Development and shelf life evaluation of Tilapia (*Oreochromis* spp.) marinades

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Abstract

Tilapia (*Oreochromis* spp.) consumption is limited due to its strong muddy odour and the difficulty of processing. In addition, consumption of tilapia is minimal in urban areas because of the low availability. There are no processed market products of tilapia available in Sri Lanka. Therefore, this study was designed to develop a new marinade for tilapia and to evaluate the shelf life of the product. Twelve different treatments of varying amounts of vinegar, salt, chili powder, white pepper and garlic powder were applied to filleted tilapia, and three best treatment combinations were selected using a sensory evaluation test. Processed tilapia was stored in the freezer at -4 °C.

Treated samples were subjected to evaluation of sensory profile: taste, odour, colour, texture and overall acceptability. Analysis of the shelf life was carried out by using the total plate count, faecal coliform test, acidity and pH at weekly intervals. Results revealed that the third treatment (vinegar 75 ml, salt 5 g, chili powder 5 g, white pepper 5 g and garlic powder 5 g) was best in terms of colour, texture, odour, taste and the overall acceptability according to the estimated medians (6, 6, 6 and 6.33 respectively). There was no significant difference between the first and the third treatment in terms of odour and overall acceptability. There was no significant difference between the three vacuum packed treatments for acidity and pH. Acidity and pH of the three treatments were at an acceptable level, which was below pH 5.3 and above 1.95% acidity. Average bacterial count was 10 colonies and 1.33×10^6 colonies respectively in vacuum packed treatments and bottled samples after one week. The acceptable level of bacterial colonies is 1.00×10^5. Vacuum packed treatments showed a one month shelf life. In conclusion, marinades can be developed from tilapia with a pleasant taste and acceptable texture.

Keywords: Tilapia marinade, vacuum packaging, sensory evaluation, shelf life

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Introduction

Tilapia is a member of the large family known as Cichlids. Of the many varieties of tilapia, the commonly cultivated species are *Oreochromis niloticus* and *Oreochromis mossambicus* (Pillay, 1990). Tilapia is a freshwater fish species, but can also live in brackish water and is found in tropical seas (Sugunan, 1997).

The large majority (over 96%) of Sri Lankans consume fresh or processed fish, which provide an estimated 65% of the mean annual animal protein intake (Sugunan, 1997) of the population. The demand for inland and marine fish, however, is highly demarcated. In poorer, rural inland areas, predominant demand is for the cheaper, fresh inland fish varieties. The estimated production of inland fishery in Sri Lanka is 20% of the total fish production. Of this, 80% is contributed by tilapia species (Sugunan, 1997). Tilapia is a highly nutritious food item providing 93 kcal of energy, 19.5 g of protein, 37 mg of sodium, 0.5 mg of iron, 1.0 g of total fat, 50 mg of cholesterol and 90 mg of omega-3 fatty acids per 100 g of fish (Anon, 2006a). It is therefore, an important source of animal protein for protein-limited inland communities in Sri Lanka.

Marinades are semi preserves; acid, usually acetic acid, and salt are added to the fish to retard the action of bacteria and enzymes, resulting in a product with a characteristic flavour and an extended but limited shelf life. The acetic acid in the form of vinegar produces the tenderness characteristic of marinades (Claucas, 1981). This is largely due to the action of some of the proteolytic enzymes that cause the partial breakdown of the protein and the release of some free amino acids which give the products its characteristic taste. The fat content of the flesh also contributes to the flavour. Some of the acetic acid combines chemically with the proteins, controls the pH and selectively allows the autolytic reactions to take place. Marinating also makes the flesh of the fish firmer in texture; the more salt that is added, the firmer the flesh becomes due to the removal of water and the coagulation of protein. The amounts of acid and salt required can be reduced if the product is kept frozen until consumed (Maclay, 2001).

Freezing is the easiest, most convenient, and least time-consuming method of preserving food. The extreme cold temperature slows the growth of microorganisms and the chemical changes that affect quality or cause spoilage. Freezing does not, however, sterilize food or destroy the organisms that cause spoilage. During freezing, enzyme action is slowed but does not stop. If not inactivated, these enzymes can cause colour and flavour changes and loss of nutrients even during storage in the freezer (Anon, 2006b).

Some bacteria and enzymes remain active in marinades throughout the storage, even in the presence of acid and salt, and eventually the flesh will dissolve completely. This residual action is desirable in some semi-preserves, for example, those in which salt alone or a mixture of salt and sugar are used to preserve the fish; the products of bacterial and enzymic action produce the typical flavour of the product (Borgstrom, 1989).

Marinades may be conveniently divided into three groups; cold marinades, cooked marinades and fried marinades. Cold marinades are preserved in a mixture of acetic acid and salt, without giving any heat treatment. In cooked marinades, fish are placed in a hot solution (85 °C solution of acetic acid and salt). Fried marinades are preserved by frying or baking before being packed in an acetic acid and salt solution. Marinades may be prepared from whole or dressed fishes (Merinodol, 1969). Normally, fish which have a high fat content is used for the preparation of marinated products (e.g. pelagic fish such as herrings and sardines). There are several marinated fish products, such as marinated herrings, mussels and milk fish, available in other counties (Maclay, 2001).

Eventhough there is comparatively a high yield of tilapia in inland areas of Sri Lanka, tilapia consumption is limited due to its strong fishy odour and the difficulty of processing. Moreover, though it is a very nutritious food, consumption of tilapia is minimal in urban areas because of the low availability. It would be appropriate, therefore, to develop a suitable novel product from tilapia as a ready to cook product, to increase its utilization (e.g. marinating). At present, there is no processed market product of tilapia available in Sri Lanka. This study was undertaken, therefore, to develop a new marinade of tilapia with a pleasant taste and medium texture, which is also safe to consume, over a reasonable period of storage. It is expected that such a product would popularize a relatively underutilized source of nutrition among the Sri Lankan population.

Materials and Methods

Tilapia (*Oreochromis* spp.) samples were obtained from the Magal Tank, Nikaweratiya and transported on ice to the laboratory. Ingredients (vinegar, table salt, white pepper, chili powder, garlic powder) used for product development were purchased locally.

In the laboratory, Tilapia fish were filleted and used for preparation of marinades. Initially, twelve treatments using varying combinations of vinegar, table salt, white pepper and chili powder were used to prepare the marinades. After storing for one weekand and being packed in glass bottles and kept in the freezer, they were subjected to sensory evaluation tests. To determine the most preferred treatments, a five point hedonic scale (5- Like very much, 1- Dislike very much) was used. The three most acceptable of the 12 treatments were selected for further studies (Table 1). Tilapia samples were subjected to these three treatments and the marinated products were bottled and vacuum packed and kept in freezers. Bottled products and vacuum packed products were subjected to sensory evaluation and shelf life analysis.

The three treatments were evaluated for sensory attributes with respect to colour, taste, texture, flavour and the overall acceptability according to Meilgaard *et al*., (1999). A panel of 25 semi-trained individuals and a modified 7-point hedonic scale were used to

evaluate the treatments (7-Like very much, 1-Dislike very much). Friedman test was used to analyze the results of the sensory evaluation using a computer statistical package (Minitab).

The shelf life of the vacuum packed and bottled marinades were evaluated by testing for acidity, pH, Total Plate Count of the product at weekly intervals; faecal coliform test were also carried out at the same time. Total Plate Count was done using nutritive agar media and the spread plate method (Garbutt, 1997). Samples from each treatment were analyzed in triplicate. Total fecal coliform test was done using the MPN technique (Garbutt, 1997).

Table 1. Different treatments applied for fish fillets (four fillets)

Treatments	**Quantity of ingredients**				
	Vinegar (ml)	Salt (g)	Chili powder (g)	White pepper (g)	Garlic powder (g)
First	37.5	5	5	-	2.5
Second	75	2.5	-	5	2.5
Third	75	5	5	5	2.5

Results and Discussion

The different treatments used in the three marinades for tilapia fish fillets are shown in Table 1. Results showed that the third treatment (vinegar 75 ml, salt 5 g, chili powder 5 g, white pepper 5 g and garlic powder 5 g) was best in terms of colour, texture, odour, taste and the overall acceptability according to the estimated median (6, 6, 6 and 6.33 respectively). There was no significant difference, however, between the first and the third treatments (Table 1) in terms of odour and the overall acceptability.

Sensory attributes

There was a significant difference between the second and the first treatment in terms of odour and the overall acceptability. When the acid and salt were combined, it gave a characteristic odour to the marinades. If there are relatively high acid levels compared to salt, the product has an unacceptable odour. The second treatment (without chili) was significantly different for odour and the overall acceptability from the other two treatments.

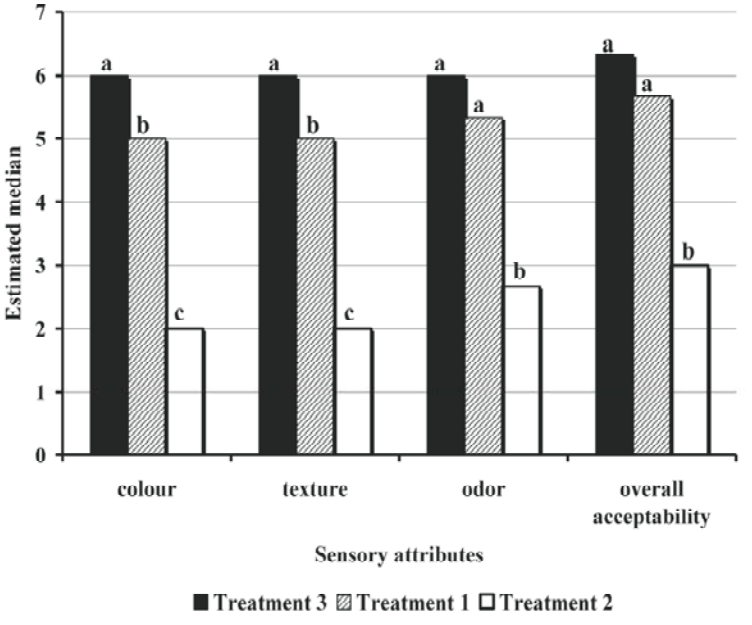

Fig. 1. Estimated median in three vacuum packed samples according to sensory attributes.

Note: Similar characters indicate no significant difference among samples with each attribute at $P<0.05$

Sensory attributes of the three different treatments are presented in Fig. 1. There was a significant difference between each of the three treatments in terms of colour and texture. Colour difference is due to additives such as chili powder, white pepper and garlic powder. The combined effect of salt and vinegar is quite different. Even a slightly acidic salt solution can make fish flesh noticeably firmer and can reduce swelling. There was a significant difference between the second and the third treatment in the texture; the likely reason being the high salt concentration though the same amount of acid was used. Conversely, addition of higher acidity levels with the same proportion of salt produces a firmer flesh (Merinodol, 1969). Thus, the first and the third treatment were significantly different with respect to texture.

Sensory evaluation was not carried out on the bottled samples due to their bad odour, cloudiness and high microbial count as a result of the spoilage of the marinades. The three main factors for spoilage of marinades are chemical factors, microbial factors and enzymatic action. Chemical action involves oxidation of fat contained within the fatty tissues of the fish. The colour and the viscocity of the fat changes and the marinades develop a strong taste. Microbial action involves bacterial decomposition of the fish flesh. This can be due to the low hygienic practices used during the processing of the product. Enzymatic action is due to the action of various enzymes found in the fish fillets. They spoil the tissue by the process of autolysis and make the fish susceptible to bacterial attack. It results in the formation of amino acids, ammonia, carbon dioxide and volatile basic compounds (Shammi and Bhatnagar, 2002) and gives a bad odour to the

product. In addition, high salt concentration causes protein solubilization and imparts cloudiness to the product.

Microbiological studies

Table 2. Microbiological changes of vacuumed packed marinades and bottled marinades

Parameter	Amount	
	Bottled	**Vacuum packed**
TPC	$1.33x10^6$ (three samples) (one week)	<30 colonies (three samples) (After 4^{th} week)
Faecal coliform	(-) ve	(-) ve

Note: Mean value of triplicates is mentioned.

Table 2 presents the microbiological changes of vacuumed packed marinades and bottled marinades. The acceptable level of bacterial colonies is $1.00x10^5$ (Garbutt, 1997). Vacuum packed marinades are safer for consumption than bottled marinades because they were negative for faecal coliforms and had an acceptable value for the total plate count. According to the total plate count results, the vacuum packed products had a shelf life of one month.

Physico-chemical studies

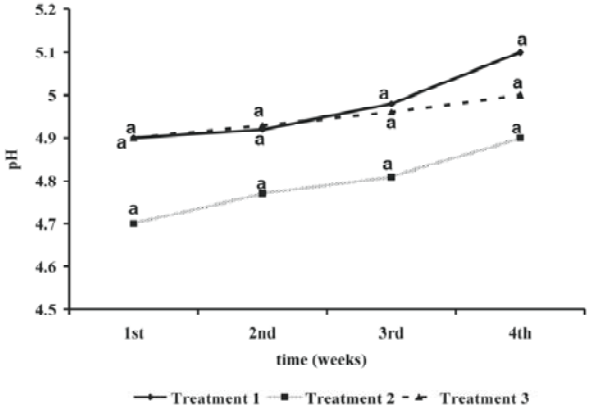

Fig. 2. pH changes of three vacuum packed samples with time.
Note: Similar letters indicate no significant difference among each sample with time at P<0.05

Changes in pH and total acidity in the three vacuum packed samples with time are presented in Fig. 2 and Fig. 3. It can be seen that there was no significant difference between the three vacuum packed treatments in these parameters. Acidity and pH of all three treatments were below 5.3 (pH) and above 1.95% acidity which are acceptable levels.

Anaerobic conditions can lead to the growth of *Clostridium botulinum,* which can produce the toxin causing botulism. Since this organism can multiply within 24 hours, the acid concentration must be high enough to ensure that the pH will be below the maximum of 5.3 within 24 hours. This can be achieved at acid levels above 1.95 % (Borgstrom, 1989).

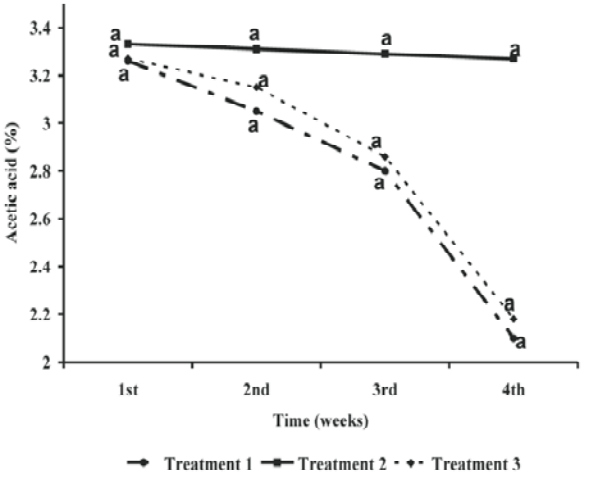

Fig. 3. Acidity value changes of vacuum packed samples with time.
Note: Similar letters indicate no significant difference among each sample with time at $P<0.05$

When preparing the marinades, there are two main considerations in choosing the concentration of salt and acid; firstly, the solution should have an equal distribution of salt in the tissue fluid of the fish and in the solution. Secondly, a part of the acetic acid should be bound by the fish fillets (Merinodol, 1969).

The shelf life of the marinades may be several months in the freezer but only a few weeks at tropical ambient temperatures. Moreover, if the autolytic enzymes and the bacteria remain active, the shelf life of marinades is limited even under frozen conditions. The shelf life can be extended by adding sugars to the marinade. This reduces the water activity of the product and inhibits the growth of bacteria. Moreover, the packaging is also important to increase the shelf life. The packaging used must be

impermeable to water vapour, oxygen and flavour volatiles. Double laminated high density polyethylene was used as packaging material in vacuum packaging to fulfill the above requirements and it is suitable for marinated products. It should be noted however, that strict hygiene and storage control practices are required with this type of product, where the acidity is not necessarily high enough to stop the growth of pathogens (Hobbs and Roberts, 1993).

Conclusion

Results of this study show that marinades with a pleasant taste and good texture can be developed from tilapia. Vacuum packed marinades were safer to consume than bottled marinades. These novel marinades developed possessed good quality and high consumer acceptance. Since only 50% of the tilapia meat can be used for the tilapia marinade production the production cost is high. Therefore, further research should be carried out to reduce the cost of production and to extend the shelf life of the product.

Acknowledgements

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