# POPS LEVELS IN FILLET OF SEA BREAM (Sparus aurata) FROM TWO OFFSHORE CAGES OF SOUTHERN ITALY

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#### Abstract

Persistent organic pollutants, such as PCDDs, PCDFs, DL-PCBs, NDL PCBs and PBDEs (13 congeners) were monitored in the fillet and in the feeds of farmed sea bream, from shallow water and deep water offshore plants, respectively, in the south Adriatic sea, in Italy. At the end of the cycle, the cumulative PCDD/F and DL-PCB averaged contamination in fillet was 0.78 pg WHO-TE/g fresh weight (*fw*), well below the reported 1.87 pgWHO-TE/g *fw* value inventoried in european farmed species. On fresh basis, the bio-accumulation trend was mitigated by the dilution due to the fish growth and resulted more evident when data were expressed on lipid basis. Bio-accumulation recorded in fillets for PBDEs and NDL-PCBs at the end of the cycle averaged around 20 ng/g lipid base (*lb*) and 220 ng/g *lb*, respectively. Cumulative feeds contamination (N = 8) spanned from 0.72 to 2.16 pgWHO-TE/g product (12% moisture) for PCCD, PCDF and DL-PCB congeners, thus indicating a certain variability in the quality of the feed materials used. The evidence of environmental contributions, other than feeds intake, was also assessed.

## Introduction

Farmed fish can be exposed to persistent organic pollutants – such as polychlorinated biphenyls (PCBs), dibenzodioxins (PCDDs), dibenzofurans (PCDFs) and polybrominated diphenyl ethers (PBDEs) – mainly via feed, this eventually resulting in accumulation levels of health concern<sup>1</sup>. To assess the correlation between the feed contamination and the accumulation in fish, a study was carried out under field conditions, according to best aquaculture practice principles. To this purpose, two offshore cages situated in the south Adriatic Sea were selected, at different distances from the coastline, respectively, in shallow and deep water. The chemical determinations were made on fish fillets and viscera of *Sparus auratus* specimens collected during their growth from both the cages and on feed samples given them in the same period. In this paper, the preliminary results are presented on fish fillets, dealing with the different accumulation patterns found, when data are reported on a lipid — or a fresh weight basis. Such data are integrated with the contamination levels found in feeds.

#### Materials and methods

The two offshore cages situated in Southern Italy called, respectively, Mattinata and Govinazzo, were selected according to their distance from the coastline (1 and 3 maritime miles far, respectively), to discriminate possible environmental contribution from anthropogenic sources. Sampling of fillet was performed on a seasonal-basis in the following months: August, November, February and May, thus encompassing a 10 month period correspondent to the growth of sea bream from 100 g to 330 g b.w. (commercial size), on average. The collected specimens presented the same size range although some differences could be observed among the individuals from the two cages, maybe related to the different temperature-regimes experienced, respectively, at shallow and deep water. The fillet portions were mixed and homogenized to prepare a pooled sample (N = 20 for each time and cage), which was then stored at below - 20 °C before the extraction and clean-up process. Pre-treatment processing to produce the matrices for analysis were reported elsewhere<sup>2</sup>. About 20 g of sample was considered for the clean-up process. It was essentially composed of fat extraction, addition of  ${}^{13}C_{12}$ -labelled internal standards, acid decomposition, and clean up on a multi-layer silica gel column followed by a power-prep separation step. The cleaned up extracts for the assessment of chlorinated and brominated analytes, were analysed by adapting the US EPA Method No 1613.3. Analytes were quantified by HRGC-LRMS(SIM) or -HRMS(SIM). Good laboratory practice and QA/QC protocols were applied throughout. As an adaptation of the widely used US EPA Method 1613 (1994), the in-house validated, congener-specific procedure utilized to quantify PCBs, PCDDs, and PCDFs in fish and feed sample extracts was based on high resolution (HR) gas chromatography (GC) combined with low (LR) or HR mass spectrometry (MS) used in the selected ion monitoring (SIM) mode  $^2$ .

#### **Results and discussion**

The contamination level of the selected contaminants in the sea bream fillets from the two different cages are reported in Figure 1 (lipid basis, *lb*) and Figure 3 (fresh weight, *fw*). It should pointed out that in the graphs of Figure 1 the analytical levels of PCDDs, PCDFs, and *dl*-PCBs are reported in pg/g (*lb*) while *ndl*-PCBs and PBDE data are expressed as ng/g (*lb*). In Figure 1, a clear accumulation temporal trend can be observed for all the chemical groups considered. The decrease of chemical concentration during the winter period (November) can be reasonably addressed to the starving status of the fish, due to the sea water Temperature below + 15 °C.



**Figure 1.** Concentrations of analytes found in fish fillets from specimens collected from Giovinazzo (left) and Mattinata (right) cages. The chemicals determined from left to right are, respectively, PCDD+PCDF, dl-PCBs, PCBs, and PBDEs. The concentrations are reported in pg/g (*lb*) for PCDD+PCDF and dl-PCBs, and in ng/g (*lb*) for PCBs and PBDEs.



**Figure 2**. Trends in fillets lipid contents of Mattinata ■ and Giovinazzo ◆ fish. Data are given in % on *fw*.

Figure 2 shows the variations observed in the lipid content of fillet, along the fish growth. The lipid dilution resulting from the growth involves an increase of an estimate of *lb* data while a decrease if the data are reported on a *fw*-basis. When reported on *fw*, according to the EU food safety regulation in place for PCDD, PCDF and DL-PCB in fish (Regulation 1881/2006/EU), all the considered pollutants show a diminishing trend (Figure 3), as consequence of the increase of the growth rate of the muscular mass and the corresponding reduction of the fat percentage in fillets (Figure 2), as already noted in trout <sup>2</sup>. On the contrary, when expressed on *lb*, the results highlight a progressive rise of POPs body burden for all the chemicals considered. This fact might bring to the consideration of different scenarios, when the progressive fish growth rate decreases, in favour of an increase fat content in the fillet.



**Figure 3.** Concentrations of dioxin-like chemicals found in fish fillets from specimens collected from Giovinazzo (left) and Mattinata (right) cages. The chemicals (given in pgWHO-TE/g, *fw*) determined from left to right are, respectively, PCDDs, PCDF, PCDD+PCDF, dl-PCBs, and total TEQ.

Among the substances determined, the chemicals with the highest concentrations are *ndl*-PCBs with a concentration range from 113 (on November) to 220 (on May) ng/g (*lb*) (Figure 1). This is in line with the contamination patterns usually observed in fish <sup>3</sup>. Even if the distance from the coast does not influence significantly the fillets concentration levels, the data on dioxin-like compounds on *fw* (Figure 3) highlight some differences in the trend between the two cages, fact that can not be addressed to the contribution from the feedingstuffs, only. These differences are particularly evident on May in the Mattinata cage, and possibly related to coastal anthropogenic activities affecting the shallow water environment.

At the end of the cycle, the fillet of fish in both farms showed almost the same levels of PCDD, PCDF and DL-PCB contamination of 0.78 pgWHO-TE/g *fw*, on average. Such value is about three time less than the average contamination reported in a recent inventory by EFSA for non fat fish  $(1.81 \text{ pg/WHO-TE/g } fw)^3$  and for farmed fish (1.87 pgWHO-TE/g fw) and well below the pertinent statutory maximum limit of 4 pgWHO-TE/g *fw*, according to Regulation 1881/2006/EU. The contamination of feeds (N = 14) administered ranged from 0.72 to 2.16 pgWHO-TE/g product (12% moisture) in compliance with the provision of law. From the risk management point of view, the regular consumption of such sea bream farmed in the low polluted south Adriatic Sea<sup>4</sup> could contribute in a relevant way to reduce the alimentary exposure to PCDD, PCDF and DL-PCBs, at least in the Mediterranean Countries, such as Italy <sup>5</sup>. The span of contamination observed in feedingstuffs indicate a possible improvement of feed quality, thus allowing a further reduction of the background levels observed in fillets.

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