

## Study of bio-based nanomaterials inflammation potential in a zebrafish embryo model

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The increasing development of new Bio-Nano-Materials (Bio-NMs) requires animal models experimentation to demonstrate their biocompatibility and their interactions in biological *milieu*. To reduce or avoid the extensive use of higher vertebrates, the zebrafish embryos can be used as a well-suited model for testing materials at nanoscale and to support a safe-by-design strategy for new chemicals and materials.

Zebrafish embryos are considered a promising bridge model between *in vitro* and *in vivo* research, achieving the requirements of reduction and replacement in conducting experiments using full-grown animals.

The Bio-NMs developed need to be examined to evaluate their safety on humans and environmental organisms, which implies the absence of adverse effects such as acute toxicity and inflammation.

To test biological interactions and bioactive effects of Bio-NMs we used different methods: the classic Fish Embryo Toxicity acute (FET) test characterized by the presence of chorion, the natural barrier that envelopes and protect a fish embryo, and a modified FET test, in which the embryos are treated without the chorion.

By the use of classic and modified FET, we compared the effects of novel bio-based nanoparticles developed within the BIOMAT project (*i.e.* SiO<sub>2</sub>-NPs from rice husk), taking into consideration the inflammation as main adverse effect.

The inflammatory process is investigated by qPCR, analyzing the level of genes as il8, il6, il1 $\beta$ , tnf $\alpha$ , nfkbia, nfk2, known to have an important role within this pathway, and by the evaluation of neutrophils recruitment, taking advantage of the Sudan Black staining. The high level of genetic homology to humans, besides the simplicity of structures, makes the zebrafish embryos model a powerful system to predict and translate the effects observed on human beings, but at the same time, the model flexibility makes the results relevant also for environmental toxicology purposes. Correspondence: C. Bragato E-mail: cinzia.bragato@unimib.it

Funding: EU-H2020 project BIOMAT, GA n. 953270.

Conference presentation: this paper was presented at the Fourth Centro 3R Annual Meeting - The role of 3Rs in the age of One Health: where we are and where we're going - 13-15 September 2023, Università degli Studi Milano-Bicocca.

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