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Comminuted Meat Emulsions:
Factors Affecting Meat Proteins as Emulsion Stabilizers

SUMMARY

An investigation was made of factors influencing the effectiveness of meat proteins as stabilizers of emulsions of fat. The effects of pH, concentrations of sodium chloride and other salts, and added water on the emulsifying capacity of meat and its water-soluble and salt-soluble proteins were determined. The emulsifying capacity of water-soluble proteins increased in increasing concentrations of sodium chloride; their capacity was maximum at *ca.* pH 5.2. The capacity of the water-soluble proteins to emulsify fat in solutions of different potassium salts was SCN $>$I $>$ NO_3 $>$ Br $>$ Cl $>$ SO_4, or the order of the Hofmeister series. At pH values approaching the isoelectric point of the salt-soluble proteins, the emulsifying capacity of the proteins increased as concentration of sodium chloride was increased. Similarly, at these pH values, the emulsifying capacity of meat increased in increasing concentrations of sodium chloride. An increase in the emulsifying capacity of meat on increasing pH from 7 to 8 was attributed to an increased extraction of proteins. Eliminating, or restricting, added water in salted meat reduced the effectiveness of the treatment with salt, and consequently the emulsifying capacity of the meat.

INTRODUCTION

Emulsion curing, the basic operation in sausage making, involves the production of emulsions that derive stability from the ability of meat proteins to enclose fat globules in matrices formed by protein membranes (Hansen, 1960). Failures and problems in producing comminuted meat products can often be traced to poor retention of fat. The capacity of meat proteins to stabilize emulsions of fat has been shown to be as high as 1.61 g fat per mg protein (Swift *et al.,* 1961). Thus, this capacity appears far greater than that required to emulsify fat in commercial products in which fat may be incorporated at levels of only 2 or 3 g fat per g protein. Consequently, the problem arises from a fractional use of the capacity of the proteins in the raw materials. The present investigation was made to obtain further knowledge of means of utilizing these meat proteins more efficiently.

A previous investigation (Swift *et al.,* 1961) showed that the factors affecting the ability of meat to emulsify fat in a saline system include the extent to which meat is comminuted, the proportion of saline phase, the rates of addition of fat and of mixing, and the temperature. The results also indicated that the effectiveness and efficiency of the meat proteins vary with type and concentration of protein. The experiments described herein were concerned with the effects of pH, concentration of NaCl, different anions, and added water on the stabilizing capacity of meat and water-soluble and salt-soluble meat proteins.

EXPERIMENTAL

Portions of longissimus dorsi, semimembranosus, and semitendinosus muscles were removed from young steers approximately one hour after slaughter. The water-soluble and salt-soluble proteins were extracted immediately afterward. Unextracted meat samples were stored at 3°C up to 5 days before use. The principal steps in the basic method used for determining the emulsifying capacity of meat involved: 1) comminuting 50 g of ground meat in 200 ml of cold 1*M* NaCl (0–3°C); 2) adding 37.5 ml of cold 1*M* NaCl (0–3°C) to 12.5 g of the resulting slurry; 3) adding 50 g of soybean oil to the 50 ml of dilute slurry; and 4) mixing-cutting in an Omni-Mixer (no endorsement implied) at *ca.* 13,000 rpm while adding oil at approximately 0.8 ml per sec until an emulsion formed, thickened, and collapsed. Modifications necessary to meet the demands of the experiments are noted in their descriptions. Application of the method to extracts of proteins involved replacing the diluted meat slurry with 50 ml of protein extracts. Details of the methods for preparing the protein extracts and determining the emulsifying capacity of meat samples and protein extracts have been described (Swift *et al.,* 1961). The water-soluble extract was prepared, as before, by extracting ground tissue, using four successive extractions and a total of 275 ml water per 100 g tissue. The residue remaining was extracted with 0.6*M* NaCl to prepare the extract of salt-soluble proteins. Use of 0.6*M* NaCl rather than 1.0*M* NaCl as the extractant was the only variation from the procedure previously described. The total amount of fat added when the emulsions collapsed is reported as emulsifying capacity, ml fat emulsified/2.5 g meat, or, in the case of protein extracts, ml fat emulsified/50 ml extract. Data presented are either averages of duplicate determinations, or experimental results were confirmed by at least one replication. Generally, experiments were repeated three or four times to ascertain that observations were qualitatively and, within reason, quantitatively repeatable, using tissues from different animals, tissues obtained pre- and post-rigor, and other protein extracts. This was done because meat tissues and extracts prepared from them can be assumed to vary somewhat in composition and, consequently, in behavior when treated to vary pH and concentration of salt.

Determining pH and NaCl effects on water-soluble and salt-soluble proteins. The emulsifying capacity of 50 ml of extracts of water-soluble and salt-soluble proteins, respectively, containing 8.56 mg protein/ml and 6.00 mg protein/ml, was determined after adjustment of pH and concentration of NaCl. NaCl was adjusted by adding appropriate amounts of solid NaCl to each of three portions of the aqueous solution of water-extracted proteins to convert them to 0.5, 1.0, and 2.0*M* NaCl. The pH of 50-ml portions of these solutions was adjusted to values ranging from 4.0 to 7.85. Portions of the solution of salt-soluble proteins extracted from meat with 0.6*M* NaCl were diluted 1:1 with either water, 0.6*M* NaCl, or 1.8*M* NaCl to provide solutions containing equal concentrations of the proteins dispersed in 0.3, 0.6, and 1.2*M* NaCl. The pH of 50-ml portions of these solutions was adjusted to obtain values in the range of 5.0 to 8.0. In this, and subsequently described experiments, a few drops of solutions of NaOH or HCl were added to the meat slurries or protein extracts to obtain approximately adjusted pH values. Then, about 1 hr later, the pH values were measured accurately, and emulsifying capacity was determined.

Determining salts effects on water-soluble proteins. The emulsifying capacity of 50-ml portions of an aqueous extract containing 4.80 mg protein/ml was determined after appropriate amounts of solid KSCN, KI, KNO_3, KBr, KCl, and K_2SO_4 had been added to produce 0.5*M* concentrations of the salts.

Determining pH and NaCl effects on meat. The emulsifying capacity of semimembranosus muscle (stored 2 days at 3°C) was determined at pH values ranging from 4.6 to 8.1 in 0.3, 0.6, and 1.0*M* NaCl. This involved applying the basic method in one series, and in the others substituting 0.3 and 0.6*M* NaCl for 1*M* NaCl throughout the procedure.

Determining water effect on salted meat. Samples of finely comminuted semimembranosus muscles were thoroughly mixed with 5.85% solid NaCl. Different volumes of cold 0.6*M* NaCl (3°C) were mixed with these to provide proportions of solution to meat ranging from 0:1 to 4:1. After storage overnight at 3°C weights of mixtures containing 2.5 g of meat were mixed with appropriate amounts of water and NaCl to produce suspensions having a 50-ml volume and 0.30 ionic strength, which were used as the saline phase in determinations of emulsifying capacity. The adjustment to 0.30 ionic strength was selected to minimize precipitation or further solubilization of proteins during emulsification, since either would tend to obscure the effects of varying the water content of the initially prepared samples of salted meat.

RESULTS AND DISCUSSION

Fig. 1 shows the results of determinations of the emulsifying capacity of water-soluble proteins in 0.5, 1.0, and 2.0*M* NaCl and at pH values ranging from 4.0 to 7.85. The results indicate that, at all pH values, the emulsifying capacity of these proteins increased with increasing concentrations of NaCl. Variation of pH had a marked effect, with maxima at approximately pH 5.2±0.1, and sharply reduced activity in either more alkaline or acidic solutions. The results indicate that water-soluble proteins exert their maximum activity at a lower pH than is normally obtained in meat, and respond favorably to higher concentrations of NaCl than those desired for flavoring purposes.

The data in Table 1 show the effect of 0.5*M* solutions of different potassium salts on the emulsifying capacity of water-soluble proteins. The results provide evidence of the mechanism of the interaction of NaCl with these proteins. The emulsifying capacity of the proteins in the 0.5*M* solutions decreased in the order KSCN, KI, KNO_3, KBr, KCl, and K_2SO_4, or the order of the Hofmeister series. In investigations of the spreading of protein films (Adams, 1941), spreading was shown to be aided by anions in the increasing order Cl^-, Br^-, I^-, and SCN. The effect was attributed to different degrees of unfolding of protein molecules. The analogous effects of anions on spreading and on the emulsifying capacity of proteins suggest that NaCl increases the emulsifying capacity of water-soluble proteins by unfolding their structure, thus extending their ability to enclose fat globules in membranes.

Fig. 2 shows the effects of varying pH and content of NaCl on the emulsifying capacity of salt-soluble proteins. The emulsifying capacity of the salt-soluble proteins was not significantly different in 0.3, 0.6, and 1.2*M* NaCl except that at pH values approaching or lower than pH 5.4,

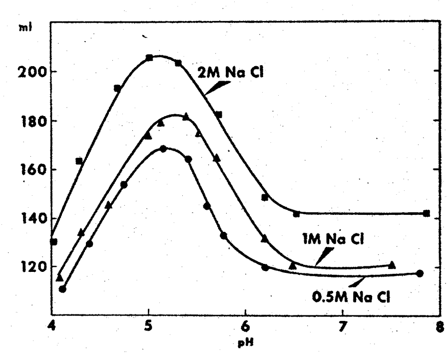

Fig. 1. Influence of pH and concentration of NaCl on the amount of fat emulsified by water-soluble proteins.

Table 1. The effect of different salts on the emulsifying capacity of water-soluble proteins.[a]

Salt	KSCN	KI	KNO_3	KBr	KCl	K_2SO_4
Fat emulsified (ml)	165	146	134	130	119	117

[a] 4.80 mg protein/ml; pH values before and after addition of salts were respectively 5.32 and 5.12.

approximating the accepted isoelectric points of the salt-soluble proteins, increasing NaCl content produced a significant increase in emulsifying capacity. In the region of this pH, decreased solubility of the proteins would be expected to decrease emulsifying capacity. Since increasing ionic strength lowers the pH at which proteins attain minimum solubility (Taylor, 1953), the effect of NaCl in the pH range of from 5.0 to 6.0 can logically be explained by assuming that an increased concentration of NaCl lowered the pH at which the salt-soluble proteins lost solubility; consequently, an increased content of NaCl enhanced emulsifying capacity by maintaining more thoroughly dispersed protein.

Fig. 3 shows the results obtained with meat on comminuting and dispersing samples in 0.3, 0.6, and 1.0*M* NaCl and then determining their emulsifying capacity after pH was adjusted to values ranging from 4.6 to 8.1. The results reflect differences in the ability of the solution both to extract proteins and to disperse and modify their characteristics. Variation of both pH and content of NaCl strongly influenced emulsifying capacity. An increasing concentration of NaCl increased emulsifying capacity at pH values ranging from approximately 5.4 to 6.0, but had no significant effect at low or high pH values (5 or 7 to 8). In general, emulsifying capacity increased markedly with increasing pH.

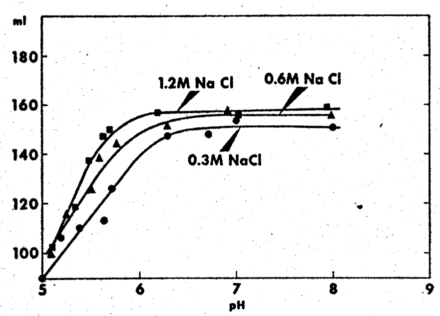

Fig. 2. Influence of pH and concentration of NaCl on the amount of fat emulsified by salt-soluble proteins.

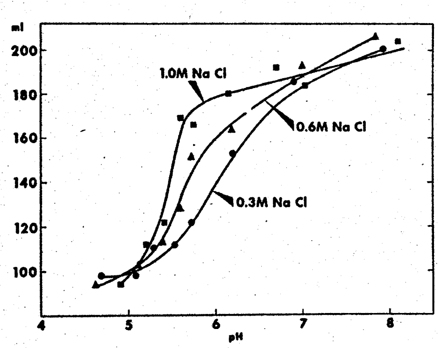

Fig. 3. Influence of pH and concentration of NaCl on the amount of fat emulsified by meat.

Table 2. The effect on the emulsifying capacity of meat of varying content of added water.

Ratio, brine to meat[a]	Emulsifying capacity (ml)[b]			
	A	B	C	D
4.0	136	153	196	150
1.0	139	149	199	153
0.5	124	129	189	......
0.25	122	137	170	131
0.125	118	......	......	......
0.0	......	106	163	126

[a] Ratio indicates ml 0.6*M* NaCl added per g meat; 0.0585 g NaCl/g meat was also added.

[b] Four samples, capacity determined on weights equivalent to 2.5 g meat in meat-brine mixture adjusted to 0.30 μ and 50 ml volume.

The significance of some of the results of varying pH and concentration of NaCl on the emulsifying capacity of meat can be understood by comparing them with the results obtained on water- and salt-soluble proteins (Figs. 1 and 2). The curves in Fig. 3 lack maxima or inflections at pH 5.2, where emulsifying capacity ranged from 102 to 110. Only a tendency to plateau is seen in the region of pH 5.0, where emulsifying capacity was approximately 100. These results indicate that the apparent contribution of water-soluble proteins throughout the entire range of pH values and at all three concentrations of NaCl accounted for an emulsifying capacity of not more than approximately 100.

A comparison of the curves in Figs. 2 and 3 indicates that the emulsifying capacities of salt-soluble protein and meat were generally similar in the region of pH 6. The explanation offered above to explain the effect of varying content of NaCl on the emulsifying capacity of salt-soluble proteins in this pH region would also seem to apply to its effect on meat, i.e., increasing NaCl minimized the effect that lowering pH had on the solubility of the proteins and, hence, favorably affected emulsifying capacity. Further comparison of the results shown in Figs. 2 and 3 indicates that the emulsifying capacity of meat, but not that of the salt-soluble proteins, increased at pH values ranging from 7 to 8. Consequently, the evidence indicates that increasing pH did not affect dispersed proteins. A logical explanation is that increasing alkalinity increased extraction of the proteins.

Table 2 shows the effect on emulsifying capacity of varying the proportion of water added to meat. The lower levels of added water approximated those employed in sausage making. The experiments were designed to determine the effect of varying the water content of meat salted before its use in emulsification. Varying the degree of dispersion of the meat during emulsification was avoided, since, as previously indicated, varied dilution of meat or proteins markedly affects emulsifying capacity (Swift *et al.*, 1961). Emulsification was conducted in an aqueous phase adjusted to 0.30 μ, a medium chosen to avoid precipitation or new extraction of proteins.

The results show that emulsifying capacity was reduced 15–30% by a drastic reduction of water content. The greater part of the meat proteins were conditioned by the treatment with NaCl so that they were dispersible in weak brine and active during the process of emulsification. The results, considered together with those previously published, indicate that the potential emulsifying capacity of meats is not obtainable when the water content of meat emulsions is restricted. Reduction of added water apparently affects the interaction of NaCl with the proteins to only a moderate extent, whereas, as pointed out above, the reduced effectiveness that follows emulsifying in a limited amount of aqueous phase, or with concentrated proteins in this phase, has been shown to be appreciable.

The results show that pH and NaCl content are extremely critical factors affecting the capacity of meat to stabilize emulsions. The pH of ordinary raw meat is approximately 5.4 to 6.0, and the usual ionic strength of sausage emulsions is approximately 0.6 μ (the 0.25 μ of meat plus the 0.34 μ of 2% NaCl), or approximately the equivalent of 0.6*M* NaCl. Under these conditions, effectiveness in emulsification is most sensitive to changes in pH and content of NaCl. The results indicate that increasing either, or both, pH and NaCl should improve the emulsification ordinarily obtained in preparing emulsion-based meat products.

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