AN ABSTRACT OF THE THESIS OF

ARMANDO CUMMINS for the degree MASTER OF SCIENCE
(Name of student) (Degree)

in Food Science and Technology presented on August 27, 1974
(Major Department) (Date)

Title: INFLUENCE OF POLYPHOSPHATES ON THE EMULSIFYING CAPACITY OF MILK AND MEAT PROTEINS

Abstract approved: _____
A. Anglemier

The influence of polyphosphates upon the emulsification of enzymatic hydrolyzates of casein and lactalbumin and upon salt-soluble meat proteins was determined by a model system in which oil-in-water emulsions were formed.

Sodium acid pyrophosphate, sodium tripolyphosphate and sodium hexametaphosphate were mixed together in a weight ratio of 4:2:4, respectively, to form a polyphosphate blend. This blend was added at a level of 0.5% (w/v) to casein or lactalbumin dissolved in 3% NaCl, pH 6.0. The polyphosphate blend was also added at the above level to meat proteins solubilized in 3% NaCl, pH 5.7. Emulsified volume (EV), total g of oil emulsified per 25 ml of protein solution, was determined for the above proteins with or without polyphosphates at varying protein concentrations. Data were also

graphically expressed as emulsifying capacity (EC) and oil phase volume (OPV).

For all proteins studied with or without polyphosphates, EV and OPV values increased with increasing protein concentration whereas EC values decreased.

Addition of the polyphosphate blend to casein solutions containing protein levels in excess of 2 mg/ml resulted in significantly ($P< 0.01$) higher EV levels than those of the controls. Conversely, the EV values of the polyphosphate-treated lactalbumin solutions were significantly ($P< 0.01$) lower than those of the controls at all protein levels tested. The diverse data obtained with these two proteins appear to be related to variations in molecular size and shape and to differences in the manner in which the protein-phosphate interactions occurred to cause the polyphosphates to enhance the emulsification of casein while depressing that of lactalbumin.

Addition of the polyphosphate blend had little or no effect upon the emulsification of meat protein extracts obtained from fresh, frozen or refrozen samples. Thus, it was concluded that the polyphosphate blend did not modify nor exert any detectable influence upon the emulsification of solubilized meat proteins as tested in a model system.

Influence of Polyphosphates on the Emulsifying Capacity of Milk and Meat Proteins

by

Armando Cummins

A THESIS

submitted to

Oregon State University

in partial fulfillment of
the requirements for the
degree of

Master of Science

June 1975

APPROVED:

Professor of Food Science and Technology
in charge of major

Head of Department of Food Science and Technology

Dean of Graduate School

Date thesis is presented August 27, 1974

Typed by Ilene Anderton for Armando Cummins

ACKNOWLEDGEMENT

The author wishes to express appreciation and gratitude to Dr. Allen F. Anglemier for his guidance and encouragement throughout the course of this work and in the preparation of the manuscript. Appreciation is also extended to Dr. Morris W. Montgomery for his help in the preparation of the thesis.

The author is indebted to the Department of Food Science for the use of its facilities in this research project.

I would also like to acknowledge the Agency of International Development for their financial support during the course of this study.

Sincere appreciation is extended to my wife, Maria Teresa, for her encouragement and understanding during my graduate study.

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INTRODUCTION

Many conventional food proteins (egg, meat, milk, etc.) are consumed for gastronomical pleasure rather than because they are the dietary source of amino acids and nitrogen essential for life. In these food systems, proteins are structural components contributing specific functional properties directly associated with their popularity as foods (Mattil, 1971). Flavor, color, solubility, rheological behavior (viscosity, gelation), water-holding ability, emulsifying capacity and foaming ability are the major functional properties of proteins of current concern to the food industry (Wuhrmann, 1972).

Due to the rapid increase in the world's population during the past decade, the demand for conventional food proteins has surpassed their availability and supply. As a result of this scarcity, the food industry has initiated research on the properties of conventional food proteins. Data from these studies should provide pertinent information about their functional roles in foods that make them so appreciated by the consumer. By characterizing the functional properties of conventional food proteins, the food industry might be able to develop new foods or reformulate current foods in which proteins can be extended and/or used more efficiently.

Furthermore, such knowledge should also provide a basis for developing the appropriate technology to simulate these properties in proteins extracted and processed from unconventional food sources (i.e., algae, bacteria, yeast, leaves, oil-seed residues, trash fish, etc.). Utilization of the latter sources should help alleviate the world's food protein shortage.

Emulsification

The ability to emulsify lipids is one of the more important functional properties of proteins. In fact emulsifying capacity (EC) is usually one of the first measurements studied when a systematic investigation of functional properties of a food component is undertaken. The EC has been defined as the maximum amount of lipid (oil) a given amount of protein will emulsify (Swift et al., 1961).

Becher (1965) defined an emulsion as a heterogeneous system consisting of at least one immiscible liquid (dispersed phase) intimately dispersed in another (continuous phase) in the form of droplets having diameters in excess 0.1 micron. Although these systems possess minimal stability, such additives as surface active agents, finely divided solids, etc., tend to increase the stability.

Sausage-type products made from meat emulsions are designated as oil-in-water (O/W) emulsions. Fat is the dispersed phase, water is the continuous phase and protein is the emulsifying

agent. Hansen (1960) found the presence of protein to lower the interfacial tension between the two phases. Protein also forms a strong protective membrane around the fat droplets or dispersed phase. The different amino acid constituents of protein account, in part, for the manner in which protein molecules interact at the oil-water interface (Swift, 1966). Nonpolar (hydrophobic) groups are attracted to the lipid phase, while polar (hydrophilic) groups are oriented to the water phase. Thus, the oil phase is attached to the water phase through the protein. These data were obtained from studies utilizing light and electron microscopy methods (Carpenter and Saffle, 1964; Mayer et al., 1964; Borchert et al., 1967). These workers also found that after thermal processing of meat emulsion products, the fat globule membranes became highly disrupted and the protein connected with the continuous phase was coagulated into dense irregular zones.

Model Systems

Swift et al. (1961) developed the first model system to measure the emulsifying capacity under controlled conditions. The basic equipment used by these workers consisted of a one-pint jar, an Omni-Mixer (13,000 rpm), and a burette containing melted lard. A meat protein extract was placed in the blender jar, and during mixing, liquid fat was added from the burette. This resulted in the

formation of a stiff emulsion similar to whipped cream which, on continued mixing and fat addition, would eventually break or collapse. This change, the end point, was detected visually since it was accompanied by a sudden decrease in viscosity, which is frequently referred to as the break-point of the emulsion. The amount of fat emulsified could be read directly from the burette while the mixing speed, rate of fat addition and the protein concentration of the meat extract could all be easily controlled.

The break-point occurs when all of the soluble protein available for emulsification is at the oil-water interface. At this point, addition of more oil causes a reversal of the phase yielding a water-in-oil (W/O) dispersion possessing very different characteristics. The creamy texture typical of an O/W emulsion is converted to a greasy texture characteristic of a W/O emulsion (Saffle, 1968).

Oil-in-water emulsions conduct electrical current whereas a W/O emulsion will not. This principle has been used by certain researchers to determine the end point more accurately (Webb et al., 1971; Satterlee et al., 1973). During the formation of an O/W emulsion the viscosity of the system increases causing a steady rise in amperage requirements of the blender motor. The sudden drop in viscosity at inversion results in a sharp drop in amperage providing a more accurate determination of the inversion or break-point (Crenwelge et al., 1974).

A variety of systems have been used by other researchers although they do not differ substantially from the original method of Swift et al. (1961). The latter procedure was modified by Hegarty et al. (1963) who used a low speed stirrer (1750 rpm) to form emulsions. They used purified muscle protein fractions to determine the effect of myofibrillar (salt-soluble) and sarcoplasmic (water-soluble) proteins upon EC. Conversely, Carpenter and Saffle (1964) employed a high speed blender (13,400 rpm) technique to study the EC of meats from a variety of sources which are normally used in sausage production. Even a microemulsifying assembly has been developed for working with very small quantities of purified proteins (Tsai et al., 1970).

Emulsions formed by different types of equipment are highly variable with gross irregularities being most apparent in fat particle size (Saffle, 1968); making it very difficult to compare data of various studies. Acton and Saffle (1972) recognized the variability in EC data obtained with different model systems and proceeded to derive an oil phase volume factor that appears to be common to all systems. Supposedly, the use of this procedure will allow for a more standard evaluation of data obtained from different studies.

Several reports indicate that model systems are not closely related to in-plant sausage production. Rongey (1965) stated that model systems serve best as a research tool rather than as a means

for evaluating commercially prepared emulsions. Emulsions prepared by model systems are difficult to break and have different textural properties than those normally formed in commercial operations. Trautman (1966) found no correlation between EC and sausage stability. However, Smith et al. (1973) reported a positive correlation between EC and emulsion stability in studying different protein additives. Saffle (1968) emphasized that caution must be employed when data obtained from model systems are compared with sausage emulsions. Nevertheless, information obtained from model systems can be very meaningful in screening new sources of proteins and to gain some foresight about contemplated changes in current meat emulsion processing procedures (Sulzbacher, 1973).

The effect of protein concentration on EC has been studied by several investigators. A number of reports (Swift et al., 1961; Hegarty et al., 1963; Sulzbacher, 1973) have shown that the efficiency of the salt-soluble proteins to emulsify fats varies curvillinearly with protein concentration. Saffle (1968) reported that the relationship between the concentration of salt-soluble protein and EC is a straight line function when tested with the model system of Carpenter and Saffle (1964). However, others (Maurer et al., 1969; Ivey et al., 1970) have stated that EC is inversely related to protein concentration under certain conditions (i.e., increasing shear forces). Borton et al. (1968) indicated that some of the variations in EC as

related to protein concentration might be due to the basis by which EC is expressed. Some of this confusion might be clarified by application of the results of Acton and Saffle (1972) who derived a common factor (oil phase volume) for analyzing EC data compiled by various researchers.

Other parameters that influence EC also have been studied. Swift et al. (1961) found a straight line correlation between the rate of oil addition and EC while Carpenter and Saffle (1964) did not. Crenwelge et al. (1974) reported that oil addition rates did not influence the EC of soy, globin and cottonseed proteins but did affect that for nonfat dry milk.

The effect of shear force on EC has been studied by numerous workers (Swift et al., 1961; Carpenter and Saffle, 1964; Ivey et al., 1970; Crenwelge et al., 1974). They all report that increasing the shear force results in a reduction of the amount of oil emulsified.

EC varies inversely with temperature (Swift et al., 1961; Carpenter and Saffle, 1964). It has been suggested (Saffle, 1968) that rigid control of emulsion temperature is more critical than any of the other factors studied.

Emulsion Proteins

The EC of several types of proteins has been determined. Hansen (1960) reported the salt-soluble proteins of meat to be good

emulsifiers of fat whereas the water-soluble proteins were not. In a more definitive study, Hegarty et al. (1963) examined various muscle protein fractions and reported the highest to the lowest EC as follows: actin in absence of salt, myosin, actomyosin, sacroplasmic proteins and actin in 0.3 M salt solution. Tsai et al. (1972) found myosin to have greater EC than either actin or sarcoplasmic proteins. From a practical production standpoint, several reports (Swift et al., 1961; Trautman, 1964; Carpenter and Saffle, 1965) have shown the EC of the salt-soluble proteins to be superior to that of the water-soluble proteins.

The ability of meat proteins to emulsify fat appears related to the size and shape of the protein molecule and the number of charges or hydrophilic groups exposed on the exterior surface. Myosin, a long rod-like molecule, has considerably more surface area than other muscle proteins which allows it to form membranes around lipid droplets (Carpenter and Saffle, 1965). DuBois et al. (1972) treated muscle proteins with hydrolytic enzymes and found that while limited proteolysis increased EC, prolonged hydrolysis caused a reduction. This work indicates that EC is closely related with the configuration or the optimum mean molecule size of protein molecules. The hydrophilic groups ionize forming effective repulsion between protein-coated droplets preventing coacervation to enhance emulsion stability (Giese, 1968).

Pearson et al. (1965) determined the EC of potassium caseinate and nonfat dry milk (NFDM). The former had greater EC at higher protein concentrations while NFDM had higher EC at lower protein levels. Morr et al. (1973) reported that different preparations of whey protein concentrate had similar emulsifying capacities. Kuehler and Stine (1974) ranked the EC of various milk protein preparations in descending order as follows: whey protein concentrate, NFDM and casein. Crenwelge et al. (1974) recently published the optimum EC conditions for comparing NFDM with other proteins.

Polyphosphates

Phosphates and polyphosphates have been widely used in the food industry for many years. These compounds have a beneficial effect on the functional properties of different food systems of which they become a part (Deman and Melnychyn, 1971).

Most of the research completed with meat proteins has been related to the effect of polyphosphates on the water holding capacity or hydration of cured meats (Hamm, 1971; Shults et al., 1972; Brotsly and Everson, 1973). Addition of polyphosphates to comminuted meat products increases the amount of salt-soluble proteins solubilized at an optimum NaCl level of 2.5 to 4% and 0.35 to 0.5% retained polyphosphate (Mahon et al., 1971). Some of the earlier research data concerning the effects of different phosphate salts on

sausage-like products do not agree with the properties and functions of phosphates as they are now known. Hellendoorn (1962) suggested that these differences might well be due to variations in the NaCl concentrations and ionic strengths of extractants used by different workers.

Addition of polyphosphates to sausage-type products results in a finer-textured, more stable emulsion as evidenced by a reduction in both fluid and fat losses during the subsequent heat-pasteurization treatment. Moreover, finished products containing polyphosphates have a greater tensile strength and withstand more physical abuse than those processed without the additive (Swift and Ellis, 1957).

Interactions between phosphates and meat proteins are not yet clearly understood in spite of intensive research in this area (Hamm, 1971). However, Swift and Sulzbacher (1963) reported that the EC of water-soluble proteins increased in the presence of different anions, particularly the Cl^- ion. They suggested that anions have some ability to enhance the unfolding of protein molecules thus providing more surface area for encapsulating fat globules. Polyphosphates are considered to be potent solubilizers since they exert a dissociating effect on certain muscle protein complexes formed during rigor (Wismer-Pedersen, 1971).

The general opinion of most researchers is that polyphosphates influence the hydration of meat proteins in three ways: (1) by

increasing pH to move proteins away from their isoelectric points; (2) by increasing the ionic strength which is a nonspecific effect based on concentration and electrical charges of the salt ions; and (3) specific effects which are due to certain interactions between the phosphate anion and proteins (Hamm, 1971; Mahon et al., 1971). The above effects may also influence the EC although much more work is necessary to clarify this situation.

The dairy processing industry has made extensive use of various phosphate salts for many years. These compounds were utilized to prevent undesirable changes occurring in products subject to high temperature - short time (HTST) processing procedures (Ellerston and Pearce, 1964).

The major casein components (α_s -, β -, and K-caseins) contain many phosphate groups that are capable of binding cations present in milk (Melnychyn and Wolcott, 1971). They also stated that the ability of casein to resist heat denaturation is likely related to phosphate action.

HTST sterile concentrated milk products gell upon storage at ambient temperature. This defect is prevented by the addition of a polyphosphate (i.e., tetrapolyphosphate) prior to heat treatment at a level of 0.56% of the milk solids. Although polyphosphates have a pronounced effect upon proteins comprising casein micelles, details are lacking with respect to interaction with specific proteins

(Carroll et al., 1968). Laboratory studies carried out at room temperature by Melnychyn and Wolcott (1965) indicated that only the long chain phosphates, tetrapolyphosphate and hexametaphosphate, have substantial interaction with milk proteins, particularly the K- and α_s-casein fractions. These authors later concluded that the mode of interaction of milk proteins with phosphates must be considered as electrostatic in character (Melnychyn and Wolcott, 1971).

The application of phosphates has increased the utilization of cheese whey proteins. These proteins can be removed undenatured from cheese whey by adjusting the system to low pH followed by isoelectric precipitation with small amounts of polyphosphate. This procedure permits the recovery of undenatured whey proteins that were previously collected in a heat-denatured form. Denatured whey proteins are completely insoluble and have only limited use in food manufacture (Wingerd, 1971).

Endo (1963) achieved promising results in adding sodium polyphosphate to whey and other milk components that were used in the preparation of emulsion-like products.

Sodium acid pyrophosphate ($Na_2H_2P_2O_7$), sodium tripolyphosphate ($Na_5P_3O_{10}$) and sodium hexametaphosphate ($NaPO_{3n} \cdot Na_2O$) are the polyphosphates commonly used in the food industry (Brotsky and Everson, 1973). Blending of these compounds yields better results with many food systems than use of a singular polyphosphate

alone. Moreover, these polyphosphates are generally recognized as safe for food use (Mahon et al., 1971).

Research Objective

As mentioned initially, the world's food protein shortage is a serious problem. Thus, several research approaches must be undertaken to resolve the situation. Sausage or emulsion-type products offer one possible means of producing highly acceptable, nutritious foods utilizing proteins from a variety of sources; many of which are not currently used in food manufacture or are not used efficiently.

Emulsification is a primary functional property for determining the utility of incorporating a new protein source into sausage emulsions. Also, the use of certain food additives (i.e., polyphosphates) may enhance the functionality of proteins, particularly those that are lacking or marginal in this respect. Since meat and milk proteins are relatively expensive food components, information about extending their capabilities in emulsions would be highly desirable.

In view of the fragmented reports in the literature, this investigation was designed to study the influence of a food grade polyphosphate blend upon the emulsification of meat and milk proteins.

A model system simulating emulsion conditions was used to collect experimental data.

EXPERIMENTAL PROCEDURE

The milk proteins, casein and lactalbumin, and the extractable salt-soluble proteins of bovine muscle tissue were the proteins used in this study.

Preparation of Milk Protein Solutions

Salt-free casein and lactalbumin powders, both enzymatically hydrolyzed preparations, were purchased from Sigma Chemical Co., St. Louis, MO.

Stock solutions containing 15 mg of protein/ml were prepared by dissolving the appropriate amounts of either casein or lactalbumin in 3% NaCl. Total nitrogen of the stock solutions were determined by the macro-Kjeldahl procedure (A.O.A.C., 1960). Total nitrogen content was multiplied by the factor, 6.38, to calculate the protein content. Subsequent dilutions were made from the stock solutions as needed.

Preparation of Meat Extracts

A section of longissimus dorsi muscle was obtained from each of three different carcasses, five days post-mortum. Carcasses were from animals processed at the Meat Science Laboratory, Oregon State University. Each muscle tissue section was divided

into three lots: lot A which was used in the fresh state; lot B was frozen and held at -15°C for three days then thawed overnight at ambient temperature; and lot C was frozen, held at -15°C for three days, thawed overnight at room temperature, refrozen and held at -15°C for two additional days before thawing as before.

A procedure similar to that of Saffle and Galbreath (1964) was used to extract the salt-soluble meat proteins. Two hundred-fifty g of lean diced (2.5 cm^3) muscle tissue was comminuted with 500 ml of 3% NaCl at maximum speed for 3 min in an Osterizer blender. The homogenate was centrifuged at 27,000 X g for 15 min at 3°C. The supernatant was filtered through glass wool and centrifuged a second time. After filtering again through glass wool, the extract was collected and analyzed for protein content by the biuret procedure of Torten and Whitaker (1964). Two 1-ml aliquots of each meat extract were mixed with 4 ml of biuret reagent. Exactly 30 min later the absorbance was read against a water-reagent blank at 540 nm in a Beckman Model B spectrophotometer. Protein values were obtained from a standard curve (Appendix I) prepared by plotting absorbance (540 nm) against protein content (mg/ml) using bovine serum albumin, A-grade, purchased from Calbiochem, La Jolla, CA. as the reference protein. Stock solutions were prepared by diluting the meat extracts with 3% NaCl to a standard protein concentration of 7.5 mg/ml. Further dilutions were made as required.

Polyphosphate Treatments

Granular food grade polyphosphates were supplied through the courtesy of Monsanto Chemical Co., St. Louis, MO. Sodium acid pyrophosphate, sodium tripolyphosphate and sodium hexametaphosphate were mixed together in a weight ratio of 4:2:4, respectively. This mixture is hereafter referred to as the polyphosphate blend.

To evaluate the effects of polyphosphates on the emulsification of milk and meat proteins, 0.5% (w/v) of the polyphosphate blend was added to the proteins dissolved in 3% NaCl.

Emulsification of casein and lactalbumin solutions with or without polyphosphates was determined in quintuplicate, two replications per treatment, at four different protein concentrations (2, 5, 10 and 15 mg/ml). The casein and lactalbumin solutions were each adjusted to pH 6.0 with 0.1 N HCl prior to the emulsification determinations.

Emulsification of the meat protein extracts with or without polyphosphates was determined in quintuplicate, three replications per treatment, at three different protein concentrations (2, 5 and 7.5 mg/ml). All meat extracts were adjusted to pH 5.7 with 0.1 N HCl prior to the emulsification determinations.

Emulsification

The emulsifying characteristics of the different proteins under study were determined by the model system of Carpenter and Saffle (1964). This system consisted of a Osterizer blender base coupled in series with a rheostat. Rotational frequency was standardized to 10,000 ± 200 rpm with a Strobatac stroboscope (Model 631-B). A 4-bladed cutter assembly was screwed onto a 250 ml jar and inverted onto the blender base. The opposite end of the jar had two holes; a 12-mm hole in the center and a 5-mm hole off to one side.

Twenty-five ml of protein solution was pipetted into the jar followed by the addition of 25 ml of cottonseed oil. The jar plus contents were held under refrigeration until the temperature of the mixture equilibrated to 15°C. The mixture was blended for 30 sec to form the initial emulsion after which, oil was added at a rate of 0.9 ml/sec from a separatory funnel connected to the jar by tygon tubing inserted through the 12-mm hole. Food grade cottonseed oil was purchased locally for use as the lipid source.

The break point (emulsion collapse) was determined by the electrical resistance method similar to that described by Webb et al. (1970) and modified by Satterlee et al. (1973). The direct current (DC) resistance of the emulsion was measured by immersing a constant sized copper wire as an electrode into the emulsion through

the 5-mm hole of the jar. The other electrode was connected to the cutter assembly base. The DC resistance of the emulsion was monitored with a Hiaki ohm meter (Model TH-L33) to which the electrodes were attached. During emulsion formation and subsequent emulsification, DC resistance ranged between 30-40 Kohm whereas the DC resistance rapidly rose to infinity as the emulsion collapsed (break point).

The amount of oil emulsified was the difference between the weight of the jar containing the protein extract before and after the addition of oil. It was assumed that the oil and protein solutions each had a weight of 1 mg/ml. The results are expressed and described subsequently on the basis of "emulsified volume" (EV) which is the g of total oil emulsified by the 25 ml of protein solution (Acton and Saffle, 1972).

For comparative purposes, the results are also presented briefly in two other ways: (1) "emulsifying capacity" (EC) which is based on the g of oil emulsified per 100 mg of soluble protein (Carpenter and Saffle, 1964); and (2) the "oil phase volume" (OPV) which is the percent oil weight of the total emulsion weight, i.e., g of added oil phase plus g of aqueous phase (Acton and Saffle, 1972).

Statistical Analysis

The EV data were statistically evaluated by the analysis of

variance procedure following the advice and guidance of Dr. Roger Peterson, Department of Statistics, Oregon State University.

RESULTS AND DISCUSSION

As indicated on the previous page, results of emulsification studies are expressed in one of three ways: (1) "emulsified volume" (EV), total g of oil emulsified; (2) "emulsifying capacity" (EC), g of oil emulsified/100 mg protein; and (3) "oil phase volume" (OPV), percent oil in total emulsion volume. EC was the term used in most of the early reports. In 1972, the terms EV and OPV appeared. Many recent reports have used the OPV term almost exclusively. In case of the latter, however, several reports have contained insufficient data and details to convert the results to the other forms for direct comparison with data of previous studies. Since it is relatively easy to convert EV data to either of the other two forms, results of this study are presented and discussed primarily on the basis of emulsified volume (EV). The emulsifying capacity (EC) and oil phase volume (OPV) data are only shown graphically.

Emulsification of Casein

The results of adding 0.5% (w/v) of the polyphosphate blend on the emulsified volume (EV) of casein dissolved in 3% NaCl, pH 6.0, at protein concentrations ranging from 2 to 15 mg/ml are summarized in Table 1. These data are also shown graphically in Figure 1 along with those plotted on the basis of emulsifying

Table 1. Effect of polyphosphates (PP) on emulsified volume (EV) of casein.

Protein conc. (mg/ml)	EV[a]					
	Rep. 1 (g)		Rep. 2 (g)		Mean (g)	
	Control	PP	Control	PP	Control	PP
2	25.0 ± 0.0	25.0 ± 0.0	25.0 ± 0.0	25.0 ± 0.0	25.0	25.0
5	25.7 ± 0.9	31.8 ± 2.0	26.4 ± 2.0	31.0 ± 1.6	26.1	31.4
10	31.8 ± 2.0	36.2 ± 2.7	31.7 ± 0.9	36.0 ± 1.6	31.8	36.2
15	36.1 ± 1.3	39.8 ± 0.8	37.4 ± 4.4	40.6 ± 1.5	36.8	40.2

[a]The mean of five determinations.

LSD (.05) = 1.512
LSD (.01) = 2.008

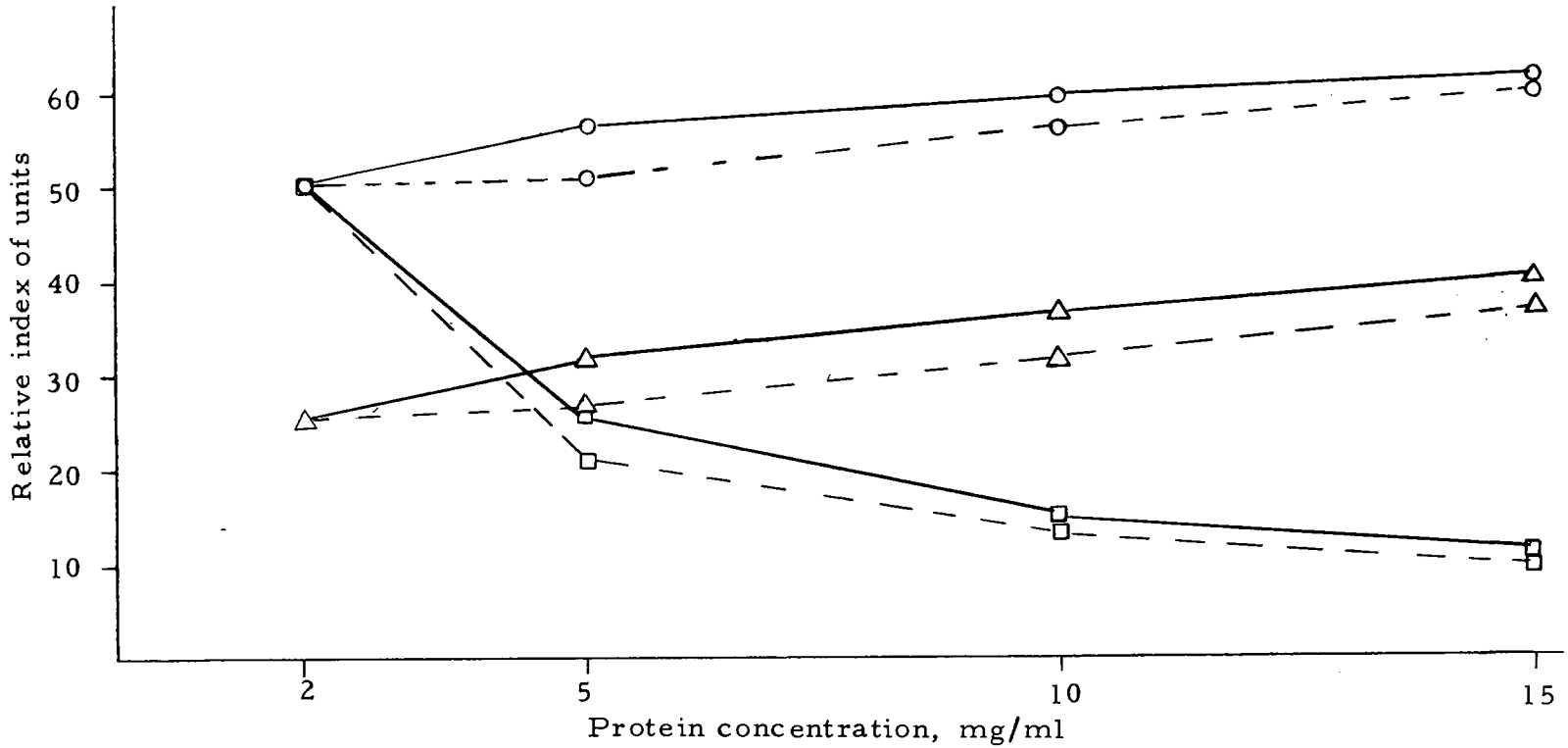


Figure 1. The effect of polyphosphates (-) on emulsifying capacity (□ , g oil emulsified/100 mg protein), emulsified volume (△ , total g oil emulsified) and oil phase volume (○ , percent oil in the total emulsion volume) of casein. Control (--).

capacity (EC) and oil phase volume (OPV).

Figure 1 shows that the addition of the polyphosphate blend enhanced the emulsification characteristics (i.e., EV, EC, OPV) of casein when the protein concentration exceeded 2 mg/ml. The EV's of polyphosphate-treated casein solutions containing 5, 10 and 15 mg protein/ml were significantly ($P < 0.01$) higher than those of the controls (Table 2). The polyphosphate blend had no apparent effect upon the emulsification ability of casein at a protein level of 2 mg/ml. Table 2 indicates that the effect of the polyphosphate blend was related to the protein concentration. The enhancing effect of phosphates on EV decreased concurrently with the decline in the phosphate:protein ratio (Figure 1). Differences in EV between the phosphate and no phosphate treatments were 5.3, 4.4 and 3.4 g of total oil emulsified for the casein protein concentrations of 5, 10 and 15 mg/ml, respectively. However, at the lowest casein level (2 mg/ml) the polyphosphate blend had no effect on EV although the phosphate:protein ratio was the highest at this protein concentration.

When these results were expressed on the basis of OPV or EC, data plotted in Figure 1 show that OPV increased while EC decreased with increasing protein concentrations. These findings agree with those reported by other investigators (Swift et al., 1961; Carpenter and Saffle, 1964; Pearson et al., 1965; Acton and Saffle, 1972; Crenwelge et al., 1974).

Table 2. The analysis of variance of the effects of polyphosphates on the emulsified volume of casein.

Source	d.f.	Sum of Squares	Mean Square	F
Phosphates	1	229.84	229.84	77.53 **
Protein conc.	3	2062.92	687.64	239.18 **
Replication	1	1.92	1.92	0.68
Phosp. X Prot.	3	87.76	29.52	9.34 **
Error	71	204.12	2.87	
Total	79	2586.56		

** ($P < 0.01$)

The EC values found in this work, 9.8 to 50 g of oil/100 mg protein at protein concentrations of 15 and 2 mg/ml, respectively, were lower than values reported by others. Pearson et al. (1965) reported the EC of potassium caseinate (pH 6.3, ionic strength of 0.3) as g of oil/mg nitrogen. By converting their data to g of oil/100 mg protein, their EC values would range from about 206 to 506 when tested at protein levels of 6 and 2 mg/ml, respectively.

The lower EC values obtained in this study might be explained in part by the findings of DuBois et al. (1972) and Kuehler and Stine (1974). They found the EC to decrease by increasing the extent or length of enzymatic hydrolysis of proteins prior to the emulsification determination. The casein used in the present study was purchased as an enzymatic hydrolyzate in order to obtain a reasonably pure preparation possessing good solubility properties. Native casein is a complex phosphoprotein of high molecular weight requiring phosphorus for its integrity. Splitting native casein enzymatically may result in the formation of many fragments which collectively may have less ability to emulsify lipids than the intact casein particles.

From this study, it is difficult to speculate on the type of protein-phosphate interaction occurring upon the addition of the polyphosphate blend to cause an increase in the emulsification characteristics of the casein hydrolyzate. Perhaps, an electrostatic-type interaction occurred between the polyphosphates and casein

(Melnychyn and Wolcott, 1973) to result in some aggregation of the casein fragments to form more appropriate sized particles conducive for the emulsification of lipids.

Emulsification of Lactalbumin

The effect of adding 0.5% (w/v) of the polyphosphate blend on the EV of lactalbumin dissolved in 3% NaCl, pH 6.0, at protein concentrations ranging from 2 to 15 mg/ml are summarized in Table 3. These data are plotted in Figure 2 along with those for EC and OPV. Figure 2 shows that the polyphosphate addition depressed the emulsification characteristics (EV, EC, OPV) of lactalbumin at each level of protein concentration tested. The EV's of the polyphosphate-treated lactalbumin solutions containing 5, 10 and 15 mg protein/ml were significantly ($P< 0.01$) lower than those of the controls (Tables 3 and 4). Data in Table 4 indicate the occurrence of an interaction between polyphosphates and protein concentration. The effect of polyphosphates in depressing EV became more pronounced with increasing protein concentration. Differences in EV between the control and polyphosphate treatments were 1.0, 4.8, 6.1 and 6.5 g of total oil emulsified for lactalbumin protein concentrations of 2, 5, 10 and 15 mg/ml, respectively. Thus as the polyphosphate:lactalbumin ratio decreased, the adverse effect of the polyphosphates upon EV increased. Although EV and OPV increased

Table 3. Effect of polyphosphates (PP) on emulsified volume (EV) of lactalbumin.

Protein conc. (mg/ml)	EV[a]					
	Rep. 1 (g)		Rep. 2 (g)		Mean (g)	
	Control	PP	Control	PP	Control	PP
2	27.0 ± 1.8	25.2 ± 0.5	28.0 ± 3.2	27.2 ± 3.2	27.5	26.5
5	35.0 ± 0.5	31.7 ± 1.6	38.2 ± 0.4	31.8 ± 2.5	36.6	31.8
10	47.4 ± 3.3	37.0 ± 0.3	44.0 ± 3.9	42.1 ± 3.7	45.7	39.6
15	51.2 ± 1.2	46.0 ± 4.8	52.6 ± 4.4	44.7 ± 2.4	51.9	45.4

[a] The mean of five determinations.

LSD (.05) = 2.621
LSD (.01) = 3.480

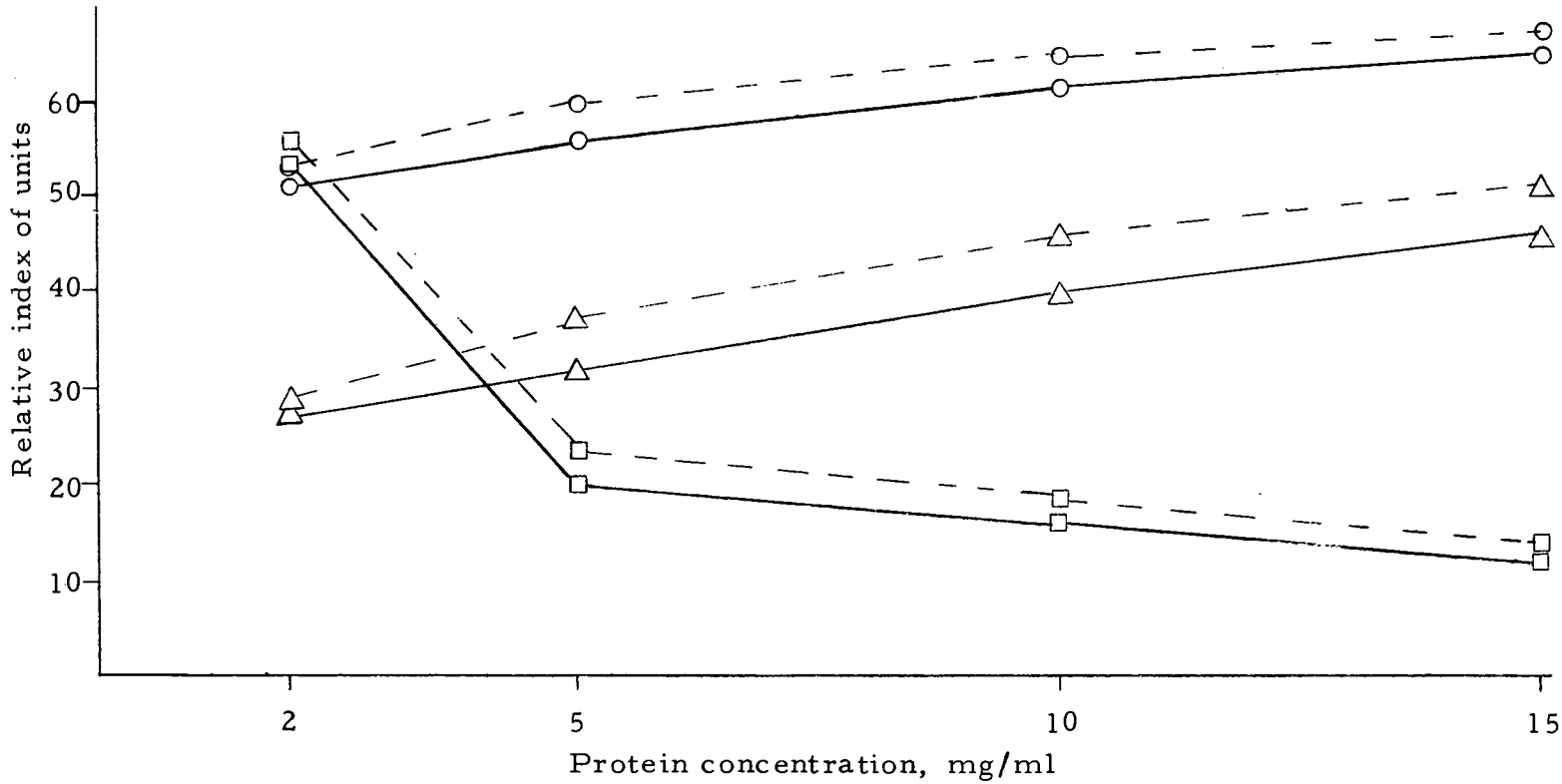


Figure 2. The effect of polyphosphates (-) on emulsifying capacity (□ , g oil emulsified/100 mg protein), emulsified volume (△ , total g oil emulsified) and oil phase volume (○ , percent oil in the total emulsion volume) of lactalbumin. Control (--).

with increasing protein concentration, EC decreased. This trend was similar to that found with casein.

Table 4. The analysis of variance of the effects of polyphosphates on the emulsified volume of lactalbumin.

Source	d. f.	Sum of Squares	Mean Square	F
Phosphates	1	427.35	427.35	49.45 **
Protein conc.	3	5412.13	1804.04	208.85 **
Replication	1	23.00	23.00	2.66
Phosp. X Prot.	3	93.05	31.02	3.59 *
Error	71	613.23	8.64	
Total	79	6568.83		

* ($P < 0.05$)
** ($P < 0.01$)

The above values are lower than those reported for native lactalbumin. Morr et al. (1973) reported EV's ranging from 34 to 66 when lactalbumin was tested at a protein level of 1 mg/ml. In this study (Figure 2), EV's of the lactalbumin tested without polyphosphates ranged from 26.8 to 45.4 at protein concentrations of 15 and 2 mg/ml, respectively. An EC (g of oil/100 mg protein) of 298 was recorded by Keuhler and Stine (1974) for lactalbumin tested at a protein level of 0.25 mg/ml. This is much higher than an EC of 53 determined for the lactalbumin tested in this study at a protein concentration of 2 mg/ml. As was noted with casein,

the enzymatically hydrolyzed lactalbumin had less ability to emulsify lipids than the native material.

The adverse effects of polyphosphates on the emulsification characteristics of lactalbumin were unexpected since these compounds are commonly used to improve protein dispersion in food systems (Ellinger, 1971). Since the dispersed phase in an emulsion is protected by a layer of soluble proteins, the particular polyphosphate blend used in this study may have reacted unfavorably with lactalbumin to reduce its availability at the oil-water interface.

Spinelli and Koury (1970) reacted various polyphosphates with soluble fish muscle proteins under acidic conditions. They found that the polyphosphates and proteins formed complexes having low solubilities in a pH range of 2.0 to 4.5. Although pH 6.0 was used in the present system, some polyphosphate-lactalbumin complexes may have been formed which reduced the amount of protein available to protect the oil-droplets at the emulsion interface.

Emulsification of Meat Proteins

The influence of adding 0.5% (w/v) of the polyphosphate blend on the EV of meat proteins solubilized in 3% NaCl, pH 5.7, at protein concentrations of 2, 5 and 7.5 mg/ml are summarized in Table 5. These data are shown in Figure 3 with those pertaining to EC and OPV. As shown in Figure 3, the polyphosphate blend had

Table 5. The effect of polyphosphates (PP) on the emulsified volume (EV) of meat extracts.

Sample	Protein conc. (mg/ml)	EV[a] Rep. 1 (g) Control	Rep. 1 (g) PP	Rep. 2 (g) Control	Rep. 2 (g) PP	Rep. 3 (g) Control	Rep. 3 (g) PP	Mean (g) Control	Mean (g) PP
Lot A (fresh)	2.0	48.8 ± 1.8	49.4 ± 2.4	51.3 ± 2.7	48.6 ± 2.4	51.3 ± 3.5	51.4 ± 2.3	50.4	49.9
	5.0	56.8 ± 3.1	57.7 ± 1.7	59.1 ± 0.8	60.1 ± 0.9	65.9 ± 1.2	70.4 ± 2.3	60.6	62.7
	7.5	80.0 ± 6.1	85.9 ± 4.2	75.1 ± 3.1	79.3 ± 2.7	93.5 ± 4.9	92.9 ± 6.9	82.9	86.0
Lot B (frozen)	2.0	46.1 ± 0.2	46.7 ± 0.8	48.6 ± 1.9	49.9 ± 1.7	53.5 ± 3.7	52.8 ± 2.8	49.4	49.9
	5.0	56.2 ± 1.7	57.2 ± 1.0	57.8 ± 1.2	60.8 ± 6.8	69.5 ± 0.8	71.0 ± 4.5	61.2	62.9
	7.5	78.6 ± 2.5	80.2 ± 6.1	84.3 ± 1.8	83.3 ± 3.2	90.3 ± 1.3	92.2 ± 1.4	84.4	85.3
Lot C (re-frozen)	2.0	52.7 ± 3.2	51.9 ± 4.7	51.7 ± 2.8	51.5 ± 2.7	50.1 ± 3.6	50.1 ± 2.8	51.6	51.4
	5.0	58.4 ± 2.0	59.6 ± 1.7	67.7 ± 2.1	63.4 ± 1.6	60.6 ± 3.7	64.1 ± 5.5	62.2	62.6
	7.5	96.2 ± 4.8	91.5 ± 5.3	89.4 ± 5.6	88.4 ± 2.3	87.9 ± 5.9	90.1 ± 5.9	91.2	90.0

[a] The mean of five determinations.

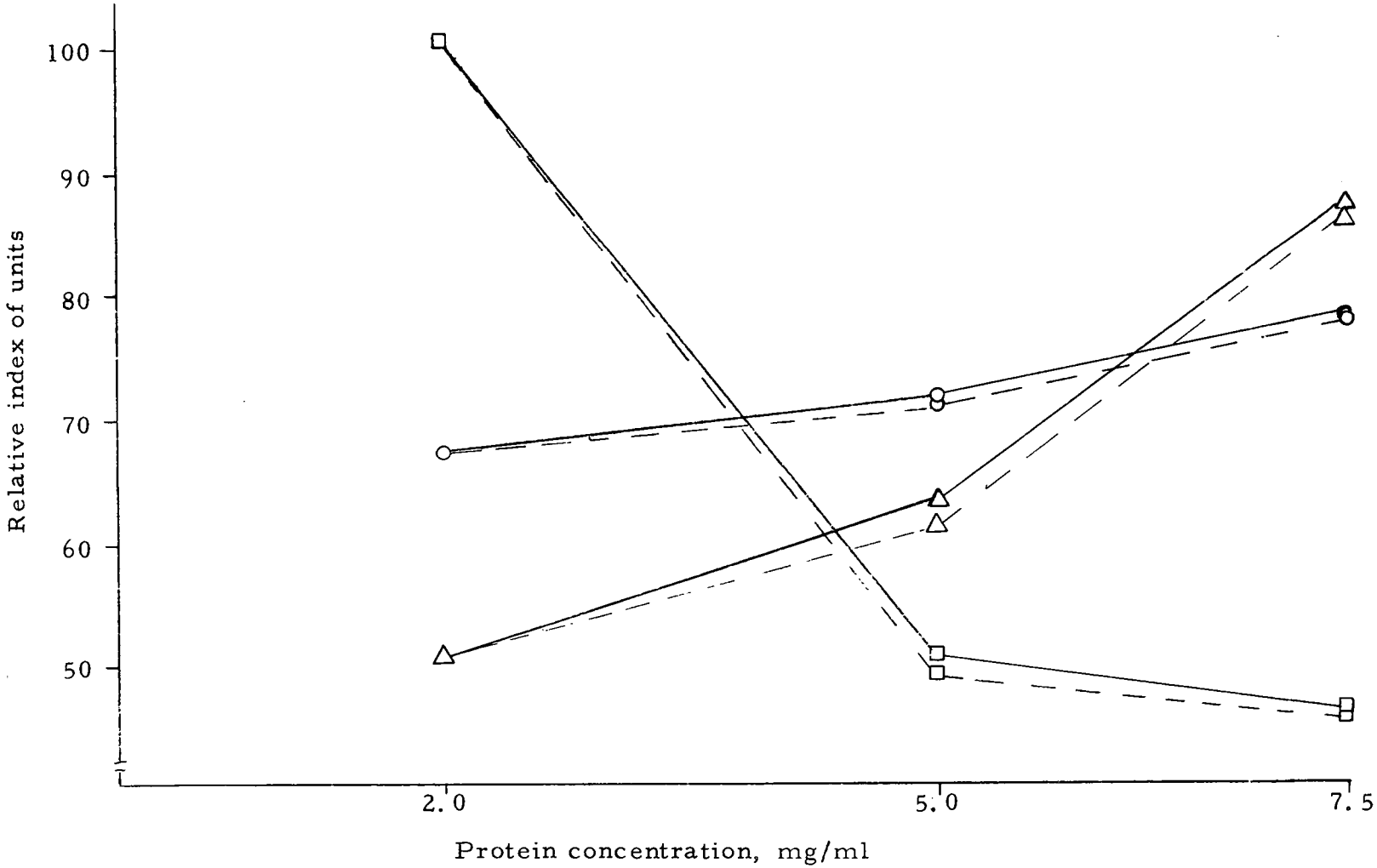


Figure 3. The effect of polyphosphates (-) on emulsifying capacity (□ , g oil emulsified/100 mg protein), emulsified volume (△ , total g oil emulsified) and oil phase volume (○ , percent oil in the total emulsion volume) of meat extract. Control (--).

little effect upon the emulsification characteristics of the meat protein extracts. Differences between the EV's of the control and polyphosphate treatment were not statistically significant ($P>0.05$) at any of the protein concentrations tested (Tables 5 and 6).

Table 6. The analysis of variance of the effects of a mixture of polyphosphates on the emulsified volume of meat extract.

Source	d. f.	Sum of Squares	Mean Square	F
Phosphates	1	41.93	41.93	0.49
Protein conc.	2	61606.17	30803.08	3361.00 **
Replication	2	1732.48	866.24	10.15 **
Freezing	2	420.68	210.34	2.46
Error	34	2901.10	85.32	
Total	269	69889.54		

** ($P < 0.01$)

Figure 3 also shows that EV and OPV increased with increasing protein concentrations while EC decreased. These results are in agreement with data reported by others (Swift et al., 1961; Carpenter and Saffle, 1964; Acton and Saffle, 1972; Sulzbacher, 1973). Graphically, the curves showing the emulsification characteristics of casein (Figure 1) and lactalbumin (Figure 2) are similar in shape and trend to those of the meat extract (Figure 3) although analogous values for the milk proteins are lower. Thus, in this study meat

protein extracts possessed greater emulsification characteristics than either the casein or lactalbumin preparations tested.

Without resorting to a mass of data and a variety of interpretations, it is sufficient to state that the results of the emulsification characteristics (EC, EV, OPV) of the meat protein extract of this study are similar to those of Carpenter and Saffle (1964), Acton and Saffle (1969), and Acton and Saffle (1972) among many others.

Analysis of variance data of Table 6 show that the replication effect was statistically significant ($P < 0.01$). This finding agrees with other reports (Carpenter and Saffle, 1964; Borton et al., 1968) in which the emulsification characteristics were found to vary significantly between different lots of beef trimmings, between different muscles within an animal, and between the same muscle from different animals, etc. Although strict attempts were made to control all of the experimental conditions, the biological variability between animals is an unknown, elusive factor.

The effect of adding the polyphosphate blend to the protein extracts from fresh and frozen meat is shown in Figure 4. To construct this bar graph, data were pooled per handling treatment with and without polyphosphates, then averaged to yield the values illustrated. It can be noted that the polyphosphate-treated protein extracts from fresh meat and meat that had been frozen and thawed once had slightly higher EV's than the control extracts. For the

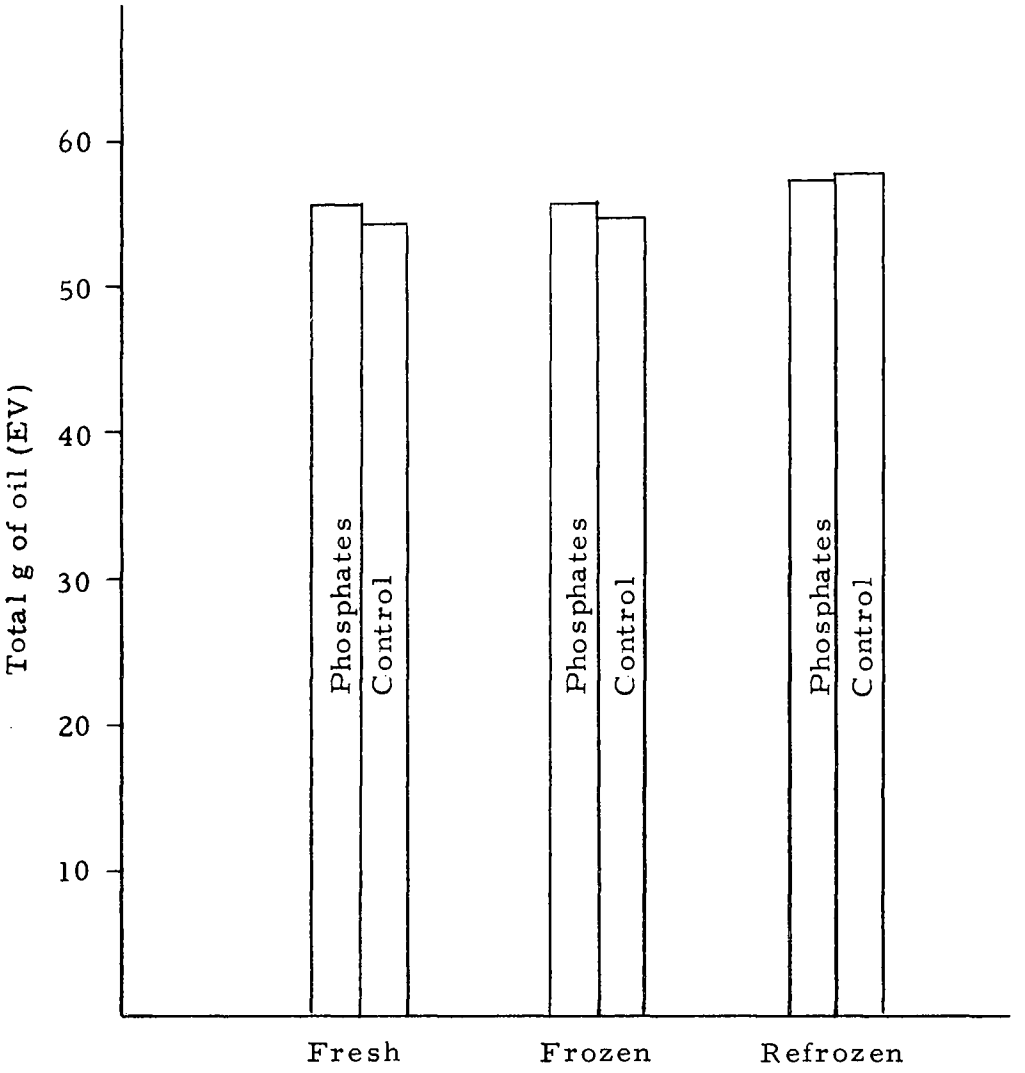


Figure 4. Effect of polyphosphates (PP) on emulsified volume (EV) of meat extracts from frozen treatments.

meat frozen and thawed twice (designated as refrozen), the EV of extracts of the control was slightly higher than that treated with the polyphosphate blend. However, none of these differences was statistically significant (Table 6).

The effect of polyphosphates on meat proteins have been reported to be due to specific changes in pH, ionic strength and protein-phosphate interactions (Hamm, 1971). In this study, the pH was adjusted initially to pH 5.7 to minimize variation in pH. The ionic strength of the protein solutions was not rigorously calculated but the approach of Brotsky and Everson (1973) was followed. They reported that the polyphosphates provide about the same degree of ionic strength as NaCl on a weight percent basis. Thus the ionic strength (I) was calculated according to the formula

$$I \simeq \frac{1}{2} \Sigma M_i Z_i^2$$

where M_i is the molarity of the ion and Z_i^2 is the total charge of the ion (Segel, 1968). With this approach, a 3% NaCl solution had an I = 0.52 while a 3% NaCl solution containing 0.5% (w/v) of the polyphosphate blend had an I = 0.60. Apparently, this small increase in ionic strength was not sufficient to increase the emulsification characteristics of the meat extracts treated with the polyphosphate blend.

The interactions of polyphosphates with meat proteins to

decrease cross-linkages between myofilbrillar proteins and to increase electrostatic repulsion between peptide chains have been offered as an explanation for the increased water-holding capacity of emulsion-like products treated with polyphosphates in the presence of salt (Hamm, 1971). DuBois et al. (1972) and Keuhler and Stine (1974) have shown that the size and shape of protein molecules protecting the dispersed phase of an emulsion do influence the emulsification characteristics. Results of this study, however, show that the addition of the polyphosphate blend had no effect, neither beneficial nor detrimental, upon the emulsification characteristics of meat proteins. Although polyphosphates increase the initial extractability of proteins from muscle tissue (Hamm, 1971), they did not affect the solubilized proteins insofar as emulsification characteristics were concerned as tested under the conditions described in this study.

SUMMARY AND CONCLUSIONS

The influence of polyphosphates upon the emulsification of casein, lactalbumin and salt-soluble meat proteins was determined by a model system.

A blend of polyphosphates was added at a level of 0.5% (w/v) to casein and lactalbumin dissolved in 3% NaCl, pH 6.0. Casein and lactalbumin were enzymatically hydrolyzed preparations. The polyphosphate blend was also added at the above level to meat proteins solubilized in 3% NaCl, pH 5.7. Emulsified volume (EV), total g of oil emulsified per 25 ml of protein solution, was determined for the above proteins with and without polyphosphates at varying protein concentrations. Data were also graphically expressed as emulsifying capacity (EC), g of oil emulsified/100 mg protein, and as oil phase volume (OPV), percent oil in total emulsion volume.

For all proteins studied with or without polyphosphates, the EV and OPV increased with increasing protein concentration whereas EC decreased.

Addition of the polyphosphate blend to casein solutions containing protein levels in excess of 2 mg/ml resulted in significantly ($P< 0.01$) higher EV's than those of the controls. Conversely, the EV's of the polyphosphate-treated lactalbumin solutions were

significantly ($P < 0.01$) lower than those of the controls at all protein levels tested. The opposing EV data obtained with these two proteins may be due, in part, to variations and differences in the size and shape existing between the casein and lactalbumin molecules or fragments at the time of emulsification. However, it is more likely that certain unidentified but specific protein-phosphate interactions occurred in a manner different with casein than with lactalbumin enhancing the EV of the former while depressing the latter.

Addition of the polyphosphate blend had no effect upon the emulsification of the meat protein extracts obtained from fresh, frozen or refrozen samples. Although polyphosphates are known to increase the extractability of proteins from muscle tissue, they apparently do not modify, react nor exert any measurable influence upon the solubilized proteins insofar as emulsification is concerned in a model system.

So far as future research is concerned, results of this study readily suggest that additional work should be completed on the use of polyphosphates with meat proteins to determine whether the emulsification properties might be enhanced: (1) by addition of polyphosphates at the time of protein extraction; and (2) by changing the weight ratio composition of the three polyphosphates comprising the final blend.

With respect to milk proteins, it would be very interesting to learn how or why the polyphosphate blend enhanced the emulsification of the casein hydrolyzate but depressed that of lactalbumin.

BIBLIOGRAPHY

Acton, J. C. and R. L. Saffle. 1969. Preblended and prerigor meat in sausage emulsions. Food Technol. 23:367-371.

Acton, J. C. and R. L. Saffle. 1972. Emulsifying capacity of muscle protein: phase volumes at emulsion collapse. J. Food Science 37:904-906.

A.O.A.C. 1960. "Official Methods of Analysis," 9th ed., p. 643. Association of Official Agricultural Chemists, Washington, D. C.

Becher, P. 1965. "Emulsions: Theory and Practice," pp. 104-123. Reinhold Publishing Corp., New York, N. Y.

Borchert, L. L., M. L. Greaser, J. C. Bard, R. G. Cassens and E. J. Briskey. 1967. Electron microscopy of a meat emulsion. J. Food Science 32:419-421.

Borton, R. J., N. B. Webb and L. J. Bratzler. 1968. Emulsion capacities and emulsion stability of dilute meat slurries from various meat trimmings. Food Technol. 22:506-508.

Brotsky, E. and W. C. Everson. 1973. Polyphosphate use in meat and other muscle foods. In: "Proceedings of the Meat Industry Research Conference," pp. 107-118. American Meat Institute Foundation, Chicago, Ill.

Carpenter, J. A. and R. L. Saffle. 1964. A simple method of estimating the emulsion capacity of various sausage meats. J. Food Science 29:774-781.

Carrol, R. J., M. P. Thompson, R. T. Boswell and P. Melnychyn. 1968. Gelation of concentrated skim milk and electron microscope appearance. J. Dairy Sci. 51:932 (Abstr.)

Crenwelge, D. D., C. W. Dill, P. T. Tybor and W. A. Landman. 1974. A comparison of the emulsification capacities of some protein concentrates. J. Food Science 39:175-177.

Deman, J. M. and P. Melnychyn. 1971. "Phosphates in Food Processing," pp. vii. AVI Publishing Co. Inc., Westport, Conn.

DuBois, M. W., A. F. Anglemier, M. W. Montgomery and W. D. Davidson. 1972. Effect of proteolysis on the emulsification characteristics of bovine skeletal muscle. J. Food Science 37:27-28.

Ellertson, M. E. and S. J. Pearce. 1964. Some observations on the physical-chemical stability of sterile concentrate milks. J. Dairy Science 47:564-569.

Ellinger, H. 1972. Phosphates in food processing. In: "Handbook of Food Additives," ed. T. E. Furia, pp. 617-780. Chemical Rubber Co., Cleveland, Ohio.

Endo, N. 1963. Mayonnaise-like products. In: Chemical Abstracts 58:10666 c.

Giese, A. C. 1968. "Cell Physiology," pp. 88-89. W. B. Saunders Co., Philadelphia, Pa.

Hamm, R. 1971. Interactions between phosphates and meat proteins. In: "Symposium-Phosphates in Food Processing," ed. J. M. Deman and P. Melnychyn, pp. 65-82. AVI Publishing Co. Inc., Westport, Conn.

Hansen, L. J. 1960. Emulsion formation in finely comminuted sausage. Food Technol. 14:565-569.

Hegarty, G. R., L. J. Bratzler and A. M. Pearson. 1963. Studies on the emulsifying properties of some intracellular beef muscle proteins. J. Food Science 28:663-668.

Hellendoorn, E. W. 1962. Water-binding capacity of meat as affected by phosphates, I. Influence of sodium chloride and phosphates on the water-retention of comminuted meat at various pH values. Food Technol. 16:119-124.

Ivey, F. J., N. B. Webb and V. A. Jones. 1970. The effect of disperse phase droplet size and interfacial film thickness on the emulsifying capacity and stability of meat emulsions. Food Technol. 24:379-382.

Kuehler, C. A. and C. M. Stine. 1974. Effect of enzymatic hydrolysis on some functional properties of whey protein. J. Food Science 39:379-382.

Mahon, J. H., K. Schlamb and E. Brotsky. 1971. General concepts applicable to the use of polyphosphates in red meat, poultry, and seafood processing. In: "Symposium-Phosphates in Food Processing," ed. J. M. Deman and P. Melnychtn, pp. 158-161. AVI Publishing Co. Inc., Westport, Conn.

Mattil, K. F. 1971. The functional requirements of proteins for foods. J. American Oil Chemists' Society 48:477-480.

Maurer, A. V., R. C. Baker and D. V. Vadehra. 1969. The influence of type of poultry and carcass part on the extractability and emulsifying capacity of salt soluble proteins. J. Poultry Science 48:994-997.

Melnychyn, P. and J. M. Wolcott. 1965. Evidence of specific interactions of inorganic ions with milk proteins. In: "Proceedings 7th Milk Concentrates Conference." U. S. Dept. Agric., Philadelphia, Pa.

Melnychyn, P. and J. M. Wolcott. 1971. Interactions of milk proteins with phosphates. In: "Symposium-Phosphates in Food Processing," ed. J. M. Deman and P. Melnychyn, pp. 49-64. AVI Publishing Co. Inc., Westport, Conn.

Meyer, J. A., W. L. Brown, N. E. Giltner and J. R. Guinn. 1964. Effect of emulsifiers on the stability of sausage emulsions. Food Technol. 22:1139-1142.

Morr, C. V., P. E. Swenson and R. L. Richter. 1973. Functional characteristics of whey protein concentrates. J. Food Science 38:324-330.

Pearson, A. M., M. E. Spooner, G. R. Hegarty and L. J. Bratzler. 1965. The emulsifying capacity and stability of soy sodium proteinate, potassium caseinate, and non-fat dry milk. Food Technol. 19:1841-1845.

Rongey, E. H. 1965. A simple objective test for sausage emulsion quality. In: "Proceedings of the Meat Industry Research Conference," pp. 99-106. American Meat Institute Foundation, Chicago, Ill.

Saffle, R. L. 1968. Meat emulsions. Advan. Food Res. 16: 105-160.

Saffle, R. L. and J. W. Galbreath. 1964. Quantitative determination of salt-soluble protein in various types of meat. Food Technol. 18:1943-1944.

Satterlee, L. D., B. Free and E. Levin. 1973. Utilization of high protein tissue powders as a binder/extender in meat emulsions. J. Food Science 38:306-309.

Segel, I. H. 1968. "Biochemical Calculations," pp. 303-306. John Wiley and Sons, Inc. New York, N. Y.

Shults, G. W., D. R. Russell and E. Wierbicki. 1972. Effect of condensed phosphates on pH, swelling and water-holding capacity of beef. J. Food Science 37:860-864.

Smith, G. C., H. Juhn, Z. L. Carpenter, K. L. Mattil and C. M. Cater. 1973. Efficacy of protein additives as emulsion stabilizers in frankfurters. J. Food Science 38:849-855.

Spinelli, J. and B. Koury. 1970. Phosphate complexes of soluble fish proteins. Their formation and possible uses. J. Agr. Food Chem. 18:284-288.

Sulzbacher, W. L. 1973. Meat emulsions. J. Sci. Food Agr. 24:589-595.

Swift, C. E. and R. Ellis. 1957. Action of phosphates in sausage products. 2. Pilot plant studies of the effect of some phosphates on binding and color. Food Technol. 11:450-456.

Swift, C. E., W. E. Townsend and L. P. Whitnauer. 1961. Comminuted meat emulsions: the capacity of meats for emulsifying fat. Food Technol. 15:468-473.

Swift, C. E. and W. L. Sulzbacher. 1963. Comminuted meat emulsions: factors affecting meat protein as emulsion stabilizers. Food Technol. 17:224-226.

Swift, C. E. 1965. The emulsifying properties of meat proteins. In: "Proceedings of the Meat Industry Research Conference," pp. 78-93. American Meat Institute Foundation, Chicago, Ill.

Torten, J. and J. R. Whitaker. 1964. Evaluation of the biuret and dye-binding methods for protein determination in meats. J. Food Science 29:168-174.

Trautman, J. C. 1964. Fat emulsifying properties of pre-rigor and post-rigor pork protein. Food Technol. 18:1065-1066.

Tsai, R. Y., R. G. Cassens and E. J. Briskey. 1970. Development of a microemulsifier. J. Food Science 35:229-230.

Tsai, R., R. G. Cassens and E. J. Briskey. 1972. The emulsifying properties of purified muscle proteins. J. Food Science 37:286-288.

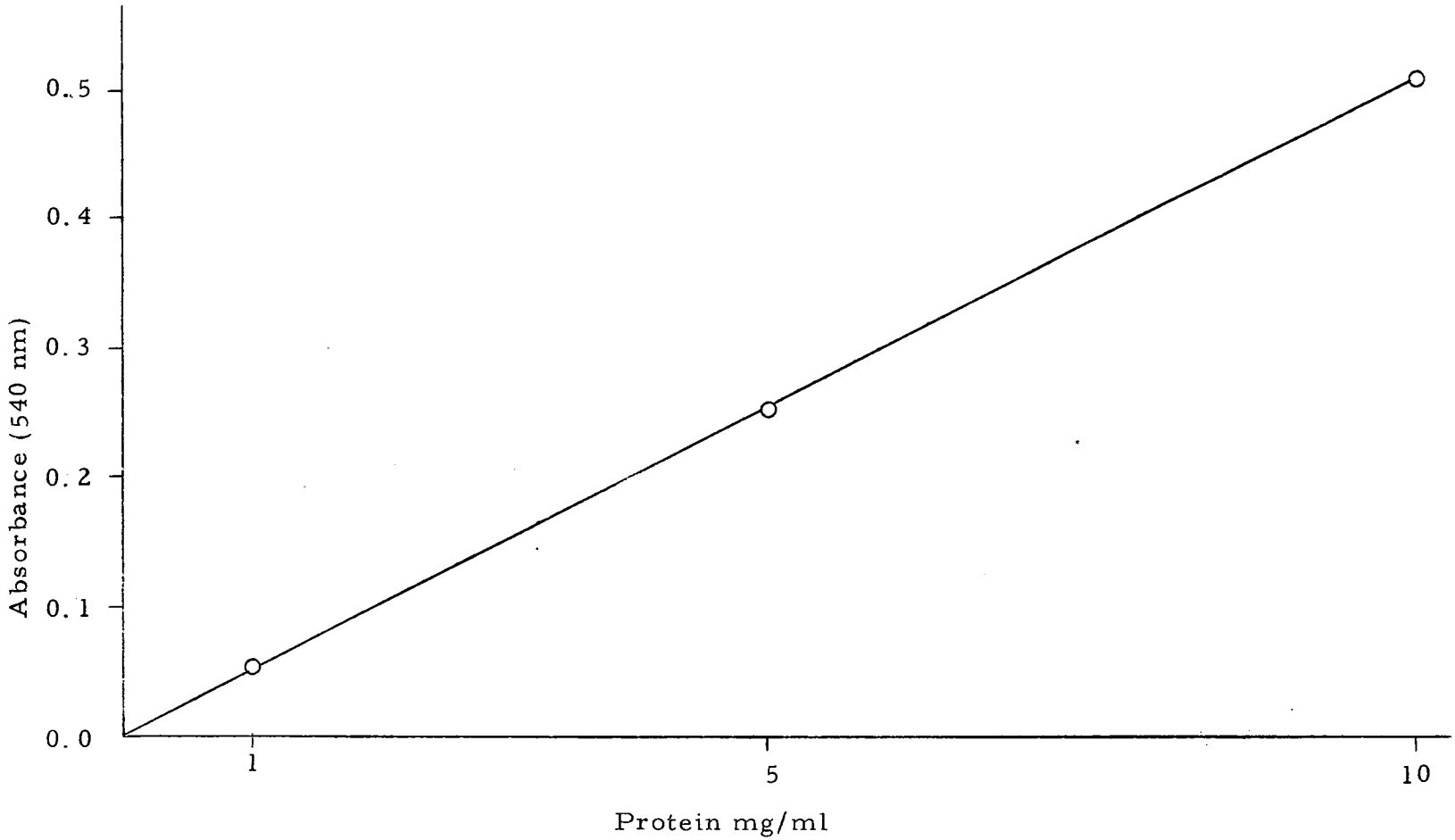
Webb, N. B., F. J. Ivey, H. B. Craig, V. A. Jones and R. J. Monroe. 1970. The measurement of emulsifying capacity by electrical resistance. J. Food Science 35:501-504.

Wingerd, H. W. 1971. Lactalbumin as a food ingredient, J. Dairy Science 54:1234-1236.

Wismer-Pedersen, J. 1971. Chemistry of animal tissues: water. In: "The Science of Meat and Meat Products," ed. J. F. Price and B. S. Schweigert, pp. 184-186. W. H. Freeman Corp., San Francisco, Calif.

Wuhrmann, J. J. 1972. Utilization of novel proteins for human food. Nestle Research News. 1972. (Lausanne, Switzerland), pp. 19-29.

APPENDIX



Appendix I. Biuret standard curve (bovine serum albumin).