Corso di Dottorato di Ricerca in Scienze della Vita e dell'Ambiente - Ciclo XXXV

Reproduction and health status in swordfish Mediterranean stock (*Xiphias gladius*) for management of sustainable fishing

PhD Student: Sara Filippi

Supervisor: Dr. Giorgia Gioacchini Co-Supervisor: Prof. Oliana Carnevali

Laboratory of Developmental and Reproductive Biology

ddRAD genotyping reveals new insight into the genetic structure and diversity and fitness of Atlantic and Mediterranean swordfish stocks

AIM – (i) confirming the current spatial stock boundaries and degree of admixture; (ii) gain deeper insights into the genetic structure of the three swordfish stocks, the North Atlantic, the South Atlantic and the Mediterranean one.

METHODS – A total of 576 swordfish were sampled from 14 sampling areas in the Atlantic Ocean and Mediterranean Sea. Genomic DNA was extracted from ~25mg of tissue. A total of 719,722,497 paired reads were sequenced and the final file was filtered for MAF> = 5 %. After filtering, 26,324 SNPs were analysed.

RESULTS

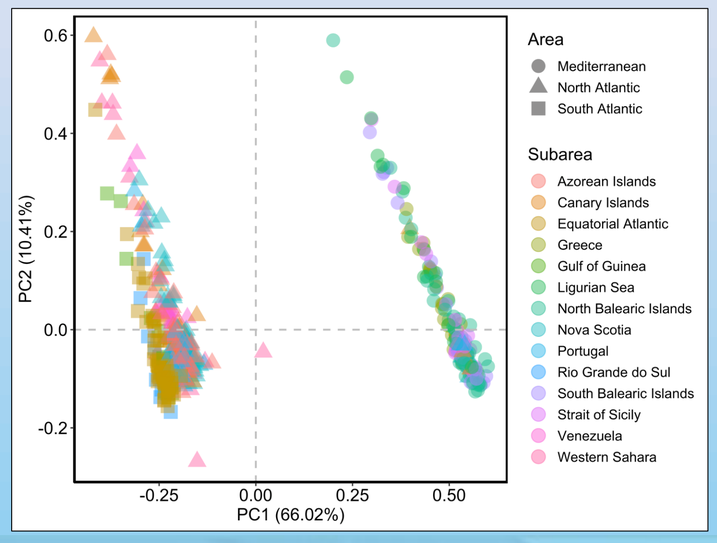

Figure 1 - The principal component analysis (PCA) well separated the samples in two main clusters, one corresponding to almost all samples from Atlantic (*n*=370) and one corresponding to all the Mediterranean samples (*n*=149) in addition to 11 swordfish from North Atlantic.

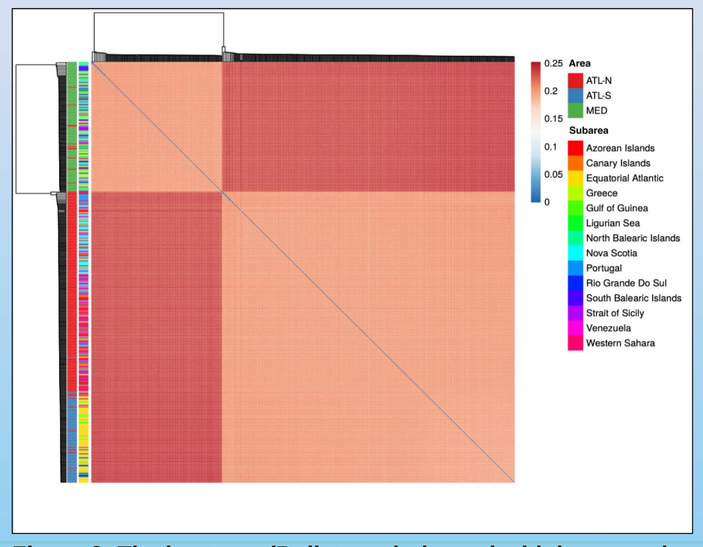

Figure 2 -The heatmap (P-distance) showed a higher genetic diversity between Mediterranean and Atlantic stocks, while a weakly genetic differentiation was evidenced between North and south Atlantic stocks. 11 specimens caught in North Atlantic were inserted into Mediterranean stock.

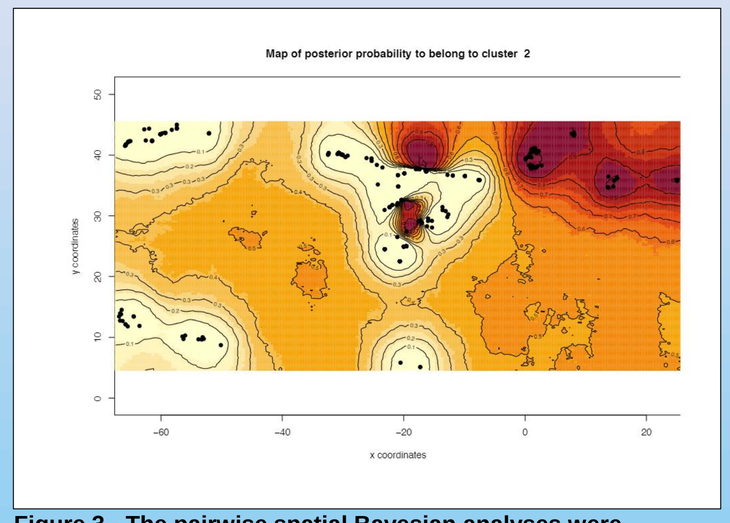

Figure 3 - The pairwise spatial Bayesian analyses were performed. The pairwise analysis between North Atlantic and Mediterranean stocks showed two cluster. Six specimens belonging to Portugal and 5 belonging to Canary Islands were assigned to cluster 2 with a high posterior probabilities (≈0.9)

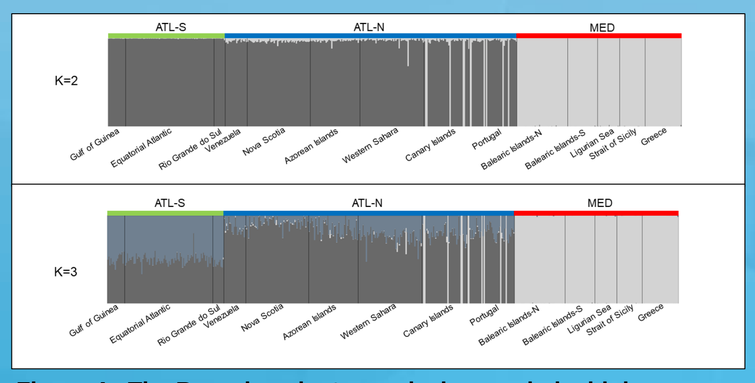

Figure 4 - The Bayesian cluster analysis revealed a high magnitude corresponding to K=2. The two clusters clearly reflected the Mediterranean and the Atlantic stocks. 11 specimens caught in the North Atlantic showed a Mediterranean genotype

Sampling Areas	Ho	Hs	Fis	TotalRichness	MeanRichness
Gulf of Guinea	0.224008	0.233515	0.031114	42060.60964	1.597804651
Equatorial Atlantic	0.227317	0.233623	0.027936	42443.72162	1.612358366
Rio Grande Do Sul	0.223649	0.231	0.022525	41603.89012	1.580454723
Venezuela	0.225631	0.235422	0.034486	42324.51908	1.607830082
Nova Scotia	0.227704	0.235864	0.032784	42585.48604	1.617743734
Azorean Islands	0.228592	0.236764	0.032531	42623.86027	1.619201499
Western Sahara	0.231611	0.237947	0.027443	42728.2161	1.623165784
Canary Islands	0.229108	0.243128	0.056633	43158.53127	1.63951266
Portugal	0.228714	0.243101	0.058279	43150.86627	1.639221481
North Balearic Islands	0.235668	0.242784	0.026899	42299.94508	1.606896561
South Balearic Islands	0.236063	0.24258	0.022598	42187.56654	1.602627509
Ligurian Sea	0.238848	0.243558	0.015949	42168.22542	1.601892776
Strait of Sicily	0.236167	0.242492	0.023165	42160.46023	1.60159779
Greece	0.234562	0.242228	0.028547	42210.72645	1.603507311

Figure 5 – Statistical analysis of heterozigosity, both expected (He) and observed (Ho), inbreeding coefficent (Fis) and total and mean allelic richness.

RESULTS and DISCUSSION

Multivariate and individual-based analyses of 26,324 SNPs, evidenced a great genetic differentiation between Mediterranean and Atlantic stocks, while a weakly differentiation between North and South Atlantic stocks was found. The results achieved in this study, confirmed and deepened the debate about stocks boundaries. Specifically, the range of the South Atlantic population extends beyond 5°N ICCAT management boundary to 25°N. Likewise the Mediterranean population extends beyond the current management boundary at the Strait of Gibraltar to approximately 27°N and 20°W. In this light, swordfish fisheries management should consider that in the East-North Atlantic area, specimens belonging to all three stocks (10-14% of Mediterranean and 15% of South Atlantic) are caught and whose presence should be considered when genetic variability is monitored in this area.

Swordfish's survey (Xiphias gladius) in central Adriatic Sea: focus on reproduction, health status and catches size

AIM – (i) evaluating the health status of immature and mature females during the reproductive and no reproductive season; (ii) evaluating the reproductive stage; (iii) determing the catches sizes in Adriatic sea.

METHODS – 48 specimens caught in Central Adriatic Sea were analysed. All specimens were collected between August and November 2021 and the total weight was measured (kg). Liver and ovaries samples were embedded in paraffin and sections of 4 µm were cut with a microtome and stained in H&E. Quantification of melanomacrophages (MMCs and MMs) and lipids were performed at 20x and 40x respectively using ImageJ/Fiji software.

RESULTS

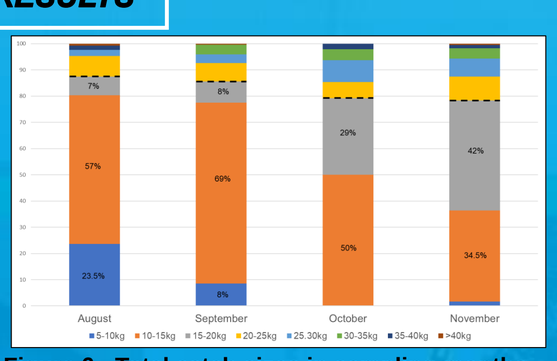

Figure 6 - Total catch sizes in sampling months

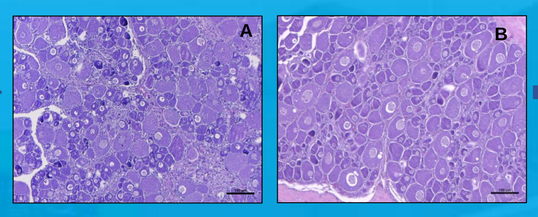

Figure 7 – Ovaries of immature (A) and mature (B) swordfish female

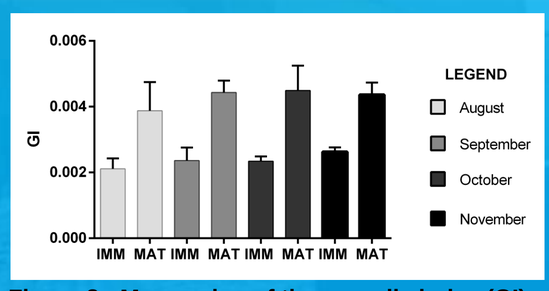

Figure 8 - Mean value of the gonadic index (GI) in the sampling months.

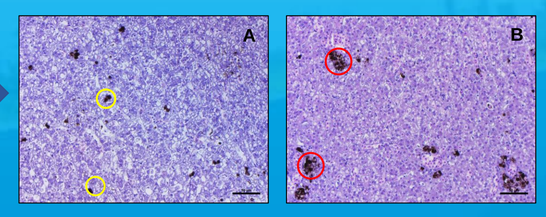

Figure 9 - Melanomacrophage centres (red circle) and single melanomacrophages (yellow circle), in liver of immature (A) and mature (B) females

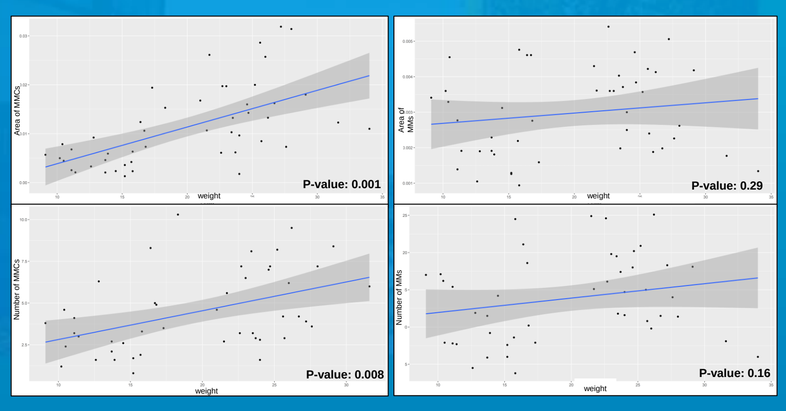

Figure 10 - Pearson correlation test between melanomacrophages parameters and weight (kg).

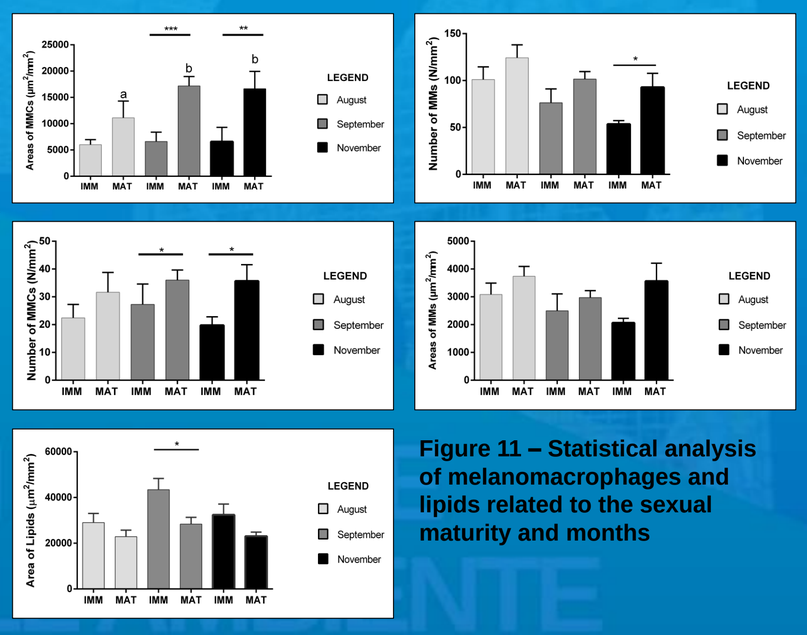

Figure 11 – Statistical analysis of melanomacrophages and lipids related to the sexual maturity and months

RESULTS and DISCUSSION

In all the sampling months, immature swordfish were caught, with a peak in the size range 10-15kg (or LJFL<120 cm) in August, September (60-70% of the landed) and November (45% of the landed). The size of first maturity is achieved by females at 131.5 cm, therefore the most of females landed correspond to immature specimens, which never reproduced, and this situation could contribute to the collapse of the stock. Moreover, the results showed the density and number of MMCs were correlated to the weight, then age of specimens. In fact, the mature females showed a higher density and number of MMCs compared to immature females. Both immature and mature females showed similar density and numbers of MMs, suggesting that MMs were not correlated to the age of specimens. Furthermore, the density and numbers of MMCs changed in relationship with the months. In fact, the mature females caught in August showed the lower density of MMCs compared to the mature females caught in September and November. Mature females caught in August probably spawned in May/June and for this reason they recovered energies to be invested in detoxification and the immune system. The density of lipids of mature females unchanged in the three months. Only significant difference was found between immature and mature females caught in September.