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The Effect of Blood Microplastic Particle Count on Corticosterone Hormone Level in Rattus Norvegicus Strain Wistar

I Made Aindrea Mahaputra Widiastawa✉, Patricia Gabrielle Tjipta Joewana, Yudhiakuari Sincihu, Shella Morina, Henry Ricardo Handoyo

Faculty of Medicine, Universitas Katolik Widya Mandala Surabaya

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Abstract

Microplastics, particularly Low-Density Polyethylene (LDPE), are environmental pollutants that can enter the body through oral exposure and potentially accumulate in the bloodstream. The presence of microplastics in systemic circulation is suspected to trigger physiological stress responses through activation of the hypothalamic–pituitary–adrenal axis. This study aimed to determine the effect of the number of microplastic particles in the blood on corticosterone hormone levels in Rattus norvegicus Wistar strain rats. This study employed an experimental design with a control group and treatment groups that received oral exposure to LDPE microplastics for 28 days. The number of microplastic particles in the blood was examined using light microscopy and polymer identification was confirmed using FTIR, while corticosterone levels were measured using the ELISA method. The results showed an increase in the number of microplastics in the blood following exposure, accompanied by elevated corticosterone levels as an indicator of physiological stress. This study recommends further investigation into the long-term effects of microplastics on the endocrine system and their implications for human health.

INTRODUCTION

Microplastics have emerged as a major global environmental concern due to the continuous increase in plastic production and inadequate waste management practices (Frias & Nash, 2019; Sharma & Chatterjee, 2017). Degradation of larger plastic debris through physical, chemical, and biological processes results in microplastic particles that persist in the environment and are readily ingested by living organisms (Galloway et al., 2017; Wright & Kelly, 2017).These particles have been detected in food, drinking water, air, and biological samples, raising serious concerns regarding their potential health effects (B. Smith, 2018). Low-Density Polyethylene (LDPE) is one of the most frequently identified microplastic polymers in the environment due to its extensive use in food packaging and single-use plastic products (Revel et al., 2018; Rochman et al., 2013). Beyond their physical presence, microplastics act as vectors for toxic substances, including heavy metals and persistent organic pollutants, thereby increasing their biological toxicity (Yong et al., 2020). Experimental studies have demonstrated that orally ingested microplastics can cross the intestinal barrier through Peyer's patches and paracellular transport mechanisms, subsequently entering systemic circulation (Deng et al., 2017; Stock et al., 2019).

Accumulation of microplastics in biological tissues has been associated with inflammatory responses, immune dysregulation, and increased production of reactive oxygen species (ROS), leading to oxidative stress (Herman et al., 2016; Wu et al., 2024). Persistent oxidative stress may disrupt cellular signaling pathways and impair endocrine function, particularly in stress-sensitive regulatory systems (Leslie et al., 2022).

One of the most sensitive systems to environmental stressors is the hypothalamic–pituitary–adrenal (HPA)

✉ yayaaindea911@yahoo.com,. Surabaya, East Java

axis, which regulates physiological stress responses through glucocorticoid secretion (Sapolsky, 2000). In rodents, corticosterone serves is the primary glucocorticoid hormone and is widely accepted as a biomarker of physiological stress induced by environmental and toxicological exposure (Anliana et al., 2025). Corticosterone secretion is regulated by circadian rhythms controlled by the central nervous system, and disruption of this regulation may exacerbate stress responses under chronic exposure conditions (Riederer & Laux, 2011). *Rattus norvegicus* of the Wistar strain is a well-established experimental model in biomedical and toxicological research due to its physiological similarity to humans, particularly in endocrine regulation and stress responses (Deng et al., 2017; Stock et al., 2019). Therefore, this model is relevant for evaluating the endocrine-disrupting potential of chronic microplastic exposure. This study was conducted to investigate the effect of microplastic particle abundance in the blood on corticosterone hormone levels in Wistar strain Rattus norvegicus rats.

RESEARCH METHOD

This study employed a post-test only control group experimental design conducted under laboratory conditions. Male *Rattus norvegicus* Wistar strain rats were divided into one control group and three treatment groups receiving oral LDPE microplastic exposure for 28 days at predetermined doses. Primary data were obtained from laboratory analysis of blood samples collected at the end of the exposure period.

Blood microplastic particles were isolated through tissue digestion, filtration, and microscopic observation using a binocular light microscope. The average particle count was calculated from multiple fields of view and expressed as particles per cubic centimeter of blood. Polymer identification was confirmed using Fourier Transform Infrared (FTIR) spectroscopy. Corticosterone hormone levels were measured using the enzyme-linked immunosorbent assay (ELISA) method, with absorbance read at 450 nm.

Statistical analysis was performed using SPSS software. Data distribution was assessed using the Shapiro–Wilk test, while homogeneity of variance was evaluated using Levene's test. Group comparisons were conducted using One-Way ANOVA for normally distributed data or the Kruskal–Wallis test for non-parametric data, followed by appropriate post hoc analyses. The relationship between blood microplastic particle counts and corticosterone levels was analyzed to evaluate exposure-related stress responses.

RESULTS AND DISCUSSION

Table 1 presents quantitative data on the number of microplastic particles detected in the blood of *Rattus norvegicus* of the Wistar strain in the control and treatment groups following 28 days of oral LDPE microplastic exposure. This table is used to compare the accumulation of microplastic particles in the systemic circulation among

Table 1
Microplastic Particle Count in the Blood

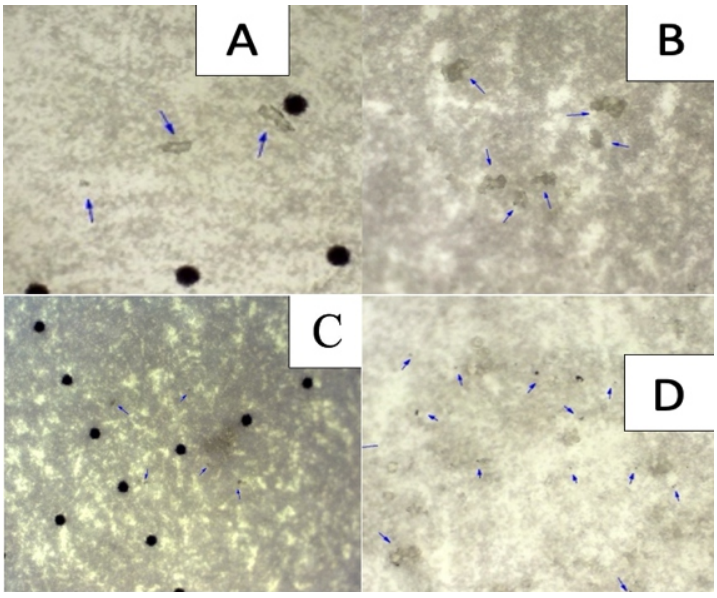
Group	Mean Microplastic Particle Count in Blood (Particles/cc of blood) (Mean±SD)
Control	**6.29 ± 1.11**
X1	**55.57 ± 3.69**
X2	**64.50 ± 2.58**
X3	**110.83 ± 11.16**

Source: Primary Data Processed, 2025

groups receiving different exposure doses. The presentation of these data aims to illustrate the distribution of blood microplastic particle counts resulting from microplastic exposure.

Based on Table 1, In the control group, the number of microplastics was the lowest compared to the experimental groups, at 6.29 particles/cc of blood, which can be considered a baseline (background level). In contrast, the treatment groups X1, X2, and X3 showed a linear increase in the number of microplastic particles in the blood as the exposure dose increased. In group X1, the mean value increased to 55.57 particles/cc of blood. Group X2 exhibited a further increase to 64.50 particles/cc of blood, while group X3 showed the highest level at 110.83 particles/cc of blood, which was markedly higher than that of the control group. These findings suggest that exposure to high doses of microplastics results in their substantial accumulation in the bloodstream. Subsequently, imaging of microplastics in the blood was performed using an XSZ-10107BN microscope and captured with an HDCE Uses camera (1280 × 1024 pixel HD CMOS) to observe the morphology of the microplastics.

The results of morphological observations of microplastic particles using microscopy in each treatment group (X0–X3) showed clear differences in the degree of physical degradation on the particle surfaces. In sample X0 (Figure A), the surface of the microplastics appeared relatively smooth and homogeneous, with only a very small number of small-sized particles observed. The identified fragments (indicated by blue arrows) were sparsely distributed and tended to have a uniform shape. This condition indicates that the microplastics were at an early stage, with no significant degradation process occurring. In sample X1 (Figure B), an increase in the number of small particles was observed, along with changes in surface texture that appeared more opaque and uneven compared to X0. This finding indicates that the microplastics in this treatment had undergone morphological degradation, characterized by the emergence of microcracks and fine fragments as a result of the exposure process. In sample X2 (Figure C), the number of observed particles increased further, with a more uniform distribution across the entire field of view. The surface of the microplastics showed more pronounced textural changes, with clear



Source: Primary Data Processed, (2025)

Figure 1
Image of Microplastic Particles In The Blood

Table 2
Results of blood corticosterone hormone level measurements

Variable	Group	Mean ± SD (µg/mL)
Corticosterone Hormone Level	X0	8.85 ± 3.07
	X1	15.32 ± 1.98
	X2	16.56 ± 4.82
	X3	19.04 ± 6.03

Source: Processed Primary Data, 2025

Table 2 presents the blood corticosterone concentrations of Wistar strain Rattus norvegicus rats in the control and treatment groups after 28 consecutive days of oral exposure to LDPE. The data presented in this table were used to compare corticosterone levels across groups exposed to different doses of microplastics. The presentation of this table aims to provide an overview of corticosterone hormone levels as an indicator of physiological stress resulting from microplastic exposure.

Based on data processing using the SPSS, the lowest corticosterone level was found in group X0, with a mean value of 8.8 and a standard deviation of ± 3.07. In group X1, corticosterone levels increased to 15.32 with a standard deviation of ± 1.98, indicating an initial response to the administered exposure. In group X2, corticosterone levels continued to rise to 16.56 with a standard deviation of ± 4.82, suggesting that the increased treatment dose began to exert a more pronounced effect on stress hormone activity. Meanwhile, the highest mean corticosterone level was observed in group X3, at 19.0 with a standard deviation of ± 6.03. Overall, an increase in corticosterone levels was evident in groups X1, X2, and X3 compared with the control group.

The average of microplastic particles in the blood showed an increasing trend with rising exposure doses. The control group had an average of 6.29 particles/cc of blood, while the treatment groups X1, X2, and X3 recorded averages of 55.57, 64.50, and 110.83 particles/cc of blood, respectively. This pattern illustrates a linear relationship between increasing microplastic doses and the number of particles detected in the systemic circulation of male Wistar rats.

The findings of this study demonstrate a clear dose-dependent relationship between blood microplastic particle accumulation and elevated corticosterone hormone levels, indicating sustained activation of physiological stress pathways. Increased corticosterone concentrations reflect prolonged activation of the hypothalamic –pituitary–adrenal (HPA) axis in response to foreign particle exposure (Herman et al., 2016; Sapolsky, 2000). Similar endocrine stress responses have been reported in experimental studies investigating chronic exposure to environmental pollutants and microplastics (Lu et al., 2018; Prata et al., 2020). The detection of microplastic particles in the control group suggests the possibility of environmental or laboratory cross-contamination, a phenomenon frequently reported in microplastic research due to their ubiquitous distribution (Chen et al., 2020; Leslie et al., 2022; Zhang et al., 2020). Microplastics are lightweight, easily aerosolized, and capable of adhering to laboratory equipment, feed, and water sources, potentially introducing background exposure even in non-treated groups (Revel et al., 2018; M. Smith et al., 2018).

Once present in systemic circulation, microplastic particles may interact directly with blood cells or adhere to erythrocyte membranes, triggering inflammatory signaling pathways and oxidative stress (Deng et al., 2017; Leslie et al., 2022). These processes may stimulate hypothalamic release of corticotropin-releasing hormone (CRH), followed by

adrenocorticotropic hormone (ACTH) secretion from the anterior pituitary, ultimately leading to increased corticosterone release from the adrenal cortex (Anliana et al., 2025; Sapolsky, 2000). Chronic disruption of gluco-corticoid regulation may impair circadian homeostasis and contribute to long-term endocrine imbalance, particularly under continuous low-dose exposure scenarios (Riederer & Laux, 2011). These findings support growing evidence that microplastics function not only as physical contaminants but also as endocrine-disrupting agents with potential implications for human health (Prata et al., 2020; Wright & Kelly, 2017).

CONCLUSION

Chronic oral exposure to LDPE increases the accumulation of microplastic particles in the blood and elevates corticosterone hormone levels in *Rattus norvegicus* of the Wistar strain. Exposure at doses exceeding 5 mg/day for 28 days induces a significant physiological stress response, indicating disruption of endocrine homeostasis through activation of the hypothalamic–pituitary–adrenal (HPA) axis. Therefore, further studies are required to evaluate long-term exposure effects, include additional endocrine and inflammatory biomarkers, assess histopathological changes, and examine the implications for human health.

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