THE GARCINIA MANGOSTANA L. PEEL EXTRACT REDUCES FECAL MICROBIOTA COUNT AND MALONDIALDEHYDE EXPRESSION OF INTESTINAL EPITHELIUM IN MICROPLASTIC-INDUCED RATS

Yudhiakuari Sincihua[1], FX Himawan Haryanto Jong[1], Alvin Julian[1], Herjunianto[1], Novera Herdiani[2], Tri Marthy Mulyasari[3], Roy Hendroko Setyobudi[4], Diah Hermayanti[4], Kusuma Scorpia Lestari[5], Prehatin Trirahayu Ningrum[6]

*Corresponding author email: yudhiakuari@ukwms.ac.id

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ABSTRACT

***Background**: Microplastics (MP) are emerging food contaminants that can penetrate the bloodstream and induce oxidative stress and inflammation. Intestinal microbiome dysbiosis and damage to the intestinal epithelial phospholipid membrane are considered key biomarkers of MP-induced toxicity in the gastrointestinal tract.* ***Objective**: This study aimed to evaluate the potential of Garcinia mangostana L. peel extract (MPE) to maintain gut microbiome homeostasis and protect the intestinal epithelium of Wistar rats exposed to low-density polyethylene (LDPE) microplastics.* ***Methods**: This was a true experimental study involving 35 Wistar rats randomly assigned to five groups: negative control (KN), positive control (KP), and three treatment groups (K1, K2, and K3). LDPE MPs were administered orally at a dose of 5 mg/day to the KP, K1, K2, and K3 groups. MPE was administered orally to the treatment groups at doses of 200 mg/kgBW/day (K1), 400 mg/kgBW/day (K2), and 600 mg/kgBW/day (K3).* ***Results**: One-way ANOVA demonstrated that MPE significantly reduced intestinal microbiome dysbiosis ($p = 0.001$; 95% CI), as indicated by mean total plate count values (CFU/mL × 10^6): KN = 8.37 ± 0.90; KP = 11.70 ± 0.99; K1 = 11.46 ± 0.84; K2 = 8.65 ± 1.20; and K3 = 9.39 ± 0.47. MPE also significantly attenuated intestinal epithelial membrane damage ($p = 0.001$; 95% CI), as shown by mean malondialdehyde expression levels (% immunoreactive area): KN = 11.84 ± 0.91; KP = 25.84 ± 3.24; K1 = 21.08 ± 6.59; K2 = 18.65 ± 3.22; and K3 = 14.09 ± 1.71.* ***Conclusion**: These findings suggest that Garcinia mangostana L. peel extract has protective potential in preserving gut microbiome homeostasis and preventing intestinal epithelial damage induced by microplastic exposure.*

***Keywords**: Dysbiosis; Garcinia mangostana L; Intestinal epithelium; Malondialdehyde; Microplastics.*

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INTRODUCTION

Every day, humans may ingest approximately 126–142 microplastic (MP) particles in adults and 106–113 particles in children.[1] These MPs originate from various food sources, including salt, sugar, canned sardines, marine fish, livestock meat, honey, tea bags, drinking water, and foods stored in plastic containers.[2,3,4] As plastic waste continues to accumulate in aquatic and terrestrial environments, human exposure to MPs through dietary intake is expected to increase steadily.[1,5] Indonesia ranks as the world's second-largest contributor of plastic waste after China,[5] and inadequate waste management accelerates MP contamination of food sources, posing a largely unrecognized risk to public health.

The primary health concern associated with MP ingestion is its long-term impact on the gastrointestinal system, which represents the main route of exposure. Evidence indicates that MPs are present in approximately 80% of human blood samples,[6] highlighting their ability to translocate

[1] *Widya Mandala Surabaya Catholic University, Jl. Kalisari No. 1, Surabaya 60272, Indonesia*
[2] *Nahdlatul Ulama University Surabaya, Jl. Raya Jemursari No. 57, Surabaya 60237, Indonesia*
[3] *Health Polytechnic of the Ministry of Health Semarang, Jl. Tirto Agung, Semarang 50268, Indonesia*
[4] *Muhammadiyah University of Malang, Jl. Raya Tlogomas No. 246, Malang 65144, Indonesia*
[5] *Airlangga University, Jl. Mulyorejo, Surabaya 60115, Indonesia*
[6] *University of Jember, Jl. Kalimantan Tegalboto No. 37, Jember 68121, Indonesia*

beyond the intestinal lumen and accumulate systemically. Following ingestion, MPs in the gastrointestinal tract may cross the intestinal barrier through phagocytosis and enter the bloodstream and lymphatic system.[7] The integrity of the intestinal epithelium therefore plays a critical role as the first line of defense against MP absorption.[8] This epithelial barrier is maintained by tight junctions between epithelial cells, which limit paracellular permeability and restrict the entry of toxic substances into the body.[6,9] The gut microbiota is a key modulator of intestinal barrier integrity, as it supports tight junction function and maintains intestinal permeability. Disruption of this microbial balance, known as intestinal dysbiosis, weakens epithelial defense mechanisms and increases intestinal permeability, thereby facilitating the uptake and endocytosis of MPs and their associated toxic compounds.[6,9] Consequently, intestinal microbiome dysbiosis represents a critical biological variable linking MP exposure to epithelial injury and systemic toxicity.

Once MPs enter the circulation, they cannot be degraded by oxygen-dependent or oxygen-independent cellular defense mechanisms.[10] Their persistence triggers oxidative stress and chronic inflammatory responses, which further compromise intestinal epithelial integrity.[9] This inflammatory cascade is driven by three major sources of toxicity, including plastic monomers, toxic additives introduced during manufacturing, and environmental pollutants adsorbed onto the MP surface.[10,11] These toxic constituents circulate throughout the body together with MPs, which have been detected in multiple organs, including the liver, lungs, heart, muscles, kidneys, and brain.[11,12] Although some MPs are excreted via feces, most are polyethylene polymers.[13] Sustained oxidative stress and inflammation can lead to widespread cellular injury and death.[12,14] One well-established biomarker of phospholipid membrane damage in epithelial cells under oxidative stress conditions is malondialdehyde (MDA) expression.[12,15]

To date, no effective strategy exists to eliminate MPs once they have entered the body, underscoring the urgency of identifying protective or mitigative interventions. Phytopharmaceutical compounds have gained attention as potential therapeutic agents due to their antioxidant and anti-inflammatory properties. Among these, mangosteen (*Garcinia mangostana* L.) peel extract has emerged as a promising candidate. This extract contains bioactive compounds such as xanthones, polyphenols, flavonoids, steroids, terpenoids, alkaloids, and saponins, which are known to exert antioxidant, anti-inflammatory, antiallergic, and microbiome-modulating effects.[16] The major constituents, α-mangostin and γ-mangostin, play a central role in mediating these biological activities.[17,18] Xanthones suppress inflammatory responses by inhibiting cyclooxygenase and lipoxygenase enzymes, while flavonoids reduce inflammation by inhibiting arachidonic acid release.[19,20] Polyphenols further contribute by neutralizing free radicals through electron donation, thereby limiting oxidative damage to cellular membranes.[21]

Based on these mechanisms, this study hypothesizes that mangosteen peel extract can reduce fecal microbiota counts as an indicator of intestinal microbiome dysbiosis and reduce MDA expression as an indicator of phospholipid membrane damage in intestinal epithelial cells in experimental animals exposed to oral microplastics. This investigation focuses on the interaction between MP exposure, gut microbiome balance, epithelial oxidative damage, and phytopharmaceutical intervention, constituting the primary objective and novelty of the present study.

METHODS

Research Design and Experimental Units

This study employed a true experimental design with a post-test-only control group. The experimental procedures were conducted from July 21 to August 26, 2025, at the Animal Laboratory of Widya Mandala Catholic University Surabaya.

The experimental units consisted of healthy male *Rattus norvegicus* (Wistar strain), approximately 12 weeks of age (sexually mature), weighing 170–180 g, and certified as disease-free by an official laboratory animal health certificate. Animals were randomly allocated into five groups using a computer-generated randomization method: negative control (KN), positive control (KP), and three treatment groups exposed to microplastics (K1, K2, and K3).

Sample size was determined using Lemeshow's formula (α = 0.05; 95% confidence interval), resulting in seven rats per group. Throughout the experimental period, animal health

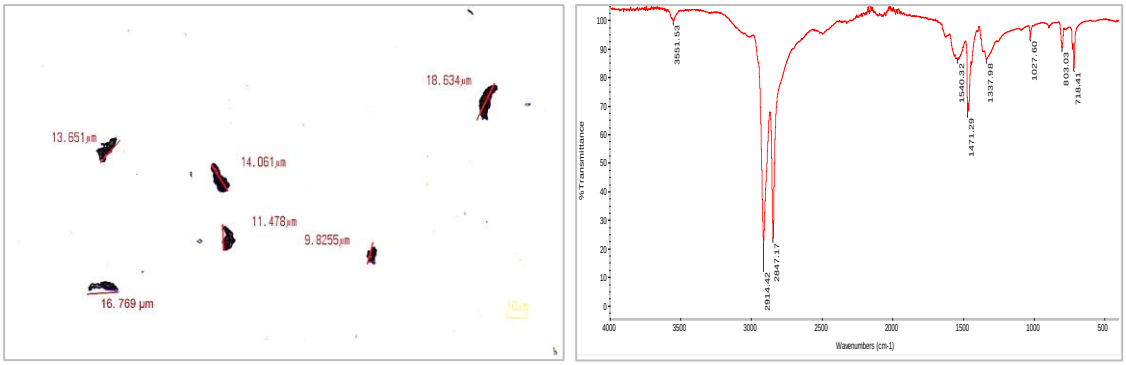


Figure 1. Diameter of LDPE Microplastics (MPs) and FTIR Analysis Results

and welfare were monitored under the supervision of a veterinarian. Normality and homogeneity of baseline body weight data were confirmed using the Shapiro–Wilk test ($p > 0.05$) and Levene's test ($p = 0.386$), indicating that the data were normally distributed with homogeneous variances.

Research Variables

Low-density polyethylene (LDPE) microplastics and mangosteen peel extract (MPE) were defined as the independent variables in this study. The dependent variables were intestinal microbiota abundance and malondialdehyde (MDA) expression in intestinal epithelial cells. Controlled variables included the route of MP administration (oral gavage), form of mangosteen peel extract (powder), experimental animals (*Rattus norvegicus*, Wistar strain), standardized mixed feed, distilled water (aquadest), and uniform animal housing and care procedures.

LDPE Microplastic Exposure Material

LDPE-type microplastics were selected because they represent one of the most prevalent plastic contaminants detected in food products.22 The MPs were produced by mechanically grinding LDPE plastic using a Bosch GWS 6-100 grinding machine, followed by particle size separation through an 800-mesh sieve (pore size 18 µm). The resulting particle size distribution was examined under a Nikon Eclipse binocular microscope at 100× magnification using a 10 µm scale bar. The polymer composition was confirmed using Fourier Transform Infrared (FTIR) spectroscopy to ensure the identity and purity of LDPE microplastics prior to experimental use.

Based on Figure 1, the LDPE microplastic (MP) particles used in this study had diameters smaller than 18.634 µm, indicating their potential to penetrate biological barriers and translocate into the bloodstream. Fourier Transform Infrared (FTIR) analysis conducted at the Institut Teknologi Surabaya further confirmed that the exposure material was composed of LDPE polymer.

Mangosteen Peel Extract (MPE) Material

Mangosteen peel extract (MPE) was obtained from PT Zena Nirmala Sentosa under the trade name Garcia® (Registration No. POM TR.113328811). Each capsule contained 400 mg of MPE powder. The extract powder was incorporated into feed dough at the BioCalorix Factory according to the predetermined dosage, then pelletized, dried, and packaged for experimental use.

MPE was administered at graded doses: 200 mg/kg body weight/day (K1), 400 mg/kg body weight/day (K2), and 600 mg/kg body weight/day (K3). The negative control (KN) and positive control

Table 1. Daily Feed Consumption Records of Experimental Rats

Group	n (rats)	$\bar{x}$ ± SD Feed Consumption (g/day)	Estimated MPE Dose Consumed (mg/day)
K1	7	17.12 ± 0.62	29.96
K2	6	17.05 ± 0.65	59.69
K3	6	17.14 ± 0.82	90.00

(KP) groups received standard feed without MPE supplementation. Dose selection was based on the study by Moongkarndi et al.[23], which demonstrated that MPE doses ranging from 200 to 600 mg/kg body weight effectively enhanced endogenous antioxidant activity and reduced free radical formation in experimental animals.

In this study, MPE was mixed into the daily feed ration (20 g per rat), yielding equivalent daily doses of 35 mg MPE/rat (K1), 70 mg MPE/rat (K2), and 105 mg MPE/rat (K3). Feed intake for each rat was recorded daily over a 28-day period to ensure accurate dose administration and compliance with the intended treatment protocol. Additional details are provided in Table 1.

Based on Table 1, the actual MPE doses administered in this study were K1 = 29.96 mg MPE/day, K2 = 59.69 mg MPE/day, and K3 = 90 mg MPE/day. These doses are consistent with previous studies that evaluated the effectiveness of mangosteen peel extract on antioxidant and anti-inflammatory activity in *Rattus norvegicus* Wistar strain experimental animals.[24]

Adaptation, Treatment, and Termination of Experimental Units

The experimental subjects were acclimatized for 7 days in the Animal Laboratory. Rats were housed individually in cages measuring 15 × 20 × 15 cm, with one rat per cage. Each cage was provided with 20 g of processed feed mixed with MPE daily. Drinking water was supplied in 100 mL rat bottles and replaced every day.

During the maintenance period, environmental conditions were kept clean and controlled, with wood shavings as bedding, stable air circulation, room temperature maintained at 20–25°C, and humidity at 50–60%. The mixture of microplastic (MPs) solution and distilled water (aquadest) was administered orally using a gavage tube according to the prescribed research dosage.

A total of three experimental animals were dropped out due to mortality during the study period. After 28 days of treatment, fecal samples were collected, and the experimental animals were terminated by a veterinarian from the Laboratorium Satwa Sehat, Malang City. Termination was performed using the cervical dislocation technique.

An abdominal dissection was then carried out to collect the small intestine, which was placed into tissue preservation tubes. Subsequently, both fecal and intestinal tissue samples were examined according to standard laboratory procedures at the Laboratorium Satwa Sehat.

After sample collection, the rat carcasses were placed in sealed containers and buried following ethical disposal guidelines.

Fecal Total Plate Count (TPC) Analysis

Quantification of intestinal microbiota was performed using the Total Plate Count method on fecal samples. Prior to sample collection, all experimental animals were clinically examined by a licensed veterinarian to ensure the absence of conditions that could affect microbiota composition. Fresh fecal samples were collected directly from

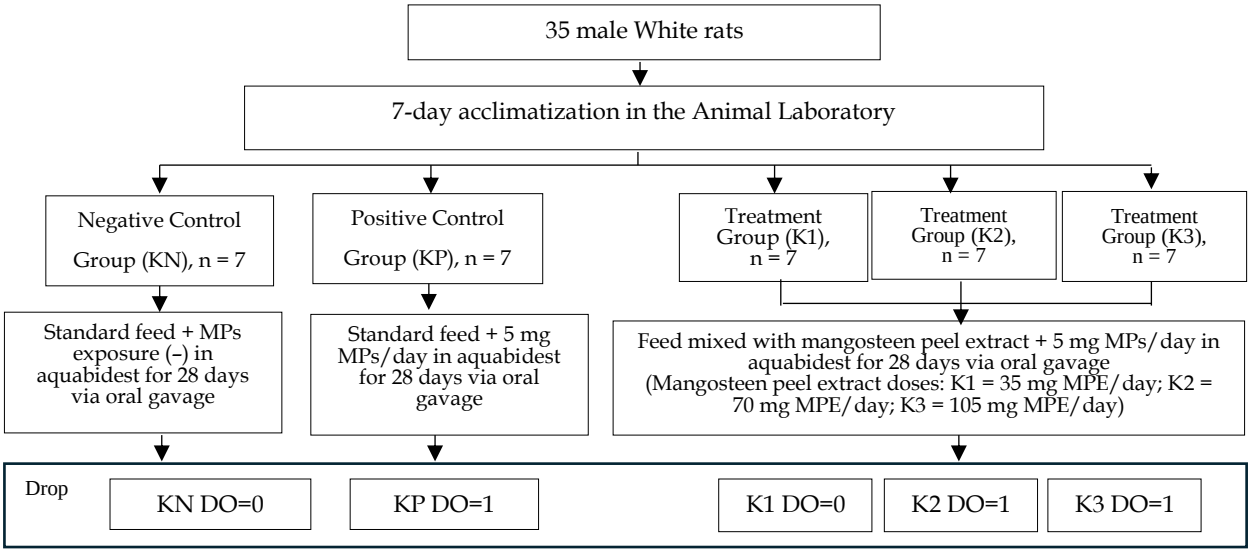


Figure 2. Experimental Animal Care, Treatment Administration, and Dropout

each rat at the end of the intervention period to minimize environmental contamination. Approximately 1 g of feces was aseptically weighed and homogenized in 9 mL of sterile physiological saline (0.9% NaCl) to obtain a 10^{-1} dilution. Serial tenfold dilutions were then prepared up to 10^{-6} under aseptic conditions. From each selected dilution, 0.1 mL of the suspension was inoculated onto Plate Count Agar (PCA) using the spread plate technique. The inoculated plates were incubated aerobically at 37°C for 24–48 hours. Colonies were counted on plates containing 30–300 colonies using a digital colony counter. The total viable bacterial count was calculated as colony-forming units per gram of feces (CFU/g) and expressed as $\log_{10}$ CFU/g for statistical analysis. All analyses were performed in duplicate to ensure data reliability. The digital colony counter functions solely as a counting device for visible colonies formed on agar plates. The instrument does not differentiate between aerobic bacteria, anaerobic bacteria, fungi, or viruses.

Immunohistochemical Analysis of Malondialdehyde (MDA) Expression

Malondialdehyde expression in intestinal tissue was assessed using immunohistochemical (IHC) staining. Intestinal tissue samples were fixed in 10% neutral-buffered formalin, embedded in paraffin, and sectioned at a thickness of 4–5 µm. Tissue sections were deparaffinized, rehydrated, and subjected to heat-induced antigen retrieval. Endogenous peroxidase activity was blocked using hydrogen peroxide, followed by incubation with a primary anti-MDA antibody (Abcam, Cambridge, UK), according to the manufacturer's protocol.

Subsequently, sections were incubated with a horseradish peroxidase–conjugated secondary antibody, and immunoreactivity was visualized using 3,3′-diaminobenzidine (DAB) chromogen (Dako, Denmark). Slides were counterstained with hematoxylin, mounted, and examined under a light microscope. MDA expression was quantified based on the intensity and distribution of positive brown staining in intestinal epithelial cells for statistical analysis.

Ethical Clearance

This study obtained ethical approval from the Health Research Ethical Clearance Commission (HRECC), Faculty of Dental Medicine, Universitas Airlangga, under approval number 0950/HRECC.FODM/IX/2025. The ethical review and approval process complied with institutional regulations and included members with relevant expertise in animal research, comprising licensed veterinarians and professors of biochemistry and biomedical sciences.

All experimental procedures were conducted in strict accordance with the National Guidelines for the Ethical Conduct of Animal Research issued by the Ministry of Health of the Republic of Indonesia. Animal handling, housing, experimental interventions, and endpoints were designed to ensure animal welfare and to minimize pain, distress, and unnecessary suffering throughout the study.

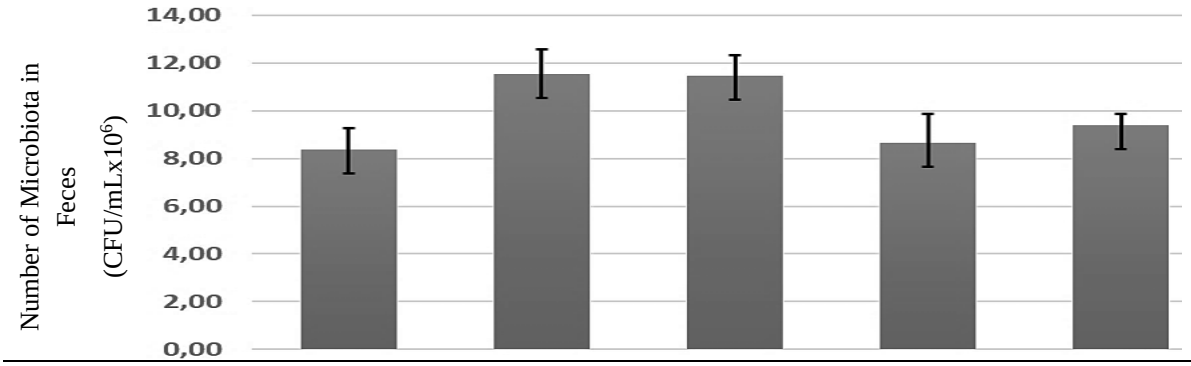
RESULT AND DISCUSSION

Effect of Mangosteen Peel Extract (MPE) on Intestinal Microbiome in Rattus norvegicus

The number of intestinal microbiota was used as an assessment variable to reflect changes in the gastrointestinal microbial environment. Intestinal microbiota plays a crucial role in maintaining gut barrier integrity, nutrient metabolism, and immune system regulation.[9] Exposure to microplastics may alter the composition and abundance of intestinal microbiota by promoting the growth of pathogenic microorganisms. These alterations are associated with increased oxidative stress and mucosal inflammation and represent one of the biological mechanisms underlying MP-induced toxicity in the gastrointestinal tract.[8]

The number of intestinal microbiota was quantified using the Total Plate Count (TPC) method on fecal samples of *Rattus norvegicus*. Microbial counts were expressed as CFU/mL×10^6. Mean ($\bar{x}$) and standard deviation (SD) values of microbiota counts are presented in Figure 3. Microscopic images of microbial colonies were obtained using Image Raster Software on a Nikon Eclipse Ei® light microscope, equipped with an Optilab SIGMA MTN020 imaging system at 40× magnification. Representative images are shown in Figure 4.

Based on Figure 3, the highest mean fecal microbiota count was observed in the positive control group (KP), which was exposed to MPs without MPE supplementation, at 11.70 ± 0.99 CFU/mL×10^6. In contrast, the negative control group (KN), which received neither MPs nor MPE, showed



Group	KN	KP	K1	K2	K3
n (rats)	7	6	7	6	6
x̄ ± SD	8.37 ± 0.90	11.70 ± 0.99	11.46 ± 0.84	8.65 ± 1.20	9.39 ± 0.47

Figure 3. Number of Microbiota in Rattus norvegicus

a lower mean microbiota count of 8.37 ± 0.90 CFU/mL×10⁶.

In the MP-exposed group receiving the lowest dose of MPE (K1), the mean fecal microbiota count was slightly lower than that of the KP group, at 11.46 ± 0.84 CFU/mL×10⁶. A decreasing trend in mean fecal microbiota counts was observed with increasing MPE dosage, with values of 11.46 ± 0.84 in K1, 8.65 ± 1.20 in K2, and 9.39 ± 0.47 CFU/mL × 10⁶ in K3. Statistical analysis was subsequently performed, and the results are presented in Table 2.

Normality testing using the Shapiro–Wilk test ($p > 0.05$) indicated that fecal microbiota counts were normally distributed, while homogeneity of variance was confirmed by Levene's test ($p = 0.593$). One-way ANOVA demonstrated a statistically significant difference in fecal microbiota counts among all experimental groups of *Rattus norvegicus* (Wistar strain) ($p = 0.001$).

The negative control group (KN), which was not exposed to microplastics (MP) or mangosteen peel extract (MPE), exhibited the lowest fecal microbiota counts, reflecting a stable intestinal microbial environment under physiological conditions. In contrast, the positive control group (KP) showed a marked increase in microbiota counts,

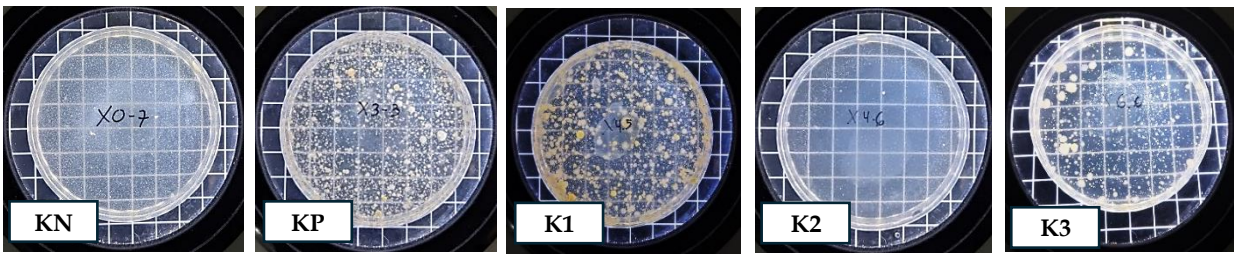


Figure 4. Abundance of Fecal Microbiota in Rattus norvegicus, 40× Magnification
Note: **KN** = negative control group; **KP** = group exposed to 5 mg MPs/day; **K1** = group exposed to 5 mg MPs + 35 mg MPE/day; **K2** = group exposed to 5 mg MPs + 70 mg MPE/day; **K3** = group exposed to 5 mg MPs + 105 mg MPE/day. Colony growth appeared as white or colored spots on the Petri plates.

Table 2. Normality Test, Homogeneity Test, and Mean Comparison of Fecal Microbiota Counts

Group	Normality test	Homogeneity test	One Way Anova
KN	.894		
KP	.467		
K1	.907	0.593	0.001
K2	.920		
K3	.334		

indicating that MP exposure alters the intestinal microbial ecosystem. This alteration may be explained by three interrelated mechanisms: oxidative stress, mucosal inflammation, and changes in microbial metabolic activity.[25,26] Collectively, these processes induce alterations in the intestinal microbial community, reflected by changes in the abundance of specific microbial taxa following gut environmental disturbance.[26,27]

The primary mechanism involves oxidative stress in the intestinal mucosa. The charged surface of MPs, enriched with carbonyl functional groups, can generate reactive oxygen species (ROS) either directly or indirectly through activation of local immune responses.[28] Elevated ROS levels activate inflammatory signaling pathways such as TLR2/NF-κB/NLRP3, leading to the release of pro-inflammatory cytokines (IL-1β, TNF-α, and IL-6) and creating a pro-oxidative intestinal environment.[25] These conditions modify luminal pH and oxygen tension, suppressing obligate anaerobic bacteria (e.g., *Bacteroides* and *Lactobacillus*) while favoring facultative anaerobes (e.g., *Escherichia* and *Enterococcus*) that are more tolerant of oxidative stress.

Secondly, MPs compromise intestinal epithelial barrier integrity by disrupting tight junction proteins, including occludin, claudins, and zonula occludens-1 (ZO-1), thereby increasing intestinal permeability (leaky gut).[27] This condition facilitates the translocation of luminal antigens, endotoxins, and microorganisms into the lamina propria, perpetuating low-grade chronic inflammation and further disturbing the intestinal microbial balance.

The third mechanism involves alterations in microbial metabolic function. MP exposure has been reported to reduce the production of short-chain fatty acids (SCFAs), such as butyrate and propionate, which are essential for maintaining mucosal integrity and immune regulation.[26] Reduced SCFA availability impairs GPR41/GPR43 signaling and disrupts enterocyte energy homeostasis, thereby sustaining inflammation and unfavorable shifts in the intestinal microbial ecosystem.

In contrast, administration of MPE at medium and high doses (K2 and K3) attenuated MP-induced alterations in fecal microbiota counts. The polyphenolic compounds in MPE, particularly xanthones, possess strong antioxidant and anti-inflammatory properties that may reduce oxidative stress and support restoration of epithelial barrier function, thereby promoting a more regulated intestinal microbial environment.[29,30] Additionally, previous studies have shown that MPE exhibits selective antimicrobial activity against pathogenic bacteria and can modulate gut microbial composition through activation of the AhR and Nrf2 signaling pathways, which play key roles in intestinal epithelial protection.[29,31,32]

Overall, these findings align with previous research, showing that medium and high doses of MPE contribute to the reduction of intestinal microbiota abundance under MP exposure conditions through combined antioxidant, anti-inflammatory, and antimicrobial mechanisms, thereby supporting intestinal mucosal integrity and function.

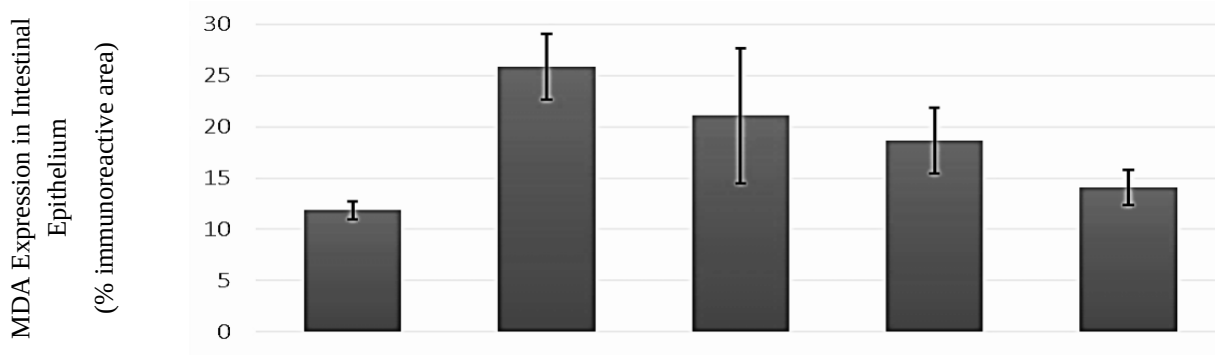
Effect of Mangosteen Peel Extract (MPE) on Malondialdehyde (MDA) Expression in Intestinal Epithelium of Rattus norvegicus

Malondialdehyde expression in the intestinal epithelium was used as an assessment variable because MDA is a terminal product of lipid peroxidation. Elevated MDA levels reflect increased oxidative stress resulting from the accumulation of free radicals that damage membrane lipids.[6,14] Therefore, MDA serves as an established biomarker for evaluating the extent of oxidative damage induced by microplastic exposure.

MDA expression in the intestinal epithelium was assessed using immunohistochemical staining. Expression levels were quantified as the percentage of immunoreactive area across five randomly selected microscopic fields. The mean ($\bar{x}$) and standard deviation (SD) of MDA expression are presented in Figure 5.

Representative microscopic images of MDA expression in intestinal epithelial cells were captured using Image Raster Software on a Nikon Eclipse Ei® light microscope, equipped with an Optilab SIGMA MTN020 imaging system, at 400× magnification. The images are shown in Figure 6.

As shown in Figure 5, the highest mean MDA expression in the intestinal epithelium was observed in the positive control group (KP), which was exposed to MPs without mangosteen peel extract (MPE), with a value of 25.84 ± 3.24%



Group	KN	KP	K1	K2	K3
n (rats)	7	6	7	6	6
x̄ ± SD	11.84 ± 0.91	25.84 ± 3.24	21.08 ± 6.59	18.65 ± 3.22	14.09 ± 1.71

Figure 5. MDA Expression in Intestinal Epithelium of Rattus norvegicus

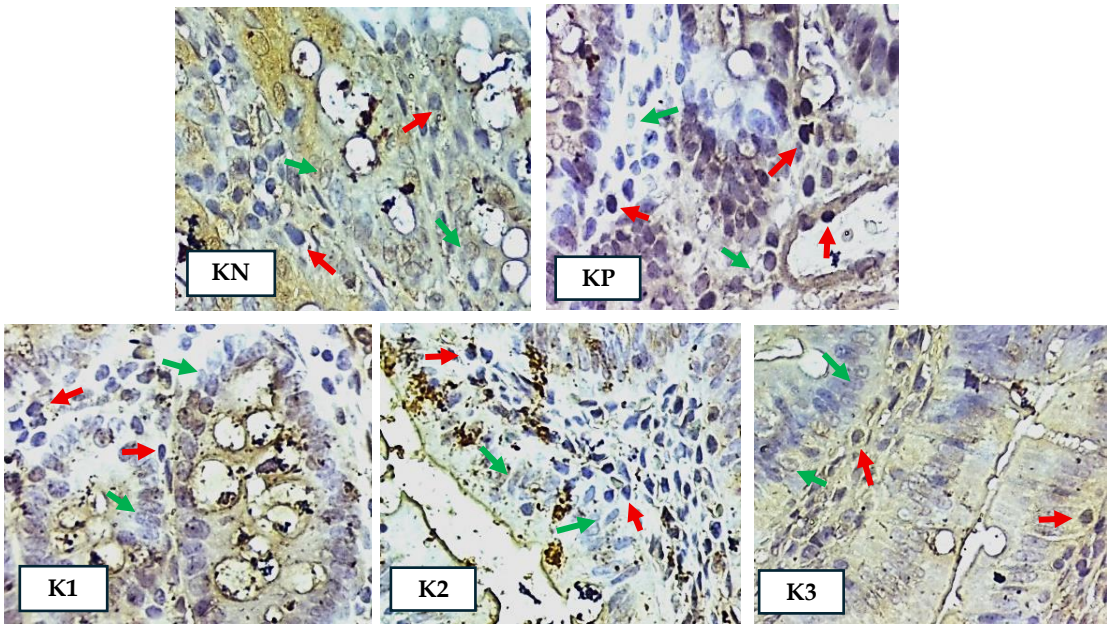


Figure 6. MDA Expression in Intestinal Epithelium of Rattus norvegicus, 400× Magnification
Note: **KN** = negative control group; **KP** = group exposed to 5 mg MPs/day; **K1** = group exposed to 5 mg MPs + 35 mg MPE/day; **K2** = group exposed to 5 mg MPs + 70 mg MPE/day; **K3** = group exposed to 5 mg MPs + 105 mg MPE/day.
Epithelial cells showing high MDA activity (red arrows) and low MDA activity (green arrows)

immunoreactive area. In contrast, the negative control group (KN), which received neither MPs nor MPE, exhibited the lowest MDA expression at 11.84 ± 0.91%.

In the MP-exposed groups receiving MPE, the mean percentage of MDA immunoreactive area was lower than that of the KP group and demonstrated a dose-dependent reduction with increasing MPE dosage: 21.08 ± 6.59% in K1, 18.65 ± 3.22% in K2, and 14.09 ± 1.71% in K3. Notably, MDA expression in the K3 group, which received the highest MPE dose, was comparable to that observed in the KN group. Statistical analyses were subsequently performed, and the results are presented in Table 3.

Normality testing using the Shapiro–Wilk test ($p > 0.05$) indicated that MDA expression in the intestinal epithelium was normally distributed. However, homogeneity of variance testing using Levene's test ($p = 0.001$) demonstrated unequal variances among groups. Therefore, the Brown–

Table 3. Normality Test, Homogeneity Test, and Mean Comparison of MDA Expression in Intestinal Epithelium

Group	Normality test	Homogeneity test	One Way Anova
KN	.487		
KP	.356		
K1	.082	0.001	0.001
K2	.962		
K3	.395		

Forsythe test was applied and revealed a statistically significant difference in MDA expression among all experimental groups of *Rattus norvegicus* (Wistar strain) ($p = 0.001$).

These findings indicate that exposure to MPs significantly increases MDA formation in intestinal tissues, reflecting enhanced lipid peroxidation as a consequence of oxidative stress. Malondialdehyde, as the terminal product of membrane lipid peroxidation induced by ROS, serves as a reliable biomarker of cellular oxidative damage. The elevated MDA expression observed in the KP group suggests that MP promote ROS accumulation in the intestinal mucosa through multiple mechanisms, including activation of the TLR4/NF-κB/NLRP3 signaling pathway and disruption of the balance between endogenous antioxidant defenses (superoxide dismutase, glutathione peroxidase, and catalase) and free radical generation.[25,27] In addition, the negatively charged and hydrophobic surfaces of MPs may directly interact with enterocyte membranes, further inducing oxidative stress and local inflammation, ultimately leading to lipid peroxidation.[33,34]

In contrast, in the groups receiving combined MP exposure and MPE supplementation, MDA expression decreased in a dose-dependent manner, indicating that higher MPE doses were associated with reduced oxidative damage in intestinal tissues. Notably, the mean MDA value in the K3 group (105 mg MPE/day) was comparable to that of the KN group, suggesting a substantial protective effect of MPE against MP-induced oxidative stress.

This protective effect is likely mediated by xanthone compounds present in mangosteen peel, including α-mangostin, γ-mangostin, and garcinone E, which exhibit strong antioxidant activity through free radical scavenging, enhancement of endogenous antioxidant enzyme expression, and activation of the Nrf2/ARE signaling pathway involved in oxidative detoxification.[27,29] Furthermore, MPE possesses anti-inflammatory properties by suppressing pro-inflammatory cytokine production, thereby reducing neutrophil activation and secondary ROS generation.[27,30]

In summary, these results demonstrate that MP exposure increases oxidative stress and lipid peroxidation in the intestinal epithelium, as evidenced by elevated MDA expression, whereas MPE supplementation effectively attenuates oxidative damage through combined antioxidant and anti-inflammatory mechanisms. The near normalization of MDA levels observed in the high-dose MPE group highlights the potential of mangosteen peel extract as a protective agent against intestinal toxicity induced by microplastics.

CONCLUSION

Exposure to LDPE microplastics significantly increased fecal microbiota counts and malondialdehyde expression in the intestinal epithelium of *Rattus norvegicus*. In contrast, supplementation with *Garcinia mangostana* L. peel extract effectively reduced both fecal microbiota counts and MDA expression in a dose-dependent manner. The highest MPE dose restored MDA levels to values comparable to those of the negative control group. These findings demonstrate that MPE exerts a protective effect against microplastic-induced intestinal disturbance by attenuating microbial imbalance and lipid peroxidation.

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