## 所属生物圈生命科学専攻氏名 AHIMBISIBWE JOHN BOSCO

## 審查委員 荒木 利芳, 加納 哲, 吉松 隆夫, 青木 恭彦

論文題目 Effect of Bleeding on the Quality of Amberjack (*Seriola dumerili*) and Red Sea Bream (*Pagrus major*) Muscle Tissues during Iced Storage and Detection of Cathepsin L in Red Cell Membranes of Fish Blood (カンパチおよびマダイ筋肉の品質に及ぼす脱血の効果および血液中の カテプシンLに関する研究)

## (要旨本文)

The aim of the study was to investigate the effect of bleeding on fish muscle freshness and spoilage in cultured amberjack and red sea bream stored on ice and the existence of cathepsin L in fish blood.

ATP-related compounds and the *K*-value were used as freshness indices and measured using High Performance Liquid Chromatography (HPLC). Trimethylamine (TMA) was measured using the picric acid method while volatile basic nitrogen (VBN) was measured using Conway's micro-diffusion method. Both TMA and VBN content was used as indicators of spoilage. Fish blood was collected immediately after killing the fish and the existence of cathepsin L was investigated using SDS-PAGE, and immunoblotting using human anti-(cathepsin L) antibody.

The levels of ATP, ADP and AMP content in the fish muscle decreased throughout iced storage and only a small amount of ADP and AMP was subsequently observed. IMP was the main degradation product in both the amberjack and red sea bream muscle. IMP content decreased from 7.36  $\mu$ mol/g on 0 day storage to 6.32  $\mu$ mol/g in bled samples and 5.6  $\mu$ mol/g in the unbled samples in the red sea bream. In the amberjack muscle, the values decreased from 7.78  $\mu$ mol/g on 0 day storage to 3.02  $\mu$ mol/g in bled samples and 2.50  $\mu$ mol/g in the unbled samples after 15 days storage. Its content remained higher than any other ATP-related compounds throughout the iced storage.

In both the amberjack and red sea bream muscles, there was no hypoxanthine observed on 0 day storage. Subsequent sampling showed a higher hypoxanthine content in the unbled samples than in the bled samples that increased throughout iced storage.

Inosine content in both species increased throughout storage and differences between bled samples and the unbled samples only became noticeable after 7 days storage. In the amberjack muscle, the *K*-value increased from 10% on 0 day storage to 54% and 58% in the bled and unbled samples, respectively, after 15 days storage. Red sea bream muscle on 0 day storage had a *K*-value of 5.8% and this increased to 31.2% in bled samples and 32.6% in the unbled samples after 15 days storage.

The TMA content was low in both amberjack and red sea bream muscles, and remained below 1 mg/100g muscle after 15 days storage. Lower values of TMA content were observed in bled samples compared to the unbled samples after 7 and 8 days storage in both fish species.

VBN content in the amberjack muscle increased from a value of 10.1 mg/100g on 0 day storage to 20.5 mg/100g in bled samples and 24 mg/100g in the unbled samples on 15 days storage. In the red sea bream muscle, VBN content increased from 15.1 mg/100g on 0 day storage to 20.1 mg/100g in bled samples and 25 mg/100g in unbled samples on 15 days storage. Bled samples had significantly lower VBN content than unbled samples after 8 days storage.

In the amberjack red cell membrane preparation, two bands appeared on the immunoblot using anti-cathepsin L antibody that corresponded to 120 and 85kDa proteins. However, only one band appeared on the immunoblot and corresponded to 30kDa protein in the red sea bream preparation. The amberjack blood contained active precursor forms of cathepsin L while red sea bream fish blood contained a mature form of cathepsin L.

These results suggest that the fish bleeding has a preferable effect on fish muscle quality.