Effect of plant-extracts on quality changes of refrigerated Nile tilapia

*Oreochromis niloticus* (Linnaeus, 1758) fillets

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**ABSTRACT:** Effect of chaplu (*Piper sarmentosum*) (1% w/v), mulberry (*Morus alba*) (1% w/v) and rang chuet (*Thunbergia laurifolia* Lindl.) (1% w/v) extracts on the quality changes and shelf-life extension of tilapia fillets kept at 4°C was investigated. Tilapia fillets dipped with rang chuet extracts had lower microbiological and chemical deterioration as evidenced by the lower microbial counts, total volatile base and thiobarbituric acid-reactive substance (TBARs), than those dipped in mulberry and chaplu extracts. Moreover, samples treated with rang chuet extracts exhibited higher likeness score for odour and flavour, compared with the samples treated with other plant extracts throughout the storage of 15 days. Therefore, rang chuet extracts treatment was shown to be the promising means to prevent the deterioration and maintain the odour and flavour attributes of tilapia fillets during prolonged storage.

**Keywords:** plant-extracts, quality, tilapia (*Oreochromis niloticus*) fillets

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**Introduction**

Nile tilapia *Oreochromis niloticus* (Linnaeus, 1758) is one of the most important cultured and exported fish in Thailand. It is commonly consumed as whole fish or as fillets. However, the short shelf-life is a limiting factor for these perishable flesh fish. Therefore, effective methods have been applied to preserve and prolong the shelf-life of fish. Nowadays, natural preservatives such as plant extracts and natural herbs have also been investigated to extend the shelf life of fish because of their safe and good preservative properties (Khalafalla et al., 2015). Plant extracts which comprise a great diversity of compounds, such as phenolic acids, flavonoids, lignans and stilbenes was found and attracted attention due to its enzyme inhibition, antiviral, antioxidant and antimicrobial activities (Zhao et al., 2013). Plant extracts such as chaplu (*Piper sarmentosum*), mulberry (*Morus alba*) and rang chuet (*Thunbergia laurifolia* Lindl.) are complex mixtures of bioactive compounds. They are the important dietary spices and herbs in Thai food and traditional medicine, which have been studied to provide good antimicrobial and antioxidant activity (Quitral et al., 2009). Although, plant extract could prevent microbial and chemical changes of fish, but the information is limited on the quality changes of tilapia treated with chaplu, mulberry and rang chuet. Thus, the objective of the present study was to investigate the effects of plant extract treatment on the quality changes of tilapia fillets during refrigerated storage.

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Introduction

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Material and Methods

Preparation of tilapia

Nile tilapia size of 5-7 individuals/kg, were purchased from a farm in Pattani. The samples were transported in ice to the Department of Food Science and Nutrition, Prince of Songkla University. Upon arrival, they were then washed, filleted and deskinned. Fillets were kept in a bag at 4 °C until used.

Preparation of plant extract and fish sample

Fresh leaves of chaplu (Piper sarmentusum), mulberry (Morus alba) and rang chuet (Thunbergia laurifolia Lindl.) with average size (6×10 cm.) were obtained from a farm in Pattani. The leaves from all plants were washed, dehydrated in a dry oven at 50 °C for 15 h and pulverized into coarse powder. The dried plant powder was kept in a polyethylene bag. Each dried plant powder (1%w/v) was extracted with hot distilled water at 95 °C for 10 min. The crude plant extract was filtrated and cooled at 4 °C. Tilapia fillets were dipped with crude extract of each plant (1%w/v) for 10 min at 4 °C. After treatment, fillets were drained for 10 min. Then fillets of each treatment were placed in pouch. The control samples were soaked in distilled water under the same condition. All samples were taken for microbiological, chemical and sensory analyses every 3 days for up to 15 days.

Microbiological analysis

Flesh samples (25g) were aseptically collected in a stomacher bag and ten volumes of sterile saline solution (0.85%) were added. After homogenizing in a stomacher, a series of ten-fold
dilutions was made using saline solution. Mesophilic bacterial counts were determined by plate count agar with the incubation at 35 °C for 2 days. Microbial counts were expressed as log CFU/ g.

**Chemical analysis**

**Determination of total volatile base (TVB)**

TVB was determined by the Conway's method as described by Conway (1950). The samples (2 g) were homogenized with 10 ml of 4% TCA. The homogenate was filtered and used for analysis. Sample extract (1 ml) was placed in the outer ring. The inner ring solution of 1% boric acid containing the Conway indicator was then pipetted into the inner ring. To initiate the reaction, $\text{K}_2\text{CO}_3$ (1 ml) was mixed with the sample extract. The Conway unit was closed and incubated at 37°C for 60 min. The inner ring solution was then titrated with 0.02 M HCl until the green color turned to be pink. The results were expressed as mg TVB/ g muscle.

**Determination of thiobarbituric acid-reactive substances (TBARS)**

Thiobarbituric acid-reactive substances (TBARS) were determined according to the method of Buege and Aust (1978). Chopped samples (0.5 g) were homogenized in 2.5 ml of the mixture containing 0.375% TBA, 15% TCA and 0.25 M HCl. The mixture was heated in the boiling water for 10 min, followed by cooling. The mixture was centrifuged at 3,600 g for 20 min and the absorbance was measured at 532 nm using spectrophotometer. TBARS were calculated from the standard curve of malondialdehyde and expressed as mg malondialdehyde /kg muscle.

**Sensory evaluation**

The sensory evaluation was performed by thirty trained panelists on the basis of previous experience. The assessment was conducted for the odour of raw fish samples using a nine-point hedonic scale. The evaluation of odour was carried out at the moment of opening the pack. For cooked samples, the samples were wrapped with aluminium foil, cooked in steaming pot until the core temperature of each sample reached 70 °C. Stick water was drained and allowed to cool to room temperature (25-28 °C). The flavour likeness of cooked samples was evaluated using a 9-point hedonic scale.

**Statistical analysis**

All experiments were run in triplicate. Data were subjected to analyze using analysis of variance (ANOVA). The least significant difference procedure was used to test for differences between means. Statistical analysis was performed using the Statistical Package for Social Science (SPSS 10.0 for Windows, SPSS Inc., Chicago, IL, USA).

**Results and Discussion**

**Microbiological analysis**

The initial prime quality of tilapia fillets used in this study was observed, as indicated by a low initial number of bacteria (<4 log CFU/g). Sample treated with rang chuet had the lowest count (2.7 log CFU/g), whereas the control contained 3.9 log CFU/g (P<0.05) (Figure 1). The plant-extract of rang chuet was the most effective in retarding the microbial growth, followed by mulberry and chaplu, respectively. Our result was in agreement
with Khalafalla et al. (2015) who reported that TVC was retarded when Nile tilapia fillets were treated with natural herbs and plant extracts. The antimicrobial properties of natural herbs and plant extracts are attributed to the phenolic constituents, flavonoid and aromatic compounds. The antimicrobial activity of rang chuet was attributed to the presence of some components, such as phenolic, carotenoid, caffeic and apigenin adsorbed in the lipid of bacterial cell membrane and mitochondria interfering with the lipid bilayer. The component activity disturbs cell structure, resulting in an increased permeability and lethal damage to cell structure. The inhibitory components such as flavonoid, rutin, quercetin and apigenin in mulberry have also been reported. The bioactive agents in chaplu including amide, flavonoids and pyrones were also reported by Amran et al. (2010). From the result, sample dipped with rang chuet had the lowest mesophilic bacterial count, especially when kept for a longer time (P<0.05). The results indicated that rang chuet inhibited the growth of bacteria effectively.

![Figure 1](image)

**Figure 1** Changes in mesophilic bacterial counts in tilapia fillets treated with different condition during storage at 4 °C. Bars represent the standard deviation from triplicate determinations.

**Chemical analyses**

Total volatile bases (TVB) are products of bacteria spoilage and their contents are often used as index to assess the keeping quality and shelf-life of fish. Generally, the higher TVB content was found in control sample when compared with plant extracts treated sample throughout storage (P<0.05) (**Figure 2(A)**). Samples treated with rang chuet showed the lowest TVB content, followed by mulberry and chaplu, respectively. For the control, TVB content increased rapidly and reached 0.32 mg/g after 9 days of storage. However, all samples dipped with plant extracts had TVB contents less than 0.30 mg/g within 12 days of storage. After 15 days of storage, TVB content of sample with plant extracts reached 0.40 mg/g. Lannelongue et al. (1982) reported that TVB limits of 0.30 and 0.35 mg/ g were established for different groups of fish species and used a limit for acceptability of bacterial spoilage for fresh fish. It was reported that TVB contents in mackerel (*Trachurus murphyi*) pretreated with extracts of oregano (*Origanum vulgare*) and rosemary (*Rosmarinus officinalis*) was increased lower than those of fish untreated with extracts (Quitral et al., 2009). Therefore, inhibitory effect of plant extracts
from rang chuet against microbial growth could decrease the production of microbial degradation products.

An increase in TBARS was observed in all samples when the storage time increased (P<0.05), indicating that formation of secondary lipid oxidation products took place during storage. Figure 2(B). Lipid in fish muscle has typically a high percentage of polyunsaturated fatty acids and is consequently prone to oxidative reaction. At each day of determining throughout storage, higher TBARS value (P<0.05) in the control samples were found in comparison to tilapia fillets dipped with other plant extracts. The plant extract of rang chuet treated sample showed the lower TBARS than those treated with mulberry and chaplu, throughout the storage. So, rang chuet was the most effective in retarding of lipid oxidation, followed by mulberry and chaplu, respectively. The antioxidant of plant extracts are attributed to the phenolic and flavonoid compounds which potent antioxidants, free radical scavengers and metal chelators to inhibit lipid oxidation (Zhao et al., 2013).

![Graph A](image1.png)

**Figure 2** Changes in total volatile base (TVB) (A) and Thiobarbituric acid-reactive substances (B) contents in tilapia fillets treated with different condition during storage at 4 °C. Bars represent the standard deviation from triplicate determinations.

**Sensory evaluation**

Figure 3 (A) and (B) shown the likeness score of tilapia fillets without and with plant extracts treatment, in comparison with the control. At day 0, all samples were generally considered to possess very high acceptability and no differences in likeness were observed between all treatments (P>0.05). The result suggested that that treatment of tilapia fillets with plant extracts had no impact on odour and flavour of treated fillets. At the end of storage, the decrease in likeness for all attributes were found for all samples during storage (P<0.05). The odour and flavour scores of the control sample decreased more rapidly than those of samples treated with all plant extracts during storage. From the result, the control samples were rejected after 6 days of storage. Generally, sensory evaluation is frequently applied in estimating the quality of fish and correlated with microbiological and chemical analyses. Samples treated with rang chuet extract exhibited the higher likeness scores for dour and flavour, compared with mulberry and chaplu throughout the 15 days storage (P<0.05). Our results indicated...
that treatment of tilapia fillets with plant extracts, particularly 1% rang chuet extract, effectively extended the shelf-life of fillets with high acceptability throughout the storage of 15 days. Therefore, it was suggested that the selected plant extracts might retard bacterial growth and lipid oxidation, leading to the shelf-life extension of tilapia fillets.

Figure 3 Changes in odour (A) and flavour (B) contents in tilapia fillets treated with different condition during storage at 4°C. Bars represent the standard deviation from triplicate determinations.

Conclusions

Appropriate plant extracts may provide the cornerstone of sanitizing formulation or processing aids that could improve the microbiological quality and safety of tilapia fillets during refrigerated storage. Microbial growth in tilapia fillets treated with 1% rang chuet extract was retarded, leading to the delayed deterioration.

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References


