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## Effects of Polystyrene Nanoplastics on Zebrafish (*Danio rerio*) Embryos



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NTNU Gongguan Campus 臺灣師範大學公館校區理學院大樓F203 Host: Prof. Li-Yih Lin 林豊益教授

**Doctoral Dissertation Defense Presentation** 

~Attendee must wear mask~ ~與會者請配戴口罩~



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## Abstract

Plastic is the most widely used material in everyday life, and negligent litter management has resulted in growing amounts in the environment. Notably, nanoplastics (NPs) minuscule bits of plastic ranging in diameter from 5 to 100 nanometers – are hard to trace and have become a major source of pollution in the twenty-first century. Polystyrene (PS) is one of the most prevalent nanoplastics in the environment. In the laboratory, PS-NPs have been shown to penetrate the skin and accumulate in the organs of fish embryos. However, little is known about the potential impacts of PS-NPs on the skin functions (such as ion regulation and lateral line sensation) of fish embryos. In this study, the zebrafish embryo was used as an animal model to explore the toxic effects PS-NPs might pose to fish embryos.

In the first part of this study, zebrafish embryos were exposed to three distinct sizes of PS-NPs (25, 75, and 200 nm) for 96h or 120h. Several categories of endpoints were examined including developmental morphology (body length, body bending, somite length, eye size, pericardial cavity size and yolk sac size), physical activity (locomotor activity, optomotor response and tough-evoked response), and lateral-line sensory (hair cell number and morphology). Moreover, the mRNA expressions of neuro-marker genes (ache, syn2a, and mbp), and eye development genes (pax6a, pax6b, otx2, and rx1) were examined. The results showed that 50 mg/L of 25 nm PS-NPs caused significant adverse effects in all examined endpoints, suggesting that embryonic development was retarded and disturbed, and physical activity and sensory hair cells were impaired. At 25 mg /L of 25 nm PS-NPs, only physical activity and hair cells were significantly impaired. At 10 mg/L of 25 nm PS-NPs and all concentrations of 75, and 200 nm PS-NPs, the effects were insignificant in most endpoints.



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In the second part of this study, zebrafish embryos were exposed to 25 nm PS-NPs for 96 h to test the effects of PS-NPs on ion regulation by the skin cells (ionocytes and keratinocytes). After exposure to 50 mg/L PS-NPs, ion (Na<sup>+</sup>, K<sup>+</sup>, and Ca<sup>2+</sup>) contents, and acid/ammonia excretion by skin cells of embryos significantly declined. The apical structure of skin keratinocytes was damaged at 10, 25, and 50 mg/L. The number and mitochondrial activity of ionocytes were reduced at 25 and 50 mg/L. Reactive oxygen species (ROS) levels indicated by CellROX staining showed that both ionocytes and keratinocytes were under oxidative **PS-NPs** reduced the mRNA expression of stress. antioxidant genes (sod1, sod2, cat, and gpx1a), and promoted apoptosis-related genes (casp3a). Taken together, study suggests that PS-NPs might this suppress antioxidative reactions and induce oxidative stress, leading to mitochondrial damage and cell death of ionocytes, eventually impairing skin functions including ion uptake, pH regulation, and ammonia excretion.

In conclusion, the present study revealed the potential risk that PS-NPs might pose to fish embryos and the toxicological mechanisms that PS-NPs might cause from molecular and cellular to individual levels.

