



## BEACHED MICROPLASTICS EXACERBATE OXIDATIVE STRESS AND INFLAMMATION IN SPARUS AURATA LIVER

Alvaro Torres-Gomez\*<sup>1</sup>, Alberto Navarro<sup>1</sup>, Ico Martínez<sup>1</sup>, Daniel Montero<sup>2</sup>, May Gomez<sup>1</sup>, and Alicia Herrera<sup>1</sup>

<sup>1</sup> EOMAR, ECOAQUA, Department of Biology, Facultad de Ciencias del Mar, ULPGC, Las Palmas de Gran Canaria, SPAIN.

*alvaro.torres@ulpgc.es, alberto.navarro@ulpgc.es, ico.martinez@ulpgc.es, may.gomez@ulpgc.es, alicia.herrera@ulpgc.es*

<sup>2</sup> GIA, ECOAQUA, Department of Biology, Facultad de Ciencias del Mar, ULPGC, Las Palmas de Gran Canaria, SPAIN.

*daniel.montero@ulpgc.es*

**Abstract:** Microplastics (MPs) pose a threat to marine organisms which mistake them for food leading to ingestion-related toxicological effects. Exposure to virgin MPs triggers activation of oxidative stress pathways and increases intestinal and hepatic inflammation. Beached MPs suffer an ageing process that enhances their ability to adsorb pollutants and therefore accentuates their toxicity. Prior studies in *Dicentrarchus labrax* determined that chemical pollutants act synergistically with MPs in triggering inflammatory responses and dysbiosis in fish intestine, and that plastic additives and contaminants bioaccumulate in fish livers and muscle tissue and can therefore be subject to trophic transfer.

Here we assess the effects of beached MP ingestion in *Sparus aurata* juveniles fed a diet containing either 10% virgin, 10% beached MPs or a control diet. Liver enzymatic activity of oxidative stress biomarkers (glutathione-S-transferase, catalase, and superoxide dismutase) was monitored over 80 days. Glutathione-S-transferase (GST) and catalase (CAT) activity remained indistinguishable from controls. In contrast, SOD activity showed a gradual increase for both MP diets, with differing dynamics. Virgin MPs peaked at day 60, whereas beached MPs exhibited consistently higher SOD levels that plateaued by day 80.

Expression of oxidative stress enzymes was quantified via RT-qPCR at day 39. GST and CAT expression remained constant, mirroring the activity data, and SOD expression was quadrupled for the beached MP diet. Oxidative stress transcription factor Hsp70 was significantly upregulated for both MP conditions, with the beached MP diet showing a 2.5-fold increase with respect to virgin MPs. Analysis of the liver cytokine expression via RT-qPCR showed a pro-inflammatory profile with significant upregulation of TNF $\alpha$  and IL-1 $\beta$  for MP-fed fish. The expression of these cytokines was doubled when beached instead of virgin MPs were used.

Overall these results demonstrate that beached MPs increase the toxicity of MPs causing significant oxidative stress and inflammation in fish livers.

**Key words:** Microplastics, Oxidative stress, Inflammation, (3 to 5 key words)

**Acknowledgments:** This work was supported by the IMPLAMAC project (MAC2/1.1a/265) Interreg MAC (European Fund for Regional Development, Macaronesian Cooperation).

### **References:**

- Espinosa, C., Cuesta, A., & Esteban, M. Á. (2017). Effects of dietary polyvinylchloride microparticles on general health, immune status and expression of several genes related to stress in gilthead seabream (*Sparus aurata* L.). *Fish & shellfish immunology*, 68, 251-259.
- Espinosa, C., Esteban, M. Á., & Cuesta, A. (2019). Dietary administration of PVC and PE microplastics produces histological damage, oxidative stress and immunoregulation in European sea bass (*Dicentrarchus labrax* L.). *Fish & shellfish immunology*, 95, 574-583.
- Herrera, A., Acosta-Dacal, A., Luzardo, O. P., Martínez, I., Rapp, J., Reinold, S., Montesdeoca-Esponda, S., Montero D & Gómez, M. (2022). Bioaccumulation of additives and chemical contaminants from environmental microplastics in European seabass (*Dicentrarchus labrax*). *Science of The Total Environment*, 822, 153396.
- Montero, D., Rimoldi, S., Torrecillas, S., Rapp, J., Moroni, F., Herrera, A., Gómez, M., Fernández-Montero, A. & Terova, G. (2022). Impact of polypropylene microplastics and chemical pollutants on European sea bass (*Dicentrarchus labrax*) gut microbiota and health. *Science of the Total Environment*, 805, 150402.
- Herrera, A., Acosta-Dacal, A., Pérez-Luzardo, O., Martínez, I., Rapp, J., Reinold, S., Montesdeoca-Esponda, S., Montero D. & Gómez, M. (2023). Trophic transfer of DDE, BP-3 and chlorpyrifos from microplastics to tissues in *Dicentrarchus labrax*. *Science of the Total Environment*, 882, 163295.