Microbial Stability of Certain Cured Fishery Products

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The microbial stability of cured fishery products processed by improved curing and packaging methods was studied. Improved drying methods resulted in lower microbial counts than trade samples. Slight to strong brown discolouration was noticed during storage in dried anchovy, *Stolephorus indicus*. The yellow discolouration of dried salted seer appeared to be checked by BHA treatment. BHA treatment in combination with a dip in 3 percent calcium propionate solution was effective in controlling fungal growth and yellow discolouration. Cooking and hot smoking of tuna, *Euthynnus affinis*, yielded a stable product with low moisture content, while the cold blanched, hot smoked fish spoiled rapidly. In pickled prawn, *Parapenaeopsis stylifera*, with condiments and pH level around 4.75, the microbial count was greatly reduced. In lactic fermented prawn pickle, there was a steep rise in staphylococci count initially which then decreased with the reduction in pH level.

In India, prevailing conditions favour the growth of fish curing industry to a great extent. It has been reported that about 69 per cent of the total fish catch is marketed as fresh and 16.7 percent is preseved by curing (Anon, 1986). The cured fishery products, a source of high quality dietary protein, is the least expensive source of animal protein available in India (Balachandran *et al*., 1978). The principles of fish drying (Waterman, 1976), salting (Zaitsev *et al*., 1969), smoking (FAO, 1970), enzyme hydrolysis and bacterial fermentation (Owens & Mendoza, 1985) have been reviewed in depth . Curing of fish by traditional methods have certain limitations. Curing is sometimes practised as a last resort on fish which is almost putrid. Dried and salted products are often sundried unhygienically on the sandy beach which yields poor quality product, contaminated with sand, dust and dirt. Uncontrolled growth of microbes in such cured products may lead to serious implications on the keeping quality and safety of the product. Various codes of practice and standards have been drawn up by FAO/WHO, and the "Recommended International Code of Practice - General Principles of Food Hygiene" as well as Codes of Practice of fresh, smoked and salted fish are all relevant (FAO/WHO Food Standards Programme Codex Alimentarius Commission). Keeping the above in view, we report the microbial stability of certain cured fishery products that have been preserved by improved curing and packaging.

Materials and Methods

All fishes used in this study were procured from Tuticorin Fishing Harbour immediately after landing in fresh condition. They were washed in running water, eviscerated (except anchovy), or peeled and deveined, washed again in running water and used for further processing.

Drying of anchovy was carried out as described below; dried fish was packed in HDPE woven bags lined with LDPE film, unless otherwise specified, and stored at ambient temperature (30 ± 1 °C).

A. Sundried: Sun drying by spreading the fish over a nylon wire meshed wooden rack (2.0 x 1.0 x 0.3 m) for 3 days.

B. Boiled and sundried: Boiling in seawater for 1 min, and dried as above.

C. Oven dried: Dried at 45°C for 15 h in an oven without air circulation.

D. Solar tent dried: Solar tent drier was fabricated as per Doe (1979) and dried by spreading the fish on a black Poly Vinyl Chloride sheet kept on a wire meshed rack for 3 days.

Sundried sardine(*Sardinella* spp.) was prepared by dipping it in seawater for 15 min and then dried for 3 days on a wire meshed wooden rack, packed in HDPE bags and stored at ambient temperature.

Salt curing of split opened seer was carried out as described below, placed in wooden box, having a velon screen opening and stored at ambient temperature.

A. Curing for 18 h at 1:3 salt to fish ratio and sundried for 48 h on a wire meshed wooden rack.

B. Curing as in 'A' followed by a dip in 3 percent calcium propionate solution for 10 min and dried as above.

C. Mixing salt with Butylated Hydroxy Anisole (BHA; 0.05% body weight of fish) and cured as in A.

D. 'C' followed by a dip in 3 percent calcium propionate solution for 10 min and dried as above.

All smoke curing operations were carried out in AFOS smoke mini kiln.

Smoked tuna (*Euthynnus affinis*) was prepared by hot blanching of tuna in 12 percent salt solution for 60 min at 96 ±1°C followed by sundrying on a wire meshed rack for 60 min and smoking at 70°C for 3 h. The smoked product was then dried until it became hard in texture, packed in HDPE bags and stored at ambient temperature.

Smoked red snapper (*Lutjanus malabaricus*) was prepared by cold blanching of fillets in 10 percent salt solution for 30 min at 32 ± 1 °C followed by smoking at 70°C for 2 h. The smoked fillets were then sundried for 24 h, packed in HDPE bags and stored at ambient temperature.

Smoked sardine (*Sardinella sirm*) was prepared by the method described for red snapper, but in whole condition.

Smoked prawns (*Metapenaeus* spp.) were prepared by the cold blanching of peeled and deveined prawns in 3 per cent salt solution for 30 min at 32 ± 1°C followed by smoking at 70°C for 2 h, sundrying for 24 h, packed in HDPE bags and stored.

Vinegar pickled prawn and lactic fermented prawn pickle were prepared as per the recipe and methodology developed by Abraham & Jeyachandran (1992) and Abraham (1988) respectively. Pickles were then packed in sterile glass bottles, covered with aluminium caps, sealed and stored.

The microbial quality of the cured fishery products were assessed by total plate count (TPC), total fungal count (TFC), Staphylococcal count (SC), total coliforms (TC) (APHA, 1976), Clostridial count (CC) and Anaerobic gas producers count (AnGC) (Coolins *et al.*, 1989).

Results and Discussion

The microbial quality of the cured fishery products are presented in Table 1 to 4. The products prepared by improved drying methods had lower microbial load than the trade anchovy wherein counts of the order of 10^4 to 10^6/g are common (Joseph *et al.*, 1983). However, the microbial load increased slightly at the end of storage (Table 1). The overall quality of the dried anchovy processed by various methods was judged in the following order; boiled and sundried > oven dried > solar tent dried > sun dried. The samples showed slight to strong brown discolouration towards the end of storage period.

Table 1. *Microbial quality of dried anchovies and sardine*

Product & treatment	Storage days	TPC g^{-1}	TC g^{-1} (MPN)	TFC g^{-1}
Stolephorus indicus	0	$3.00x10^3$	4.00	$3.30x10^1$
	30	$4.00x10^3$	13.00	$5.00x10^1$
A. Sundried	60	$4.80x10^3$	40.00	$7.67x10^1$
(15.91 & 16.71)	90	$5.70x10^3$	95.00	$1.13x10^2$
	120	$7.10x10^3$	140.00	$1.40x10^2$
B. Boiled &	0	$1.33x10^3$	<2.00	$1.30x10^1$
sundried	30	$1.47x10^3$	<2.00	$1.30x10^1$
(15.79 & 18.20)	60	$2.10x10^3$	<2.00	$3.33x10^1$
	90	$3.02x10^3$	<2.00	$8.00x10^1$
	120	$4.63x10^3$	<2.00	$1.00x10^2$
C. Oven dried	0	$1.83x10^3$	<2.00	$2.00x10^1$
(15.62 & 16.25)	30	$2.27x10^3$	4.60	$2.00x10^1$
	60	$2.67x10^3$	17.00	$6.00x10^1$
	90	$3.70x10^3$	94.00	$9.00x10^1$
	120	$5.00x10^3$	120.00	$1.16x10^2$
D. Solar tent dried	0	$2.83x10^3$	3.00	$2.67x10^1$
(15.31 & 16.23)	30	$3.73x10^3$	4.00	$5.33x10^1$
	60	$4.37x10^3$	34.00	$7.33x10^1$
	90	$5.27x10^3$	58.00	$9.67x10^1$
	120	$6.33x10^3$	120.00	$1.20x10^1$
Sun dried sardine	0	$4.35x10^4$	9.00	$4.40x10^2$
(*Sardinella* spp)	30	$5.90x10^4$	14.00	$5.20x10^1$
	60	$8.20x10^4$	20.00	$5.50x10^1$

Values in parenthesis are moisture content in percent immediately after preparation and at the end of storage

Table 2. *Microbial quality of salt cured seer fish (Scomberomorus commerson)*

Product code*	Storage days	TPC g^{-1}	TC g^{-1} (MPN)	TFC g^{-1}
A. (46.40 & 32.52)	0	$5.70x10^3$	<2.00	$2.00x10^1$
	30	$8.90x10^3$	<2.00	$2.10x10^3$
	60	$5.80x10^4$	<2.00	$1.20x10^4$
	90	ND	ND	ND
	120	ND	ND	ND
B. (44.96 & 22.62)	0	$4.40x10^3$	<2.00	$<1.00x10^1$
	30	$8.10x10^3$	<2.00	$<1.00x10^1$
	60	$3.20x10^4$	<2.00	$1.10x10^2$
	90	$8.70x10^4$	<2.00	$5.00x10^2$
	120	ND	ND	ND
C. (47.65 & 18.52)	0	$3.50x10^3$	<2.00	$<1.00x10^1$
	30	$6.70x10^3$	<2.00	$<1.00x10^1$
	60	$1.10x10^4$	<2.00	$3.50x10^1$
	90	$3.40x10^4$	<2.00	$2.60x10^2$
	120	$5.70x10^4$	<2.00	$4.10x10^2$
D. (46.36 & 18.92)	0	$3.70x10^3$	<2.00	$<1.00x10^1$
	30	$5.90x10^3$	<2.00	$<1.00x10^1$
	60	$8.50x10^3$	<2.00	$2.00x10^1$
	90	$3.10x10^4$	<2.00	$1.60x10^2$
	120	$6.70x10^4$	<2.00	$3.60x10^2$

* Refer Materials and Methods; ND - Not done: spoiled as a result of visible white fungal colonies on the surface of the fish and yellow discolouration.; Values in paranthesis are moisture content in percent immediately after preparation and at the end of storage.

Salted fish is less susceptible to insect attack but more susceptible to halophilic bacteria introduced by salt used in curing and halophilic mould and yeasts. The major deteriorative change observed in salted seer was yellow discolouration at the beginning of spoilage which spread further on the surface of fish with increase in storage period. (Table 2)

BHA treatment appeared to check yellow discolouration of the product to a great extent. However, a dip treatment in 3 percent calcium propionate solution in combination with BHA treatment was found to be more effective in controlling the fungal growth as well as yellow discolouration (Table 2). Sen & Sripathy (1967) recommended a preservative mixture containing BHA to retard brown discolouration in salt curing of mackerel.

Smoking in improved smoke kiln reduces the process time and final moisture

Table 3. *Microbial quality of smoke-cured tuna, red snapper, sardine and prawns*

Product	Storage	TPC g^{-1}	TC g^{-1}	TFC g^{-1}
Smoked tuna (Masmin)	0	$3.25x10^3$	21.00	$<1.00x10^1$
(12.25 & 8.00)	30	$3.60x10^3$	23.00	$<1.00x10^1$
	60	$4.10x10^3$	28.00	$<1.00x10^1$
	90	$6.15x10^3$	43.00	$1.50x10^1$
Smoked red snapper,	0	$1.50x10^4$	2.00	$<1.00x10^1$
(Lutjanus malabaricus)	30	$1.70x10^5$	3.00	$3.00x10^1$
(42.80 & 33.00)	60	$1.20x10^6$	24.00	$2.40x10^2$
Smoked sardine	0	$1.50x10^4$	2.00	$<1.00x10^1$
(Sardinella sirm)	30	$6.20x10^4$	15.00	$1.00x10^1$
(20.09 & 12.00)	60	$4.20x10^5$	15.00	$3.50x10^1$
Smoked prawns,	0	$4.00x10^4$	2.00	$<1.00x10^1$
(Metapenaeus spp.)	30	$3.60x10^4$	2.00	$3.00x10^1$
(62.54 & 59.61)	60	$8.40x10^4$	2.00	$4.50x10^1$
	90	$1.08x10^5$	6.00	$1.00x10^2$

Values in parenthesis are moisture content in percent immediately after preparation and at the end of storage.

content of the product than the traditional smoking. In the present study hot blanching and hot smoking yielded a stable product with low moisture content and microbial load, while the cold blanching and hot smoking yielded products with high moisture content and TPC, ranging from 33 to 62 percent and 10^4 to 10^6/g respectively (Table 3). These levels are well within the limits prescribed for cold smoked fish (ICMSF, 1986). The process of smoking fish is reported to impart a degree of microbiological stability to the product which is a function of reduced water activity, heating, smoking and dehydration (Eklund *et al.*, 1988).

Pickled products with a pH level around 4.50 are expected to have a shelflife of more than 6 months. Erichsen (1967) reported that the pickled fish normally carried microbes in the range of 10^1 to 10^3/g of meat unless spoiled. The results in Table 4 revealed that the counts were within the normal range for pickled prawn. Clostridial count and anaerobic gas producers count increased with increase in storage period. The growth of these organisms are normally favoured because of anaerobic condition maintained in the pickle and hence needs further investigations on their safety.

During the initial stage of fermentation, at near neutral pH, the plate counts of the order of 10^6 to 10^8 are common. Suppression of bacterial growth including

Table 4. *Microbial quality of pickled prawns*

Product	Storage days	TPC g^{-1}	SC g^{-1}	TFC g^{-1}	CC g^{-1} (MPN)	AnGC g^{-1} (MPN)
Vinegar pickled prawn	0	$1.55x10^3$	$1.00x10^1$	$<1.00x10^1$	<2.00	<2.00
(pH 4.79 & 4.75)	60	$1.00x10^2$	$<1.00x10^1$	$<1.00x10^1$	20.00	4.00
	120	$1.00x10^2$	$<1.00x10^1$	$1.50x10^1$	278.00	7.00
	180	$2.00x10^2$	$<1.00x10^1$	$2.00x10^1$	390.00	70.00
Lactic fermented prawn pickle	0	$3.7\text{-}0x10^7$	$1.00x10^2$	$1.00x10^2$	ND	ND
(pH 6.80 & 5.40)	30	$1.09x10^8$	$3.70x10^6$	$3.00x10^2$	ND	ND
	60	$1.34x10^6$	$4.00x10^3$	$2.50x10^2$	ND	ND

Values in parenthesis are end product pH immediately after preparation and at the end of storage
ND - not done

pathogens, if present, is expected with the reduction in pH levels. An increase in microbial count (TPC & SC) was observed upto 30 days, therafter the count decreased with reduction of pH in the product (Table 4).

The results of this study revealed that the fungal population was under control in almost all products and improved curing and handling could eliminate the problems of mycotoxin and/or mycotoxicosis. In summary, the improved preservation and protection methods resulted in microbiologically better cured fishery products than the traditional ones.

Authors are thankful to Dr. M. Devaraj, Dean, Fisheries College and Research Institute for encouragement and facilities provided.

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