

EFFECTS OF FROZEN STORAGE ON FUNCTIONALITY OF MEAT FOR PROCESSING

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ABSTRACT

Frankfurter quality was monitored to evaluate effects of frozen storage on meat components. After fresh meat controls were tested, beef, pork, and pork fat were frozen and stored at -17.8°C for 1–37 wk. At 6-wk intervals, functional and quality tests were performed on thawed and control meat samples and on frankfurters made from the samples. Frozen storage significantly affected beef and pork lean (drip loss, % solids and N in drip, extractable protein, water binding, emulsifying capacity) and fat (beef-thiobarbituric acid and pork-peroxide values); frankfurters produced from these ingredients were also affected (cooking tests, penetration force, sensory panel scores).

INTRODUCTION

FREEZING AND FROZEN STORAGE can produce profound effects on the structural and chemical properties of muscle foods, including changes in muscle fibers, lipids, and proteins, all of which have the potential for significantly influencing the quality attributes of meat and meat products.

Early studies by meat researchers focused on relationships between drip volume and freezing conditions of beef muscle (Cook et al., 1926; Moran, 1932; Ramsbottom and Koonz, 1939, 1940). A concurrent line of investigation that continues today is the effect of freezing on meat tenderness (Paul and Child, 1937; Hankins and Hiner, 1941; Winger and Fennema, 1976).

The structural changes that occur in muscle were reported by Luyet (1959), who, in a series of experiments, demonstrated the damaging effects of ice crystal growth on muscle fiber morphology.

Lipids have been studied with regard to oxidative and degradative changes that result from frozen storage. Awad et al. (1968) reported a slight increase in extractable lipids from beef during 8 wk of frozen storage at -4°C . Cholesterol levels were unaffected by the treatment, but free fatty acids (FFA), thiobarbituric acid (TBA), and peroxide values (PV) increased. Keller and Kinsella (1973) observed no change in lipid extractability but did observe increases in carbonyls, TBA, and PV values. Also, phospholipid extraction decreased during the -18°C storage of ground beef.

Alterations in proteins have been observed previously in frozen meat. Sarcoplasmic proteins are less soluble after freezing of muscle tissue; moreover, there is a loss of specific components separated by electrophoresis and ultracentrifugation (Kronman and Winterbottom, 1960). Awad et al. (1968) found that total extractable protein (TEP), sarcoplasmic protein, and actomyosin extractability decreased with frozen storage. These investigators observed significant changes in banding patterns on electrophoretic polyacrylamide gels for sarcoplasmic and urea-treated actomyosin proteins. These degradative transformations appear to be evidenced by changes in the hydration of meat (Deatherage

and Hamm, 1960) and its water-holding capacity (Wierbicki et al., 1957a, b).

To date, investigators have studied these alterations mostly with regard to fresh meat products. Little attention has been focused on the relationship between frozen storage and the use of meat for processing. This study reports on the functional changes that occur in the storage of frozen meat block components and the relationship of these properties to changes within a comminuted meat product.

EXPERIMENTAL

Materials

Boneless beef chuck, boneless pork shoulders, and pork back fat were randomly selected from freshly cut lots from two commercial slaughterhouses. The beef came from ungraded mature dairy cows, while the pork and pork fat were obtained from United States #1 young swine. The raw materials were transported to the U.S. Department of Agriculture's Meat processing Laboratory where they were ground through a 1.91 cm plate on a Butcher Boy B-52 grinder. Aliquots of each were ground 2 \times through a 0.31 cm plate and were retained for proximate analysis. Eight sets of beef (6.5 kg), pork (5 kg), or pork fat (5 kg) were packaged in polyethylene (Cry-O-Vac) bags.

All samples which were to be frozen were placed in a -31.7°C freezer for 24 hr and then transferred to a -17.8°C freezer for the allotted storage time. Thawing was accomplished by transferring frozen bags to a 4.4°C cooler for 72 hr. Unfrozen meats were stored in the 4.4°C cooler until utilized. Frozen meat thaw exudate (drip) was measured, and a 50 ml sample was retained for Kjeldahl nitrogen and moisture determinations.

Product preparation

Proximate analyses for moisture, lipid, and protein were performed according to AOAC procedures (1975). Frankfurters were formulated to meet the following constraints: lipid $\leq 30\%$, protein $\geq 11\%$ (65% of the protein to come from beef), % moisture $\leq 4\text{X}$ protein + 10%, NaCl 2.5%, sugar 1.5%, commercial spice mixture 1% (Baltimore Spice Co., #FF3118), 150 ppm NaNO_2 , and 550 ppm sodium ascorbate.

Meats were ground through a 0.48 cm plate immediately prior to the manufacture of the frankfurters. The beef and pork, spices, cure, salt, and 3/4 water (added as ice) were comminuted for 2 min at 2500 rpm in a Koch-Alpina Pb-50 chopper. The remaining ice and the pork fat were then added, and the mixture was comminuted to a temperature of 15.6°C . The total time required for comminution was recorded. Emulsion was stuffed into #29 No-Jax casings (Union Carbide Corp.), linked into 15 cm lengths, and cooked and smoked for 90 min in an air conditioned smokehouse according to the following schedule: 10 min at 54°C dry bulb (DB), 30 min at 63°C DB and 54°C wet bulb (WB), and 45 min at 74°C DB and 63°C WB. The products were cooked to an internal temperature of 71°C and then showered to 32°C . Processing losses were determined as the difference in weight of raw and cooked sausage expressed as percentages. The frankfurters were stored in plastic bags in a 0.5°C cooler until they were tested within 48 hr.

Chemical and physical tests on meats and fat

Functionality and quality tests were performed on the raw components in investigating their relationship to the characteristics of finished products as follows:

The pH values of beef, pork, and pork fat were determined in duplicate on a Radiometer model 25 pH meter, with the electrode inserted into the tissue.

Water-holding capacity (WHC) was measured in triplicate according to the method of Wierbicki and Deatherage (1958).

Emulsifying capacity of the meats was determined in triplicate by the method of Swift et al. (1961).

Total extractable protein (TEP) was measured in duplicate by the method of Awad et al. (1968).

Thiobarbituric acid (TBA) values of fresh meat samples were obtained in triplicate according to the method of Witte et al. (1970). Values are expressed as absolute absorbance units.

Peroxide values (PB) of pork adipose tissue were determined in triplicate by the AOAC procedure (1975).

Product evaluation

Frankfurters were tested for cooking losses by the consumer cooking and severe cooking tests of Tauber and Lloyd (1947). The pH was measured with a combination electrode inserted into two sections of randomly selected product. A penetrometer (Marine Colloids, Inc.) was used to determine peak force required to penetrate cross sections of 2.54 cm frankfurter sections. A 7.8 mm blunt plunger was utilized at a crosshead speed of 20 cm/min. In addition, subjective evaluation of fat capping was conducted.

Sensory evaluation

Nine days after the products were manufactured they were evaluated by a taste panel. Frankfurters were cooked in boiling water for 10 min and kept between 50–55°C in an apparatus similar to that described by Caperaso (1978). A panel of 10 experienced tasters determined differences between control (fresh raw materials) and treated (frozen raw materials) frankfurters by a triangle test. Additional comments were solicited from the panelists concerning the basis for determining differences between test samples. Results were analyzed by use of tables published by Roessler et al. (1978).

Experimental design

Experiment 1 was conducted with a batch of beef, pork, and pork fat divided into 8 lots. Seven lots of samples were stored frozen for 1, 7, 13, 19, 25, 31, and 37 wk prior to manufacture. The fresh and frozen lots were analyzed as indicated above. All tests except the sensory evaluation were performed. A second experiment was initiated in which the fresh meat and fat controls were included at each of the seven frozen storage testing increments. The controls were purchased fresh from the same sources and were subjected to the full complement of chemical and physical tests; frankfurters were also manufactured from these samples. Sensory triangle tests were performed on controls and treated products. This brought the total number of replicates to $n = 9$ for control samples and to $n = 2$ for experimental samples. Except for taste panel determinations, the results are expressed as the means of the two experiments.

Statistical analysis

Data were analyzed by analysis of variance and polynomial regression statistical treatments. Significance was determined by the F-test at the 5% level of probability (Steel and Torrie, 1960).

RESULTS

THE EFFECTS of frozen storage on pork and beef thaw exudate volume are shown in Figure 1. Both species lost increasing amounts of exudate as storage time progressed, although beef drip losses remained higher ($P < 0.05$) throughout the majority of the testing increments. Previous reports have shown that there is no simple relationship between frozen storage time and drip volume. The data presented here are in general agreement with Rahelic et al.

Table 1—Effect of frozen storage on meat and product pH and processing parameters

Wk in frozen storage	Beef pH	Pork pH	Frankfurters pH	Comminution time (sec)	Processing loss (%)
0	5.8 (0.0)*	6.2 (0.1)	6.0 (0.0)	594 (41)	8.4 (0.6)
1	5.8 (0.0)	6.1 (0.1)	5.9 (0.2)	527 (117)	8.8 (0.4)
7	5.9 (0.1)	6.0 (0.0)	5.9 (0.1)	430 (30)	7.6 (0.1)
13	5.9 (0.0)	6.0 (0.1)	6.0 (0.1)	485 (63)	8.0 (0.5)
19	5.9 (0.0)	6.1 (0.0)	6.0 (0.1)	573 (92)	7.5 (2.3)
25	6.0 (0.1)	6.1 (0.1)	5.7 (0.1)	543 (53)	8.3 (1.7)
31	5.9 (0.1)	6.0 (0.1)	5.8 (0.1)	622 (22)	8.4 (0.4)
37	5.9 (0.0)	5.9 (0.0)	5.7 (0.0)	563 (39)	8.4 (2.1)

* Standard error (SE) in parentheses.

(1974), Jakobsson and Bengtsson (1973), Awad et al. (1968), Law et al. (1967), and Moran (1932). Investigators have also attributed thaw exudate losses to other factors. These include pH of meat (Empey, 1933; Sair and Cook, 1938; Ramsbottom and Koonz, 1940), storage temperature (Moran and Hale, 1932), time that meat spends in the range of -1 to -5°C (Reay, 1934), and time of freezing postmortem (Rahelic et al., 1974).

The relationship between drip volume (Fig. 1) and meat pH (Table 1) was determined by a linear regression analysis of the data, and correlation coefficient was calculated; the correlation coefficients for beef and pork were $r = 0.32$ and $r = -0.09$, respectively. These values were not significant at the 95% level.

The percentage of solids in the thaw exudates are presented in Figure 2. The values were expressed as the difference between 100 and % moisture in the thaw exudate. Increases in % solids in thaw exudates in beef and pork were closely parallel, both being significantly correlated

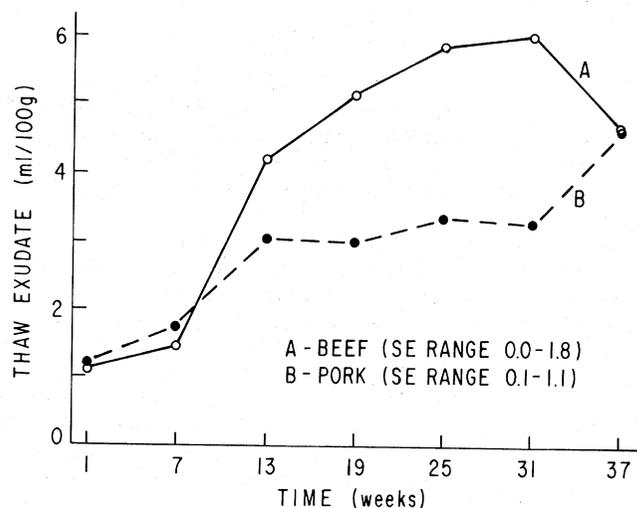


Fig. 1—Effect of frozen storage of meats on thaw exudate volume.

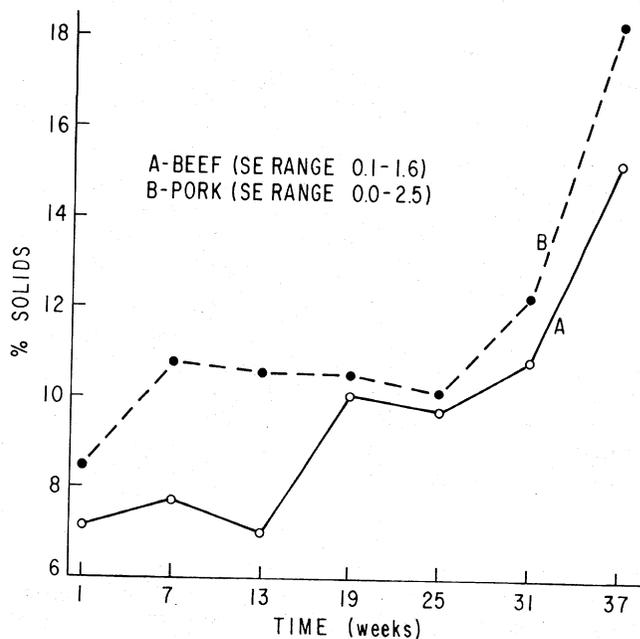


Fig. 2—Effect of frozen storage of meats on solids in thaw exudate.

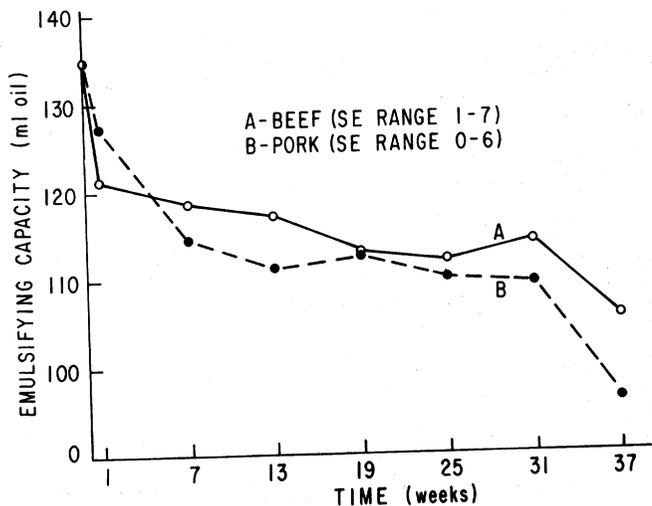


Fig. 6—Effect of frozen storage on meat emulsifying capacity.

beef ($P < 0.01$) and pork ($P < 0.01$). Dramatic diminution of emulsifying capacity occurred between 0 and 7 wk in pork and 0 and 1 wk in beef. Swift et al. (1961) showed that higher levels of salt soluble protein increased emulsifying capacity. The concurrent decrease in muscle protein solubility explains the decrease in emulsifying capacity observed.

It has been recognized for some time that lipids in frozen meat are likely to undergo oxidative deterioration (Ramsbottom, 1947). The common indicators of these changes are PV and TBA tests. In this investigation, the peroxide values (meq peroxide/kg lipid) of adipose tissue from pork rose from initial measurements of 0.9 to 2.37 ($P < 0.05$) after 25 wk of -17.8°C storage (see Fig. 7). The remaining 14 wk of storage resulted in a decrease of detectable peroxides. A rise followed by a decrease in PV was noted by Awad et al. (1968, 1969) on bovine and fish muscle. Degradation of hydroperoxides to carbonyl compounds has been postulated (Awad et al., 1968, 1969). TBA values, a measure of malonaldehyde and rancidity, of stored frozen pork and beef are shown in Figure 8. Observed values from both species remained quite close up to 13 wk in storage; thereafter, beef malonaldehyde production in relation to time of freezer storage accelerated ($P < 0.01$) at a higher rate than that of pork ($P > 0.05$). This phenomenon is contrary to expected results since pork lipids contain a higher percentage of unsaturated lipids than those of beef. However, evidence has also been presented by Shamberger et al. (1977) which agrees with our results. Recently, Brown et al. (1979) found that TBA values increased in beef muscle stored 180 days at -18°C . This increase is perhaps explained by the greater concentration of iron-containing pigments in beef muscle, which can act as prooxidants.

Table 1 contains results of observations of meat pH and tests made during and after frankfurter manufacture. Frozen storage duration did not appear to have an effect on variations in chopping time to 15.6°C . The variations observed in this parameter could be a function of ingredient temperature. Average comminution time was 544 sec (standard error, SE = 21.7). Processing losses averaged 8.1%, with an SE of 0.1. Mean frankfurter pH was 5.9. These parameters were not affected by ingredient freezing. The size and incidence of fat caps increased as frozen storage progressed. However, there was no evidence of severe emulsion breakdown.

Cooking tests showed that frozen storage did affect

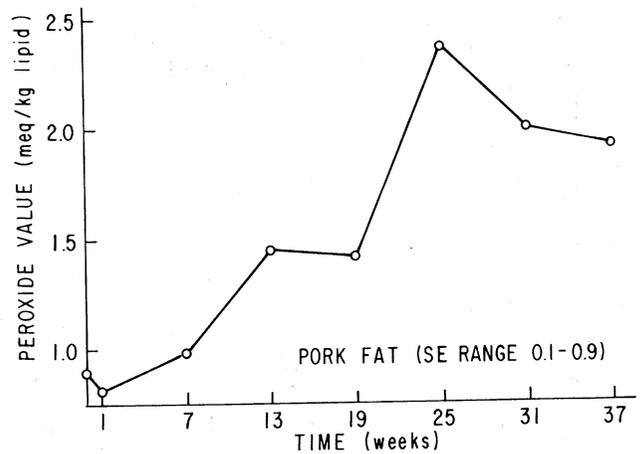


Fig. 7—Effect of frozen storage of pork fat on peroxide value.

product quality. The consumer cooking test (Fig. 9) demonstrated that as storage time increased the comminuted products lost their ability to retain moisture and fat ($P < 0.05$). This trend was more evident in the severe cooking test ($P < 0.01$). The greatest decrease in product quality appeared between weeks 1 and 19.

Cross sectional peak force required to puncture cores showed a significant decrease ($P < 0.01$) with greater storage time (Fig. 10), indicating a marked textural change. From an initial value of 900g, a decrease to 555g was recorded after the 37 wk testing period. Lee and Toledo (1976) reported that factors contributing to textural characteristics of a comminuted fish muscle product included presence of NaCl and/or polyphosphates, temperature of cooking, and type of heating medium used. In addition, these investigators found a significant relationship ($P < 0.01$) between product texture and comminution time and WHC. To test the hypothesis that decreased penetration force was the result of variations in chopping time, the linear regression and correlation coefficient between these factors was determined. Results indicated that the relationship was not significant at the $P < 0.05$ level ($r = -.44$) for beef and pork frankfurter manufactured with commercial

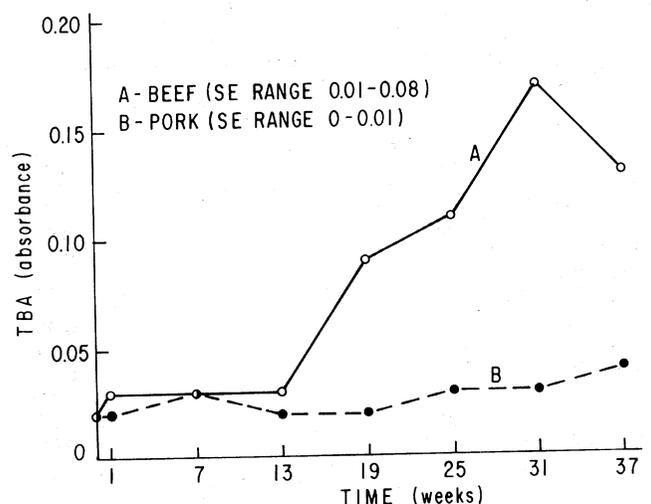


Fig. 8—Effect of frozen storage on meat thiobarbituric acid.

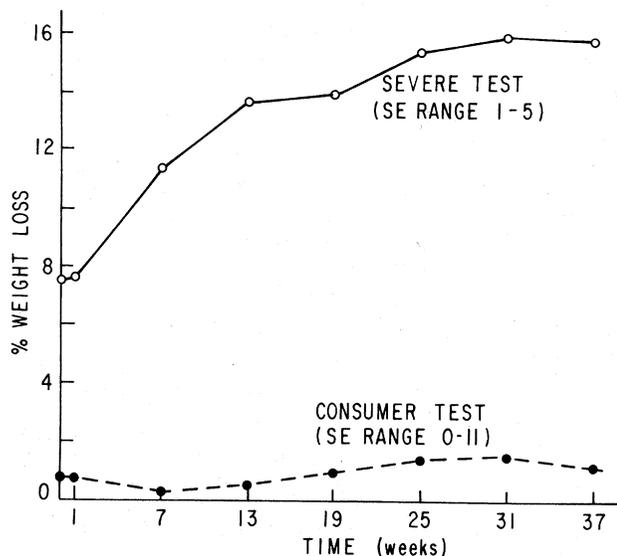


Fig. 9—Effect of frozen storage of meats and fat on frankfurter cooking tests.

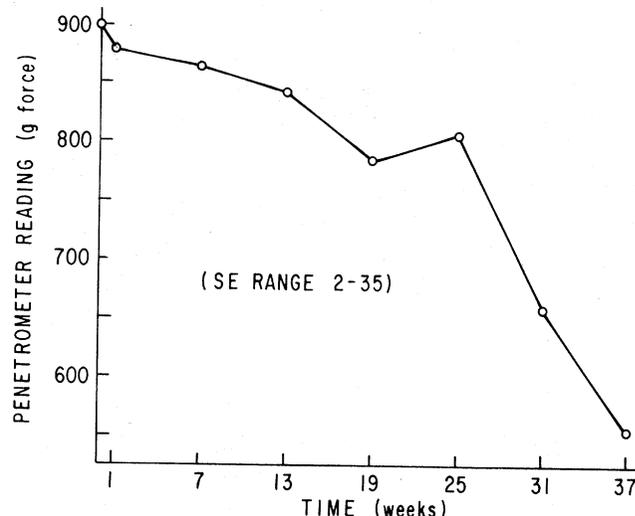


Fig. 10—Effect of frozen storage of meats and fat on frankfurter texture.

equipment. Correlation coefficients for various factors compared to penetrometer readings are presented in Table 2. The following characteristics were shown to relate significantly with these scores: beef emulsifying capacity ($r = 0.75$, $P < 0.05$), pork emulsifying capacity ($r = 0.81$, $P < 0.05$), pork WHC ($r = -0.75$, $P < 0.05$), beef TEP ($r = 0.83$, $P < 0.05$), pork TEP ($r = 0.93$, $P < 0.01$), beef TBA ($r = -0.87$, $P < 0.01$), severe cooking test ($r = -0.75$, $P < 0.05$), and triangle test ($r = -0.77$, $P < 0.05$). These comparisons suggest that the state of the myofibrillar proteins directly affect textural quality of emulsified meat products.

Triangle sensory tests, comparing fresh meat (control) frankfurters with frozen meat (treatment) frankfurters, upheld the evidence that significant changes had occurred during the 37 wk freezing period (see Table 3). After meat and fat were stored for 7 wk at -17.8°C , the resulting frankfurters were discernable ($P < 0.01$) from controls. This tendency continued throughout the course of storage, progressing by 31 wk to differences which all of the panelists were able to distinguish. Solicited comments added insight into the factors that panelists used to discern differences between samples. The majority of the comments were related

to differences in texture between frozen and fresh product. In addition, there were comments related to rancid flavor in the treatment samples and a lack of overall flavor as time progressed. Two panelists were especially sensitive to oxidative notes, and one of these members detected rancidity as early as 1 wk of storage of meat materials. The other panelists did not, by and large, recognize an oxidative note before 31 wk of storage.

Comparing PV and sensory scores (% correct choice) by regression analysis it was determined that a highly significant ($P < 0.01$) correlation coefficient (0.89) existed between these parameters. Panelists, however, did not, on the whole, perceive rancidity until the 31 wk sample, suggesting that lipid alterations were perceived as secondary defects until the later stages of the experiment. During this time textural changes were the predominant characteristics recognized by panel members. It is quite possible that these changes were induced by the lipids.

DISCUSSION

IN THE PRESENT STUDY, we evaluated changes in beef, pork, and pork fat stored at -17.8°C for 37 wk. The data

Table 2—Correlation coefficient between mean penetration scores and various parameters

Parameter	Correlation coefficient (r)
Beef emulsifying capacity (ml)	0.75*
Pork emulsifying capacity (ml)	0.81*
Beef WHC (%)	0.47
Pork WHC (%)	0.75*
Beef TEP (%)	0.83*
Pork TEP (%)	0.93**
Beef TBA (absorbance)	-0.87**
Pork TBA (absorbance)	0.47
Comminution time (sec)	-0.44
Consumer cook test (%)	-0.62
Severe cook test (%)	-0.75*
Triangle test (% correct responses)	-0.77*

* Correlation coefficient significant ($P < 0.05$).

** Correlation coefficient significant ($P < 0.01$).

Table 3—Triangle sensory evaluation of frankfurters made from frozen beef, pork, and pork fat

Wk in frozen storage	% Choosing correct pair ^a	Comments
0	—	—
1	50	Flavor-1 ^b , texture-5, rancidity-1
7	80**	Flavor-1, texture-6, rancidity-2
13	80**	Flavor-1, texture-3, rancidity-1
19	80**	Flavor-3, texture-5, rancidity-2
25	90***	Flavor-1, texture-8, rancidity-2
31	100***	Flavor-1, dryness-1, texture-9, rancidity-5
37	100***	Texture-6, rancidity-4

^a $n = 10$.

^b Number of responses.

** Significant at probability level $P < 0.01$.

*** Significant at probability level $P < 0.001$.

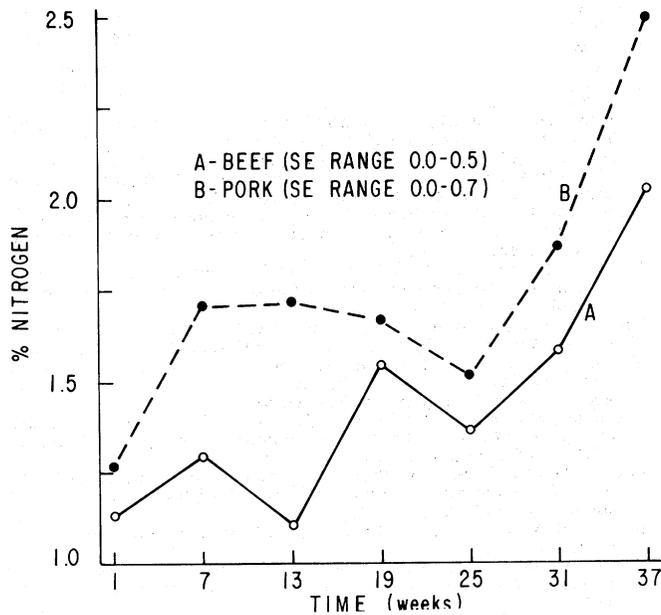


Fig. 3—Effect of frozen storage of meats on nitrogen in thaw exudate.

with longer freezing time ($P < 0.01$). These levels ranged from 8.5% in pork and 7.1% in beef exudates after 1 wk to 18.3% in pork and 15.2% in beef after 37 wk. Toward the latter storage times there was an accelerated increase of solids in the thaw exudate. This appeared between 25 and 31 wk of storage of sample pork, and 31–37 wk of storage of sample beef.

The changes in the concentration of dissolved nitrogenous compounds in the thaw fluid are shown in Figure 3. As with drip volume (Fig. 1) and percent solids in the exudate (Fig. 2), contents of nitrogen in the exudate increased with frozen storage time ($P < 0.05$), with the increasing nitrogen losses in the meat closely following the course of solids formation. In the pork samples there were two periods dur-

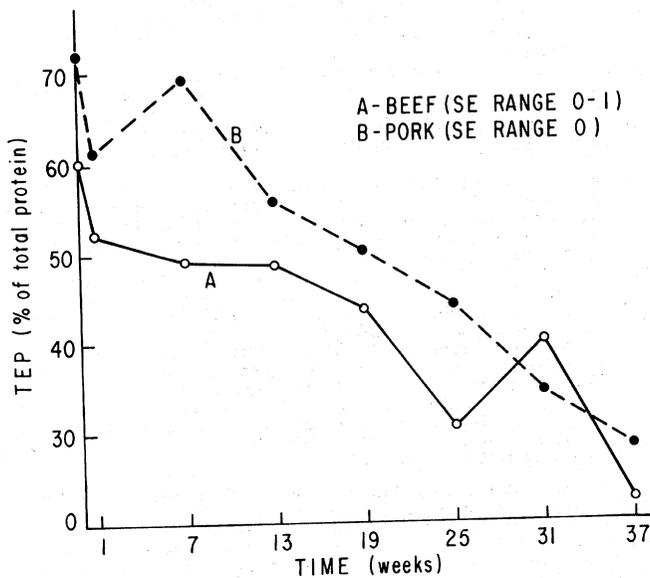


Fig. 4—Effect of frozen storage of meats on total extractable protein.

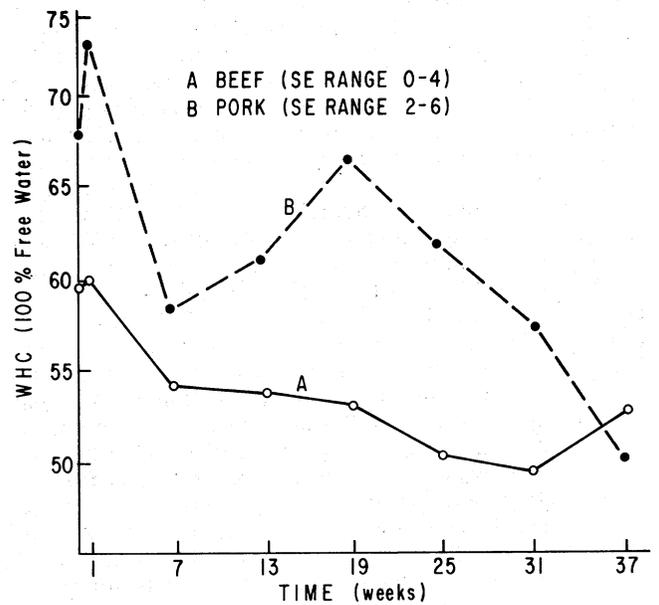


Fig. 5—Effect of frozen storage of meats on water holding capacity.

ing which nitrogen losses sharply increased. This occurred between the first and seventh week and then after 25 wk of storage. Nitrogen content of beef thaw exudate gradually increased up to 19 wk, then there was a very sharp rise between the last two test periods. According to Lawrie (1979), the solubilized nitrogenous compounds in this fluid are sarcoplasmic proteins, creatine and creatinine, free amino acids, peptides, nitrogenous bases and their corresponding nucleosides and nucleotides, purine and pyrimidine degradation products, porphyrin containing compounds, and metabolic cofactors. Of these, amino acid, peptides, proteins, and vitamins represent potential nutritional losses.

The total extractable protein (TEP) values determined from the eight sampling increments are presented in Figure 4. From initial means of 72% TEP for pork and 60% TEP for beef muscle, decreases to 28% ($P < 0.01$) and 22% ($P < 0.01$), respectively, were observed. For both species of meat a gradual, almost linear, decrease in TEP was observed. Other investigators likewise have found that the extractable protein or nitrogen decreases with increased storage time. Awad et al. (1968) observed a change from 90.99% to 50.84% TEP in beef muscle over an 8 wk storage period at -4°C . According to Khan et al. (1963), the % total extractable nitrogen of chicken pectoralis muscle decreased from 88 to 56% over a 50 wk period at -4°C .

The water-holding capacity (WHC) of the meat of both species decreased with frozen storage time ($P < 0.01$). These data are presented in Figure 5. WHC of both beef and pork samples produced an increase after 1 wk of freezing. This was followed by a sharp decrease in samples frozen for 7 wk. Beef WHC decreased gradually throughout the remainder of the testing periods. Pork WHC increased between weeks 7 and 19. The significance of this observation is not clear. From 19–37 wk pork WHC markedly decreased. Deatherage and Hamm (1960) reported decreased WHC for beef which had been slow frozen. This phenomenon has been attributed to a mechanical loosening of the muscle tissue by the formation of ice crystals inside the cells. Extensive splitting of the sarcolemma has also been observed (Hiner et al., 1945).

Figure 6 shows a decrease in emulsification capacity in

presented here indicate that alterations occurred in the chemical and physical properties of these components, especially in proteins and lipids. Theories regarding the frozen storage induced deterioration of muscle have been proposed. Callow (1955) attributed the phenomenon to the presence of high concentrations of natural salts in the juice of frozen meat. A second explanation postulates interaction between proteins and lipid degradation products. Free fatty acids, the products of hydrolytic rancidity, have been shown to bind to proteins in model systems (Bull and Breese, 1967a) and will denature proteins (Bull and Breese, 1967b). In addition, Buttkus (1969) has shown that malonaldehyde, a product of polyunsaturated fatty acid oxidation, reacts with muscle proteins to produce a denaturation. Moreover, observations we have made on the possible effects of peroxides in meat emulsion systems suggest that these compounds may exhibit deleterious effects on emulsion stability, implying a protein alteration. Sikorski et al. (1976), in a review of protein changes in frozen fish, included partial dehydration and the formation of formaldehyde as additional factors influencing the denaturation of muscle proteins. The observed changes in the present study support the theory that degradation products formation and interaction with muscle tissue result in decreased protein solubility, emulsifying capacity, and WHC. Moreover, it is plausible to suggest that the observed increases in solids and nitrogen in the thaw exudates were the result of the endogenous salt solubilization of these substances.

The factors involved in the stabilization of emulsified meat products include the solubilization of myofibrillar proteins, especially myosin, and their inclusion as an emulsifier, interfacing hydrophobic fat globules, and a hydrophilic aqueous phase. Once heat denaturation of the proteins occurs during cooking, a stable water, protein, and lipid matrix is set. Any factors upsetting this balance could result in deleterious alterations in the final product (Kramlich, 1971). Evidence obtained here indicates that the quality of proteins and fat deteriorated and that the comminuted products manufactured from the frozen meat block components gradually decreased in quality characteristics. While we found no acute emulsion instability during the 37 wk study, the trend indicated an eventual failure through further storage. The first signs of change were evident after only 1 wk of freezing, and by 25 wk all observed characteristics showed significant degradative changes. By 7 wk, panelists could readily discern differences between treatment and control products. For this reason it is recommended that short frozen storage periods be used for meat block components intended for use in comminuted meat products.

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