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# The effect of powdered thyme sprinkling on quality changes of wild and farmed gilthead sea bream fillets stored in ice

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### ABSTRACT

The effect of powdered thyme sprinkling on the quality characteristics of fresh and ice-stored wild and farmed gilthead sea bream fillets was assessed. Initially, significant differences in proximate composition and quality attributes were found between wild and farmed fresh sea bream flesh. Throughout ice storage, biochemical alteration appeared more pronounced in farmed fish fillets with significantly higher levels of TVB-N, TMA-N, and TBA; and a lower liquid-holding capacity (LHC). Thyme powder addition (1% w/w) exhibited a preservative effect in both fish lots since significant lower levels of TVB-N, TMA-N, free amino acids (NPS), TBA and LHC were observed in thyme-treated fillets during ice storage. However, thyme inhibitory effect was more marked in wild than farmed fish. As revealed by partial least square regression, LHC in both groups was positively influenced by storage time and trimethylamine accumulation factors, while it was negatively influenced by thyme treatment and fish origin. Hence, LHC was suggested to be related to spoilage bacterial growth. The use of dried thyme extended the shelf life of fish fillets by about 5 days and appeared to be highly valuable to the fish industry as a natural preservative.

# 1. Introduction

Consumer attitudes towards any food product are a major factor influencing its future development. Thus any new wave of products on the market has to meet the needs of the consumer, such as foods that offer specific health-oriented benefits or ready-to cook food including fish and seafood products. However, due to its high perishability, the quality and shelf life of fish products are restricted and are defined by several factors including both handling practices and origin. For instance, wild fish acceptability is generally higher than cultured fish (Grigorakis, 2007). Thus existing literature noticeably specifies that significant organoleptic differences always exist when comparing wild and cultured flesh of the same species. Among studied parameters, technological quality attributes were found among the most discriminating parameters between wild and reared fish (Mairesse, Thomas, Gardeur, & Brun-Bellut, 2005). Thus, the determination of liquid-holding capacity (LHC) in meat and fish is important for economical reasons (weight decrease due to water loss) and for sensory properties (colour, juiciness and tenderness) (Olsson, Olsen, & Ofstad, 2003a). In general, the flesh of farmed fish tends to be softer in texture than wild fish. Therefore it is essential to make an evaluation of high quality fish available to the consumer whether there are differences related to the various post-mortem handling procedure and/or the origin of the fish.

Additionally, and because of its greater awareness and safety concern regarding synthetic chemical additives, food preserved with natural additives has become more popular. For instance, the antimicrobial and antioxidant properties of essential oils, and their active constituents derived from various plant organs have been empirically recognised (Burt, 2004; Lee & Shibamoto, 2002). However, any processing technologies used in the production of such compounds have to prove their technical/scientific efficiencies and the product quality to meet the basic requirement of hygiene and safety standards. It would therefore be economically more convenient to use powdered spices/herbs as ingredients rather their extracts to preserve food including fish fillets (Smid & Gorris, 1999) as they are generally recognised as safe to consume. Beside it is well known that at the core of the traditional Mediterranean cooking, spices and fresh/dried herbs were used for centuries, which current research has confirmed its tremendous health benefits (Burt, 2004; Lee & Shibamoto, 2002). Among the various aromatic plants, thyme (Thymus vulgaris) is a characteristic herb of the Tunisian and Mediterranean environment with various beneficial effects, e.g., antiseptic, carminative, antimicrobial, and antioxidative properties (Baranauskiene, Venskutonis, Viskelis, & Dambrauskiene, 2003). In the seafood sector, such preservative compounds including spices/herbs were equally used to extend the shelf life of fish fillets, but at low levels to avoid strong flavours imparting unpleasant sensorial characteristics (Mejlholm & Dalgaard, 2002).

Despite the numerous comparative investigations on postmortem quality alteration in both wild and cultured sea bream

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through handling, distribution and storage conditions (Alasalvar, Taylor, & Shahidi, 2002, 2005; Grigorakis, 2007; Kuley, Özogul, & Özogul, 2005; Mnari et al., 2007; Özogul, Kuley, & Özogul, 2007), no studies have investigated the changes in liquid-holding capacity in *Sparus aurata* muscle during ice storage.

The aims of the study was to compare the biochemical changes including liquid-holding capacity in wild and farmed gilthead sea bream (*S. aurata*) flesh, and to determinate the effect of dried-powdered thyme (1% w/w) as a natural and low-cost preservative on the quality of sea bream.

#### 2. Materials and methods

### 2.1. Materials

Thirty cultured gilthead sea bream (S. aurata), (average weight and length:  $208 \pm 22$  g and  $241 \pm 11$  mm, respectively) used in this study were bought in January 2005 from the farming unit "Aquaculture Tunisienne", located in Hergla, Tunisia. The commercial feed (Mistral-21, EWOS, Dueñas, Palencia, Spain) used for fish feeding, contained 45% protein, 21% fat, 14.5% carbohydrates, 11% ashes and 1.5% crude fibre. Wild sea bream (30 fishes with average weight and length:  $191 \pm 18 \,\mathrm{g}$  and  $262 \pm 14 \,\mathrm{mm}$ , respectively) were caught by commercial longline fishing gear during the same day, from the coastal water of the same area (Hergla, Tunisia). The aerial part of wild-thyme (T. vulgaris) from Zaghouan Mountain (North of Tunisia), was dried, ground to powder and sterilised by gamma irradiation at 10 kGy (National Center of Nuclear Sciences and Technologies, Sidi Thabet, Tunisia). All chemicals were obtained from Sigma-Aldrich-Fluka Company Ltd. (Poole, Dorset, UK), unless otherwise specified.

# 2.2. Sample preparation and storage conditions

Wild/farmed gilthead sea bream were killed by immersing in ice cold water (hypothermia) and delivered (packed into an insulated polystyrene box with ice) to the laboratory, within 3 h of harvesting. Upon arrival, fishes were immediately weighed, gutted, headed, washed, and filleted. Six farmed (F) and wild (W) fish fillets were immediately sampled (day 0), a portion of tissues from each fillet was cut, deep-immersed in liquid nitrogen and stored at -80 °C until proximate/biochemical analysis. The following treatment of fish fillets (n = 30 in each group) were prepared: control lots (W) and (F), and sprinkled with irradiated thyme powder (1% w/w) lots (WT) and (FT). All fillets were packed in polyethylene films (not sealed) separately and than stored in flake ice (ratio of 1:1 (w/w)) into polystyrene boxes provided with holes for drainage. Boxes were covered and stored in a refrigerator (2-4 °C) for up to 15 days. Tissues sampling and storing from each lot was performed as previously reported on days 0, 3, 7, 10 and 15.

# 2.3. Proximate analysis

The fish samples were analysed for proximate composition: moisture content was determined by air-drying of a portion of minced fish fillet at  $103 \pm 2$  °C for 24 h (method 950.46), crude protein by the Kjeldahl method using potassium sulphate and copper (II) sulphate as the catalysts and 6.25 nitrogen-to-protein conversion factor (method 981.10), and ash by incineration in a muffle furnace at  $550 \pm 2$  °C for 24 h (method 938.08), according to the official methods of the Association of Official Analytical Chemists (AOAC, 1995). Lipids were extracted by using chloroform:methanol (2:1 v/v) extraction solution according to the Bligh and Dyer method (Bligh & Dyer, 1959).

### 2.4. Determination of TVB-N, TMA-N and total free amino acids

For this analysis, 0.5 g of tissue were homogenised (DI-25, IKA, Germany) on ice in 500  $\mu$ l ultra pure water for 1 min, 0.5 ml of 6% perchloric acid was added and the extract was homogenised for a further 2 min. Homogenates were centrifuged at 14,000g for 20 min and the supernatants were used for total volatile bases (TVB-N) (Ruiz-Capillas & Horner, 1999), trimethylamine (TMA-N) (Sadok, Uglow, & Haswell, 1996) and total free amino acids measured as ninhydrin-positive substances (NPS) (Sadok, Uglow, & Haswell, 1995).

# 2.5. Determination of thiobarbituric acid value (TBA)

TBA values were determined spectrophotometrically according to the procedure described by Hamre, Næss, Espe, Holm, and Lie (2001). The homogenised sample (0.5 g) was weighed into screwcapped glass tubes and 4.0 ml of chloroform:methanol 2:1 (v/v) with 0.005% butylated hydroxytoluene (BHT) was added. Samples were purged with nitrogen gas; the tubes were closed and incubated with constant shaking for 30 min at room temperature. Thereafter, 2.0 ml of a saturated EDTA solution was added and the tubes were centrifuged for 20 min at 1500g. A 2.0 ml aliquot of the methanol:water layer was transferred to clean screw-capped glass tubes, mixed with 2.0 ml TBA-reagent (1% thiobarbituric acid in 5% trichloroacetic acid) and heated for 30 min at 100 °C. The absorption was measured at 532 nm with a Smart Spec-plus spectrophotometer (Bio-Rad, Hercules, CA) and TBA values quantified by reference to an external standard. The standard was prepared from 50 µl of 1,1- to 3,3-tetraethoxypropan diluted to 50 ml with 0.1N HCl and heated at 100 °C for 10 min. A volume of 2.4 ml of the hydrolysed acetal was then diluted to 100 ml with distilled water, giving a stock-solution equivalent to 0.1 mM malondialdehyde. TBA values were expressed in units of mg malonaldehyde/ kg sample.

# 2.6. pH measurement

Measurement of pH was performed at room temperature using a digital calibrated pH metre (Inolab pH-720, WTW, Weilheim, Germany) on homogenised fillet samples in distilled water (1:2 w/v) (method 981.12, AOAC, 1995).

## 2.7. Determination of liquid-holding capacity

To evaluate the liquid-holding capacity (LHC) of fillets, a centrifugation test as described by Rørå, Regost, and Lampe (2003) was performed. Chopped frozen muscle samples (2 g) were weighed and placed in a centrifuge tube with weighted (V1) filter paper (Whatman, Maidstone, England). Following muscle thawing, the tubes were centrifuged (3K30 Sigma Centrifuge, Osterode am Harz, Germany) at 4000g for 10 min at 10 °C, and the wet paper was weighted (V2) before drying at 50 °C to constant weight (V3). The percentage value of liquid loss (LL) was calculated as  $100 \times (V2 - V1)/S$ , where S = weight of muscle sample, water loss as  $100 \times (V2 - V3)/S$  and fat loss as  $100 \times (V3 - V1)/S$ . All losses were expressed as percentage of muscle wet weight.

# 2.8. Statistical analysis

All biochemical analysis were carried out in duplicate and averaged for each fillet. For each lot and at each sampling time, the results were presented as mean  $\pm$  standard deviation of n = 6 fillets. All the data were statistically treated using ANOVA with an SPSS Statistical Software System 11.01 (SPSS Chicago, IL, USA). Tukey's test was used to determine the possible significant differences

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