>5.4</td>
</tr>
<tr>
<td>2</td>
<td>4.37</td>
<td>4.44</td>
<td>1.6</td>
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<tr>
<td>3</td>
<td>4.41</td>
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<tr>
<td>4</td>
<td>4.38</td>
<td>4.56</td>
<td>4.1</td>
</tr>
<tr>
<td>5</td>
<td>4.40</td>
<td>4.55</td>
<td>2.5</td>
</tr>
<tr>
<td>Flank</td>
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<td></td>
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</tr>
<tr>
<td>1</td>
<td>3.62</td>
<td>3.77</td>
<td>4.1</td>
</tr>
<tr>
<td>2</td>
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<td>3.64</td>
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<tr>
<td>4</td>
<td>3.77</td>
<td>3.83</td>
<td>1.6</td>
</tr>
<tr>
<td>Av.</td>
<td>3.76%</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* Standard Parawax household wax.
* 500-mg. samples at 500 p.s.i. per min. pressure.

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**Table V. Standard Curve Free Moisture Area vs. Free Moisture**

<table>
<thead>
<tr>
<th>Juice, Fresh Beef</th>
<th>Juice, Beef after Freezing and Thawing</th>
<th>Juice, Fresh Pork</th>
<th>Distilled Water</th>
</tr>
</thead>
<tbody>
<tr>
<td>H₂O, Mg.</td>
<td>Area</td>
<td>H₂O, Mg.</td>
<td>Area</td>
</tr>
<tr>
<td>90</td>
<td>1.60</td>
<td>42</td>
<td>0.70</td>
</tr>
<tr>
<td>92</td>
<td>1.68</td>
<td>65</td>
<td>1.16</td>
</tr>
<tr>
<td>125</td>
<td>2.15</td>
<td>111</td>
<td>1.84</td>
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<tr>
<td>153</td>
<td>2.59</td>
<td>158</td>
<td>2.49</td>
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<td>187</td>
<td>3.08</td>
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<td>2.99</td>
</tr>
<tr>
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<td>3.30</td>
<td>211</td>
<td>3.10</td>
</tr>
<tr>
<td>211</td>
<td>3.40</td>
<td>283</td>
<td>4.02</td>
</tr>
<tr>
<td>221</td>
<td>3.70</td>
<td>317</td>
<td>5.31</td>
</tr>
<tr>
<td>258</td>
<td>4.53</td>
<td>399</td>
<td>6.10</td>
</tr>
<tr>
<td>265</td>
<td>4.60</td>
<td>399</td>
<td>6.49</td>
</tr>
<tr>
<td>298</td>
<td>5.02</td>
<td>192</td>
<td>3.11</td>
</tr>
<tr>
<td>357</td>
<td>5.61</td>
<td>245</td>
<td>4.06</td>
</tr>
<tr>
<td>362</td>
<td>5.78</td>
<td>261</td>
<td>4.56</td>
</tr>
<tr>
<td>385</td>
<td>6.38</td>
<td>270</td>
<td>4.42</td>
</tr>
<tr>
<td>389</td>
<td>6.45</td>
<td>261</td>
<td>4.56</td>
</tr>
</tbody>
</table>

* Pressure, 500 p.s.i.
* Mg. juice × 0.883 (moisture of the juice = 88.3%).
* Mg. juice × 0.890.
* Mg. juice × 0.892.
* Regression coefficient, k: X = kY where X = area, sq. in., and Y = mg. free H₂O.
* k = value for all 43 meat samples = 61.105 ± 0.311.
This version of the method is not as fast and is less accurate than the free moisture estimation by the area measurement. It requires a standard drying of the filter paper before use and the immediate weighing of the moist paper after pressing. Otherwise the moisture evaporated very rapidly from the filter paper on exposure to air. Also, removal of the filter paper from the muscle residue could not always be achieved. As various meat juices contain from 10 to 12% solids and as the method gives the same relationship between the wetted area and the free moisture for the juices containing as low as 6% solids, an admixture of 5 to 40% water to meat does not affect the accuracy of the determination. This was confirmed in this investigation and by Grau and Hamm (17, 18).

Increasing the solids in the meat fluids by adding various meat additives is without effect on the spreading of water on the filter paper (17). However, the additives which increase the viscosity of fluids, like Graham salt (metaphosphates) tended to decrease the wetted area for the same weight of water in the fluid (17, 18). On the other hand, the presence of the visible fat particles in the meat sample being pressed increases the moisture area around the meat film.

Calculation of Results. For the calculation, the free moisture area is determined by subtracting the surface of the meat film from the total moisture area. The difference multiplied by the regression coefficient of 61.10 mg of water per square inch gives the amount of free water in the meat sample being pressed. Another sample of the same meat should be run for the total moisture content. The results are best expressed as the per cent of the free water out of the total moisture content of the meat.

\[
\text{Per cent free H}_2\text{O} = \frac{\text{total area - total moisture (mg.) in muscle sample}}{\text{meat film area}} \times 61.10 \times 100
\]

The per cent of bound water equals 100 minus per cent of free water. The amount of free or bound water can be also expressed as per cent of the meat weight, or as the amount of bound or free water per unit weight of protein of the muscle.

Application of Method. The method can be successfully used for studying the degree of water holding of fresh lean muscles. As very small muscle samples are required for the determination, the method is adequate for studying the relative water holding of different muscles of the same animal or of the same muscles of different animals. It can also be used for studying the relative changes of water holding of muscles under different physiological and experimental conditions—i.e., setting in and resolution of rigor mortis, and effect of various meat additives or processing techniques.

Hamm (16) used this method for studying the biochemistry of muscular

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**Table VI. Free Moisture Content of Some Meats**

<table>
<thead>
<tr>
<th>Cereus No.</th>
<th>Muscle</th>
<th>Age Post-mortem</th>
<th>Total Moisture, %</th>
<th>Free H_2O as % of Total H_2O</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Vastus medialis</td>
<td>2</td>
<td>75.52</td>
<td>38.5</td>
</tr>
<tr>
<td>2</td>
<td>Rectus femoris</td>
<td>2</td>
<td>75.80</td>
<td>35.1</td>
</tr>
<tr>
<td>3</td>
<td>Biceps femoris</td>
<td>2</td>
<td>74.40</td>
<td>43.3</td>
</tr>
<tr>
<td>4</td>
<td>Adductor</td>
<td>2</td>
<td>74.16</td>
<td>42.5</td>
</tr>
<tr>
<td>5</td>
<td>Semimembranous</td>
<td>0.5</td>
<td>76.32</td>
<td>29.5</td>
</tr>
<tr>
<td>6</td>
<td>Longissimus dorsi</td>
<td>2</td>
<td>74.80</td>
<td>38.0</td>
</tr>
<tr>
<td>7</td>
<td>Rectus femoris</td>
<td>2</td>
<td>73.29</td>
<td>41.0</td>
</tr>
<tr>
<td>8</td>
<td>Semimembranous</td>
<td>1</td>
<td>73.10</td>
<td>39.6</td>
</tr>
<tr>
<td>9</td>
<td>Cured hams</td>
<td></td>
<td>74.96</td>
<td>39.6</td>
</tr>
<tr>
<td>10</td>
<td>Semimembranous</td>
<td>3</td>
<td>76.60</td>
<td>49.0</td>
</tr>
<tr>
<td>11</td>
<td>Biceps femoris</td>
<td>3</td>
<td>76.62</td>
<td>46.8</td>
</tr>
<tr>
<td>12</td>
<td>Leg</td>
<td>2</td>
<td>76.80</td>
<td>52.1</td>
</tr>
<tr>
<td>13</td>
<td>Leg</td>
<td>2</td>
<td>77.01</td>
<td>50.9</td>
</tr>
</tbody>
</table>

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contraction and relaxation by measuring the meat film area for the same weight of muscle. A very small area was found for the muscle in rigor, which then gradually increased with the resolution of the rigor caused either by the muscle aging or by the addition of adenosine triphosphate, analogous with the changes in the rigidity and elasticity of the glycogen isolated muscle fibers commonly used in studies of biochemistry and physiology (35).

A few examples of the application of the method are given in Table VI. The water-holding properties of different muscles of meat animals vary from muscle to muscle, from animal to animal, and with the post-mortem aging of the muscles.

The complimentary nature of the centrifugal method, as reported from this laboratory, and the filter paper method is shown in Figures 4 and 5. Figure 4 shows the changes in water-holding capacity of meat with post-mortem age. The upper curve is for the shrinkage at 70° C. and confirms earlier data obtained in Ohio by the centrifugal method (37), and the lower curve shows the changes in water-holding capacity of the same but unheated meat as determined by the filter paper method. Grau and Hannam (17-13) and the authors (39, 40) have previously reported on the effect of sodium chloride on the water-holding capacity of meat. Using the same meat, the two methods give the results shown in Figure 5. The parallelism of the data obtained is apparent. Each method can give useful and reproducible information on the water-holding capacity of meat. There remains much work to determine if information from one method carries directly to the other and to determine the quantitative relationship to consumer quality attributes, such as tenderness, shrinkage on cooking, and drip on freezing.

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