FISHING INDUSTRY RESEARCH TRUST ACCOUNT

FINAL REPORT

1. Title of project: Investigation of key factors in the maintenance of quality from catching to consumer.

2. Organisation: CSIRO, Division of Food Research.

3. Section: Tasmanian Food Research Unit (TFRU) Hobart. As from 1.4.86 Seafood Technology Group, CSIRO, Division of Fisheries Research.

4. Persons responsible for programme:

   Dr. June Olley, D.Sc., Leader, TFRU.

   Mr. H. Allan Bremner, M.Appl.Sc. Senior Experimental Scientist.

5. Location of operation: The work was based at the Tasmanian Food Research Unit but some experiments were carried out on two cruises of the FRV 'Soela' to the North-West Shelf.

6. Work schedule:

   Commencement date: 1.7.83

   Completion date: 30.6.86.

7. Final Report: 5 papers have already been published from the work. The FIRTA travel funds enabled the microbiologist and the engineer employed with FIRTA money to accompany the permanent staff of TFRU to a joint CSIRO-DSIR Workshop on seafood technology from the 7-11th April. The workshop was held in Nelson and included industry visits. The main results of the work from the current grant were presented as 15 papers at this workshop and arrangements had been tentatively made for the proceedings to be published by Fishing News (Books) Ltd. (A grant application will be made to FIRTA to cover the costs of preparing camera ready copy.) The Abstract of each paper is included in Appendix 1 to this report.

The main findings over the three year period, either published or delivered at the Workshop, are given below.

The quality of a resource depends on the intrinsic properties of the species, the subsequent handling at sea and at the wholesale and retail level.

The work has focused particularly on fresh seafoods due to a marked swing back to a preference for unfrozen products by the general public. However, in the main, products for export must be frozen and a comprehensive study has therefore been undertaken on the effects of freezing seafood products in the commercial fibreboard packages in which they are to be ultimately shipped.

INTRINSIC PROPERTIES

Finfish.

a) Tropical species: It has been widely held that tropical species of fish have a longer shelf-life on ice than fish from temperate waters. Mesophilic spoilage bacteria from fish in tropical waters are suppressed by the unfavourable temperature of melting ice and the small numbers of psychrotrophic bacteria present initially on the fish or added with the ice take a considerably longer time to cause spoilage. In temperate water species they are already present and adapted to multiply immediately on capture. Examination of four commercial species from the North-West shelf has shown this to be a half truth. Because the fish do keep well on ice the loss of acceptability is determined by the intrinsic properties of the species and not by bacterial spoilage. The nucleotide adenosine triphosphate which controls the energy flow in all living matter is degraded by a series of enzymes in finfish by the following pathway.

ATP	->	ADP	- >	AMP	- >	IMP	- >	In	- >	Hx.
Adenosine triphosphate		Adenosine diphosphate		Adenosine monophosphate		Inosine monophosphate		Inosine		Hypoxanthine

Inosine monophosphate is a flavour enhancer and hypoxanthine is a bitter compound. The other compounds appear to be flavourless. In those species of fish in which IMP breaks down rapidly and the conversion of inosine to hypoxanthine is also rapid, flavour acceptability deteriorates quickly, while in species where IMP breaks down slowly and inosine accumulates the acceptability remains high for long periods on ice.

b) *Sterile temperate water species*: Experiments on sterile trumpeter, mullet and snapper carried out in collaboration with a visiting scientist from DSIR, New Zealand has shown that techniques which attempt to extend the shelf life of seafoods such as irradiation, modified atmosphere packaging, and treatment with sorbate or other preservatives - cannot extend the shelf life indefinitely due to the intrinsic nucleotide changes mentioned above and due to oxidative rancidity changes in the fattier species. It is important to determine which factor is likely to limit the shelf life of individual species.

Combining the taste panel assessment of the fish from these experiments and those of the fish from the North-West shelf has shown that 70% of the variability in the flavour acceptability of widely different species is accounted for by the IMP and Hx content of the flesh.

RIGOR MORTIS IN TROPICAL SPECIES

Workers at the Tropical Products Institute, London, U.K. have studied pond held species of tropical fish and claim from their work that tropical species suffer cold shortening when chilled immediately on capture. The marked

temperature differential causes an energised release of calcium from the sarcoplasmic reticulum of muscle producing a rapid stiffening of the fish when they are iced. This phenomenon precedes rigor mortis and has deleterious effcts on filleting yields and drip loss. The U.K. workers recommend different codes of practice for tropical species to prevent this. Experiments on species trawled on the North West shelf show that these conclusions are erroneous. Trawled fish have struggled sufficiently in the net to markedly reduce their pH and ATP levels. In the opinion of meat researchers at CSIRO, Cannon Hill, no cold shortening would be possible. One large species of fish (*Diagramma pictum*) which survived the haul alive may have had sufficient energy reserves to cold shorten. Further work is required on the larger species; this may have implications for the tropical tuna fisheries. Many tropical fishing operations involve surround netting and more recently trapping which would not exhaust fish and here again cold shortening could occur.

A NEW TROPICAL RESOURCE

It seldom happens that the storage life both fresh and frozen is studied in detail before a resource becomes commercially available in extensive quantities. This grant and the availability of accommodation on the FRV 'Soela' facilitated a thorough study of three species of scampi *Metanephrops andamanicus, M. australiensis* and *M. boschmai* caught on the North West shelf. Preliminary data has been published in Australian Fisheries (Appendix 2) and two full papers were presented at the CSIRO-DSIR workshop.

EXTENDING THE SHELF-LIFE OF FRESH SEAFOODS.

Packaging techniques were developed with the previous FIRTA grant and shelf life extension was obtained with potassium sorbate, a GRAS (generally recognised as safe) substance in the USA, but not generally accepted as a food additive in Australia. Attention was therefore turned to modified atmosphere packaging. The work commenced with a review of the background literature (Appendix 3). Storage trials on scallops, trevalla and morwong held in 100% CO_2 at 4° C were completed and combinations of CO_2 and sorbate were tried with morwong (Appendices 4 and 5). The efficacy of CO_2 depends on the partial pressure, so trials with 100% CO_2 gave the maximum shelf life possible. It is usual to include some oxygen in the pack due to fear of anaerobic conditions and development of *Clostridium botulinum*. This can lead to a feeling of false security and no packaged product should be held above 4° C. Consequently a survey of the temperature histories of retail cabinets is being undertaken with the current FIRTA grant.

BOTULISM HAZARDS IN SORBATE TREATED SEAFOODS

Because of the great success in vacuum packaging scallops in the presence of K - sorbate during the previous grant and due to the presence of a possible lucrative market in the USA, extensive storage trials on scallops inoculated with *Clostridium botulinum* were undertaken. The results were presented at the CSIRO-DSIR workshop. Scallops should be safe providing the chill storage chain is carefully controlled and periodically monitored.

TASTE PANEL EXPERTISE

The TFRU has built up a formidable expertise in analytical and consumer taste panels, the latter using the General Foods of America "Smiley Scale" (Fig. 1). Extensive odour and flavour profiling has also been done. The taste panel is run by Miss Maria Ottenschlaeger who now has six years

experience in the techniques and is employed with FIRTA funds. Papers specifically concerned with taste panel techniques were presented at the CSIRO-DSIR workshop.

SENSORY EVALUATION OF SEAFOOD PRODUCTS

A program was first developed for a CASIO computer to grade fish on a system of demerit points (Appendix 6). Due to the world wide interest in this concept a local company Minex Pty.Ltd. has designed and built a commercial model.

FREEZING OF SEAFOODS

Extensive studies have been carried out on the effect of various commercial fibre-board-packs on the freezing rates of tylose blocks. The results were presented at the CSIRO-DSIR workshop. The work on the freezing of commodities such as abalone, lobster tails and fish fillets was almost completed by the end of the grant and will be finalised in the first few months of the current grant.

EFFECT OF ON BOARD HANDLING ON QUALITY OF FROZEN STORED FINFISH

Storage trials on gemfish frozen after five on-board handling treatments (previous FIRTA GRANT) were continued during one year of storage at -18° C. Sensory and chemical assessment of the products were discussed in two papers given at the CSIRO-DSIR Workshop.

CONCLUSIONS

The main findings and areas where further work follows directly from them are listed in Appendix 7.

APPENDIX 1

Abstracts of papers from the joint CSIRO-DSIR Workshop, Nelson, New Zealand. The full text of these papers is available on request.

Prediction of Freezing Times of Packaged Foods in Air Blast Freezers.

S. J. SYKES

ABSTRACT

A previously developed modification of Plank's equation for freezing time is used to investigate the effect on freezing time of fibreboard packaging of products frozen by air blast. Freezing experiments carried out on a range of commercial fibreboard cartons containing Karlsruhe test substance show that a product in fibreboard takes between 60 and 150% longer to freeze than an identical product frozen under the same conditions but without packaging. The equation used underpredicts the freezing time of packaged products by up to 20%.

Influence of Nucleotide Catabolism on the Storage Life of Tropical Species of Fish from the North West Shelf of Australia.

A.M.A. VAIL, H.A. BREMNER, J. OLLEY AND J.A. STATHAM.

ABSTRACT

The storage life of four species of fish from the North West Shelf was examined by means of nucleotide catabolism and sensory evaluation. It was found that the shelf life was related to the rate of inosine monophosphate (IMP) breakdown rather than to bacterial spoilage, because the endemic mesophilic bacteria were unable to adapt to ice storage conditions. The results established that the IMP level was fundamentally related to both flavour intensity, and acceptability and was not merely circumstantially related to time of storage.

See also Appendix 6.

The Shelf-life of Sterile Yellow-eyed Mullet (*Aldrichetta forsteri*) at 4° C

G.C. FLETCHER and J.A. STATHAM

ABSTRACT

The relative effects of microbial and non-microbial spoilage on the shelf-life of yellow-eyed mullet were studied. Sterile portions of fish flesh were held for 16 days at a temperature of 4° C. Results for their sensory and chemical analyses were compared with fillets which had either naturally spoiled while held at 4° C, or frozen fillets held at -18° C. Within 6 days, significant changes were observed in the frozen flesh and after 69 days it was considered unacceptable.

At 4° C, nucleotide catabolism resulted in a rapid production of inosine from inosine monophosphate (IMP), followed in sterile flesh by a slower breakdown to hypoxanthine. Hypoxanthine production from inosine was rapid in the presence of bacteria. A bitter flavour associated with hypoxanthine appeared to contribute to loss in acceptability. Other off odours and flavours also developed at a slower rate in the absence of bacteria. Their development was not sufficiently slow to result in a significant extension in shelf-life for this species. The increase in pH normally observed during spoilage did not occur in the absence of bacteria.

The Shelf-life of Sterile Trumpeter (*Latridopsis forsteri*) at 4° C.

G.C. FLETCHER AND J.A. STATHAM

ABSTRACT

Sterile flesh of bastard trumpeter (*Latridopsis forsteri*) spoiled within 7 days at 4° C, due to autolysis and oxidation. The rate of nucleotide catabolism was such that, even in the absence of bacteria, autolytic changes could contribute significantly towards the spoilage of this species. Oxidative changes were prevented in frozen material by vacuum packaging.

Inosine Monophosphate, Hypoxanthine and and Fish Flavor Acceptability.

G. FLETCHER, J. OLLEY, J.A. STATHAM AND A.M.A. VAIL

ABSTRACT

The adenosine triphosphate catabolites, inosine monophosphate (IMP) and hypoxanthine (Hx), are measures of fish flavour acceptability. Japanese workers have related flavour acceptability to K-value, which is in effect a measure of IMP breakdown. Seventy one samples of fish from temperate and tropical species were assessed for flavour acceptability on a "Smiley" scale. The samples were stored iced, frozen or sterile and were assessed from immediately after capture through to the point of rejection. A multiple regression analysis showed that 70% of the variance in flavour acceptability was accounted for by the concentration of IMP

and Hx in the flesh at the time of cooking. 'Fresh', 'boiled' and 'creamy' descriptors in steamed fresh fish are not accounted for by IMP and Hx nor are rancid, astringent and sour notes associated with the later stages of spoilage.

Time scale of cold shortening and rigor mortis in North West shelf species caught under semi-commercial conditions.

A.M.A. VAIL AND J. OLLEY

ABSTRACT

Workers at the Tropical Development and Research Institute (TDRI), U.K. have recently described cold shock in aquarium reared tropical carp and tilapia. They claim that the FAO/WHO International Codes of Practice for chilling fish quickly have important commercial implications as cold shock should be avoided. Experiments with five commercial species from the North-West shelf have shown that only samples of *Diagramma pictum* of the family Haemulidae were sufficiently large (up to 3 kg) to survive trawling and to be hauled live. These fish contained sufficient adenosine triphosphate (ATP) and adenosine diphosphate (ADP) for cold shock to occur, while the smaller fish become exhausted in the trawl and ATP levels and pH were too low for cold shortening. A generalisation from aquarium specimens with high ATP levels should be treated with caution and further experiments on cold shortening and rigor in commercial tropical species are required.

Chill-storage Trials on Three Species of Scampi

H. ALLAN BREMNER

ABSTRACT

Three species of scampi *Metanephrops andamanicus, M. boschmai* and *M. australiensis* were frozen at sea in two forms, as whole animals and as scampi tails. After thawing they were stored at a temperature of 4° C for up to eight days, and evaluated by chemical microbial and sensory methods.

There were no differences in the microbial flora or the rate of nucleotide breakdown between the two forms, but the whole scampi exhibited greater discoloration and more deteriorative textural change possibly due to enzymes from the digestive gland. Although characteristic odors and flavors decreased and off odors and off flavors increased during storage, the tails remained as acceptable after 8 days storage as they were when just thawed. Whole scampi deteriorated but remained acceptable even at the end of the storage period.

Frozen Storage Trials on Three Species of Scampi.

H. ALLAN BREMNER

ABSTRACT

THREE SPECIES OF scampi, *Metanephrops andamanicus, M. boschmai* and *M. australiensis*, were frozen on board soon after catching in the whole form and as tails. These samples were evaluated after 2, 6 and 12 months frozen storage at a temperature of −18° C.

The proteins denatured only slowly as indicated by slight decreases in saline extractable protein. The K-value decreased slightly and the total nucleotide pool decreased during storage. There were only minor textural changes but there was some loss of characteristic odors and flavors and increases in off odors and off flavors during storage. The acceptability was significantly lower after 12 months storage than after 6 months storage. Scampi tails were more stable in storage then whole scampi.

The scampi remained high in acceptability and are a relatively stable seafood in frozen storage.

Shelf-life extension of packaged seafoods - a summary of a research approach +

J.A. STATHAM AND H.A. BREMNER

ABSTRACT

A theoretical step by step approach to packaging of fish has been adopted, based on the knowledge gained in meat packaging technology. The investigation includes the modification of the intrinsic properties of fish flesh, the use of vacuum and modified atmosphere packaging and the addition of lactic acid bacteria and potassium sorbate. The inherent biochemical properties of fish flesh have been considered and the insights gained will enhance commercial developments and prevent undue optimism where conditions are not optimal.

+ *The full text of this paper was Appendix F of the 1985/86 Progress Report.*

Packaging of Scallops with Sorbate: An Assessment of the Hazard from Clostridium botulinum

G.C. FLETCHER, W.G. MURRELL, J.A. STATHAM, B.J. STEWART AND H.A. BREMNER.

ABSTRACT

Toxin production in shucked scallops by 10 strains of *Clostridium botulinum* types A, B, E and F was studied using mouse bioassay. Scallops, with or without 0.1% potassium sorbate, were packaged in air-permeable film or vacuum packaged in

impermeable film. Two trials were carried out in which packages were inoculated with different spore levels and held at 4, 10 and 27° C. The appearance and odour of the packaged scallops were recorded along with bacterial counts on various selective media and the potential hazard from botulism in packaged scallops was considered.

Only type A toxin was observed and this only in evacuated packages held at 27° C. Toxin developed more rapidly in the presence of sorbate. All packages in which toxin was recorded had deteriorated beyond absolute rejection by sensory evaluation. Packs were rejected on the basis of off-odour before the appearance deteriorated.

Naturally-occurring *Clostridium perfringens* accounted for some mouse deaths not caused by *C. botulinum*.

Scallops packaged under the experimental conditions did not represent a significant botulism hazard. Qualifications to this conclusion were: 1. Initial levels of competitive flora were high. Contamination with *C. botulinum* in the presence of lower levels of competitive flora might increase the botulism hazard. 2. Scallops packaged in air-permeable film with sorbate were not held to the point of absolute sensory rejection. In this case, it is possible that toxin may have developed before the scallops were unacceptable.

Taste Testing - Examples of Design and Analysis

H. ALLAN BREMNER AND RICHARD G. JARRETT

ABSTRACT

Taste testing is an important tool for the evaluation of seafoods and it is essential that careful consideration is given to the design and analysis of tasting experiments. This paper outlines examples of approaches taken at the Seafood Technology Group (STG) and discusses the use of analysis of variance, whether taster are random or fixed effects, problems with randomisation and the confounding of duration of storage with session, substitution of samples and the construction of a two phase experiment.

Free choice flavour and odour profiling of fish spoilage: Does it achieve its objective?

A.R. QUARMBY And D.A. RATKOWSKY

ABSTRACT

Free choice odour and flavour profiling of fish spoiling under different spoilage regimes leads to a wide range of descriptors which are difficult to collate and quantify. An attempt was made to gain an insight into the spoilage process by applying a wide range of data-analytical techniques to the profile data, including cluster analysis, ordination and generalised procrustes analysis. However, the conclusion is reached that manipulation of free choice profiles does not provide any additional information about the spoilage of fish that cannot be obtained from analytical taste panels, simple chemical tests and a cursory examination of the profiles themselves.

A Dedicated Hand-Held Device for the Sensory Assessment of Fish and Fish Fillets

A.M.A. VAIL AND A.C. BRANCH

ABSTRACT

A dedicated hand held computer designed to grade and estimate the shelf-life of whole fish, gutted fish and fish fillets is described. The grading criteria are based on sensory score sheets featuring a demerit point regime that adds up obvious defects due to chemical, bacterial and enzymic changes that occur during storage of product. The user is guided through a series of prompts, and scores each attribute by entering a value using the numeric keypad contained in the unit. Calculations for remaining shelf-life are based on the ice-time principle. Ice-time was developed as a descriptor of the relative rates of psychrotrophic spoilage between 0° and 20° C. Experimental data obtained from spotty trevalla (*Seriollela punctata*), blue rock whiting (*Haletta semifasciata*) and blue grenadier (*Macruronus novaezelandiae*) gave a change of 1.71 demerit points per day with 91% of the variance accounted for. This slope factor is one of the default values programmed into the computer and is used to estimate the grade and shelf-life of fish species from temperate waters. The four tropical species *Nemipterus furcosus, Argyrops spinifer, Diagramma pictum* and *Lutjanus vittus* when stored on ice gave a slope of 0.74. This accounts for 89% of the variance and is the recommended factor for tropical fish species. The value of 1.36 demerit points per day was obtained for blue grenadier and blue rock whiting fillets. The user has the option to change the default values and customising by the manufacturer is available. Arbitrary grading into "prime condition", "good condition", "fair condition" and "poor condition" is displayed along with score and shelf-life at the completion of the test.

Demerit Point score sheets - Scampi and Prawns

MARIA OTTENSCHLAEGER

ABSTRACT

The demerit point score sheet, a principle developed for the grading of finfish, has been reconstructed for the non-destructive spoilage grading and physical condition assessment of scampi, held at 4° C, and prawns held at 4° C and in chilled sea water (CSW) over a period of time. This system gives a linear spoilage response over time, with both whole scampi and a peeled tail product, reaching their maximum possible demerit scores after 3 days and 7 days at 4° C respectively. The prawns reached their maximum possible score after 6 days at 4° C and 8 days in CSW. These asessment sheets provide an example of what can be achieved with a demerit point scoring system showing the versatility when assessing treatments or new products for visual acceptability.

A Comparison of Chemical and Organoleptic Assessment Techniques for Frozen-stored Gemfish (*Rexea Solandri*)

S.J. THROWER

ABSTRACT

The results from an experiment to examine the effect of differing post-catch handling treatments on the long term frozen storage life of gemfish (*Rexea solandri*) have been used to compare the sensitivity of sensory and chemical techniques for assessing the quality of fish. A visual and olfactory assessment of the fish which assigns demerit points as the product deteriorates, failed to distinguish between good and poor handling treatments. Three different types of taste panel were used, an expert analytical panel, a consumer acceptability panel, and a profile panel. The consumer panel gave equivocal results. The profile panel separated out those treatments which had been left on deck for 24 hours from those that were chilled immediately after capture, but failed to distinguish any other treatments.

The expert panel, which required a great deal of logistical support, showed small but statistically significant differences between treatments. Of the chemical tests used, trimethylamine analysis showed clear differences of an order of magnitude between treatments.

A cost effectiveness comparison between the assessment techniques clearly demonstrates that for inspection purposes, a simple trimethylamine analysis can give a good indication of the temperature history of fish quality. Organoleptic assessment, to give unequivocal results, needs elaborate planning and organisation which is costly in time and money.

CSIRO food researchers look at scampi

by H. A. Bremner

Allan Bremner is with the CSIRO Division of Food Research, Tasmanian Food Research Unit, Hobart.* He was part-supported by funds from the Fishing Industry Research Trust Account to be on the fisheries research vessel *Soela* on its last cruise to the scampi grounds on the north-west shelf.

THE CSIRO Division of Fisheries Research has now identified fishing grounds for three potentially commercial species of scampi — *Metanephrops andamanicus*, *M. australiensis* and *M. boschmai* (see References 1 and 2).

During the recent survey of the grounds samples were taken for evaluation of the storage characteristics of the species at the Tasmanian Food Research Unit (TFRU) of the CSIRO Division of Food Research. This article contains results of these evaluations.

Scampi is a prized seafood in Europe where it is caught in shallow waters close to shore both in pots and by trawling. In contrast the Australian scampi is caught by trawling in deep water offshore in a remote area (Refs 2 and 3) and in this respect the Australian fishery is similar to the one off South Africa.

In the European fisheries the scampi is often tailed at sea and the tails stored on ice. Storage on ice permits easier removal of the raw meat from the shell at the factory. Frozen tails (shell-on) are also received at the main factories for further processing and thawed tails are easier to peel than fresh tails.

A large proportion of the catch has been marketed as frozen tail meat but recently an expanding proportion of the UK catch is sold as whole scampi (either fresh or frozen) to continental Europe.

Because of the remoteness of the catching area of the Australian fishery it is likely that freezing at sea will be the best technique for preserving the catch. While other options, such as stowage in ice or in refrigerated sea water (RSW), are possible these options are less likely to be adopted because of the offshore distances and the lack of suitable processing facilities near the catch area.

Although the tail contains most of the edible meat it seems probable that the main marketable form of the Australian species will be as frozen whole scampi. Whole cooked animals may also be the major form in which scampi are served in restaurants. This is particularly true of the species *M. australiensis* and *M. boschmai*, which are very attractive in appearance.

Therefore it was considered important to investigate the keeping qualities of scampi in frozen storage and during chilled storage after thawing. From this information a reasonable shelf life can be deduced.

Samples of both whole animals and tails of the three potentially important commercial species were placed in the blast freezer on the CSIRO fisheries research vessel *Soela* within an hour of them landing on deck. These scampi were brought frozen to the TFRU in Hobart for chemical, microbiological and taste-panel evaluation. Also whole scampi (*M. andamanicus*) from the last haul of the cruise were stored in ice and air-freighted to Hobart.

This article will concentrate on general observations on handling the catch and the obvious patterns of deterioration which occur both in iced storage and in chilled storage of thawed scampi.

The biochemical work and identification of bacteria found on scampi is still in progress. Frozen storage trials are continuing, even though at present scampi are sold almost as soon as they are caught. Indeed the demand generated by the publicity given to scampi has resulted in increased imports of frozen scampi tails from Denmark.

Observations

It is essential that the scampi be cleaned after catching since they live in or on the bottom. (*M. andamanicus* is often covered in dark silty mud which tenaciously adheres to the grooves in the carapace.) Some form of rotary brushing may be necessary (but with care, so the relatively fragile claws and legs are not broken).

The wash water could be chilled to help cool the scampi but the important step is to ensure they are put in the freezer as soon as possible after catching.

Damaged or moulting scampi is best stored as tails. The tails can be readily removed by hand. Consideration must also be given to tailing at sea those animals that show developing gonads in the cephalothorax or 'head' region, since the blue shadow imparted by the gonad is considered to detract from the appearance.

In ovigerous females the bright blue of the eggs turns to a yellow pink colour when they are cooked. Although this is quite an attractive colour some consumers may prefer scampi without eggs, and before freezing the scampi the eggs could be removed by rotary brusher or rotary rubber fingers.

When frozen the legs and claws of scampi are particularly fragile; a

*'Stowell', Stowell Avenue, Hobart, Tasmania 7000.

special pack to prevent damage is necessary and careful handling of the frozen packs is also essential.

Iced scampi

Storage of seafoods in ice provides a good reference to their pattern of deterioration at other chill temperatures and in this case the scampi were kept in ice for a maximum of 17 days after catch. Although this time may seem extreme it was necessary to determine practical storage limits.

Seafoods deteriorate at a rate proportional to the square of the storage temperature; that is rate of deterioration = $(10 + T°C)^2$. (See Ref 4.) This means that 16 days at 0° C are equivalent to eight days at 4° C, four days at 10° C and only 1.8 days at 20° C. Note that 10° C is a relatively mild temperature in the region where scampi are caught. Therefore speedy handling and chilling is essential if significant deterioration is to be avoided.

The deteriorative changes that were noted in the iced scampi are listed in Table 1. Although they are less prone to melanosis (blackening) than many species of prawns, the scampi show this phenomenon after about three days at 0° C.

Melanosis starts as small black areas on the head, the uropods, and the abdomen particularly near where the legs join the body. As it progresses the dark black often changes to a dull bilious green reminiscent of an old bruise. In the early stages the meat is not affected and the shell can be trimmed, but after a few days on ice the flesh is also stained, particularly on the underside.

In some countries, where it is approved, a two-minute dip in 2 per cent sodium meta-bisulphite has proved effective in preventing melanosis. Treatment at this level is said to leave a residue in the flesh of below 30 parts-per-million sulphur dioxide.

With increasing time of storage the butt end of the tails softened and was stained brown due to leakage of powerful enzymes from the adjacent digestive gland. The pH of the tail flesh increased during storage (Fig 1); at 17 days after catching it was quite alkaline (pH 8.4).

Table 1. Appearance and odour of *M. andamanicus* stored in ice.

Time of storage (days)	*Appearance*	*Odour*
0	Bright colour, clear eyes	Only slight amount of fresh sea odour
4	Blackish green head, eyes opaque, dark patches where legs join body, soft inside head, soft meat on butt of tail	Fresh seaweed, fresh fish
7	Yellow green patches on carapace, black patches on uropods, very dark in gill area, digestive gland intact but soft	Fishy, seafoody, but no off odours
11	Continued darkening and yellow stains on shell, yellow fluid in head area, brown gills	Very little odour
13	Black patches over whole of carapace more noticeable in moulting animals, eyes clouded and loose on their stalks, some tails very loose, dull general appearance	Astringent, antiseptic, oyster like, sour odours in heads
17	Eggs of berried females loose and bleached in colour, legs easily shed	Antiseptic (iodine), strong sulphide, stale old smell when tailed, ammoniacal, old drains, dog faeces

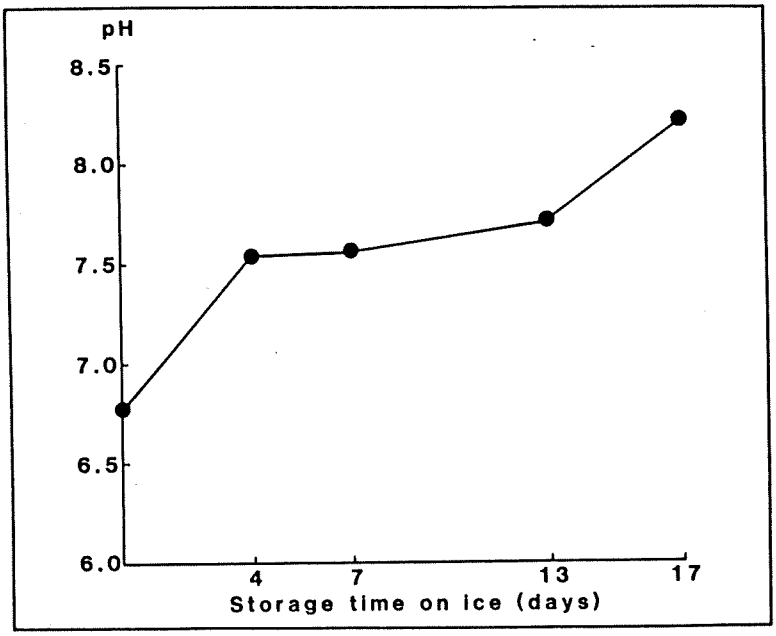

***Figure 1:** pH of tail flesh of* M. andamanicus *stored on ice.*

The scampi were sampled for bacteria on the boat and the plates incubated at about 27° C and 4° C. Further samples were taken during the storage trial at TFRU and these samples were incubated at 22° C.

No growth was observed on the plates incubated at 4° C, indicating negligible numbers of psychrotropic bacteria.

The bacteria that grew at 27° C were difficult to enumerate since they tended to spread and cover the whole plate. A rough estimate of about 3 x 10^3 colony forming units per gram was obtained.

The plates were returned to TFRU and some 60 per cent of the isolates were found to be *Bacillus* spp. and about 25 per cent were *Moraxella*

spp. The rest either have not yet been identified or were reluctant to grow in subculture media. *Bacillus* organisms are common in sea muds and these organisms probably were present on the shells of the scampi.

Similar erratic counts and spreading organisms were found on samples taken at TERU on the fourth day after catch. The counts for days seven, 13 and 17 were 1×10^6, 2.5×10^5 and 5×10^6 cfu per g respectively.

It had been the intention to identify the bacteria present at spoilage but too few organisms grew sufficiently well in subculture media for identification.

Tails removed from the iced scampi were cooked for 2.5 minutes in unsalted boiling water then served hot to a profile panel similar to that used previously to evaluate gemfish (Ref 5). This was done four, seven, 13 and 17 days after catch.

Whole scampi stored for four and 17 days in ice were frozen and, after thawing at a later date, the cooked tail meat was served cold on a bed of lettuce to an acceptability panel. Cooked tails from scampi frozen on board boat were similarly served and the three samples (zero, four and 17 days in ice) were thus tasted in the one session presented in balanced order to the panellists.

The profile results are shown in Table 2. In this panel it was mandatory for panellists to score these attributes marked by asterisks in Table 2 and so the scores shown are an average for the whole panel of 12 persons.

It was not mandatory to score the other attributes and these were selected by the panellists as free choices. The results shown in Table 2 for these attributes are arrived at by adding the scores given to a particular attribute and dividing by the number of panellists who scored that attribute.

The acceptability scores are marked on the seven-point Smiley scale (Ref 5), while the profile attributes are scored on an unstructured nine-point scale from 0 (absent) to 9 (strong).

Apart from its typical odour the *M. adamanicus* was described as having a seaweedy, shellfish, wet-straw and slight sulphide aroma at the first taste session, four days after catching.

Table 2. Odour and flavour profiles of *M. andamanicus* stored in ice.

	Days post-catch stored in ice			
	4	*7*	*13*	*17*
Odour				
* Typical odour	5.5	4.5	2.8	2.6
* Off odour	0.5	0.8	2.5	3.2
Seaweedy	2.4	3.0	3.0	3.0
Shellfish	3.6	4.4	3.2	3.3
Boiled clothes	—	3.0	3.0	3.5
Wet straw	2.0	1.8	2.8	3.0
Mousey	—	—	2.0	4.0
Grassy	—	—	—	2.5
Sulphide	1.0	—	—	4.0
Ammonia	—	1.0	2.0	2.5
Acrid	—	1.0	1.3	2.0
*† Odour acceptability	4.7	4.8	2.8	2.7
Flavour				
* Typical flavour	4.4	5.0	2.8	2.1
* Off flavour	0	0.3	1.4	4.0
Sweet	4.6	4.6	3.1	2.9
Salty	3.0	2.9	1.3	1.3
Butter	1.5	—	2.5	3.7
Carrots	3.2	2.2	3.0	2.0
Astringent	1.0	2.0	2.5	3.0
Soapy	—	—	2.5	2.5
Greasy	—	—	4	3
Creamy	3.2	2.8	2.0	2.0
Sulphide	—	—	—	4.0
Rubber	—	2.0	2.0	3.0
Blood	2.0	2.0	4.0	2.3
*† Flavour acceptability	5.6	5.3	3.7	2.7
*† Overall acceptability	5.5	5.2	4.3	3.0

* Mandatory score
† 7-point Smiley scale

During storage the typical odour decreased, off-odour increased and mousey, grassy, sulphide, ammoniacal and acrid odours were detected, and the acceptability of the odour decreased. The detection of ammonia by the panel is consistent with the increase in pH (Fig 1) and changes in the odour of the raw scampi (Table 1).

Similarly the typical flavour decreased and the off-flavour increased during storage (Table 2). The scoring for some attributes changed only slightly but those for sweet, salty and creamy decreased whereas those for rubber, sulphide, soapy, greasy and astringent either increased or made their appearance with storage.

The flavour acceptability and the overall acceptability of the scampi served hot decreased quite markedly with duration of storage. The nature of the deterioration that occurred on storage is consistent with bacterial spoilage, although the effects of autolysis must not be discounted.

It was also noted that the flesh of the scampi appeared to become softer with increasing time of storage, but this was not reflected in the scores given by the panel for textural attributes.

Softening occurred in the smaller muscles closest to the shell rather than in the main abdominal extensor muscles, and so this softer meat tends to be left in the shell when the scampi is eaten and thus does not influence the assessment of texture. Indeed the panel considered that with increasing time of storage the meat became slightly drier and springier when first bitten but was more succulent after chewing.

The flavour, texture and overall acceptability of the scampi served cold with salad were slightly lower for samples stored four days in ice than it was for those frozen on board straight after catching (Fig 2).

These tests indicated that there are few undesirable changes in the flesh up to an equivalent storage time of near seven days at 0° C but the external appearance of the scampi is poor at this stage. Although it was possible to examine only one species stored in ice, it is likely that the other species would undergo similar changes under similar conditions.

Since the initial overall acceptability scores were not a maximum (that is, seven) for scampi served either hot or cold, the impression could be gained from this and from the descriptor terms chosen that scampi is poor fare. Such is not the case, since at a Federal Parliamentary luncheon it was highly praised, and who would argue with that opinion?

Plain boiled seafoods served hot or cold without all the trimmings are never very exciting, particularly if they have been stored for some time before cooking.

Thawed scampi

Scampi tails and whole scampi of all three species (frozen on board) were thawed and held at 4°C for zero, four and eight days.

Similar changes in appearance to those found in the iced scampi were evident (Fig 3). Dark areas were noticeable several hours after thawing. Storage of whole thawed scampi allowed breakdown of the digestive gland, with consequent staining and softening of the butt end of the tail flesh. Melanosis was noted on the tails, but it was not as prevalent as on the whole animals.

After four days at 4° C the staining was only slight, the appearance dull, the flesh was firm and there was only a slight trace of staleness in the odour.

On the other hand, whole scampi thawed and held at 4°C for four days had patches of brown-black-green discoloration on their heads, their eyes were grey dull and loose, the uropods were stained black, and the carapace was stained. The odour was slightly sour and faecal, the flesh was stained brown at the butt end and had black areas at the edges of the body segments. The developing roes in the body of the scampi often stained the flesh blue but this was not so noticeable once the flesh was cooked.

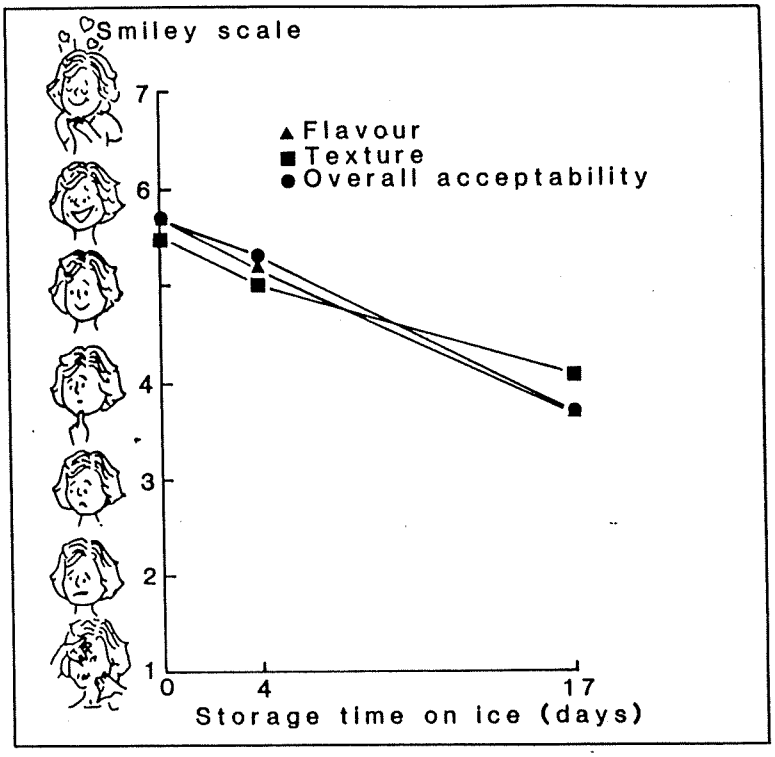

Figure 2: *Acceptability of* M. andamanicus *frozen on board or stored on ice for four and 17 days then frozen. All three samples were tasted in the one session.*

It is recommended that tails be removed if there is to be any significant delay between thawing and usage.

Moulting and non-moulting males and berried and non-berried females of *M. andamanicus* from the one haul were also evaluated by the taste panel. There were negligible differences between them, and the apparently softer flesh of the moulting scampi when raw was rated as having similar textural properties to the non-moulting scampi when it was cooked.

Conclusions

This work was designed to describe the pattern of deteriorative changes that occurs with chill stored fresh and thawed scampi and so the bulk of the results pertain to deteriorating scampi. This tends to take emphasis away from the desirable properties of scampi and its wide appeal.

The results indicate that scampi flesh is fairly robust in chilled storage but that the appearance of chilled or thawed scampi can rapidly deteriorate after two to three days at 0° C or equivalent.

If thawed scampi needs to be kept for any length of time it is preferable it be kept as tails. Careful handling is required at all stages to keep intact all the legs and claws.

References

1. Anon. (1983): 'CSIRO finds scampi and carid prawns on N.W. shelf', *Aust. Fish.* **42** (3) 13.
2. Davis, T. L. O. and Ward, T. J. (1984): 'CSIRO identifies two new scampi grounds off the North West Shelf, *Aust. Fish.* 43 (8): 41-45
3. Carter, D., Maxwell, J. G. H. and Bowtell, C. (1983): 'Cautious optimism over potential for scampi fishery on NW shelf', *Aust. Fish.* **42** (11): 2-12
4. Bremner, H. A. (1984): 'Quality an attitude of mind', in *The Australian Fishing Industry — today and tomorrow*, 10-12 July, Launceston, Australian Maritime College. Seminar Papers pp. 244-269.
5. Quarmby, A. R., Bremner, H. A. and Thrower, S. J. (1982): 'On-board handling of gemfish Part II: Sensory profiles', *Aust. Fish.* **41** (11): 42-45.

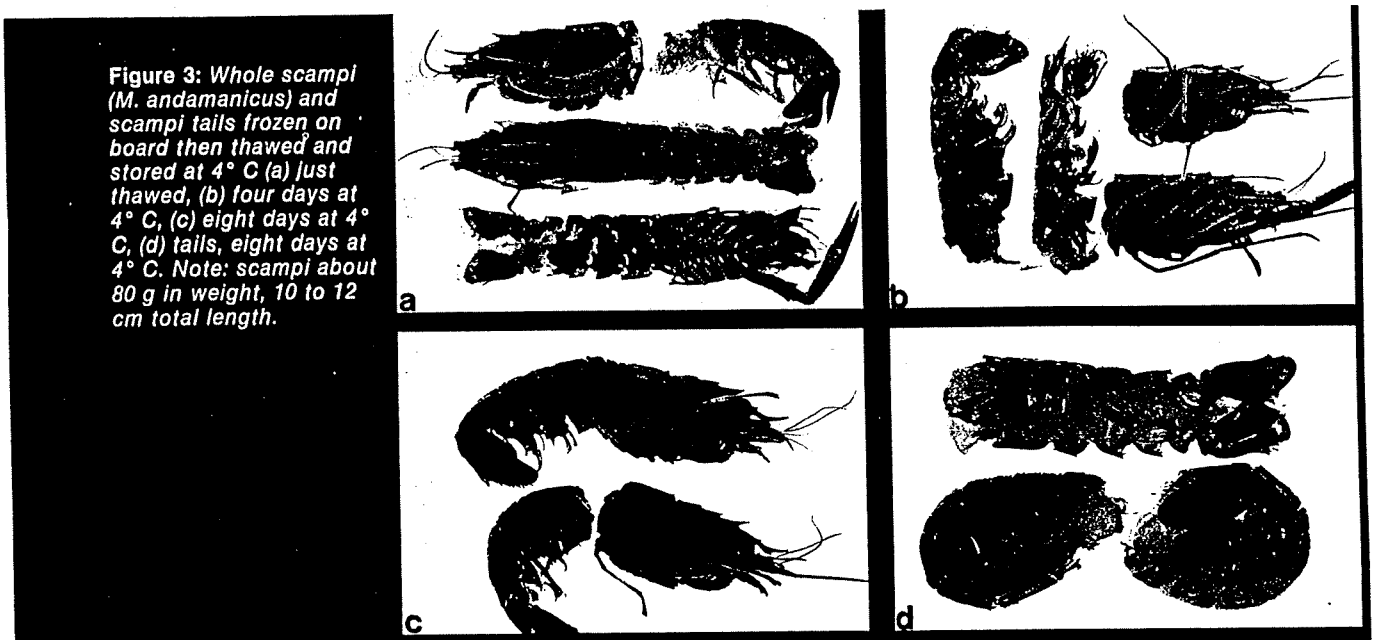

Figure 3: *Whole scampi (M. andamanicus) and scampi tails frozen on board then thawed and stored at 4° C (a) just thawed, (b) four days at 4° C, (c) eight days at 4° C, (d) tails, eight days at 4° C. Note: scampi about 80 g in weight, 10 to 12 cm total length.*

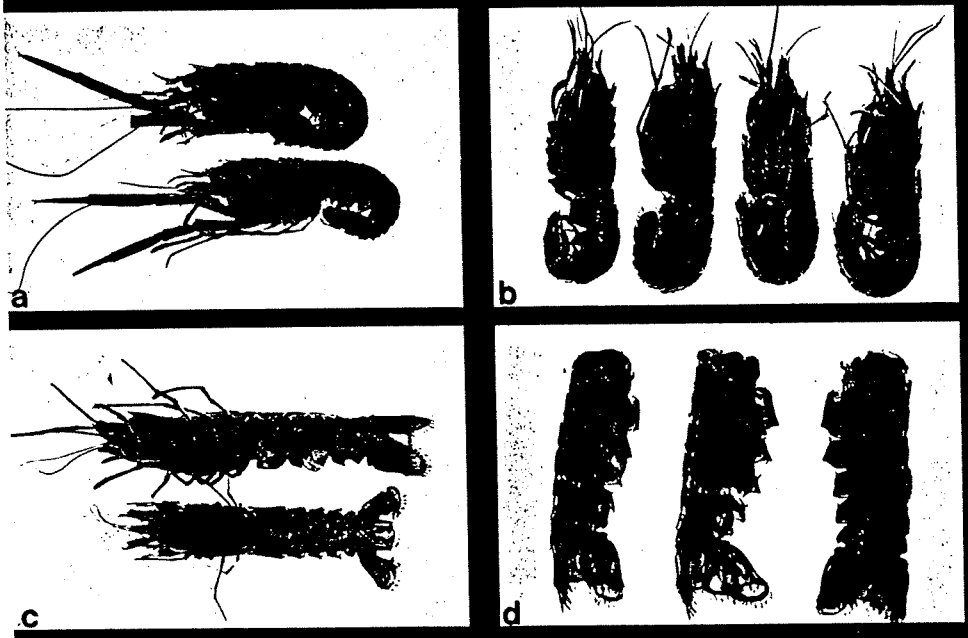

Figure 4: *Whole scamp (M. australiensis) and scamp tails frozen on board then thawed and stored at 4° C (a) just thawed, (b) four days at 4° C, (c) eight days at 4° C, (d) tails, eight days at 4° C.*

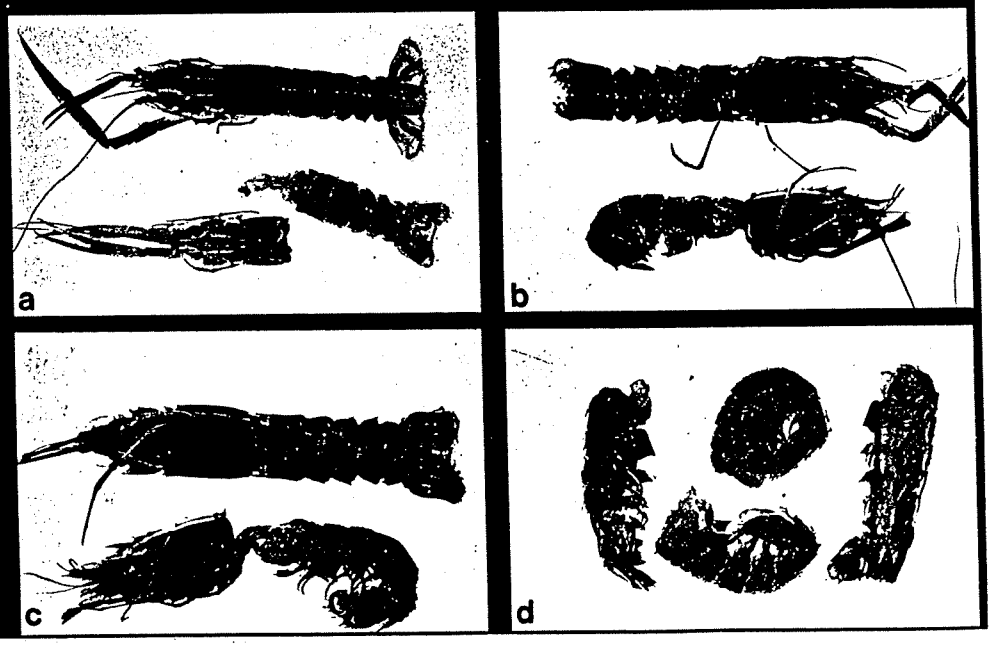

Figure 5: *Whole scampi (M. boschmail) and scampi tails frozen on board then thawed and stored at 4° C (a) just thawed, (b) four days at 4° C, (c) eight days at 4° C, (d) days, eight days at 4° C.*

Modified atmosphere storage of fisheries products: the state of the art

J.A. STATHAM

The search for the maximisation of the shelf-life of chill-stored fish and other seafoods has regenerated interest in the preservative action of storage in carbon dioxide and modified atmospheres. This paper reviews the potential of such systems in terms of their physical, chemical and antimicrobial effects and their role in the transportation and distribution of chilled-stored fish.

Packing of products in a modified gas atmosphere involves the replacement of air by other gases, usually CO_2, N_2 and O_2, alone or in combination. The aim is to inhibit physical, chemical and microbiological changes that lead to product deterioration; the composition of the modified atmosphere depends on the type of product being packed. In order to maintain the correct atmospheric composition around the product, packing materials with low gas permeabilities must be used. The effects of headspace volumes and gas to product ratios on shelf-life extension have not yet been satisfactorily investigated.

Nitrogen has a negligible effect on bacterial growth and on the shelf-life of flesh foods (Coyne 1932, Enfors, Molin & Ternstrom 1979, Fey 1980, Savell *et al.* 1981, Tiffney & Mills 1982b); it serves as an inert filler to balance a gas mix. High levels of oxygen (75%) have been successfully used with CO_2 to extend the shelf life of beef (Savell *et al.* 1981). Using white fish, Tiffney and Mills (1982b) showed some extension of quality as judged by organoleptic assessment, but the storage life in 100% O_2 did not exceed that of similar fish in vacuum or overwrap packs. Bacterial growth rates were increased and the storage life of lamb was considerably shorter in 80% O_2 than in low-O_2 or O_2-free atmospheres (Newton, Harrison & Smith 1977). In general, the proportion of O_2 used in modified atmosphere storage (MAS) systems is either equal to or lower than that naturally present in the atmosphere. Low levels of carbon monoxide, not an approved food additive (Finne 1982), have also been included in modified atmospheres to prevent discoloration of flesh caused by the production of metmyoglobin. The carboxymyoglobin formed by association of CO with myoglobin is spectrally similar to the red pigment oxymyoglobin, but is more stable and therefore maintains a bright flesh colour. High levels of CO (10–30%) have inhibitory effects on the growth of some bacteria (Gee & Brown 1981); they would therefore have a selective effect on mixed cultures. However, the level proposed for use in packaging flesh foods (1%) appears to have negligible effects on bacterial growth (Brown *et al.* 1980)

Sulphur dioxide, ethylene oxide and ozone have antimicrobial properties (Clark & Takacs 1980, Silva 1982) but are not commonly included in MAS systems for various reasons, such as stability of the gas, limited approval for use in foods and the formation of toxic residues. Carbon dioxide has been found to be the most effective gas for preserving fresh foods and most discussions of modified atmospheres refer to those with elevated levels of this gas.

Jo Statham is an experimental officer with CSIRO Division of Food Research, Tasmanian Food Research Unit, "Stowell", Stowell Ave, Hobart, Tas. 7000.

An address to the 4th Australian Food Microbiology Conference, *Sydney 26–27 October 1983.*

Mechanism of bacterial inhibition by CO_2

Although the preservative action of CO_2 in foodstuffs has been utilised for many years (Kolbe 1882, Killefer 1930) its mechanism of antibacterial action has not been fully elucidated. Valley and Rettger (1927) suggested that CO_2 acts by lowering substrate pH; however, Coyne (1932) and Haines (1933) demonstrated that pH alone does not account for the inhibitory effect of CO_2. Two main hypotheses have recently been put forward (Parkin & Brown 1982). The first considers the inhibitory effect of CO_2 to be on enzyme systems necessary for growth. King and Nagel (1975) showed that a 50% CO_2 atmosphere had a mass action effect on certain enzymatic decarboxylation reactions. *Pseudomonas aeruginosa* was found to have decreased isocitrate dehydrogenase and malate dehydrogenase activity. Kritzman, Chet and Henis (1977), working with *Sclerotinium rolfsii*, found CO_2 to inhibit the tricarboxylic acid cycle, thus affecting types and amounts of carbohydrate intermediates.

The second theory suggests that CO_2 acts on the cell membrane, altering contact between the cell and its external aqueous environment by redistribution of lipids at the interface. This phenomenon has been demonstrated using a model system (Sears & Eisenberg 1961) and has been proposed as the mechanism by which CO_2 inhibits bacterial spore germination (Enfors & Molin 1978, cited in Enfors & Molin 1980).

Regardless of mechanism, the overall effect on aerobic bacterial growth is a linear increase in lag phase and decrease in growth rate with increasing CO_2 levels (King & Nagel 1967, Sutherland *et al.* 1977, Gill & Tan 1979, Gee & Brown 1981). Enfors and Molin (1980) showed that for *Ps. fragi* the effects of the presence of increased CO_2 and limited O_2 were purely additive; no synergism between CO_2 inhibition and O_2 limitation occurred.

Chemistry of CO_2 in flesh foods

The solubility of CO_2 in water (1.71 mL CO_2/mL H_2O at 760 mm Hg and 0°C, Clark & Takacs 1980) is inversely related to temperature. The relationship is not linear and the change in CO_2 solubility per degree change in temperature increases as the temperature is lowered (Ogrydziak & Brown 1982). Only a small fraction of the CO_2 dissolved in water is hydrated to carbonic acid (Daniels & Alberty 1975) which dissociates in two stages, the first yielding hydrogen and bicarbonate ions, the second yielding another hydrogen ion and a carbonate ion. The pK values for the two dissociations are 6.37 and 10.25 respectively. Hence, under normal physiological conditions the concentration of carbonate is negligible, the first dissociation being of greater significance. Weak acids are known to have antimicrobial activity in their undissociated form; therefore, carbonic acid is unique as a microbial inhibitor since at pH

0015-6647/84/36/0233/07e 1984
The Council of Australian Food Technology Associations, Inc

values near neutrality at least one-half of the acid is in the undissociated form (Silliker 1982). In contrast, sorbic, acetic and formic acids have pKs of 4.8, 4.76 and 3.75 respectively and therefore require low pH for maximal antimicrobial activity.

Chemical and physical effects of CO_2 on fish muscle

pH

In general the dissociation of carbonic acid in fish flesh results in a slight drop in pH. Buffering capacity of the material (Cutting 1953) and the composition of the spoilage flora determine the magnitude of pH change. Barnett *et al.* (1982), for example, found no significant change in the pH of salmon flesh stored in 90% CO_2. The extent to which pH decreases is proportional to the concentration of CO_2 in the atmosphere (Lannelongue *et al.* 1982a, Tiffney & Mills 1982b). CO_2 is absorbed rapidly and pH drops over the first two days. Parkin, Wells and Brown (1981) noted in rockfish muscle a drop from initial pH 6.7 to pH 6.3 which was maintained throughout the storage period, while Fey and Regenstein (1982) found that after an initial decrease flesh pH rose and after 27 days had reached a level similar to its starting point.

Colour

One of the major problems with using elevated levels of CO_2 for storage of red meats is that discoloration occurs, partly due to the formation of metmyoglobin. Fish muscle contains lower levels of myoglobin and discoloration is a less significant problem. Whole fish may suffer due to greying of the cornea, bleaching of the skin and damage to bloom at high (>60%) CO_2 concentrations (Coyne 1933, Stansby & Griffiths 1935, Anon. 1950). Goodfellow (1982) found it necessary to restrict the CO_2 level to 25% due to discoloration (species of fish not stated). Parkin *et al.* (1981) observed no difference between rock fish fillets stored in 80% CO_2 and fresh samples, while those stored in air became significantly darker. Undesirable browning of salmon was reduced by the addition of an antioxidant dip and reduction of O_2 levels to 5% (Fey 1980).

Drip loss

A further problem caused by elevated CO_2 levels is the development of excessive amounts of drip during storage. This may be caused in part by a lowering of pH and a consequent reduction in water binding capacity of the flesh (Tarr 1962). Fey and Regenstein (1982) found increased drip losses from red hake, chinook salmon and to a lesser extent, sockeye salmon, stored in 60% CO_2, 21% O_2, 19% N_2. Tiffney and Mills (1982b) found that 100% CO_2 increased the rate and quantity of drip produced and found it necessary to decrease the level of CO_2 to below 60% to prevent the level of drip production becoming unacceptable before adverse organoleptic changes occurred in stored codling. The quantity of drip produced was higher at 0°C than 5°C; they concluded that the problems could be overcome by:

- using an atmosphere of 40% CO_2, 30% O_2, 30% N_2
- inclusion of polyphosphates which delay drip production but are needed in high concentrations
- placing absorbent pads in packs.

Organoleptic changes

Organoleptic changes may also be induced by storage of fish in high CO_2 atmospheres. These include peculiar acidic-type odours noticed when the packs are first opened but which dissipate with time (Stier *et al.* 1981, Tomlins *et al.* 1982). Textural changes such as powdery (Haard & Lee 1982), dry, tough (Tiffney & Mills 1982b) and grainy flesh (Wang & Brown 1983) have been reported. Flavour may also be affected. Jensen *et al.* (1980) noted an acidic taste in cod packed in high CO_2 or vacuum, while Haard and Lee (1982) found salmon steaks to have a carbonated, bland taste after storage in 100% CO_2.

Pack collapse

As previously discussed, CO_2 is highly soluble in water and dissolves readily in fish tissues. This solubility can lead to collapse of rigid retail packs if a high CO_2 level has been used (Jensen *et al.* 1980, Mills & Tiffney 1982). The problem can be eliminated by reducing the proportion of CO_2 in the atmosphere (Mills & Tiffney 1982). When using flexible barrier film the collapse of the pack is acceptable as it then looks like a vacuum pack.

Bacterial growth on fish stored in modified atmospheres

Although the mechanism by which CO_2 affects bacterial growth has not been fully elucidated, the overall effect of modified atmosphere storage is suppression of bacterial growth and a subsequent extension of shelf-life. Lag phases lasting for 6-10 days have been induced in brown shrimp (Lannelongue *et al.* 1982a), swordfish (Lannelongue *et al.* 1982b), finfish (Lannelongue *et al.* 1982c), freshwater crayfish (Wang & Brown 1983) and perch, sea trout, croaker and blue fish (Gray, Hoover & Muir 1983). In other cases no growth occurred during the full extent of the storage period: 14 days (Parkin *et al.* 1981), 26 days (Fey 1980), and 21 days (Barnett *et al.* 1982). In addition to extending lag phase and suppressing subsequent growth of spoilage organisms, high levels of CO_2 have bactericidal effects on the initial flora (Jensen *et al.* 1980, Parkin *et al.* 1981, Lannelongue *et al.* 1982a). Microbial growth in CO_2-enriched atmospheres is inversely related to the CO_2 tension in the atmosphere.

As a result of bacterial growth being suppressed, levels of chemical spoilage indicators, such as trimethylamine (TMA) and total volatile nitrogen (TVN) are generally reduced in fresh fish by atmospheres containing elevated levels of CO_2 (Jensen *et al.* 1980, Banks, Nickelson & Finne 1980, Haard & Lee 1982, Lannelongue *et al.* 1982a,b, Wang & Brown 1983). Jensen *et al.* (1980) found suppression of TMA production in CO_2 atmospheres during early storage but by 12 days the level equalled that found in cod stored in air. At CO_2 levels above 20% Lannelongue *et al.* (1982c) found TVN production inversely proportional to CO_2 concentration.

Banks *et al.* (1980) suggested that TVN levels are lowered by the effect of suppressing bacterial growth and by the reduction of O_2 concentration, since the reactions involved are mainly those of oxidative deamination. Lannelogue *et al.* (1982b) suggested that CO_2 selectively inhibits TVN-producing bacteria and that TVN should not be used as a spoilage indicator of fish products stored in modified atmospheres. Gorczyca, Cohen and Sumner (1983) showed that hypoxanthine production from inosine monophosphate is also restricted, due to a reduction in the growth rate of *Alteromonas putrefaciens,* but they did not eliminate the possibility that CO_2 may have inhibited dephosphorylating enzymes in the fish flesh.

Selective inhibition by CO_2

Increased CO_2 in atmospheres used for fish storage not only limits overall microbial growth but also changes the distribution of bacterial types. Susceptibility to inhibitory action of CO_2 varies between bacterial genera. Coyne (1933) showed *Achromobacter, Flavobacter, Micrococcus, Bacillus* and *Pseudomonas* to be susceptible to CO_2 while the growth of *Proteus* was unaffected. Enfors and Molin (1980) noted increasing resistance to the effects of CO_2 through the order *Ps. fragi, B. cereus* and *Streptococcus cremoris.*

Freshly caught fish normally supports a predominantly Gram negative flora composed of *Pseudomonas, Moraxella, Acinetobacter* and *Flavobacteria/Cytophaga* species; however, some tropical fish and shellfish may support significant populations of coryneforms and micrococci. *Pseudomonas* and *Alteromonas* are generally selected by reduced storage temperatures; but employment of elevated CO_2 levels as a further selective agent shifts the Gram negative flora to a predominantly Gram positive one (Ogrydziak & Brown 1982).

The flora of CO_2-stored fish and meat is generally dominated by lactic acid bacteria (Banks *et al.* 1980, Silliker & Wolfe 1980, Finne 1982, Lannelongue *et al.* 1982a,b, Mokhele *et al.* 1983, Oberlender *et al.* 1983). This shift may be regarded as beneficial since lactic acid bacteria are known to be inhibitory to other bacterial genera and are not such active spoilers. *Brochothrix thermosphacta* has also been isolated from fish stored in CO_2 (Lannelongue *et al.* 1982b, Oberlander *et al.* 1983.)

L s *et al.* (1980) postulated that outgrowth of lactic acid bacteria may result from a combination of anaerobic conditions and increased acidity. Lannelongue *et al.* (1982b) found the shift from Gram negative to Gram positive occurred rapidly in 100% CO_2 and in CO_2/N_2 mixtures. In atmospheres containing CO_2/O_2 the rate of change in population distribution was related to O_2 tension.

There have been some reports of Gram negative bacteria comprising a major proportion of the spoilage flora. Cann, Smith & Houston (1983) found *Pseudomonas* and *Alteromonas* predominating in the final flora of cod and herring and *Moraxella* in smoked mackerel. Lee (1982) found *Lactobacillus* species and *Alteromonas* species were the predominant types in CO_2-treated Dover sole. The presence of *Alteromonas* species was unexpected, but using artificial media he showed that *Alteromonas* species were capable of growth equal to or better than that of *Lactobacillus* in 20% and 60% CO_2. The growth of *Pseudomonas, Moraxella, Flavobacterium/Cytophaga* and *Arthrobacter* was significantly slower in CO_2 enriched atmospheres.

Contrary to the above findings, Jensen *et al.* (1980) found that trimethylamine oxide-reducing *Pseudomonas* species predominated on cod stored in both air and CO_2 enriched atmospheres. They attributed this to the inhibition of other bacteria by CO_2 and the presence of trimethylamine oxide allowing growth of *Pseudomonas.* They did not speculate on how *Pseudomonas* overcame the inhibitory effect of 100% CO_2. Valid comparisons between the undefined systems are impossible, because pH was not determined in either of the latter two experiments.

Extension of shelf-life resulting from MAS

Shelf-life is a difficult term to define since it may be assigned based on criteria derived from chemical, microbiological or organoleptic assessment employed singly or in combination. Regenstein and Regenstein (1981) suggested that the ultimate measure of shelf-life is consumer satisfaction and consider this more significant than defining the endpoint as when arbitrarily defined levels of spoilage compounds or bacterial numbers are reached.

The definition of the time at which shelf-life begins is also uncertain. It may be taken as the time of catch or death or the time at which processing is completed. These inconsistencies make comparisons between sets of data of different origin difficult.

Table 1 shows the shelf-life obtained in studies of MAS of various types of fish. Carbon dioxide–enriched atmospheres result in an extension of shelf-life of between 50% and 100%. The importance of good temperature control is stressed by the work of Mills and Tiffney (1982) and Cann *et al.* (1983). The condition of the fish before packing is also of great importance and it is generally understood that the lower the bacterial count the better. The use of MAS will not disguise poor initial handling of a product. Some workers, however, have questioned the value of CO_2 storage of fish for the first few days after catching. Stansby and Griffiths (1935) suggest that CO_2 storage is of little value during the first few days after fish are caught as bacterial growth is restricted until rigor mortis is resolved. The "delay-pack" hypothesis of Regenstein and Regenstein (1981) suggests that during the initial lag phase additives will destroy only some of the bacteria. The remainder being tolerant of the selective agent will continue to grow. If the product is packed while the bacteria are in the log phase, substances antagonistic to other bacteria may have been produced, for example by lactic acid bacteria, which would reduce the number of spoilage bacteria able to grow at this stage.

Residual effect of CO_2 storage

Clark and West (cited by Silliker 1982) showed that CO_2 must be present continuously to prevent microbial growth. But there are conflicting reports on the continued inhibition that may occur when muscle tissue is removed from a modified atmosphere. Silliker *et al.* (1977) call this phenomenon a post-treatment or residual effect resulting in a reduced rate of microbial growth subsequent to MAS. This effect becomes important when considering the use of MAS for bulk storage of fish which would be removed from the modified atmosphere before repacking and retail sale. Post-treatment effects have been demonstrated with pork (Spahl, Reineccius & Tatini 1981) and beef (Silliker *et al.* 1977).

The mechanism by which bacterial inhibition extends into post-treatment is not well understood. Silliker *et al.* (1977) suggest the reduced bacterial growth may result from a lag phase induced by CO_2 treatment, while Mitsuda, Kawai & Yamamoto (1975) speculate that CO_2 may be bound to amino acid residues and also dissolve in sarcoplasmic fluid. The release of CO_2 after removal from the modified atmosphere may then result in the observed post-treatment effect.

Residual effects of CO_2 treatment are not observed consistently. Banks *et al.* (1980) reported a rapid increase in the growth rates of Gram negative bacteria following removal of fish from 100% CO_2, and a concomitant decrease in numbers of Gram positive types. Similar results have also been reported by Goodfellow (1982). Veal, beef, pork and poultry removed from a CO_2 atmosphere spoiled 1–2 days faster than similar material that had been vacuum packaged.

Table 1. Shelf-life obtained by using modified atmosphere packing

Author	Atmosphere	Species	Storage temperature (°C)	Shelf-life (days)	Shelf-life extension (%)
Coyne (1933)	100% CO_2	Cod, whiting, haddock	0	10–12	50–100
Stansby & Griffiths (1935)	25% CO_2	Haddock	0	25	100
Fey (1980)	60% CO_2:21% O_2:19% N_2	Red hake	0–1	>27	–
	60% CO_2:5% O_2:35% N_2	Red hake	0–1	>27	–
	20% CO_2:21% O_2:59% N_2	Red hake	0–1	>27	–
Lee (1982)	20% CO_2:20% O_2:60% N_2	Dover sole	0		100
	60% CO_2:20% O_2:20% N_2	Dover sole	0		200
Stier *et al.* (1981)	100% CO_2	King salmon	4.4	12	100
	100% CO_2	King salmon	22	<2	<100
Barnett *et. al.* (1982)	90% CO_2:10% air	Chum and Coho salmon	0	>21	–
Haard and Lee (1982)	100% CO_2	Salmon	3	>20	>100
Mills & Tiffney (1982)	40% CO_2:30% N_2:30% O_2	Various white fish	0	15	70
			2	8	60
			5	6	50
Cann, Smith & Houston (1983)	40% CO_2:30% N_2:30% O_2	Cod	0	8.5–12.5*	40–80*
Wang & Brown (1983)	80% CO_2: 20% air	Freshwater crayfish	4	>21	>50

* *Depending on quality criterion used*

Effect of MAS on growth of pathogens

MAS effectively increases storage life and maintains quality of a number of products, but the question of microbiological safety must also be considered. The effect of CO_2 on the growth of various pathogens at both chill and abuse temperatures has been widely studied. Modified atmosphere storage does not appear to increase the hazards from *Salmonella, Staphylococcus aureus, Clostridium perfringens, Yersinia, Campylobacter, Vibrio parahaemolyticus* and *Enterococcus* above those expected for air stored products (Silliker & Wolfe 1980, Goodfellow 1982, Lee 1982, Silliker 1982). Silliker (1982) showed that CO_2 actually caused some reduction in numbers of staphylococci at temperatures up to 20°C. Contrary to these findings Tomlins *et al.* (1982) showed that levels of CO_2 from vacuum up to 60% stimulated the growth of *S. enteritidis* on an artificial medium above that observed in air at 10°C. They concluded that high CO_2 levels or the addition of potassium sorbate would be needed to reduce the risk of *Salmonella* multiplication.

The pathogen of most concern is the anaerobe *Cl. botulinum* type E which is capable of growth at temperatures as low as 3.3°C (Schmidt, Lechowich & Folinazzo 1961) along with non-proteolytic types B and F. As with the other pathogens listed above, the presence of CO_2 is not considered to increase the probability of growth of *Cl. botulinum* in packaged fish (Silliker 1982), even though CO_2 promotes spore germination and growth of some clostridia in artificial media (Eyles & Warth 1981). The main concern is that CO_2 may inhibit the normal spoilage flora sufficiently to allow toxin formation during the extended shelf-life of the product.

The results of a number of studies undertaken to assess the botulism risk of MAS fisheries products are shown in Table 2.

The effect of temperature on the formation of toxin before spoilage can be seen clearly, demonstrating the increased hazard brought about by temperature abuse of an MAS product. It is for this reason that the development of MAS is proceeding with caution and the process has not been widely put into practice.

Direct comparisons between various reports cannot be made due to different inoculum levels, sites of inoculation and gas atmospheres. Eklund (1982) demonstrated the effect of inoculum size on the rate of toxin production (Table 3). During the first seven days of storage, 60% CO_2 and 90% CO_2 had a slight inhibitory effect on growth and toxin production at lower inoculum levels. A similar effect was noted at 25°C, but no inhibition occurred at inoculum levels greater than 10^2 spores/100 g. Lindsay (1982a) also demonstrated that higher spore

Table 3. Salmon stored in modified atmospheres for 7 days: relationship of spoilage to toxin production by type E *Clostridium botulinum* at various levels of inoculation in salmon stored at 10°C in modified atmospheres for 7 days

		No. of toxic samples/total no. of samples tested		
Atmosphere	Spoilage*	10^2 spores/100 g sample	10^3 spores/100 g sample	10^4 spores/100 g sample
Air	+	1/3	3/3	3/3
60% CO_2	-	0/3	3/3	3/3
90% CO_2	-	0/3	0/3	3/3

After Eklund (1982)
** + = spoiled, - = acceptable*

Table 2. Growth and toxin production by *Clostridium botulinum* in MAS fish products

Author	Species	Storage temperature (°C)	Atmosphere	Inoculum	Toxicity (storage time)	Spoiled before being toxic
Anon. (1979)	Salmon	25	Stored in ?% CO_2† for 17 days then inoculated and stored at 25°C or 10°C	Types A & E — method of inoculation not specified		+
		10	As above	As above		+
		25	?% CO_2†	Type E	+ (24 h)	-
		25	?% CO_2†	Type A	+ (48 h)	-
		10	?% CO_2†	Type E	+ (14 d)	+
		10	Air	Type E	+ (7 d)	+
		10	CO_2 or air	Type A		+
		5	CO_2 or air	Type A		+
Huss *et al.* (1980)	Smoked herring	15	99.5% CO_2	Type E 10^3 spores/g, surface inoculum	+ (4 d)	*
		15	46.1% CO_2:53.8% N_2	As above	+ (4 d)	
		15	23% CO_2:77% N_2	As above	+ (4 d)	
		15	99% N_2	As above	+ (4 d)	
		15	Vacuum	As above	+ (4 d)	
		15	48% CO_2:52% O_2	As above	+ (10 d)	
		15	99% O_2	As above	+ (9 d)	
		15	Air	As above	+ (6 d)	
Lindsay (1982a)	Rock-fish	27	Unsealed vacuum, partial CO_2 and 100% CO_2	Mixed type E strains 5 & 50 spores/g	+ (24 h)	*
		21	As above	Site of inoculum not specified	+	*
		7.2	As above	As above	- (29 d)	
		4.4	As above	As above	- (29 d)	+
		1.7	As above	As above	- (29 d)	+
Stier *et al.* (1981)	Salmon	22.2	60% CO_2:25% O_2:15% N_2	Types A, B and E spores — type E cells 10^4/g, surface inoculum	+ (2–3 d)	+
		4.4	As above	As above	- (28 d)	+
		22.2	Air	As above	+ (2–3 d)	+
		4.4	Air	As above	- (28 d)	+

Continued

Table 2 — continued

Author	Species	Storage temperature (°C)	Atmosphere	Inoculum	Toxicity (storage time)	Spoiled before being toxic
Eklund (1982)	Salmon	25	60% CO_2:25% O_2: 15% N_2	Type E, 10^2 spores/ 100 g — site of inoculum not specified	+ (48 h)	-
		25	90% CO_2:10% air	As above	+ (48 h)	-
		25	Air	As above	+	+
		10	60% CO_2	Type E 10^2 spores/ 100 g	+ (10 d)	-
		10	90% CO_2	As above	+ (10 d)	-
		10	Air	As above	+ (10 d)	+
		5	60% CO_2	Type E 10^2 spores/ 100 g	+ (21 d)	*
		5	90% CO_2	As above	+ (21 d)	*
Cann, Smith & Houston (1983)	Cod	10	40% CO_2:30% O_2:30% N_2	Types B & E 10^2 spores/g deep muscle	11 d	+
		10	Vacuum	Surface	10 d	+
		10	Vacuum	Site of inoculum not specified	8 d	+
		20	40% CO_2:30% O_2:30% N_2	Deep muscle	2 d	*
		20	40% CO_2:30% O_2:30% N_2	Surface	2 d	*
		20	Vacuum	Site of inoculum not specified	2 d	*
	Herring	10	40% CO_2:30% O_2:30% N_2	Deep muscle	7 d	+
		10	40% CO_2:30% O_2:30% N_2	Surface	9 d	+
		10	Vacuum	Site of inoculum not specified	7 d	+
		10	60% CO_2:40% N_2	Deep muscle	6 d	+
		10	60% CO_2:40% N_2	Surface	8 d	+
		10	Vacuum	Site of inoculum not specified	6 d	+
		15	40% CO_2:30% O_2:30% N_2	Deep muscle	2 d	*
		15	40% CO_2:30% O_2:30% N_2	Surface	2 d	*
		15	Vacuum	Site of inoculum not specified	1 d	*
		15	60% CO_2:40% N_2	Deep muscle	2 d	*
		15	60% CO_2:40% N_2	Surface	2 d	*
		15	Vacuum	Site of inoculum not specified	1 d	*
		20	40% CO_2:30% O_2:30% N_2	Deep muscle	1 d	*
		20	40% CO_2:30% O_2:30% N_2	Surface	1 d	*
		20	Vacuum	Site of inoculum not specified	1 d	*
		20	60% CO_2:40% N_2	Deep muscle	2 d	*
		20	60% CO_2:40% N_2	Surface	2 d	*
		20	Vacuum	Site of inoculum not specified	1 d	*
	Smoked mackerel	10	40% CO_2:30% O_2:30% N_2	Deep muscle	-	+
		10	40% CO_2:30% O_2:30% N_2	Surface	-	+
		10	Vacuum	Site of inoculum not specified	-	+
		10	60% CO_2:40% N_2	Deep muscle	-	+
		10	60% CO_2:40% N_2	Surface	-	+
		10	Vacuum	Site of inoculum not specified	12 d	-
		15	40% CO_2:30% O_2:30% N_2	Deep muscle	5 d	*
		15	40% CO_2:30% O_2:30% N_2	Surface	-	*
		15	Vacuum	Site of inoculum not specified	6 d	*
		15	60% CO_2:40% N_2	Deep muscle	6 d	*
		15	60% CO_2:40% N_2	Surface	-	+
		15	Vacuum	Site of inoculum not specified	4 d	*
		20	40% CO_2:30% O_2:30% N_2	Deep muscle	3 d	*
		20	40% CO_2:30% O_2:30% N_2	Surface	4 d	*
		20	Vacuum	Site of inoculum not specified	3 d	*
		20	60% CO_2:40% N_2	Deep muscle	2 d	*
		20	60% CO_2:40% N_2	Surface	3 d	*
		20	Vacuum	Site of inoculum not specified	2 d	*

† *Proportion of CO_2 not stated* - *Acceptable*
+ *Spoiled* * *Not stated*

loads cause toxigenesis more rapidly than low spore loads, but concluded that a definite risk is associated with low inoculum levels if elevated temperatures occur. As with general spoilage of flesh foods, the relationship between time, temperature and inoculum levels (Pooni & Mead 1984) are the key factors controlling the safety of these products. The site of inoculation, surface or intramuscular, might also influence toxin production. Stier *et al.* (1981) showed that stacking fillets with inoculum between them did not create conditions more conducive to growth and toxin production. However, Cann *et al.* (1983) showed that fish inoculated intramuscularly became toxic before fish which had been surface inoculated.

The actual composition of the gas mix is much less important than temperature when considering the botulism risk in MAS products (Cann *et al.* 1983). Toxin may be produced in atmospheres containing nearly 100% oxygen (Huss *et al.* 1980), yet oxygen is sometimes included in modified atmosphere gas mixes in the belief that aerobic conditions will be maintained and toxin production inhibited. Lindsay (1982b) suggests that the presence of O_2 may offer a false sense of security. Surface conditions may become sufficiently anaerobic and have sufficiently low oxidation-reduction potentials brought about by respiration of tissues and aerobic bacteria to allow spore germination and outgrowth (Goodfellow 1982, Silliker 1982, Sperber 1982).

Applications for MAS

There are three methods by which modified atmosphere packing may be employed for transportation and distribution (Bell 1982). The first method involves bulk transportation in refrigerated seavans, railcars or trailers. The container is loaded with pre-cooled material and the atmosphere injected and sealed in. This system allows continuous monitoring and management of temperature during transport and minimises risks of temperature abuse. Using this method Pacific salmon (Veranth & Robe 1979, Bell 1980) and Alaskan salmon (Schwartz 1982) have been successfully transported.

The second method, the master pack concept, employs permeable over-wrap packs of the type used in supermarkets placed in a large impermeable master pouch flushed with CO_2. It holds promise in that it provides shelf-life and convenience since the master pack is maintained under strict control during transport and retailers open the master cartons as required. The system eliminates the need for re-packing in the supermarket and protects the consumer from any misconceptions about the stability of the product. This system has been successful for a Gulf-coast area processor in the USA (Bannar 1979).

The third type of package is the individual consumer pack intended for direct retail sale. Bell (1982) sees this system as hazardous since each pack is a potential 'leaker' susceptible to mishandling and abuse. In view of the botulism hazards associated with temperature abuse it is generally recommended that at this stage modified atmospheres should not be used in retail packs of fish products (Wilhelm 1982, Anon. 1979). In Great Britain the incidence of *Cl. botulinum* type E in the marine environment is low (Shewan 1970), but the overall incidence in farmed trout is 9.4% (Cann, Taylor & Hobbs 1975). In view of the low degree of contamination, modified atmospheres are being used for pre-packaged cod, haddock, plaice, mackerel, Dover and lemon soles, crab, lobster, scampi (Tiffney & Mills 1982a) and smoked fish lines (Anon. 1983). The development of adequate processing procedures (Cann & Taylor 1979) has also allowed the safe use of modified atmospheres with trout products.

Very little work has been done on the development of this type of packaging system for use in the Australian fishing industry. The industry's needs must be carefully considered before appropriate systems can be applied on a commercial basis. The benefits to be gained for species caught in Australian waters must be weighed against possible drawbacks of the system, for example increased freight charges due to increased pack volumes, the apparent necessity to modify currently available packaging machinery to deliver adequate gas volumes, and the risk of temperature abuse. It appears, however, that MAS of fisheries products holds some promise as a means of maintaining quality and may prove to be a useful method of transportation and distribution of chill-stored fish.

Acknowledgement

This paper was prepared during the tenure of a grant from the Fishing Industry Research Trust Account to the Tasmanian Food Research Unit.

References

Anon. London: Department of Scientific and Industrial Research; 1950. Report Food Investig. Board: 22-3.

Anon. Preliminary results on the potential public health safety of fishery products stored in modified atmosphere. Seattle: NW Alaska Fisheries Center, Utilization Research Division; 1979. Activities Report 21 December.

Anon. CAP is the fashion to follow. Fish. News Int. 22(2) : 58; 1983.

Banks, H. Nickelson, R. & Finne, G. Shelf-life studies on carbon dioxide packaged finfish from the Gulf of Mexico. J. Food Sci. 45: 157-62; 1980.

Bannar, R. Vacuum-packaging for fresh fish — scaling the first hurdle. Food Eng. Int. 4: 56-8; 1979.

Barnett, H.J., Stone, F.E., Roberts, G.C., Hunter, P.J., Nelson, R.W. & Kwok, J. A study in the use of a high concentration of CO_2 in a modified atmosphere to preserve fresh salmon. Mar. Fish. Rev. 44(3) : 7-11; 1982.

Bell, L. Keep fresh foods fresh. Package Eng. 25: 72-5; 1980.

Bell, L.D. Commercial perspectives on various modes of modified and controlled atmosphere applications for the seafood industry. Martin, R.E., ed. Proceedings of the first national conference on modified and controlled atmosphere packaging of seafood products, San Antonio, 1981. Washington, DC: National Fisheries Institute; 1982: 304-21.

Brown, W.D., Albright, M., Watts, D.A., Heyer, B., Spruce, B. & Price, R.J. Modified atmosphere storage of rockfish (*Sebastes miniatus*) and silver salmon (*Onorhynchus kisutch*). J. Food Sci. 45: 93-6; 1980.

Cann, D.C., Smith, G.L. & Houston, N.C. Further studies on marine fish stored under modified atmosphere packaging. Aberdeen: Ministry of Agriculture, Fisheries and Food; 1983.

Cann, D.C. & Taylor, L.Y. The control of botulism in hot-smoked trout and mackerel. J. Food Technol. 14: 123-9; 1979.

Cann, D.C., Taylor, L.Y. & Hobbs, G. The incidence of *Clostridium botulinum* in farmed trout raised in Great Britain. J. Appl. Bacteriol. 39: 331-6; 1975.

Clark, D.S. & Takacs, J. Gases as preservatives. ICMSF. Microbial ecology of foods. Vol. 1. Factors affecting life and death of microorganisms. New York: Academic Press; 1980: 170-92.

Coyne, F.P. The effect of carbon dioxide on bacterial growth with special reference to the preservation of fish. Part 1. J. Soc. Chem. Ind. 51: 119T-21T; 1932.

Coyne, F.P. The effect of carbon dioxide on bacterial growth with special reference to the preservation of fish. Part II. Gas storage of fresh fish. J. Soc. Chem. Ind. 52: 19T-24T; 1933.

Cutting, C.L. Changes in pH and buffering capacity of fish during spoilage. J. Sci. Food Agric. 12: 597-603; 1953.

Daniels, F. & Alberty, R.A. Physical Chemistry. 4th ed. New York: John Wiley & Sons; 1975.

Eklund, M.W. Significance of *Clostridium botulinum* in fishery products preserved short of sterilization. Food Tech. 36(12) : 107-12, 115; 1982.

Enfors, S.O. & Molin, G. The influence of high concentrations of carbon dioxide on the germination of bacterial spores. J. Appl. Bacteriol. 45: 279-85; 1978.

Enfors, S.O. & Molin, G. Effect of high concentrations of carbon dioxide on growth rate of *Pseudomonas fragi, Bacillus cereus* and *Streptococcus cremoris*. J. Appl. Bacteriol. 48: 409-16; 1980.

Enfors, S.O., Molin, G. & Ternstrom, A. Effect of packaging under carbon dioxide, nitrogen or air on the microbial flora of pork stored at 4°C. J. Appl. Bacteriol. 47: 197-208; 1979.

Eyles, M.J. & Warth, A.D. Assessment of the risk of botulism from vacuum-packaged raw fish: a review. Food Technol. Aust. 33: 574-80; 1981.

Fey, M.S. Extending the shelf life of fresh fish by potassium sorbate and modified atmospheres at 0-1 degree C. Ithaca, NY: Cornell University; 1980. PhD thesis.

Fey, M.S. & Regenstein, J.M. Extending shelf-life of fresh wet red hake and salmon using CO_2-O_2 modified atmosphere and potassium sorbate ice at 1°C. J. Food Sci. 47: 1048-54; 1982.

Finne, G. Modified- and controlled-atmosphere storage of muscle foods. Food Technol. 36(2) : 128-33; 1982.

Gee, D.L. & Brown, W.D. The effect of carbon monoxide on bacterial growth. Meat Sci. 5: 215-22; 1981.

Gill, C.O. & Tan, K.H. Effect of carbon dioxide on growth of *Pseudomonas fluorescens*. Appl. Environ. Microbiol. 38: 237-40; 1979.

Goodfellow, S.J. An overview of controlled atmosphere packaging of foods. Martin, R.E., ed. Proceedings of the first national conference on modified and controlled atmosphere packaging of seafood products; San Antonio, 1981. Washington DC: National Fisheries Institute; 1982: 205-18.

Gorczyca, E., Cohen, D. & Sumner, J.L. The chemistry and microbiology of shelf-life extension of fish species. Abstract, 16th Annual Convention, AIFST, May-June; 1983. Melbourne, Australia.

Gray, R.J.H., Hoover, D.G. & Muir, A.M. Attenuation of microbial growth on modified atmosphere-packaged fish. J. Food Prot. 46: 610-3; 1983.

Haard, N.F. & Lee, Y.Z. Hypobaric storage of Atlantic salmon in carbon dioxide atmosphere. Can. Inst. Food Sci. Technol. J. 15: 68-71; 1982.

Haines, R.B. The influence of carbon dioxide on the rate of multiplication of certain bacteria, as judged by viable counts. J. Soc. Chem. Ind. 52: 13T-17T; 1933.

Huss, H.H., Schoeffer, I., Pedersen, A. & Jepsen, A. Toxin production by *Clostridium botulinum* type E in smoked fish in relation to the measured oxidation reduction potential (E_h), packaging method and associated

icroflora. Connell, J.J., ed. Advances in fish science and technology. Torry Research Station Jubilee Conference. Farnham: Fishing News (Books); 1980; 476–9.
Jensen, M.H., Petersen, A., R ø ge, E.A. and Jepsen, A. Storage of chilled cod under vacuum at various concentrations of carbon dioxide. Connell, J.J., ed. Advances in Fish Science and Technology. Torry Research Station Jubilee Conference. Farnham: Fishing News (Books); 1980; 294–7.
Killefer, D.H. Carbon dioxide preservation of meat and fish. Ind. Eng. Chem. 22: 140–3; 1930.
King, A.D. & Nagel, C.W. Growth inhibition of a *Pseudomonas* by carbon dioxide. J. Food Sci. 32: 575–9; 1967.
King, A.D. & Nagel, C.W. Influence of carbon dioxide upon the metabolism of *Pseudomonas aeruginosa*. J. Food Sci. 40: 362–6; 1975.
Kolbe, H. Antiseptic properties of carbon dioxide. J. Prakt. Chem. 26: 249–55; 1882.
Kritzman, G., Chet, I. & Henis, Y. Effect of carbon dioxide on growth and carbohydrate metabolism in *Sclerotium rolfsii*. J. Gen. Microbiol. 100: 167–75; 1977.
Lannelongue, M., Finne, G., Hanna, M.O., Nickelson R. & Vanderzant, C. Storage characteristics of brown shrimp (*Penaeus aztecus*) stored in retail packages containing CO_2-enriched atmospheres, J. Food Sci. 47: 911–13, 923; 1982a.
Lannelongue, M., Finne, G., Hanna, M.O., Nickelson, R. & Vanderzant, C. Microbiological and chemical changes during storage of swordfish (*Xiphias gladius*) steaks in retail packages containing CO_2-enriched atmospheres J. Food Prot. 45: 1197–203; 1982b.
Lannelongue, M., Hanna, M.O., Finne, G., Nickelson, R. & Vanderzant, C. Storage characteristics of finfish fillets (*Archosargus probatocephalus*) packaged in modified gas atmospheres containing carbon dioxide. J. Food Prot. 45: 440–4; 1982c.
Lee, J.S. Selection and growth of seafood microorganisms under carbon dioxide environment. Martin, R.E., ed. Proceedings of the first national conference on modified and controlled atmosphere packaging of seafood products; San Antonio, 1981. Washington DC: National Fisheries Institute; 1982: 304–21.
Lindsay, R. Modified atmosphere packaging systems for refrigerated fresh fish providing shelf life extension and safety from *Clostridium botulinum* toxigenesis. Martin, R.E., ed. Proceedings of the first national conference on modified and controlled atmosphere packaging of seafood products; San Antonio, 1981. Washington DC: National Fisheries Institute; 1982a: 30–51.
Lindsay, R. Controlled atmosphere packaging. Proceedings of the fourth annual international seafood conference; Munich, 1981. Washington, DC: International Seafood Conference; 1982b: 80–91.
Mills, A. & Tiffney, P. The use of controlled atmosphere packaging for the storage of fish. Chilled Foods 1: 22–3; 1982.
Mitsuda, H. Kawai, F., Yamamoto, A. & Nakajima, K. Carbon dioxide-protein interaction in a gas-solid phase. J. Nutr. Sci. Vitaminol. 21: 151–62; 1975.
Mokhele, K., Johnson, A.R., Barrett, E. & Ogrydziak, D.M. Microbiological analysis of rock cod (*Sebastes* spp.) stored under elevated carbon dioxide atmospheres. Appl. Environ. Microbiol. 45: 878–83; 1983.
Newton, K.G., Harrison, J.C.L. & Smith, K.M. The effect of storage in various gaseous atmospheres on the microflora of lamb chops held at −1°C. J. Appl. Bacteriol. 43: 53–9; 1977.
Oberlender, V., Hanna, M.O., Miget, R., Vanderzant, C. & Finne, G. Storage characteristics of fresh swordfish steaks stored in carbon dioxide-enriched controlled (flow-through) atmospheres. J. Food Prot. 46: 434–40; 1983.
Ogrydziak, D.M. & Brown, W.D. Temperature effects in modified atmosphere storage of seafoods. Food Technol. 36(5): 86–96; 1982.
Parkin, K.L. & Brown, W.D. Preservation of seafood with modified atmospheres. Martin, R.E., Flick, G.J., Hebard, C.E. & Ward, D.R. eds. Chemistry and biochemistry of marine food products. Westport, CT: AVI Publishing Co.; 1982: 453–65.
Parkin, K.L., Wells, M.J. & Brown, W.D. Modified atmosphere storage of rockfish fillets. J. Food Sci. 47: 181–4; 1981.
Pooni, G.S. & Mead, G.C. Prospective use of temperature function integration for predicting the shelf-life of non-frozen poultry-meat products. Food Microbiol. 1: in press; 1984.
Regenstein, J.M. & Regenstein, C.E. The shelf-life extension of fresh fish. Boston. Paris: Bull. Int. Inst. Refr.; 1981. Annex 1981–4: 357–64.
Savell, J.W., Smith, G.C., Hanna, M.O. & Vanderzant, C. Packaging of beef loin steaks in 75% O_2 plus 25% CO_2. I. Physical and sensory properties. J. Food Prot. 44: 923–7; 1981.
Schmidt, C.F., Lechowich, R.V. & Folinazzo, J.F. Growth and toxin production by type E *Clostridium botulinum* below 40°F. J. Food Sci. 26: 626–30; 1961.
Schwartz, W. The M.A.P. program of Pace Fish Co. Martin, R.E., ed. Proceedings of the first national conference on modified and controlled atmosphere packaging of seafood products; San Antonio, 1981. Washington, DC: National Fisheries Institute; 1982: 243–9.
Sears, D.F. & Eisenberg, R.M. A model representing a physiological role of CO_2 at the cell membrane. J. Gen. Physiol. 44: 869–87; 1961.
Shewan, J.M. Bacteriological standards for fish and fishery products. Chem. Ind.: 193–9; 1970.
Silliker, J.H. The efficacy of controlled atmospheres in extending the shelf life of meat, poultry and fish. Martin, R.E., ed. Proceedings of the first national conference on modified and controlled atmosphere packaging of seafood products; San Antonio, 1981. Washington DC: National Fisheries Institute; 1982: 219–39.
Silliker, J.H. & Wolfe, S.K. Microbiological safety considerations in controlled atmosphere storage of meats. Food Technol. 34(3) : 59–63; 1980.
Silliker, J.H., Woodruff, R.E., Lugg, J.R., Wolfe, S.K. & Brown, W.D. Meat Sci. 1: 195–204; 1977.
Silva, M. Alternatives to modified atmosphere packaging. Martin, R.E., ed. Proceedings of the first national conference on modified and controlled atmosphere packaging of seafood products; San Antonio, 1981. Washington DC: National Fisheries Institute; 1982: 267–74.
Spahl, A., Reineccius, G. & Tatini, S. Storage life of pork chops in CO_2-containing atmospheres. J. Food Prot. 44: 670–73; 1981.
Sperber, W.H. Requirements of *C. botulinum* for growth and toxin production. Food Tech. 36(12) : 89–94; 1982.
Stansby, M.E. & Griffiths, F.P. Carbon dioxide in handling fresh fish — Haddock. Ind. Eng. Chem. 27: 1452–8; 1935.
Stier, R.F., Bell, L., Ito, K.A., Shafer, B.D., Brown, L.A., Seeger, M.L., Allen, B.H., Porcuna, M.N. & Lerke, P.A. Effect of modified atmosphere storage on *C. botulinum* toxigenesis and the spoilage microflora of salmon fillets. J. Food Sci. 46: 1639–42; 1981.
Sutherland, J.P., Patterson, J.T., Gibbs, P.A. & Murray, J.G. The effect of several gaseous environments on the multiplication of organisms isolated from vacuum-packaged beef. J. Food Technol. 12: 249–55; 1977.
Tarr, H.L.A. Changes in nutritive value through handling and processing procedures. Borgstrom, G. ed. Fish as food. Vol. 2. Nutrition, sanitation and utilization. New York: Academic Press; 1962: 235–60.
Tiffney P. & Mills, A. Cooling the catch — a package system for chilled fish. Fish. News Int. 21(4) : 20–1; 1982a.
Tiffney, P. & Mills, A. Storage trials of controlled atmosphere packaged fish products. Sea Fish Industry Authority, 1982b. Technical Report No. 191.
Tomlins, R.I., Gray, R.J.H., Muir, A.M., Hoover, D.G. & Elliot, P.H. Microbial variations during carbon dioxide modified atmosphere packaging in a model system and on fresh fish. Martin, R.E., ed. Proceedings of the first national conference on modified and controlled atmosphere packaging of seafood products; San Antonio, 1981. Washington DC: National Fisheries Institute; 1982: 158–85.
Valley, G. & Rettger, L.F. The influence of carbon dioxide on bacteria. J. Bacteriol. 14: 101–37; 1927.
Veranth, M.F. & Robe, K. CO_2-enriched atmosphere keeps fish fresh more than twice as long. Food Process. 40(4) : 76–9; 1979.
Wang, M.Y. & Brown, W.D. Effects of elevated CO_2 atmosphere on storage of freshwater crayfish (*Pacifastacus leniusculus*). J. Food Sci. 48: 158–62; 1983.
Wilhelm, K.A. Extended fresh storage of fishery products with modified atmospheres: a survey. Mar. Fish. Rev. 44(2) : 17–20; 1982.

REPRINTED FROM FOOD TECHNOLOGY IN AUSTRALIA
VOLUME 37 NO. 5 PP 212-214

Acceptability of trevalla (*Hyperoglyphe porosa* Richardson) after storage in carbon dioxide

J.A. STATHAM and H.A. BREMNER

The effect of a 100% carbon dioxide atmosphere on the storage life of trevalla (*Hyperoglyphe porosa* Richardson) (initial pH 6.2) at 4°C was investigated. Frozen trevalla and trevalla stored in air were used as comparisons for organoleptic assessment. Trevalla stored in air remained acceptable for eight days; trevalla stored in CO_2 remained acceptable and was not scored significantly lower than the frozen control until after 16 days in store. Bacteria grew rapidly in trevalla stored in air. Bacterial growth was slow during storage under 100% CO_2 and the resulting flora was dominated by lactic acid bacteria.

The preservative effects of CO_2 on flesh foods were demonstrated in 1882 by Kolbe. Fifty years later, after showing that atmospheres rich in CO_2 extended the shelf life of whitefish, Coyne (1933) concluded that "in general distribution, i.e. in transport and in retail stores, gas storage should be of considerable value". Another half century later the practice is not widely used for packing of fish although in recent years has been the subject of much investigation. Recent literature reviews by Parkin and Brown (1982) and Statham (1984) cite reports that fish storage in atmospheres containing 40–100% CO_2 results in shelf life extension of between 50–100%. This is due mostly to suppression of the Gram-negative spoilage bacteria resulting in a population shift towards Gram-positive bacteria, mainly lactobacilli (Banks, Nickelson & Finne 1980, Silliker & Wolfe 1980, Finne 1982, Lannelongue *et al.* 1982a,b, Ogrydziak & Brown 1982, Mokhele *et al.* 1983, Oberlender *et al.* 1983).

Shelf life extension gives a commercial impetus to the research, yet a broad range of interrelated problems still has to be solved. These problems range from the fundamental aspects of the mechanism(s) of suppression of bacteria by CO_2 to the composition of the gas atmosphere, the packing technology involved, and to the commercial and consumer acceptability of the product and how this changes with time of storage.

In a CO_2-rich environment fish flesh can absorb up to 1.3 times its own volume of CO_2 within 24 h (Statham & Bremner, unpublished data). The dissolution of CO_2 in the flesh causes the pack to collapse and for this reason some workers have diluted the CO_2 with other gases to maintain pack integrity. Dissolved CO_2 is in complex equilibrium of CO_2 (aq.), H_2CO_3, HCO_3^- and $CO_3^{=}$. It is the undissociated H_2CO_3 that is likely to be the active bacterial suppressant (Bell, Etchells & Borg 1959) although other mechanisms have been proposed.

Undissociated carbonic acid concentration is proportional to the partial pressure of CO_2 in the atmosphere and is independent of flesh pH when the system is in equilibrium with gaseous CO_2 (Umbreit, Burris & Stauffer 1964). This means that under the same conditions, dilution of CO_2 in the pack atmosphere produces lower H_2CO_3 concentration. Much of the recent work has been concerned with establishing the minimum proportion of CO_2 consistent with shelf life extension without so much regard to this relationship. Nevertheless, fish packed in various gas mixtures is on retail sale in the UK (Tiffney & Mills 1982a, Anon. 1982a,b, 1983a,b). To the authors' knowledge no modified atmosphere-stored fish in on sale in Australia and snapper *(Chrysophrys auratus)* is the only Australian species known to have been packed experimentally (Scott, Fletcher & Summers 1984). It was decided that as a reference point packing in a known volume of 100% CO_2 should first be investigated before any particular gas mixture was chosen from the variety of possible combinations employed by other workers (Statham 1984, Table 1). There are no published reports of the acceptability of fish stored in CO_2 as judged by a consumer-type panel. Accordingly, the present experiment was designed to investigate the acceptability of trevalla *(Hyperoglyphe porosa)* fillets packed in 100% CO_2.

Materials and methods

Trevalla caught off St Helens, on the east coast of Tasmania, were stored in ice and taken to the Laboratory within 48 h of landing. Three fish were sampled to determine bacterial numbers on the starting material. The fish were filleted, skinned and cut into pieces of approximately 150 g and allocated randomly to one of three treatments, viz. packed in polyethylene bags tied loosely at the neck and stored at 4°C, vacuum packed in Cryovac U gauge barrier bags (Registered trade name of W.R. Grace Pty Ltd, O_2 transmission rate 3.5 mL m^{-2} 24 h^{-1} atm $^{-1}$ at 2.5°C and 75% RH, CO_2 transmission rate 12 mL m^{-2} 24 h^{-1} atm^{-1} at 3.5°C and 75% RH) and stored at −18°C or packed in Cryovac U gauge barrier bags evacuated and then backflushed with sufficient 100% CO_2 to give a ratio in the sealed pack of 4 volumes of CO_2 to 1 volume of fish. Fish and total pack volumes were estimated by displacement; fish stored in CO_2 was kept at 4°C.

Packing equipment

A chamber vacuum-packing machine (Boss, Regina 2/100, Germany) was modified for gas flushing by (a) replacing the existing time switch with one having a 60 s time cycle (Omron Tateisi Electronics Co., Japan) to extend the backflush cycle and allow packs to be filled with CO_2 at a pressure near atmospheric and (b) installing independent pressure lines to the pneumatically-activated sealing bars which otherwise would not operate at the low pressure differentials between the chamber and atmosphere.

Sampling

Six packs from each of the frozen and modified atmosphere treatments were sampled at intervals of 1, 3, 6, 9, 13, 16, 20 and 23 days. Spoilage of the samples stored in air was evident after 9 days; accordingly fresh trevalla was substituted for these to balance samples presented to taste panels, except for day 23 when frozen morwong *(Nemadactylus macropterus)* was used. At each sampling time, the appearance and odour of the raw fish were

Jo Statham and Allan Bremner are experimental officers at the CSIRO Division of Food Research, Tasmanian Food Research Unit, Stowell Avenue, Hobart, Tas. 7000.

0015-6647/85/37/0212/04

recorded by three experienced assessors and drip volume measured. Bacterial numbers were determined in small portions of three pieces of fish stored in air or CO_2. Surface pH of each piece of fish was measured by an Orion contact pH electrode and, after dividing each piece into 3 x 50 g portions for taste panel assessment, cut surface pH was measured. Fish pieces for taste panel assessment were coated in flour, beaten egg and breadcrumbs, then deep fried in oil (Salfry, Provincial Traders Pty Ltd, Australia) at 177°C for 2.25 min yielding 72°C centre temperature.

Taste panel

The taste panel consisted of 18 members of the CSIRO Tasmanian Regional Laboratory staff unaware of the details of the experiment. At each sampling time the panel was divided into three sittings of six tasters and each taster was asked to taste three samples, one from each of the three treatments. At the first taste session the six possible taste orderings were randomly allocated to the six tasters in a sitting. The order of tasting was then varied cyclically within a sitting over the remaining seven sessions so as to ensure effective balance. Tasters scored each sample for flavour, texture and overall acceptability on a pictorial hedonic 'Smiley' scale of 1 to 7 (Quarmby, Bremner & Thrower 1982). Statistical analysis of these three variates was performed using the analysis of variance facilities of the GENSTAT Statistical Package (Alvey *et al.* 1977).

Microbiology

Flesh samples (5.1 cm^2) were homogenised in sterile saline for 3 min using a Colworth stomacher. Serial decimal dilutions were spread-plated on tryptone soya agar (Oxoid) containing glucose (2 g/L), yeast extract (2 g/L) and NaCl (5 g/L). The medium of de Man, Rogosa and Sharpe (MRS) (1960) was used similarly to estimate numbers of presumptive lactic acid bacteria on samples stored under CO_2 for 20 and 23 days. All plates were incubated in air (22°C, 3–5 days).

Results and discussion

Raw odour and appearance

As judged by odour and appearance of the raw fish, the fish in all treatments remained acceptable until the ninth day when strong off odours were detected in aerobic packs (Table 1). Although some bleaching and staining occurred in fish stored in CO_2, they were considered acceptable. These observations indicate that spoilage in CO_2 follows a different mechanism from that in air.

Drip and pH

Figures 1a and 1b show changes in internal and external pH values. After 9 days pH of fish stored in air had risen significantly and spoilage had occurred. Samples stored in CO_2 showed a slight rise in external pH above the initial value after 13 days in storage, while the internal value did not rise significantly until after 20 days in storage. Frozen samples' pH did not change significantly until after 16 days in storage, when both internal and external values decreased. Frozen fish pH was not determined at the subsequent sampling time; therefore no conclusions can be drawn as to the significance of this change. No significant difference was noted between the internal and external pH values of fish stored frozen or in CO_2 until after 20 days in storage when surface pH values of CO_2-stored fish were higher than internal ones. The amount of drip from individual packs varied from 2% to 11%. Drip loss was not affected by storage time; however, it was positively correlated ($r = 0.619$) with the hydrogen ion concentration of the fish from which it was lost. This results from a decrease in water binding capacity as pH approaches the isoelectric point of fish muscle (approximately pH 5.6) (Buttkus & Tomlinson 1966, Wagenknecht & Tulsner 1974). In commercial practice drip loss is likely to be controllable by the use of polyphosphates, but these at present are only permitted in frozen fish.

Microbiology

Increases in aerobic bacterial numbers on fish stored in air and in CO_2 are shown in Figure 2. Bacterial numbers increased rapidly on fish stored in air, reaching a maximum of 10^9 cfu/cm^2 after 9 days. The bacteria on fish stored in CO_2 multiplied less rapidly, reaching a maximum of 10^7 cfu/cm^2 after 13 days. A lag phase of around 3 days was evident in the CO_2 treatment. Large standard errors associated with some estimates from fish stored in CO_2 suggest variations between conditions in individual packs rather than errors in sampling technique because the standard errors associated with all samples stored in air are small. Similar variability was also found with packaged snapper fillets (Scott *et al.* 1984). At 20 and 23 days storage lactic acid bacteria represented 95% of the total flora on fish stored in CO_2.

Table 1. Changes in raw odour and appearance of raw trevalla during chilled storage in air or CO_2

Storage time (days, 4°C)	Treatment	Odour	Appearance
0	—	Fresh	Firm, translucent
9	Aerobic	Floury, cheesy, stale, sulphides	—
	CO_2	Bland	Flesh slightly soft and spongy
13	CO_2	Bland, slightly pickled, meaty, metallic	Some white or bleached, others with slight brown staining where fillets were folded
16	CO_2	Slightly stale, fishy	—
20	CO_2	Meaty, slightly fishy, pickled	Marinated, white
23	CO_2	Slightly fishy	Marinated, white

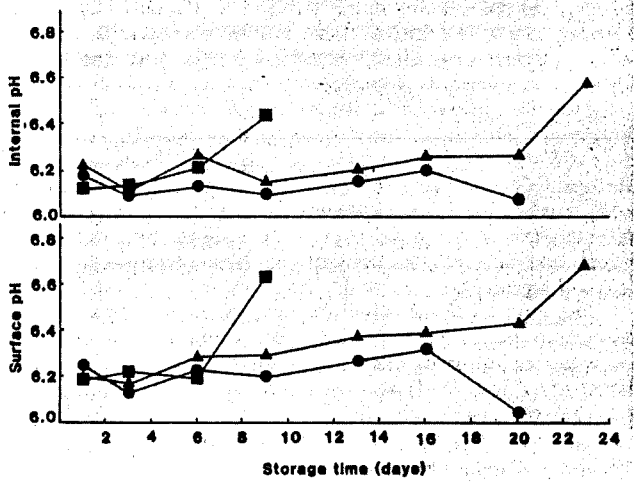

Figure 1. pH of trevalla fillets stored at 4°C or -18°C; a) internal, b) surface; ●, frozen; ■, air; ▲, CO_2

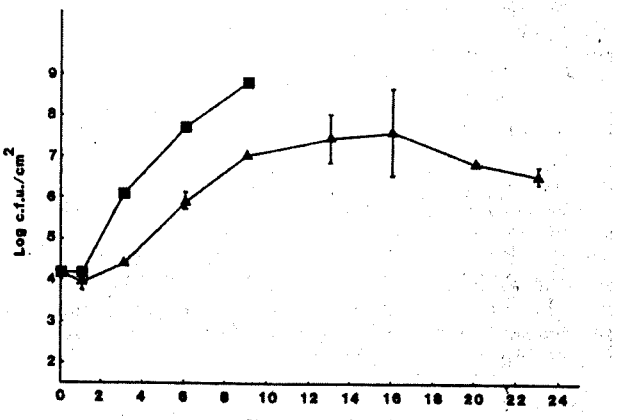

Figure 2. Bacterial numbers on trevalla fillets stored at 4°C. ■, air; ▲, CO_2. Each point represents the mean of three individual packs

Taste panel

Figures 3, 4 and 5 show the results of the taste panel assessment. Flavour, texture and overall acceptability of trevalla stored in air decreased significantly during 9 days in storage. Texture and overall acceptability scores for CO_2-stored fish dropped less dramatically and it was not until after 13 days that the scores for these attributes differed significantly from those for frozen fish. The flavour scores for fish stored in CO_2 differed from those for the frozen fish at 16 days, but apparently this had less effect than texture on the overall acceptability. If a drop in overall acceptability score to 4 or less is taken as the point of rejection, then the samples stored in air were acceptable for 8 days, while the fish in the modified atmosphere remained acceptable for 16 days representing doubling of shelf life.

Conclusion

This experiment has established that the shelf life of trevalla stored at 4°C can be increased from 8 to 16 days by packaging the fish in 100% CO_2. At 0°C, an overall shelf life of at least 30 days could be expected, due directly to the effect of reduced temperature on bacterial growth (Ratkowsky *et al.* 1982) and indirectly to the greater solubility of CO_2 in the flesh (Ogrydziak & Brown 1982). The flexible packs used in this experiment would not be practical for retail sale because the gas-to-fish ratio makes them unattractive in appearance and difficult to handle. A possible solution would be to use a rigid moulded base with a flexible top, similar to those developed in the UK (Tiffney & Mills 1982a,b). When first filled, the top would 'balloon' out but after the CO_2 had dissolved in the tissue it would retract to lie flush. Irrespective of the approach chosen, the development of suitable packs and packaging systems requires the co-operation of retailers, packaging equipment suppliers and processors.

Acknowledgements

The authors thank Alex Vail and Stephen Sykes, CSIRO Tasmanian Food Research Unit, for modifications made to packaging equipment, CIG for supplying CO_2 and Dr R.G. Jarrett and Mr R.K. Lowry, CSIRO Division of Mathematics and Statistics, who assisted with experimental design and analysis of results. A grant from the Fishing Industry Research Trust Account supported this work.

References

Alvey, N.G. and others. GENSTAT. A general statistical program. Harpenden, Herts: Rothamstead Experimental Station, UK; 1977.

Anon. CAP-set to fill that grocery gap? Chilled Foods October: 13, 17; 1982a.

Anon. A 'new era' of packaging. Fish. News Int. 21 (12): 32–3; 1982b.

Anon. CAP is the fashion to follow. Fish. News Int. 22(2): 58; 1983a.

Anon. CAP helps move to the supermarkets. Fish. News Int. 22(12): 50; 1983b.

Banks, H., Nickelson, R. & Finne, G. Shelf-life studies on carbon dioxide packaged finfish from the Gulf of Mexico. J. Food Sci. 45: 157–62; 1980.

Bell, T.A., Etchells, J.L. & Borg, A.F. Influence of sorbic acid on certain species of bacteria, yeasts and filamentous fungi. J. Bacteriol. 77: 573–80; 1959.

Buttkus, H. & Tomlinson, N. Some aspects of postmortem changes in fish muscle. Briskey, E.J., Cassens, R.G. & Trautman, J.C., eds. The physiology and biochemistry of muscle as a food. Madison, WI: University of Wisconsin Press; 1966: 197–203.

Coyne, F.P. The effect of carbon dioxide on bacterial growth with special reference to the preservation of fish. Part II. Gas storage of fresh fish. J. Soc. Chem. Ind. 52: 19T–24T; 1933.

de Man, J.C., Rogosa, M. & Sharpe, M.E. A medium for the cultivation of lactobacilli. J. Appl. Bacteriol. 23: 130–5; 1960.

Finne, G. Modified- and controlled-atmosphere storage of muscle foods. Food Technol. 36(2): 128–33; 1982.

Kolbe, H. Antiseptische Eigenschaften der Kohlensäure. J. Prakt. Chem. 26: 249–55; 1882.

Lannelongue, M., Finne, G., Hanna, M.O., Nickelson, R. & Vanderzant, C. Storage characteristics of brown shrimp *(Penaeus aztecus)* stored in retail packages containing CO_2-enriched atmospheres. J. Food Sci. 47: 911–13; 1982a.

Lannelongue, M., Finne, G., Hanna, M.O., Nickelson, R. & Vanderzandt, C. Microbiological and chemical changes during storage of swordfish *(Xiphias gladius)* steaks in retail packages containing CO_2-enriched atmospheres. J. Food Prot. 45: 1197–203; 1982b.

Mokhele, K., Johnson, A.R., Barrett, E. & Ogrydziak, D.M. Microbiological analysis of rock cod (*Sebastes* spp.) stored under elevated carbon dioxide atmospheres. Appl. Environ. Microbiol. 45: 878–83; 1983.

Oberlender, V., Hanna, M.O., Miget, R., Vanderzant, C. & Finne, G., Storage characteristics of fresh swordfish steaks stored in carbon dioxide-enriched controlled (flow-through) atmospheres. J. Food Prot. 46: 434–40; 1983.

Ogrydziak, D.M. & Brown, W.D. Temperature effects in modified atmosphere storage of seafoods. Food Technol. 36(5): 86–96; 1982.

Parkin, K.L. & Brown, W.D. Preservation of seafood with modified atmospheres. Martin, R.E., Flick, G.J., Hebard, C.E. and Ward, D.R., eds. Chemistry and

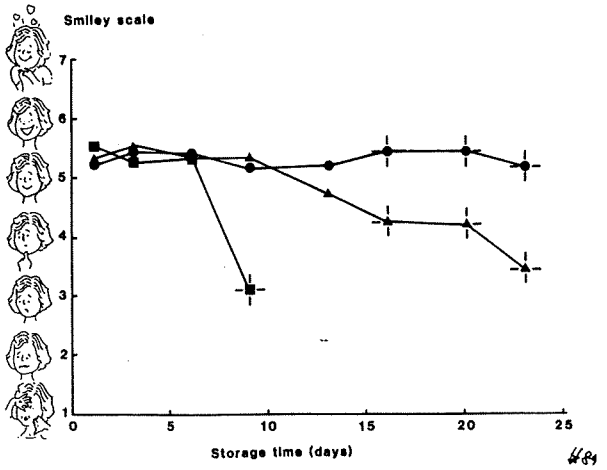

Figure 3. Flavour acceptability of trevalla fillets stored at 4°C or −18°C. ●, frozen; ■, air; ▲, CO_2. Treatments differing significantly at any one sampling ($p<0.05$ by analysis of variance) are marked with ÷

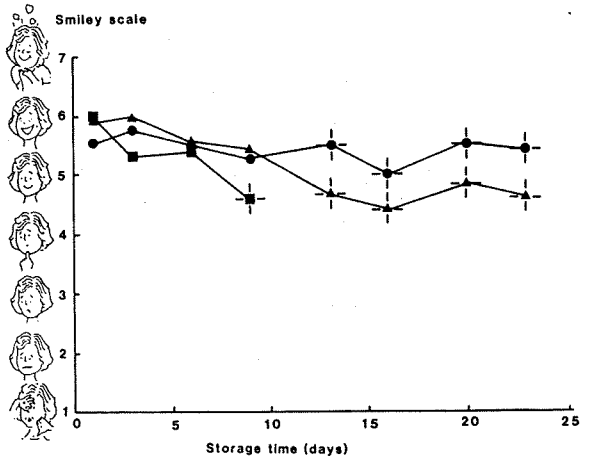

Figure 4. Texture acceptability of trevalla fillets stored at 4°C or −18°C. ●, frozen; ■, air; ▲, CO_2. ÷ As in Fig. 3

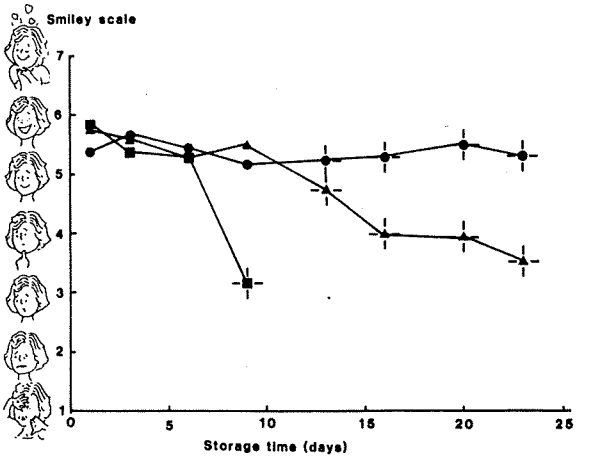

Figure 5. Overall acceptability of trevalla fillets stored at 4°C or −18°C. ●, frozen; ■, air; ▲, CO_2. ÷ As in Fig. 3

biochemistry of marine food products. Westport, CT: AVI Publishing Co.; 1982: 453–65.
Quarmby, A.R., Bremner, H.A. & Thrower, S.J. On board handling of gemfish. Part II: Sensory profiles. Aust. Fish. 41: 42–5; 1982.
Ratkowsky, D.A., Olley, J., McMeekin, T.A. & Ball, A. Relationship between temperature and growth of bacterial cultures. J. Bacteriol. 149: 1–5; 1982.
Scott, D.N., Fletcher, G.C. & Summers, G. Modified atmosphere and vacuum packing of snapper fillets. Food Technol. Aust. 36: 330–4; 1984.
Silliker, J.H. & Wolfe, S.K. Microbiological safety considerations in controlled atmosphere storage of meats. Food Technol. 34(3): 59–63; 1980.
Statham, J.A. Modified atmosphere storage of fisheries products — the state of the art. Food Technol. Aust. 37: 233–9; 1984.
Tiffney, P. & Mills, A. Cooling the catch — a package system for chilled fish. Fish. News Int. 21(4): 20–1; 1982a.
Tiffney, P. & Mills, A. Storage trials of controlled atmosphere packaged fish products. Edinburgh, UK: Sea Fish Industry Authority; 1982b. Technical Report No. 191.
Umbreit, W.W., Burris, R.H. & Stauffer, J.F. Manometric techniques. Minneapolis, MN: Burgess Publishing Company; 1964: 18–27.
Wagenknecht, W. & Tulsner, M. Die Wasserbindung im Fischmuskel und ihre Veränderungen. Fischereiforschung 12: 69–83; 1974.

Reprinted from J. Food Sci.
50(6): 1580–1584,& 1587

Storage of Morwong (*Nemadactylus macropterus* Bloch and Schneider) in Combinations of Polyphosphate, Potassium Sorbate and Carbon Dioxide at 4°C

JO A. STATHAM, H. ALLAN BREMNER, and ANTHONY R. QUARMBY

ABSTRACT

Combinations of potassium sorbate and polyphosphate, in conjunction with vacuum or modified atmosphere packaging, were assessed for their preservative effects on morwong fillets (pH 6.75) stored at 4°C. Vacuum packaged and/or frozen fillets were used as comparisons for microbiological and taste panel analysis. A combination of potassium sorbate, polyphosphate and 100% CO_2 was the most effective packaging regime. Potassium sorbate on vacuum packaged fish was more effective than a 100% CO_2 atmosphere alone. Polyphosphate had no apparent additional effect on fillets stored under vacuum with or without potassium sorbate.

INTRODUCTION

A SATISFACTORY METHOD for extending the shelf-life of chilled fish and shellfish has long been sought, to ensure quality, continuity of supply and minimal wastage. Preservatives such as sorbic acid and its potassium salt have met with varying success. The reported effects of potassium sorbate dips range from being minimal (Fey and Regenstein, 1982) to extending the shelf-life of vacuum packed scallops from 4–5 days to 28 days at 4°C (Bremner and Statham, 1983). The use of modified atmospheres incorporating high proportions of CO_2 has also had success, with shelf-life extension ranging from 50-100% (Statham, 1984). A combination of these two treatments may give rise to additive or synergistic effects, providing greater assurance of product quality and stability.

Used in conjunction with atmospheres containing 60% CO_2, potassium sorbate dips or potassium sorbate incorporated into ice were effective in extending the shelf-life of red hake and salmon to 4 wk at 1°C (Regenstein, 1982; Fey and Regenstein, 1982). Pathogens such as *Salmonella enteriditis* (Elliot and Gray, 1981) and *Staphylococcus aureus* (Elliot et al., 1982) are inhibited to a greater extent in media containing potassium sorbate and incubated in CO_2 enriched atmospheres than by either treatment alone.

Fish packaged in atmospheres containing high levels of CO_2 (>60%) have been reported to suffer from excessive drip loss (Mills and Tiffney, 1982). Nitrogen may be used as an inert filler to lower the proportion of CO_2. Oxygen may also be added if the product is likely to undergo color changes or bleaching (Cann, 1984). However pre-mixed gases are not always readily available, therefore the use of 100% CO_2 in conjunction with polyphosphate dips to prevent drip may be a more viable alternative. Statham and Bremner (1985) discuss the importance of high partial pressures of CO_2 in MAP fish. Polyphosphate dips could be used to overcome this problem. There is some evidence that polyphosphates may themselves have an inhibitory effect on the bacterial flora of flesh foods (Elliot et al., 1964; Van Wazer, 1971); however, increases in shelf-life, when they occur, are in the order of 1–2 days (Spencer and Smith, 1962). Some suppression of toxin production by *Clostridium botulinum* in beef/pork frankfurter emulsions may occur as a result of synergism between sodium acid pyrophosphate, potassium sorbate and sodium nitrite (Wagner and Busta, 1983).

The purpose of this experiment was to assess the efficacy of a potassium sorbate dip, when used in conjunction with a 100% CO_2 atmosphere, to extend the chill-storage life of morwong (*Nemadactylus macropterus* Bloch and Schneider). Potassium sorbate, CO_2 and polyphosphate treatments were also used individually to allow the estimation of additive or synergistic effects of various combinations of treatments.

Authors Statham, Bremner, and Quarmby are with CSIRO, Division of Food Research, Tasmanian Food Research Unit, "Stowell," Stowell Ave., Hobart 7000, Tasmania, Australia.

MATERIALS & METHODS

FRESH MORWONG FILLETS (pH 6.75) were bought from the Melbourne fish market the day after catching and were air freighted in polyethylene bags, surrounded by ice, to Hobart the same evening. On arrival at the Tasmanian Food Research Unit (TFRU) the fillets were skinned by hand and stored in a 0 ± 0.5°C cool room overnight. Morwong is a medium to low-priced white fish which is representative of the perch family, having a composition of 19% protein, 1% fat, 78% moisture and 1.5% ash (unpublished results from this laboratory). These fish are widely distributed through Australian and New Zealand waters and are available for most of the year. At present the stocks are under-exploited in some areas (Wankowski, 1984).

Preparation of dip solutions

A 10% polyphosphate dip solution was prepared containing 5.3% sodium tripolyphosphate and 4.7% sodium pyrophosphate (Albright and Wilson (Australia) Limited) to give a pH of 6.2 in the final solution. A 1.2% potassium sorbate solution was prepared and the pH adjusted to 6.2 using hydrochloric acid. This concentration had previously been found to give a residual level of near 0.1% (w/w) potassium sorbate on the fillets. A composite polyphosphate/potassium sorbate dip with the solutes at the above concentrations was prepared and its pH adjusted to 6.2 using hydrochloric acid.

In all cases the fillets were immersed in the dips for 1 min as recommended by manufacturers of polyphosphates for the treatment of fish (Albright and Wilson (Australia) Ltd., Technical Bulletin - 'Mera' polyphosphate for treating seafood) and were allowed to drain for 5 min before packaging. Preliminary experiments showed that polyphosphate did not affect the rate or amount of sorbate uptake, that sorbate uptake had reached a maximum after 1 min in the dip and that 2 days after dipping surface levels were still higher than at the center of fillets 25 mm in thickness.

Packaging

In this experiment seven different treatments were employed, as shown in Table 1. Fillets were packed three to a bag (approximate total weight 240g). Twenty-eight packs were prepared for each treatment, except for treatment F, where extra packs were prepared. Cryovac U gauge barrier bags (W.R. Grace Pty. Ltd., Melbourne), a laminate of EVA/PVDC/EVA having a nominal OTR and CO_2TR of 3.5 and 12 mL m^{-2} $24hr^{-1}$ atm^{-1} at 3.5°C and 75% RH were used. Packaging was carried out using a Boss, Regina 2/100 Germany, vacuum packaging machine equipped for gas flushing. Fillets were packaged side by side with the minimum degree of overlap. In those treatments which were vacuum packed (V, F, SP, P and S) the film was in firm contact with the fillets. In those packs flushed with CO_2 (SPC and C) the gas initially held the film away from the upper surface of the fillet; however, as CO_2 was absorbed by the flesh the bags deflated. By 24

Table 1—Packaging treatments

Treatment	Code	Atmosphere	Dip	Storage temp (°C)
Vacuum	V	vacuum	nil	4
Frozen	F	vacuum	nil	−18
CO_2	C	100% CO_2	nil	4
Polyphosphate	P	vacuum	10% polyphosphate	4
Sorbate	S	vacuum	1.2% potassium sorbate	4
Sorbate + polyphosphate	SP	vacuum	1.2% potassium sorbate + 10% polyphosphate	4
Sorbate + polyphosphate + CO_2	SPC	100% CO_2	1.2% potassium sorbate + 10% polyphosphate	4

hr after packaging the film was in contact with the surface but not as tightly as in the vacuum packaged treatments.

Four packs from treatments V, F, SP and SPC and three packs from treatments C, P, and S were assessed after 0, 3, 6, 10, 13, 17, and 20 days in storage.

Sorbate analysis

Seventeen sorbate-treated fillets were frozen (−18°C) for analysis. After 4 months' storage the fillets were thawed, diced then extracted and the sorbate levels determined by U.V. spectrophotometry (AOAC, 1980). Previous experiments done in this laboratory indicate that potassium sorbate is stable on fish flesh for at least 10 months at a temperature of −18°C (Bremner, 1984, unpublished data).

Microbiology

Sampling. Treatments V, P, S, and SP were sampled immediately after packaging. Those samples subjected to the same dips but flushed with CO_2 or vacuum packaged were taken as being the same at that stage (i.e. SP = SPC, V = C). At subsequent sampling times all chill-stored treatments were sampled. Three packs from each treatment were selected at random and one fillet from each pack was sampled. From these fillets a 16 cm^2 surface sample was excised using a sterile template and scalpel. Following homogenization in a Colworth Stomacher, serial decimal dilutions were prepared in sterile saline and spread plated on to Tryptone Soya Agar (Oxoid). The medium was enriched by the addition of 0.2% yeast extract (Sigma), 0.2% glucose (M and B) and 0.5% NaCl to encourage growth of fastidious bacteria, particularly lactobacilli. Plates were incubated at 22°C for 3–5 days.

Identification of isolates. Twenty colonies were picked at random from plates from treatment V at time 0 and after 6 days, treatments SP and C after 10 days and treatment SPC after 20 days. The isolates taken from untreated fish at time 0 were considered to be representative of the initial flora, while the isolates taken later in storage were considered to represent the spoilage flora of each treatment. Gram negative isolates were identified using the scheme of Hendrie and Shewan (1979). Gram positive isolates were identified according to *Bergey's Manual of Determinative Bacteriology* (Buchanan and Gibbons, 1974).

pH, raw odor and appearance

The surface pH was measured at four positions on each fillet from each pack using an Orion surface pH electrode. Internal pH of three fillets from each treatment was measured at four positions on the cut surface directly below the area sampled for microbiological analysis. The appearance and odor of the raw fish was recorded by two people experienced in judging fish quality, and the types and intensities of odors were used to assess the suitability of the fish for taste panel analysis. The amount of drip produced in each pack was measured volumetrically, if present in sufficient quantity.

Profile panel

Frozen fish was used by the taste panel as reference "fresh fish" samples and as substitutes for treatments that were withdrawn from the tasting sessions as they became unacceptable. Two fillets from the four packs from treatments V, SP, SPC, and F, plus reference samples, were halved and placed in individual, previously coded, plastic bags. All bags were strung on a rod in the correct taste order for each panelist and the bags were suspended in a waterbath set at 84°C. The fish was cooked for 20 min to a center temperature of 75°C. When cooked the fish was removed from the bags and placed in heated, coded glass jars and served to the taste panel.

The profile panel consisted of 16 staff members who were unaware of the details of the experiment but were experienced in assessing seafoods and in the profile technique. One taster was absent at the first and fifth sessions and two tasters were absent at the seventh session. At each sampling time the 16 tasters were divided into two sittings of eight tasters and each taster was asked to taste an identified reference sample, then four coded samples, one from each of treatments F, V, SP, and SPC. The five samples were served at one time and the panelists were asked to taste them according to the order marked on their profile sheets. The 24 possible taste orderings were randomly allocated to the 16 tasters at the first taste session. The order of tasting was then varied cyclically within a session over the remaining six sessions so as to minimize taste order effects. Samples from treatments V and SP became unsuitable for tasting by the 10th and 13th days of storage, respectively, and were replaced by frozen fillets (equivalent to treatment F) to maintain sample numbers at all subsequent taste sessions.

The panelists were asked to assess the fish by the odor and flavor profiles used previously in this laboratory (Quarmby et al. 1982; Bremner and Statham, 1983; Statham and Bremner, 1983) with the following differences in methodology. The panelists were in isolated taste booths and they marked their score sheets independently with no round table discussion. Instead of the 6-point structured scale used previously, a 10-point unstructured scale (0 = absent, 9 = strong) was used for scoring the intensity of descriptors listed on the profiles. Scoring for the attributes of odor, off odor, flavor and off flavor was obligatory. Texture was assessed by scoring the initial characteristics, wetness, firmness, springiness, and the secondary characteristics, toughness, succulence and fibrousness (Howgate, 1977) on this same scale. The panelists were also asked to score for odor, flavor and overall acceptability of the samples on the 7-point facial 'Smiley Scale' (Bremner and Statham, 1983).

Profile data analysis

The scores for the attributes which panelists were obliged to score, viz. fish odor, off odor, fish flavor and off flavor were subjected to analysis of variance. For the remaining 'free choice' attributes the total panel score or total panel response was transformed to a proportion of the total possible panel response, i.e. if all panelists had responded and scored 9 for that attribute. These data are plotted as integer values on a 0 to 100 scale. This technique can thus be used to compare the responses from panels of different sizes. When an attribute at any one session was scored by only one panelist, then this datum has been deleted from the data set.

RESULTS

Sorbate levels

The mean potassium sorbate level was 0.08%. Morwong fillets of the size range used in this experiment (80–120g) have an area-to-weight ratio from approx. 0.45 to 0.55 g/cm^2. This means that for a 100-g fillet, potassium sorbate was applied at an average level of 0.4 mg/cm^2.

pH changes

Internal and surface pH values of the fillets were not significantly different ($p<0.05$) and were therefore combined to give the means plotted in Fig. 1. One fillet from each of two packs in treatment SPC sampled at 20 days had pH values significantly lower than the others in that treatment measured at the same time (mean pH 5.2 compared to 6.4). These values were considered to be unrepresentative and were therefore not included in the final means.

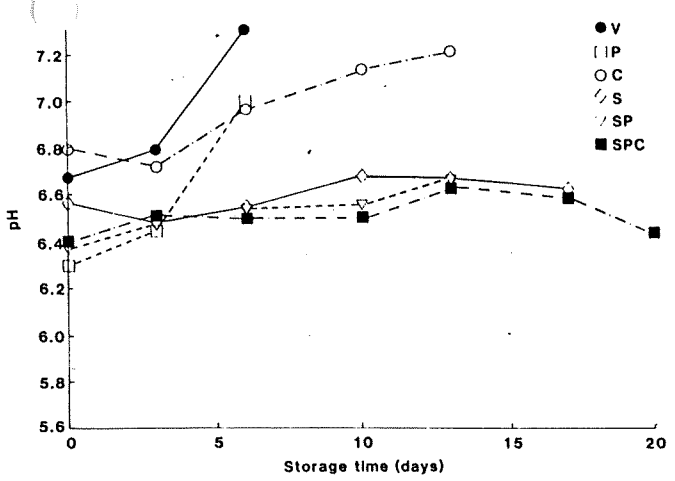

Fig. 1—pH of morwong fillets stored at 4°C. (See Table 1 for Code).

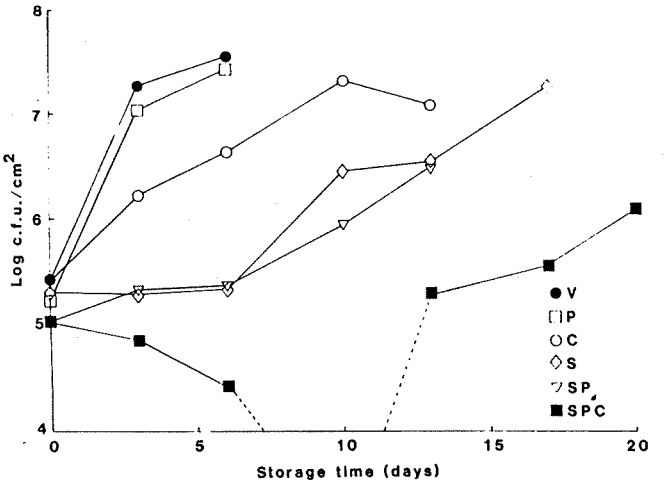

Fig. 2—Estimated bacterial numbers on morwong fillets stored at 4°C. (See Table 1 for code.)

The pH of fish in all packs on day 0 was between 6.8 and 6.2. Dipping in polyphosphate lowered the pH by near 0.4 units. The pH of fish from treatments V, C, and P increased rapidly as spoilage progressed, while those of fish in treatment S remained constant during the storage period.

Bacterial numbers

Changes in the estimated numbers of bacteria after each treatment are shown in Fig. 2. Rapid increases in bacterial numbers occured in treatments P and V, reaching around 5×10^7 cfu/cm^2 after 6 days. Bacterial numbers in these treatments did not differ significantly ($p<0.05$), showing that polyphosphate alone had no significant effect on the bacterial flora. Fish stored in carbon dioxide showed a slower increase in bacterial numbers, reaching around 4×10^7 cfu/cm^2 after 10 days. Treatments S and SP gave a lag period of 6 days, followed by a rise to around 5×10^6 cfu/cm^2 after 13 days. The bacterial numbers in these two treatments differed significantly ($p<0.05$) only initially and at 10 days. Polyphosphate therefore had little or no effect on the bacterial flora when used in conjunction with potassium sorbate. Treatment SPC had an initial bactericidal effect, with bacterial numbers being reduced to below the minimum level of detection (1000 cfu/g) during the first 10 days. By 13 days bacterial numbers had risen to around 2.5×10^5 cfu/cm^2 and continued to rise slowly during storage, reaching 10^6 cfu//cm^2 at the end of the storage period.

Bacterial flora

The bacterial flora of untreated morwong was composed of *Moraxella* species (60%) and *Alteromonas putrefaciens* (30%) (Table 2). *Lactobacillus* were present at low levels (5%). After 6 days in vacuum packs the proportion of *A. putrefaciens* increased to 45%. Fifty percent of the colonies selected showed only limited growth and could not be cultured for isolation and identification. *Vibrio/Aeromonas* species were present at a low level (5%). Addition of 100% CO_2 caused the development of a flora composed mainly of *Vibrio/Aeromonas* species. *Brochothrix thermosphacta* comprised 65–75% of the flora in treatments SP and SPC. The pH (6.4) and the low glucose levels in fish flesh may have given *B. thermosphacta* a competitive advantage over the *Lactobacillus* species.

Appearance and odor of raw fish

The only noticeable changes in the appearance of the fish in any of the treatments was a slight opacity of fish in treatment SPC after 3 days. This changed to a bleached appearance later in storage. No significant volumes of drip were noticed in any treatment. The changes in the odor of raw fish are listed in Table 3.

Profile panels

The profile results for the seven sessions at which fresh/frozen fillets were tasted have been consolidated (Fig. 3a, 4a). The major attributes contributing to the odor profile of the fresh/frozen morwong were sweet, shellfish, seaweedy and metallic with traces of starchy, boiled clothes, cardboard, wet feathers, musty, rubbery, pungent and putrid. For flavor the attributes of sweet, cardboard, chicken, salty and buttery were important which, with the exception of cardboardy, could be considered as desirable attributes. Odor intensity scores for all the treatments were uninformative and have been deleted from the profile results. The fish in treatment V deteriorated rapidly, undesirable off odors and off flavors were evident within 6 days of storage (Fig. 3b, 4b). The ammoniacal odor and flavor, the sour pungent acrid odors and sour, bitter, sulfide flavors that developed are typical for fish spoiled by *A. putrefaciens*. Fillets in treatment SP spoilt less rapidly than those in treatment V, off odors and off flavors increased, the desirable buttery flavor declined and astringent soapy flavors formed (Fig. 3c, 4c). There were few consistent changes in the odor profiles for the fish in the treatment SPC but pungent, acrid and putrid notes were quite noticeable after 20 days storage (Fig. 3d, 3e). The off flavor intensity increased and the desirable flavor characteristics such as sweet, buttery, chicken and broiled potato decreased while rancid, sour and bitter notes increased (Fig. 4d, 4e). Some of these changes may in part be due to autolysis and it seems unlikely that the rancid note would have arisen from oxidation but is more probably a result of lipolysis.

Texture

Very few significant textural changes were detected in or between treatments F, SP and SPC. Treatment V showed the most marked changes, becoming more tender, less succulent, less firm, less springy and less fibrous (Fig. 5). These changes may have resulted from the effect of increasing pH (Fig. 1) on protein structure (Love et al., 1979) or from bacterial proteolysis (Shewan, 1974). The texture of treatments SPC and F remained similar throughout the 20 days of storage.

Odor, flavor and overall acceptability

The results for odor and flavor correlated highly with those for overall acceptability and only the latter has been plotted (Fig. 6). After 6 days in storage the scores for treatment V were significantly lower ($p<0.05$) than scores for the frozen

Table 2—Changes in bacterial flora on morwong stored at 4 C Genera present, % of total

Storage time (Days)	Treatment	*Moraxella*	*A. putrefaciens*	*Vibrio/ Aeromonas*	*Enterobacteriaceae*	*Pseudomonas*	*Lactobacillus*	*B. thermosphacta*	Unidentified
0	V-Vacuum packaged	60	30	—	—	—	5	—	5
6	V-Vacuum packaged	—	45	5	—	—	—	—	50 no growth after isolation
13	SP-Polyphosphate + K-sorbate dip, vacuum packaged	5	—	5	5	—	10	75	—
	C-Packaged in 100% CO_2	—	—	90	—	10	—	—	—
20	SPC-Polyphosphate + K-sorbate dip, packaged in 100% CO_2	—	—	5	5	—	20	65	5

Table 3—Odor of raw fish
Storage time (days) at 4°C

Treatment*	3	6	10	13	17	20
V	slightly sour, fishy	pungent, rotting seaweed	—	—	—	—
C	fresh	sour	sour	sour, sickly		
P	fresh	sour, unacceptable	—	—	—	—
S	fresh	bland, slightly floury	very slightly spoiled	slightly stale, musty	smelly	—
SP	shellfish, sweet scallops	fresh seaweed, slightly marinated	slightly spoiled	sour	—	—
SPC	shellfish, mussels, seaweed	fresh seaweed	seaweed, fishy	fresh shellfish	slightly fishy	seaweed, not spoiled

* See Table 1 for treatment codes.

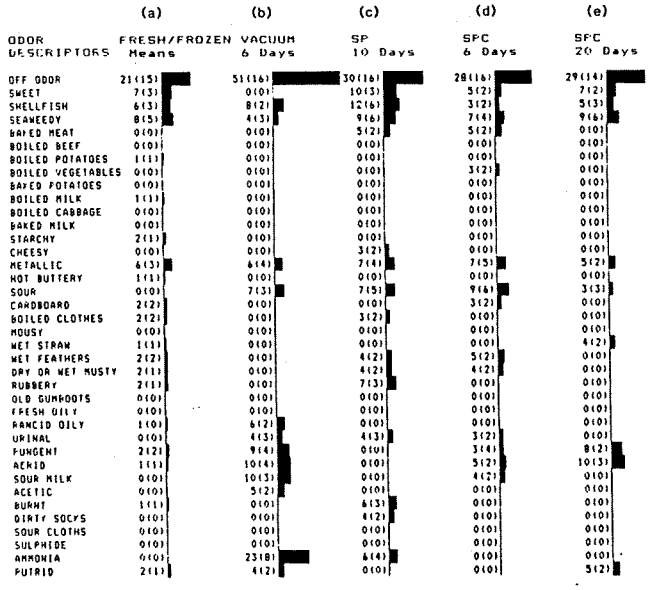

Fig. 3—Odor profiles of stored morwong: (a) fresh/frozen, (b) treatment V after 6 days, (c) treatment SP after 10 days, (d) treatment SPC after 6 days, (e) treatment SPC after 20 days. Total panel scores (0–100 scale) for each descriptor are shown, followed in parentheses by the number of panelists who marked that particular descriptor. In (a) the mean of all the sessions is shown to the nearest integer value.

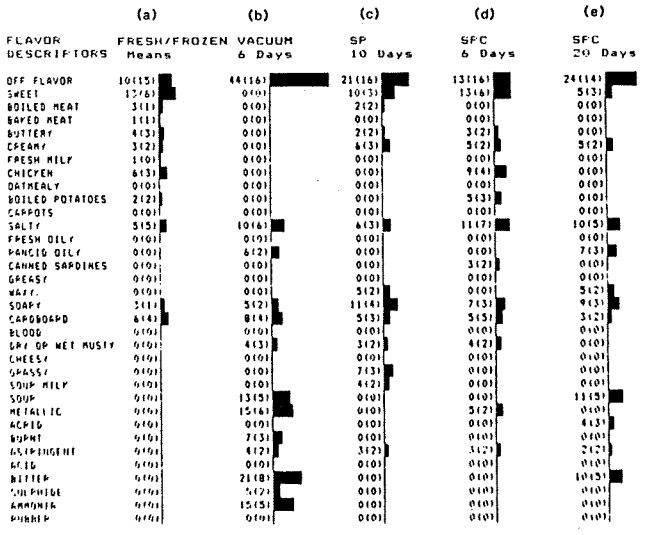

Fig. 4—Flavor profiles of stored morwong: (a) fresh/frozen, (b) treatment V after 6 days, (c) treatment SP after 10 days, (d) treatment SPC after 6 days, (e) treatment SPC after 20 days. Total panel scores (0–100 scale) for each descriptor are shown, followed in parentheses by the number of panelists who marked that particular descriptor. In (a) the mean of all the sessions is shown to the nearest integer value.

material. Flavor and overall acceptability scores for treatment SP were significantly lower than those for the frozen material after 10 days storage. Fish in treatment SPC remained as acceptable as the frozen fish for 13 days after which scores were significantly lower.

It could be suggested that the fish at the start of the experiment was of low quality even though it was supposedly chilled and obtained as soon as practical after catching. Experience

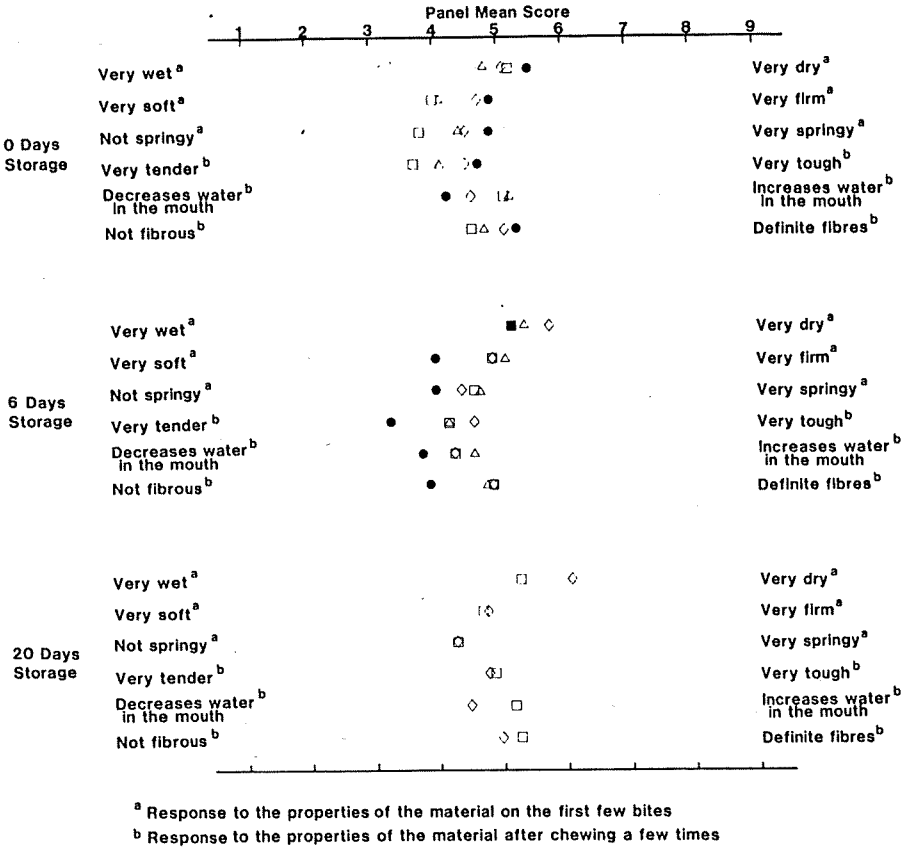

Fig. 5—Texture profiles of morwong stored for 0, 6 and 20 days; △ = SP, □ = SPC, ◇ = F, ● = V. (See Table 1 for Code).

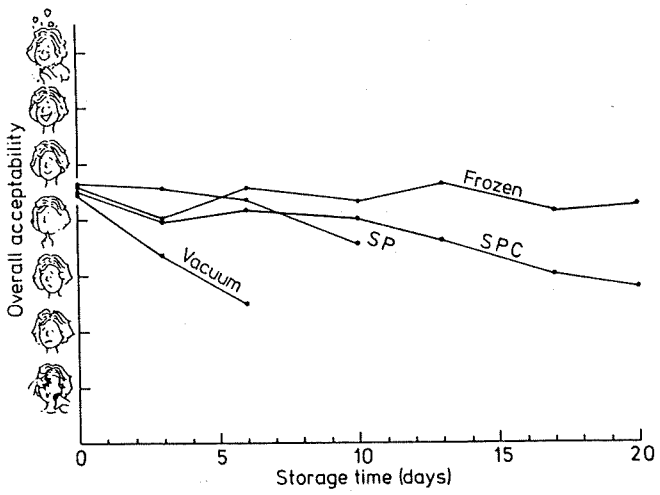

Fig. 6—Acceptability of stored morwong.

with these profile panels using this scoring method has shown that fairly low ratings are invariably given to plain cooked fish. The results for the frozen fish indicate the panel was consistent in its judgement.

DISCUSSION

THE COMBINED ACTION of CO_2 and sorbate (with polyphosphate) had an inhibitory effect on the bacteria (Fig. 2). Nevertheless, the fish still deteriorated steadily as indicated by increases in off odor and off flavor, decreased acceptability and, to a lesser extent, by the profile results (Fig. 3,4). The surviving bacterial flora was dominated by *Lactobacillus* and *B. thermosphacta*; genera which are regarded as having a low spoilage potential. Low numbers of *Vibrio/Aeromonas* also survived and it is likely that these organisms were a cause of the steady deterioration. *Vibrio* are well known to be spoilage organisms on other seafoods.

In addition, it is also possible that other spoilage organisms could have been present in numbers too low to be detected by the methods employed here which entail using high dilutions to enumerate 'total plate counts' on a non-specific medium. When the majority of the normal spoilage organisms have been suppressed and the shelf-life has been extended then even relatively low numbers of surviving spoilage organisms have the time and opportunity to produce significant amounts of deleterious byproducts which accumulate in the pack and migrate into the flesh. To enumerate low numbers of spoilage organisms would require the enormous workload involved with sequential sampling using a wide range of dilutions and a wide variety of selective media. Other mechanisms that may also have time to express their influence are degradation due to inherent autolytic or catheptic processes and bacterial enzymes released from cells no longer viable. When bacteria are killed by processes that do not denature their enzymes then the enzymes can remain active even though cell numbers are in decline.

Any or all of these mechanisms may account for the steady deterioration of the fish in treatment SPC. All of them place emphasis on the conventional wisdom of the need to obtain and package fish in as fresh a condition as possible. Given these circumstances the treatment of fillets with potassium sorbate, CO_2 and polyphosphate effectively extended the shelf-life of morwong fillets. Processors and regulatory authorities should determine if such treatment is safe and worthwhile on a commercial basis.

SUMMARY

THE VARIOUS COMBINATIONS of potassium sorbate, polyphosphate and CO_2 had markedly different effects on the keeping quality of morwong. The efficacy of the treatments used was: V = P <C <S = SP <SPC as indicated by changes

—Continued on page 1587

in pH, raw odor and bacterial numbers and confirmed by taste panel results for treatments V, SP and SPC.

The use of polyphosphate, either by itself or in combination with sorbate, had no effect on bacterial growth.

REFERENCES

AOAC. 1980. "Official Methods of Analysis." 13th ed. Association of Official Analytical Chemists, Washington, DC.

Bremner, H.A., and Statham, J.A. 1983. Effect of potassium sorbate on refrigerated storage of vacuum packed scallops. J. Food Sc. 48: 1042.

Buchanan, R.E. and Gibbons, N.E. 1974. "Bergey's Manual of Determinative Bacteriology". 8th ed. Williams and Wilkins Co. Baltimore, MD.

Cann, D.C. 1984. Packaging fish in a modified atmosphere. Torry Advisory Note No. 88. Torry Research Station, Aberdeen, Scotland.

Elliot, P.H., and Gray, R.J.H. 1981. *Salmonella* sensitivity in a sorbate/modified atmosphere combination system. J. Food Prot. 44: 903.

Elliot, P.H., Tomlins, R.I., and Gray, R.J.H. 1982. Inhibition and inactivation of *Staphylococcus aureus* in a sorbate/modified atmosphere combination system. J. Food Prot. 45: 1112.

Elliot, R.P., Straka, R.P., and Garibaldi, J.A. 1964. Polyphosphate inhibition of growth of pseudomonads from poultry meat. Appl. Microbiol. 12: 517.

Fey, M.S., and Regenstein, J.M. 1982. Extending shelf-life of fresh wet red hake and salmon using CO_2-O_2 modified atmosphere and potassium sorbate ice at 1°C. J. Food Sci. 47: 1048.

Hendrie, M.S., and Shewan, J.M. 1979. The identification of Pseudomonads. In "Identification Methods for Microbiologists," p.l, F.A. Skinner and D.W. Lovelock (Ed.) Academic Press, London.

Howgate, P. 1977. Aspects of fish texture. In "Sensory Properties of Foods," p. 249, J.G. Brennan and K.J. Parker (Ed.) Applied Science Publishers Ltd., London.

Love, R.M., Ratkowsky, D.A., Lowry, R.K., and Olley, J. 1979. Quoted in Olley, J. 1979. Structure and proteins of fish and shellfish. Part 1. "Advances in Fish Science and Technology," p. 65. Torry Research Station Jubilee Conference, Fishing News Books Ltd. Farnham. London.

Mills, A., and Tiffney, P. 1982. The use of controlled atmosphere packaging for the storage of fish. Chilled Foods 1(4): 22.

Quarmby, A.R., Bremner, H.A., and Thrower, S.J. 1982. On-board handling of gemfish Part II: Sensory profiles. Aust. Fish. 41(11): 42.

Regenstein, J.M. 1982. The shelf-life extension of haddock in carbon dioxide-oxygen atmospheres with and without potassium sorbate. J. Food Qual. 5: 285.

Shewan, J.M. 1974. The biodeterioration of certain proteinaceous foodstuffs at chill temperatures. In "Industrial Aspects of Biochemistry", p. 475. Federation of European Biochemical Societies. North-Holland Pub. Co., Amsterdam.

Spencer, J.V. and Smith, L.E. 1962. The effect of chilling chicken fryers in a solution of polyphosphates upon moisture uptake, microbial spoilage, tenderness, juiciness and flavor. Poult. Sci. 41: 1685.

Statham, J.A. 1984. Modified atmosphere storage of fisheries products: the state of the art. Food Technol. Aust. 36: 233.

Statham, J.A., and Bremner, H.A. 1983. Effect of potassium sorbate on spoilage of blue grenadier (*Macruronus novaezelandiae*) as assessed by microbiology and sensory profiles. J. Food Prot. 46: 1084.

Statham, J.A. and Bremner, H.A. 1985. Acceptability of trevalla (*Hyperoglyphe porosa* Richardson) after storage in carbon dioxide. Food Technol. Aust. 37: 212.

Van Wazer, J.R. 1971. Chemistry of the phosphates and condensed phosphates. In "Symposium: Phosphates in Food Processing," p. 1. J.M. Deman and P. Melnychyn (Ed.). Avi Publishing Co., Westport, CT.

Wagner, M.K. and Busta, F.F. 1983. Effect of sodium and pyrophosphate in combination with sodium nitrite or sodium nitrite/potassium sorbate on *Clostridium botulinum* growth and toxin production in beef/pork frankfurter emulsions. J. Food Sci. 48: 990.

Wankowski, J.W.J. 1984. Provisional estimates of population parameters, standing stock and sustainable catch for school whiting, tiger flathead and jackass morwong in Eastern Bass Strait. Internal Report No. 68, Department of Conservtion, Forests and Lands, Marine Science Laboratories, Victoria, Australia.

Ms received 2/11/85; revised 6/17/85; accepted 6/27/85.

The authors thank Ms. Maria Ottenschlaeger for skilled technical assistance with the taste panels and sorbate analysis. This work was funded by a grant from the Fishing Industry Research Trust Account.

REPRINTED FROM 'FOOD TECHNOLOGY IN AUSTRALIA'
VOLUME 37 NO 8 PP 352-355

156

Bringing fish inspection into the computer age

A.C. BRANCH and A.M.A. VAIL

Inspectors in the fishing industry generally acquire their skills through experience rather than systematic training. A prototype pocket sized computer described here provides a systematic basis for collecting information on fish quality attributes to assess the condition and likely shelf life of the catch. The technique can be used at any stage, from catching to market place or beyond. The advantages for experienced inspectors are ease of use and confidence in the results because of the uniformity of the approach to assessment. These factors also mean that reliable results can be obtained by the novice. Furthermore the information programmed into the computer can assist decision-making where the differences between grades are borderline. The program was derived from sensory score sheets for the assessment of fish quality developed at the CSIRO Division of Food Research, Tasmanian Food Research Unit, Hobart.

The benefits of ensuring the quality control of fish harvesting have been discussed by Bremner (1984) and relate directly or indirectly to increased productivity for both the fisherman and intermediaries in the fishing industry. Psychologically, simple but effective methods for assessing the result of quality control measures will be more acceptable than more complex or less effective methods.

Quantitative analysis of metabolites formed during the storage of fish (determination of K-value) is considered to be effective but is a time consuming and costly approach (Saito, Arai & Yajima 1959, Tomioka & Endo 1984). The setting up and operation of formalised taste panels are also time consuming and costly. Of considerable interest is the non-destructive process of allocating a rating for a number of easily identifiable features of stored fish. If the selection of these features identifies attributes which change as the fish deteriorates, then these features can be called quality attributes. If each feature or quality attribute is rated after simple examination, the resultant total of these ratings may be called a quality score.

Background

A practical rating system has been developed at the CSIRO Division of Food Research, Tasmanian Food Research Unit (TFRU) (Table 1) in which fish are inspected and the characteristics listed on the sheet are assessed and the appropriate demerit point score is recorded. The scores for the separate characteristics are summed to give an overall sensory score. This system gives scores of zero (or near zero, see later) for very fresh fish while increasingly larger totals result as fish deteriorate. Thus scores increase as deterioration increases. There are several sensory methods for evaluating fish which use scales converse to those described here, i.e. ratings which *decrease* for decreased quality of the product. Such systems are used by the EEC (Anon. 1970) and de Zylva (1974).

The system described here is considered to be simpler to use in that perfect specimen of fish are not required to train inspectors in the appropriate standards since the scheme evaluates the presence or absence of obvious defects. The system was developed over a number of years with the aid of numerous colour photographs of spoiling fish. The attributes chosen for

Table 1. Sensory assessment score sheet

Fish identification		
Appearance		V.bright/Bright/Sl.dull/Dull 0 1 2 3
Skin		Firm/Soft 0 1
Scales		Firm/Sl.loose/Loose 0 1 2
Slime		Absent/Sl.slimy/Slimy/V.slimy 0 1 2 3
Stiffness		Pre-rigor/Rigor/Post-rigor 0 1 2
Eyes	Clarity	Clear/Sl.cloudy/Cloudy 0 1 2
	Shape	Normal/Sl.sunken/Sunken 0 1 2
	Iris	Visible/Not visible 0 1
	Blood	No blood/Sl.bloody/V.bloody 0 1 2
Gills	Colour	Characteristic /Sl.dark/V. dark /Sl.faded/V.faded 0 1 2
	Mucus	Absent/Moderate/Excessive 0 1 2
	Smell	Fresh oily, metallic, seaweed / Fishy/Stale/Spoilt 0 1 2 3
Belly	Discoloration	Absent/Detectable/Moderate/Excessive 0 1 2 3
	Firmness	Firm/Soft/Burst 0 1 2
Vent	Condition	Normal /Sl.break /Excessive /Exudes /opening 0 1 2
	Smell	Fresh/Neutral/Fishy/Spoilt 0 1 2 3
Belly cavity	Stains	Opalescent/Greyish/Yellow-brown 0 1 2
	Blood	Red/Dark red/Brown 0 1 2

Allan Branch is a Director of Branch & Associates Pty Ltd, High Technology Consultants, 49 Grove Road, Glenorchy, Tas. 7010 and Alex Vail is a Technical Officer, CSIRO Division of Food Research, Tasmanian Food Research Unit, Stowell Avenue, Hobart, Tas. 7000.

0015-6647/85/37/0352/04

Table 2. Program for pocket fish freshness computer

```
5       VAC
10      PRINT "IS THE FISH GUTTED? XXX";
15      X$ = KEY
17      IF X$ = "Y" THEN 90
18      IF X$ = "N" THEN 90
19      GOTO 15
90      GOSUB 900
100     FOR Z = 1 TO 5
105     PRINT CSR 4; "XXXXXX";
110     PRINT CSR 0; A$(Z);
115     IF Z = 1; PRINT CSR 7; "NCE";
117     IF Z = 5; PRINT CSR 7; "SS";
120     PRINT CSR 11; "X";
130     GOSUB 800
190     NEXT Z
200     FOR Z = 6 TO 9
210     PRINT CSR 0; "EYES — ";
220     PRINT A$(Z); "XXX";
230     GOSUB 800
290     NEXT Z
300     FOR Z = 10 TO 12
310     PRINT CSR 0; "GILLS — ";
320     PRINT A$(Z); "XXX";
330     GOSUB 800
390     NEXT Z
400     IF X$ = "N" THEN 500
410     FOR Z = 13 TO 14
420     PRINT CSR 0; "BELLY CAVITY —";
430     PRINT A$(Z); "XXXX";
440     GOSUB 800
490     NEXT Z: GO TO 700
500     FOR Z = 15 TO 16
510     PRINT CSR 0; "BELLY — ";
520     PRINT A$(Z);
525     IF Z = 15; PRINT "URATION XXX";
527     IF Z = 16; PRINT "SXX";
530     GOSUB 800
590     NEXT Z
600     FOR Z = 17 TO 18
610     PRINT CSR 0; "VENT — ";
620     PRINT A$(Z);
625     IF Z = 17; PRINT "ONXX";
627     IF Z = 18; PRINT "XXXX";
630     GOSUB 800
690     NEXT Z
700     PRINT CSR 0; "XXXXXXXXXXXX";
710     PRINT CSR 0; "SCORE = "T";
715     FOR P = 1 TO 200: NEXT P
720     PRINT CSR 0; "XXXXXXXXXXXX";
730     GO SUB 2000
790     END
800     GOTO 810
810     B$ = KEY
820     IF B$ "0" THEN 810
830     IF B$ "3" THEN 810
840     PRINT CSR 11; B$;
845     T = T + VAL (B$)
850     FOR W = 1 TO 100: NEXT W
860     RETURN
900     A$(1)  = "APPEARA"
910     A$(2)  = "SKIN"
920     A$(3)  = "SCALES"
930     A$(4)  = "SLIME"
940     A$(5)  = "STIFFNE"
950     A$(6)  = "CLARITY"
960     A$(7)  = "SHAPEXX"
970     A$(8)  = "IRISXXX"
980     A$(9)  = "BLOODXX"
990     A$(10) = "COLOUR"
1000    A$(11) = "MUCOUS"
1010    A$(12) = "SMELLX"
1020    A$(13) = "STAINS"
1030    A$(14) = "BLOOD"
1040    A$(15) = "DISCOLO"
1050    A$(16) = "FIRMNES"
1060    A$(17) = "CONDITI"
1070    A$(18) = "SMELL"
1090    RETURN
2000    IF T < 5 : PRINT CSR 0; "FISH QUALITY IS PRIME": END
2010    IF T < 18; PRINT CSR 0; "FISH QUALITY IS GOOD": END
2020    IF T < 24; PRINT CSR 0; "FISH QUALITY IS FAIR": END
2030    PRINT CSR 0 : "FISH QUALITY IS POOR"
2050    RETURN
```

X: Designates a space

listing are common to many other schemes and the individual choice of scores in Table 1 never exceed 3, so no attribute can unduly imbalance the score; untrained personnel are thus unlikely to give unduly discrepant answers. The method is easy to understand, simple and fast to use; it gives as good or better correlation with time of storage than sophisticated laboratory testing of chemical change, i.e. K-values, which are a measure of nucleotide breakdown (Bremner 1984). This paper describes a computerised version of the sensory score sheet which may encourage a more enthusiastic attitude towards quality assessment.

Computer software

The scheme listed in Table 1 was suitable for programming into a computer and there were obvious advantages in using a pocket sized portable computer. Accordingly the software was written for a Casio PB100 pocket computer equipped with 1.5K memory (Table 2). The program prompts the user with specific questions for each attribute to which the user keys in the appropriate integer number as answer. No computer experience is necessary. The responses are totalled to give an overall sensory score.

A keyboard overlay based on Table 1 has been designed that lists quality attribute descriptors, score ranges and other user instructions. It is expected that this version of the program will be modified considerably as knowledge of user requirements is increased by field trial experience. The portable hand held computer in its waterproof case is shown in Figure 1. A mini thermal printer is available, giving the user a hardcopy of the results if required. The computer can be programmed to use the sensory score for grading the fish. In Table 2 the fish was arbitrarily graded as follows: <5 prime quality, 6–18 good quality, 19–24 fair quality, >24 poor.

Demerit points for a temperate water species

Figure 2 shows the sensory score SS as demerit points for spotty

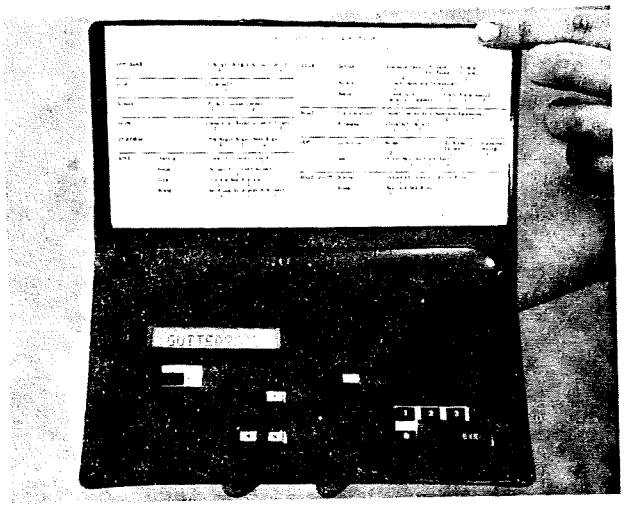

Figure 1. Fish freshness computer

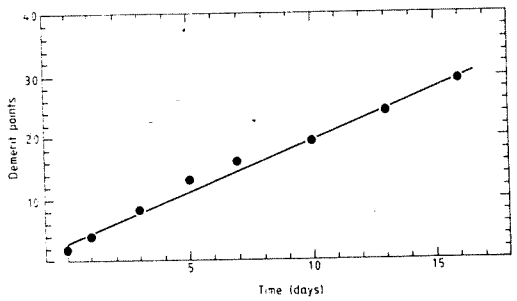

Figure 2. Changes in demerit points when spotty trevalla is stored on ice

Table 3. Attributes contributing to total sensory score for spotty trevalla

Demerit points	Storage time (days)	Quality	Attribute involved in score change
< 5	2	Prime	Rigor; slime on body and gills
6–18	7	Good	As above plus appearance; scales; stiffness; eyes — clarity, shape and iris; gills — colour, mucus and smell; vent — condition and smell
18–24	13	Fair	As above plus eyes — shape; gills — colour and smell; vent — smell
> 24	14	Poor	As above plus appearance; slime; gills — mucus; belly — discoloration and firmness; vent — condition and smell

Table 4. Effect of temperature on demerit point daily changes and K–value slope for four tropical species

Species	Change in demerit points/day* 24°–26°C (a)	Ice (b)	a/b	Change in K–value/day† 24°–26°C (a)	Ice (b)	a/b
Nemipterus peronii	36.0	0.87	41	55.2	2.0	28
Argyrops spinifer	31.2	0.86	36	45.6	1.3	35
Plectorhynchus pictus	33.6	0.73	46	158.4‡	3.7	43
Lutjanus vittus	26.4	0.77	34	160.8‡	2.3	70

* *Data of Bremner, Statham and Sykes (1985)*
† *A.M.A. Vail and P. Kearney (unpublished results)*
‡ *Note that the value given is a rate: K-values cannot exceed 100%*

Table 5. Equivalent days on ice corresponding to the actual number of days of storage (ice time)

Temp (°C)	Time of storage at given temperature (days) 1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	Relative spoilage rate
0	1.0	2.0	3.0	4.0	5.0	6.0	7.0	8.0	9.0	10.0	11.0	12.0	13.0	14.0	15.0	1.00
1	1.2	2.4	3.6	4.8	6.0	7.2	8.4	9.6	10.8	12.0	13.2	14.4	15.6	16.8	18.0	1.20
2	1.4	2.8	4.2	5.6	7.0	8.4	9.8	11.2	12.6	14.0	15.4	16.8	18.2	19.6	21.0	1.40
3	1.7	3.3	5.0	6.6	8.3	9.9	11.6	13.2	14.9	16.5	18.2	19.8	21.5	23.1	24.8	1.65
4	1.9	3.8	5.7	7.6	9.5	11.4	13.3	15.2	17.1	19.0	20.9	22.8	24.7	26.6	28.5	1.90
5	2.2	4.4	6.6	8.8	11.0	13.2	15.4	17.6	19.8	22.0	24.2	26.4	28.6	30.8	33.0	2.20
6	2.5	5.0	7.5	10.0	12.5	15.0	17.5	20.0	22.5	25.0	27.5	30.0	32.5	35.0	37.5	2.50
7	2.8	5.6	8.4	11.2	14.0	16.8	19.6	22.4	25.2	28.0	30.8	33.6	36.4	39.2	42.0	2.80
8	3.2	6.4	9.6	12.8	16.0	19.2	22.4	25.6	28.8	32.0	35.2	38.4	41.6	44.8	48.0	3.20
9	3.6	7.2	10.8	14.4	18.0	21.6	25.2	28.8	32.4	36.0	39.6	43.2	46.8	50.4	54.0	3.60
10	4.0	8.0	12.0	16.0	20.0	24.0	28.0	32.0	36.0	40.0	44.0	48.0	52.0	56.0	60.0	4.00
11	4.5	9.0	13.5	18.0	22.5	27.0	31.5	36.0	40.5	45.0	49.5	54.0	58.5	63.0	67.5	4.50
12	5.0	10.0	15.0	20.0	25.0	30.0	35.0	40.0	45.0	50.0	55.0	60.0	65.0	70.0	75.0	5.00
13	5.5	11.0	16.5	22.0	27.5	33.0	38.5	44.0	49.5	55.0	60.5	66.0	71.5	77.0	82.5	5.50
14	6.0	12.0	18.0	24.0	30.0	36.0	42.0	48.0	54.0	60.0	66.0	72.0	78.0	84.0	90.0	6.00
15	6.5	12.9	19.4	25.8	32.3	38.7	45.2	51.6	58.1	64.5	71.0	77.4	83.8	90.3	96.8	6.45
16	6.9	13.8	20.7	27.6	34.5	41.4	48.3	55.2	62.1	69.0	75.9	82.8	89.7	96.6	103.5	6.90
17	7.3	14.5	21.8	29.0	36.3	43.5	50.8	58.0	65.3	72.5	79.8	87.0	94.3	101.5	108.8	7.25
18	7.6	15.2	22.8	30.4	38.0	45.6	53.2	60.8	68.4	76.0	83.6	91.2	98.8	106.4	114.0	7.60
19	7.9	15.8	23.7	31.6	39.5	47.4	55.3	63.2	71.1	79.0	86.9	94.8	102.7	110.6	118.5	7.90
20	8.1	16.2	24.3	32.4	40.5	48.6	56.7	64.8	72.9	81.0	89.1	97.2	105.3	113.4	121.5	8.10

Table 6. Equivalent days on ice corresponding to the actual number of hours of storage

Temp (°C)	Time of storage at given temperature (h) 2	4	6	8	10	12	14	16	18	20	22	24	Relative spoilage rate
0	0.1	0.2	0.3	0.3	0.4	0.5	0.6	0.7	0.8	0.8	0.9	1.0	1.00
1	0.1	0.2	0.3	0.4	0.5	0.5	0.7	0.8	0.9	1.0	1.1	1.2	1.20
2	0.1	0.2	0.4	0.5	0.5	0.7	0.8	0.9	1.1	1.2	1.3	1.4	1.40
3	0.1	0.3	0.4	0.6	0.7	0.8	1.0	1.1	1.2	1.4	1.5	1.7	1.65
4	0.2	0.3	0.5	0.6	0.8	1.0	1.1	1.3	1.4	1.6	1.7	1.9	1.90
5	0.2	0.4	0.6	0.7	0.9	1.1	1.3	1.5	1.7	1.8	2.0	2.2	2.20
6	0.2	0.4	0.6	0.8	1.0	1.3	1.5	1.7	1.9	2.1	2.3	2.5	2.50
7	0.2	0.5	0.7	0.9	1.2	1.4	1.6	1.9	2.1	2.3	2.6	2.8	2.80
8	0.3	0.5	0.8	1.1	1.3	1.6	1.9	2.1	2.4	2.7	2.9	3.2	3.20
9	0.3	0.6	0.9	1.2	1.5	1.8	2.1	2.4	2.7	3.0	3.3	3.6	3.60
10	0.3	0.7	1.0	1.3	1.7	2.0	2.3	2.7	3.0	3.3	3.7	4.0	4.00
11	0.4	0.8	1.1	1.5	1.9	2.3	2.6	3.0	3.4	3.8	4.1	4.5	4.50
12	0.4	0.8	1.3	1.7	2.1	2.5	2.9	3.3	3.8	4.2	4.6	5.0	5.00
13	0.5	0.9	1.4	1.8	2.3	2.8	3.2	3.7	4.1	4.6	5.0	5.5	5.50
14	0.5	1.0	1.5	2.0	2.5	3.0	3.5	4.0	4.5	5.0	5.5	6.0	6.00
15	0.5	1.1	1.6	2.2	2.7	3.2	3.8	4.3	4.8	5.4	5.9	6.5	6.45
16	0.6	1.2	1.7	2.3	2.9	3.5	4.0	4.6	5.2	5.8	6.3	6.9	6.90
17	0.6	1.2	1.8	2.4	3.0	3.6	4.2	4.8	5.4	6.0	6.6	7.3	7.25
18	0.6	1.3	1.9	2.5	3.2	3.8	4.4	5.1	5.7	6.3	7.0	7.6	7.60
19	0.7	1.3	2.0	2.6	3.3	4.0	4.6	5.3	5.9	6.6	7.2	7.9	7.90
20	0.7	1.4	2.0	2.7	3.4	4.1	4.7	5.4	6.1	6.8	7.4	8.1	8.10

trevalla *(Seriolella punctata)* stored on ice. The rate of increase of demerit points is linear with storage time. The demerit points contributing to score and grade are shown in Table 3. The line of best fit accounted for 99% of the variance in the data and gave a slope of 1.65 demerit points per day and an intercept of 3 demerit points. When sufficient trials have been conducted it is envisaged that the operator can key in the appropriate slope for each particular species. However, present work indicates that reliable results can probably be obtained from just two factors, one for fish from temperate waters and the other for tropical

s es.

Some fish even when very fresh do not yield a zero score because of some inherent characteristics. The stargazer (*Kathetostoma* sp.) for example is naturally very slimy which would result in a zero time intercept of three demerit points.

Demerit points over a wide range of temperatures

So far the demerit point concept has been discussed for fish held on ice. In practice fish may be subjected to delayed or inefficient icing. It is therefore important to know the relationship between the accumulation of demerit points of iced fish and the same species at other temperatures. The work of Bremner, Statham and Sykes (1985) provided a suitable example. An alternative method of measuring spoilage (K-values) was used for comparison. The demerit points and K-values both at ambient (24°–26°C) and on ice were obtained for four tropical species from the North-West shelf. The change in demerit points per day together with the change in K-values for these species are shown in Table 4. The relative rates of spoilage, as measured by demerit points, at ambient and in ice storage were respectively 34 to 46 with a mean of 40 (S.E. ± 2.7). K-value relative rates ranged between 28 and 70 with a mean of 44 (S.E. ± 9.2). This indicates that demerit points can be used for time-temperature integration and are more useful than K-values, which have a more variable response to temperature changes, for predicting remaining shelf-life after storage at various temperatures.

Calculation of remaining shelf-life

A fish merchant may want to know how long his purchase will remain saleable if the fish are stored on ice immediately. A buyer at a fish market might be interested in the equivalent number of days on ice for which the fish has been stored since it was caught and thus how much marketable time on ice is left. All of these condition indicators can be extracted for a fish sample with a known rate of change of demerit points with the present sensory method. When the post mortem history of the fish is unknown, demerit points can be used to evaluate the normalised time-temperature data for the sample, (equivalent number of days on ice); if 'icetime' is calculated in terms of days on ice to reach the maximum score. For example, spotty trevalla having 22 demerit points represents an icetime of 12 days. The fish would have been caught 12 days ago if it had been stored on ice from the time it was caught. From Table 5 which shows the equivalent storage time on ice for a range of times and temperatures it can be seen that an equal number of demerit points would be obtained from 2 days at 14°C, 3 days at 10°C or approximately 10 days at 1°C. In practice, fish are either warming or cooling and Table 6 which lists icetimes in hours may be more appropriate. Tables 5 and 6 are taken from Bremner, Olley and Thrower (1978) and are derived from the relative rate curve of Olley and Ratkowsky (1973).

The icetime tables were developed as a descriptor of the relative rates of psychrotrophic spoilage between 0° and 20°C. In the temperature range 24°–26°C the relative rate of psychrotrophic spoilage would be expected to be approximately 12.25 (c.f. Pooni & Mead 1984). The values ranging from 34–46 in Table 3 indicate markedly increased rates of spoilage due to autolysis and presumably to mesophilic organisms. Despite this a linear accumulation of demerit points was still obtained for tropical species (Bremner, Statham & Sykes 1985).

Species variability of shelf life in ice

In its present form the 'fish freshness computer' provides the user with a number which is equivalent to the sum of individual demerit points and an arbitrary grading into 'prime condition', 'good condition', 'fair condition' and 'poor condition'. Inspectors or processors may wish to alter the cut off points for grading to suit their individual requirements. Plastic colour-coded tags corresponding to the four quality grades could be used by the fisheries inspectors to identify individual sample lots. Storage (shelf) life of fish species vary (Lima dos Santos 1981) and while the prototype computer is programmed for fish from temperate waters there is no difficulty in introducing scaling factors for tropical species which often have a longer shelf life, or for very fatty species which have a shorter shelf life.

Summary

This paper describes a method for recording and utilising data obtained from the sensory analysis of fish. It is stressed that the electronic device outlined here is a prototype. Before a dedicated field version can be designed, more data on the storage life of Australian temperate and tropical fish are needed.

Acknowledgement

This work was partly funded by the Fishing Industry Research Trust Account.

References

Anon. Fixing common marketing standards for certain fish, fresh or chilled. Off.J. of the European Communities L Series: Legislation 264: 1–11; 1970.

Bremner, H.A. Quality — an attitude of mind. Proceedings of the Seminar on The Australian fishing industry — today and tomorrow. Launceston, 10–12 July 1984. Launceston: Australian Maritime College; 224–69; 1984.

Bremner, H.A., Olley, J. & Thrower, S.J. CSIRO Tasmanian Regional Laboratory. Occasional Paper No. 4; 1978.

Bremner, H.A., Statham, J.A. & Sykes, S.J. Tropical species from the North-West shelf of Australia. Sensory assessment and acceptability of fish stored on ice. Proceedings of the 6th Session of the IPFC Working Party on Fish Technology and Marketing, Melbourne, 22–26 October 1984. FAO Fisheries Report No. 252 Supplement 1: 1985.

de Zylva, E.R.A. Effect of fish handling at sea on storage quality of New Zealand snapper (*Chrysophrys auratus*). N.Z. J. Sci. 17: 309–18; 1974.

Lima dos Santos, C.A.M. The storage of tropical fish in ice — a review. J. Trop. Sci. 23: 97–127; 1981.

Olley, J. & Ratkowsky, D.A. Temperature function integration and its importance in the storage and distribution of flesh foods above the freezing point. Food Technol. Aust. 25: 66–73; 1973.

Pooni, G.S. & Mead, G.C. Prospective use of temperature function integration for predicting the shelf-life of non-frozen poultry meat products. Food Microbiol. 1: 67–78. 1984.

Saito, T., Arai, K. & Yajima, T. Changes in purine nucleotides of red lateral muscle of rainbow trout. Nature, Lond. 184: 1415; 1959.

Tomioka, K. & Endo, K. K-value — increasing rates and IMP-degrading activities in various fish muscles. Bull. Jap. Soc. Sci. Fish. 50: 889–92; 1984.

APPENDIX 7

HIGHLIGHTS OF THE WORK

1. Factors limiting the shelf-life of different chilled fish species have become much more clearly defined.
2. The controversy as to whether tropical fish keep longer on ice than temperate water species has been partially explained.
3. The limits to storage life of chilled Australian seafoods packaged under modified atmospheres or with K-sorbate can now be predicted and the botulism risk assessed.
4. Sensory analysis of seafoods has been studied in depth and a commercial computerised scoring system based on demerit points is now being marketed. This is particularly valuable for iced tropical species where the rapidly melting ice leaches out the usual water soluble chemical compounds used to monitor spoilage.
5. The cold-shortening phenomenon in tropical species when rapidly chilled needs further study particularly if the trapping of tropical fish increases in importance.
6. The effects on the freezing rate of prepackaging commodities in commercial fibre-board packs prior to freezing can now be predicted.

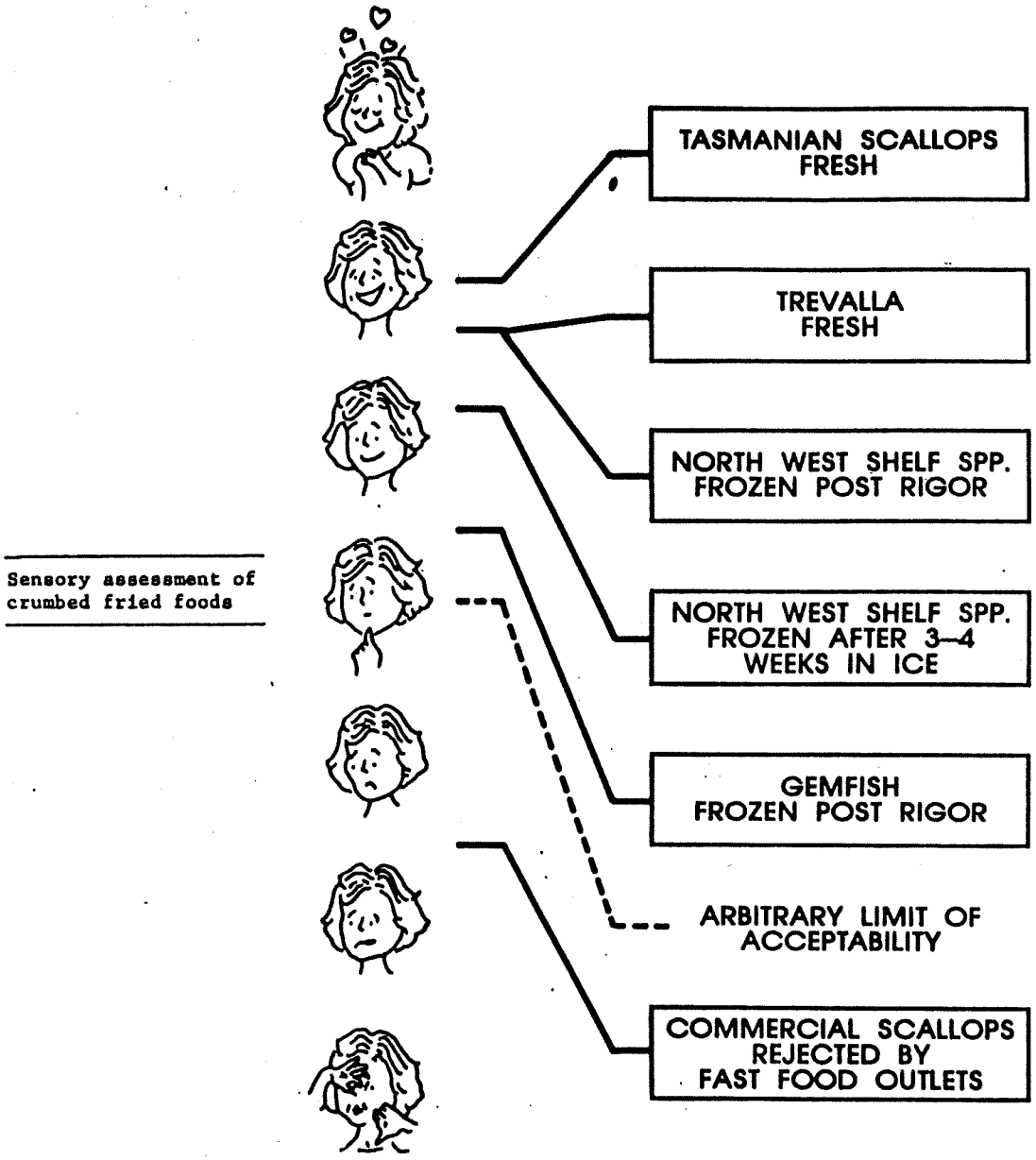
Sensory assessment of crumbed fried foods
TASMANIAN SCALLOPS FRESH
TREVALLA FRESH
NORTH WEST SHELF SPP. FROZEN POST RIGOR
NORTH WEST SHELF SPP. FROZEN AFTER 3–4 WEEKS IN ICE
GEMFISH FROZEN POST RIGOR
ARBITRARY LIMIT OF ACCEPTABILITY
COMMERCIAL SCALLOPS REJECTED BY FAST FOOD OUTLETS

Figure 1