CSIRO DIVISION OF FOOD RESEARCH

Tasmanian Food Research Unit, Hobart

74/015

Report to the Fishing Industry Research Trust Account

Research on Technology of Separating Meat from Rock Lobster and Fish. - Completed.

Date commenced - 1/7/74 Date finished - 30/6/77.

1) Separating meat from rock lobster

Cooked rock lobster 'spiders' (the ventral portion of the cephalothorax and attached legs) were obtained in the frozen state from a local lobster processor. After thawing, they were passed through a Bibun meat separator equipped with a 5mm screen. This preliminary operation resulted in a 23% yield of separated minced flesh. The flesh, however, contained visible pieces of carapace and unsightly brown gill material. This flesh was then passed through a Bibun strainer fitted with a 2.5 mm screen, resulting in a 70% yield or 16% of the original material (the spider). Much of the carapace grit and gill was removed in the straining process, however some was finely ground and distributed through the product giving it a distinctly pink appearance. The cleanup step through the strainer was obviously effective, but the product still contained 4.5% carapace grit. While much of the grit was fine, small hardened cones from the tips of the legs and the armoured projections from the leg joints, passed through the screen into the product. A screen with smaller size holes was not available for test.

Flotation of the mince in brine solution showed that the majority of the shell fragments were attached to pieces of flesh and hence it was not possible to separate them by differential flotation.

Mixing the mince with an equal weight of water and centrifuging the mixture resulted in the heavier shell fragments being deposited at the bottom of the centrifuge tube. This lower portion containing the grit was easily removed. A batch of flesh was prepared in this way as test material. Some of the lobster flavour was leached out in the centrifuging step and the flesh texture itself became rather tough and gritty.

This material was made into a pâté and into lobster balls. (Table 1) While some minor shell fragments remained in the flesh it was found that these were neither detectable in the pâté if it was served on dry cracker biscuits, nor in the lobster balls after frying where they were disguised by the crisp fried breadcrumbs.

Lobster flesh can be separated from the 'spiders', however it is not easy to obtain a product free from grit. The problem is not so much free carapace particles, but mainly those pieces of carapace that have adhering flesh and/or connective tissue such as arises from the joints of the legs and where they attach to the cephalothorax.

Brine flotation is not the answer and a centrifugal procedure appears likely to be the most successful. Such procedures are being investigated overseas to recover flesh

from crab waste (see Torry Annual Report 1975 p.13). The equipment involved is expensive and sophisticated. Modification of this equipment and its operating conditions may be suitable for flesh recovery from rock lobster.

2) Separating meat from fish

This report takes the form of the attached papers with particular attention drawn to the pre-publication manuscript entitled "Mechanically separated fish flesh from Australian species - a summary of results of storage trials".

Two further papers are in preparation.

FISHEXPO '76

FISH FINGER TASTE TRIALS AT FISHEXPO '76

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SUMMARY

During the Australian Fisheries Exposition representatives of both the fishing industry and the general public were invited to taste samples of fish fingers made from the mechanically separated flesh of four Australian species.

This paper reports the results for over 400 people who each tasted two fish fingers made from different species of fish and answered an accompanying questionnaire.

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INTRODUCTION

The Tasmanian Food Research Unit of the CSIRO has been engaged in studying ~~on~~ the storage properties of the mechanically separated fish flesh of 16 Australian species. The Unit has made fish fingers from this flesh as examples of typical commercial products. The Australian Fishexpo '76 (September 1976, Melbourne) was taken as a unique opportunity to survey the response of both industry and public to four of these products; such surveys normally being outside the scope of the Unit.

The results of this survey are reported here.

MATERIALS AND METHODS

Fish fingers were made from the flesh of four species of fish, ocean perch (*Helicolenus papillosus*), gemfish (*Rexea solandri*), blue grenadier (*Macruronus novazealandiae*) and southern frostfish (*Lepidopus lex*). The blue grenadier and the gemfish were caught by the CSIRO's FRV Courageous in Bass Strait while the ocean perch and frostfish were caught off the New South Wales coast by the NSW State Fisheries FRV Kapala. The details of fish processing and manufacture of the fish fingers have been given previously (Bremner, 1976, 1977a,b).

At Fishexpo, fish fingers were heated for 30 min. in a pie warmer in batches - two species at a time - in as random a combination of types as practical under the conditions. Pairs of samples,

identified as Y or O, were presented to each taster along with a questionnaire. It was considered that conditions prevented serving more than two samples at a time and that more than two samples would result in less completely filled in questionnaires. The fish fingers were often not served at their best due to the method of heating, and the delays when there were few visitors at the stand, which meant that some samples had tended to dry out.

Circumstances at Fishexpo prevented a rigorously designed experiment since the exhibition extended over three days and few people assessed more than one pair of samples and for any given batch the tasters may have been predominantly from either industry or public and may have varied in their frequency of eating fish fingers: neither was each sample pair presented an equal number of times. In two instances, samples Y and O were identical, the tasters being offered only blue grenadier or frostfish. The nature of the trials has curtailed statistical analysis of the results. On the questionnaire (Appendix 1) tasters were asked whether they were associated with the fishing industry (Industry tasters) or not (Public tasters); how frequently they ate fish fingers (Frequency); how they liked the product (How like) and how they thought the product compared with commercial fish fingers (How compare).

Commercial fish fingers were not offered as a reference. There are a wide range of types on the market made from different fish, both fillets and mince, and coated with a variety of batters of differing colour and texture. To have selected one brand would not only have been difficult, but wrong.

To enable statistical comparison of the data, scores were arbitrarily assigned to the responses in the 'How like' questions from like very much =1 to dislike very much =5, and in the 'How compare' question better =1, equal =2 and poorer =3.

RESULTS AND DISCUSSION

Four hundred and twenty nine people (219 Industry and 210 Public) filled in the questionnaire sufficiently adequately to provide usable data. Table 1 shows the frequency results. There was a tendency for Industry tasters to eat fish fingers less frequently, nevertheless over 50% of all respondents ate fish fingers at least occasionally while some 10% never ate them.

The results of the 'How like' and 'How compare' answers are displayed as bar graphs in Figures 1-3, plotted as a percentage of the number of answers; the number is written beside each group. In this manner the preferences of each of the frequency categories can be noted. The information is also shown in an alternative manner as numbers of answers in Tables 2-6 where the answers to the 'How like' and 'How compare' questions are broken down into the frequency of eating.

The overall results (all fish types, all tasters) contained in Fig. 1 (and Table 2) show that the tasters liked the fish fingers very much (36%) or slightly (34%) and that 36% thought they were better than commercial varieties, 41% thought they were equal and 23% considered them poorer. The majority results were thus favourable. It is

interesting to note (Fig. 1), that those people who never eat fish fingers show a different response pattern, with only 25% thinking that the samples would be better than commercial varieties and 42% saying they were worse. While this group had no experience of commercial fish fingers, it seems to indicate that their expectations are high. Considering only the responses from the experienced regular eaters of fish fingers, then the overall result is encouraging with only 8% (12 people) disliking them very much and 43% liking them very much, while in the 'How compare' question 73% thought them equal to, or better than, commercial fish fingers.

On the other hand of the 34 people who disliked the fish fingers very much, 12 of them regularly eat fish fingers, while a further seven eat fish fingers occasionally. It is impossible to tell whether this result could be expected by chance, alternatively these people could be used to eating a style of fish finger different to the types presented, since as previously stated commercial fish fingers vary widely in many attributes. Using the scores assigned, the 'How compare' answers are better than chance (x^2 test) and as is evident from Figs. 1-4, this is due mainly to the public. The industry were more non-committal tending to rate the samples equal to commercial fish fingers rather than considering them better.

Analysis of variance of the scores showed that the public liked the ocean perch samples the best and thought they compared most favourably with commercial products; while the industry preferred the gemfish samples. There were no differences between industry and public opinion for the blue grenadier or the frostfish samples.

On the two occasions where the pair of samples presented were the one fish species then the judgements were consistent (paired sample t-test).

CONCLUSION

These results agree with those of a previous smaller survey held during ANZAAS, where the tasters were mostly scientists (Bremner, Lewis and Quarmby, 1976b) and appear to suggest that fish fingers made from Australian fish species are well liked and compare favourably with commercial products on the market today. It is the role of those in the market place to consider the value of the information contained in this report.

REFERENCES

Bremner, H.A. (1976a). 'Fishexpo '76 Seminar - Report of Proceedings.' Australian Government Publishing Service, p.309.

Bremner, H.A., Lewis, T.L. & Quarmby, A.R. (1976b). Tasmanian Regional Laboratory, Occasional Paper No.2.

Bremner, H.A. (1977a). Food Technology in Australia, 29, 000.

Bremner, H.A. (1977b). Food Technology in Australia, 29, 000.

Table 1. Frequency of eating fish fingers

	All tasters %	Industry %	Public %
Regular	19	17	21
Occasional	43	40	46
Infrequent	28	28	28
Never	10	15	5

Table 2a. Breakdown of 'How like' preferences into frequency of eating for the four fish finger types combined

Frequency	All tasters					Industry					Public				
	Like very much	Like slightly	Neither like nor dislike	Dislike slightly	Dislike very much	Like very much	Like slightly	Neither like nor dislike	Dislike slightly	Dislike very much	Like very much	Like slightly	Neither like nor dislike	Dislike slightly	Dislike very much
Regular	67	42	23	12	12	30	19	13	4	2	37	23	10	8	10
Occasional	147	119	51	28	7	59	61	26	15	3	88	58	25	13	4
Infrequent	61	98	36	32	8	30	54	21	13	4	31	44	15	19	4
Never	24	21	16	12	7	17	19	12	9	5	7	2	4	3	2
Total no.	299	280	126	84	34	136	153	72	41	14	163	127	54	43	20

Table 2b. Breakdown of 'How compare' answers into frequency of eating for the four fish finger types combined

Frequency	All tasters			Industry			Public		
	Better	Equal	Poorer	Better	Equal	Poorer	Better	Equal	Poorer
Regular	53	55	39	21	28	16	32	27	23
Occasional	123	145	62	47	71	33	76	74	29
Infrequent	76	92	48	31	52	29	45	40	19
Never	14	18	23	12	11	18	2	7	5
Total no.	266	310	172	111	162	96	155	148	76

Table 3a. Breakdown of 'How like' preferences into frequency of eating for fish fingers made from ocean perch

Frequency	All tasters					Industry					Public				
	Like very much	Like slightly	Neither like nor dislike	Dislike slightly	Dislike very much	Like very much	Like slightly	Neither like nor dislike	Dislike slightly	Dislike very much	Like very much	Like slightly	Neither like nor dislike	Dislike slightly	Dislike very much
Regular	14	9	4	1	4	7	5	1	0	1	7	4	3	1	3
Occasional	43	21	18	9	2	16	10	10	7	1	27	11	8	2	1
Infrequent	19	22	14	6	2	9	12	11	2	1	10	10	3	4	1
Never	7	5	5	4	1	4	4	5	2	1	3	1	0	2	0
Total no.	83	57	41	20	9	36	31	27	11	4	47	26	14	9	5

Table 3b. Breakdown of 'How compare' answers into frequency of eating for fish fingers made from ocean perch

Frequency	All tasters			Industry			Public		
	Better	Equal	Poorer	Better	Equal	Poorer	Better	Equal	Poorer
Regular	10	13	7	4	8	2	6	5	5
Occasional	30	43	14	10	23	7	20	20	7
Infrequent	20	25	11	7	17	8	13	8	3
Never	5	4	5	3	3	4	2	1	1
Total no.	65	85	37	24	51	21	41	34	16

Table 4a. Breakdown of 'How like' preferences into frequency of eating for fish fingers made from gemfish

Frequency	All tasters					Industry					Public				
	Like very much	Like slightly	Neither like nor dislike	Dislike slightly	Dislike very much	Like very much	Like slightly	Neither like nor dislike	Dislike slightly	Dislike very much	Like very much	Like slightly	Neither like nor dislike	Dislike slightly	Dislik very much
Regular	24	8	6	4	1	11	3	4	2	0	13	5	2	2	1
Occasional	38	24	3	6	0	14	18	1	3	0	24	6	2	3	0
Infrequent	10	22	7	9	3	6	17	3	5	0	4	5	4	4	3
Never	9	4	1	2	1	7	4	1	1	0	2	0	0	1	1
Total no.	81	58	17	21	5	38	42	9	11	0	43	16	8	10	5

Table 4b. Breakdown of 'How compare' answers into frequency of eating for fish fingers made from gemfish

Frequency	All tasters			Industry			Public		
	Better	Equal	Poorer	Better	Equal	Poorer	Better	Equal	Poorer
Regular	15	17	8	6	7	5	9	10	3
Occasional	27	33	7	12	19	2	15	14	5
Infrequent	12	16	15	8	10	7	4	6	8
Never	2	7	4	2	5	2	0	2	2
Total no.	56	73	34	28	41	16	28	32	18

Table 5a. Breakdown of 'How like' preferences into frequency of eating for fish fingers made from blue grenadier

Frequency	All tasters					Industry					Public				
	Like very much	Like slightly	Neither like nor dislike	Dislike slightly	Dislike very much	Like very much	Like slightly	Neither like nor dislike	Dislike slightly	Dislike very much	Like very much	Like slightly	Neither like nor dislike	Dislike slightly	Dislik very much
Regular	11	9	5	3	3	6	5	3	1	0	5	4	2	2	3
Occasional	37	50	17	5	1	14	22	9	2	1	23	28	8	3	0
Infrequent	18	37	8	10	2	8	15	3	2	2	10	22	5	8	0
Never	5	5	8	3	3	4	5	5	3	2	1	0	3	0	1
Total no.	71	101	38	21	9	32	47	20	8	5	39	54	18	13	4

Table 5b. Breakdown of 'How compare' answers into frequency of eating for fish fingers made from blue grenadier

Frequency	All tasters			Industry			Public		
	Better	Equal	Poorer	Better	Equal	Poorer	Better	Equal	Poorer
Regular	12	5	11	6	4	4	6	1	7
Occasional	41	43	20	12	19	14	29	24	6
Infrequent	24	36	13	7	15	8	17	21	5
Never	5	4	6	5	2	4	0	2	2
Total no.	82	88	50	30	40	30	52	48	20

Table 6a. Breakdown of 'How like' preferences into frequency of eating for fish fingers made from frostfish

Frequency	All tasters					Industry					Public				
	Like very much	Like slightly	Neither like nor dislike	Dislike slightly	Dislike very much	Like very much	Like slightly	Neither like nor dislike	Dislike slightly	Dislike very much	Like very much	Like slightly	Neither like nor dislike	Dislike slightly	Dislike very much
Regular	18	16	8	4	4	6	6	5	1	1	12	10	3	3	3
Occasional	29	24	13	8	4	15	11	6	3	1	14	13	7	5	3
Infrequent	14	17	7	7	1	7	10	4	4	1	7	7	3	3	0
Never	3	7	2	3	2	2	6	1	3	2	1	1	1	0	0
Total no.	64	64	30	22	11	30	33	16	11	5	34	31	14	11	6

Table 6b. Breakdown of 'How compare' answers into frequency of eating for fish fingers made from frostfish

Frequency	All tasters			Industry			Public		
	Better	Equal	Poorer	Better	Equal	Poorer	Better	Equal	Poorer
Regular	16	20	13	5	9	5	11	11	8
Occasional	25	26	21	13	10	10	12	16	11
Infrequent	20	15	9	9	10	6	11	5	3
Never	2	3	8	2	1	8	0	2	0
Total no.	63	64	51	29	30	29	34	34	22

CSIRO

TASMANIAN FOOD RESEARCH UNIT
HOBART

FISHEXPO '76 FISH FINGER TASTING

Please tick the appropriate box.

Are you associated with the fishing industry

Yes ☐

No ☐

Do you eat fish fingers

Regularly (e.g. once a fortnight) ☐

Occasionally (e.g. once every 2-3 months) ☐

Infrequently (e.g. twice a year) ☐

Never ☐

You are given two samples of fish fingers identified as Y or O.

Please taste them separately and answer the questions by placing a tick or cross in the appropriate column.

How did you like the product	Y	O
Like very much		
Like slightly		
Neither like nor dislike		
Dislike slightly		
Dislike very much		

How do you think the fish finger compares with commercial products

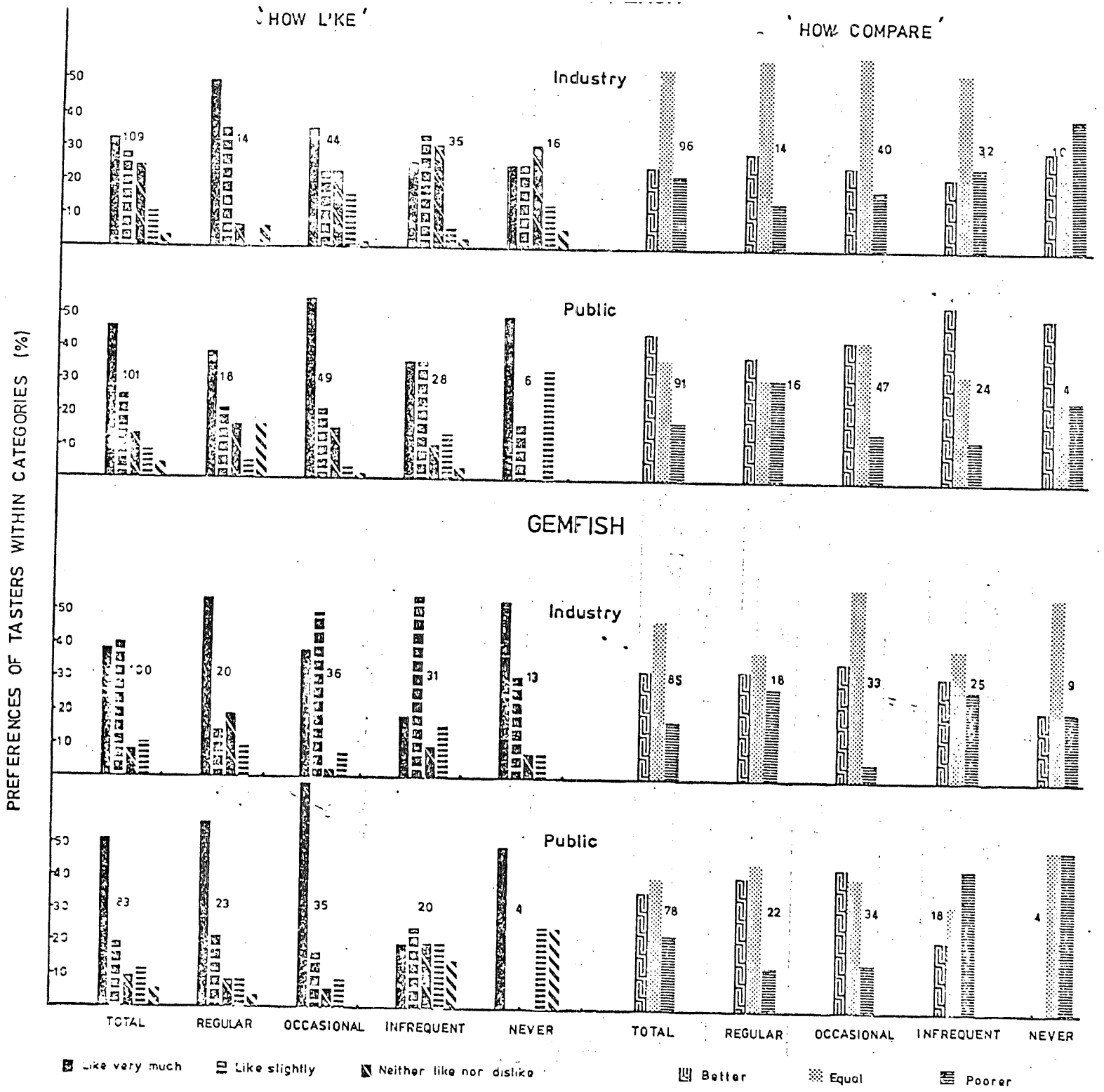
Better		
Equal		
Poorer		

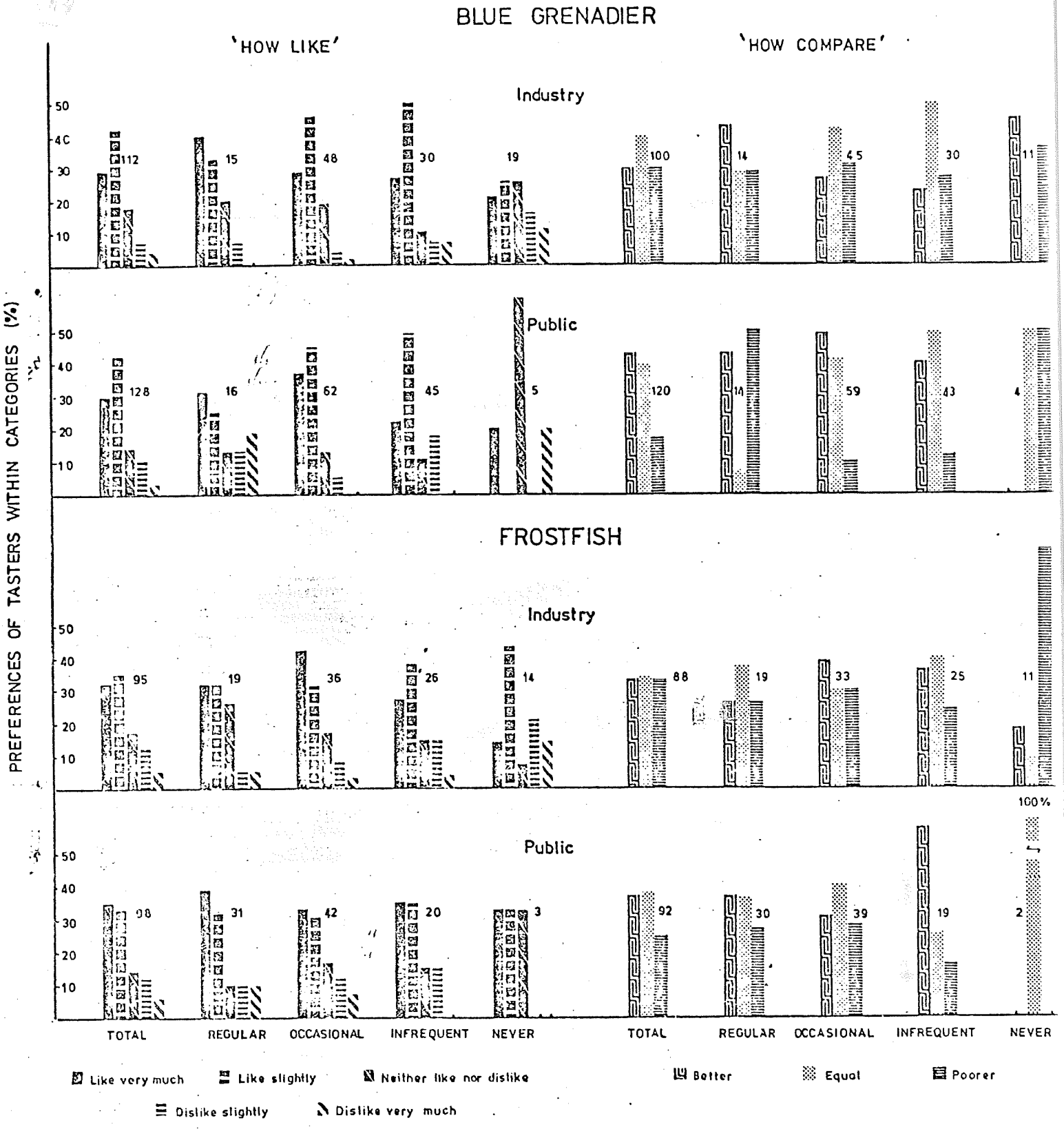
If you answer poorer could you describe in what way this product was inferior.

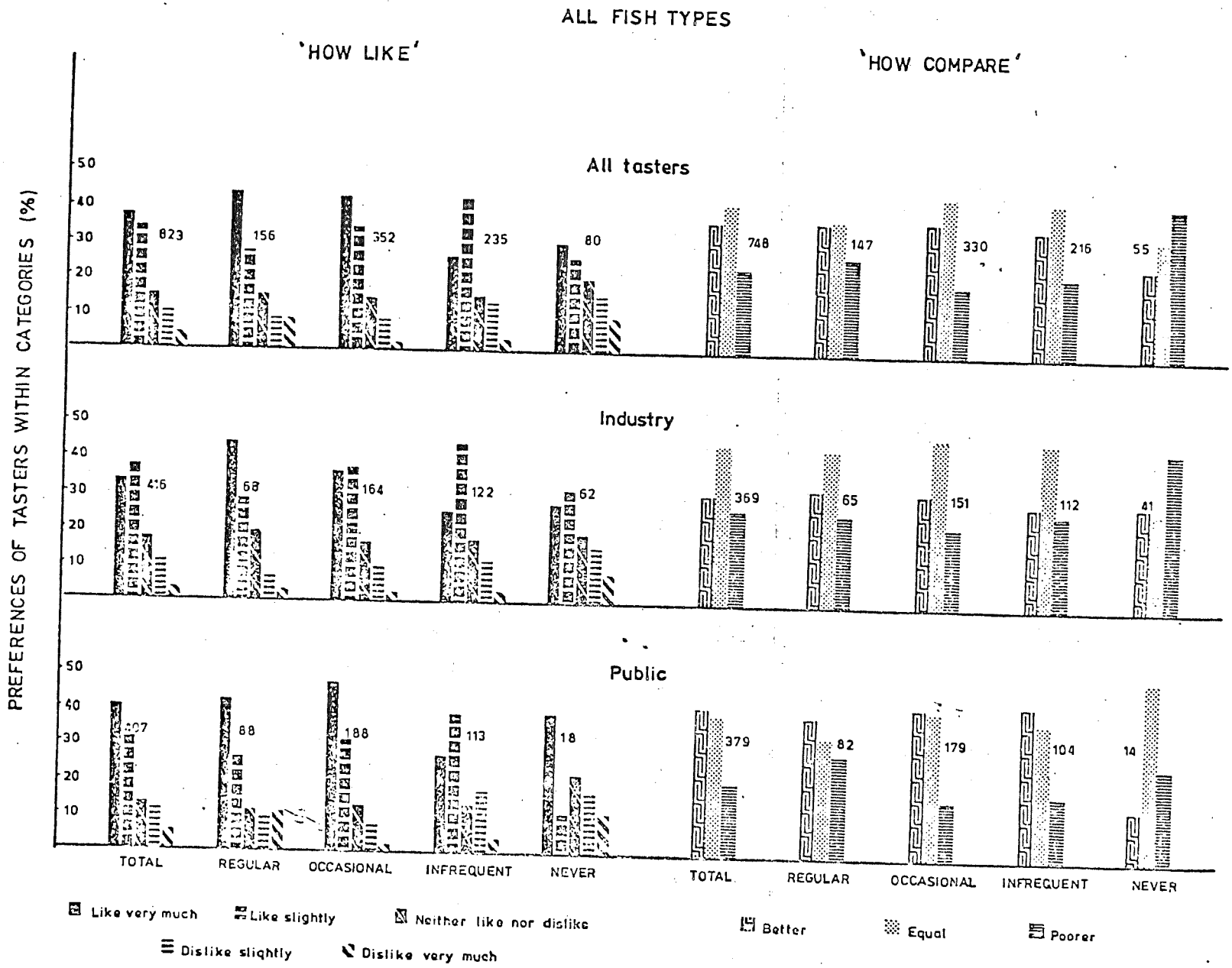
e.g. too fishy, not fishy enough, too wet, too dry, unpleasant aroma or flavour etc.

Y

O


'HOW L'KE'
'HOW COMPARE'
Industry
Public
GEMFISH
Industry
Public
PREFERENCES OF TASTERS WITHIN CATEGORIES (%)
50
40
30
20
10
109
14
44
35
16
96
14
40
32
10
101
18
49
28
6
91
16
47
24
4
100
20
36
31
13
85
18
33
25
9
23
23
35
20
4
78
22
34
18
4
TOTAL
REGULAR
OCCASIONAL
INFREQUENT
NEVER
TOTAL
REGULAR
OCCASIONAL
INFREQUENT
NEVER
Like very much
Like slightly
Neither like nor dislike
Better
Equal
Poorer


BLUE GRENADIER
'HOW LIKE'
'HOW COMPARE'
Industry
Public
FROSTFISH
Industry
Public
PREFERENCES OF TASTERS WITHIN CATEGORIES (%)
50
40
30
20
10
112
15
48
30
19
100
14
45
30
11
128
16
62
45
5
120
14
59
43
4
95
19
36
26
14
88
19
33
25
11
98
31
42
20
3
92
30
39
19
2
100%
TOTAL
REGULAR
OCCASIONAL
INFREQUENT
NEVER
TOTAL
REGULAR
OCCASIONAL
INFREQUENT
NEVER
Like very much
Like slightly
Neither like nor dislike
Dislike slightly
Dislike very much
Better
Equal
Poorer


ALL FISH TYPES
'HOW LIKE'
'HOW COMPARE'
PREFERENCES OF TASTERS WITHIN CATEGORIES (%)
All tasters
Industry
Public
50
40
30
20
10
823
156
352
235
80
748
147
330
216
55
416
68
164
122
62
369
65
151
112
41
307
88
188
113
18
379
82
179
104
14
TOTAL
REGULAR
OCCASIONAL
INFREQUENT
NEVER
TOTAL
REGULAR
OCCASIONAL
INFREQUENT
NEVER
Like very much
Like slightly
Neither like nor dislike
Dislike slightly
Dislike very much
Better
Equal
Poorer

COMMONWEALTH SCIENTIFIC & INDUSTRIAL RESEARCH ORGANISATION – TASMANIAN FOOD RESEARCH UNIT
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Storage trials on the mechanically separated flesh of three Australian mid-water fish species 1. Analytical Tests

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The mechanically separated flesh from the three Australian fish species: cucumber fish *(Chlorophthalmus nigripinnis)*, ocean perch *(Helicolenus papillosus)* and spiny flathead *(Hoplichthys haswelli)* was frozen stored as small blocks at -18° C and evaluated by various tests at regular intervals. The results indicate that the flesh of ocean perch and spiny flathead does not significantly deteriorate in six months at -18° C and would make a suitable manufacturing material. Cucumber fish produced formaldehyde in frozen storage resulting in loss of salt extractable protein and water holding capacity. Its use is not recommended for frozen fish products.

Mechanical separation gives a greater yield of edible flesh from the available fish catch than conventional means such as filleting (Regier, 1974). Either headed and gutted fish or frames after filleting can be used to produce a coarse mince suitable for incorporation directly into a variety of products or into frozen blocks for later processing, *eg* into fish fingers.

The mechanical separators used on fish employ the belt and drum principle in which fish, headed and then gutted, are crushed by a ridged roller onto a flexible moving belt. The belt squeezes the fish against a perforated steel drum revolving in the same direction but at lower speed; the shearing action of the sharp edges of the holes and the simultaneous tearing action of the belt squeezes the flesh through holes into the drum from which it is removed by a helical screw. Bone, fin and skin which by contrast with the flesh are tougher, continue with the belt and are scraped off the outer surface of the drum by a doctor blade.

During the mechanical separation the disruption of tissue integrity allows access of oxygen, spreading of bacteria and novel contacts of intracellular and extracellular components; enzymes are also released. The overall effect is the promotion of a variety of reactions, bio-chemical, physical and chemical, at rates greater than occur in fillets or in whole fish.

Marine fish contain the osmoregulatory compound trimethylamine oxide (TMAO) which can be reduced by bacteria to trimethylamine (TMA) (Beatty & Gibbons, 1937) and TMA levels have been proposed as indicators of post-catch handling before freezing (Connell, 1975). In the frozen state in some species the TMAO is converted enzymatically to dimethylamine (DMA) and formaldehyde (FA) (Amano & Yamada, 1963). The neutral fat of frozen fish and the phospholipids can be hydrolysed by lipases and phospholipases to yield free fatty acids (FFA) with phospholipases generally predominating (Olley, Pirie & Watson, 1962). These FFA are more prone to oxidation than the parent compounds, yielding malonaldehyde (MA) as an end-product of oxidation. Estimation of MA forms the basis of the thiobarbituric acid (TBA) test for rancidity.

The myofibrillar proteins actin and myosin are soluble in salt solutions but become insolubilised during frozen storage due to a variety of reactions. The major chemical reactions implicated in the insolubilisation of proteins are the formation of FA, FFA and MA (Sikorski, Olley & Kostuch, 1976). These compounds readily react with proteins and alter the natural configuration of the protein helix by forming cross links and hydrophobic interactions, the general result being textural changes, loss of water holding capacity, poorer manufacturing properties and development of off-flavours and aromas, particularly where oxidation is implicated.

Exploratory mid-water and deep-water trawling in 200-400 fathoms (366-732 m) along the continental shelf of south eastern Australia by the N.S.W. State Fisheries vessel FRV Kapala has discovered large fish stocks (Gorman & Graham, 1975). These stocks are at present not commercially exploited but could provide raw material for locally manufactured fish products such as fish fingers which currently are either wholly imported or made from imported frozen fish blocks.

While many of the species caught by mid-water trawling may be suitable for sale on the fresh fish market, they are unfamiliar to the public, often unattractive in appearance and hence fetch only a low price. To a large processor many of the species yield only a small fillet with high wastage, and since mechanical filleting machines cannot handle the variety of fish shapes and sizes which comprise the mixed catches from mid-water trawling, the greater yields from mechanical separators on low-priced fish could provide an economic situation where

these species could compete favourably with imported material.

There is no information, however, on the manufacturing characteristics of the flesh or its frozen storage properties, and apart from some information on related species listed in Jowett & Davies (1938) even the basic gross composition of the species has not been reported. A research project was thus initiated to supply information on the mechanically separated flesh of Australian mid-water species and their behaviour in frozen storage. This paper reports experiments on the first three species made available by courtesy of the New South Wales State Fisheries.

The three species of fish were cucumber fish *(Chlorophthalmus nigripinnis),* ocean perch *(Helicolenus papillosus)* and spiny flathead *(Hoplichthys haswelli).* Despite being relatively small, cucumber fish enjoyed some popularity over twenty years ago but is no longer in demand. The ocean perch is unattractive in appearance and because of its sharp dorsal spines requires cautious handling, and while of reasonable size yields only a comparatively small fillet. Spiny flathead has a somewhat grotesque appearance and its double row of spines or bony bucklers on the lateral lines of the sides make it extremely difficult to fillet. These three species thus make mechanical processing appear an attractive proposition as an avenue to commercial exploitation.

MATERIALS AND METHODS

Fish mince production

The fish caught in early December 1974 off the N.S.W. coast by the State Fisheries vessel FRV Kapala were sorted on board, blast frozen (-20°C) and despatched on landing by refrigerated transport to the CSIRO, Hobart Laboratory where they were held at -18°C for 6 weeks before processing. After leaving the fish over-night in air, thawing was completed by immersing the fish in running tap water. The fish were weighed, scaled if necessary, headed and gutted, then scrubbed by hand to remove the dark belly lining (if present) and as much remaining kidney as possible. After washing, draining and weighing they were held in ice before processing.

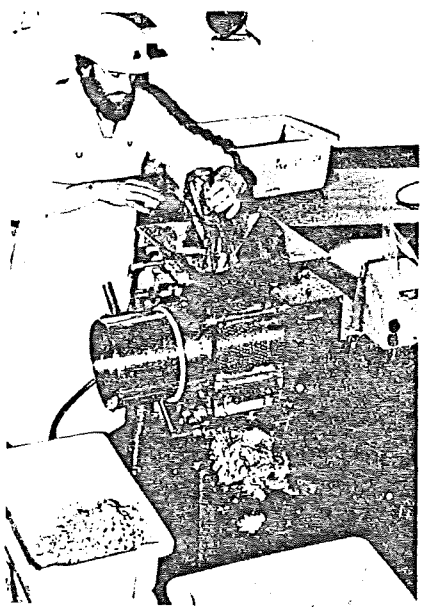

Figure 1. The fish-meat separator in action. Input capacity 450 kg/hr.

The fish meat separator used was a Japanese "Bibun" SDX13 similar to that described by Okada (1975) and Figure 1 shows it in operation. The separator was used with screens having either 5-mm or 10-mm holes. The minced flesh was collected from the separator, blended and weighed. A set of samples was packaged as a standard pack in aluminium dishes (40 mm deep x 100 mm wide x 130 mm long) and placed inside coded polyethylene pouches and frozen on racks in an open-end wind tunnel with solid sides set in a freezer at -18°C, a fan at one end providing the air flow. Other samples were placed in open polyethylene sachets which were vacuum sealed into cans and frozen as above. After 4 hr in the tunnel the temperature in the centre of the trays was -5°C and after 16 hr it was -16°C. The samples were stored at -18°C after 24 hr in the tunnel.

SAMPLING OF FISH BLOCKS

A block from each batch was tested after approximately 1, 2, 3, 4.5 and 6 months in store. It was partially thawed overnight at 0-1°C and then cut into approximately 200 cubes (3 cuts laterally, 7 longitudinally and 10 vertically) which were mixed and distributed at random into three 200-g portions. The first lot was further divided, remixed and weighed out for the various analyses, while the other two were retained for taste panel evaluation (Bremner, 1977).

ANALYTICAL METHODS

pH of the mince: a cheese electrode fitted to a Radiometer pH Meter 29 was pressed into the thawed mince.

Moisture: oven drying over-night at 105°C of three 10-g samples.

Crude protein (N x 6.25): Kjeldahl-Wilfarth-Gunning method.

Fat: extraction method of Hanson & Olley (1963).

Salt extractable protein: method of Anderson & Ravesi (1968) on three 10-g samples, extractant ionic strength 0.8.

Perchloric acid extract: method Mackie & Thomson (1974), three 20-g samples of flesh extracted with 0.6 M perchloric acid with the addition of 200 mg EDTA and 200 mg propylgallate per 80 ml solution.

FA: method of Nash (1953) on the perchloric acid extract.

DMA: method of Dowden (1938) on the perchloric acid extract after removing the acid by precipitation and neutralisation with KOH.

TMA: on a trichloroacetic acid extract of the flesh (Bystedt, Swenne & Aas, 1959) by the method of Dyer (1959) as modified by Tozawa, Enokihara & Amano (1971).

TMAO: Similarly, following reduction to TMA with $TiCl_3$ by the method of Yamagata, Horimoto & Nagaoka (1969).

Extractable malonaldehyde (TBA value): modification of the method of Vyncke (1970) by heating 5 ml of perchloric acid extract with 5 ml aqueous 0.02 M thiobarbituric acid, 20 min in boiling water bath, cooling and reading developed colour at 532 nm in a spectrophotometer. Perchloric acid did not lead to high or erratic blanks or recoveries such as obtained by heating the TBA reagent with other oxidising agents (Tarladgis, Pearson & Dugan, 1962).

FFA: on the extracted fat by the colorimetric method of Dowden (1963) after removing phospholipids by shaking in chloroform with

Table 1
Fish weight/length data, mince yields, proximate analysis of the mince and pH changes in frozen storage

Fish	Weight/length data[a] mean weight (g)	weight range (g)	mean length (cm)	length range (cm)	Yield of mince expressed on whole ungutted fish (%)	expressed on headed gutted fish (%)	screen size (mm)	pH initial	pH after 326 days	Proximate analysis[b] crude protein (N x 6.25) (%)	moisture (%)	fat (%)
Cucumber	197	161–225	24.2	22–27	37.2	55.6	5	6.7	6.9	18.5	80.0	2.08
						60.0	10			(0.2)[c]	(0.1)	(0.2)
Ocean perch	603	375–835	28.5	25–32	33.0	58.3	10	6.6	6.5	16.3	81.6	1.21
	772	615–920	35.0	33–38		60.0	10			(0.2)	(0.2)	(0.1)
Spiny flathead	436	256–539	40.0	37–43	34.7	61.1	5	6.5	6.5	17.0	82.1	0.50
					34.7	59.0	10				(0.0)	(0.1)

[a] Data obtained on 5 ungutted samples.

[b] Means of 3–20 replicates. The analytical data for two mincings of cucumber fish, three of ocean perch and two of spiny flathead have been combined, the replicate mincings being only a few days apart and from the same catch of fish.

[c] S.E. in brackets.

silicic acid activated beforehand for 24 hr at 120°C, the silicic acid being removed by centrifuging 10 min at 5000 rpm to prevent high reagent blanks, caused by carry-over into the colorimetric determination.

Water holding capacity (WHC): determined centrifugally on 10 g flesh in a weighed centrifuge tube at 20 000 rpm (50 000 *g*) for 1 hr the tube was then decanted, drained and tube plus the fish pellet weighed.

WHC = 100 x weight of remaining pellet/10 g.

RESULTS AND DISCUSSIONS

Yields and Analysis

The yields of mince, pH and proximate analysis are presented in Table 1. The yields based on the whole fish are similar to those reported for some American species (Crawford *et al.*, 1972; Miyauchi & Steinberg, 1970). The pH change in cucumber fish on storage was assumed to be due primarily to formation of DMA. The spiny flathead flesh was notably low in fat (0.5%), this amount representing mainly structural lipid.

The analytical data for two mincings of cucumber fish, three of ocean perch and two of spiny flathead have been combined, the replicate mincings being only a few days apart and from the same catch of fish.

The analytical results for TMAO, FA and DMA are presented in Table 2. The TMAO levels reported here are in the median range of those reported in the literature (Love, Lovern & Jones, 1959). The TMA levels were low, indicating good handling practice before freezing. Cucumber fish flesh produced large amounts of FA such that after 11 months in storage 85% of the initial TMAO present had been broken down. By comparison, FA production in the ocean perch and spiny flathead was neglible and only 0.3% of the initial TMAO present was degraded. Two further catches of cucumber fish (ex Kapala) and two very small Tasmanian catches approximately 2-3 kg of juveniles from about 50 fathoms (92 m) have also produced FA in frozen storage; but FA production is not a characteristic of many species. From the 17 species investigated to date the only other to produce FA is the blue grenadier *(Macruronus novaezealandiae)* a whiptail of the family *Macruridae.*

The FA determined in cucumber fish decreased with time, indicating that after an initial period the rate of reaction of FA with the proteins exceeded the rate of production from TMAO, a reaction which follows first order kinetics. This is in agreement with Babbitt, Crawfor & Law (1972) and Tokunaga (1974) who also showed a decrease in production rate in pacific hake *(Merluccius productus)* and alaska pollack *(Theragra chalcogramma)* respectively. The vacuum-packed samples contained the same levels of FA and DMA as the corresponding packs stored in the aluminium dishes in oxygen-permeable polyethylene pouches.

Changes in salt-extractable protein during storage are shown in Figure 2. The cucumber fish protein rapidly became almost totally inextractable in salt solution; even the water soluble sarcoplasmic proteins were insoluble, presumably being entrained in the toughened muscle matrix (Anderson & Ravesi 1968). The level of extractable protein of the spiny flathead decreased only slowly, whereas that for ocean perch fell between the other two species. The level of extractable protein tended to fall more rapidly with those minces pro-

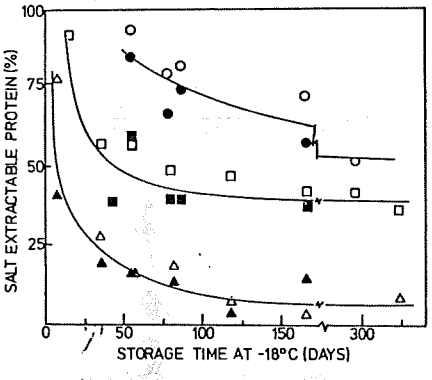

Figure 2. Change in salt extractable protein of minced fish flesh during frozen storage at –18°C.

○, ● Spiny flathead; □, ■ Ocean perch; △, ▲ Cucumber fish.

Open or closed symbols denote values for mince produced using 10-mm screen and 5-mm screen respectively.

duced using the 5-mm screen rather than the 10-mm screen; however the inter-species difference was far greater. At the point of catch, fish flesh proteins are vitually totally extractable in dilute salt solution and the initial values obtained here were taken as an indication of denaturation occuring in transit and during storage before processing.

After 6 months the FFA values for the three species were in the range 40-45 mg/100 g flesh, a figure which would be regarded as low (Olley, Pirie & Watson, 1962; Olley, Farmer & Stephen, 1969). TBA values progressively decreased. For both ocean perch and spiny flathead after 300 days in store, the TBA values of the standard pack were exactly the same as for the vacuum pack, namely 0.55 and 0.7 mg malonaldehyde/kg fish flesh respectively, whereas the values for the standard packs 4 months earlier had been 0.7 and 1.2 mg malonaldehyde/kg fish flesh, indicating that the initially higher MA levels were the result of oxygen inclusion up to and during processing. The malonaldehyde became inextractable during storage presumably due to reaction with protein (Buttkus, 1967). Values determined on small samples of fresh ocean perch have been of the order of 0.1-0.2 mg malonaldehyde/-kg flesh. The TBA levels in cucumber fish also decreased slightly over the period 120-300 days in store from 1.9 to 0.7 mg malonaldehyde/kg flesh. TBA levels in fresh cucumber fish are in the region of 0.3-0.8 mg malonaldehyde/kg flesh.

Loss of WHC for cucumber fish is evident in Table 3, but this test is obviously not sensitive since the salt extractable protein had already reached its minimum long before the WHC began to decline; nor was the WHC test sensitive enough to detect a change in the loss of salt extractable protein in ocean perch or spiny flathead.

CONCLUSION

Ocean perch should offer the best opportunity for mechanical separation because of the yield, the quality of its flesh and its stability in frozen storage. The flesh of the spiny flathead is also of good quality and is stable in frozen storage, but yields are low. The cucumber fish is not stable in frozen storage, producing FA, which explains the resultant toughening, the loss of water holding capacity and the rapid loss in salt extractability.

While it would be wise not to market products made from cucumber fish, FA production may not preclude its use since other fish of commerce, *eg* atlantic cod, pacific hake and alaska pollack, also produce FA in frozen storage where the problem is minimised by correct handling, freezing and storage from point of catch to the market place.

The mild increase in FFA and the same decrease in TBA value for

Table 2
Breakdown of trimethylamine oxide to formaldehyde and dimethylamine during storage at –18°C

Fish	*Determination*[a]	*Storage time (days)* 7	54	118	164	326
Cucumber	TMA μ mole/100 g	267				
	FA μ mole/100 g	127	697 [607][b]	1717	1077	910
		(44)[c]	(57)	(90)	(47)	(22)
	DMA μ mole/100 g		1774 [1606][b]	3055	2797	3565
			(95)	(196)	(43)	(109)
	FA/DMA ratio		0.39	0.56	0.39	0.26
	% TMAO converted (initially 4190 μ mole/100 g)		42.4	72.9	66.8	85.0
Ocean perch	TMA μ mole/100 g	74				
	FA μ mole/100 g	3.7	1.7	3.6	4.4	3.3 [3.3][b]
		(0.9)	(0.3)	(0.1)	(0.2)	(0.1)
	DMA μ mole/100 g		9.4	16.9	7.1	20.3 [2.2][b]
			(0.9)	(0.6)	(2.2)	(0.8)
	FA/DMA ratio		0.18	0.21	0.62	0.16
	% TMAO converted (initially 6580 μ mole/100 g)		0.14			0.31
Spiny flathead	TMA μ mole/100 g	51				
	FA μ mole/100 g	0.5	1.2		4.3	3.3 [3.3][b]
		(0.1)	(0.2)		(0.2)	(0.1)
	DMA μ mole/100 g		8.4		1.9	19.4. [19.0][b]
			(0.3)		(0.1)	(0.7)
	FA/DMA ratio		0.14		2.26	0.17
	% TMAO converted (initially 6820 μ mole/100 g)		0.12			0.28

[a] Means of 3–6 replicates. The combined results of two m incings of cucumber fish, three of ocean perch and two of spiny flathead have been combined, the replicate mincing being only a few days apart and from the same catch of fish.
[b] Values for vacuum packed samples in square brackets.
[c] S.E. in curved brackets.

Table 3
Changes in water holding capacity during frozen storage (−18°C) of minced fish blocks

Fish		*Storage time (days)* 81	85	118	132	164	326
Cucumber	Mean[a]	47.6		43.3		18.2	14.6
	S.E.	2.4		1.5		3.2	1.8
Ocean perch	Mean[a]	42.2	42.5		44.6	35.5	40.1
	S.E.	1.2	1.5		2.6	1.6	0.7
Spiny flathead	Mean[a]	39.1	40.7		42.5		37.5
	S.E.	3.7	1.7		2.4		0.8

[a] Means of at least 4 replicates.

both vacuum packed and standard samples indicate that lipase activity, phospholipase activity and rancidity are not major problems during the cold storage of these species.

Taste panel results in the following paper (Bremner, 1977) are consistent with the analytical results presented here.

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Storage trials on the mechanically separated flesh of three Australian mid-water fish species 2. Taste panel evaluation*

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The mechanically separated flesh from the three Australian mid-water fish species cucumber fish *(Chlorophthalmus nigripinnis)*, ocean perch *(Helicolenus papillosus)* and spiny flathead *(Hoplichthys haswelli)* was frozen and stored at -18°C in the form of small blocks and evaluated by a taste panel at regular intervals. Neither ocean perch nor spiny flathead deteriorated significantly in six months, whereas cucumber fish decreased in fish aroma and fish flavour, developed off-odours and off-flavours, toughened markedly and decreased considerably in acceptability. These results agree with the chemical tests which showed that ocean perch and spiny flathead would be suitable for new frozen fish products, but that it would be more difficult to prepare stable products from cucumber fish.

In 1974-75 over $A15m worth of fresh and frozen fish products and $A4.5m dollars worth of fish fingers were imported into Australia (Anon., 1976). Before locally caught species can fill this demand there is a need for information on the storage properties of their flesh.

This paper reports the results of taste panel evaluation on the frozen stored flesh of three mid-water fish species.

MATERIALS AND METHODS

Sample preparation

Three species of fish were used, cucumber fish *(Chlorophthalmus nigripinnis)*, ocean perch *(Helicolenus papillosus)* and spiny flathead *(Hoplichthys haswelli)*. The process of mechanical separation to produce a coarse mince, the packaging and freezing of this mince in small blocks and the sampling procedure has been described (Bremner, 1977). 'Fish fingers' were made from some of the frozen blocks as an example of familiar frozen fish products. Blocks were sawn into fish finger blanks (as near a standard size as possible, 15 mm x 25 mm x 85 mm), dipped in batter and breadcrumb mix (Table 1) and deep fried for 1 min at 190-195°C, in a vegetable oil ('Frymasta', Vegetable Oils Pty Ltd, Sydney). The fish fingers on trays covered with aluminium foil were frozen in the wind tunnel, then 24 hr later packaged in polyethylene bags and stored on racks at -18°C.

Taste panel assessment

The taste panel was used to provide some information on the inherent properties of each species, such as degree of fishiness, characteristic odour or flavour; and to indicate whether gross change occurred in the flesh during storage. The panel consisted of ten members of staff — five women, five men; it was untrained since thorough training was beyond the scope of this investigation and reference samples could not be provided, as a regular supply of these fish is not available at present.

Portions of the fish mince (200 g) were heated for 1 hr in a stainless steel dish fitted with a lid, in a water bath set at 60°C. The fish was heated in its own juices to near 60°C, a temperature reported by Kushtalov & Saduakasov (1971) as optimal to retain the water holding capacity of the flesh. Solids and any juices were placed in pre-heated glass jars marked with the symbols △, □, X and Y and presented to the panel in random order. Fish cooked in this manner is rather plain fare, but it is generally accepted that taste panels can detect subtle texture and flavour changes more readily in plain cooked or steamed fish than in fish which has been dipped in batter and fried (c.f. Dyer *et al.* 1964).

The design of the presentation of the cooked minces to the taste panel is shown in Table 2 and a summary of the score sheet, descriptive terms used and corresponding numerical scores in Table 3. The panel marked the appropriate descriptive term on the score sheet with the sample symbols. These were converted to numerical scores and punched on to data cards. Comments were freely invited but not actively solicited.

Since each panellist had to set his own scale of reference for each attribute, the statistical method of Steel & Torrie (1960) was chosen. The method assumed linear changes and examined the data for each individual panellist, calculated a regression for each panel member for each attribute, gave a measure of the fit of his data to this regression, then compared regressions between panellists to ascertain whether the panel as a whole detected a change in the attribute with time. The criterion suggested by Dahloff & Jul (1965) that a change of 1 unit represents a 'Just Noticeable Difference' was employed in interpreting the significance of the results.

Two batches of fish fingers were made from cucumber fish and ocean perch blocks resp. after 18 and 180 days in store. Only one batch was made from spiny flathead blocks after 180 days. The first batch of fish fingers made from cucumber fish and ocean perch were tasted together

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Table 1
Batter mix and breadcrumb mix used for fish fingers

Batter mix (Parts by weight)		*Breadcrumb mix* (Parts by weight)	
Self raising flour	93.8	Self raising flour	40.0
Dried whole egg powder	2.0	Dried whole egg powder	10.0
Skim milk powder	2.0	Skim milk powder	2.5
Salt	2.0	Salt	1.0
Mono sodium glutamate	0.2	Mono sodium glutamate	0.65
	100.0	Breadcrumbs (white)	45.85
Water (approx.)	175.0		100.00

(two fish fingers x two sessions) the day after making. The second batch of fish fingers made from cucumber fish and ocean perch were tasted with the first batch of spiny flathead fish fingers (three fish fingers x two sessions) the day after making and again after 4 months in store at -18°C.

The fish fingers were presented to the panel on individual stainless steel wire grids on which they had been heated in the oven for 45 min at 180°C. The same marking and scoring system was used as for the mince.

RESULTS

The results presented in Table 4 are a summary of the pooled data obtained on two mincings of cucumber fish, three mincings of ocean perch and two mincings of spiny flathead. The results were combined on the basis that differences (t-tests) between minces made with either the 5-mm screen or the 10-mm screen were only minor and not of any consequence when using the criterion of Dahloff & Jul (1965). Analyses of variance showed no significant bias between morning and afternoon replicates, and the panel's judgement of ocean perch was not apparently influenced by whether it was tested at the same session as spiny flathead (which softened) or cucumber fish (which toughened).

The initially low panel mean scores in the categories of fish aroma and fish flavour are a true reflection of the degree of 'fishiness' of the three species. In other trials the panel has marked the stronger flavoured species, such as tuna and salmon much higher.

The panel considered that cucumber fish deteriorated markedly in frozen storage. This species decreased in 'fishiness' and its characteristic mild, sweetish, sharp, almost turnip-like odour which most panellists classed as off-aroma increased in intensity during storage. Other off-aromas described as cardboardy, stale and ammoniacal developed as did off-flavours described as sickly, flat and musty. The texture score increased, i.e. the flesh toughened and was described as fibrous and rubbery, despite an upward change in pH, the effect of which would tend to oppose this trend (Bosund & Beckeman, 1972).

Table 2
Presentation of cooked mince samples to the taste panel

Fish	Processing date *	Screen size (mm)	Samples taste session (No.)	Occasions and sessions (No.)
Cucumber	(1)	5		5 Occasions over 6 month period
Cucumber	(1)	10	3	2 Sessions on occasions 1, 4 and 5
Ocean perch	(2)	10		1 Session on occasions 2 and 3
Ocean perch	(2)	5	—	Unsuccessful run, machine seized)
Ocean perch	(3)	5		5 Occasions over 6 month period
Ocean perch	(3)	10	4	2 Sessions on occasions 1, 4 and 5
Spiny flathead	(3)	5		1 Session on occasions 2 and 3
Spiny flathead	(3)	10		
Ocean perch	(3)	10		
Ocean perch vacuum packed	(3)	10	4	1 Occasion after 10 months in store 2 Sessions
Spiny flathead	(3)	10		
Spiny flathead vacuum packed	(3)	10		

* (1) 14/1/75 (2) 15/1/75 (3) 22/1/75

Table 3
Numerical score corresponding to descriptive terms for each attribute

Taste panel score	Fish aroma	Off-aroma	Fish flavour	Off-flavour	Texture	Moisture	Acceptability
9	Very strong	Very strong	Very strong	Very strong	Tough	Very wet	Very good
7	Strong	Strong	Strong	Strong	Slightly tough	Wet	Good
5	Moderate	Moderate	Moderate	Moderate	Preferred texture	Normal moisture	Moderate
3	Weak	Weak	Weak	Weak	Slightly soft	Dry	Poor
1	Very weak	None	Very weak	None	Soft	Very dry	Very poor

The moisture category in Table 4 for this fish requires further explanation. When toughened fish flesh is chewed much of its moisture readily escapes, leaving a dry fibrous wad in the mouth. Some panellists took note of the initial impression of moisture and marked the sample wet, others noted the dry wad and marked the sample dry. It is thus impractical to pool the data.

The results for ocean perch and spiny flathead agree with the subjective impression that neither of these fish changed greatly in storage; the only significant change were in texture, where both fish were judged to have become softer. In the case of ocean perch, this texture was described in the comments as floury, powdery and granular, and for spiny flathead as gritty, powdery, stringy and woolly; the slow denaturation presumably was accompanied by a breakdown of fibre length. Olley *et al.* (1969) have pointed out that where denaturation is slow other reactions may precede the insolubilisation, leading in extreme cases to sloppiness during cold storage (Olley, Farmer & Stephen, 1967). Fish muscle is known to have greater catheptic activity than

Table 4
Summary of changes[a] detected by taste panel occurring in three species of minced fish[b], stored as blocks over a 6 month period at -18° C

Fish		Fish aroma Decrease **	Off-aroma Increase ***	Fish Flavour Decrease **	Off-flavour Increase ***	Texture toughness Increase ***	Moisture	Acceptability Decrease ***
Cucumber	Initial mean panel score	4.55	4.78	5.13	4.26	6.91	Refer to text	4.80
	S.E.[c]	0.39	0.58	0.36	0.56	to 0.32		0.34
	Panel mean change over 180 days	-1.07	+2.09	-1.04	+2.36	+1.54		-2.61
		Decrease ns	Negligible change ns	Negligible change ns	Negligible change ns	Decrease *	Increase ns	Increase ns
Ocean perch	Initial mean panel score	4.27	3.63	4.10	3.26	4.75	5.02	4.70
	S.E.	0.23	0.35	0.21	0.27	0.32	0.30	0.20
	Panel mean change over 180 days	-0.25	-0.03	0.13	-0.10	-0.95	+0.39	+0.27
		Increase ns	Decrease ns	Negligible change ns	Decrease ns	Decrease *	Increase ns	Negligible change ns
Spiny flathead	Initial mean panel score	3.84	4.31	4.41	4.28	4.69	5.23	4.31
	S.E.	0.22	0.32	0.25	0.39	0.32	0.30	0.30
	Panel mean change over 180 days	+0.29	-0.26	-0.03	-0.45	-0.97	+0.39	+0.10

[a]The symbols ***, **, * and ns represent changes which are significant at $p<0.001$, $p<0.01$, $p<0.05$ and not significant respectively.
[b]Samples presented to the panel as cooked mince. [c]Standard error of mean.

mammalian muscle (Siebert, 1958) and to possess high concentrations of enzymes responsible for proteolysis and amino acid metabolism (Siebert Schmitt & Bottke, 1965). More recently Obanu, Ledward & Lawrie (1975) have demonstrated breakdown in the proteins of meat flesh in the period before the over-riding cross-linking reactions. The state of the proteins depends on the balance between these reactions.

The flesh of ocean perch has a milky, slightly cabbage-like odour and flavour. In storage some slight musty ammoniacal overtones and a stale bland flavour developed. Spiny flathead has a sharp aroma and sweet flavour, and here some slight stale milky ammoniacal odours and stale musty flavours developed. In neither case did this adversely affect the mean off-odour or off-flavour panel score, nor the acceptability.

Analysis of variance comparing panel results of standard pack samples of ocean perch and spiny flathead with their vacuum packed counterparts — from the same mincing and all stored at -18°C for 300 days — showed that the vacuum packed spiny flathead was judged softer.

Taste panel results on fish fingers

The results presented in Table 5 show that the panel considered that fish fingers (batch 2) from blocks of cucumber fish had less fish aroma, less fish flavour and were less acceptable than fish fingers made from freshly processed blocks (batch 1). When these were tasted again after storage for 120 days, toughening was evident, but the other deteriorative trends had not progressed. As with the mince, texture was described as being chewy and rubbery. These results are an improvement over those obtained on the mince where the panel considered that significant deterioration had occurred in all categories.

The second batch of fish fingers made from ocean perch were judged to have less off-aroma, less off-flavour than the original batch, be more acceptable and be firmer in texture in contrast to the mince. No further changes were detected on storage of this second batch of fish fingers.

After 120 days in store the aroma of fish fingers made from spiny flathead had decreased and off-flavour increased, but these effects were not sufficient to change overall acceptability.

The taste panel scores for cooked minces and fish fingers made from blocks after comparable times of storage listed in Table 6 show the improvement in acceptability obtained by conversion of the fish into a fish finger. The process of battering, breading and frying masks off-aromas and off-flavours and improves fish texture, as commented on by Bramsnaes (1969). Acceptable fish fingers can be made from otherwise unacceptable fish: the fish fingers made from 6-months old blocks of cucumber fish stored for a further four months were as acceptable to the panel as a commercial brand (Bremner, 1976).

It should be noted that these trials were conducted on fish that had already been frozen and stored some six weeks before processing, and the results could only but improve if fresh unfrozen fish were used. It remains also to be seen whether other catches of the same fish at different seasons or from different grounds show the same properties.

Follow-up work on these three species to ascertain the affect of various treatments on their storage properties is being done. Further work is also well underway on several other species.

CONCLUSION

The taste panel detected storage changes in accordance with the findings of the previous analytical tests (Bremner, 1977). Formaldehyde production by cucumber fish flesh explains the increase in toughness detected by the panel; although the mince deteriorated, this deterioration could be masked when fish fingers were made from it. The previous conclusion based on

Table 5
Mean taste panel scores for fish fingers compared by analysis of variance

Fish	Batch (No.)	Storage (-18°C) as minced block (days)	Storage (-18°C) as fish fingers (days)	Fish aroma (score)	Off-aroma (score)	Fish flavour (score)	Off-flavour (score)	Texture (toughness) (score)	Moisture (score)	Acceptability (score)
Cucumber	1	18	2	4.80^a	3.48^a	5.64^a	3.46^a	5.84^a	3.76^a	5.27^a
	2	180	2	3.57^b	3.63^a	4.43^b	3.62^a	6.23^a	4.01^a	4.31^b
	2	180	120	4.26^{ab}	3.24^a	4.69^b	4.00^a	7.19^b	3.85^a	4.63^{ab}
			S.E.	0.27	0.27	0.25	0.33	0.24	0.29	0.21
			D.F.	13	12	13	9	14	12	14
Ocean perch	1	18	2	4.35^a	3.77^a	4.71^a	3.52^a	3.95^a	5.05^a	4.80^a
	2	180	2	3.61^a	2.63^b	4.43^a	1.88^b	4.77^b	5.10^a	6.24^b
	2	180	120	3.70^a	2.59^b	5.05^a	2.42^b	5.21^b	4.99^a	6.47^b
			*S.E.**	0.24	0.15	0.33	0.33	0.15	0.12	0.19
			*D.F.***	13	9	13	11	14	11	13
Spiny flathead	1	180	2	3.60^a	1.90^a	4.40^a	1.60^a	4.10^a	4.50^a	5.35^a
	1	180	120	3.15^b	2.50^a	4.05^a	2.65^b	4.30^a	4.40^a	5.60^a
			S.E.	0.12	0.21	0.14	0.14	0.15	0.10	0.16
			D.F.	9	9	9	9	9	9	9

$^{a, b}$ Values followed by same superscript not significantly different ($p<0.05$) for each attribute of each fish.

* *S.E. Standard error of mean*
** *D.F. Degrees of Freedom*

Table 6
Taste panel results on cooked minces and fish fingers made from comparable blocks stored for 180 days at -18°C

Fish		Fish aroma (score)	Off-aroma (score)	Fish flavour (score)	Off-flavour (score)	Texture (score)	Moisture (score)	Acceptability (score)
Cucumber	Mince[a]	3.48	6.87	4.09	6.62	8.45		2.19
	Fish fingers	3.57	3.63	4.43	3.62	6.23		4.31
Ocean perch	Mince[a]	4.05	3.60	4.23	3.16	3.80	5.41	4.97
	Fish fingers	3.61	2.63	4.43	1.88	4.77	5.10	6.24
Spiny flathead	Mince[a]	4.13	4.05	4.38	3.83	3.72	5.62	4.41
	Fish fingers	3.60	1.90	4.40	1.60	4.10	4.50	5.35

[a] Calculated from Table 4

analytical data that ocean perch and spiny flathead flesh would be suitable for new frozen fish products has been confirmed. Cucumber fish would require pre-treatment and demand careful handling in the frozen food chain before it could be used to produce stable products.

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Taste panel assessment of textural properties of fish minces from Australian species

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Summary

The relationship between taste panel scores for texture (toughness) and moisture (succulence), flesh pH and salt extractable protein for sixteen Australian species of fish has been investigated using an approach outlined by Cowie & Little (1966; 1967) for frozen stored cod.

By graphing their data, Cowie & Little (1967) showed that a line could be drawn which divided the results into those samples with soft flesh and those samples with tough flesh. Remarkably, this dividing line was found to fit the present data accumulated in the course of a series of experiments on frozen stored minced fish flesh.

This approach emphasises the underlying relationship of texture with pH and salt extractable protein.

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Introduction

An investigation into the properties of the separated flesh of a wide variety of Australian fish species (Bremner, 1977a) afforded a unique opportunity to examine the resulting data for underlying relationships between texture, moisture as measured by taste panel, pH, and salt extractable protein.

Cowie & Little (1966) employed a texture-moisture matrix in their taste panel work on frozen stored cod and established the importance of the relationship between muscle pH and toughness. They also showed (Cowie & Little, 1967) that a dividing line could be drawn between the taste panel results obtained on tough cooked cod fillets of low pH and/or protein extractability and softer fillets of high pH and/or greater protein extractability. Bosund & Beckeman (1972) reported a strong negative correlation between toughness and pH for cod stored at $-30^{\circ}C$. This correlation still held at $-10^{\circ}C$, but was lessened by other factors which overrode the pH effect at the higher temperature; for example, cod forms formaldehyde during cold storage at higher temperatures and this in turn leads to the denaturation or inextractability of myofibrillar proteins in salt solutions (Sikorski, Olley & Kostuch, 1976).

The present study extends the approach outlined by Cowie & Little (1966; 1967) and demonstrates the remarkable fact that their concept of a dividing line between tough and soft cod fillets, applies to the stored minces and fish fingers made from sixteen different Australian species of fish. Using the parameters pH and salt extractable protein, 46% of the variance was accounted for; the further incorporation of moisture (taste panel) increased the explanation of the variability to 71%.

In view of the importance of the underlying concepts, it seems worthwhile to describe the data in detail.

Materials and methods

The preparation of the comminuted fish minces, the frozen blocks and the fish fingers have been described previously, together with the sampling and analytical methods (Bremner, 1977a; 1977b). The composition of the untrained taste panel (10 members) and the preparation, presentation of the samples in two sessions - morning and afternoon - after each storage period, and the scoring system have also been reported (Bremner, 1977c).

Inextractable protein was calculated as total protein (N x 6.25, Kjeldahl) minus the extractable protein determined by the biuret method.

The symbols for the different species and their common and Latin names are shown in Table 1.

The data for fish fingers is restricted to those which had been freshly prepared, although the mince from which they were made had often been stored for considerable periods.

The taste panellists were required to score the cooked minces and fish fingers for texture, moisture, and acceptability on a nine point scale: from very soft (1) to very tough (9) and from very dry (1) to very wet (9), with 5 as preferred texture and normal moisture (Bremner, 1977c). Cowie & Little (1966) used a similar but less extended scoring system.

The discrepancy in numbers of samples mentioned in the text is due to the lack of complete data on some samples.

Results and discussion

Texture-moisture matrix

Taste panel means (2 sessions x 10 tasters) have been employed throughout. The texture-moisture results for each individual fish are displayed as a matrix in Fig.1, identified by the symbols in Table 1.

Attention is drawn to those species which were judged to have a soft texture even though their moisture was judged normal, i.e. nannygai, shark and silver trevally.

The Cowie & Little line

Taste panel texture scores are shown in Fig. 2 plotted with pH and protein extractability % (g extractable protein/100g total protein) as ordinates, to provide a comparison with Fig. 5 in the paper by Cowie & Little (1967), however in the present case, nonprotein nitrogen was not determined and hence the protein extractability values are erroneously low by a small amount. The Cowie and Little dividing line is drawn on the figure as a solid line where it extends over the range of pH and protein extractability encompassed by their data. The present results embrace a wider range, and the line can be seen to fit, such that it misclassified only one point on the basis that texture scores below 4.0 are soft and above 6.0 are tough.

It is also worth noting that while Cowie and Little (1967) used a 4 point texture scale they reported only two results which were below 2 (marginally). Their line drawn using a panel mean texture score of 3 is then at the mid point of their data set and this accords with a score of 5 in the present work.

Despite the fact that the total protein (N x 6.25) content of the flesh of the various species ranged from 22% (tuna) to 15.3% (ocean perch), the extractable protein and the percentage extractable protein, having a correlation coefficient of 0.98, were almost of equal value in predicting the texture score, accounting for 31 to 38% of the variance. Therefore the data is displayed in alternate form in Fig. 3 with pH and extractable protein (g extractable protein/100g mince) as ordinates to

show the general applicability of the concept, since extractable protein is more commonly and more readily determined than percentage protein extractability. In this case the line has been fitted by eye. It misclassified only one point using the criteria that scores below 4.0 are soft and those above 6.0 are tough.

It is evident from both Figs. 2 & 3 that the nannygai, for example, are softer than would be expected from their pH and extractable protein levels; even so, they are still on the appropriate side of the Cowie and Little dividing line. The data for fish fingers has not been shown graphically because the smaller number of tastings provided less data for plotting, and the presentation to the panel of the fish mince in this breaded and battered form resulted in fewer texture scores above 5 (c.f. Bremner, 1977c). However, the Cowie and Little line, again separated the tough from the soft fish fingers.

Robustness of the dividing lines

There is no suitable objective statistical method of drawing dividing lines for a continuum of data such as presented here. The nearest accordant method is that of discriminant analysis and this first requires an arbitrary judgement on the criteria for misclassification. Application of discriminant analysis to the data in Fig. 2 resulted in a 'family' of dividing lines depending on which criteria were chosen; none performed better than the Cowie and Little line on the basis of misclassifying soft samples (below 4.0) and tough samples (above 6.0).

Likewise in the case of Fig. 3 other lines could probably be drawn; since, it is unusual to encounter fish samples with a pH much greater than 7 that are not spoilt, the line drawn seems the most suitable.

Data from previous experiments on three Australian species (Bremner, 1977b,c) were also found to be in accord with the Cowie and Little line

(Fig. 2), except in those few cases where samples softened with storage, probably due to catheptic activity. Further experimental results on six tropical Malaysian species are also discriminated by the line (one out of six points misclassified). The results from these earlier (three) and latter (six) species were consistent with the texture-moisture matrix.

Shaw and Botta (1977) expressed surprise at the good textural properties of capelin (*Mallotus villosus*) stored at -23°C for up to two years, however, this is readily explained in terms of this present concept, in that their pH was high (range 7.2 to 6.7) and their extractable protein did not decrease sufficiently to place them below the Cowie and Little line.

Relationship between variables

The correlation coefficients relating texture scores with pH, protein extractability and moisture for the fish minces are shown in Table 2: values for fish fingers, based on smaller numbers of samples, are shown for comparison. Multiple linear regression analysis showed that pH and extractable protein could account for 46% of the variance in the minces. Inextractable protein was highly negatively correlated with extractable protein and proved to be of almost equal value in accounting for the variance.* Incorporation of taste panel moisture scores as a sensory measurement of water holding capacity - influenced by both pH and protein extractability (Hamm, 1960) - increased the variance accounted for to 71%. This suggests that an experimental variable related to

* Percentage variance accounted for $\equiv 100\ (1 - \sigma_e^2/\sigma_y^2)$, where σ_y^2 is the variance of the texture scores and σ_e^2 is the mean square residual error after fitting a regression model. An Analysis of Variance on the individual texture scores demonstrated that the maximum possible percentage variance of the mean texture scores which could be accounted for was 93%: random (uncontrolled) variation contributes the rest.

water holding capacity would be valuable in conjunction with pH and salt extractable protein - as a predictor of texture.

Inclusion of sample identity, as a variable, raised the 'variance accounted for' to 90%. Although this figure is irrelevant as an indicator of accuracy when attempting to predict the texture of untried species, the analysis indicated that the soft samples, with normal moisture (Fig. 1), are most responsible for introducing the variability.

It is impossible to ascertain whether a better correlation exists for one species, cod, than for the variety of species reported on here, since Cowie and Little (1967) did not include texture scores on their figure; neither did they make use of their texture-moisture matrix, as an aid to explaining the variability in their results.

Robustness of the relationship

A number of common sources of variability in raw material is taken into account in the relationship. Variables such as season, feed, fishing ground, catching and handling techniques all affect either pH or level of salt extractable protein, or both. These in turn affect water holding capacity - measured here sensorily as taste panel moisture.

Moreover, where fish continue to toughen even after their level of salt extractable protein has dropped almost to zero (Connell, 1968) then, this too, is reflected in the strong negative correlation (Table 2) between texture and taste panel moisture, implying that, at least as far as the senses are concerned, fish tend to become drier as they toughen (see also Fig. 1).

The complex influence of the state of rigor in which fish are frozen (Amlacher, 1961) is not directly taken into account; neither will the relationships hold where fish soften markedly in frozen storage due to catheptic activity.

Texture and acceptability

Cowie and Little (1967) equated a lower texture score with greater acceptability, without considering that acceptability may decrease with fish of too soft a texture. The present results showed no linear correlation between texture and taste panel acceptability scores ($r = 0.07$; d.f. 67) or moisture (succulence) and acceptability ($r = 0.034$; d.f. 67). The present study showed two reasons why fish may have acceptability scores unrelated to, or nonlinearly related to, texture; either they are too soft (c.f. Fig. 1) or when they toughened on cold storage other attributes such as off-flavour had a predominant influence on acceptability (Laslett & Bremner, unpublished).

While texture may not be the prime determinant of acceptability of cooked minces and fish fingers, this is not so for fish sausages and heat gelled fish products, where quality depends on correct pH and high protein extractability to provide the emulsifying and water holding capacities necessary to obtain a product with suitable rheological properties (Sadowska & Sikorski, 1977).

Conclusions

The relationship between texture, pH, salt extractable protein and moisture explored by Cowie and Little (1966; 1967) on frozen stored cod has been re-examined and extended to results on a random mixture of sixty-two samples from sixteen species of fish. The striking similarity in the results obtained shows the validity of the underlying concept of the relationship of texture with pH and salt extractable protein and the interrelated phenomenon of succulence.

As traditional stocks become over-fished and new species enter the trade, this information may be of considerable value, particularly in framing investigations on newer species.

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Table 1. Key to species

Symbol	Common name	Generic name
A	Australian salmon	*Arripis trutta esper*
B	Barracouta	*Leionura atun*
C	Cucumber fish	*Chlorophthalmus nigripinnis*
E	Saw shark	*Pristiophorus cirratus*
G	Gemfish (hake or king barracouta)	*Rexea solandri*
L	Ling	*Genypterus papillosus*
M	Morwong (jackass fish)	*Nemadactylus macropterus*
N	Nannygai (redfish)	*Centroberyx affinis*
O	Ocean perch	*Helicolenus papillosus*
P	Perch*	
R	Red gurnard	*Currupiscis kumu*
S	Silver trevally	*Usacaranx nobilis*
H	Spiny or deep sea flathead	*Hoplichthys haswelli*
T	Tuna	*Katsuwonus pelamis*
F	Tiger flathead	*Neoplatycephalus richardsoni*
W	Blue grenadier (whiptail)	*Macruronus novaezealandiae*

* Generic name not identified

Table 2. Correlation matrix between variables for fish minces and fish fingers[+]

	Fish minces (d.f. = 59)				Fish fingers (d.f. = 20)			
	Texture	pH	Extractable protein g/100g flesh	Inextractable protein g/100g flesh	Texture	pH	Extractable protein g/100g flesh	Inextractable protein g/100g flesh
H	-0.16				-0.21			
xtractable protein	-0.57***	0.37**			-0.42*	0.26		
nextractable protein	0.63***	0.19	-0.94***		0.50*	-0.08	-0.91***	
aste panel noisture	-0.78***	0.32*	0.34**	-0.46***	-0.74***	0.36	0.33	-0.50*

*, **, *** significant at the 5%, 1% and 0.1% level respectively

[+] Note the similarity between the correlation coefficients between variables for fish minces and fish fingers

Fig. 1 Taste panel mean scores displayed as a texture-moisture matrix, for the minces from sixteen species of fish caught in Australian waters, and stored for varying times up to a year at -18°C. Each species is marked with a symbol (Table 1).

Fig. 2 The relationship between taste panel score for texture (shown in diagram against symbol for each species) for fish minces and the pH and percentage of salt extractable protein. The Cowie & Little line is taken from their paper and divides tough from acceptable fillets. See legend to Fig. 1.

Fig. 3 The relationship between taste panel texture score (shown in diagram against symbol for each species) for fish minces and the salt extractable protein in g/100g mince. See legend to Fig. 1.

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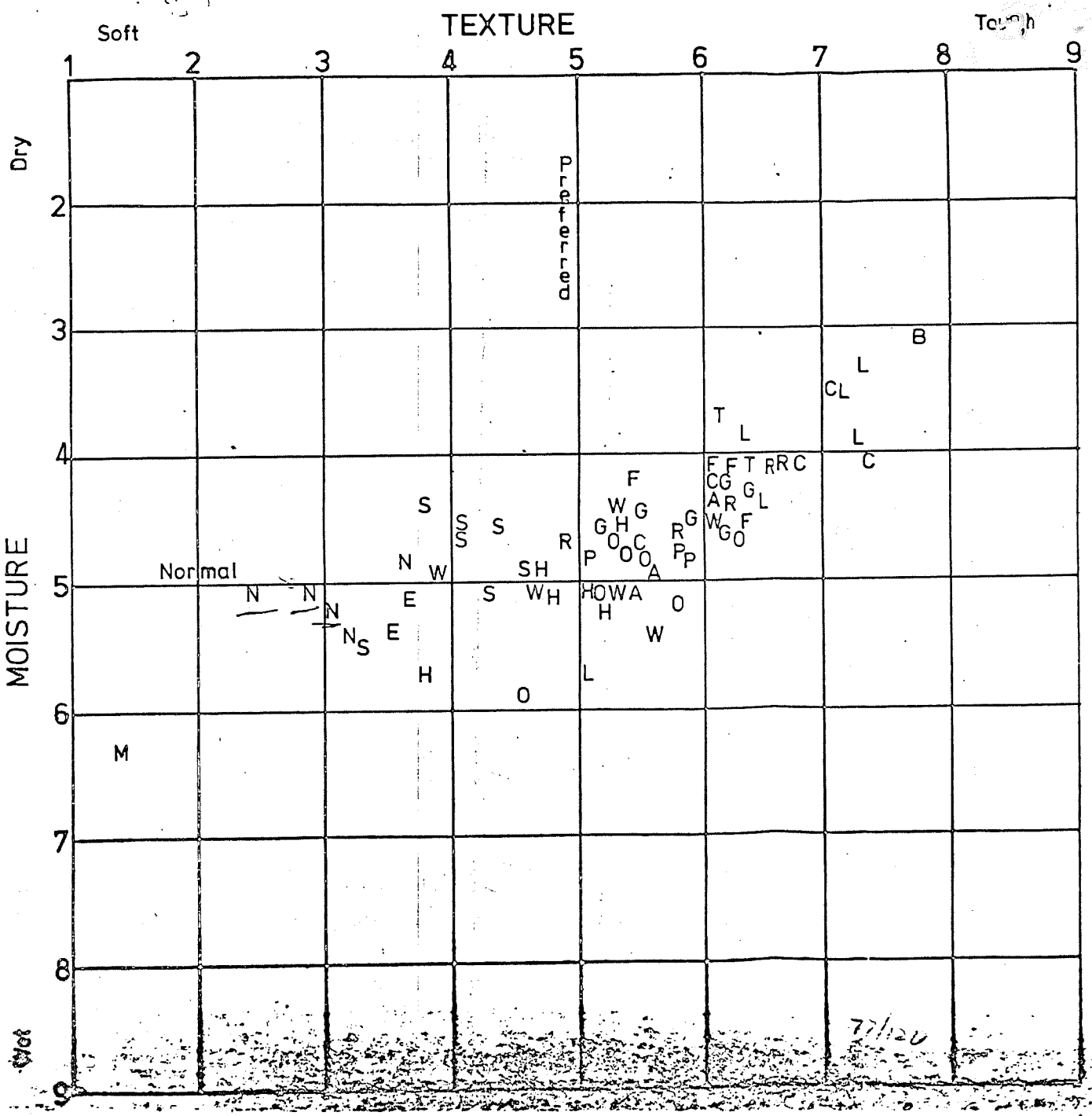
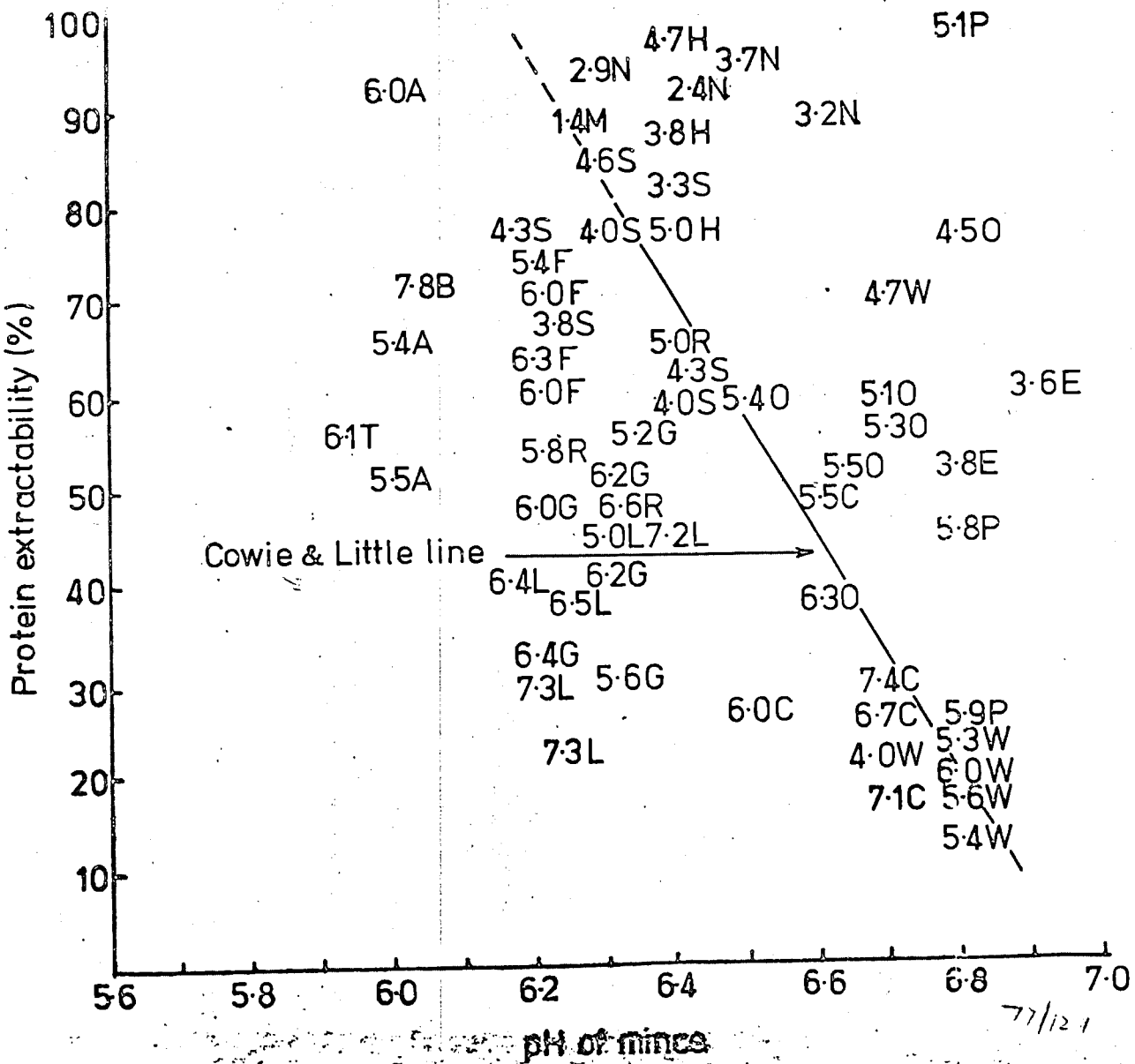

TEXTURE
Soft
Tough
1
2
3
4
5
6
7
8
9
Dry
MOISTURE
Wet
Preferred
Normal


Protein extractability (%)
100
90
80
70
60
50
40
30
20
10
5·6
5·8
6·0
6·2
6·4
6·6
6·8
7·0
pH of mince
Cowie & Little line
4·7H
5·1P
2·9N
3·7N
6·0A
2·4N
3·2N
1·4M
3·8H
4·6S
3·3S
4·3S
4·0S
5·0H
4·5O
5·4F
7·8B
6·0F
4·7W
3·8S
5·4A
5·0R
6·3F
4·3S
6·0F
4·0S
5·4O
5·1O
3·6E
6·1T
5·2G
5·3O
5·8R
5·5O
3·8E
5·5A
6·2G
5·5C
6·0G
6·6R
5·8P
5·0L
7·2L
6·4L
6·2G
6·5L
6·3O
6·4G
7·3L
5·6G
7·4C
6·0C
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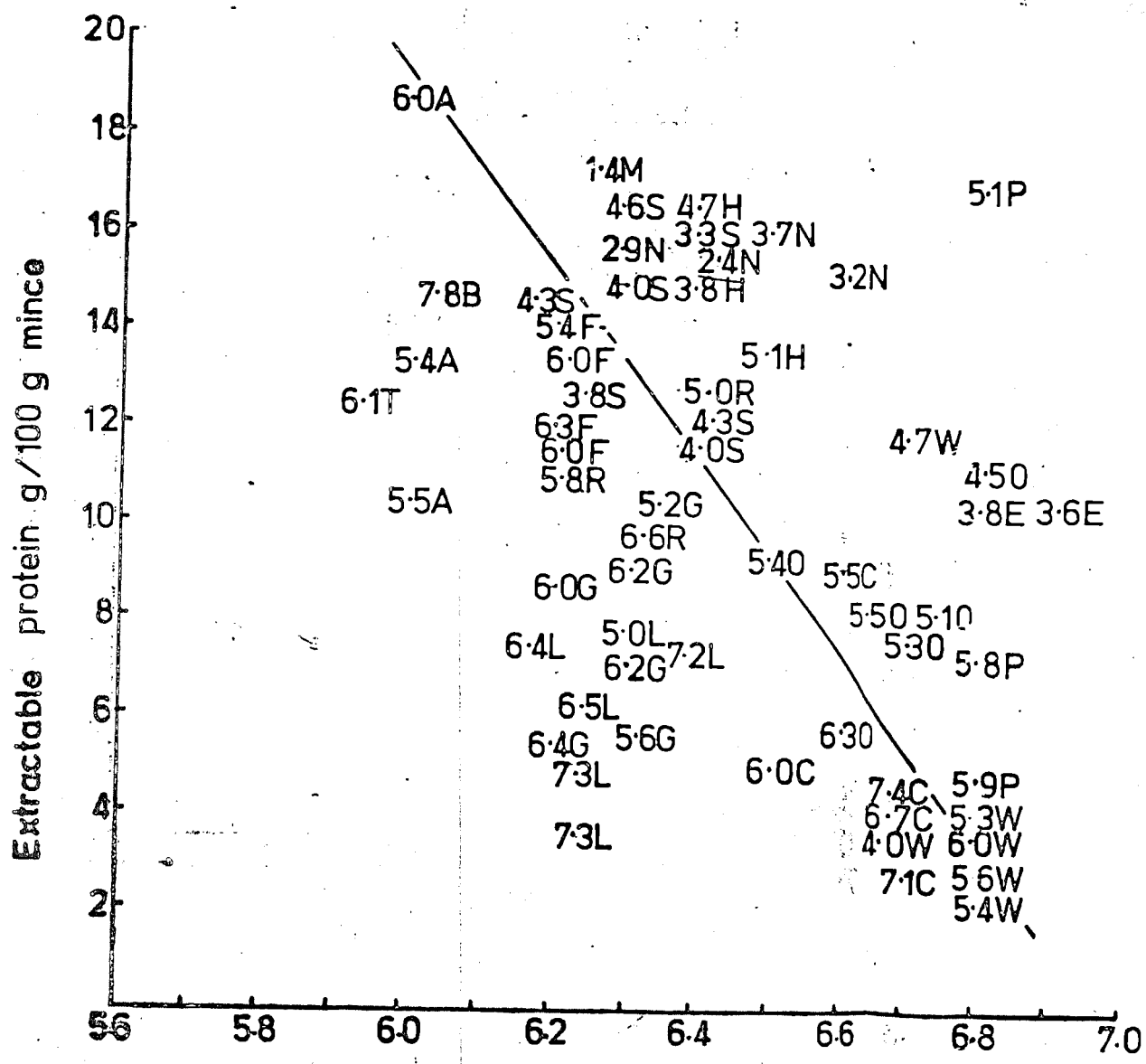
6·0W
7·1C
5·6W
5·4W

Fig 3



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Mechanically separated fish flesh from Australian species - a summary of results of storage trials*

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* Presented in part at the AIFST Eleventh Annual Convention as "Species difference in mechanically separated fish flesh."

ABSTRACT

A number of separate storage experiments were carried out on frozen blocks of mechanically separated (minced) flesh from a variety of Australian species. Each experiment was conducted using one species, and dressed fish (after heading and gutting) were used to prepare the minced blocks.

When minced fish is stored in the frozen state degradative changes occur. The changes that occurred in these experiments varied in nature and degree according to the species: in some the amount of saline extractable protein decreased markedly, in others the decrease was slight; the samples from some species became tougher, others softened. A taste panel detected that off-aromas and off-flavours had formed in the stored samples with consequent decrease in their acceptability. The amount of bone in the mince varied with both the species and the size of hole in the screen of the separator.

Attention is drawn to key processing factors in obtaining quality and the overall results and process of mechanical separation is discussed relevant to Australian conditions.

INTRODUCTION

A pretty kettle of fish

The proceedings of three international conferences on the technology and utilization of mechanically separated (minced) fish flesh, held in recent years at Oakbrook USA (Martin 1972), Boston USA (Martin 1974) and Aberdeen UK (Keay 1976), attest to the current interest in this field. While many of the techniques, results and observations reported at these conferences are relevant to Australian conditions, Australian species are different and the processing characteristics of their flesh unknown.

Previously unutilized fish stocks are only now being caught by the developing Australian trawl-fishing industry, as yet in its infancy. Unlike catches from the large fishing grounds in other areas of the world, local catches are generally comprised of several species, the proportion varying with the season.

Mechanical separation provides a means of recovering the maximum amount of available edible flesh from such species. Separated flesh is normally frozen into blocks, and these blocks become manufacturing material - mainly for the production of fish fingers. Many brands of fish fingers on the Australian market are made from minced fish; they are imported as retail packs or made from imported frozen minced fish blocks. There is a possibility that given certain conditions, minced

flesh from Australian species could supplant these imports if there was a sufficient regular supply of fish with suitable flesh; if that flesh were stable in storage; if the products were acceptable to the Australian consumer; and most importantly, if it was an economic proposition.

At the outset of the work reported here (January 1975) the number of potential species that could be caught was not clear, nor was there any information as to the properties of the flesh of each particular species. Therefore a separate experiment on the minced flesh of each species was done, as that species became available (i.e. was caught). The whole of the dressed fish (after heading and gutting) was processed, to provide information on the properties of the flesh, since, at this stage, estimates of the nature and volume of off-cuts were impossible to predict (and indeed still are).

The major results from the separate, but similar, experiments are reviewed here and attention is drawn to overall trends and the marked differences in some properties between the species. Observations on the relevance of some analytical procedures and on the nature and use of minced fish are also discussed, with particular application to the Australian industry.

MATERIALS AND METHODS

Materials

The species investigated are listed in Table 1 and a more comprehensive background, explanatory details and some preliminary results have already been given (Bremner 1977a-c).

In each experiment minced fish was stored at -18°C in the form of frozen blocks for periods of up to one year. Blocks were withdrawn from store at intervals, thawed and divided into three, one portion being used for analysis and the other two for taste panel evaluation.

Analytical Methods

The methods for pH, crude protein, fat, formaldehyde, free fatty acid, extractable malonaldehyde (TBA value) and saline extractable protein (SEP) have been described in detail previously (Bremner 1977b).

The saline extractable protein (SEP g/100g flesh) is often alternately expressed as percentage extractability (SEP/total protein X 100). During extraction of SEP it was found necessary to keep the homogeniser chamber cool by immersion in an ice-salt mixture (-7°C) otherwise the friction heat generated, resulted in some denaturation of the protein and a lower SEP.

The TBA estimation was done by heating an aliquot of a perchloric acid extract of fish muscle with the thiobarbituric

acid reagent, and it was noted that cloudiness and precipitation tended to occur in those extracts from the fish in which formaldehyde was produced. Therefore readings were taken on the solution immediately following the heating process, after cooling to 20°C.

Three methods were tested to analyse fish mince for bone content; an aqueous flotation method (Patashnik *et al.* 1974), dissolution of the flesh in urea (Yamamoto & Wong 1974) and alkaline digestion of the flesh with subsequent separation of bone by differential flotation in $CHCl_3$ (Dingle *et al.* 1974). The first method, although quick, did not give reproducible results; the second was time-consuming and the large volumes of solution involved made it unwieldy; the third method, even though it dissolved some of the collagen of the bone, gave fairly reproducible results with a reasonable sample throughput and was adopted as the most suitable for routine analysis.

Taste panel assessment

Details of design, sampling and presentation of samples to the panel have been given previously (Bremner 1977c). The panel were asked to score the minces and fish fingers for aroma, off-aroma, flavour, off-flavour, texture (toughness), moisture and acceptability on a nine point scale; the higher the score the greater the attribute. In the case of texture

and moisture a score of five was designated as the preferred texture and normal moisture respectively (see Bremner 1977c). Comments on the samples were invited but not actively solicited.

RESULTS AND DISCUSSION

Texture

During frozen storage changes occur in fish flesh; this is particularly the case with minced fish, where the opportunity for reaction to occur is much greater because cellular disruption has brought reactants into proximity. Textural changes are regarded as arising from denaturation of the native structure of the myofibrillar protein and the reader is referred to a recent review of this topic (Sikorski *et al.* 1976) for a full discussion of the factors involved.

In general, denaturation results in a decrease in the amount of protein which is extractable into dilute saline solution. The results for SEP are shown in Table 1 for samples before and after storage, together with the first and final taste panel texture scores and descriptive comments on texture, given by the taste panel. Since there was no compulsion for panellists to provide comments, the typical examples are merely reported; not listed with the frequency of their occurrence. Comments, such as are reported, give a

valuable description of the nature of the samples and may act as pointers to the direction of subsequent change e.g. as with cucumber fish. With the exception of the perch and the Australian salmon the fish were received in the frozen state, stored for four to six weeks at -18°C and then thawed before processing, and in part this may account for the examples of initially low protein extractability. In most species reported in the literature, the decrease in SEP follows a first order reaction (Love & Olley 1965): i.e. most change occurs initially, and the typical patterns exemplified by spiny flathead (slight decrease), ocean perch (medium decrease), and cucumber fish (rapid decrease to almost total inextractability) have been pointed out previously (Bremner 1977b). This first order reaction is then followed by a slow reaction, which in whole fish may take considerable time to reach completion. Kim *et al.* (1977) have recently described this phenomenon in lemon sole stored for seventeen years.

The level of SEP is an important indicator of the manufacturing property of the flesh and Sorenson (1976) has shown that to ensure satisfactory binding in heat-gelled products, a minimum level of 8.5-11% SEP (g/100 g flesh) is required, particularly where no other binders such as starch are present. Of the freshly prepared minces only the ling had an SEP below this minimum. Several other species decreased in SEP to below the required level over the period of storage.

Two of the species, shark and nannygai, were judged by the taste panel to be soft in texture (low score) when first examined, while the salmon became softer on storage as did

one of the two batches of ocean perch and one of the two batches of spiny flathead. Softening of stored fish flesh is considered to be due to catheptic activity and has been reported previously (e.g. Olley *et al.* 1967).

The taste panel judged that nearly all the samples became tougher (increase in texture score) during frozen storage with marked change in cucumber fish, blue grenadier, ling and red gurnard. Cucumber fish and blue grenadier both form formaldehyde in frozen storage (Bremner 1977b) while ling forms malonaldehyde and these compounds presumably account for the denaturation and resultant toughening (Sikorski *et al.* 1976). Red gurnard produced free fatty acid (FFA) in storage (195 mg FFA/100 g flesh in 200 day) and FFA has been implicated in the denaturation of fish myofibrillar protein with consequent loss of SEP and toughening of fish flesh (Sikorski *et al.* 1976). On the other hand, the nannygai produced 200 mg FFA/100 g flesh over the same period and not only did it remain soft but its protein extractability remained high. The level of FFA in other species remained low, except for the blue grenadier which produced 275 mg FFA/100 g flesh after 200 days in store: it also produced formaldehyde which reacts more rapidly with the protein than FFA (Sikorski *et al.* 1976).

While in general as SEP decreased most samples became tougher, analysis of the data by multiple regression showed that it was necessary to consider flesh pH and taste panel moisture scores as well as SEP to explain the variability in texture (Bremner *et al.* 1978).

Since there are different reasons, either singly or in combination, why fish flesh toughens in storage, it is not surprising that different comments were given by the panel to describe samples of the same toughness score; and it is reasonable to consider that the ultrastructure of the flesh is reflected in mouthfeel. Using phase contrast microscopy, Olley *et al.* (1967) presented some interesting micrographs of different patterns of myofibrillar aggregation in different species. In aggregated fibres where the water holding capacity is reduced, due to an increase in the hydrophobic nature of the protein (Aitken & Connell 1977), and where catheptic activity has weakened fibre bundles at the Z lines, the result could be flesh which is both tough and pasty.

Sensory characteristics and acceptability

Examination of the results from the taste panel indicates that the acceptability of the fish, both as a mince and as fish fingers is most affected positively by the amount of fish flavour (Fig. 1) and negatively by the degree of off-aroma

and off-flavour (Figs. 2,3). The interrelationship of flavour, off-aroma, off-flavour, moisture and texture with acceptability has been investigated further (Laslett & Bremner in preparation) and the results show the relatively greater importance of aroma and flavour characteristics over textural characteristics. This is an important point since much work in the past has been on the textural characteristics of fish, and fish mince. It points to the need for further investigations on both the natural flavours and those flavours formed during storage; such investigations require sophisticated expensive analytical equipment whereas much practical information can be gained from the use of taste panels.

There is a tendency in the trade to prefer bland tasting fish for incorporation into products, however this adult panel favoured samples with a stronger fish flavour.

Fish flavour and oxidation

Table 2 is arranged with the fish listed in descending order of fat content, and all the available data on thiobarbituric acid (TBA) values have been displayed graphically to show the trends with time, the vertical scale for each is, of necessity, different and the maximum and minimum TBA values are given alongside to illustrate the magnitude of the trends. The off-aromas and off-flavours that arise during the frozen storage of fish are generally regarded as being

due to lipid oxidation (McGill *et al.* 1977) and hence the taste panel comments are included in Table 2, and, like the texture comments the frequency is not listed, only the occurrence.

The TBA test measures malonaldehyde, an end product of lipid oxidation, and it is apparent that the TBA value is not a function of the fat level in the fish. Furthermore the high TBA values for the fatty fish, salmon and tuna, may be due to accumulation of malonaldehyde in the lipid, which, by dilution, hinders the secondary reaction with protein; the malonaldehyde remains chemically extractable. An analogous mechanism was invoked by Anderson & Steinberg (1964) to explain why added FFA did not cause the expected decrease in SEP if sufficient diluting lipid was present. In the case of the ling (which was low in lipid) the decrease in malonaldehyde is presumed to be due to reaction with the protein, resulting in toughening of the flesh, since malonaldehyde can react with food constituents to become inextractable under the test conditions. Buttkus (1967) demonstrated that malonaldehyde reacts with trout myosin at appreciable rates at -20°C and Botta *et al.* (1973) and Botta & Shaw (1975) have reported decreases in TBA values in both frozen and iced fish respectively. At any one time the TBA value is therefore a reflection of the balance between the rates of formation

and reaction of malonaldehyde with proteins and amino acids.

The taste panel scores (Table 2) for off-aroma and off-flavour increase with storage in most species and the phenomenon of "cold store odour" or "cold store flavour" developed to varying degrees as indicated by the comments. McGill *et al.* (1974) and McGill *et al.* (1977) established that hept-*cis*-4-enal is the major component of cold store odour; using pure solutions of the compound they found that even experienced tasters gave differing descriptions of its odour and that these descriptions varied with the concentration. Similar differences were found repeating the experiment using the *trans* isomer. In concentrations as low as 0.0004% the hept-*cis*-4-enal was described variously as cardboardy, turnipy, oily, rancid, creamy or astringent, comments the same, or similar, to those described in Table 2. They also found cabbagey odours in the GLC separation of volatiles from stored cod while rancid, oily, nutty, bitter, flat and stale off-flavours are commonly associated with lipid oxidation, particularly of the phospholipids.

While different comments were given by the taste panel to describe off-aromas and off-flavours that were formed in each of the species, it is not certain whether there is a true species difference, or whether there is just a

different individual response to the same end products of oxidation, as is found with hept-*cis*-4-enal.

When extracted cod lipids were oxidised, the amounts of aldehydes produced increased with time, whereas in stored cod some aldehydes increased, some decreased (McGill *et al.* 1977), indicating the complex nature of cold store odours and flavours and their changing patterns with time: simple tests - for example estimation of total carbonyls as dinitrophenyl hydrazones - may be misleading.

Taste panels, however, can be used to advantage and Howgate (1976) has shown that musty, turnipy, and metallic flavours and musty, earthy, fungal odours are prevalent in minces from the backbone areas of the fish, while mushroom flavours mentioned by the panel and which may be due to oct-1-ene-3-ol which has been detected in oxidised milk fat (Kinsella 1969).

Bones of contention

Many of the minces reported on here (Table 1) were produced using a screen with 10 mm holes until screens with 5 and 7 mm holes became available. A screen with 10 mm holes results in a coarse mince which retains more of the natural texture, but normally it would not be used because it allows too high a proportion of bone to pass through into the product. Smaller holes in the screen yield a finer mince freer of bone,

but one in which biochemical and oxidative reactions occur faster. As might be expected, results for both size and content of bone vary with species and screen size, as is evident in Fig. 4. The results for the sample of nannygai mince (put through a 5 mm screen) are comparable with those obtained on a sample of commercial fish fingers made from minced fish where the bone content was found to be 0.015g/100g flesh. It is not only the level of bone content but the size of the pieces that is of concern and the proposed standards of the FAO/WHO Codex Alimentarius Commission place emphasis in this area. The standard which covers frozen minced blocks classifies as defective a 1 Kg sample of mince if it contains two or more bones of a size greater than 5 mm in any dimension; while the standard covering fish fingers classifies as defective a sample (equivalent to 250g fish flesh) if it contains even one bone 'capable of piercing or hurting the palate after the product is cooked'.

Bone content can be minimised using good manufacturing practice, i.e. care in preparation of the fish, selection of screen size and adjustment of belt tension on the separator. Alternately the mince can be passed through a second machine, known as a strainer to remove excess bone (see below).

PROCESSING IMPROVEMENTS TO OBTAIN QUALITY

Storage life of mince depends greatly on the preparation of the fish. Belly cavity walls should be unstained, free from traces of gut, scrubbed clean of black membrane and any traces of kidney removed. The kidney, which contains a number of enzymes capable of causing deteriorative changes, is situated close to, or around, the backbone. It is therefore preferable to remove much of the backbone, kidney and associated blood vessels using a bone cutter. Haem pigments in the blood not only result in a greyish mince but are responsible for bitter flavours and catalysis of oxidative reactions (Howgate 1976). Further removal of traces of skin, scales, connective tissue, bone and fin can be accomplished by passing the mince through a strainer. The hole size in the screen of the strainer is generally about 2 mm diam. and the resulting product tends to be rather pasty. Another technique applied, particularly in Japan (Okada 1975), is that of washing the mince in ice water to remove blood, deteriorative enzymes and substrates e.g. trimethylamine oxide. This is effective depending on the degree of washing but at the same time flavour losses are great, the texture tends to become rubbery and, in all, the disadvantages probably outweigh the advantages (Bremner unpublished;

Miyauchi *et al.* 1977). Washing also gives rise to effluent problems. Peroxide bleaches have been used to lighten the mince colour (Ravichander & Keay 1976), since it has been the opinion (Elston 1975) that white fish flesh is essential for success in marketing fish fingers. This is perhaps more a reflection that the public identifies both the product with the species and a species with its natural colour. A wide range of variously coloured fish is already eaten in Australia and it is possible that products in which the fish is not white will appeal to the palate if they are also tasty and succulent. There is an obvious need for market research in this area. Where familiar species are naturally dark in colour and are labelled accordingly then the consumer accepts this. The local product must stand or fall on its own merits and costly attempts to produce a poor imitation of cod are not likely to end in success for the local fish industry.

PERSPECTIVES AND CONCLUSION

The impression could be gained that none of the Australian species examined are satisfactory as potential manufacturing material. This is not the case. Only blue grenadier and cucumber fish formed formaldehyde and careful handling of the ling may inhibit malonaldehyde formation. In fact, it is

possible that further catches with different antioxidant status in the tissue (due to feed?) may not produce such initially large amounts of malonaldehyde.

It should be remembered that during frozen storage of cod, a fish in great demand on the world market, both FFA and formaldehyde are released, SEP decreases, the flesh toughens, the flavour deteriorates, and cold store aromas and flavours develop. Other important commercial species display similar phenomena. The problems are minimized by careful handling and processing in the light of the known likely changes.

Indeed Australia is fortunate that most of the species examined, which may be caught in quantity, are those in which the changes occurring during storage are slow; and that acceptable products can be made from them. Results of two consumer acceptability trials on fish fingers (with 66 and 429 tasters) have proved encouraging. In both cases the fish fingers, made from fish mince, were well liked, with over seventy percent of respondents rating them better than, or equal to, commercial varieties (Bremner *et al.* 1976; Bremner 1977d).

The trawling effort increases, but it remains to be seen what use is made of the diverse species available, and whether mechanical separation will be employed to optimise

the potential yield from the catch. Nevertheless the information gained from this project will now be available to Government and industry alike as an aid to determining future policy.

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LEGENDS TO FIGURES

Fig. 1 The relationship acceptability versus flavour for fish minces and fish fingers indicating greater acceptability at higher flavour scores. Each letter symbol represents a mean taste panel score (Bremner 1977c). The symbol key is contained in Table 1.

Fig. 2 The relationship acceptability versus off-aroma for fish minces and fish fingers indicating lower acceptability at high off-aroma scores. Each letter symbol represents a mean taste panel score (Bremner 1977c). The symbol key is contained in Table 1.

Fig. 3 The relationship acceptability versus off-flavour for fish minces and fish fingers indicating lower acceptability at higher off-flavour scores. Each letter symbol represents a mean taste panel score (Bremner 1977c). The symbol key is contained in Table 1.

Fig. 4 Bone fragments contained per 100g fish mince, produced at maximum belt tension. Determination by the method of Dingle *et al.* (1974).

Table 1

Change in protein extractability, taste panel texture scores and typical taste panel comments on texture in frozen stored minces (-18°C)

Description of samples					Chemical and organoleptic changes during storage						
					Protein extractability		Texture scores		Storage time of mince	Typical taste panel texture comments	
					Sampling		Sampling			Sampling	
Fish[†]	Symbol	Date caught	Screen size	Total protein	first	final	first	final		first	final
			(mm)	(g/100g mince)	(%)	(%)			(days)		
Tuna	T	Mar.1975	10	22.3	56*	15	6.1	6.5	260+	sticky	granular, dry
Gemfish	G	Jul.1975	7	17.9	56*	29	5.2	6.2	200	clingy	pasty, tough
Perch	P	Mar.1975	10	17.2	99*	27	5.1	5.9	112	stringy	spongy
Salmon	A	Mar.1975	10	20.4	92*	51*	6.0	5.5	112	rubbery, crumbly	no comments
Cucumber fish (2)	C	May 1975	10	18.3	49*	17	5.5	7.1	182	cotton wool, soft, rubbery	tough, rubbery, fibrous
Blue grenadier	W	Aug.1975	7	17.0	70*	19	4.7	6.1	181	no comments	fibrous, pasty
Cucumber fish (1)	C	Dec.1974	10	17.8	76*	4	6.9	8.4	163	dry, rubbery	dry, rubbery
Silver trevally	S	Mar.1975	10	19.4	82*	61*	3.3	4.4	217	no comments	chewy, succulent
Tiger flathead	F	Apr.1975	10	19.2	74*	60*	5.4	6.3	90	no comments	chewy
Ocean perch (1)	O	Dec.1974	10	15.9	77*	40	4.8	3.8	205	no comments	granular, floury, powdery
Ocean perch (2)	O	Apr.1975	10	15.4	91*	36	4.5	6.3	162	sloppy	granular
Red gurnard	R	May 1975	10	20.0	64*	47*	5.0	6.6	182	no comments	rubbery, grainy
Saw shark	E	Mar.1975	10	18.7	59*	53*	3.6	3.8	259+	gelatinous	pap, porridge, mushy
Nannygai	N	May 1975	10	17.0	95*	88*	3.2	2.9	200	no comments	pasty, mushy
Ling	L	Jul.1975	7	17.0	47	22	5.0	7.3	193	granular, fibrous	dry, mealy, gritty, woodchip
Spiny flathead (1)	H	Dec.1974	10	16.6	88*	65*	4.7	3.7	196	no comments	woolly, powdery
Spiny flathead (2)	H	Apr.1975	10	17.4	84*	71*	3.8	5.3	167	fibrous	clingy, fibrous

[†]Species listed in order of decreasing fat content (see Table 2). Fish stored whole frozen 4-6 weeks before processing, except perch and salmon (see text).

*These samples contain more than the minimum level of 11% SEP (g SEP/100g flesh) required for satisfactory binding properties (Sorenson 1976).

+The final taste for these samples was conducted at a storage time of 38 days.

Table 2

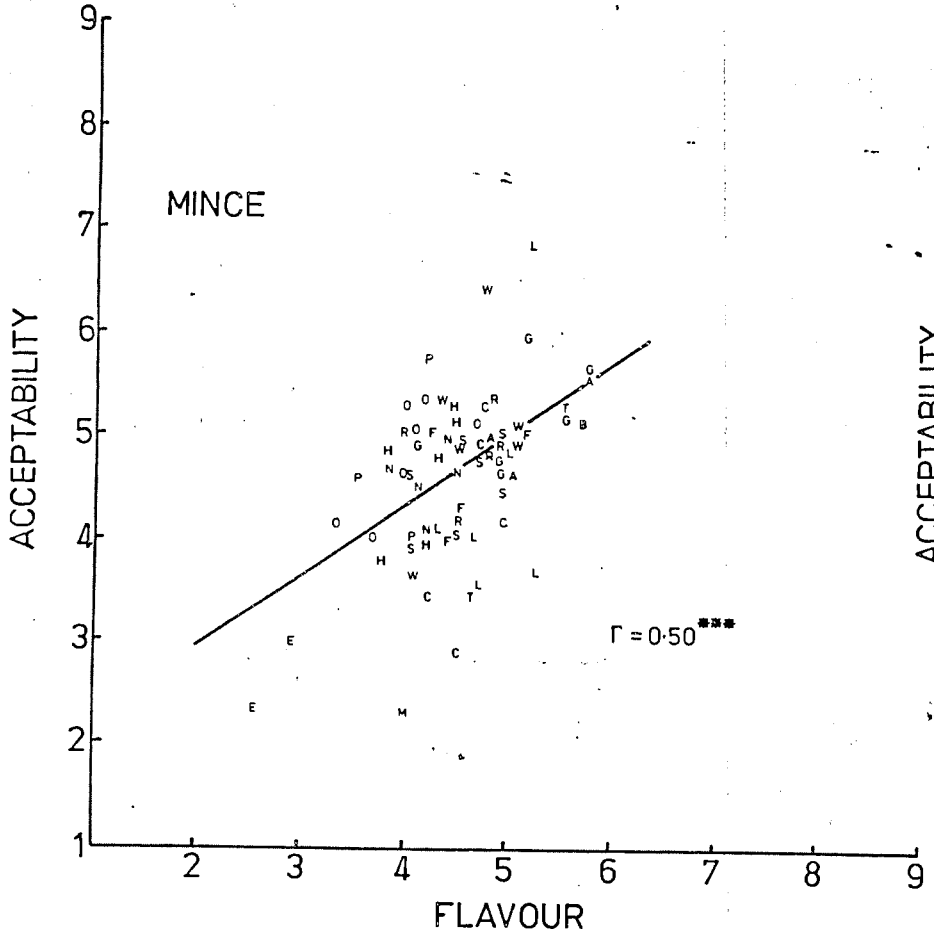
Fat levels, trends in TBA values[+], maximum and minimum TBA values, taste panel scores for off-aroma and off-flavour and typical taste panel comments on off-aroma and off-flavour in frozen stored minces (-18°C)

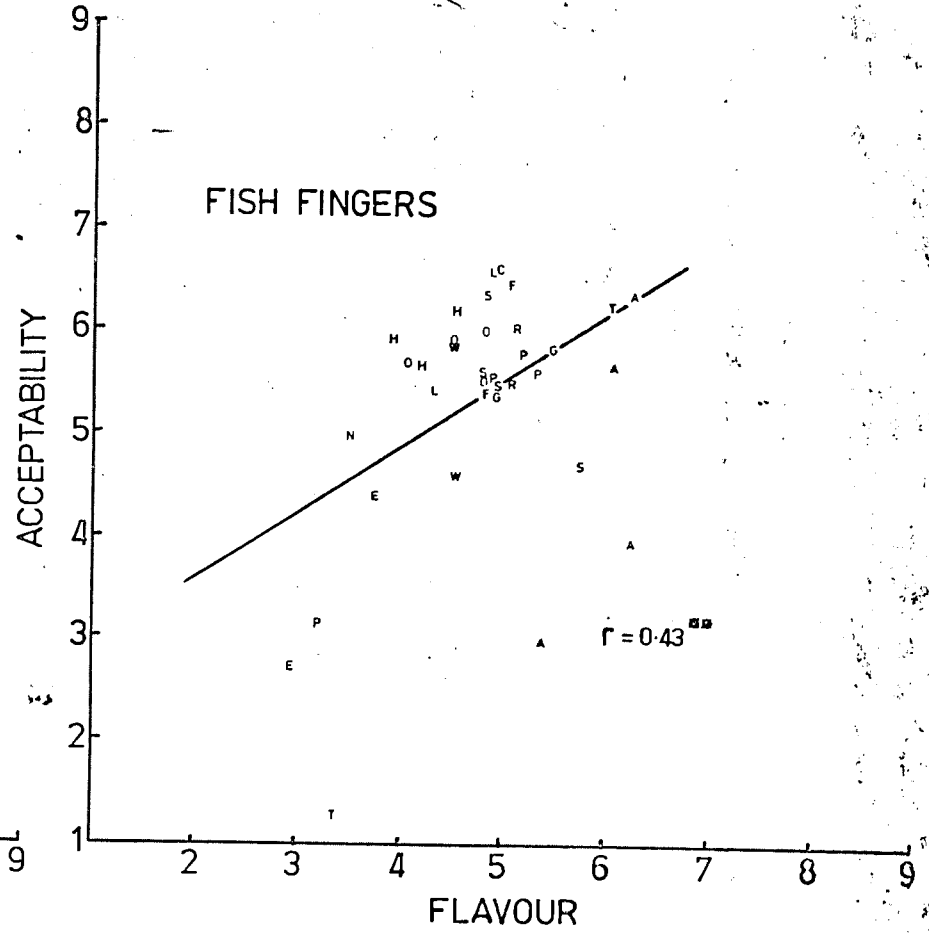
Fish	Fat level	TBA trends over 300 days[*]	TBA levels (mg malonaldehyde /kg flesh)		Taste panel scores: Off-aroma sampling		Taste panel scores: Off-flavour sampling		Storage time of mince	Typical taste panel comments at final sampling	
	(%)	0 100 200 300	max.	min.	first	final	first	final	(days)	off-aroma	off-flavour
Tuna	7.5		8.8		4.1	3.4	3.9	4.9	38	oily, stale, musty	bitter, rancid
Gemfish	5.3		2.0	0.4	2.7	2.6	2.3	3.6	200	musty	oily, bitter
Perch	4.5		0.8	0.7	3.0	4.5	2.3	2.9	112	cabbage-like, sharp	stale, mushroom-li
Salmon	4.2		9.0	4.8	3.7	3.4	3.6	3.3	112	rancid, oily	sour-acid, rancid
Cucumber fish (2)	2.5		1.5	0.3	2.4	5.2	2.6	4.2	182	stale, acrid	stale, mushroom-li
Blue grenadier	2.4		0.8	0.5	1.8	3.9	2.0	3.0	181	milky, grassy	stale, carrot-like
Cucumber fish (1)	2.1		1.7	0.7	4.8	6.9	4.3	6.6	163	sweet, turnip-like	cardboardy, flat
Silver trevally	2.0		2.2	1.1	2.1	3.9	3.7	3.8	217	ammoniacal	salty, bitter
Tiger flathead	1.6		0.9	0.4	3.5	2.9	2.7	3.9	90	chemical	rancid
Ocean perch (1)	1.5		1.1	0.6	3.6	3.6	3.3	3.2	205	nutty, milky	cooked vegetable, bland
Ocean perch (2)	1.4		1.4	0.6	3.0	3.1	2.0	2.3	162	milky	stale, bland
Red gurnard	1.4		1.7	1.0	3.5	3.4	2.2	3.1	182	sharp	no comments
Saw shark	1.0		0.6		4.3	4.0	4.0	3.8	38	ammoniacal	bitter, cardboardy
Nannygai	0.8		0.8	0.4	3.0	4.4	1.8	3.3	200	musty	meaty, stale, swee
Ling	0.7		5.7	2.0	2.3	2.5	2.3	2.6	193	musty	milky, oily
Spiny flathead (1)	0.7		1.6		4.3	4.0	4.3	3.8	196	musty, stale	nutty, stale
Spiny flathead (2)	0.7		0.8	0.5	3.0	4.4	1.8	3.3	167	milky	musty

[+]Means of at least three analyses.

[*]The vertical scale is not the same for each species. To assess the magnitude of the trends use the maximum and minimum values in the adjacent column.

[+]Dots are not joined where only one or two measurements were made.


MINCE
ACCEPTABILITY
FLAVOUR
r = 0·50***
1
2
3
4
5
6
7
8
9


FISH FINGERS
ACCEPTABILITY
FLAVOUR
r = 0·43**
1
2
3
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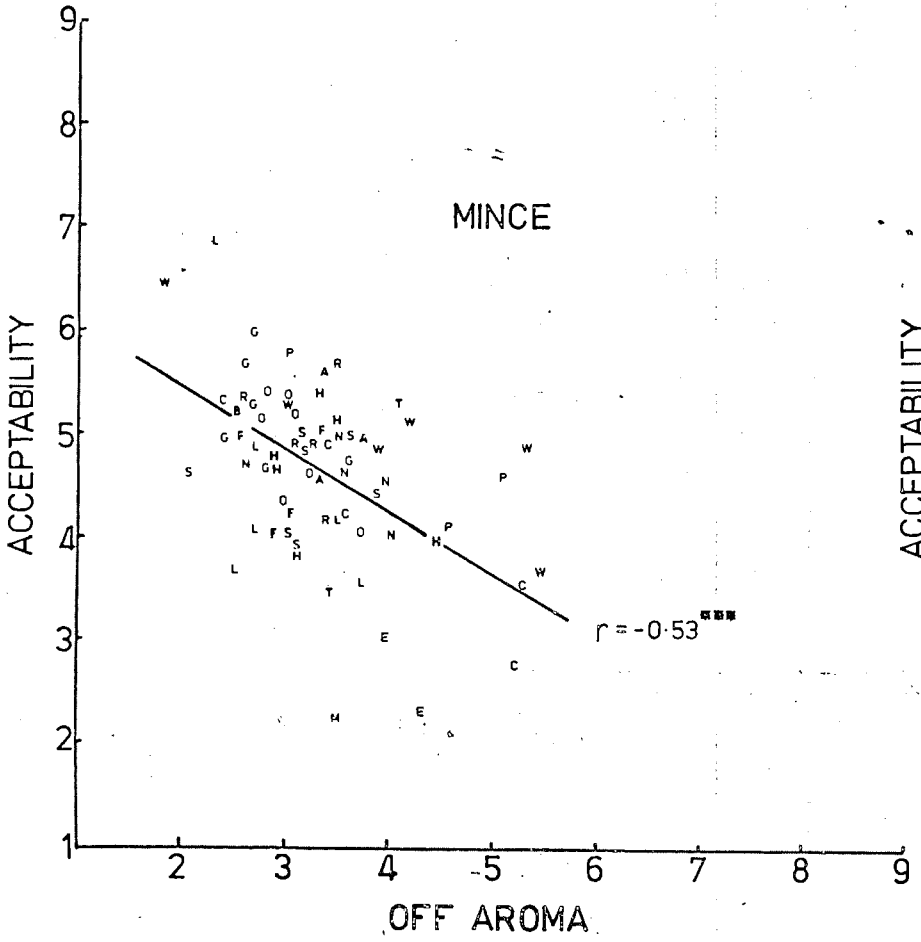
7
8
9

Fig. 1


MINCE
ACCEPTABILITY
OFF AROMA
r = -0·53***
1
2
3
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5
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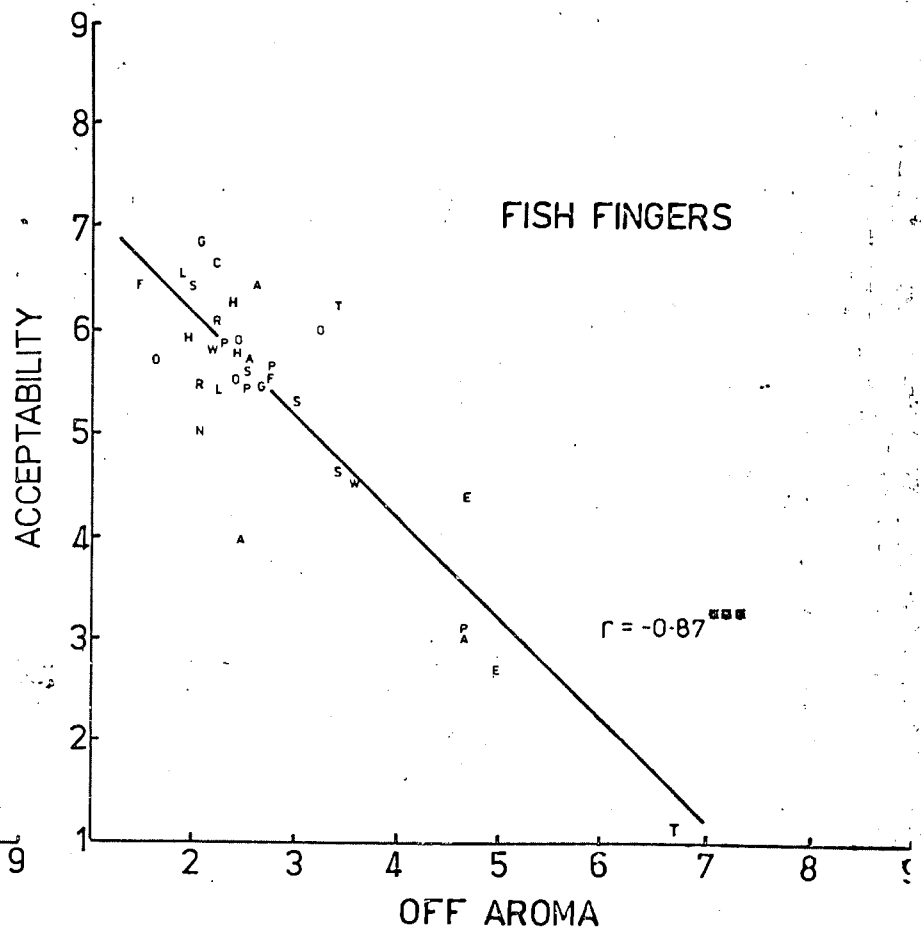
7
8
9


FISH FINGERS
ACCEPTABILITY
OFF AROMA
r = -0·87***
1
2
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8
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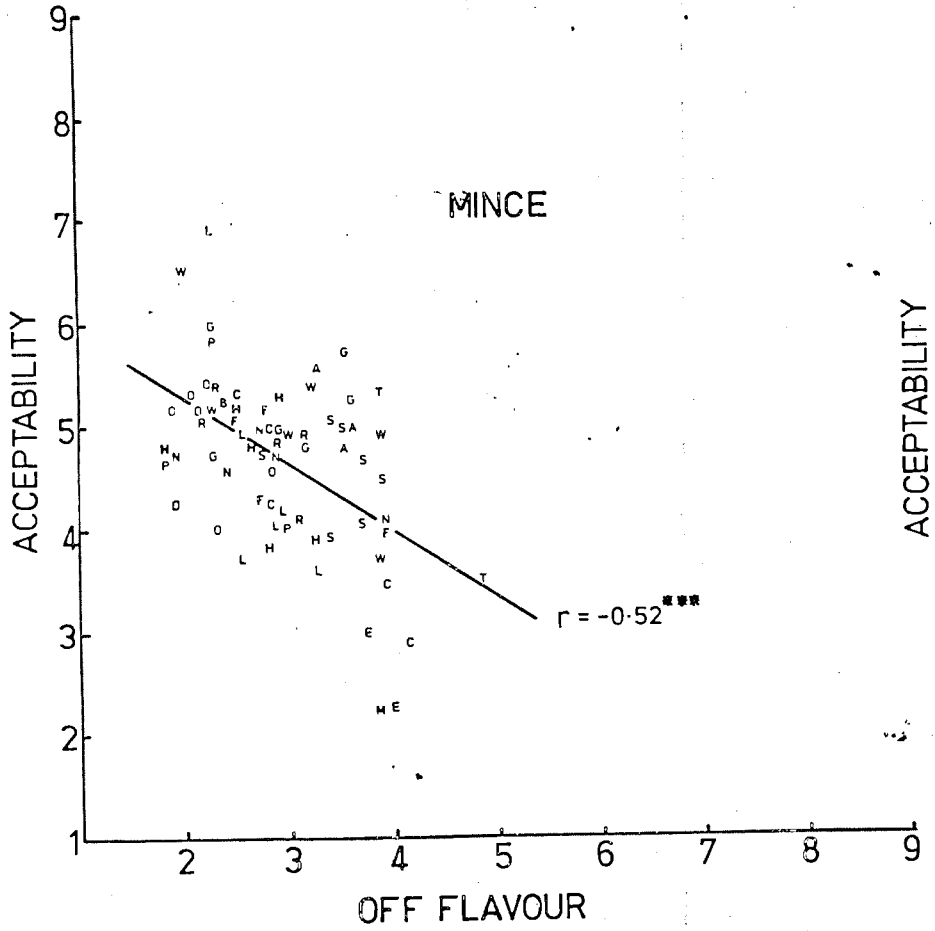
Fig. 2


MINCE
ACCEPTABILITY
OFF FLAVOUR
r = -0·52***

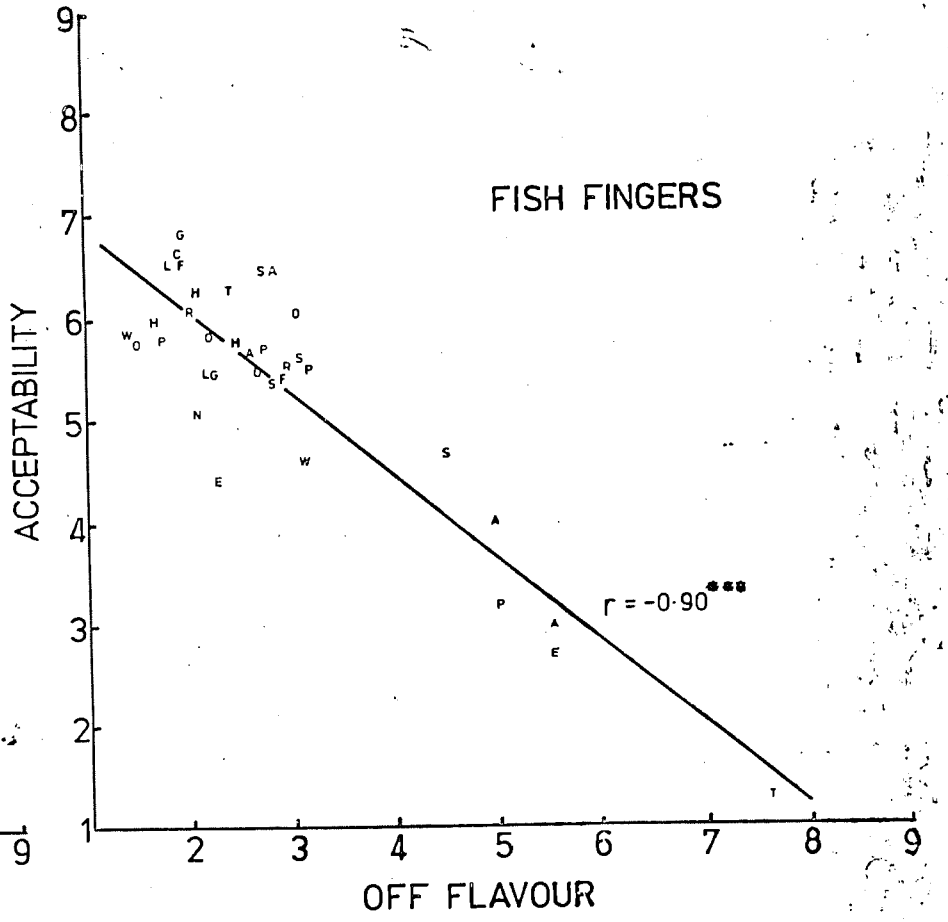
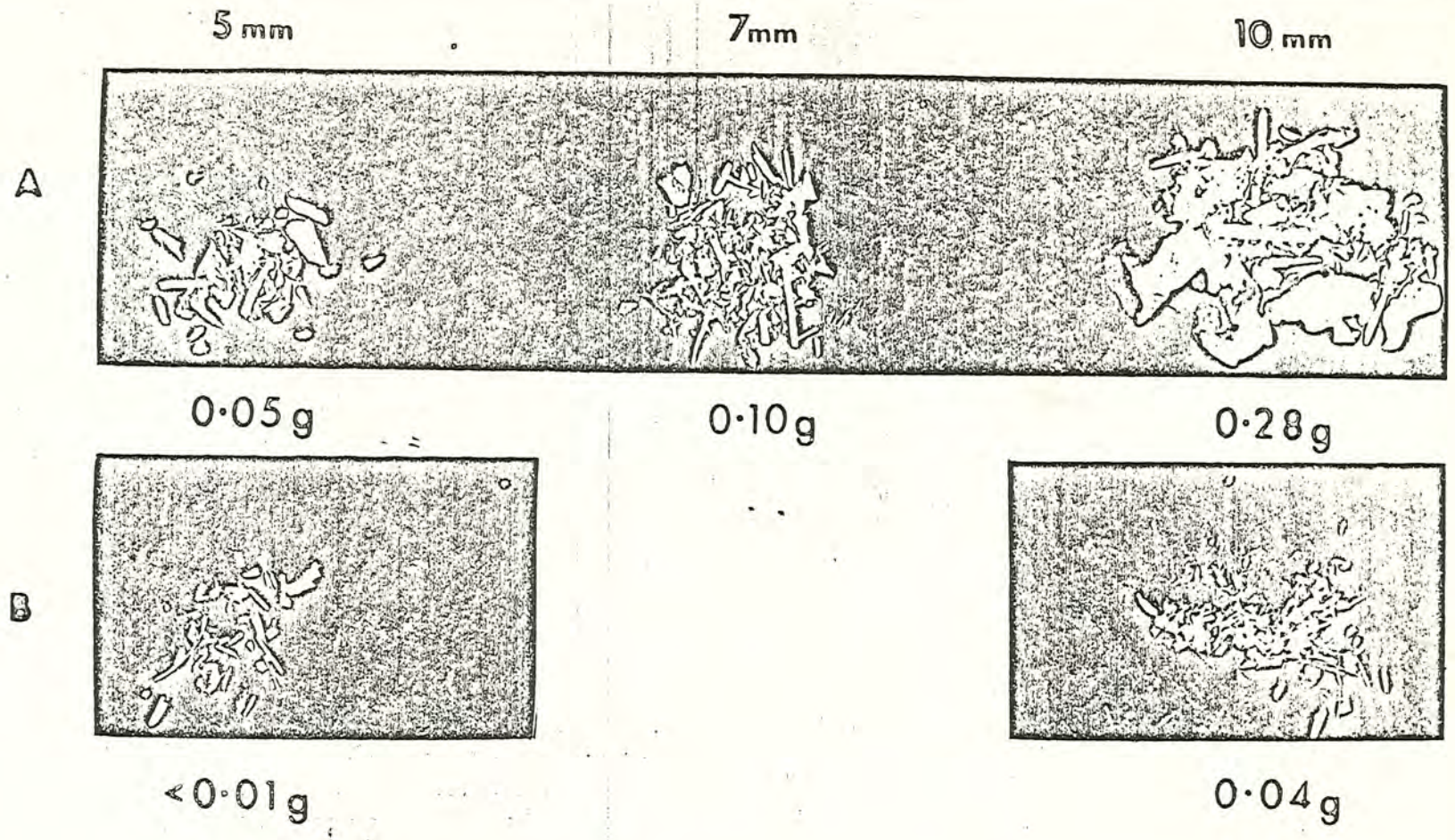

FISH FINGERS
ACCEPTABILITY
OFF FLAVOUR
r = -0·90***

Fig. 3



A SPINY FLATHEAD
B NANNYGAI

SCALE
10 mm

Fig. 4

74/15

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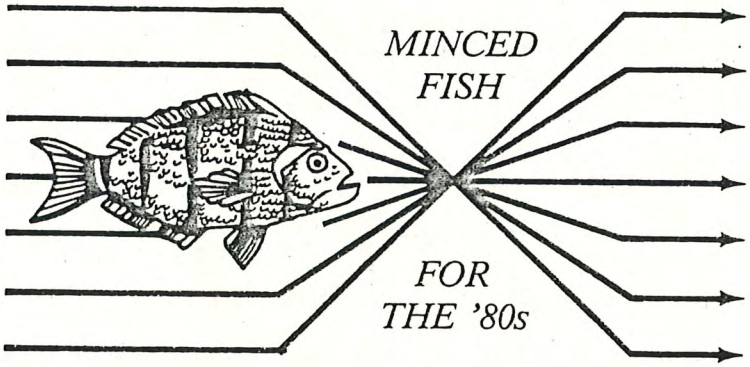
FISH FLESH

Raleigh, North Carolina
December 1-3, 1980



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MINCED FISH IN AUSTRALIA - USAGE AND RESEARCH

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Introduction

The production of mechanically recovered (minced) fish flesh in Australia is low. There are a number of reasons for this and it is necessary to cover some of them before proceeding. In global terms the Australian fishing industry is small, the main catching effort is centered on low-volume high-priced species such as prawns, lobsters and abalone, most of which are exported to Japan, Southeast Asia, and the U. S. A. There has been little in the way of a concerted effort in catching high volumes of low priced fish. This is partly because Australia has always had adequate cheap supplies of sheep and beef meat (Farrer, 1980) that are well promoted and compete advantageously with fish; so the need to exploit fish as a food source has not arisen. Furthermore, while Australia has an extensive coastline, there are insufficient upwellings of nutrient rich ocean water to support large populations of fish, thus there are few rich fishing grounds as in other continental waters.

With the exception of the northern prawn interests in the Gulf of Carpentaria, there are no company owned fleets of vessels working in an organized manner. Generally, the fishing boats are small owner-skipper operated or are family ventures. Our long and

diverse coastline means that many boats operate from small ports where the processing facilities are not geared to handle large volumes. The majority of the Australian population lives in a few main cities, the state capitals, and thus efficient transport to these markets is necessary.

Recently, there has been considerable promotion of packaged fish and fish fillets in supermarkets and much of this fish, to date, has been imported. As this market becomes established, the domestic supply of fish is expected to increase to replace these imports.

Fishermen are gradually capitalizing on the results of government sponsored exploratory trawling operations on new grounds and the declaration of the 200-mile Australian fishing zone is expected to give the local industry the necessary breathing space to develop. If the domestic catch continues to increase and more diverse and unfamiliar species are landed, then the industry is more likely to consider processes such as mechanical separation, in order to make fuller use of the catch.

Imported Minced Blocks

It is unfortunate that statistics on the import of frozen minced blocks are not recorded separately, but are combined with those for fillet blocks. It is certain, however, that the principal use of minced blocks has been for fish finger production. In the period 1965-1976, only one company operated as a contract producer of fish fingers and they claim their share of the market

rose to about 25% (Bingham, 1980). Some 20% of their product was made from minced fish blocks. In the last five years, all fish fingers sold have been wholly imported, but now two major Australian companies are manufacturing them and a third has a plant under construction. Only one of the two companies now in production is using minced as well as fillet blocks. To an extent, the proportion of minced blocks to fillet blocks used is a matter of price, and while fillet blocks remain relatively cheap, minced blocks will not be competitive. At present, the market for fish fingers is not stable because of the alleged practice of dumping imported fish fingers made from mince into Australia, and also the transition to local manufacture. The manufacturers of the local product expect to replace the need for an annual import of 6000 tons (A$12 million) of fish fingers, (Best, 1979) of which about one-third is made from minced fish.

It is difficult to ascertain the fate of those imported minced blocks that have not been used for fish fingers, they presumably have found their way into a variety of products depending on shortfalls of local supply. There are, on the market, a variety of fast food items and party-starter products such as fish bites, fishburgers, fish rolls, fish dips, etc., which incorporate minced fish. But, in most of these products other materials, e. g. rice, soy isolates, potato, flour, etc., provide much of the binding properties, with the minced fish lending the product a fish flavor rather than some unique textural characteristic.

Domestic Production

As far as can be determined, the domestic production of minced fish has not been in the form of frozen blocks and, thus, as a corollary, there has been no production of fish fingers from local species. There is a steady production of mince which seems mainly to be from fillet trimmings and the nature of this material varies from season to season and state to state. The mince may be used fresh, but is often in plastic bags for later incorporation into fishburgers, fish bites, fish rolls and canned products. Other material is used as soup stocks while the poorest grades go for pet food. There appear to be no unique products which have been developed from minced fish, nor have the potential manufacturing properties, particularly of the freshly prepared minces, been exploited.

Industrial Research

There are few food technologists employed within the Australian fishing industry, correspondingly there is not a great deal of research carried out and even less is published. There have been no technical reports from industry on minced fish, but it is known that at least one Australian company has done a considerable amount of investigation on the production of mince and now successfully incorporates mince into their products.

University/College Research

No university in Australia has a group working in the field

of fish technology, nor has there been the type of promotional product development outlined by Regenstein (1980). However, at Hawkesbury Agricultural College, New South Wales, interesting work has been done comparing the viscosity and gelling properties of several species using a Brabender amylograph and viscograph (Baumgartner, 1979). There were substantial viscosity differences between species in homogenates of their proteins and differences also between fresh and thawed materials. Emulsification capacity was measured and some studies aimed at isolating protein factors using different pHs and salt solutions of different strengths were also carried out (D'Mello, A. F. personal communication).

State Research

The fisheries authorities in each state are geared more towards biological and management problems in fisheries. With the exception of New South Wales, where investigations into the production of fish sausages from minced flesh of trawl species has been carried out, there has been no other product development fostered under state research.

A variety of types of sausage can be produced successfully (Parish, personal communication) and one batch at an informal taste trial was reasonably well received (Bremner, Lewis and Quarmby, 1976).

Federal Research (CSIRO)

The Tasmanian Food Research Unit (TFRU) is that organ of the

Division of Food Research within the Institute of Animal and Food Science which has responsibility for research into seafoods. In 1975, a program of research into the storage properties of minced fish was started. At that time, supplies were only available from the N. S. W. State Fisheries research vessel FRV 'Kapala' which was exploring the waters off the southeast coast for midwater and deepwater trawling grounds. Many of the species investigated were commercially unknown and it was decided to produce a mince from the whole fish rather than guess at the nature of the off-cuts. The details of these experiments have been published, Bremner 1977a, 1977b, 1977c, a brief summary follows.

A list of the main species of fish investigated are listed in Table 1. The fish were received frozen at TRFU, thawed in running water, beheaded, gutted, scaled and then passed through a Bibun SDX13 meat separator.

A portion of the minces from some species were washed three times in about three times their volume of icewater. The minces were stored at -18 degrees C in aluminum trays and over a 6 month period samples were taken from storage and analyzed chemically for pH, trimethylamine oxide (TMAO), trimethylamine (TMA), dimethylamine (DMA), formaldehyde (FA), free fatty acid (FFA) and saline extractable protein (SEP). Samples were also scored by a taste panel according to the score sheet abbreviated in Table 2.

The minces from a majority of the species, representing a range of orders, became tougher and drier with time in frozen storage (Bremner, 1978). No single factor or group of factors

could be inferred from the data to have a causative effect. By regression techniques, some 45% of the variance in the toughness scores could be accounted for using the parameters pH and SEP (Bremner, Laslett and Olley, 1978). When the taste panel moisture score was included in the regression, the variance accounted for was increased to 81%. The moisture score alone accounted for over 50% of the variance in toughness. Principal component analysis on the data confirmed the close inverse relation between moisture and toughness. This relation may be purely a function of the taste panel results or it may indicate that - if it is assumed that the taste panel moisture score is a sensory measurement of water holding capacity - some physical measurement of water holding capacity may be useful as a predictor of texture.

None of the chemical measurements taken in this study were useful either alone or in combination in forming a predictive model for the moisture scores.

It was noticeable that samples given the same scores for toughness or moisture exhibited differences in the nature of these parameters. For instance, those species such as cucumber fish and blue grenadier which formed formaldehyde in large quantities in frozen storage, tended to exhibit the wet-dry phenomenon (Love, 1968) where the initial impression of wetness in the mouth soon changes to that of dryness as the moisture freely escapes and a tough, dry wad remains. More detailed texture score sheets on which the initial and later impressions in the mouth are recorded, such as that outlined by Howgate (1977)

are necessary. This is particularly so where attempts are made to group species according to similarities in edibility characteristics.

In frozen fish trimethylamine oxide (TMAO) can be broken down enzymically to FA, which reacts with proteins, and DMA, which accumulates and can be readily determined (Amano and Yamada, 1965; Harada, 1975). In some species, the reaction occurs at chill temperatures, e. g. silver hake (Castell, Neal and Dale, 1973), cod, saithe and haddock (Mackie and Thomson, 1974) and at super-chill temperatures, e. g. Alaska pollock (Tokunaga, 1965). Significant production of FA and DMA was found in the gadiform blue grenadier and the myctophiforms cucumber fish and lizard fish (Saurida tumbil) (Bremner, 1978; Bremner and Snell, 1978). The gadiforms are well-known as producers of FA and DMA but the myctophiforms are less well-known in this regard. Small scale experiments have confirmed this property in two other species of the Saurida genus (S. undosquamis and S. micropectoralis) both of which are also known as lizard fish - and in two zeiformes spiky dory (Neocyttus rhomboidalis) and a commercial sample labeled antarctic dory (Allocyttus sp.). Harada (1975) also reported FA and DMA production in lizard fish (S. tumbil and S. undosquamis) and the myctophiforms Synodus hoshinonus and Trachinocephalus myops as well as in the Japanese dory (Zeus japonicus).

The results of further laboratory scale experiments, where minces were prepared by hand from a variety of non-commerical species obtained from the Australian museum, are listed in Table 3.

These samples had been in frozen storage for different times and direct comparisons of the amounts of DAM formed are not valid. Some species were very small (5-10 cm) and it cannot be guaranteed that all of the kidney tissue was removed. Kidney tissue promotes DMA formation. Since DMA formation by bacteria is slight even in spoiled fish, it is most likely that the DMA was formed enzymically, particularly where DMA levels rose on further storage. If it is assumed that FA is formed in equimolar quantities to DMA (cf. Harada, 1975) then considerable amounts must have been formed in many of the species (see calculated levels, Table 3). Levels of 100 mg/kg, whether present naturally or added to fish proteins, have a dramatic denaturing effect (Castell, 1971; Castell, Smith and Dyer, 1973; Connell, 1975; Ohnishi and Rodgers, 1980; Poulter and Lawrie, 1979; Tokunaga, 1965). Thus, each unfamiliar species should be checked for this property when its use is being considered, particularly if minces from different species are to be blended. Dingle, Keith and Lall (1977) have reported the hazards of indiscriminately mixing different species.

While the cucumber fish, blue grenadier and lizard fishes were the most prolific producers of FA and DMA, it was noted that all the other teleost species produced DMA slowly, but steadily, in frozen storage, (e. g. see examples in Table 5). This raises the possibility that FA is indeed produced in all fish but that the rates of production vary considerably from species to species by as much as two orders of magnitude. Variation of rate of production within one taxonomic order, is already well established

(Castell, Neal and Dale, 1973).

In all but two of the species listed in Table 1, which normally would not be classed as DMA producers, the flesh toughened on storage. The possibility that even very small amounts of formaldehyde were sufficient to destabilize the protein structure was considered and the data examined using multiple regression. However, only some 15% of the toughness score could be accounted for by DMA and the hypothesis can, at best, be regarded as unproven.

In those species that produced large amounts of DMA, the pH increased by 0.1-0.2 units. It would be expected that an increase in pH would cause a softening in texture due to opening of the protein helix, but either this does not occur if the protein is denatured or its effects are slight or localized in comparison to those effects holding the proteins together. Sodium metabisulfite (0.15%) was added to some cucumber fish mince. The result (Table 5) was increased production of FA and more protein denaturation.

The large amount of data from the taste panel was investigated to determine whether a relationship existed between the sensory variables scored by the panel and the acceptability of the samples. The analyses showed the importance of scoring samples for the 'off' variables such as off aroma and off flavor, since these were important determinants of acceptability. The intensity of fish flavor was also important, and in combination with the off variables, proved to be equally as important as the texture variables, toughness and moisture (Laslett and Bremner, 1979). This is an important conclusion since much of the research work on fish has been directed

towards changes in textural attributes. The results emphasized the usefulness and versatility of taste panels for this work.

Two opportunities were taken to conduct acceptability trials on fish fingers made from the mince of Australian species. In both trials, with 66 and 429 tasters respectively, the results were encouraging and over 70% of tasters rated the fish fingers better than, or equal to, commercial varieties (Bremner, Lewis and Quarmby, 1976; Bremner, 1977d).

Minces from seven different species were washed in icewater. The washing lightened the color and improved the appearance of the minces. Some fat and protein was lost in the wash water. The taste panel results for four of these minces are shown in Figures 1-4. In general, the washed minces were rated lower in aroma and flavor, higher in toughness and were less acceptable. Off aromas and flavors developed at similar rates in both washed and unwashed minces and the overall patterns of change with time in storage were similar for both the washed and the unwashed minces.

This was generally true also for fish fingers where those made from washed minces were less acceptable (Table 4). Each species gave slightly different taste panel results which were not necessarily predictable from the results obtained on the minces. This confirms that storage trials should be done on the finished product, not the raw materials.

In three species, a number of fish fingers made from the freshly prepared minces were kept in frozen storage to enable comparison with fish fingers made from stored minces. With silver

trevally the fish fingers that had been stored were rated more highly than those made from stored mince, whether it was washed or unwashed. With ocean perch, there was no difference between the methods of storage except that for the washed mince, storage as fish fingers was slightly better (higher acceptability score). For spiny flathead, there was a marginal preference for the fish fingers stored as minces particularly from the washed mince. These few results show the need for experimentation with each species to find the most stable form for storage.

Some of the results of the chemical analyses are listed in Table 5. In the washed samples, the proportion of SEP was lower initially and decreased more rapidly than in the comparable unwashed samples. Washing removed some of the materials, salts, etc., which stabilize the structure (Sikorski, Kostuch and Kolodziejska, 1975). The production of FFA was unaltered by washing, while the production of DMA was diminished but not stopped, whether the species were prolific producers or not. Washing the minces is thus of mixed benefit, some effects remain the same, and others increase and decrease. In general, the washed minces, and fish fingers made from them, are less acceptable, but color and appearance improve. Thus, washing is no panacea and it is necessary to investigate its effects on each species for the particular advantages and disadvantages it may have.

The binder formulation of Teeny and Miyauchi (1972) was used in experiments with red gurnard and nanny-gai, and a modified binder with half the amount of sucrose (0.5%) for gemfish. Unfor-

tunately, some panelists were sensitive to the surface active agents used to disperse the anti-oxidant and could detect them as a bitter taste, furthermore, the sucrose in the binders made the product unacceptable to the taste panel. A much simpler binder using only 1% salt and 0.15% polyphosphate was used with silver trevally and ocean perch. For these reasons only, comments on the textural attributes will be considered here.

In the silver trevally minces treated with binder, the toughness scores for both washed and unwashed minces were notably lower and the moisture scores slightly higher. Ocean perch minces treated with binder had an initially lower toughness score and higher moisture score with the effects being more noticeable in the washed minces. In the red gurnard and nannygai, the binders had no apparent effects on toughness and moisture scores. However, in the gemfish, the minces treated with binder were tougher and drier, but the effects were less noticeable in the washed minces.

This conflicting information again stresses the importance of doing experiments on each individual species. It is not sufficient to select one species which is prone to change and find how to improve it, then extend the results to other species.

Identification of Species

There is considerable interest in the need to identify the species of origin in a minced, or fillet, block. Polyacrylamide gel electrophoresis (PAGE) has proven itself useful as a tool for this analysis. Nevertheless, since the proteins denature faster

in minces than in whole blocks, a system based on extraction and separation of the water soluble proteins may fail if sufficient change has occurred to make these proteins extractable. This could occur particularly in fish where FA is formed rapidly as, for example, red hake, where after only 65 days at -10 degrees C some 78% of its protein was inextractable even in 1% SDS (Dingle, Keith and Lahl, 1977). In these circumstances, the water soluble proteins may be either 'locked' in a matrix of denatured myfibrillar protein or be inextractable due to direct reaction with FA.

In experiments on cucumber fish mince, it was found that even after 3 years, stored at -18 degrees C, when the SEP was only 3%, that the PAGE of an aqueous extract still gave patterns that were easily identifiable. With blue grenadier, however, some of the slower moving bands were lost, but because of the distinctive pattern of some faster moving bands, identification is still possible (Figure 5). It seems likely that FA at a sufficient level can crosslink the slow-moving sarcoplasmic proteins in this fish.

Fundamental Aspects of Protein Denaturation

At the same time as the practical experiments were in progress, consideration was being given to the fundamental nature of the changes that occur in proteins in frozen storage. These deliberations occurred while Professor Z. E. Sikorski of the Poletechnika Gdanska was on sabbatical leave at TFRU and a review was published (Sikorski, Olley and Kostuch, 1976). This review was based on the concepts of Lewin (1974) in his book 'Displacement of water and its

control of biochemical systems': the basic tenet being that water provides a supportive and buttressing role to protein structure and that any ion or molecule coming within the configuration of a protein molecule will alter the properties of the protein, the water monolayer and the bulk water. Fennema (1977) has also written that "Proteins influence the properties of vicinal water, and water, in turn, dictates to a considerable degree the properties of protein."

The agents or phenomena likely to cause change, to influence, or to measure the results of influences on protein molecules have been covered recently by Olley (1980). These are primarily the phenomena that could be classed as physical, e. g. water activity, specific conductivity, electrical charge, diffusion, ionic strength, osmolality, dipole relaxation and secondly, the phenomena best described as being chemical or biochemical, e. g. amino acid levels, rigor mortis, formaldehyde, buffering capacity, pH, salt concentrations, nucleotides, adhesion of myofibrils, electron acceptors, lipid oxidation, presence of cryoprotectants, fatty acids, reducing agents, disulfide bond splitting agents, deconformation of myosin helix and removal of specific water molecules at very low temperatures. Many of these phenomena are related, to a greater or lesser extent, and experimentally many need to be taken into account. With such a comprehensive number of diverse phenomena, it is most unlikely that there is any simple single reason for protein denaturation and that a solution for the prevention of denaturation will be discovered. In recent studies, Tsuchiya, Tsuchiya and

Matsumoto (1980) have shown that many different types of bonds, ionic, disulfide, hydrogen and hydrophobic were all involved in the process of denaturation. Indeed, the question that could be asked is not, why do some proteins denature in frozen storage, but why do some not denature? A low and steady storage temperature to restrict water and solute movement, and restricted access of oxygen are still the main requirements for stability in storage.

The future of minced fish production in Australia depends not only on the continued expansion of the trawling industry, but also on the fluctuations in price of blocks of mince and fillets on the international market. It is certain, however, that each potential species and the way each process interacts with it requires separate study.

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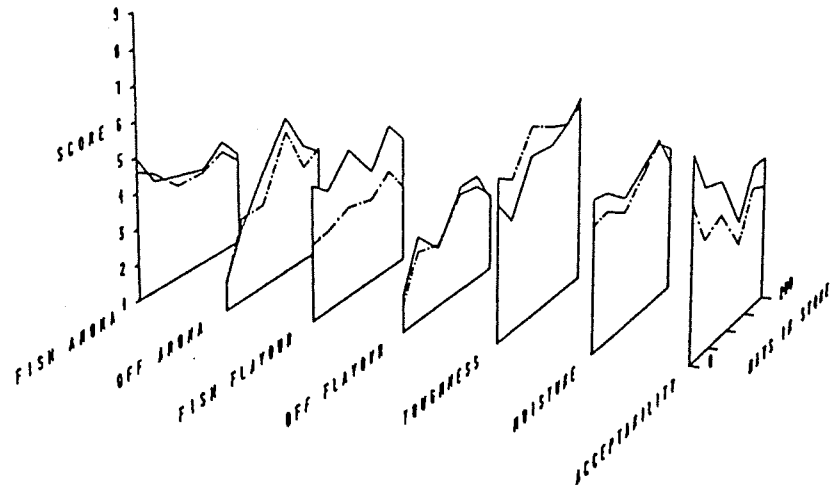

Fig. 1 Taste panel scores obtained with stored blue grenadier minces.

Solid line, untreated mince.
Broken line, washed mince.

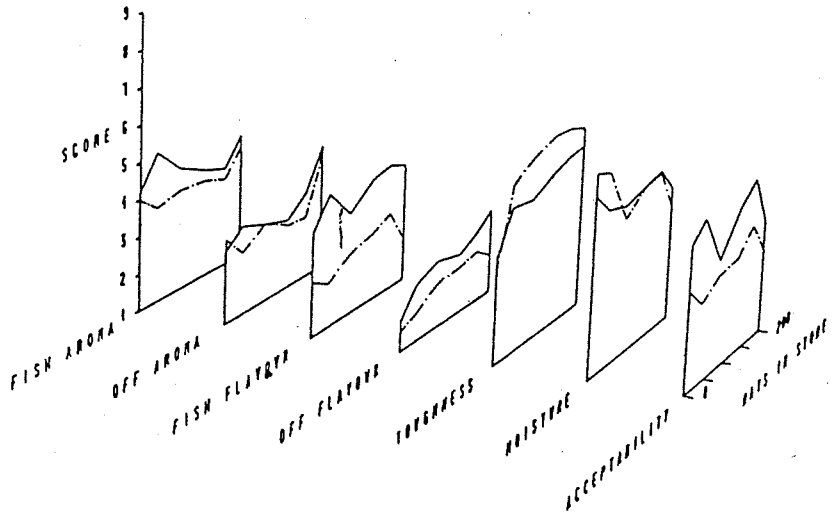

Fig. 2 Taste panel scores obtained with stored ocean perch minces.

Solid line, untreated mince.
Broken line, washed mince.

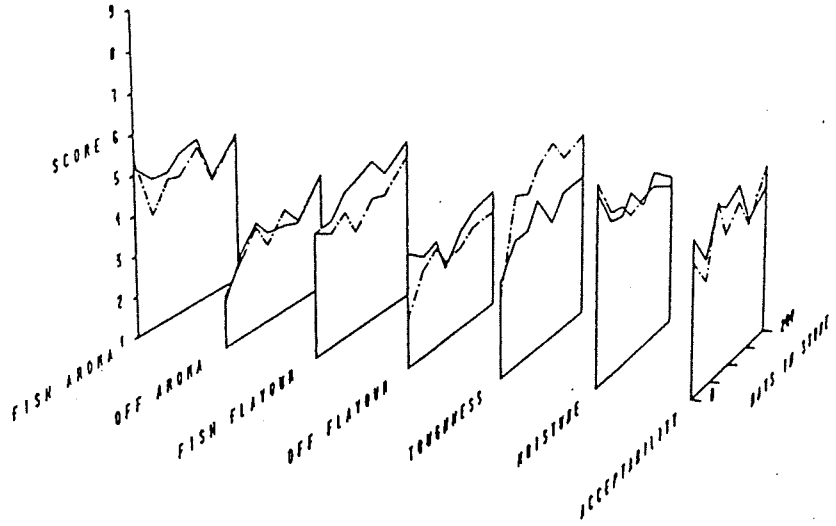

Fig. 3 Taste panel scores obtained with stored silver trevally minces.

Solid line, untreated mince.
Broken line, washed mince.

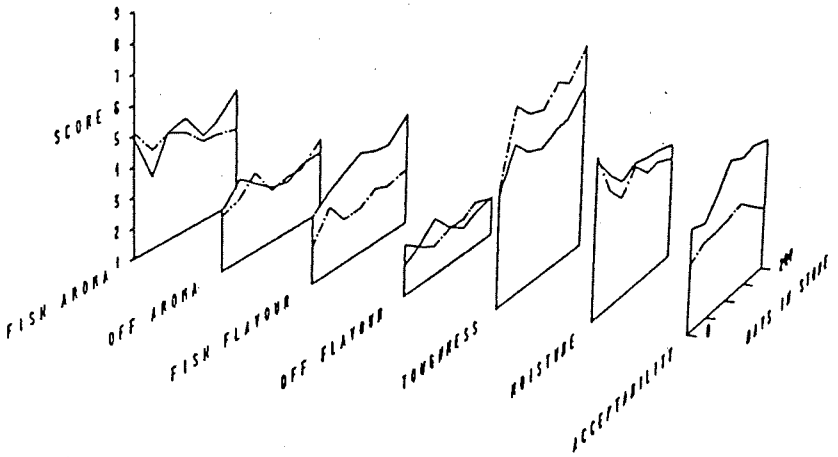

Fig. 4 Taste panel scores obtained with stored spiny flathead minces.

Solid line, untreated mince.
Broken line, washed mince.

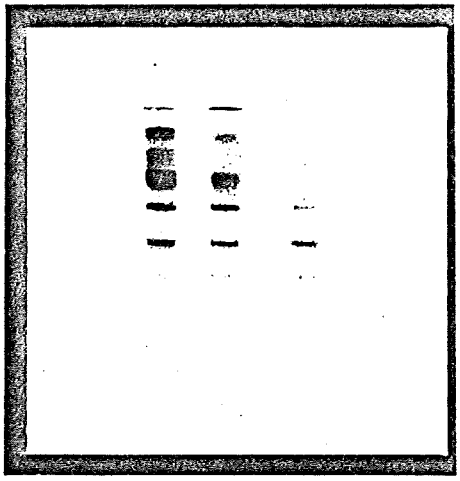

Fig. 5 Electrophoretograms of (from left to right) fresh blue grenadier, stored blue grenadier fillet (thoroughly denatured) and blue grenadier mince stored nearly 4 years.

Table 1. List of Species Investigated

Common Name	Generic Name
Australian Salmon	Arripis trutta esper
Barracouta	Leionura atun
Cucumber fish	Chlorophthalmus nigripinnis
Saw shark	Pristiophorus cirratus
Gemfish (hake or king barracouta)	Rexea solandri
Ling	Genypterus blacodes
Morwong (jackass fish)	Cheilodactylus macropterus
Nannygai (redfish)	Centroberyx affinis
Ocean perch	Helicolenus papillosus
Perch*	
Red gurnard	Currupiscis kumu
Silver trevally	Usacaranx nobilis
Spiny or deep sea flathead	Hoplichthys haswelli
Tuna	Katsuwonus pelamis
Tiger flathead	Neoplatycephalus richardsoni
Blue grenadier (whiptail)	Macruronus novaezelandiae

*Generic name not identified

Table 2. Taste panel score sheet

Taste panel score	Fish aroma	Off aroma	Fish flavor	Off flavor	Toughness	Moisture	Acceptability
9	Very strong	Very strong	Very strong	Very strong	Tough	Very wet	Very good
7	Strong	Strong	Strong	Strong	Slightly tough	Wet	Good
5	Moderate	Moderate	Moderate	Moderate	Preferred texture	Normal moisture	Moderate
3	Weak	Weak	Weak	Weak	Slightly soft	Dry	Poor
1	Very weak	None	Very weak	None	Soft	Very dry	Very poor

Table 3. Laboratory experiments on FA and DMA production

Order	Family	Species	DMA (mg/kg) When mince prepared	DMA (mg/kg) After 30 days at -18°C	Estimated FA (mg/kg) equivalent
Myctophiformes	Myctophidae	*Electrona rissoi*	122	185	122
		Symbolophorus bernardi	270	417	275
		Lampanyctus australis	140	160	106
		Lampanyctus intricarius	200	250	167
		Scopelopsis multipunctatus	330	-	220
		Diaphus kapalae	360	950	633
		Diaphus danae	230	290	193
Gadiformes	Moridae	*Euclicthys polynemus*	234	247	165
		Tripterophycis intermedius	977	1170	780
	Macrouridae	*Lepidorhynchus denticulatus*	175	205	137
		Malacocephalus laevis	377	442	295
Cetomimiformes	Ateleopidae	*Ateleopus*	123	117	78
Lophiiformes	Ogcocephalidae	*Halieutaea*	175	218	145
	Chaunacidae	*Chaunax fimbriatus*	390	390	260

Table 4. Description of differences in taste panel scores between fish fingers made from washed mince, compared to those made from untreated mince from both freshly prepared minces and from stored minces (-18°C)

Blue grenadier	Initially the fish fingers made from washed mince were slightly tougher but after 182 days in store the scores for fish fingers made from both washed and unwashed material were very similar. After storage off aroma and off flavor had increased, they were tougher, drier and less acceptable (cf. Bremner 1980).
Cucumber fish	Initially the fish fingers from washed mince had a lower aroma and flavor, were tougher, drier and less acceptable. Storage trials were not done.
Gemfish	No real differences between fish fingers made from washed or unwashed minces either initially or after 217 days in store when they were equally tougher, drier and markedly less acceptable.
Ling	Initially the fish fingers made from washed mince were less acceptable but after 205 days the ratings for fish fingers made from washed and unwashed minces were identical. Both were considered drier, tougher with more off-aroma and off-flavor, and were less acceptable than the initial samples.
Ocean perch	Initially fish fingers from washed mince had slightly lower flavor, were slightly tougher and slightly less acceptable, while those from stored minces (216 days) had less aroma, less flavor, were tougher, drier and less acceptable. The initial slight differences were accentuated on storage.
Silver trevally	Initially those fish fingers from washed mince had less flavor, were tougher, drier and less acceptable, while those from stored mince (200 days) were also lower in flavor, had higher off-flavor, were tougher and much less acceptable, viz. the fish fingers from washed material deteriorated to a greater extent.
Spiny flathead	No difference initially but those from stored minces (200 days) were slightly less acceptable.

Table 5. Effects of washing minces on SEP, FFA and DMA levels in frozen storage (-18°C)

Species and time in store	Treatment	SEP g/100g flesh initial	SEP g/100g flesh final	FFA mg/100g flesh final	DMA mg/kg flesh initial	DMA mg/kg flesh final
Blue grenadier	-	11.9	3.8	274	106.0	900.0
182 days	washed	9.8	1.6	206	48.0	410.0
Cucumber fish	-	9.0	3.2	159	173.0	816.0
182 days	washed	7.2	2.6	141	47.0	90.0
	+0.15% $Na_2S_2O_5$	9.0	0.0	57	173.0	1405.0
Gemfish	-	10.3	9.1	79	9.1	15.1
194 days	washed	7.6	3.9	84	6.6	16.3
Ling	-	7.9	3.8	62	5.4	9.6
193 days	washed	7.9	2.8	64	3.8	6.2
Ocean perch	-	11.9	5.8	56	2.3	4.5
205 days	washed	11.3	3.9	54	1.4	2.2
Silver trevally	-	16.1	11.9	60	4.8	9.5
217 days	washed	12.2	6.7	60	1.9	3.4
Spiny flathead	-	15.5	11.4	46	2.3	4.9
196 days	washed	13.3	8.9	41	2.0	2.5