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**FAT EXTRACTION FROM MECHANICALLY DEBONED BEEF WITH VARIOUS  
pH AND ALKALIS**

A thesis presented in fulfilment of the requirements for the degree of Master of  
Technology at Massey University, NEW ZEALAND.

SAN-DER WU  
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## ABSTRACT

The study showed that meat surimi with a 1.3% fat content could be prepared from mechanically deboned beef. The process used in the laboratory to prepare the meat surimi was a relatively simple process requiring few unit operations, unit operations which are already used for the manufacture of fish surimi. It is therefore expected that the meat industry would have few problems in preparing meat surimi from mechanically deboned beef given the fact that the technology has already been demonstrated for the commercial production of fish surimi. The key processing steps are firstly the preparation of a mechanically deboned slurry with cold water to assist in the centrifugal removal of the "free" fat present in mechanically deboned meat. The centrifugal separation also removes the sarcoplasmic proteins which could be used for the production of meat flavours, soup stock and possibly pharmaceuticals. The second crucial step in the process is a sieving operation of the myofibrillar/collagen slurry to remove the collagen and "bound" fat from the myofibrillar protein. The subsequent collagen free myofibrillar protein could be concentrated by either further centrifugation or by pressing.

The study also showed that most alkali washes had no significant impact on the fat removal efficiencies of the process, with the possible exception of sodium carbonate, compared to the use of fresh, potable water. It was further demonstrated that it was unnecessary to increase the pH of the wash water beyond a pH of 7.0 as no additional fat separation efficiencies were obtained at the higher pH's. The neutral pH requirements of the process would reduce chemical costs, and possibly also limit equipment wear compared to high wash treatments of pH 9.0 advocated by other researchers. The low pH requirements of the process could also be expected to minimise protein damage which can occur, if held for extended periods at the higher pH's of 9.0 or higher.

The present study has only demonstrated the feasibility of producing meat surimi from mechanically deboned beef. Other uses for the sarcoplasmic and collagen fractions should be established and then a financial feasibility of the whole process should be carried out to establish whether the outlined process is commercially feasible.

## DEDICATION

This thesis is dedicated to my parents, POE CHENG and JER-JIAHN WU, who cared for me, loved me and educated me for the last **thirty years!**

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# CONTENTS

	<u>Page</u>
Title Page	i
Abstract	ii
Dedication & Acknowledgements	iii
Contents Page	iv, v
List of Tables	vi
List of Figures	vii
List of Appendices	viii
<b>1. INTRODUCTION</b>	<b>1</b>
<b>2. LITERATURE REVIEW</b>	<b>3</b>
<b>2.1. Mechanically Deboned Meat</b>	<b>3</b>
2.1.1. Potential production	3
2.1.1.1. Yield	3
2.1.1.2. Machine types and production mechanism	4
2.1.1.3. Utilization in food products	6
2.1.2. Composition	7
2.1.2.1. Proximate composition	7
2.1.2.2. Bone particle and calcium content	9
2.1.2.3. Fatty acid	11
2.1.2.4. Nucleic acid and purine content	12
2.1.2.5. Mineral content	13
2.1.2.6. Cholesterol content	14
2.1.3. Functionality	15
2.1.3.1. Emulsion capacity & stability	15
2.1.3.2. Water holding capacity and texture	16
2.1.3.3. Binding properties	17
2.1.4. Disadvantages & advantages	18
2.1.4.1. Bacterial consideration	18
2.1.4.2. Oxidative rancidity and off flavour	19
2.1.4.3. Nutritional and health implications	20
<b>2.2. Separation of Mechanically Deboned Meat</b>	<b>23</b>
2.2.1. Fat removal techniques	23
2.2.1.1. Supercritical fluid extraction	23
2.2.1.2. Washing procedure	25
2.2.1.3. Solvent extraction	26
2.2.2. Fish surimi	28
2.2.2.1. Basic concepts and history	28
2.2.2.2. The surimi process	29
2.2.2.3. Gel quality evaluation	31
2.2.2.4. Production of surimi-based products	32
2.2.3. Meat surimi	34
2.2.3.1. Why red meat and poultry surimi	34
2.2.3.2. Varieties of meat surimi	35
2.2.3.3. Quality evaluation of meat surimi	39
2.2.3.4. Utilization of meat surimi	41

	<b>Page</b>
<b>2.3. Functional Testing</b>	<b>42</b>
2.3.1. Water holding capacity	42
2.3.2. Gel strength	44
2.3.3. Protein solubility	46
<b>2.4. Rationale for Present Study</b>	<b>47</b>
<b>3. MATERIALS &amp; METHODS</b>	<b>49</b>
<b>3.1. Raw Materials</b>	<b>49</b>
<b>3.2. Analytical Techniques</b>	
3.2.1. Proximate analysis of MDB and by-product streams	49
3.2.2. Water holding capacity ( WHC )	50
3.2.3. Gel strength	50
3.2.4. Protein solubility	51
<b>3.3. Fat Separation Procedure</b>	<b>51</b>
3.3.1. Preparation	51
3.3.2. Washing and centrifugation	52
3.3.3. Preparation for functional testing	52
<b>4. RESULTS &amp; DISCUSSION</b>	<b>53</b>
<b>4.1. Fat Separation</b>	<b>53</b>
4.1.1. The effect of various alkalis and pH treatments on fat removal from mechanically deboned beef	53
4.1.2. Distribution of various MDB constituents in various layers after centrifugation	56
4.1.3. Modified separation process for mechanically deboned beef	57
<b>4.2. Functional Tests On Myofibrillar/Collagen Fraction</b>	<b>59</b>
4.2.1. Water holding capacity of myofibrillar/collagen fraction	59
4.2.2. Gel strength of myofibrillar/collagen fraction	60
4.2.3. Protein solubility of the myofibrillar/collagen fraction	61
4.2.4. The interrelationship between functional tests and initial treatment factors	63
<b>4.3. Summary</b>	<b>64</b>
<b>5. SUGGESTED FURTHER WORK</b>	<b>66</b>
<b>6. REFERENCES</b>	<b>67</b>
<b>7. APPENDICES</b>	<b>82</b>

## LIST OF TABLES

	Page
<b>Table 1:</b> Yields, Mechanically Deboned Beef Bones. ( Dudley, 1975 )	3A
<b>Table 2:</b> Yields, Mechanically Deboned Pork. ( Dudley, 1975 )	3A
<b>Table 3:</b> Meat Products In Which MDM Has Been Successfully Incorporated. ( USDA, 1976; Martin, 1976 )	6A
<b>Table 4:</b> Examples of Composition of Mechanically Deboned Meat. ( Froning, 1970; Froning et al. 1971; Grunden et al. 1972; Crawford et al. 1972; Froning and Johnson, 1973; Field et al. 1974; Field et al. 1976; Ackroyd, 1979)	7A
<b>Table 5:</b> Requirements For Mechanically Deboned Red Meat & Poultry. ( USDA, 1982 )	8B
<b>Table 6:</b> Effect Of Mechanical Meat Recovery On Total Bacterial Numbers. ( Newman, 1980 )	18A
<b>Table 7:</b> Properties Proposed For Grading Meat Surimi. ( Ockerman & Hansen, 1988 )	39A
<b>Table 8:</b> Fat Removal Efficiency Of MDB With Various pH & Alkaline Treatments	53A
<b>Table 9:</b> Proximate Analysis Of MDB	56B
<b>Table 10:</b> Solid Content Distribution Of MDB In Various Layers After Centrifuge	56B
<b>Table 11:</b> MDB Fat Content Distribution In Various Kinds Of Protein	57A
<b>Table 12:</b> Solid Contents Of Various Proteins In Modified Fat Extraction	58A
<b>Table 13:</b> WHC Of Myofibrillar/Collagen Fraction From Various pH & Alkaline Treatments	59A
<b>Table 14:</b> Gel Strength Of Cooked Myofibrillar/Collagen Fraction From Various pH and Alkaline Treatments	60A
<b>Table 15:</b> Protein Solubility Of Myofibrillar/Collagen Fraction From Various pH and Alkaline Treatments	61A
<b>Table 16:</b> Interrelationships Between Functional Tests & Initial Treatment Factors	63A

## LIST OF FIGURES

	Page
Fig 1: Effect Of Broiler Back Skin Content On The Moisture & Fat Levels Of The Mechanically Deboned Chicken Meat Obtained. ( Satterlee et al. 1971 )	8A
Fig 2: Denaturation Of Myofibrillar Protein As Influenced By Temperature & pH. ( Konagaya et al. 1978 )	46A
Fig 3: Fat Removal Efficiency Of MDB With Various pH & Alkaline Treatments	53B
Fig 4: Flow Chart Of MDB Separation Process	56A
Fig 5: Flow Chart For Modified Separation Process For MDB	57B
Fig 6: WHC Of Myofibrillar/Collagen Fraction From Various pH & Alkaline Treatments	59B
Fig 7: Gel Strength Of Cooked Myofibrillar/Collagen Fraction From Various pH And Alkaline Treatments	60B
Fig 8: Solubility Of Myofibrillar/Collagen Fraction From Various pH & Alkaline Treatments	61B

## LIST OF APPENDICES

	<b>Page</b>
<b>Appendix 1:</b> Fat Removal Efficiency Of MDB & Standard Deviation	<b>82</b>
<b>Appendix 2:</b> WHC Of Myofibrillar/Collagen Fraction & Standard Deviation	<b>83</b>
<b>Appendix 3:</b> Gel Strength Of Cooked Myofibrillar/Collagen Fraction & Standard Deviation	<b>84</b>
<b>Appendix 4:</b> Raw Data Of Fat Removal Efficiency Of MDB	<b>85</b>
<b>Appendix 5:</b> Raw Data Of WHC Of Myofibrillar/Collagen Fraction	<b>86</b>
<b>Appendix 6:</b> Raw Data Of Gel Strength Of Cooked Mrofibrillar/Collagen Fraction	<b>87</b>

## 1. INTRODUCTION

Mechanically deboned meat ( MDM ) was first commercialized for human food consumption in 1976. Since that time considerable research has been conducted to expand its uses in food products. The main application for MDM is in ground meat products, such as sausages, salami and restructured steaks. A large number of researches have demonstrated that MDM possesses good functionality and can improve the texture of restructured meat products. However, its high microbial count, high fat content and off flavour development have long been its main drawback, especially the oxidative rancidity development from fat has limited its use and storage properties dramatically.

Since 1980, work has been conducted on the extraction of fat from meat to make it more stable and versatile. Chao et al. ( 1991 ) used CO<sub>2</sub> at various pressures and temperatures to extract the fat from ground beef. They found that pressure, rather than temperature, was the predominant factor in determining power for fat extraction.

McLachlan et al. ( 1990 ) extracted fat and cholesterol from partially dried mince with supercritical fluid CO<sub>2</sub> ( SCO<sub>2</sub> ), and they found that the optimal fat removal was achieved when the moisture content of meat was reduced to 30 ~ 50 %. In this patent they used calcium carbonate as an adsorbent to selectively remove cholesterol from SCO<sub>2</sub> - dissolved fats.

Wehling ( 1991 ) extracted samples of spray-dried beef and chicken powders. He found that substantially more fat was extracted at 55 °C than at 45 °C. The fat from chicken chunks was more readily extracted at the lower temperature.

King et al. (1989) and Meat Research Corporation of Australia (1991) reported that maximum fat extraction was achieved when samples were finely minced and dehydrated.

Recently, surimi-type processes have been investigated as suitable method for the removal of fat. Various authors have investigated the effects of aqueous and saline washings at different pH conditions on lipid removal from washed MDM. Lin and Chen (1989) partially removed the lipids and pigments in the production of poultry Kamaboko type products by washing mechanically deboned poultry meat (MDPM) with NaCl or phosphate buffers at PH 8.0. Dawson et al. (1988) reported that fat removal was highest when mechanically deboned chicken meat (MDCM) was washed with an 0.5% sodium bicarbonate solution (pH 7.9 for meat-water mixture). The bicarbonate treatment was shown to be better than washing with tap water or an 0.1% acetate buffer at PH 5.1. However, the extent of fat extractions in various pH and alkalies were not studied yet.

The objectives of this study were to examine the effects of fat extraction from the mechanically deboned beef (MDB) with various PH and alkalies. Also, the functionalities of the residue surimi from the various treatments were determined.

## 2. LITERATURE REVIEW

### 2.1. MECHANICALLY DEBONED MEAT

#### 2.1.1. Potential production

##### 2.1.1.1. Yield

Mechanical deboning has become a widely accepted and economical means of removing meat from carcasses. Mechanically deboned poultry meat (MDPM) has been used in the United States since 1965. The waste "bone" material typically contains 13 ~ 15% protein, 16 ~ 20% fat and 8 ~ 11% ash (Lawrence, 1981). Although the majority of the protein is collagen, over 18% of the wasted protein is alkali extractable sarcoplasmic and myofibrillar protein that could be a valuable ingredient for luncheon meats and similar processed meat products (Jelen et al. 1982).

The use of MDPM in further processed products reached 18 billion kg in 1984 (Schuler, 1985), and it has been estimated that enough raw material is available to easily double that volume if the demand were present. (Prince, 1986).

Yields to be expected from red meat bone residues, according to Dudley's (1975) statement, are 1.4 ~ 1.9 kg of pork meat per carcass and 5.8 ~ 7.5 kg of beef per carcass (Table 1 & Table 2). Even more startling figures have been recorded in tests on the Protecon MRS 15 machine. Processing bacon pig neck bones in one UK plant, an average meat yield of 51 % was recovered (KATZ & ACKROYD, 1977). Pietraszek (1975) reported that 335 million kgs of beef, 170 million kgs of pork and 8.6 million kgs of lamb and mutton could become available for human consumption, i.e. a total of 513 million kgs!

**Table 1: Yields, Mechanically Deboned Beef Bones**

Types of Bones	Approximate Kg Weight per Carcass	Meat Yields per Carcass ( Kg )	Deboning Yield Percent ( % )
Neck	4.54	1.36 ~ 1.82	30 ~ 40
Chine	1.14	0.36 ~ 0.45	32 ~ 39
Loin Rack	4.99	1.00~ 1.50	20 ~ 30
Chuck Rib	4.10	0.82 ~ 1.00	20 ~ 24
Rib Rack	5.44	1.00 ~ 1.18	18 ~ 22
Plate Rack	6.36	1.27~ 1.54	20 ~ 24
<b>TOTAL</b>	<b>26.57</b>	<b>5.81~ 7.49</b>	<b>22 ~ 28</b>

Source: Dudley ( 1975 )

**Table 2: Yields, Mechanically Deboned Pork**

Types of Bones	Approximate Kg Weight per Carcass	Meat Yields per Carcass ( Kg )	Deboning Yield Percent ( % )
Neck	0.45	0.14 ~ 0.23	31 ~ 51
Back	0.73	0.32 ~ 0.36	44 ~ 49
Blade	0.45	0.09 ~ 0.14	20 ~ 31
Ham	2.36	0.50 ~ 0.73	21 ~ 31
Picnic	1.45	0.32 ~ 0.41	22 ~ 28
<b>TOTAL</b>	<b>5.44</b>	<b>1.37 ~ 1.87</b>	<b>25 ~ 34</b>

Source: Dudley ( 1975 )

### 2.1.1.2. Machine types and production mechanism

Although mechanical meat separators have improved tremendously since their introduction, the mode of action of the earliest machines is still the basis of much of today's machinery.

The relatively soft bones from fish and poultry carcasses permit machines of similar action to be used for the separation of both. The recovery process is as follows (Newman, 1979):

A rubber belt is power-driven and rotates against a perforated steel drum which counter-rotates, but at a speed somewhat slower than that of the rubber belt. The bone material is forced into the nip between belt and drum and the softer meat passes through the drum perforations, while the harder bone is retained. The pressure of the belt can be altered to suit the material.

An example of a machine using this process is the BAADER model 694. One problem associated with the belt/cylinder separators is the build-up of material and formation of bulges in the flexible rubber belt especially at the edges. The introduction of pressure rollers, as in the BAADER model 695, even out the distribution of the material and prevent the lateral escape of processed material.

Other machines incorporate alternative refinements which are claimed to improve meat-bone separation. The Bibun range of separators have an additional straining device sieving out any residual bone. Poultry meat has also been the subject of a large number of patented machines using alternative techniques including the cutting centrifuge of Lapeyre( 1975 ), shot blasting with ice particles ( Lindall, 1963 ) and cutting with water jets ( Draper & Rejsa, 1971 ).

Machines modified from existing technology for red meat recovery include the paoli ( Models 19-21 ), Yieldmaster ( RC ) and Beehive deboners ( AU & AUX range ). These all use an auger feed and recover the meat, either through stainless steel screens ( Yieldmaster ) or via microgrooves ( Paoli ). This type of machine can incorporate cooling plant to reduce the temperature rise caused by processing and can be readily adapted for separating fish, poultry or red meat, usually by a simple change of the screening mesh and readjustment. However, an essential prerequisite is that the bones must be preground in a bone grinder.

In Britain and Europe, the development of an alternative hydraulic piston-type separator has been successful. Unilever first developed the concept which was Russian Patents in 1949 and 1962 and the basis of their apparatus is described in British Patent 1451128 ( Unilever, 1976 ). It is similar in design and action to the Protecon system ( Berger, 1977 ). Bones need not be ground, merely broken to a convenient size. Unlike the machines of the first group which are continuous in operation, these are batch producers, each batch being processed within a 30 ~ 60 s cycle. An automatic hopper delivers a pre-determined weight of meat/bones into a thick-wall steel cylinder. A hydraulically-powered piston compacts the meat and bones under pressures of 100 ~ 250 atm. Under this extreme pressure the meat flows off the bones and is recovered through a multiplicity of microgrooves. The harder bone is unable to enter into the grooves and is either compacted into a puck, as in the KP system, and ejected, or removed by a further forward movement of the piston as in the Protecon system.

### 2.1.1.3. Utilization in food products

The MDM recovery process imparts a fine, more uniform texture to the meat. This is due, in part, to the grinding action common to many mechanical separators, as well as the partial removal of the connective tissue from the meat. This does not affect the acceptance of products such as fish spreads and red meat patties, where a finer texture will improve the product, but it can lead to lower acceptance ratings in products where a coarser texture is usually encountered. For example, in the U.S.A. MDM is not permitted in hamburgers, as the particle size of MDM is considered too small when compared to that usually encountered in conventional hamburgers. **Table 3** lists some meat products in which MDM has been successfully incorporated.

Marshall et al. ( 1977 ) reported high consumer acceptance levels for frankfurters containing up to 40% mechanically deboned goat or mutton, but much lower acceptance when made with mechanically deboned pork ( MDP ). Pork, particularly, exhibited poor colour stability and produced rapid destruction of preservatives.

Joseph et al. ( 1978 ) used mechanically deboned beef ( MDB ) in cooked salami. They found that 10 ~ 20% substitution levels of MDB for skeletal muscle can be successfully employed in cooked salami, whilst a 30% MDB level may produce some undesirable attributes in this type of product. Miller ( 1986 ) indicated that the inclusion of 20% MDB yielded a spiced luncheon loaf which was higher in eating quality than an all-beef control. Miller et al. ( 1986 ) showed that extending restructured steak with MDB was superior to textured soy protein or vital wheat gluten extended steak. Field and Riley ( 1974 ) noted that the use of mechanically separated spleen in bologna improved emulsion stability, texture score and reduced shrink. Krol et al. ( 1975 ) showed that the addition of mechanically deboned poultry meat ( MDPM ) to products such as Gelderse Rookworst ( addition rate up to 47% ), Frankfurter type sausage ( addition rate up to 20% ) improved the colour and texture of these products. They also found that an addition of 20% MDPM enhanced the organoleptic qualities of canned luncheon meat. Froning et al. ( 1971 ) showed no consumer difference for frankfurters containing all red meat or up to 15%

**Table 3: Meat Products In Which MDM Has Been Successfully Incorporated**

Beef Patties	Fish Sausage
Breakfast Sausage	Fish Patties
Bologna	Ham, Tongue Spreads
Braunschweiger	Hash
Bockwurst	Luncheon Meat
Chilli Con Carne	Liver Sausage
Corned Beef Hash	Meat Loaf
Chop Suey	Potted Meat
Canned Corned Beef	Pizza
Chow Mein	Smoked Pork Sausage
Devilleed Meat	Scrapple
Fresh Pork Sausage	Spaghetti and Meatballs
Fish Cakes, Sticks etc.	Stuffings
Fish Spreads	Salami
Fresh Beef Sausage	Tamales
Frankfurters	Whole Hog Sausage

Sources of information: USDA ( 1976 ); Martin ( 1976 ).

replacement with recovered turkey meat, providing fresh material was used. There is general agreement that the addition of low levels of deboned poultry meat enhances the acceptability of many red meat formulations. Lampila ( 1984 ) indicated that the use of texturized MDT in restructured turkey meat products may have excellent potential. Meanwhile, Laughren & Maurer have proved that the use of MDT in a sloppy Tom recipe was successful and economic. Scopes ( 1970 ) suggested that MDPM is high in protein, especially myofibrillar protein, therefore it should be a good ingredient for surimi-based foods.

Fish sticks manufactured from fish surimi have been popular for many years now. New innovations with mechanically deboned fish meat have met with increasing consumer acceptance. These include fish cakes and patties ( Mendelsohn & Connors, 1974 ), fish pastes and spreads ( Pataschnik et al. 1973 ), and fish sausage, already widely accepted in Europe and Japan ( Bond, 1975 ).

## **2.1.2. Composition**

### **2.1.2.1. Proximate composition**

Considerable proximate compositional data for MDM has been reported ( Froning, 1970; Froning et al. 1971; Grunden et al. 1972; Crawford et al. 1972; Froning and Johnson, 1973; Field et al. 1974; Field et al. 1976; Ackroyd, 1979 ) (Table 4 ).

High variability seems to be common in the above studies. Field ( 1974 ) argued that the composition of MDM depended on the material entering the boning machine and the setting at which the machine was operating. Field et al. ( 1976 ) reported the percent protein in MDM was lower and the fat content was higher than similar hand deboned meat. This difference in composition was due to bone marrow which was extracted as deboned meat and to connective tissue which was removed from the meat during mechanical deboning.

**Table 4: Examples of Composition of Mechanically Deboned Meat**

Bone source	Percentage of Total Composition		
	Fat	Crude protein	Ash
<b>Pork</b>			
Ham	39.0	10.2	4.0
Picnic	42.3	9.1	3.7
Loin	29.5	14.0	1.8
Neck	27.3	14.7	1.5
Ribs	23.0	15.5	1.2
<b>Cow</b>			
Ribs	23.2	16.4	1.2
Rump	41.9	10.0	4.3
Short loin	33.4	11.6	4.3
Neck	13.7	17.2	3.4
Plate	32.7	11.4	4.3
<b>Mutton</b>			
Whole carcass	19.7	19.1	1.4
Breasts	36.5	15.0	1.2
<b>Chicken</b>			
Neck and Back	22.0	12.3	X
Spent layers	22.3	14.1	X
Broiler necks	17.6	12.4	0.7
<b>Turkey</b>			
Frames	13.6	12.8	X
<b>Fish</b>			
English Sole	2.6	13.4	4.9
Trude Code	2.1	15.6	4.3
Orange Rockfish	7.3	16.2	6.5

Sources of Information: Froning, 1970; Froning et al. 1971; Grunden et al. 1972; Crawford et al. 1972; Froning and Johnson, 1973; Field et al. 1974; Field et al. 1976; Ackroyd, 1979.

" X "..... data unavailable.

Field (1976) stated that pork bones with small amounts of meat adhering to them, e.g. pork ham and picnic bones, yield higher fat and lower protein contents, whilst lean beef back bones yield high protein and low fat (Goldstrand, 1975). However, when whole animal carcasses are mechanically recovered (beef, lamb, and pork), the overall composition is close to that of hand boned meat (Field, 1974; Meiburg et al, 1976). Field (1976) suggested bones from the vertebral column, ribs and sternum are most suitable for mechanically deboning because they usually have more lean attached and thus will yield a greater percentage of mechanically separated tissue, whilst round bones were least suitable for mechanical deboning since they have very little lean attached and the marrow is primarily fat.

Satterlee et al. (1971) investigated the effect of skin content of chicken broiler backs on the composition of the resultant MDM. As the skin content of the backs increased in relation to muscle and bone content, the fat content of the deboned meat increased and the moisture & protein contents decreased (Fig. 1).

Goodwin et al. (1968) defatted necks and backs by trimming before deboning, and observed reduced fat and increased protein in the final deboned product.

Koolmees and Bijker (1986) reported that mechanically deboned poultry meat (MDPM) had a higher crude protein and lower fat content than mechanically deboned pork (MDP) and veal (MDV).

Mechanically deboned meat that is used as an ingredient in meat food products must meet maximum limits on calcium content (as a measure of bone solids content), bone particle size and fat content, also a minimum protein quality requirement (Table 5). In America these are controlled by U.S.D.A. regulations. (U.S.D.A. 1982)

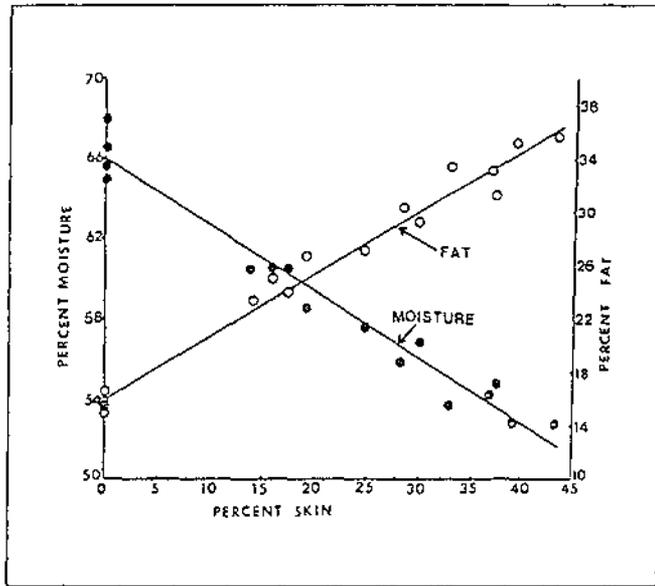


Fig 1: Effect of Broiler Back Skin Content on the Moisture & Fat Levels of the Mechanically Deboned Chicken Meat Obtained ( Source: Satterlee et al. 1971 )

Table 5: Requirements For Mechanically Deboned Red Meat and Poultry

	Minimum % Protein		Minimum Protein Quality		Maximum % Fat
	Raw	Cooked	PER	AA	
Beef	15	21.4	2.5	33.0	30.0
Lamb	15	21.4	2.5	33.0	30.0
Pork	14	20.0	2.5	33.0	30.0
Chicken	15	21.0	2.5	33.0	30.0
Turkey	16	22.0	2.5	33.0	30.0

Minimum protein quality should be on the basis of either Protein Efficiency Ratio (PER) or the percent of the following amino acids expressed as a percentage of the total protein: phenylalanine, isoleucine, leucine, methionine, tryptophan, valine, and threonine.

PER, if used, should be adjusted to 2.5 for casein.

Source: USDA ( 1982 )

### 2.1.2.2. Bone particle and calcium content

Interest in measuring the calcium content of meat products has risen sharply since the 1982 relaxation of federal regulations governing the maximum allowable levels of residual bone fragment in U.S.A. The legal upper limits on permissible fragment in meat have been raised to the point where separation of meat and bone can now be done mechanically in the United States (in spite of the larger percentage of bone fragments introduced into the final meat product via the increase in sample "abrasion" occurring with the more violent mechanical separation approach, Wichman, 1986 ).

Mechanically deboned red meat is limited to a maximum of 0.75% calcium ( representing a measure of " total bone solids " equal to 3% by weight ). Mechanically deboned poultry meat was previously unregulated, but is now limited to a maximum of 1% bone solids ( nominally representing 0.175% Ca by weight for young chickens and ducks, and nominally 0.235% Ca for mature chickens and all turkeys ).

There is general agreement that bone must be reduced to particles of 0.5mm diameter or less in MDM if they are to be undetectable by mouth ( Chant et al. 1977 ), but when properly adjusted, the meat recovery equipment currently on the market prevents the passage of organoleptically detectable bone. There is evidence that incorrect adjustment or lack of maintenance can cause the production of an unacceptable material ( Field, 1974; Mawson & Collinson, 1974 ).

Field et al. ( 1977 ) found a particle size of 77 ~ 120  $\mu\text{m}$  in MDM ( Beehive ). Baum et al. ( 1980 ) investigated the size of the bone particles at different filter apertures ( continuous pressure system ) histologically. A linear correlation between the size of the particles and the size of the filter openings was not always present. Reducing the filter aperture from 1.1 to 0.6mm caused a reduction in maximum bone particle size from 750 down to 260 $\mu\text{m}$  and a decrease of the average diameter from 50 down to 42 $\mu\text{m}$  in both beef and pork MDM.

Field et al. ( 1974 ) stated that calcium content increased when the Beehive deboner's ring valve was tightened to get a higher yield. Ranged from 0.09% to 0.20% as the yield adjusted from 52% to 70%. Field also reported that the calcium content in MDM was always higher than that for hand deboned meat because some fine bone particles went through the holes in the deboner ( Field, 1976 ).

Koolmees and Bijker ( 1986 ) found that press-type deboners produced MDM with a lower calcium content and a lower hard bone residue than auger-type machines. However, bone particle size found in MDM processed by press-type machines were larger.

Patashnik et al. ( 1974 ) pointed out that bone particle content appears to be a function of processing methods and raw material, not of species. Corrao et al. (1983) compared the calcium content of products made from mechanically deboned beef and mechanically deboned poultry. The poultry products ranged in calcium content from 0.007% to 0.023%. Beef products ranged from 0.012% to 0.15%. This research coincided with Patashnik et al. ( 1974 )'s statement.

### 2.1.2.3. Fatty Acid

Mechanically deboned meat has been shown to be more susceptible to oxidation than whole muscle tissue (Froning, 1976; Dawson and Gartner, 1983). Suggestions as to the cause of this oxidation have centered on the increased concentrations of polyunsaturated fatty acids of the phospholipids (Moerck & Ball, 1974; Kunsman & Field, 1976). Earlier Dugan (1961) has stated that the common feature of oxidative rancidity is the reactivity of the unsaturated fatty acid moieties in the lipids.

Kunsman and Field (1976) reported that the polyunsaturated fatty acid content of the phospholipid fraction from mechanically deboned red meat was 25% or higher. In beef and pork MDM, phosphatidylethanolamine accounted for a large portion of the polyunsaturated phospholipid, whereas in lamb MDM phosphatidylcholine was the most common polyunsaturate.

Cross et al. (1978) found MDM from beef bones (aged or fresh) oxidized at about the same rate as lipids in ground beef. They also reported that mechanically deboned red meat (MDRM) had a much higher level of hemo-proteins (haemoglobin and myoglobin) and a lower polyunsaturated fatty acid content than mechanically deboned chicken meat (MDCM). Therefore the relative concentration ratio of polyunsaturated fatty acids to hemo-proteins was different in MDRM compared with MDCM. Changes in the ratio of polyunsaturated fatty acids to hemo-proteins were probably responsible for the difference in storage characteristics between MDRM & MDCM.

However, Dawson and Gartner (1983) argued that MDCM was composed of relatively high levels of unsaturated fatty acids and low levels of natural tocopherol, making it relatively unstable. But the comparison between MDRM and MDCM was not made in their experiment.

Meiburg et al. ( 1976 ) found that mechanically deboned pork was between mechanically deboned beef and mechanically deboned poultry as far as stability was concerned and recommended that mechanically deboned pork should be stored only for a short period of time in the frozen state before TBA values became too high and taste panellists objected to the flavour of this product.

#### **2.1.2.4. Nucleic acid and purine content**

High levels of nucleic acids and purine compounds are undesirable in food products because their metabolic end products include uric acid. Persons afflicted with hyperuricemia or gout are unable to control their serum urate content. When the serum urate exceeds physiological solubility, the urate precipitates forming tophi or calculi, stone-like structures in the joints, kidneys, or in other organs. Therefore before accepting MDM as a new protein source, it would be an advantage to know the nucleic acid & purine contents.

Young ( 1985 ) found that the variations of nucleic acid & purine content in mechanically deboned poultry meat were relatively small. These variations were mostly associated with type of input material and day to day differences. Also, the content of these compounds in bone residue was generally the same or lower than is found in edible meat and poultry products. So it does not appear that the nucleic acids or purines in bone residue will preclude its use as a source of protein for human consumption.

Savaiano et al. ( 1983 ) reported that DNA and total nucleic acid levels were higher in both mechanically deboned beef (MDB) and veal (MDV) whereas RNA levels were higher only in MDB compared to hand deboned counterpart. Total purine content of MDB did not differ from hand deboned beef, whereas the purine content of MDV was slightly higher than hand deboned veal. They also said the addition of MDM to the diet would not significantly alter total purine consumption and hence should pose no risk to hyperuricemic individuals.

Arasu et al. ( 1981 ) quantified the nucleic acid content in bone marrow and established whether nucleic acid levels in MDM, which contained marrow were high enough to be nutritionally hazardous. They found marrow, MDM and muscle averaged 20.3, 7.6, and 1.4 mg DNA/g, and 1.9, 0.7, and 0.4 mg RNA/g respectively, indicating that increased levels of marrow in MDM would increase nucleic acid content. However, because the addition rate of MDM to processed meats is limited to 20% of the meat block, it is unlikely that products made from MDM would be nutritionally hazardous. However, in New Zealand, where there are no upper limits for MDM, it could pose a potential nutritional hazard!

#### **2.1.2.5. Mineral content**

The mineral content may differ substantially between commercial mechanically deboned poultry meat ( MDPM ) and muscle tissue, because small amounts of bone and other tissues ( blood, collagenous and nerve tissues ) are introduced into MDPM from such parts as backs, necks and frames. The U.S.A.'s Agricultural handbook No. 8-5 ( Consumer and Food Economics Institute, 1979 ) lists mean values for nine mineral elements in 3 to 19 samples of MDPM from broiler backs and necks ( unspecified proportions ).

MacNeil et al. ( 1978 ) reported calcium, iron and zinc contents, for MDPM from broiler necks without skin and from broiler backs. Essary ( 1979 ) also reported values for about 40 elements in MDPM from broiler rib-neck ( 1:1 ) and turkey racks. Data for the same element in the same tissue differ widely, as much as 400% in one case, between reports.

Hamm and Searcy ( 1980 ) determined the contents of 10 mineral elements in nine commercial MDPM samples, and found high variability in mineral composition because of the many uncontrollable production factors. Therefore, they suggested the need for frequent monitoring of production lots of these products for mineral elements of nutritional interest.

Klose (1980) reported that MDPM from broilers and young turkeys contained little fluoride while MDPM from old fowl contained levels of fluoride ( $< 30 \mu\text{g/g}$  raw product) that should preclude using this type product in baby foods.

#### 2.1.2.6. Cholesterol Content

Because of the strong public concern over cholesterol and fat content of foods, in 1976 the U.S.D.A convened a select panel to investigate the health and safety of MDM. They analyzed 18 samples of MDM from beef and reported cholesterol levels ranging from 28 ~ 202mg per 100g tissue. (Kolbye et al. 1977)

Sweeney and Workrauch (1976) reviewed the cholesterol content of foods, and reported that beef lean ranged from 42 ~ 78mg per 100g of tissue and beef fat from 76 ~ 131mg per 100g of tissue. Weyant et al. (1976) reported cholesterol values for chuck, round, heart and liver from hay-fed cows of 52, 50, 103, and 222mg per 100g tissue, respectively.

Kritchevsky and Tepper (1961) found veal, beef steak and ground beef contained 85, 114, and 116 mg per 100g tissue, respectively. Moerck and Ball (1974) determined the cholesterol contents of MDM from chicken, and found it went as high as 700mg per 100g tissue.

Kunsman et al. (1985) found that cholesterol content of marrow was significantly different when diet or anatomical locations were compared. Bovine marrow from grass-fed animals averaged 119.6 mg/100g marrow and marrow from grain-fed animals averaged 150.6 mg/100g marrow. The cholesterol content of marrow from the cervical, lumbar and femur was 190.1, 124.1, and 91.0 mg/100g marrow respectively.

MDM and beef lean had a mean cholesterol content of 153.3 and 50.9 mg/100g tissue. Therefore, the MDM which came from back bones which contains large amount of marrow probably can account for the increased concentration of cholesterol in some MDM samples over the values of lean and marrow.

### **2.1.3. Functionality**

#### **2.1.3.1. Emulsion capacity and stability**

In a study of the effect of chopping time and temperature on the emulsion stability of chicken and turkey meat, Froning (1970) found that mechanically deboned chicken formed its most stable emulsion when chopped between +7 °C and +13 °C, whereas hand boned broilers formed emulsions best when chopped at temperatures above +13 °C.

Maurer (1973) said that the mixtures of hand boned and MDM have high emulsifying capacity. Froning (1970) reported that the emulsifying capacity of mechanically deboned broiler backs and necks mixtures was less than mixtures of breasts, legs and thighs, although the comparison between the two mixtures was the same for hand boned meat. He also found the emulsion capacity formed from poultry light meat was significantly better than that from dark meat.

Satterlee et al. (1971) showed that increased skin content in broiler backs, prior to deboning, yielded meat with a higher fat content. This is the probable explanation for the results of Froning et al. (1973) and Schnell et al. (1974) who demonstrated that higher skin content in the starting material led to significant decreases in the emulsifying capacity and the emulsifying stability. The additional skin will have reduced the proportion of actomyosin in the formulation, and this may have led to the lowered emulsion capacity and stability. Similar reasons account for the improved stability and capacity reported by Froning & Johnson (1973) and Dhillon & Maurer (1975) with mechanically deboned poultry meat (MDPM) whose fat content had been reduced by centrifugation.

Increases in pH have also been reported to increase the extractability of protein in MDM ( Anderson & Gillet, 1974 ). Bone marrow content significantly increases pH and this affects the emulsion capacity and stability in MDM ( Field, 1976 ). Neelakantan & Froning ( 1971 ) demonstrated substantial increases in the emulsion stabilities of actomyosin, myosin and myofibrils in MDM by raising pH towards 7.0.. However, the emulsion stabilities of sarcoplasmic proteins fell remarkably at pH 7.0 in the same time, otherwise the functional characteristics of MDM should be more excellent than whole muscle tissue whose pH is around 5.8 ~ 6.2. ( Field, et al. 1974 ).

#### **2.1.3.2. Water holding capacity and texture**

A number of studies have been carried out with various types of ground meat products whose muscle proteins have been replaced by MDM. Pisula & Rejt ( 1979) have experimented with added pork backbone MDM to meat model blends. They found that the water holding capacity ( WHC ) increased when 10 or 20% muscle protein was replaced with MDM protein. The increase in WHC was shown to be higher in mixed batches only. The product with 100% MDM was shown to have very poor WHC. The same trend was found when determining the cooking losses.

Marshall et al. ( 1977 ) studied the effect on cooking loss and texture of adding increasing amounts of mechanically deboned pork ( MDP ) to Frankfurter sausages. They manufactured Frankfurters comprising 10%, 25% or 40% MDP, calculated on total batch weight, and found that the cooking loss increased with increased MDP concentration. However, when sausages were judged for texture by sensory evaluation, the batch containing 10% MDP was ranked the best, followed by the control batch and the batch containing 25% MDP. The batch containing 40% MDP was rated as the poorest and assigned the description " Very mushy ".

Cross et al. ( 1977 ) produced beef patties containing up to 30% mechanically deboned beef ( MDB ). They were unable to prove any constant relationship between MDB concentration and cooking loss. Texture was evaluated by a consumer panel. The results were that texture was judged to be better with increasing MDB.

Tomsen & Zeuthen ( 1988 ) reported that the addition of MDP increased the pH and the WHC, and that moderate addition of MDM resulted in greater yield stress and elasticity modulus, while higher concentrations of MDM resulted in very soft texture. The pH of the meat blends was very important for the functional properties of the pasteurized products.

#### **2.1.3.3. Binding properties**

The inherent problem of mechanical deboning is complete disruption of the muscle fibril, which results in a mealy, coarse texture upon heat treatment of 100% MDM. Because protein binding is essential for a product with a texture acceptable to consumers, the greatest problem involving the use of MDM alone is protein binding.

Acton ( 1973 ) studied the effect of heat on the binding properties of mechanically deboned turkey. He demonstrated that longer heating times ( 0 to 12 mins at 100 °C ) increased resistance to shear with good binding quality development. The heat-initiated binding of meat proteins has long been recognized. The characterization of exudate proteins involved in the binding of beef chunks has been reported by Booreen et al.(1982).

Lampila ( 1985 ) incorporated texturized mechanically deboned turkey at levels of 0~ 50% to turkey roasts. He found at higher addition levels of texturized mechanically deboned turkey, the turkey roasts exhibited good binding and textural properties, and puncture compression testing did indicate increased firmness, too.

Taking full advantage of functional proteins and enhancing these properties with modified treatments may promote the utilization of MDM in restructured meat products!

#### **2.1.4. Disadvantages and advantages**

##### **2.1.4.1. Bacterial consideration**

Use of MDM has been limited because the meat has a pH close to neutral, high moisture content, very small particle size and large surface area, and is high in blood, iron, calcium and lipids. These characteristics predispose MDM to microbiological and enzymatic deterioration if associated with inferior raw material, unsatisfactory processing or inadequate refrigeration. Some reports have been published where MDM contained potential pathogens. Zwingman ( 1980 ) found 300 ~ 500 staphylococci per gram in samples of MDM, while Smelter & Ramsay ( 1981 ) reported that they isolated 24 serovars of Salmonella from 40 of 55 MDM samples tested.

A greater problem exists for mechanically deboned poultry than for other meat as, generally, they contain much higher bacterial levels in the raw state (Terbijhe, 1976). Mulder & Dorrestein ( 1975 ) have studied the transmission of pathogenic bacteria present in poultry during the different stages of processing, using different mechanical recovery equipment. They concluded that bacterial passage into the final product was frequent and not affected by the separation technique used. Newman ( 1980 ) in a review of the microbiology of MDM before and after processing ( **Table 6** ) showed that microbial numbers often increase during the production of MDM.

Table 6: Effect of Mechanical Meat Recovery on Total Bacterial Numbers			
Material	Before Processing	After Processing	Reference
Mutton			
Pre-rigor	$1.4 \times 10^4$	$1.4 \sim 3.0 \times 10^6$	Mawson & Collinson, 1974
Hot ( 33 °C )		$1.8 \times 10^4$ ( Hand Boned )	Field, Riley&Corbridge,1974
		$2.2 \times 10^4$ ( Machine Boned )	Field, Riley&Corbridge,1974
Cold ( -1 °C )		$3.2 \times 10^5$ ( Hand Boned )	Field, Riley&Corbridge,1974
		$4.3 \times 10^5$ ( Machine Boned )	Field, Riley&Corbridge,1974
Chilled	$1.1 \times 10^4$	$1.6 \times 10^5$ ( Start of run )	Mawson & Collinson, 1974
		$5.5 \times 10^4$	
	$1 \times 10^3 \sim 1 \times 10^4$ , *	$3.0 \times 10^6$	Meiburg, 1976
Pork	$1 \times 10^3 \sim 1 \times 10^4$ , *	$1.0 \times 10^5$	Goldstrand, 1975
		$5.0 \times 10^4$	Meiburg, 1976
	$1 \times 10^5 \sim 6 \times 10^6$	$1.3 \times 10^5 \sim 6.0 \times 10^6$	Ackroyd, 1978
Beef	$1 \times 10^3 \sim 1 \times 10^5$ , *	$1 \times 10^5 \sim 1.0 \times 10^6$	Goldstrand, 1975
		$5 \times 10^4 \sim 5.0 \times 10^5$	Meiburg, 1976
	$9 \times 10^4$	$7.0 \times 10^4 \sim 1.0 \times 10^5$	Ackroyd, 1978
Chicken	$3.3 \times 10^5$	$9.3 \times 10^6$ ( after 12 days at + 3 °C )	Froning, 1976
		$1.8 \times 10^4$ ( after 90 days at - 15 °C )	Froning, 1976
			Ingram & Robert, 1976, *

Sources: Newman ( 1980 )

To minimise bacterial numbers in MDM, Wittmann ( 1977 ) suggested only certain meat-bearing bones should be used and these must be processed immediately after they are obtained or within 24 hrs if stored at a temperature of -1 to +2 °C. If stored for longer then the storage temperature must be at least -18 °C. Transport of the raw material is only possible in the deep-frozen condition. Head bones, feet bones, marrow bones and bones with little meat on them should be excluded, as the yield of residual meat obtained is too low and its bacterial quality too poor. The residual meat obtained should only be transported in the deep frozen state. Packaging materials should be very firm and stable. Moreover, the deboned meat should be used immediately after separation, or within 24 hrs if stored at -1 to +2 °C or within 3 months at the latest if stored at -18 °C. Only by following strict hygienic control, can the functional characteristics of MDM be maintained at their optimum.

#### **2.1.4.2. Oxidative rancidity and off flavour**

MDM is highly susceptible to deteriorative changes during storage, with oxidative rancidity and off flavour being the major problem ( Mooerck & Ball, 1974 ). The high oxidative potential of MDM is due in part to the shearing and mixing action of the deboning machine which results in a highly aerated product ( Dawson & Gartner, 1983 ). Also, the mechanical deboning process has been reported to incorporate heme and lipid components from bone marrow and skin into the resulting meat. Therefore, lipids from these sources might also affect the flavour quality of the meat and be responsible for the stability problems found in subsequent utilization and storage of the meat. The high levels of polyunsaturated fats in some types of MDM are also responsible, in part, for the production of off-flavour, particularly in stored mechanically deboned fish and poultry meat.

Many attempts have been made to slow down or prevent oxidative rancidity in MDM. Generally, oxygen, light and temperature all act as determining factors on the rate of lipid oxidation ( Solberg, 1968 ).

Barnett, et al. ( 1971 ) showed that adding carbon dioxide to refrigerated sea water used as a chilling medium significantly extended the storage life of marine fish, and Groninger ( 1972 ) successfully applied the techniques to mechanically deboned fish meat. Johnson et al. ( 1974 ) said that long storage at temperature below  $-13^{\circ}\text{C}$  had no effect on the eating quality of mechanically deboned turkey meat ( within limited time ). Also, increasing storage time and higher storage temperature resulted in increases in TBA values. Schnell et al.( 1971 ) reported that the TBA values of mechanically deboned chicken with the smallest particle size increased most rapidly following processing. Therefore, this partly means mechanical deboning with greater yield ( smaller particle size ) needs to consider the oxidative problem at the same time!

#### **2.1.4.3. Nutritional and health implications**

The addition of MDM into the human diet must be viewed in relation to its effect on the existing balance of protein, fat and essential mineral nutrients, and any imbalance that result from its inclusion.

The amino acid composition of the MDM is of great importance in estimating its nutritional value. Essary & Ritchey ( 1968 ) found the amino acid content of mechanically deboned turkey meat is comparable to hand boned meat, while Chang & Field ( 1977 ) reported that sulphur amino acids and isoleucine were limiting factors in mechanically deboned red meat. Therefore, these two amino acids were a good indicator of protein quality. The best quality protein came from bones with large amounts of muscle adhering to them.

Nutritionally, the essential amino acid complement in mechanically deboned fish ( Crawford et al. 1972; Meinke et al. 1975 and Wong et al. 1975 ), poultry ( Essary & Ritchey, 1968 ) and red meat ( Field, 1976 ) is more than satisfactory. The very nature of the material, being essentially meat, indicates that it is unlikely that the quality of the protein will be below recommended standards ( Alsmeyer et al. 1974 ). However, in compact bone, Eastoe ( 1955 ) has shown that bone collagen has less than 16% of essential amino acids. Thus when bone levels in MDM are very high, the protein efficiency ratio is often 2.0 or less, below the minimum recommended standards, 2.5. Therefore, partial removal of collagen during processing would enhance the food value of MDM. Meanwhile, as processed meat products form only a small portion of most people's diets, any amino imbalance due to the inclusion of MDM is unlikely to present a serious nutritional threat!

The level of iron in MDM can be as much as twice that of hand boned meat ( Field et al. 1976 ). Eastoe ( 1961 ) said this additional iron originated from the bone marrow.

Skeletal bone and bone marrow accumulate a number of other mineral elements including lead and other heavy metals ( Schroeder & Tipton, 1968 ), barium and strontium ( Sowden & Stich, 1957 ) and vanadium ( Soremark & Anderson, 1962 ). Lead, heavy metals and strontium are particularly undesirable as once accumulated they are never lost. However, at present none of these elements at the concentration found in MDM is likely to pose a health problem.

Fluorine readily accumulates in bone, particularly areas of active bone growth ( Weidemann & Wetherall, 1959 ). It is required for the maintenance of a normal skeleton and reduces the incidence of osteoporosis ( decrease in bone density ) in the mature adult ( Underwood, 1973 ). As there is an increased rate of physiological fluorine uptake in children and infants, it has been recommended that MDM should not be incorporated into junior or baby food formulations until further studies on long term fluorine intakes are available.

Many research have demonstrated the possible beneficial effects of bone in the diet( Drake et al. 1949; Davidson & Passmore, 1963 and Fleischmann et al. 1966 ). Indeed, calcium supplementation of foods such as flour have been considered desirable for many years and is required by law in the U.K. Therefore, the presence of a certain amount of calcium in the MDM should be beneficial to the nutritional quality.

Using MDM in the human diet can be seen to be generally beneficial. However, additional research is needed to ensure that there are no long term problems associated with the consumption of MDM. The very young and old are two groups who should be followed closely to assess the effects of MDM in their diets.

## 2.2 Separation of Mechanically Deboned Meat

### 2.2.1. Fat removal techniques

#### 2.2.1.1. Supercritical fluid extraction

Supercritical fluids exhibit physicochemical properties intermediate between those of liquids and gases which enhance their efficacy as solvents. The relatively high gas density gives good solvent power, while the relatively low viscosity and high diffusivity provide appreciably higher gas permeability into the solute matrix. These properties impart higher rates of mass transfer of solutes into a supercritical fluid than a liquid (Rizvi et al. 1986). In addition the inclusion of a relatively small amount of ethanol in the supercritical fluid can improve both the solubility and selectivity of the solvent (Brunner & Peter, 1982). The most important thing is that supercritical fluid extraction (SFE) has minimal effects on the properties of the proteins (Duwe et al. 1986; Eldridge et al. 1986).

CO<sub>2</sub> is a useful solvent in supercritical extractions of food components because it is non-toxic, non-flammable, inexpensive and readily available. It also has relatively low critical temperature (31.1 °C) and pressure (1070 psig, 7.38 MPa).

Many researchers have employed superfluid extraction to extract oil from oil seeds or lipids from fish muscle and red meat (Stahl et al. 1980, 1984; Friedrich & Pryde, 1984; Snyder et al. 1984; Christianson et al. 1984; Bulley et al. 1984; Taniguch et al. 1985 and Lee et al. 1986). Hardardottir & Kinsella (1988) extracted 97% fat and 99% cholesterol from fish muscle with a combination of ethanol as an entrainer.

Chao et al. ( 1991 ) used supercritical - CO<sub>2</sub> ( SCO<sub>2</sub> ) to extract the fat from ground beef with pressures ranging from 103 ~ 310 bar at 30 ~ 50 °C. They found higher pressures were needed to increase the yield of extracted fat, and that pressure, rather than temperature was the determining factor for extracting lipid from ground beef.

Wehling ( 1991 ) reported using SCO<sub>2</sub> with a pressure at 303 and 308 bar respectively at 45 ~ 55 °C to extract fat from dehydrated beef and chicken. He found that fat was removed from dehydrated chicken more readily at the lower temperature. More significantly fat and cholesterol were extracted at 55 °C than at 45 °C. He also found that because of the powder-like samples which were used, they tended to pack tightly and restrict the CO<sub>2</sub> flow. This greatly reduced the extraction efficiency!

King et al. ( 1989 ) and a report for the Meat Research Corporation of Australia ( 1991 ) showed that during SCO<sub>2</sub> extraction maximum yield could be achieved when samples were finely comminuted and dehydrated. This partly coincided with McLachlan et al.'s ( 1990 ) report. They said maximum extraction was accomplished if the moisture content of the meat was reduced to 30 ~ 55%. They used partial freeze drying to remove the moisture, partly to minimise the formation of sterol oxides which were not readily soluble in SCO<sub>2</sub>.

In Reid's ( 1993 ) thesis report, he used SCO<sub>2</sub> to extract objectionable compounds and their precursors from mutton fats and tallow. It has been shown to be an energy-efficient, environmentally friendly process to deodorize milkfat, beef fats and vegetable oils. In his experiment he also suggested that at higher pressure (250 bar) relatively large quantities of fat were extracted.

### 2.2.1.2. Washing procedure

The aqueous washing of mechanically separated fish meat has been successfully used to increase the value of underutilized fish mince. More recent interest has been directed towards washing meat, particularly MDM from other species to increase its use and value. The results have proved that aqueous washing not only reduces the fat content, but also improves the lightness. Undesirable flavour compounds may also be eliminated by aqueous washing. Furthermore, because the washing process may also increase the concentration of myofibrillar proteins, thereby, improve the gel strength and elasticity of meat products ( Lee, 1984 ). All of these advantages of washing could greatly promote the value of MDM in the food industry.

Ball et al. ( 1984 ) used tap water ( pH 6.8 ), a sodium bicarbonate solution ( pH 8.45 ), and acetate buffer ( pH 5.25 ) to wash intact and blade-tenderized broiler thigh meat. All solutions significantly reduced the fat content of treated thighs and resulted in a significantly lighter meat. Dawson et al. ( 1988 ) tested solutions of tap water ( pH 6.8 ), sodium bicarbonate ( pH 8.5 ), and acetate buffer ( pH 4.5 ) in washing MDCM. All the treatments resulted in a significant reduction of fat but only the sodium bicarbonate solution significantly increased lightness and reduced redness. Based on these results, Dawson et al. ( 1989 ) designed a pilot-plant extraction procedure to evaluate the efficiency in removing fat and colour from MDCM produced from skineless broiler breast frames. In their experiments, 91 kilogram, batches of MDCM were extracted with a .75% sodium bicarbonate solution ( pH 8.0 ) followed by a rinse with tap water. The pH of the water/meat slurry was adjusted to 6.8 with 1 N HCl to facilitate dewatering. The meat slurries were pumped through a continuous decanting centrifuge after the washing and rinsing steps. The raw washed ( RW ) MDCM was found to be lighter in colour than the raw unwashed ( RU ) MDCM. The cooked washed ( CW ) MDCM was also lighter than the cooked unwashed ( CU ) MDCM. The RU MDCM fat content ( 12.8 % ) was reduced by washing to 1.5% in the RW MDCM. The fat content of CU MDCM ( 13% ) was also significantly reduced to 2.5% in the CW MDCM. Therefore, this pilot plant washing procedure was proved to be effective in removal of fat and colour from MDCM.

Dawson et al. (1990) adopted the above pilot-plant washing procedure to determine the changes in the phospholipid and neutral-lipid fractions of MDCM due to washing. They found washing did remove 88.3% of the total fat from MDCM, yet this meat had higher TBA values and oxidized to a greater extent during storage than the unwashed MDCM.

Synowiecki and Shahidi (1991) washed mechanically separated seal meat (MSSM) with water three times. They found the lightness (Hunter colour meter, L value) was increased from 14.8 to 32.4. Meanwhile, the total lipid content was reduced by between 30.7 to 57.5%, depending on the original fat content of the samples. Samples with a higher initial fat content (21.8%, on a dry basis) lost only 30.7%. Reversely, samples with a lower initial fat content (12.7%, on a dry basis) lost 57.5% of total fat.

#### 2.2.1.3. Solvent extraction

A number of solvents have been recommended for fat extraction of animal and plant tissues. Dowigiallo (1975) and Christie (1982) suggested a 2:1 mixture of chloroform-methanol, as it supposedly removes lipids more completely than other solvents (Hagan et al. 1967). Diethyl ether, petroleum ether, and chloroform have been recommended for extracting neutral lipids (Dowigiallo, 1975). Blem (1976) also suggested petroleum ether or petroleum ether followed by a second solvent for neutral lipids extraction, while Sawicka-Kapusta (1975) stated that ether was the best and most convenient solvent to use, but gave no supporting data.

Though Hagan et al. (1967) said chloroform-methanol is likely to be the most effective solvent for removing total fat, Christie (1982) argued that it also may extract some non-lipid material, e.g; amino acids and carbohydrates, when used in a fat extractor. Dobush et al (1985) also suggested chloroform-methanol is an inappropriate solvent for studies dealing with body composition while used in a Goldfish or Soxhlet fat extractor.

The solvent used should depend on the type of lipid being quantified (Nelson 1975; Christie, 1982). The two major types of lipids are neutral lipids and phospholipids. They both dissolve in solvents of different polarities (Nelson, 1975; Christie, 1982).

Chung et al. (1980) reported the extractability of total lipids from hard red winter wheat flour increased linearly with extraction temperature, and any increases in amounts of total extracted lipids, were mainly due to increases in the extracted polar lipids. They also suggested proper selection of solvent and/or extraction temperature was of major importance for polar lipid extractability.

Chung and Ferrier (1991) reported hexane-isopropanol (77:23, w:w) was most effective for removing the fat from egg yolk powder. It extracted more than 50% of the lipid. However, protein solubility and emulsifying activity of the extracted powder decreased after extraction.

Erickson (1993) extracted catfish minced muscle using 9 solvent systems including: chloroform : methanol (2:1), hexane : isopropanol (3:2), chloroform : isopropanol (7:11), dichloromethane : methanol (2:1) or chloroform : methanol : water (2:2:1). Only a few minutes were required to extract a tissue sample by these methods as compared to a few hours for Soxhlet extraction (using petroleum ether).

## 2.2.2. Fish surimi

### 2.2.2.1. Basic concepts and history

Surimi is a Japanese term for mechanically separated fish flesh that has been washed with water and mixed with cryoprotectants for a good frozen shelf life. It is used as an intermediate product for a variety of fabricated seafoods. Washing not only removes fat and undesirable matters, such as blood, pigments, and odorous substances, but, more importantly, increases the concentration of myofibrillar protein (actomyosin), thereby improving the gel strength and elasticity. Surimi, because of its high concentration of myofibrillar protein, produces an elastic and chewy texture which can be made to resemble that of shellfish. Due to this unique property, surimi has been used extensively in Japan for many centuries to develop a variety of fabricated products. It appears that surimi has great potential as a functional protein ingredient which can be substituted for a variety of traditional animal and vegetable proteins ( Lee, 1984 ).

Traditionally, Japanese surimi was freshly prepared from fresh fish and immediately processed into Kamaboko products. Kamaboko is a generic term which includes a variety of products prepared from surimi. The technique for making Kamaboko products from minced and washed fish evolved around A.D. 1100, when Japanese fishermen discovered that they could keep the product longer if washed minced fish was mixed with salt, ground, and steamed or broil-cooked. Although all products made from surimi are generally called Kamaboko, strictly speaking, Kamaboko are those mounted on a wood plate and steamed and/or broiled. There are other narrowly defined products such as Chikuwa, which is broiled, and tempura, which is fried.

The traditional surimi production was run on a day-to-day basis, depending upon the supply of fresh fish. Consequently, the surimi industry could not expand to any great extent and remained in a limited capacity. However, in 1959, the surimi industry took a new turn when a group of scientists discovered a new technique to stabilize frozen surimi ( Matsumoto, 1978 ). This discovery was made from an incidental finding of a cryoprotectant which kept the surimi from freeze denaturation during frozen storage. This technique enabled Japanese manufacturers to stock pile surimi, previously most of the surimi was produced on shore, but subsequently about half of the surimi has been produced on processing ships as a result of an intensive joint effort by government and industry to mechanize on board surimi production. Subsequently, frozen surimi production increased from 32 metric tons in 1964 to 250,000 metric tons in 1987 in Japan, and 400,000 metric tones in the world ( Okada, 1963; Matsumoto, 1978; Knight, 1992 ).

#### **2.2.2.2. The surimi process**

##### **Raw material preparation**

The fish should be handled carefully since fish freshness affects the quality of the surimi ( Ohshima et al. 1933 ); fresher fish results in higher-quality products. The body temperature of the fish should be kept just above the freezing point prior to processing, stored in crushed ice or in refrigerated sea water. In addition, the fish should be delivered for subsequent processing within 130 hours of harvest in order to produce the highest quality surimi. Holmes ( 1987 ) said washing the fish by dipping in refrigerated seawater for 60 hours prior to evisceration results in a high quality product. Then, the head, viscera and a major part of the backbone are removed with a header/gutter, followed by filleting with a mechanical filleter. Once the fish is cut open, the opportunity for biochemical interactions and bacterial contamination leading to loss of protein functionality becomes apparent. Suzuki ( 1981 ) said proteolysis by enzymes found in the viscera will reduce the quality of the surimi. The accumulation of high levels of fat from the viscera is also undesirable for producing high quality surimi. For fish oils are rich in polyunsaturated fatty acids, surimi containing a high level of fat tends to undergo lipid oxidation ( Ackman, 1989 ).

During subsequent processing, the muscle tissues are separated and removed from the skin tissues of the fish fillets using a mechanical deboner. The crude muscle tissues thus obtained are then extruded over a rotating stainless steel drum with small pores to obtain the minced meat.

### **The washing process**

Washing with water is the central process in surimi production. It removes enzymes, pigments/blood, lipids, and haem compounds which cause lipid oxidation leading to protein denaturation. In addition to removing these undesirable compounds, washing concentrates the actin and myosin to give a product with good gel forming properties. The number of wash cycles, contact time and water:mince ratio is dependent on the raw material preparation. In general, 3 cycles of 10 minutes contact with water:mince ratio of 3:1 or 4:1 would be adequate for most applications. The water temperature is usually 5 ~ 10 °C to prevent muscle protein denaturation, although this will depend on the fish species, tropical fish should be capable of withstanding higher temperatures ( Hall, 1992 ).

Water quality is important as a high pH will lead to water retention in the mince. Hard water with metal ions present, such as calcium/magnesium and iron/manganese will affect texture and colour respectively. Saeki et al. ( 1986 ) found that high concentrations of calcium and magnesium ions in the wash water not only decreased the temperature tolerance of the minced meats during the washing process, but also accelerated the denaturation of actomyosin in the frozen surimi during storage ( Tamoto, T. 1971 ). Salt ( up to 0.2 % ) is usually added to the last wash to remove water but should not be at a level to solubilize the actin and myosin.

### **Refining and dehydration**

The washed meats are passed through a refiner to remove any remaining small bones, dark muscle tissues and skin tissues. The refiner is a high speed rotary spiral surrounded by a screen with many small pores 1.2 ~ 3.2 mm in diameter. It

is important to operate the refiner at a low temperature to maintain the protein quality. Excess water being held in the washed meats is then removed using a screw press; the moisture content is reduced from 90 % in the refined meats to 80~84 % in the dehydrated meats. Actually, the moisture level after the dehydration process depends on the washing steps: the temperature, pH and ionic strength of the washing water, the mixing ratio of meat and washing water, etc. ( Ohshima, et al. 1993 ).

#### **Use of additives**

In general, 4 % sucrose or 4 ~ 5 % sorbitol is added to the dehydrated meats in addition to 0.2 ~ 0.3 % polyphosphate. Although polyphosphate alone does not appear to be an efficient cryoprotectant, polyphosphate together with sucrose or sorbitol is more effective than sucrose or sorbitol alone ( Ohshima et al. 1993 ). Added sucrose is sometimes undesirable as it imparts a sweet taste to the surimi and causes browning during frozen storage ( Lanier and Akahane, 1986 ); therefore Lanier (1986) suggested polydextrose be used to avoid browning.

The surimi can now be packed in trays and frozen quickly to below -25 °C using a contact freezer.

#### **2.2.2.3. Gel quality evaluation**

Surimi is highly concentrated with myofibrillar protein, primarily actomyosin, which is solubilized by salt during chopping. The solubilized protein sol then gels upon heating. The gel forming ability, measured by the water binding capacity and gel strength, are determined by the level of functional actomyosin present. The level of functional actomyosin increases with an increase in the number of washing cycles and decreases as the freshness of the fish decreases; such a decrease in extractable actomyosin can be explained by tissue autolysis, which increases with extended storage ( Makinodan et al. 1980; Kim et al. 1982 ). In addition, the quality of surimi during frozen storage is also affected by storage temperature, the level of remaining moisture, and the type and level of cryoprotectants used ( Lee, 1984 ).

Iwata et al. (1968, 1971) found the gel forming ability of surimi made from fresh fish (1 to 2 days) does not change significantly for up to one year when held at a constant temperature below  $-20^{\circ}\text{C}$ . However, when the surimi is stored at  $-10^{\circ}\text{C}$ , the gel forming ability gradually decreases, and the surimi becomes useless after three months; this is attributed to a decrease in extractable actomyosin (Iwata et al. 1971).

Okada et al. (1968) reported that temperature fluctuation during short term transportation (less than a week) does not significantly reduce surimi quality; however, extended periods of temperature fluctuation (more than three weeks) cause significant loss in quality.

Iwata and Okada (1971) found surimi containing salt does not deteriorate as fast as surimi without salt. However, surimi containing salt may undergo gelation during storage as a result of the setting of solubilized protein; this makes surimi less functional for making surimi based products.

#### **2.2.2.4. Production of surimi - based products**

Surimi-based products are prepared by extruding the surimi paste into various shapes which resemble shellfish meat such as crab, lobster, scallop, or shrimp. The closer the simulation desired, the greater sophistication of extrusion technique is required. The products may be divided into four major categories according to their fabrication and structural features: molded, fiberized, composite-molded, and emulsified (Lee, 1984).

#### **Molded**

These products are produced by molding the chopped surimi paste into the desired shape and allowing it to set and form an elastic gel. Molding may be done by

either a single extrusion or a coextrusion. For single extrusion, the paste is extruded through a single opening of the nozzle without cocurrent texturization. For coextrusion, the paste is extruded through a nozzle having many separate openings such that strings of extrudate are laid over one another during forming. Coextrusion therefore gives a meat-like texture, whereas the single extrusion results in a uniform and rather rubbery mouthfeel. Restructured shrimp from broken or odd shaped shrimp of low value is in this category ( Lee, 1984 ).

### **Fiberized**

These products are produced by extruding the paste into a thin sheet through a rectangular nozzle having a narrow opening ( 1 ~ 3 mm ). The extruded sheet is then partially heat set and cut into strips of desired width by a cutter, similar to a noodle cutter. Strip width is determined by the type of finished product desired. Fine strips are preferred for the fibrous crab-leg product, whereas wider strips are more suitable for the simulated shellfish in the form of sea flake and chunk. The surimi used in this process should be of top grade so that the paste remains sufficiently cohesive and elastic while it is stretched, cut, and pulled ( Lee, 1984 ).

### **Composite-molded**

For these products, the strings of desired length are mixed with or without surimi paste and extruded into a desired shape. This type of product tends to be rubbery and uniform in texture. Composite-molded products are found in chunk form and sold mixed with fiberized products.

Another type of composite-molded product called " fish ham " is prepared by mixing the dice of cured tuna and pork into the fish paste before extrusion ( Lee, 1984 ).

### **Emulsified**

To make this type of product, surimi is treated in a manner similar to that used when meat is processed for emulsion products. The level of fat added is usually

less than 10 %, and the type of fat used is not limited to animal fat ( Lee, 1984). In fact, vegetable oil is often added, since, unlike red meat, fish meat readily produces a stable emulsion with oil ( Lee et al. 1981 ). For wiener-type products, the resulting paste is stuffed into casings and steam or smoke-cooked. A variety of these products have been developed and successfully marketed in Japan for more than twenty years.

### **2.2.3. Meat surimi**

#### **2.2.3.1. Why red meat and poultry surimi**

Due to the successful development of large scale fish surimi technology and the increasing market share of fish surimi-based products throughout the world, research has been prompted in applying the surimi process to red meat and poultry. Knight ( 1992 ) has listed a number of potential advantages of using red meat and poultry surimi in new meat products :

- (1) lower fat and cholesterol content than other manufacturing meats;
- (2) reduced risk of rancidity development;
- (3) reduced risk of microbial spoilage;
- (4) bland-tasting raw material to which any flavour can be added;
- (5) almost colourless raw material for incorporation into a range of products;
- (6) improved rheological properties compared with other manufacturing meat;
- (7) a base raw material that can be used as the major component of a product, therefore providing a wider product range than is possible from other manufacturing meats.
- (8) upgrading low valued raw material such as MDM and trimmings to produce high quality meat ingredients for further processing.

### 2.2.3.2. Varieties of meat surimi

The extraction of pigment and fat from red meat and poultry for the production of surimi-like raw material has been investigated by many researchers since the early 1980s. A variety of red meats have been used to produce red meat surimi, and include pork, beef, mutton and poultry. The different species meats are known to have differing functionalities and features, and therefore the surimi derived from the various species are likely to require different methods of production and moreover their functional characteristics could be different as well.

#### **Pork surimi**

Lee et al. ( 1987 ) examined the effects of pH, ionic strength, temperature, and number of washings on the colour and yield of pork surimi. They found that as the number of washings increased, the fat and salt soluble protein content decreased, but the water content and colour lightness values ( L, a, b scale ) increased. Increasing the pH value above 5.5 also increased the water content of the surimi. The maximum colour lightness was obtained from pork treated between pH 5.0 and 5.5; however, the salt-soluble protein content of the surimi was lowest in this pH range, compared with higher pH treatment ( pH 7.5 ). Knight ( 1992 ) said this was due to the fact that the isoelectric point of myosin lies between pH 5.0 ~ 5.5. The protein concentration of the pork surimi they produced ranged from 8 ~ 18 %, and fat content was less than 0.5 %. This composition is similar to fish surimi ( Lee, et al. 1987 ).

Mckeith et al. ( 1988 ) used chopped lean pork as a raw material to make pork surimi. They filtered the slurry through a metal screen with 2 mm mesh to remove connective tissue, then centrifuged the filtrate at 2,000 g for 15 minutes. Then collected the pellet and rewashed twice with water. They found the yield was 45%, higher than the traditional fish surimi yield, 25 % ( Ohshima et al. 1993 ). The pork surimi contained 23.5 % protein and 0.1 % fat, an excellent raw material for a protein product ( Lee, et al. 1987 ).

McCurdy et al. (1987) extracted the protein residue from mechanically deboned pork with alkali at pH 10.5, followed by acid precipitation at pH 5.3. They found in laboratory scale experiment, 11 % of the starting protein was recovered in a protein coagulate. Also, the use of the basket centrifuge in the pilot scale process greatly reduced the fat level in the final protein product in comparison to the horizontal decanter centrifuge process.

### **Beef surimi**

A large amounts of research on beef surimi has been conducted by Leatherhead Food Research Association, LFRA (Knight, 1992). They produced beef surimi with fat content ranging from 0.5 ~ 1.8 % and protein from 15.8 ~ 19.4 %, comparable with pork surimi produced by Lee et al. (1987). In the test of compressibility they found that pH had a significant influence on compressibility, producing softer gels with increasing pH value. The results also indicated that sodium tripolyphosphate causes a small reduction in compressibility, and that there can be interactive effects between pH and sodium chloride and between pH and sodium tripolyphosphate (Knight, 1992).

In the evaluation of tensile adhesive strength (TAS), LFRA found that pH is the most significant factor affecting the TAS values, with a small increase in TAS with increasing pH (Knight, 1992).

When LFRA determined the effects of cryoprotectant on beef surimi, they vacuum - packaged, blast-froze and transferred the beef surimi to - 18 °C overnight before thawing, then they found the thawed material was paler in colour than fresh surimi.. It also had a " bitty" or loose feel to the touch and felt less cohesive than the fresh material ( Knight, 1992 ). However, they also found that when salt and phosphate were chopped into the frozen-thawed surimi, it became more cohesive and doughy! It also became light-tan in colour( Knight, 1992 ).

McKeith et al. (1988) used lean beef and beef heart, weasand meat, head meat, and tongue as the raw materials and followed the same procedure of pork surimi production as stated previously to produce beef surimi. They found beef surimi was significantly darker than commercial fish surimi after being cooked. The textural characteristics of lean beef surimi, measured by compression test, were similar to those of pork surimi. However, they also found lean beef surimi gels were slightly harder than the pork surimi gels if both of them are at the same protein level. Salt soluble protein content of lean beef surimi was 7.8 %, compared with 7.9 % for pork surimi and both are higher than the salt soluble protein content (3.6%) of fish surimi (Knight, 1992).

### **Mutton surimi**

Limited research on the production of mutton surimi has been done at this stage. However, because New Zealand is a major sheep meat exporting country, every effort should be made to upgrade low value sheep meat off-cuts and one potential way of doing this is to produce mutton surimi. A group of scientists at the Meat Industry Research Institute of New Zealand (MIRINZ) have evaluated the feasibility of producing mutton surimi since 1987 (Knight, 1992).

MIRINZ used two methods for mutton surimi production. Firstly, Torley et al.(1988) suspended the mutton mince in water, allowing the fat to float and to be removed prior to centrifugation. This method reduced the fat from 30 % to 5 ~ 10 % and the mutton surimi contained 75 % moisture. In the second method, they chopped the mutton to finely particles prior to mixing with water. This time they could produce the mutton surimi with only 1 % fat and 80 % moisture (Knight, 1992).

Torley et al. (1988) conducted frozen storage trials on the mutton surimi, and found that after the first week the gel strength fell by about 20 %, even when cryoprotectants were used. However, after the first week, little change in gel strength was detectable with additional frozen storage.

## **Poultry surimi**

Mechanical deboning of poultry began in the 1950's, and provided a way of dealing with over supply of necks, backs, thighs, and drumsticks. However, mechanically deboned poultry meat (MDPM) contains higher heme and fat content than hand deboned poultry (Froning, 1981). Additionally, the darker colour, poor texture and short storage life have all limited its usage in poultry meat products directed towards the more desirable white meat market. Surimi processing of MDPM has generated considerable interest and has demonstrated the potential for new poultry products (Ball, 1988; Dawson et al. 1988).

The surimi processing of MDPM involves a repeated washing process with an aqueous solution for removing heme pigments, fat and other undesirable substances. The beneficial effects of surimi processed MDPM include increased myofibrillar protein concentration and removal of colour of the washed meat. The higher content of the myofibrillar proteins in the washed poultry meat has been shown to substantially improve its textural properties, thereby increasing the value of MDPM (Elkhalifa, et al. 1988; Dawson et al. 1988; Froning and Niemann, 1988).

Froning and Johnson (1973) reported that most of the pigment in MDPM is loosely held and can be removed readily by water washing followed by centrifugation.

Phosphate buffer was used by Warris (1976, 1977) to extract pigment. He found maximum extraction could be achieved by buffers having a pH above 6.8. Hernandez et al. (1986) studied the extraction of pigment from turkey meat with phosphate buffers at pH 6.4, 6.8, 7.2 and 8.0. They found pH 8.0 produced the lightest coloured surimi.

Dawson et al. (1988) and Ball and Montejano (1984) all used bicarbonate buffer (pH 8.5 and 8.45 respectively), acetate buffer (pH 4.5 and 5.25 respectively) and tap water to remove the pigments from chicken meat. Both experiments revealed that bicarbonate buffer was more effective in removing pigment from chicken meat than washing with acetate buffer or neutral tap water.

Yang and Froning (1992) washed mechanically deboned chicken meat (MDCM) with tap water, 0.5% sodium bicarbonate, 0.5% sodium phosphate buffer and 0.5% sodium chloride. They found all the selected washing solutions affected the lightness, water holding capacity and textural properties. The scanning electron microscopy revealed that washed meat showed a fibrous protein network structure resulting from protein gelation.

#### **2.2.3.3. Quality evaluation of meat surimi**

Criteria for assessing the quality of meat surimi have been proposed by Ockerman and Hansen (1988, Table 7). Knight (1992) suggested that in addition to the physical and chemical tests listed in Table 7, the microbiological status of surimi should be assessed at the same time.

Meat surimi quality is based on colour/blandness of flavour (affected by impurities) and gel forming ability (actin and myosin properties). Other tests which should be performed are chemical (pH and moisture) and physical (drip loss and viscosity).

Gel-forming ability is assessed by the use of a standard gelling method after which the gel is tested for strength by a special gel-testing instrument (Okada gelometer), which has been developed together with sensory testing (Suzuki, 1981). The sensory tests include folding the gel between the thumb and index finger and looking for the extent of cracking, and a bite test (Hall, 1992). The gel quality so measured will reflect:

Table 7: Properties Proposed for Grading Meat Surimi	
Chemical and visual	
	Moisture level
	pH
	Whiteness - Hunter colour meter
	Impurities - black skin and bones
Physical properties	
	Expressible drip - pressed
	Viscosity - in 3/5 % NaCl solution
Gel forming ability	
	Gel strength - plunger
	Folding test - crack when folded
	Firmness - sensory
	Chewiness - energy used with repeated compressions
	Elasticity - tensile force to break sheet
	Water binding - slope of gel strength versus moisture
Frozen storage	
	Freeze - thaw cycles - pressed fluid

Source: Ockerman and Hansen, 1988.

- (1) the quality of the raw material;
- (2) the effect of the washing cycles in concentrating the actin and myosin;
- (3) any changes brought about by frozen storage ( Hall, 1992 ).

Very limited information is available on the potential source of microbial contamination during commercial surimi production. Himelbloom et al. ( 1991 ) reported that freezing and thawing of surimi reduced microbial counts to 45 ~ 67 % of the prefrozen load; however, the composition of the microbial flora was maintained. They also said plant environment was a potential source of microbial contamination. Strict hygienic control of plant environment would help to improve the quality of meat surimi.

Though the proposed meat surimi grading system of Ockerman and Hansen ( 1988 ) does not include the fat content, Dawson et al. ( 1988 ) have found high thiobarbituric acid ( TBA ) values from chicken surimi produced in a decanter-centrifuge based process. They believed that the amount of fat remaining in surimi will influence the extent of rancid flavour development in both red meat and poultry surimi.

Textural quality improvement in fish surimi has been achieved by vacuum chopping and the incorporating egg white or potato starch. This procedure also reduced the extent of rancidity development ( Verrez et al. 1988; Babbitt and Reppond, 1988 ). A similar procedure could be applied to meat surimi production.

#### 2.2.3.4. Utilization of meat surimi

There are many potential uses of red meat and poultry surimi. It can be divided into three categories ( Knight et al. 1990 ):

##### (1) Traditional meat products

The addition of meat surimi to traditional meat products, e.g. sausages, burgers, has shown improvements in rheology and shelf-life stability, compared with other raw material used in traditional products. Jelen et al. ( 1982 ) found that addition of up to 20 % poultry surimi did not significantly alter the quality of luncheon meat. Caldironi and Ockerman ( 1982 ) produced a sausage at the level of 15 % beef surimi, and found it very acceptable.

##### (2) Novel meat products

Knight et al. ( 1990 ) said meat surimi could be fabricated into similar shaped and sized products as crab sticks which at present are produced from fish surimi. Meat surimi could be used as an ingredient in a range of novel meat products.

##### (3) Others

Scott ( 1988 ) has listed a large number of products in which meat surimi would be used as a major ingredient. These include: beverages, baby foods, bakery products and soup.

Martin ( 1986 ) said that the US consumer was generally well disposed to surimi based products, seeing them as wholesome foods with many recipe opportunities, with even the additives used in the production of final products being acceptable. This unique advantage of surimi should be used as a positive element in their marketing!

## 2.3. Functional Testing

### 2.3.1. Water holding capacity ( WHC )

The water-uptake property of meat proteins is a result of protein-water interactions and may result in swelling of the protein. This water-uptake property of meat proteins has been recognized as one of the most important features of meat. Tenderness, juiciness, colour, taste, shrinkage on cooking, and drip on freezing and thawing all appear to be directly related to this property of meat ( Deatherage, 1955). Factors affecting this property are amino acid composition, pH, solution ionic strength, particle size, protein concentration and surface topography ( Hermansson, 1986). Various terms have been used to describe the water-uptake properties of meat ( Hall, 1992 ): (1) water absorption capacity ( WAC ); (2) water binding capacity ( WBC ); (3) water holding capacity ( WHC ); (4) water hydration; and (5) swelling capacity. These terms have their own specific definitions.

Since no methods are available for determining the hydration of proteins of meat directly, the WHC is obtained by subtracting free water content from total moisture content ( Asselbergs and Whitaker, 1961 ). The total moisture content is determined by the standard oven drying technique ( 8 hours, 110 °C ). The free moisture content of meat is commonly determined by either a hydraulic-pressure technique ( for raw meat, Wierbicki and Deatherage, 1958 ), or a centrifugal-force technique ( for cooked meat, Wierbicki et al. 1957 ), or a filter paper press method ( Grau, 1978; Bechtel, 1986; Barge et al. 1991 ). Asselbergs and Whitaker ( 1961 ) found that the three methods produced different WHC values for the same meat samples. However, differences in relative WHC can be determined provided the same WHC technique is used.

Hamm (1957) reported that WHC could be increased by the addition of salt and water. He found an approximate 80 % increase in WHC with 60 % added water and 8 % sodium chloride. With 5 % sodium chloride and no added water, WHC increased by 25 %. The elevated salt concentration may explain some of the difference; however, the water content effect should be considered, also. Chung and Lee (1991) stated that the increase in WHC resulting from the addition of sodium chloride was caused by an increase in negative charges ( " salting-in effect " ) on the protein molecules which in turn resulted in an increase in loosely bound water and so enhanced the WHC.

Though many researchers have reported that pH is the primary factor influencing WHC of meat proteins ( Hamm, 1957; Wierbicki et al. 1957; Gillett, et al. 1977; Acton, et al. 1983; Offer and trinick, 1983; Trout and Schmidt, 1984; Paterson et al. 1988 ), Beuschel et al. ( 1992 ) found that the WHC of wheat protein concentrate heated to 90 °C showed no difference at pHs 6.0, 7.0, and 8.0 respectively.

Yang and Froning ( 1992 ) used tap water, sodium chloride, sodium phosphate and sodium bicarbonate to wash mechanically deboned chicken meat ( MDCM ). They found tap water, sodium chloride and sodium phosphate all increased the WHC of washed MDCM, the one exception being sodium bicarbonate. They stated that the reason for the increase in WHC was due to the elevated concentration of myofibrillar proteins after leaching out of fat and sarcoplasmic proteins, so that a greater amount of water could be held ( Hamm, 1960; Yang and Froning, 1992 ).

### 2.3.2. Gel Strength

Gel formation occurs by intermolecular bonding, which gives a solid matrix holding water, with flavour and textural implications, through a well ordered tertiary network (Hall, 1992). Busk (1984) has discussed the nature and terminology of gel formation, which may involve a combination of heat, salts, pressure and pH to achieve the network. Many food materials give gels, particularly carbohydrates such as starch, pectins, alginates and plant gums. Proteins that have been investigated for gelling include muscle, blood, egg, soya and milk proteins (Ziegler and Acton, 1984; Gossett et al. 1984; Schmidt and Morris, 1984). Gels have been formed in many ways and assessed by a range of strength tests, which include resistance to penetration, breaking strength, gravimetric methods. Machines such as the Instron Universal Testing machine apply forces in a controlled manner and with a variety of shaped plungers and give a force/resistance curve that is said to describe the gel texture in objective terms (Hermansson, 1982).

The gel strength of surimi and further processed fish and meat products has been shown to be influenced by the freshness of the meat and fish that the products were made from. For instance, Okada and Tamoto (1986) found that surimi produced from frozen fish had a gel strength considerably lower than the top grade ship-processed surimi. Scott et al. (1988) also found that freezing and thawing pollock resulted in surimi with significantly lower gel strength than that from fresh pollock. Park et al. (1987) reported that sausage batters manufactured from pre-rigor beef showed significantly higher gel strength than those prepared from post-rigor beef.

The storage condition and process followed to manufacture surimi have been shown to affect the gel strength. Hsu (1989) demonstrated that the gel strength of surimi products was significantly affected by storage (for more than six months) and the four major unit operations (leaching, grinding, setting and heating).

Yang and Froning (1992) washed mechanically deboned chicken meat (MDCM) with tap water, sodium chloride, sodium phosphate and sodium bicarbonate. They found that there was a profound increase in the gel strength of heat induced washed MDCM as compared to the unwashed MDCM. However, the increase for the sodium bicarbonate treatment was lower than that of other washed meat. The above authors argued that they had expected extensive hydration with the bicarbonate washing treatment and this extensive hydration was expected to decrease the gel strength of the washed MDCM as compared to the other treatments. Furthermore, because bicarbonate is a reducing agent which interferes with -SH bonding of meat proteins, this reduction in gel strength was not unexpected (Niwa and Musato, 1971).

Gel strength of surimi-based products is affected not only by the quality of the surimi but also by the ingredients incorporated during its preparation (Lee, et al. 1992). Ascorbate is being considered as a potential gel-strengthening ingredient based on reports by Yoshinaka et al. (1972) and Nishimura et al. (1990). Lee et al. (1992) also demonstrated that sodium ascorbate significantly improved gel cohesiveness and sensory firmness of fiberized products with maximum strengthening effect at a 0.2% level. The above authors explained this gel strengthening effect as the formation of S-S bonds through oxidation of sulfhydryl (-SH) groups by reducing oxidized ascorbate (Yoshinaka et al. 1972; Nishimura et al. 1990; Lee et al. 1992).

Tanchotikul et al. (1989) found that the addition of 3.5 % surimi to restructured beef roasts did not improve tensile strength. They said that the small amount of surimi may have restricted any potential for improved functionality. Therefore, they suggested that higher levels of surimi might be effective, but unacceptable fish flavour would have to be prevented or masked.

### 2.3.3. Protein solubility

Protein solubility is usually considered the premier functional property because of its relevance to other properties such as viscosity, gelation, foaming and emulsification. Solubility of the protein molecule is often a pre-requisite for these other properties to be observed ( Hall, 1992 ). Protein solubility is an indication of whether or not denaturation has taken place in the myofibrillar proteins. A decrease in solubility of myofibrillar protein is observed with the establishment of rigor mortis in warm blooded animals ( Bate-Smith, 1948; Zender, 1958; Migita, 1961 ).

A method commonly used to determine protein solubility is to measure the quantity of myofibrillar protein extracted from the muscle by salt solution with 0.45 ~ 0.6 ionic strength ( Suzuki, 1981 ). Several parameters are known to affect protein solubility. These include pH, temperature, ionic strength and the presence of other materials capable of binding to the protein ( Hall, 1992 ). Konagaya et al. ( 1978, 1979 ) found that low pH and high temperature cause protein denaturation resulting in low solubility ( **Fig. 2** ).

Kenney and Hunt ( 1990 ) said that although the maximal salt concentration ( ionic strength) necessary to optimize protein dissolution is not acceptable from a sensory standpoint, this obstacle can be overcome by preblending. Preblending involves subjecting the lean part of the product formula to all of the salt that would be added to the product. Following a holding period of 8 ~ 24 hours, this mixture was further blended to an acceptable salt level with the remaining raw material specified by the formulation. This combination of an elevated salt concentration ( 4 ~ 6 % sodium chloride ) and prolonged exposure time was conducive to more efficient use of the potential functional salt - soluble myofibrillar proteins ( Kenney and Hunt, 1990 ). More recent industry practices involve formulating preblends based on composition and subsequently correcting to the product's target composition, depending on the fatness or leanness of the preblend. LaBudd and Selfridge ( 1981 ) found this practice streamlines production and reduces time and labour requirements.

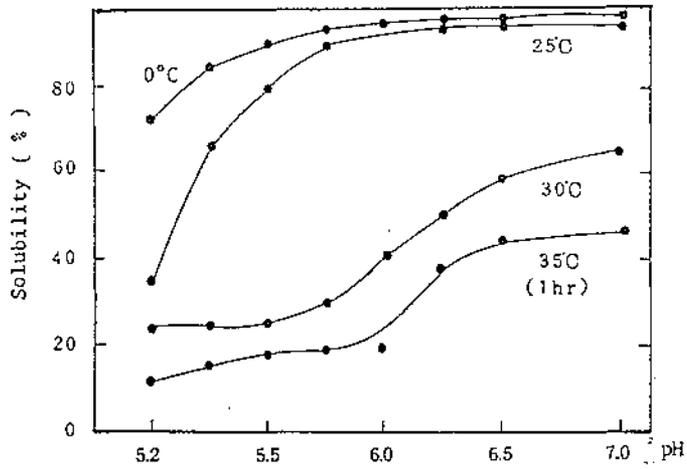


Fig 2: Denaturation of myofibrillar protein as influenced by temperature and pH ( Source: Konagaya et al. 1978 ).

## 2.4. Rationale for Present Study

The literature indicated that three potential fat extraction procedures could be used for the present project. These included supercritical fluid extraction (SFE), solvent extraction and various washing procedures.

Supercritical fluid extraction was rejected for the following reasons:

- (1) The high capital cost of plant (Reid, 1993).
- (2) High operating costs (Reid, 1993).
- (3) The fact that there is little or no information on the use of SFE for the extraction of fat from animal tissues. Supercritical fluid extraction though energy efficient and environmentally friendly is unlikely to be adopted by New Zealand meat companies given the lack of poor data to make commercial decisions (Reid, 1993).

Similarly, solvent extraction was rejected for the following reasons:

- (1) Solvent(s) often extract non-lipid materials (Christie, 1982).
- (2) Protein functionality can be jeopardised by various solvents (Chung and Ferrier, 1991). Bobush et al.(1985) even suggested that certain kinds of solvents, e.g. chloroform-methanol, are inappropriate for dealing with animal tissues.

Consequently, it was decided that some washing procedure should be used in the current project to remove fat from MDB. Previous research has indicated that several washing solutions are available for surimi processing of MDM: tap water (Ball and Montejano, 1984); phosphate buffer solution with pH 5.8 to 8.0 (Hernandez et al. 1986; Elkhalfa et al. 1988); 0.5% sodium bicarbonate solution (Ball and Montejano, 1984; Dawson et al. 1988, 1989); and 0.1M sodium chloride solution (Froning and Niemann, 1989).

The previous studies indicated that alkali washing achieved more effective fat removal than just tap water or phosphate buffer. Furthermore, Yang and Froning (1990) observed that alkali washing more effectively removed the heme pigments and increased the lightness of the washed meat. Whilst acidic washing was found to cause myofibrillar protein denaturation (Konagaya et al. 1978). Therefore, alkali washing procedures seemed to be a more appropriate area of research for the current project.

Several investigators have studied the feasibility of alkali washing procedure to remove fat from MDM (Duerr and Earle, 1974; Young, 1976; Jelen et al. 1978, 1979, 1982; Lawrence, 1981; Lawrence et al. 1982; Palka et al. 1985; McCurdy et al. 1986). However, none of the studies have systematically assessed the effectiveness of a range of alkalies over the pH range pH 7.0 ~ pH 11.5.

The aim of the current study was to determine the effectiveness of a number of alkalis over the pH range 7.0 ~ 11.5 on:

- (1) The removal of fat from MDB.
- (2) The protein functionalities of the resulting meat surimi, and finally
- (3) The proximate composition of the various by-products of the surimi processing.

### 3. MATERIALS AND METHODS

#### 3.1. RAW MATERIALS

Two twenty kg cartons of frozen mechanically deboned beef (MDB) were obtained from Richmonds Freezing Co. Ltd. (Hastings). The cartons were thawed in a chiller (3 °C) and then blended in a food processor for three minutes to firstly reduce the particle size to 2 ~ 3 mm and secondly to ensure that the twenty kg blocks were of a uniform composition. The blended material was then packed into fifty gram bags and stored in a -30 °C freezer for later use.

On the day before a run the required number of bags of pre-blended MDB were removed from the freezer and tempered in a cold room (3 °C) overnight.

#### 3.2. ANALYTICAL TECHNIQUES

##### 3.2.1. Proximate analysis of MDB and by-product streams

MDB was analyzed by AOAC (1980) methods for its moisture, fat, protein and ash content. The by-product streams of surimi processing were also analyzed by AOAC (1980) methods for their solid contents (fat, protein and ash).

##### Moisture

Moisture assays were conducted on 5 g duplicate samples dried in a forced air oven at 97 ~ 99 °C to constant weight.

##### Fat

Fat content was determined on 5 g duplicate samples with Soxhlet apparatus, using diethyl ether (boiling point, 34.5 °C) as solvent. An eight hour continuous extraction procedure was used. At the end of the extraction cycle the samples were allowed to cool overnight. A rotarvapor was used to remove the solvent from the Soxhlet flasks after which time the flasks were dried in a 97 °C oven to constant weight.

## **Protein**

Nitrogen content was assayed on 1 g duplicate samples via the automated Kjeldahl method (digested with concentrated sulphuric acid for one hour, distilled into a 20% solution of boric acid for six minutes then titrated with 0.1 N hydrogen chloride). Protein content was calculated by multiplying nitrogen content with 6.25.

## **Ash**

Ash was determined by a stepwise charring of 5 g duplicate samples at 200 °C for two hours, 300 °C for two hours, and 600 °C for eighteen hours, until constant weight was achieved.

### **3.2.2. Water holding capacity ( WHC )**

WHC was determined by the method described by Holmquist et al. (1984). One filter paper was dried then weighed precisely. Half a gram of protein sludge was placed on the dried filter paper which was then placed between two plates. A five kg weight was then placed on top of the plates and this weight applied for five minutes. The weight and plates were removed and a pair of tweezers used to remove the sample. Filter paper was immediately weighed and the WHC was expressed as grams of water held per gram of protein sludge.

### **3.2.3. Gel strength**

Gel strength was determined by pulling heat set surimi rings to breaking point with the aid of an Instron Universal Testing Machine ( Model 4500 ).

The protein sludge obtained at the bottom of the centrifuge bottles after centrifugation was mixed with 1.5 % sodium chloride and 0.3 % tetra potassium pyrophosphate for one minute in a beaker. The mixture was then placed in a PVC mould to produce rings with an internal diameter of 0.8 cms and an external diameter of 1.4 cms ( i.e. 0.6 cms thickness ). The filled moulds were placed in a vacuum bag and evacuated for two minutes at twenty-eight inches of mercury to remove any entrained air. After evacuation the mixture was smoothed and re-evacuated and the bags heat sealed. The plastic packaged moulds were heated for twenty minutes at eighty °C in a water bath and then allowed to cool overnight.

The meat-rings were removed from the moulds for the pulling-apart tests on the Instron machine at the cross speed of 50mm/min. The heat gel strength was recorded as the maximum force (newton) required to pull apart the gel as read off the force deformation curve.

#### **3.2.4. Protein solubility**

The protein content of each sample was determined first. Then ten grams of sample were taken and placed in a centrifuge tube. Thirty mls of distilled water were added plus enough salt to make the overall salt concentration either 0.2M or 0.6M. The mixture was then stirred and left overnight. The tubes were centrifuged at nine thousand rpm for fifteen minutes. The weight of supernatant was recorded and protein content of the supernatant was determined. The 0.2M salt treatment provided a measure of the sarcoplasmic protein concentration whilst the 0.6M salt treatment provided a measure of both the sarcoplasmic and myofibrillar protein content. The difference in protein content between the 0.2M and 0.6M treatments gave the myofibrillar protein solubility of the sample.

### **3.3. FAT SEPARATION PROCEDURE**

#### **3.3.1. Preparation**

The following eight alkalis were used in the trial: sodium carbonate ( $\text{Na}_2\text{CO}_3$ ), sodium bicarbonate ( $\text{NaHCO}_3$ ), sodium hydroxide ( $\text{NaOH}$ ), potassium carbonate ( $\text{K}_2\text{CO}_3$ ), potassium hydroxide ( $\text{KOH}$ ), ammonia water ( $\text{NH}_4\text{OH}$ ), magnesium carbonate ( $\text{MgCO}_3$ ) and calcium hydroxide [ $\text{Ca}(\text{OH})_2$ ].

### **3.3.2. Washing and Centrifugation**

A number of mechanically deboned slurries were prepared using fifty grams of MDB mixed with 400 mls of tap water (ratio 1:8). The pH of the mixtures were adjusted to pH7, 8, and 9 respectively with one of 20% alkali solutions prepared previously. A control sample containing just fifty grams of MDB and 400 mls of tap water was also prepared at the same time. Duplicate samples of each treatment were used for all experiments. Once the pH of each mixture had been adjusted, the samples were held in a cold room ( 3 °C ) for one hour. The mixtures were then transferred to centrifuge bottles and centrifuged at 9,000 rpm ( Sorvol model: RC5C ) for fifteen minutes. The supernatant was then filtered through a pre-weighed filter paper ( Calbiochem Miracloth, quick filtration material for gelatinous grindates ) to collect the fat. The fat remaining on the sides of the centrifuge bottle was recovered using three previously weighed balls of cotton wool soaked in ethanol. The filter paper and cotton wool balls were then placed on a watch glass and dried in an oven at 97 °C until constant weight. The difference in weight between the original filter paper and cotton wool weight and the final weight of material on the watch glass provided the weight of recovered fat.

The solids content, protein, ash and fat content, were determined for the fat layer, supernatant and protein sludge according to the methods listed earlier in this chapter. The weight of the mixture prior to centrifugation plus the weights of the fat layer, supernatant and protein sludge layers were also recorded with the objectives of establishing where the various components of the original MDB mixture ended up( i.e. protein, fat, ash and total solids ).

### **3.3.3. Preparation for functional testing**

The protein/sludge mixture collected on the bottom of each centrifuge bottle was removed from the centrifuged bottle, weighed and then placed in a plastic bag. The bag was stored in a -30 °C freezer until needed for the various functional tests, i.e. gel strength, protein solubility, and water holding capacity.

## 4 RESULTS AND DISCUSSION

### 4.1 FAT SEPARATION

#### 4.1.1 The Effect Of Various Alkalis And pH Treatments

##### On Fat Removal From Mechanically Deboned Beef

A summary of the fat removal results from a number of different alkali treatments at three separate pH's (i.e., pH 7.0, 8.0, and 9.0) have been presented in **Table 8** and **Figure 3**. An examination of all the results clearly shows that there was no consistent improvement in fat removal efficiency with increasing pH, i.e. with some alkalis, fat removal efficiency improved with increasing pH, whilst for other alkalis the converse was true. However, the pH 7.0 results for practically all alkali treatments were significantly different to the respective pH 9.0 treatments ( 95% probability), irrespective of trend. The results showed that treatment with some alkalis at pH 7.0 brought about only marginal improvements in fat removal compared to the non-pH adjusted controls, and that for most alkali treatments pH adjustment had no significant effect on fat removal from slurried mechanically deboned beef.

The results indicated that for strong alkalis, i.e., NaOH and KOH, fat removal efficiency improved as the pH of the mechanically deboned beef slurry was raised from pH 7.0 to pH 8.0. However, there were no significant improvements in fat removal efficiency at pH's greater than 8.0. A comparison between the non-pH adjusted control and the strong alkali treatments showed that the fat removal efficiencies achieved in the pH 8.0 and 9.0 treatments were no better than for the control, and that the pH 7.0 results for both strong alkalis were significantly worse than the controls, i.e., that pH adjustment of mechanically deboned beef slurries with strong alkalis were at best no better than the distilled water controls.

With some of the weak alkalis, i.e. sodium carbonate, and ammonium hydroxide fat removal efficiency declined with increasing pH. In the case of sodium carbonate, a decline in fat removal efficiency occurred between the pH 8.0 and 9.0 treatments, whilst for the ammonium hydroxide treatments fat removal efficiency deteriorated between pH 7.0 and pH 8.0. The pH 7.0 treatments of both intermediate alkalis were both significantly better in terms of fat removal efficiency than the controls. The remaining alkalis, i.e. calcium hydroxide, potassium carbonate, and sodium bicarbonate, appeared to have no significant effect on fat removal compared to the non-pH-adjusted controls (pH 6.5), irrespective of pH.

Table 8: Fat removal efficiency of MDB with various pH and alkaline treatments									
pH & alkali	KOH a	NaOH b	Ca(OH) <sub>2</sub> c	K <sub>2</sub> CO <sub>3</sub> d	Na <sub>2</sub> CO <sub>3</sub> e	NH <sub>4</sub> OH f	NaHCO <sub>3</sub> g	MgCO <sub>3</sub> h	Control i
pH 7 (O)	71.3 ( )	76.65 ( )	80.62 di (X)	79.54 cgi ( )	85.70 ( )	84.02 ( )	78.94 di (X)	75.28	79.85 cdg
pH 8 (X)	78.77 gi (▲)	81.51 cf (▲)	80.33 bfi (O)	82.74 ( )	84.35 ( )	80.62 bci (▲)	78.65 ai (O)	NIL	79.85 acfg
pH 9 (▲)	78.23 cd (X)	81.15 ef (X)	78.28 ad ( )	78.19 ac ( )	80.88 bfi ( )	81.37 be (X)	NIL	NIL	79.85 e

\* Every letter labelled under the alkali represents the alkalie treatment. Means within a row having the same letter as labelled under the alkali are not significantly different (P>0.05).

\* Symbols, O, X and ▲, represent pH 7, pH 8 and pH 9 respectively. Means within a column having the same symbol as labelled under the pH, in the "( )", are not significantly different (P>0.05).

\* NIL---- the alkali can not reach the pH.

\* The pH of control which goes with the other treatments is 6.25.

\* Values represent means of duplicate observations.

### Fat Removal Efficiency of MDB with various pH and alkaline treatment

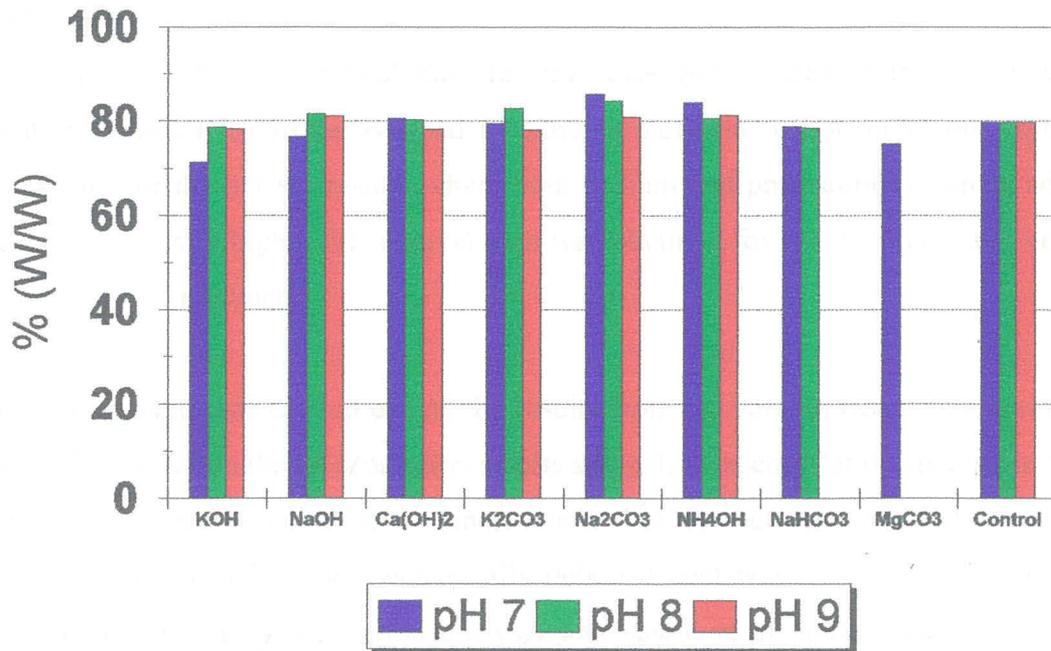


Fig 3: Fat removal efficiency of MDB with various pH and alkaline treatments

A comparison of all the pH 7.0 fat removal results for the eight different alkalis showed that fat removal efficiencies were highest for sodium carbonate and ammonium hydroxide and lowest for potassium hydroxide followed by magnesium carbonate and then sodium hydroxide.

The fat removal efficiencies for potassium carbonate, calcium hydroxide and sodium bicarbonate were the same as the results for the water (pH 6.25 distilled water) control treatments.

An examination of the fat removal data for the three pH's indicated that there was no consistent trend which could be ascribed to either a particular cation or anionic species - except possibly for the pH 8.0 results where both sodium and potassium carbonate addition resulted in significantly higher fat removal than was obtained for the distilled water controls and the other alkali treatments.

A correlation analysis was carried out on the results from the various treatments to establish the relationships between the fat separation results and pH. The correlation analysis indicated that there was a positive correlation, albeit small (0.475), between pH and fat removal efficiency, i.e., as the pH of the mechanically deboned beef was raised so the fat removal efficiency improved. This correlation analysis was based on all alkali treatments. The overall correlation between pH and fat removal was low because some alkali treatments either showed no improvement with increasing pH, whilst others indicated a contrary trend, i.e. as pH increased so fat removal efficiency decreased.

It would appear from the results that only at pH 7.0 could the various alkalis exert some effect on the centrifugal removal of fat from the mechanically deboned beef slurry. It is very difficult to compare the results of this study with other studies as no other researchers have systematically examined the effect of various alkalis on the removal of fat from mechanically deboned meat. However, Ball et. al. (1986) and Dawson et al. (1988) showed that the extraction of lipid and meat pigments from MDC was higher at pH 8.5 (sodium bicarbonate solution) than for a distilled water control (pH 6.8). Hernandez et. al. (1986) also showed that pigment removal increased with increasing pH of phosphate washing solutions. No consistent fat removal trends with increasing pH were observed in the current study. The current study indicated that there was little advantage in increasing solution pH above 7.0 as the highest fat removal was achieved with a pH 7.0 sodium carbonate solution.

It was observed that the addition of strong alkalis (e.g. sodium hydroxide and potassium hydroxide) to the mechanically deboned beef slurry (pH 7.0) resulted in significantly lower fat removal results than the water controls. This could be attributed to either increased swelling of the myofibrillar proteins, which then passively trapped the free fat particles and so prevented their centrifugal separation or it could be attributed to increased solubilisation of the myofibrillar proteins by the strong alkalis and this soluble protein then coated the free fat particles to produce an emulsion which was in turn difficult to separate centrifugally. The results could also be due to a combination of the above listed causes.

The series of experiments showed that much of the fat in mechanically separated beef was "free" fat (78 - 85% of the total fat) which could be readily recovered by the addition of cold water followed by centrifugation. Fat removal could be enhanced by the Addition of sodium carbonate solution to raise the solution pH to 7.0, though the increased fat separation was marginally improved in comparison to the controls. A portion of the fat appeared to be "bound" fat, and this could not be separated from the myofibrillar/collagen fraction by centrifugation. This bound fat could be either emulsified fat or fat which was still held in fat cells embedded in the collagen matrix of the mechanically deboned beef. According to other researchers' work the fat emulsion should have been broken at pH's above 8.0 (Ball et al., 1986; and Dawson et. al.; 1988). The current study failed to show that fat removal increased with increasing pH and so largely negated the idea that the poor fat recoveries were due to the trapping of the "bound" fat in an emulsified state which in turn meant that the "bound" fat should be associated with the collagen fraction. The concept that the "bound" fat was associated with the collagen fraction could be readily tested by carrying out an experiment where the meat surimi/collagen fraction was screened after centrifugation to remove the collagen fraction and then carrying out fat analyses on the material caught on the screen and the material passing through the screen. The results of such an experiment are reported later in this chapter.

#### 4.1.2 Distribution of Various MDB Constituents in Various Layers After Centrifugation

A generalised flow process chart of the process for separating MDB is shown below **Fig 4**: A mass balance for the controls has been presented in **Tables 9** and **10**. The total solids content of the starting mechanically deboned beef was 43.26% which consisted of 27.62 grams of fat, 14.70 grams protein and 0.94 grams of ash per 100 grams of MDB. The figures compare favourably with data quoted in the literature, namely: 39.7% - 55.1% for pork; 34.3 - 56.2% for cow; 40.2 % - 52.7% for mutton; 30.7% - 37% for chicken; and 26.4% - for turkey; 22% - 30% for fish. The figures vary extensively because they are largely dependent on the pre-processing which has occurred to the raw materials entering the deboner, age of the animal, settings on the deboner, and type of raw materials being processed. (Field, 1974).

## Fig 4. FLOW CHART OF MDB SEPARATION PROCESS

Thawed MDB  
(43.26% solids)

Added 8 parts water (10°C) to 1 part MDB  
(pH modification with alkali)

Stirred for 30 minutes

Centrifuged at 9000 r.p.m. for 15 minutes

	Fat fraction (16.75% solids)	Sarcoplasmic fraction (6.64% solids)	Myofibrillar/collagen fraction (19.87% solids)
Analyses(dry weight basis):			
Fat	98.9 %	41.3	41.8
Protein	1.1%	48.2	57.0
Ash	0	10.5	1.2

**Table 9: Proximate analysis of MDB**

Moisture %	Fat %	Protein %	Ash %	Total 99.47%
56.21	27.62	14.70	0.94	<b>Total solid 43.26 %</b>

**Table 10: Solid content distribution of MDB in various layers after centrifuge**

	Floating layer	liquid	protein sludge	Total %
Fat %	16.57	2.74	8.31	27.62
Protein %	0.18	3.20	11.32	14.70
Ash %	0	0.70	0.24	0.94
<b>Solid content%</b>	<b>16.75</b>	<b>6.64</b>	<b>19.87</b>	<b>43.26</b>

The mass balance indicated that 60% of the fat in the MDB was recovered in the fat fraction, 9.9 % in the sarcoplasmic fraction and the balance, i.e., 30.1 % of the fat in the meat myofibrillar/collagen fraction. Most of the protein, i.e., 77% was collected in the myofibrillar/collagen fraction, a further 21.8% of the protein in the sarcoplasmic fraction and the remainder, 1.1%, in the fat fraction. Almost 75% of the ash ended up in the sarcoplasmic fraction with the remainder in the myofibrillar/collagen fraction.

An examination of the figure above indicates that the fat fraction was largely composed of fat, i.e. 98.9% on a dry weight basis with the remainder consisting of a small amount of protein. The sarcoplasmic fraction consisted of almost an equal weight of fat and protein on a dry weight basis with the balance made-up of ash. The myofibrillar/collagen fraction consisted of 57% protein, 41.8% fat and the remainder ash. This represented a slight improvement on the original mechanical deboned beef which comprised 63.8% fat, 34% protein and 2.2% ash ( on a dry weight basis). As indicated in the previous section much of the fat was "bound" fat and little more of the fat could be removed by sequential washing (private communication). Clearly, if the myofibrillar/collagen fraction is to have some commercial value then the remaining fat must be removed in some way so that the resulting 'meat surimi' has similar proximate composition to the fish surimi now being traded internationally as a food ingredient.

#### **4.1.3 Modified Separation Process for Mechanically Deboned Beef**

The results of the experiment to establish whether the fat which appeared to be "bound" by the myofibrillar/collagen fraction was associated with the collagen fraction or whether it was held by the myofibrillar fraction have been presented in **Table 11**. The flow chart for the process is presented in **Fig. 5** below.

As can be seen from the above flow chart, the modified process was identical to the original process up to the myofibrillar/collagen fraction collection step. The additional steps were aimed at removing the collagen fraction from the myofibrillar protein fraction to determine which fraction was responsible for " binding" the fat in the collagen/myofibrillar fraction.

Table 11: MDM Fat content distribution in various kinds of protein			
	Collagen + Myofibrillar	Collagen	Myofibrillar
Control	18.47%	16.67%	1.30%
Na <sub>2</sub> CO <sub>3</sub> ( pH 7 )	13.15%	12.02%	0.90%

# Fig. 5 Flow Chart for Modified Separation Process for MDB

Thawed MDB

Added 8 parts cold water (10°C) to one part MDB

Stirred for 30 minutes

Centrifuged at 9000 r.p.m. for 15 minutes

Fat Fraction

Sarcoplasmic fraction

Myofibrillar/collagen fraction

Re-suspend in cold water  
2.5 parts water 1 part protein  
(alkali pH adjustment to pH 7.0)

Stirred for 20 seconds

Sieved through 2mm mesh

Centrifuged slurry at 4000 r.p.m. for 25 minutes

Wash water fraction

Myofibrillar protein fraction  
(Meat surimi)

A sodium carbonate (pH 7.0) treatment was compared with a distilled water control (pH 6.8) to establish whether alkali treatment resulted in an improved fat separation compared to the control. The results firstly provided clear proof that the "bound" fat was associated with the collagen and that once the collagen fraction was removed by sieving the resulting myofibrillar fat levels were very low (1.77% for the sodium carbonate treatment and 3.4% for the distilled water control, **Table 12**). These figures were similar to those reported by Knight (1992). The results also showed that an initial wash with sodium carbonate (pH 7.0) marginally improved the fat separation as shown above..

Only 0.9% of the original MDB fat ended up in the myofibrils obtained by the sodium carbonate treatment compared to 1.3% for the distilled water controls(**Table 11**), i.e. most of the original fat in the MDB was collected in the fat fraction and the collagen fraction leaving little fat in the myofibrillar fraction.

**Table 12: Solid contents of various proteins in modified fat extraction**

	Collagen + Myofibrillar		Collagen		Myofibrillar	
	Control	Na <sub>2</sub> CO <sub>3</sub>	Control	Na <sub>2</sub> CO <sub>3</sub>	Control	Na <sub>2</sub> CO <sub>3</sub>
Fat %	6.67	4.35	3.98	2.75	3.40	1.77
Protein %	13.45	12.50	5.13	4.81	39.87	32.95
Ash %	0.70	0.40	0.24	0.5	0.02	0.2
Solid Content%	20.82	17.25	9.35	8.06	43.29	34.92

## 4.2 FUNCTIONAL TESTS ON MYOFIBRILLAR/COLLAGEN FRACTION

### 4.2.1 Water Holding Capacity of Myofibrillar/collagen Fraction

The results of the various water holding capacity results of the myofibrillar/collagen fractions obtained from the various alkali extraction procedures have been summarised in **Table 13** and **Fig 6**. An examination of the results shows that there was no consistent increase in water holding capacity of the myofibrillar/collagen fraction with pH, the one exception was calcium hydroxide washes where the water holding capacity increased from 55.4% at pH 7.0 to 63.9% at pH 9.0. Most of the other alkali treatments either showed no increase with increasing pH (sodium hydroxide) and in many cases actually showed a decline in water holding capacity with increasing pH (sodium carbonate, potassium carbonate, ammonium hydroxide and potassium hydroxide). Ammonium hydroxide treatments showed the most significant decline with increasing pH - from a water holding capacity of 59.1% at pH 7.0 to 46.1% at pH 9.0. The distilled water controls in a majority of cases had a higher water holding capacity than most of the alkali treatments.

The experimental results were totally unexpected as they were completely different to the work of Hamm (1962), Ishioroshi et al., (1979) and others who have all shown that water holding capacity of meat increases with increasing pH. The filter paper method for determining water holding capacity (Grau, 1978; Bechtel, 1986; Barge, et al., 1991) was used in this study as it has been shown by Holmquist (1984) and the above listed authors to be a reliable method for determining water holding capacity, though producing different water holding capacity results compared to the centrifugal-force and hydraulic-pressure methods (Asselbergs and Whitaker (1961). The failure of the results to show any consistent increase in water holding capacity with increasing pH could be a result of at least two causes, the first connected with the lack of precision of the method and the second tied up with the method for producing the myofibrillar/collagen fraction. With some treatments the filter press method produced very high standard deviations and because of this imprecision it was almost impossible to actually say whether water holding capacity had actually increased or decreased with pH with any certainty. The myofibrillar/collagen fraction was collected at the bottom of either centrifuge bottle or tubes after centrifugation at 9000 r.p.m. for 15 minutes -

pH & alkali	KOH a	NaOH b	Ca(OH) <sub>2</sub> c	K <sub>2</sub> CO <sub>3</sub> d	Na <sub>2</sub> CO <sub>3</sub> e	NH <sub>4</sub> OH f	NaHCO <sub>3</sub> g	MgCO <sub>3</sub> h	Control i
pH 7 (O)	50.0 be ( )	48.7 a (▲)	55.4 dhi ( )	56.9 cfi ( )	51.9 agh ( )	59.1 d ( )	52.6 eh (X)	54.1 cegi	54.9 cdh
pH 8 (X)	42.8 ( )	53.7 cfgi ( )	53.1 bfgi ( )	46.9 e ( )	48.8 d (▲)	53.3 begi ( )	53.7 bcfi (O)	NIL	54.9 bcfg
pH 9 (▲)	46.3 ef ( )	49.3 de (O)	63.9 ( )	49.9 be ( )	48.4 abd (X)	46.1 a ( )	NIL	NIL	54.9

\* Every letter labelled under the alkali represents the alkaline treatment. Means within a row having the same letter as labelled under the alkali are not significantly different ( P>0.05 ).

\* Symbols, O, X and ▲ , represent pH 7, pH 8 and pH 9 respectively. Means within a column having the same symbol as labelled under the pH, in the " ( )", are not significantly different ( P>0.05 ).

\* NIL---- the alkali can not reach the pH.

\* The pH of control which goes with the other treatments is 6.25.

. Values represent means of duplicate observations.

### WHC of Myofibrillar/collagen Fraction from various pH & alkaline treatments

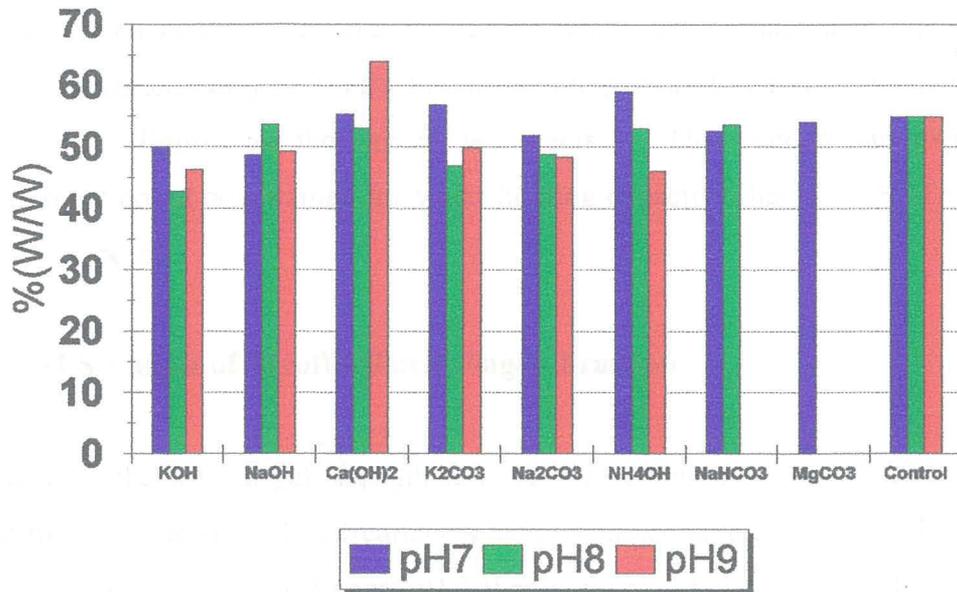


Fig 6: WHC of myofibrillar/collagen fraction from various pH and alkaline treatments

centrifugal force conditions which were greater than those prescribed for the centrifugal-force method for determining the water holding capacity of meat. As a consequence of the severe centrifugal-force conditions during the preparatory stage it could be expected that the myofibrillar/collagen fraction would give up the free water during centrifugation and as a consequence the results which were obtained in this study for the water holding capacity may be a reflection of sampling procedure rather than the actual water holding capacity figures for the various treatments. This may in part explain why no consistent trend could be obtained for water holding capacity when the pH of the extraction solutions was raised.

#### 4.2.2 Gel Strength of Myofibrillar/collagen Fraction

The results for the Instron gel strength tests on the various myofibrillar/collagen fractions obtained from the various alkali treatments have been presented in **Table 14** and **Fig. 7**.

With the exception of one treatment (pH 7.0 potassium carbonate ) it was found that the gel strength of all treatments was between 25% - 50% lower than the gel strengths of the distilled water controls. Moreover, gel strength appeared to fall with increasing pH for all alkali treatments, though the differences in gel strength with increasing pH were more marked with potassium hydroxide and potassium carbonate washes than for the other alkalis. No consistent trend with either added cation or added anion could be discerned in the results.

The gel strength results were more consistent with the results of other research workers. For instance, Yang and Froning, (1991) found that gel strength decreased with increasing pH, a similar result to the present study. Ishioroshi et al., (1979) also found that gel strength decreased with increased pH and they argued that the decreased gel strength of the high pH meat samples was a result of the increased swelling of the meat fibres at the higher pH's which in turn led to fewer meat fibres per unit area and hence a lower gel strength. In the present study it is difficult to say whether the reduction in gel strength with increased pH was a result of increased protein swelling because the water holding capacity results provided no consistent trend with increasing pH.

pH & alkali	KOH a	NaOH b	Ca(OH) <sub>2</sub> c	K <sub>2</sub> CO <sub>3</sub> d	Na <sub>2</sub> CO <sub>3</sub> e	NH <sub>4</sub> OH f	NaHCO <sub>3</sub> g	MgCO <sub>3</sub> h	Ctrl i
pH 7 (O)	2.210 f ( )	1.666 ceh ( )	1.775 bfh (X)	2.954 i ( )	1.449 b (X)	2.031 ach ( )	1.007 ( )	1.853 bcf	2.734 d
pH 8 (X)	1.060 bdf ( )	1.002 afg (▲)	1.701 e (O)	1.294 a (▲)	1.610 c (O)	1.018 ab ( )	0.743 b ( )	NIL	2.734
pH 9 (▲)	0.716 ef ( )	1.242 cd (X)	1.257 bd ( )	1.135 bce (X)	0.949 ad ( )	0.602 a ( )	NIL	NIL	2.734

\* Every letter labelled under the alkali represents the alkaline treatment. Means within a row having the same letter as labelled under the alkali are not significantly different (P>0.05).

\* Symbols, O, X and ▲, represent pH 7, pH 8 and pH 9 respectively. Means within a column having the same symbol as labelled under the pH, in the "( )", are not significantly different (P>0.05).

\* NIL---- the alkali can not reach the pH.

\* The pH of control which goes with the other treatments is 6.25.

\* Values represent means of duplicate observations.

### Gel Strength of Cooked Protein from various pH & alkaline treatments

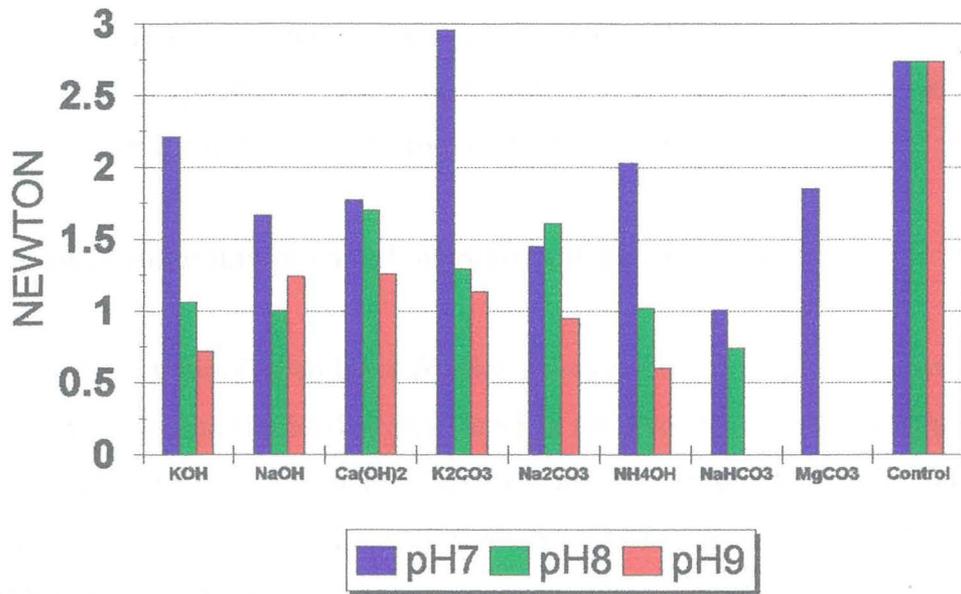


Fig 7: Gel strength of cooked myofibrillar/collagen fraction from various pH and alkaline treatments

The gel strength of the sodium bicarbonate treated MDB was noticeably lower for the pH 7.0 treatments compared to the other alkali treatments. Niwa and Musato, 1971 have argued that sodium bicarbonate acts as a reducing agent in meat systems and as a consequence interferes with the formation of sulphhydryl bonds which are known to be important for the development of meat gels at temperatures above 55°C. This interference of sulphhydryl bond formation could explain the lower than expected gel strength results for the sodium bicarbonate washed MDB compared to the other treatments.

#### 4.2.3 Protein Solubility of the Myofibrillar/collagen Fraction

In this study the myofibrillar/collagen fraction from each of the alkali treatments was obtained after centrifugation and the total protein level in the fraction was then determined for each treatment. Ten grams of the fraction was then dissolved in 0.2 M sodium chloride solution and a further 10 grams dissolved in an 0.6 M sodium chloride solution. The mixtures were then stirred and left overnight. The samples were centrifuged at 9000 rpm for 15 minutes and the protein content of the supernatant from each salt concentration was then determined.

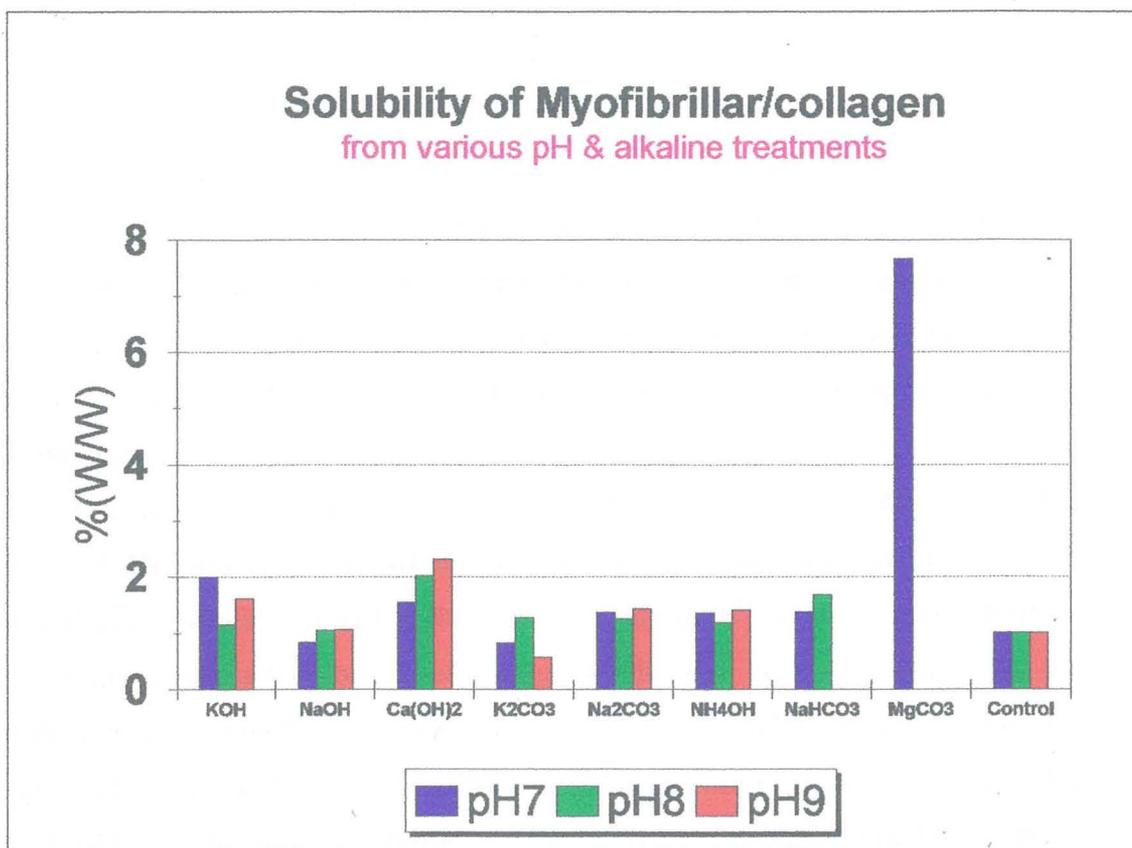
The protein content of the 0.2M salt solution gave a measure of the sarcoplasmic protein still trapped in the myofibrillar/collagen layer, whilst the 0.6M sodium chloride gave an indication of the total amount of soluble protein in the myofibrillar/collagen fraction. The difference between the 0.2M and 0.6M solutions provided an indication of the amount of soluble myofibrillar protein. The results for the soluble myofibrillar protein have been provided in **Table 15** and **Fig 8**.

The most striking result of the study was the significantly higher soluble myofibrillar protein obtained with the magnesium carbonate washed MDB compared to the other treatments - almost four times higher at 7.67% compared to the other treatments. Whilst there was no consistent trend evident for increased protein solubility with increasing pH across all alkali treatments, a majority of alkalis tended to increase protein solubility with increasing pH, if the pH 7.0 results are compared with the pH 9.0 results. Some alkalis, and in particular calcium hydroxide, showed a marked increase in protein solubility with increasing pH from 1.55% at pH 7.0 to 2.32% at pH 9.0. In some cases protein solubility actually decreased, as evidenced by the potassium hydroxide results where protein solubility dropped from 2%

**Table 15: Protein solubility of myofibrillar/collagen fraction from various pH and alkaline treatments**

pH & alkali	KOH	NaOH	Ca(OH) <sub>2</sub>	K <sub>2</sub> CO <sub>3</sub>	Na <sub>2</sub> CO <sub>3</sub>	NH <sub>4</sub> OH	NaHCO <sub>3</sub>	MgCO <sub>3</sub>	Control
pH 7	2.00	0.84	1.55	0.83	1.37	1.36	1.38	7.67	1.01
pH 8	1.16	1.05	2.02	1.28	1.25	1.19	1.69	NIL	1.01
pH 9	1.62	1.06	2.32	0.58	1.43	1.42	NIL	NIL	1.01

\* NIL----- the alkali can not reach the pH.



**Fig 8: Solubility of myofibrillar/collagen fraction from various pH & alkaline treatments**

at pH 7.0 to 1.62% at pH 9.0.

Protein solubility should increase with increasing pH, firstly because of the increased swelling of the protein which should occur as the pH of the protein is moved away from its isoelectric point (Dawson et al., 1988), and secondly because as the pH is increased so the ionic strength of the protein solutions is increased as the pH is increased with the respective alkalis - though the increase in ionic strength is not expected to be a major influence in this regard due to the small amounts of each alkali treatment that had to be added to increase the pH of the protein solutions.

However, in the present study the pH of the various solutions was changed right at the start of the MDB process. The MDB slurry was then stirred and centrifuged. Any protein solubilisation which occurred at the outset would not be picked up by the protein solubility test simply because the solubilised protein would have been collected in the sarcoplasmic protein fraction which was discarded. This could explain why pH adjustment had no significant effect on protein solubility as tested in the present study. In hindsight, the 0.2M and 0.6M sodium chloride solutions for the various treatments should have been pH adjusted by the respective alkalis to truly assess the effects of pH modification on protein solubility.

The intriguing result of the solubility test results were the substantially higher solubility results for the magnesium carbonate treatments compared to the other treatments and the controls.

The solubility results were substantially higher than for any other treatment and suggest that the magnesium carbonate treatment affected the myofibrils in some way enabling a substantially higher amount of myofibrillar protein to be extracted from the myofibrillar/collagen fraction compared to the other alkalis. The magnesium ion is known to play a significant role, as is the calcium ion in the contraction/relaxation process of myofibrils. It is possible that the high magnesium ion concentration, relative to physiological concentrations in the muscle, resulted in de-coupling of the actomyosin of the filaments and thus enabled more myofibrillar protein to be extracted from the fraction once the higher ionic strength (0.6 M) salt solution was added compared to the other treatments.

#### 4.2.4. The Interrelationship between Functional Tests and Initial Treatment Factors

A correlation analysis was carried out on the results from the various treatments to establish the relationships between gel strength, water holding capacity, and protein solubility with pH and fat removal efficiency. The results of the correlation analyses have been presented in **Table 16**.

The results of the correlation analyses indicated firstly that there was a strong negative correlation between gel strength and pH (-0.780), i.e., as the pH was increased so the gel strength decreased. The correlation analyses are based on data for all alkali treatments and so reflect an overall trend. As shown earlier some alkalis showed no trends in gel strength with increasing pH. The correlation analyses also showed that there was a marginal increase in gel strength with increasing water holding capacity. The correlation analysis was low (0.362). The analyses also showed that gel strength decreased with fat removal i.e., the more fat that was removed the lower the gel strength (-0.464). This last result was unexpected as it could be expected that as the fat was removed so the gel strength should be increased given the fact that the protein content should increase as more fat is removed.

Water holding capacity was seen to decrease with increasing pH (-0.309) which was totally unexpected given the fact that most workers have shown that water holding capacity increases with increasing pH. Fat removal hardly had not any effect on the water holding capacity (-0.161), which too was unexpected given the fact that water content of meat is inversely related to fat content, i.e. as the fat content of meat increases so water content and protein content are known to decrease.

Final protein solubility was neither affected by the original pH of the alkali treatment (-0.079) nor by the amount of fat removed from the sample during the extraction procedures (0.082). The two results were contrary to expectations given the fact that more protein should be available in the sample if the fat content is lowered.

**Table 16: Interrelationships between Functional Tests & Initial Treatment Factors**

	( 1 )	( 2 )	( 3 )	( 4 )	( 5 )
(1)Gel Strength	1				
(2)pH	-0.780	1			
(3)WHC	+0.362	-0.309	1		
(4)Protein Solubility	+0.013	-0.079	+0.141	1	
(5)Fat Removal Efficiency	-0.464	+0.475	-0.161	+0.082	1

In summary, the correlation analyses point out the futility of carrying out complex statistical analyses on data such as was obtained in the present study. The effects of pH on fat separation and the various functional characteristics of the collagen/myofibrillar fraction were highly dependent on the individual alkalis used in the study where one alkali demonstrated that fat separation and functional characteristics improved with increasing pH, whilst a second demonstrated a contrary trend and a third no relationship at all with increasing pH.

### 4.3 SUMMARY

The study showed that meat surimi with a 1.3% fat content could be prepared from mechanically deboned beef. The process used in the laboratory to prepare the meat surimi was a relatively simple process requiring few unit operations, unit operations which are already used for the manufacture of fish surimi. It is therefore expected that the meat industry would have few problems in preparing meat surimi from mechanically deboned beef given the fact that the technology has already been demonstrated for the commercial production of fish surimi. The key processing steps are firstly the preparation of a mechanically deboned slurry with cold water to assist in the centrifugal removal of the "free" fat present in mechanically deboned meat. The centrifugal separation also removes the sarcoplasmic proteins which could be used for the production of meat flavours, soup stock and possibly pharmaceuticals. The second crucial step in the process is a sieving operation of the myofibrillar/collagen slurry to remove the collagen and "bound" fat from the myofibrillar protein. The subsequent collagen free myofibrillar protein could be concentrated by either further centrifugation or by pressing.

The study also showed that most alkali washes had no significant impact on the fat removal efficiencies of the process, with the possible exception of sodium carbonate, compared to the use of fresh, potable water. It was further demonstrated that it was unnecessary to increase the pH of the wash water beyond a pH of 7.0 as no additional fat separation efficiencies were obtained at the higher pH's. The neutral pH requirements of the process would reduce chemical costs, and possibly also limit equipment wear compared to high wash treatments of pH 9.0 advocated by other researchers. The low Ph requirements of the process could also be expected to minimise protein damage which can occur, if held for extended periods at the higher pH's of 9.0 or higher.

The present study has only demonstrated the feasibility of producing meat surimi from mechanically deboned beef. Other uses for the sarcoplasmic and collagen fractions should be established and then a financial feasibility of the whole process should be carried out to establish whether the outlined process is commercially feasible.

## 5. SUGGESTED FURTHER WORK

The present study has only demonstrated the feasibility of producing meat surimi at the laboratory scale. A further series of experiments must be conducted in the laboratory to:

- \* Assess the effect of multiple washings with either sodium carbonate or sodium bicarbonate on not only the removal of free fat, but also on the colour, texture, gel strength and flavour of the meat surimi as well as on the sarcoplasmic and collagen/fat fraction.
  
- \* Establish whether the sieving unit operation should be carried out on the slurried mechanically deboned meat as one of the first unit operations of the process, or whether the sieving operation should be one of the last operations of the process as used in the current study. This phase of the study should not only consider the fat separation efficiencies of the respective processes, but should also consider the recovery rates for the respective fractions so that the process with the highest meat surimi recoveries and possibly lowest fat content are selected for piloting and subsequent commercialisation of the process.

Once a suitable process has been demonstrated on a bench scale then further work should be conducted to assess the feasibility of the process at a pilot plant level. Detailed costings, yields, man-power requirements and equipment requirements must be obtained so that a financial costing of the proposed process can be made before decisions are arrived at as to whether the process is commercial.

The financial viability of the proposed process will largely depend on establishing uses for the fat, collagen, and meat surimi fractions as the process viability may not be able to stand on the sale of just one product, i.e. the meat surimi. Prices for each stream as well as potential market volumes must also be obtained if the process feasibility is to be assessed objectively.

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# Appendices

Appendix 1: Fat removal efficiency of MDB & standard deviation									
pH & alkali	KOH	NaOH	Ca(OH) <sub>2</sub>	K <sub>2</sub> CO <sub>3</sub>	Na <sub>2</sub> CO <sub>3</sub>	NH <sub>4</sub> OH	NaHCO <sub>3</sub>	MgCO <sub>3</sub>	Control
pH 7	71.3 ±1.65	76.65 ±0.76	80.62 ±0.04	79.54 ±1.17	85.70 ±0.74	84.02 ±0.43	78.94 ±0.13	75.28 ±1.49	79.85 ±0.43
pH 8	78.77 ±0.64	81.51 ±0.16	80.33 ±0.98	82.74 ±1.70	84.35 ±0.57	80.62 ±0.37	78.65 ±0.15	NIL	79.85 ±0.43
pH 9	78.23 ±0.37	81.15 ±0.07	78.28 ±0.14	78.19 ±0.28	80.88 ±1.09	81.37 ±0.36	NIL	NIL	79.85 ±0.43

Appendix 2: WHC of myofibrillar/collagen fraction & standard deviation									
pH & alkali	KOH	NaOH	Ca(OH) <sub>2</sub>	K <sub>2</sub> CO <sub>3</sub>	Na <sub>2</sub> CO <sub>3</sub>	NH <sub>4</sub> OH	NaHCO <sub>3</sub>	MgCO <sub>3</sub>	Control
pH 7	50.0 ±1.56	48.7 ±4.10	55.4 ±1.27	56.9 ±8.34	51.9 ±3.82	59.1 ±7.00	52.6 ±2.97	54.1 ±3.18	54.9 ±4.45
pH 8	42.8 ±2.26	53.7 ±0.28	53.1 ±0.07	46.9 ±0.14	48.8 ±0.28	53.3 ±2.40	53.7 ±2.19	NIL	54.9 ±4.45
pH 9	46.3 ±3.04	49.3 ±2.40	63.9 ±0.35	49.9 ±0.78	48.4 ±3.68	46.1 ±0.92	NIL	NIL	54.9 ±4.45

Appendix 3: Gel strength of cooked myofibrillar/collagen fraction & standard deviation									
pH & alkali	KOH	NaOH	Ca(OH) <sub>2</sub>	K <sub>2</sub> CO <sub>3</sub>	Na <sub>2</sub> CO <sub>3</sub>	NH <sub>4</sub> OH	NaHCO <sub>3</sub>	MgCO <sub>3</sub>	Ctrl
pH 7	2.210 ±0.19	1.666 ±0.26	1.775 ±0.08	2.954 ±0.46	1.449 ±0.05	2.031 ±0.09	1.007 ±0.10	1.853 ±0.10	2.734 ±0.14
pH 8	1.060 ±0.05	1.002 ±0.002	1.701 ±0.25	1.294 ±0.08	1.610 ±0.15	1.018 ±0.13	0.743 ±0.04	NIL	2.734 ±0.14
pH 9	0.716 ±0.07	1.242 ±0.02	1.257 ±0.03	1.135 ±0.02	0.949 ±0.12	0.602 ±0.03	NIL	NIL	2.734 ±0.14

Appendix 4: Raw data of fat removal efficiency of MDB									
pH & alkali	KOH	NaOH	Ca(OH) <sub>2</sub>	K <sub>2</sub> CO <sub>3</sub>	Na <sub>2</sub> CO <sub>3</sub>	NH <sub>4</sub> OH	NaHCO <sub>3</sub>	MgCO <sub>3</sub>	Control
pH 7	72.47	77.19	80.65	80.37	86.22	84.32	79.03	76.33	80.15
	70.13	76.11	80.59	78.71	85.18	83.72	78.85	74.23	79.55
pH 8	79.22	81.62	81.02	83.94	84.75	80.88	78.76	NIL	80.15
	78.32	81.40	79.64	81.54	83.95	80.36	78.54		79.55
pH 9	78.49	81.20	78.38	78.39	81.65	81.62	NIL	NIL	80.15
	77.97	81.10	78.18	77.99	80.11	81.12			79.55

Appendix 5: Raw data of WHC of myofibrillar/collagen fraction									
pH & alkali	KOH	NaOH	Ca(OH) <sub>2</sub>	K <sub>2</sub> CO <sub>3</sub>	Na <sub>2</sub> CO <sub>3</sub>	NH <sub>4</sub> OH	NaHCO <sub>3</sub>	MgCO <sub>3</sub>	Control
pH 7	51.10	51.60	56.30	62.80	54.60	64.05	55.80	56.35	58.05
	48.90	45.80	54.50	51.00	49.20	54.15	51.51	51.85	51.75
pH 8	44.40	53.90	53.15	47.00	49.00	55.00	55.25	NIL	58.05
	41.20	53.50	53.05	46.80	48.60	51.60	52.15		51.75
pH 9	48.45	51.00	64.15	50.45	51.00	46.75	NIL	NIL	58.05
	44.15	47.60	63.65	49.35	45.80	45.45			51.75

Appendix 6: Raw data of Gel strength of cooked myofibrillar/collagen fraction									
pH & alkali	KOH	NaOH	Ca(OH) <sub>2</sub>	K <sub>2</sub> CO <sub>3</sub>	Na <sub>2</sub> CO <sub>3</sub>	NH <sub>4</sub> OH	NaHCO <sub>3</sub>	MgCO <sub>3</sub>	Ctrl
pH 7	2.340	1.846	1.835	3.286	1.489	2.091	1.077	1.923	2.834
	2.080	1.486	1.715	2.624	1.409	1.971	0.937	1.753	2.634
pH 8	1.100	1.003	1.881	1.354	1.720	1.108	0.773	NIL	2.834
	1.020	1.001	1.521	1.234	1.500	0.928	0.713		2.634
pH 9	0.766	1.252	1.277	1.145	1.029	0.622	NIL	NIL	2.834
	0.666	1.232	1.237	1.125	0.869	0.582			2.634