# **Supplementary Information**

# Assessment of the Cytotoxicity Micro- and Nano-Plastic on Human Intestinal Caco-2 Cells and the Protective Effects of Catechin

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20 nm polystyrene nano-plastics at a concentration of 20 mg/mL.

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**Figure S1.** The morphology of PS plastic particles used in this study. (A) TEM image of 3  $\mu$ m PS-NP in culture medium; (B) TEM image of 0.3  $\mu$ m PS-NP in culture medium, (C) TEM image of 80 nm PS-NP in culture medium, (D) TEM image of 20 nm PS-NP in culture medium.



**Figure S2.** The composition of PS plastic particles used in this study. (A) FTIR spectrographs for the 3  $\mu$ m PS plastic particles, (B) FTIR spectrographs for the 0.3  $\mu$ m PS plastic particles, (C) FTIR spectrographs for the 80 nm PS plastic particles, (D) FTIR spectrographs for the 20 nm PS plastic particles.



**Figure S3.** The particle sizes distributions of the four types of PS plastic particles in culture medium. (A) The particle sizes distribution for 3  $\mu$ m PS-MP; (B) The particle sizes distribution for 0.3  $\mu$ m PS-MP, (C) The particle sizes distribution for 80 nm PS-NP; (D) The particle sizes distribution for 20 nm PS-NP.



**Figure S4.** Scavenging effect of different concentrations of N-acetyl-L-cysteine (NAC) on ROS induced by 20 nm polystyrene nano-plastics. The results presented are the means  $\pm$  SD from three independent experiments. \* means p < 0.05 (\*\* means p < 0.01), compared with the negative control group.



Figure S5. The protective effects of the four catechins against cytotoxicity induced by 20 nm polystyrene nano-plastics at a concentration of 20 mg/mL. The results presented are the means  $\pm$  SD from three independent experiments. \* means p < 0.05 (\*\* means p < 0.01), compared with the negative control group.

#### Text S1. Hazard quotient (HQ).

The hazard quotients (HQ) were calculated as follows <sup>1-2</sup>:

 $HQ = Measured serum level \times Uncertainty factor (UF) / PoD$ 

PoD is the lowest observed adverse effect level (LOAEL) (3  $\mu$ m: 0.01  $\mu$ g/mL, ~ 674 items/mL; 0.3  $\mu$ m: 0.01  $\mu$ g/mL, ~674,000 items/mL;80 nm: ~ 35.5 million items/mL; 20 nm: ~ 2.27 billion items/mL). The generally accepted default UF = 300 (3 × for inter-species interpolation, 10 × for human variability, and 10 × for LOAEL to NOAEL extrapolation), and that was used. An HQ below 1 indicates an absence of risk for the particular endpoint considered, whereas HQ greater than 1 indicates exposure that may be regarded as being of concern.

## **REFERENCES:**

1. Stoner, K. A.; Eitzer, B. D., Correction: Using a Hazard Quotient to Evaluate Pesticide Residues Detected in Pollen Trapped from Honey Bees (Apis mellifera) in Connecticut. *PLoS One* **2016**, *11* (7), e0159696.

2. Ludwicki, J. K.; Goralczyk, K.; Strucinski, P., et al., Hazard quotient profiles used as a risk assessment tool for PFOS and PFOA serum levels in three distinctive European populations. *Environ. Int.* **2015**, *74*, 112-8.